ECOPHYSIOLOGICAL STUDIES IN FRY AND FINGERLINGS OF SOME FRESHWATER FISHES WITH SPECIAL REFERENCE TO TEMPERATURE TOLERANCE

by

H. Mohamad Kasim, M. Sc.,
School of Biological Sciences, Madurai University,
Madurai-625 021, India.

A thesis submitted to the Madurai University for the Degree of
Doctor of Philosophy

1978

CONTENTS

ACKNOWLEDGMENTS

I am greatly indebted to Professor M.N.Kutty, who was a constant source of inspiration and encouragement throughout the course of this study. I am extremely grateful to him for rendering liberal help and advice at all times.

I am also grateful to Professor S.Krishnaswamy and Prof. A.Gnanam for providing necessary facilities and advice. I am extremely thankful for the valuable suggestions given by Professor F.E.J.Fry. I express my appreciation for the valuable suggestions extended by Dr E.T.Garside, Dr S.V.Job and Dr T.J.Pandian.

The Tamilnadu Fisheries Department has not only seen fit to grant extended leave of absence to the author, but has also supplied him the materials for the present study. It is a pleasure to acknowledge this support.

I gratefully acknowledge the help rendered by my colleagues in the preparation of this thesis.

Thanks are due to Mr K.V.Ravindran and Mr K.K.Hariharan for the typing work.

The financial support given by the U G C in the form of Junior Research Fellowship is gratefully acknowledged.

LIST OF FIGURES

LIST OF TABLES

Page

Introduction

Studies on survival of fish fry and fingerlings of the major cultivated freshwater species are important especially in view of the present stress on development of aquaculture throughout the world (Hickling, 1971; Bardach *et al*., 1972; Jhingran, 1975). This is specially so for India and other countries where certain developments in this direction have already been taken place such as the technology concerned with composite fish culture (Jhingran, 1975). In this context information on the influence of ecological factors such as temperature, salinity and oxygen on the survival of the fry and fingerlings is a major necessity. Such studies on Indian fishes are meagre and it is well known that mortality of fishes in the younger stages is a serious impediment in advancing fish culture.

There is also an additional importance for studying thermal requirements of fish and other aquatic organisms which inhabit especially the everchanging environment affected in many cases by thermal pollution owing to the increasing number of thermal plants. Precise knowledge of the thermal requirements of the economically important fishes is needed specially for recommending conservation measures. With these points on aquaculture and environment in view, the present study emphasizing thermal tolerance of fry and fingerlings of some important fishes of the region was taken up.

This investigation includes studies on two Indian major carps 'mrigal', *Cirrhinus mrigala* (Hamilton) and 'rohu', *Labeo rohita* (Hamilton), the fringe-lipped carp, *Labeo fimbriatus* (Bloch), the common carp, *Cyprinus carpio* (var-*communis*, Bangkok strain, Linnaeus) the freshwater mullet, *Rhinomugil corsula* (Hamilton) and the cichlid fish, *Tilapia mossambica* Peters. The former four species (only a few trials have been made with *L. fimbriatus*) are usually included in composite fish culture in India and the latter two are also important in fish production in this country.

There is no information on the thermal tolerance on mrigal, rohu and fringe-lipped carp. But some literature is available on certain aspects of thermal requirements of common carp, *Cyprinus carpio* and *Tilapia mossambica* (Black, 1952; Pitt *et al*., 1956; Allanson and Noble, 1964; Kutty and Murugapoopathy, 1969; Ananthakrishnan and Kutty, 1974). Thermal requirements of freshwater mullet to swim at specific speeds (temperatures causing swimming inhibition) have been studied by Kutty and Sukumaran, (1975); whereas thermal tolerance of this species has not been investigated so far. The present study will throw light on these aspects of thermal adaptation. Besides some aspects of salinity adaptations of these fishes have also been studied.

The objectives of the present study are to estimate the upper and lower lethal temperatures and to describe and compare the thermal resistance and tolerance of the selected species and also to study the effect of salinity on survival of the five species and the interaction of salinity with lethal temperature in one species.

Much of the literature available on the temperature tolerance of poikilotherms are based on studies on fishes (Gibson, 1954; Arai *et al*., 1963; Tyler, 1966; Strawn and

Dunn, 1967; Charlon, 1968; Garside and Jordan, 1968; Brett 1944, 1952, 1970; Charlon *et al.*, 1970; Fry *et al.*, 1942, 1946, 1971; Coutant, 1970; Brown and Feldmeth, 1971; Cairns, 1972; Fahmy, 1972, 1973, Garside and Chin-Yun Kee, 1972; Becker, 1973; Ihssen, 1973; Holland *et al.*, 1974; Feldmeth *et al.*, 1974; Ananthakrishnan and Kutty, 1974, 1976; Ananthakrishnan and Srinivasan, 1977; Otto and Rice, 1977). As early as in 1895 Regnard pointed out the ability of fish to survive in temperatures very close to freezing point of water. Maurel and Lagriffe (1899) studied both upper and lower levels of temperature tolerance in freshwater fish and commented on their better adaptability to low temperature than to high temperature.

Early workers measured the lethal temperature of fishes by heating or cooling the test medium at a constant rate (Huntsman and Sparks, 1924). This method was supplanted by the precise method of abrupt transfer to a series of preset temperatures from which percentage mortality and rate of dying could be obtained (Loeb and Wasteneys, 1912; Hathaway, 1924; Sumner and Doudoroff, 1938). The latter method was employed because it clearly demonstrates the effect of the thermal history (i.e., temperature acclimation) of fish (Fry *et al.*, 1942). Though a great deal of work has been executed without adequate regard for the conditioning effects of temperature, i.e., acclimation, many workers have contributed to the knowledge on temperature relations with due attention to this phenomenon (Binet and Morris, 1934; Sumner and Doudoroff, 1938; Heilbrunn, 1943). Subsequent studies on thermal tolerance clearly indicate that the influence of acclimation temperature is greater on thermal response than any other factors like salinity, photoperiod, endocrine activity, season, diet and size.

Further investigations by Fry et al., (1942) on systematic relationship between acclimation temperature, lethal temperature and exposure time enabled Fry (1946) to define a zone of thermal tolerance. This zone is bounded by upper and lower lethal temperatures within which the species concerned could be expected to survive the primary influence of the extreme temperatures i.e., biokinetic range. In addition to this McErlean et al., (1969) have proposed the thermal triangle which could be expected to give the area of upper thermal tolerance independently from the zone of thermal tolerance. Thus the study on thermal tolerance has progressively evolved from a measure of single value to a series of sequential response figures and then to an area of tolerance.

These temperature limits are often characteristic of the species and are genetically controlled, with minor variations among the individuals or populations of the species i.e., sexes, different life history stages, with different physiological status and geographical distributional differences (Fry, 1957). Aside from these small variations it is possible to divide fishes roughly into two groups: those which are restricted to a narrow range of temperature i.e., 'stenothermic' animals, and those which tolerate a wide range of temperature changes i.e., 'eurythermic'. As would be expected there are intergrades between these two groups (Brett, 1970).

Perhaps next to temperature, salinity of water is a factor of great importance. It is obvious that these two environmental factors, temperature and salinity are inseparably interdependent that the physiology of most aquatic organisms, especially those from brackish-water environment, is a functional outcome of the differential interaction of these two. Added to this, distribution and orientation of most aquatic animals are determined and restricted by these two factors (Hora, 1934; Raj, 1954; Bapat, 1970; Vernberg and Vernberg, 1971). Animals which occupy the everchanging environments like estuaries and those which migrate from freshwater to sea or vice-versa should possess an intricate mechanism of physiological adjustments in response to the changing salinity and temperature. The aspect of salinity tolerance at different temperatures forms the basic requisite for the understanding of the complex osmoregulatory mechanism.

Studies on salinity tolerance and adaptation have been carried out since as early as 1920 to date. Salinity tolerance, acclimation and effect on various aspects, such as growth, development, metabolism and iono-osmoregulation in fishes have been studied by several workers (Armitage and Olund, 1962; Potts and Parry, 1964; Potts and Evans, 1967; Nelson, 1968; Rao, 1969, 1971; Parvatheswararao, 1970; Prosser _et al_., 1970; Feldmeth and Waggoner, 1972; Mackay, 1974; Ahokas and Duerr, 1975 and others).

In the present study on salinity tolerance at different temperatures has been carried out on freshwater mullet and in the case of mrigal, fringe-lipped carp, common carp and _Tilapia mossambica_ the same study has been carried at ambient temperature (30° ± 1°C) alone. The genetically controlled tolerance to different salinities is very narrow in some species and very wide in some other species. This range of plasticity could be altered by non-genetic adaptations i.e., by acclimation to different factors (Black, 1962; Parvatheswararao, 1970; Prosser and Brown, 1965).

Salinity and temperature interactions on fishes have been investigated by many workers (Gibson, 1954; Blaxter, 1960; Blaxter and Holiday, 1963; Craigie, 1963; Brown, 1960; Lewis, 1966; Schlieper et al., 1952; Arai et al., 1963; Morris, 1960; Strawn and Dunn, 1967; Alabaster, 1967; Forrester and Alderdice, 1966; Garside and Jordan, 1968; Garside and Chin-Yun Kee, 1972). A review of these literature indicates that several studies have been made on larval, post-larval and adult form of fishes with different combination levels of salinity and temperature and specific influence of certain cations (Mg^{2+} and Ca^{2+}). The response exhibited by the fish are multiple and complex. There are indeed several unsolved questions in this specific field of ecophysiology.

In the present study the freshwater mullet R. corsula was used to assess the displacement of resistance to lethal temperature at various concentrations of salinity from different acclimation levels. This work is expected to explain the multiple response of this species, which is apparently a brackish-water species capable of survival and reproduction in fresh water (Pakrasi and Alikunhi, 1952; Narayanan, 1974) to various salinity and temperature levels.

Materials and Methods

Fishes used for the present study are : 1. Cirrhinus mrigala (Hamilton), an Indian major carp known as 'Mrigal' 2. Labeo rohita (Hamilton), another major carp known as 'Rohu', 3. Labeo fimbriatus (Bloch), known also as 'Fringe-lipped carp', a species commonly present in South Indian rivers, 4. Cyprinus carpio (var) communis, the common carp (Bangkok strain) Linnaeus, cultured widely all over the world, 5. Rhinomugil corsula (Hamilton), the fresh water mullet and 6. Tilapia mossambica Peters, a cichlid fish introduced in India over two decades ago (Panikkar and Tambi, 1954). In the case of carps, only fry were used. The fry of carps were procured from the Induced

Spawning Centre of Tamil Nadu Fisheries Department at Vaigai Dam located 30 miles Northwest of Madurai. The fingerlings of R. corsula were caught from Vaigai Reservoir itself and the hatchlings of T. mossambica were obtained from the mouth of parent fish collected from Avaniapuram pond which is located 10 miles South of Madurai. The details of size, weight source of collection types of experiments done on various species of fish and the periods of experiments are given in Table I. It can be noted in Table I that the freshwater mullet tested are much larger in size than the others. While the carps fry could be obtained from the Induced Breeding Centre, it was difficult to procure the smaller (fry) size of mullet from the reservoir because the 'natural breeding' takes place in the upper reaches of the river and the young ones descend to the reservoir only when they are over 4 cm (Narayanan, 1974). Eventhough some tests have been made, 'Induced Breeding' technique to produce the young ones of freshwater mullet has so far not been successful and an in vivo method for monitoring gonadial maturity was, however successfully worked out (Kasim, 1974).

As indicated in Introduction and also in Table I three separate series of experiments were made:

Table I

Details of size, source of collection and types of experiments size indicated are mean + one Standard Deviation (SD). The lengths and weight are given in Appendix from <u>Aa1</u> to <u>Ce13</u> in

No.	Species and Common Name		Weight (mg) mean + one (SD)	Size (cm) mean + one SD Fry/Fingerlin
1	<u>Cirrhinus mrigala</u> Indian major carp 'Mrigal'	TT ST	685.80 ± 162.24 15.97 ± 4.09	4.30 ± 0.31 (4 1.26 ± 0.09 (
2	<u>Labeo rohita</u> Indian major carp 'Rohu'	TT	628.97 ± 457.13	3.79 ± 0.86 (4
3	<u>Labeo fimbriatus</u> 'Fringe-lipped' carp	TT ST	331.93 ± 259.36 41.76 ± 15.98	2.97 ± 0.64 (5 1.76 ± 0.19 (
4	<u>Cyprinus carpio</u> 'Common carp' (Bangkok strain)	TT ST	225.20 ± 116.58 109.43 ± 36.15	2.55 ± 0.40 (4 2.24 ± 0.22 (
5	<u>Rhinomugil corsula</u> 'Freshwater mullet'	TT ST SE.TT	7211.47 ± 4230.52 2395.25 ± 1225.73 6825.12 ± 4609.93	9.46 ± 1.85 (5 6.77 ± 1.11 (2 9.35 ± 2.20 (3
6	<u>Tilapia mossambica</u> 'Tilapia'	ST	7.58 ± 1.06	0.77 ± 0.07 (6

TT:Temperaturature tolerance; ST=Salinity tolerance; SE.TT=Salinity effect on thermal tolerance.

Table I

done on the various species of fishes used in the study. The number of fish used are given within brackets. Individual fish raw data

Source of collection	Experiments done	Period of Study
Induced spawning centre, Dept. of Fisheries, Tamilnadu, Vaigai dam	a. Thermal tolerance b. Salinity tolerance	December 1976, January and February 1977
" "	a. Thermal tolerance	September 1975, Sept. October, November and December 1976
" "	a. Thermal tolerance b. Salinity tolerance	October, November and December 1976; Jan. and February 1977
" "	a. Thermal tolerance b. Salinity tolerance	Aug, Sept. 1974; Jan, Sept, and Oct. 1975; July, Aug, Sept, Oct, 1976; January and Feb. 1977
Vaigai reservoir Vaigai dam	a. Thermal tolerance b. Salinity tolerance c. Salinity effect on thermal resistance	Oct, Nov. 1975; Jan, Feb, Mar, May, June, Sept. Oct. 1976; Jan, Feb, 1977
Avaniyapuram pond, Madurai	a. Salinity tolerance	November 1975; April, May 1976; Feb, March, April, 1977

I Thermal tolerance

This study has been carried out in five species; of which four are carp, mrigal, rohu, fringe-lipped carp and common carp and the fifth one is freshwater mullet.

II Salinity tolerance

Experiments on salinity tolerance have been done on five species. Three of them are carps, mrigal, fringe-lipped carp and common carp and the other two are freshwater mullet and the cichlid fish, Tilapia mossambica.

III Salinity effect on thermal resistance

This work has been done only in the freshwater mullet.

Details of transportation, maintenance, acclimation and lethal baths are given under respective headings hereunder.

Transportation

Approximately 2000 fry of carp or 25 fingerlings of mullet were introduced into a polythene bag, half-filled with water and kept inside a square tin carrier (size 1.5'x1'x1'). The polythene bags were filled with oxygen to compensate for the loss of oxygen in the medium due to high respiratory rate of the fish consequent to handling(Brett, 1964;

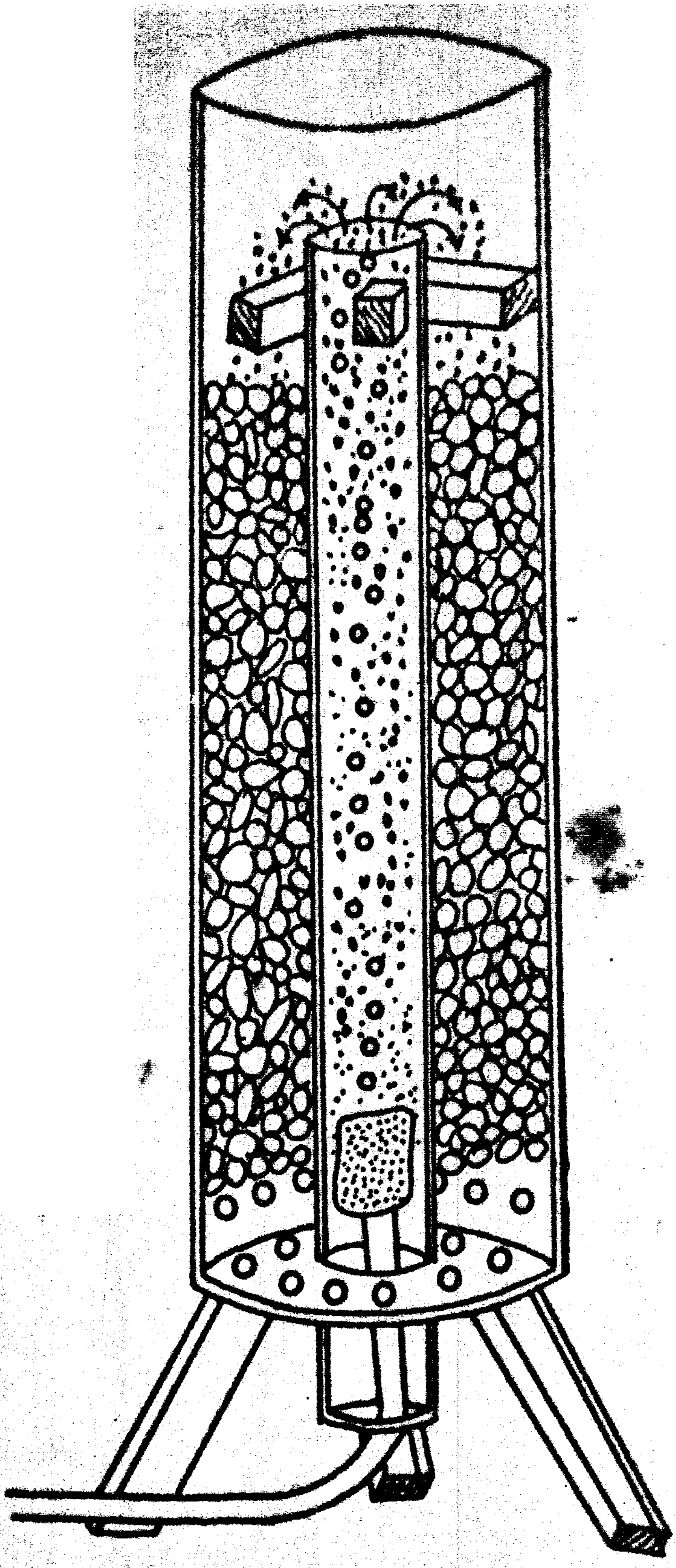
Fry, 1971). The percentage of mortality during transit was almost nil in the case of carps and it was observed that there was considerable level of mortality among mullet. This mortality among the mullet was apparently caused more by the handling effect due to cast netting than due to transportation.

Maintenance

The fish were stocked separately species-wise in 1500 lit. plastic collapsible tanks outside the laboratory. The stock tanks were well aerated to maintain the oxygen content near air saturation by passing compressed air through air diffuser made out of coral stones. Thousand to 3000 carp fry or 250 to 500 fingerlings of freshwater mullet were kept in each tank and the water in the holding tanks was recirculated by a pump through an overhead reservoir which has a 'Biological filtering-unit' (Fig.1) (Kutty et al., 1977). A constant water level was maintained in the upper reservoir (Tank 1) by using an electronic relay which pumps up water from a lower reservoir (Tank 2) (Fig.2). The pump was operated by an electronic water level control device (Electric Control Equipment Company, Madurai).

Fig.1

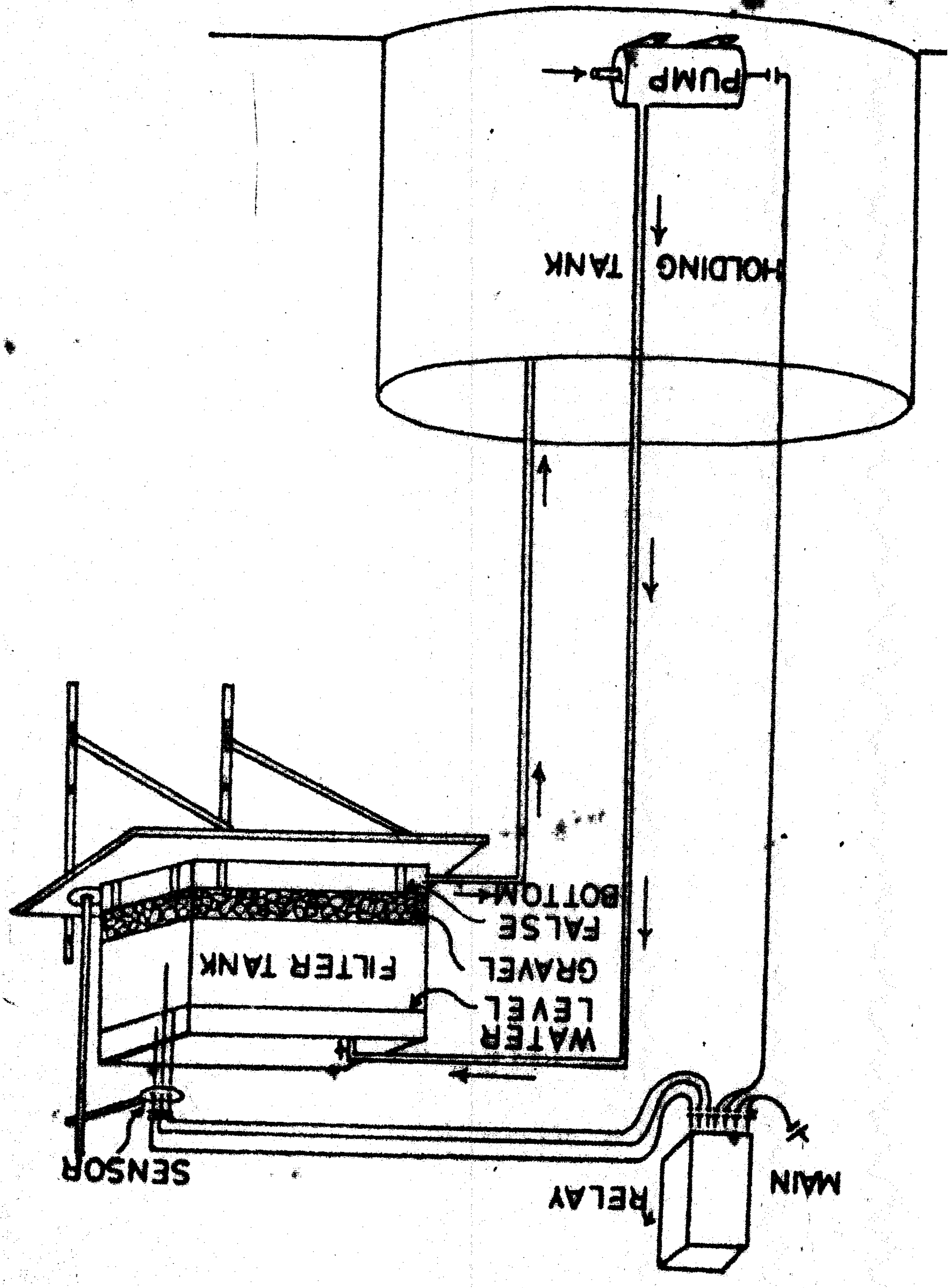
Diagrammatic representation of Biological filter-cum aeration unit



Biological Filter

Fig.2

Diagrammatic representation of water recirculating system and holding tank used for the maintenance of experimental fish during holding period


PUMP
HOLDING TANK
BOTTOM
FALSE
GRAVEL
WATER LEVEL
FILTER TANK
SENSOR
RELAY
MAIN

During holding period the fish were fed once in a day with a formulated diet prepared out of wheat heart (Rava), groundnut oil cake and boiled egg yolk in the ratio of 1:1:0.25. This formulated fish meal was found to be suitable for carps and mullet. The fish were kept in the holding tanks for not less than 15 days prior to their transfer to acclimation tanks. Temperature was not controlled in the holding tanks and the fluctuation was recorded to be ± 1.0°C from ambient level.

Acclimation

The physico-chemical properties of the University tap water in which the fish were acclimated are presented in Table II. Fry of carps and fingerlings of mullet were transferred from holding tanks to 75 lit. acclimation glass tanks, each tank holding about 100 fry of carps or 20 fingerlings of mullet. The water in the acclimation tank was stagnant when the acclimation was carried out at high temperatures, but the system was constantly aerated to maintain the dissolved oxygen concentration near air saturation. However, the water in the acclimation tanks was recirculated as explained under 'maintenance' at low temperatures. The acclimation water was changed by addition of fresh water on alternate days. Since accumulation of ammonia excreted by the fish

Table II

Physico-chemical content of Madurai University tap water used for the experiments in the present study

Temperature	30°C
Dissolved oxygen	6 mg/l
Dissolved carbon dioxide	2.86 mg/l
pH	7.7
Conductivity (in µMHOS/cm at 30°C)	425
Total hardness	236
Alkalinity Total (in terms of $CaCO_3$)	200 mg/l
Alkalinity phenolphthalein	30 mg/l
Chlorides	15.2 mg/l
Flurides	0.004 mg/l
Nitrites	0.04 mg/l
Phosphates	0.013 mg/l
Silicate	4.00 mg/l

Source: Unpublished data - Anandavalli,M. Department of Environmental Sciences, Madurai University, Madurai 625 021 in ambient water is toxic to fishes (Black, 1957; Kawamoto, 1961; Lloyd, 1961; and Burrows, 1964), a biological filter (Saeki, 1958; Kutty, 1966) as the one described by Kutty <u>et</u> <u>al</u>., (1977) was kept inside each acclimation tank to remove suspended particulate materials and ammonia in the ambient water.

The biological filtering unit (Fig.1) was made up of two cyolindrical acrylic plastic tubes open at both ends. The small inner tube (2.5 cm in diameter and 30 cm in length) was attached by 3 acrylic plastic strut inside the large cylinder (7.5 cm in diameter and 32 cm in length). The upper opening of the inner tube was about 2 cm lower than that of the outer big cylinder. The whole unit stood on three plastic legs on the floor of the aquarium. The space between the inner and outer tubes was filled with gravel and the water level in the aquarium was maintained at a level slightly lower than the upper end of the inner tube. **An air diffuser stone** was kept inside the inner tube near its base. Water was air lifted by vigorous bubbling and the upward displacement of air and the water spilled over into the outer tube and was filtered while flowing down the gravel bed. It is known that such gravel filters in due course develop ammonia fixing bacteria which can remove the excreted ammonia from the water (Saeki, 1958; Kutty, 1968). Kutty, Narayanan and Sukumaran (1977) have shown that the filters used were efficient in removing ammonia in water.

The photoperiod was not controlled; the fish were exposed to natural day light normally, but occasionally the night lights were on for a few extra hours.

The feeding schedule during the acclimation period was the same as it was for the fishes in the stock tanks.

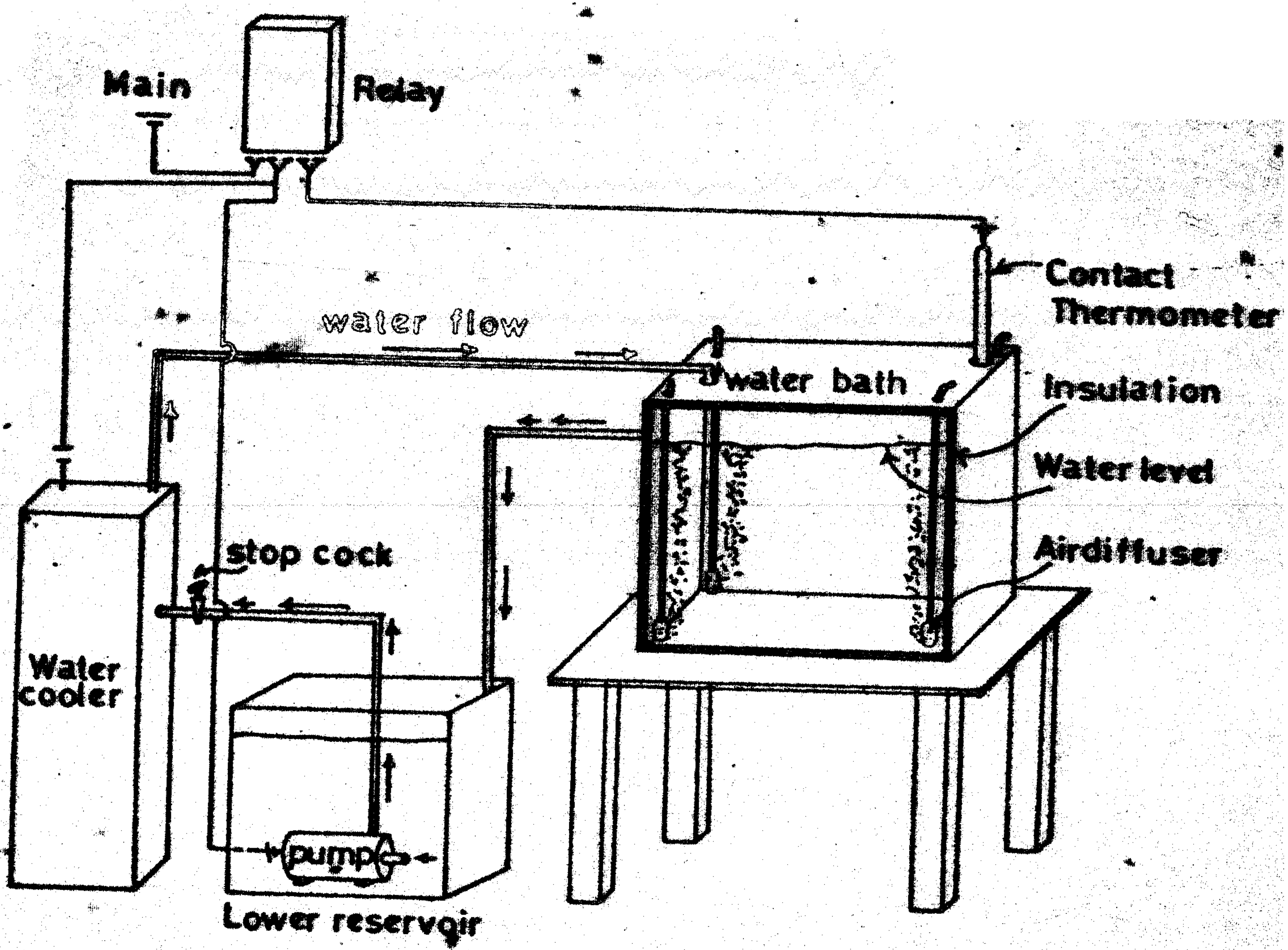
Temperature acclimation

The fry and fingerlings were acclimated to 15, 20, 25, 30, 35 and 38°C for thermal tolerance studies. The highest acclimation level for common carp and freshwater mullet was 35°C and for others it was 38°C. For salinity tolerance studies the temperature was not controlled in the acclimation tank for all the five species and the temperature variation was recorded to be ± 1°C from the mean ambient temperature, i.e., 28 and 30°C in the case of freshwater mullet and carps respectively. Among all the six species, fresh water mullet alone was tested for the effect of salinity on thermal tolerance (Table I) and was acclimated to 20, 25, 30 and 35°C prior to experimentation.

During the high temperature acclimations the ambient water temperature was maintained at 30 and 35°C with the help of 'Jumo' contact thermometer and a thermostatic relay (Electric Control Equipment Company, Madurai) which controlled an immersion heater. For thermal adaptation, the temperature was raised or lowered by 1°C per day from ambient temperature before attaining the desired acclimation temperature (Brett, 1952; Fry, 1957; Beamish, 1964). Thereafter the temperature was maintained constant for a minimum period of one week (Brett, 1964; Beamish, 1964). The temperature variation during high temperature acclimation was ± 0.1°C. For low temperature acclimations i.e., 25°C and below a common water cooler (Voltas Limited, Bombay), a submersible pump (Little Giant Pump Co., USA), an electronic relay (Electric Control Equipment Co., Madurai) and a 'Jumo' contact thermometer were used (Kasim et al., 1977). Two glass aquaria 70 lit. capacity served as the upper acclimation tank and a lower reservoir. The immersion pump in the lower reservoir pumped water through the cooler into the upper acclimation tank. The overflow of the acclimation chamber was drained back into the lower reservoir; thus a constant water circulation was maintained. The 'Jumo' contact thermometer in the acclimation chamber activated the cooler and the pump simultaneously whenever

Fig.3

Diagrammatic representation of low temperature water bath and connected water recirculating system used. as low temperature experiments



CONSTANT LOW TEMPERATURE RECIRCULATING WATER BATH the temperature went up due to heat gain from the surrounding atmosphere and the temperature fluctuation was ± 0.05°C After attaining the required acclimation level, the temperature was maintained constant for a minimum period of two weeks.

Lowest temperature in which acclimation was done was 15°C for all the species studied except common carp (20°C). Such a limitation could not be overcome because of the difficulties of regulating required lower test temperatures over extended period of time under the local conditions (Kasim _et al._, 1977) and also because it was felt that a reasonable picture of thermal response can be obtained inspite of this limitation owing to the general higher thermal adaptation of tropical fishes (Kutty and Murugapoopathy, 1969; Allanson and Noble, 1964; Brett, 1970; Ananthakrishnan and Kutty, 1974, 1967) (see also following text).

Salinity acclimation

Among the five species tested for salinity tolerance (Table I) _L. fimbriatus_ alone was subjected to salinity acclimation. Sudden exposure of this species from fresh water to different higher salinities showed that there was no mortality at 5‰ (S) for a period of 10,000 min.

Hence, this species was kept at 5‰ (S) for a minimum period of 10 days in a 70 lit. aquaria. The temperature was not controlled and the temperature remained at 30° ± 1°C. The water was well aerated with air diffuser stones to keep the oxygen level near air saturation. Same feeding schedule was maintained as for fish under various temperature acclimations. Salt water was prepared by mixing commercial salt (Anil Brand Salt Co., Madurai) with university tap water (Table II) (Raynold, 1974; Ahokas and Duerr, 1975) and the salt water was replaced every alternate day in the acclimation tank.

Experimental set-up

Two types of test tanks were used for lethal baths. One (12" x 18" x 12") was used as a low temperature test tank and is made of acrylic plastic transparent sheets with a built-in filtering unit. The others were glass aquaria (14" x 24" x 14") mainly used for high temperature experiments. These tanks were insulated by 3" thick 'thermocole' to minimise the heat dissipation. The temperature in the test tanks was maintained as explained for acclimation (both high and low). The temperature fluctuation during the tests at high temperature was ± 0.1°C and at low temperature it was ± 0.05°C. A thorough mixing of water was obtained by bubbling air on all the corners of the test tanks through air-diffuser stones. This, apart from preventing thermal stratification in the test tanks, maintained the oxygen level near air saturation.

Temperature in all these tanks were counter-checked with a standard thermometer (Gold E-Mil Line, England EN 13884) at frequent intervals throughout the experiments. The water was stagnant in all the high temperature tests and water loss due to evaporation in high lethal baths were equalled by addition of water of the same temperature when required. As already explained under 'acclimation' the biological filtering unit (Fig.1) in the test tanks prevented accumulation of toxic nitrogenous waste materials and suspended particles in the test water.

temperature it was ± 0.05°C. A thorough mixing of water was obtained by bubbling air on all the corners of the test tanks through air-diffuser stones. This, apart from preventing thermal stratification in the test tanks, maintained the oxygen level near air saturation.

Temperature in all these tanks were counter-checked with a standard thermometer (Gold E-Mil Line, England EN 13884) at frequent intervals throughout the experiments. The water was stagnant in all the high temperature tests and water loss due to evaporation in high lethal baths were equalled by addition of water of the same temperature when required. As already explained under 'acclimation' the biological filtering unit (Fig.1) in the test tanks prevented accumulation of toxic nitrogenous waste materials and suspended particles in the test water.

Experimental procedure

I Thermal tolerance

Two methods are available to approach the problem of describing the lethal limits of temperature. The first method involves exposing the sample of animals in a lethal bath in which the medium is warmed or cooled from ambient temperature at a constant rate until all the animals succumbed (Vernon, 1899; Huntsman and Sparks, 1924; Battle, 1926; Cocking, 1959). This method has been widely criticised for two reasons. Steady and constant warming or cooling of the medium involves a time factor which could lead the animals to further adaptation to high or low temperatures. As an example Gibson (1954) working with Lebistes reticulatus obtained a median lethal level 7 to 8°C lower than that for heat-coma, as recorded for the same species by Tsukuda (1960). Similar differences occured between the 'critical thermal maximum' (heating at 0.5°C/min) and the lethal temperatures for 3 species of shallow water marine fish (Heath, 1967). Secondly the data could not be adequately explained satistically (Fry, 1948; Bovee, 1949). The second method involves exposing the samples acclimated at different non-lethal temperatures abruptly to high and low lethal temperatures which ranges from rapidly lethal to sublethal limits. The latter method which eliminates the two problems encountered in the first method has been employed in the present study.

This method of lethal temperature assessment has been employed by Fry and his associates and other workers (see Fry, 1971). Eventhough the principles of the method remains basically the same; the maximum time of exposure varied notably with different workers. The data have been statistically treated and graphically interpreted by various workers (Fry et al., 1942; Brett, 1944, 1952; Doudoroff, 1945; Fry, 1941; Fry et al., 1946; Hart, 1947, 1949; McLeese, 1956; Edney, 1964; Allanson and Noble, 1964; Kutty and Murugapoopathy, 1969; Ananthakrishnan and Kutty, 1974, 1976). An extensive discussion on the statistical treatment of the data and widely acceptable terminology that are used to describe the observations of the events of lethal temperature determination has been given by Brett (1952). The terms and explanations given herein are taken mainly from Brett (1952).

Lethal temperature

Upper: The carp fry and freshwater mullet fingerlings acclimated to 15, 20, 25, 30, 35 and 38°C were exposed to different high lethal temperatures ranging from sharp lethal to sublethal levels at an interval of 1 or 0.5°C. The shorter temperature interval was adopted for tests near sublethal levels. This interval was found to be adequate for enabling the reasonable interpretation of data (Miller and Tainter, 1944). The level of lethal and sublethal temperatures were worked out by preliminary random exposures of the above materials before proper finer tests were made. Time to death of individual fish of all the samples were noted down by close inspection. Complete arrest of opercular beats and the onset of muscular contraction, followed by complete loss of response to external stimuli are decisive symptoms to conclude that the animal is dead (Brett, 1944, 1952; Hart, 1947). Length and weight of the dead fish were recorded immediately after removing from lethal baths.

Lower: Low lethal temperature tests were conducted at an interval of 0.5°C from 6° to 21°C. Temperatures lower than 6°C were not tested, for the reasons explained before, namely, limitations of the techniques and facilities for maintaining low temperatures precisely for long periods, eventhough the apparatus adaptation did provide the needed accuracy (Kasim et al., 1977), but not the range of temperature required and also the reason that the temperature range tested and the techniques available would suffix the requirements of delineation of thermal range of tropical fishes reasonably, as already shown in earlier studies (Allanson and Noble, 1964; Kutty and Murugapoopathy, 1969; Ananthakrishnan and Kutty, 1974, 1976).

Relatively it was easier to decide when a fish died from high temperature. In high lethal temperature the fish swim wildly around the test chamber immediately when exposed and the death is sudden with a comparatively shorter period of time from the time of loss of balance to the time of death. On the contrary in low lethal temperature the fish appears to be in a stage of 'suspended animation' (Brett, 1952) and there is a long lapse of time between loss of balance and death.

Exposure time

Various investigators have used different durations of exposure in lethal baths to determine the lethal temperatures and this has been a matter of controversy. Fry *et al*., (1942) exposed their fish for a period of 14 hrs and Brett (1944) shortened the exposure time to 12 hrs. Subsequently, Brett (1952) has carried out experiments for Pacific salmons even as long as 7 days. Edney (1964) found the upper lethal level is lowered when isopods were exposed to 12hrs instead for only 30 min. However, the mortality due to temperature as a primary cause ceases after a particular resistance time in many animals (Brett, 1952; Fry, 1947). Beyond this period 50% or more of the exposed population survive for an indefinite period of time and this varies

Samples of 10 fish were drawn from acclimation tank and sudden exposures were made at different preset lethal salinity and temperature combinations. Table III gives the details of different combinations of salinity and temperature used for the freshwater mullet. Temperature of the test medium was controlled at different levels only for R. corsula. But for other four species including salt-water acclimated L. fimbriatus the experiments were carried out at ambient room temperature (30 + 1°C) and the salinity range was from 5 to 35‰ (S). The saline test medium was prepared as mentioned for salinity acclimation. The elevation in salinity due to evaporation was compensated by addition of distilled water and it was constantly checked with a 'salinometer' (Yellowspring Instrument Co., USA). The salinity fluctuation was within 0.5‰ (S) for any set of experiments. Time to death of individual fish was recorded by close inspection and the length and weight of individual fish were noted down.

III Salinity effect on thermal resistance

Salinity effect on thermal resistance was studied only in R. corsula at high temperatures. Fingerlings of R. corsula previously acclimated to four different temperatures (see 'acclimation') were tested to potentially lethal

Table III

Different salinity and temperature combinations employed for salinity tolerance study among the fingerlings of R. *corsula*. Places (x) marks indicate the specific salinity temperature combinations in which tests have been made

Test Temp. (°C)	Salinity ‰											
	45.00	40.00	35.00	30.00	26.25	25.00	20.00	18.00	17.50	15.00	12.50	10.00
35.0	-	x	x	x	x	-	-	x	x	x	x	x
30.0	-	x	x	x	x	-	-	x	x	x	x	x
26.0	-	x	x	x	-	x	x	x	x	x	x	x
20.0	-	x	x	x	-	x	x	-	-	x	x	x
17.5	x	x	x	x	-	x	x	-	-	x	x	x

temperature and salinity combinations. The different combinations of lethal temperature and salinity employed for this study are presented in Table IV. Samples of fish from different acclimations were exposed abruptly, as indicated in the Table IV, and the time to death of individual fish were recorded to assess the salinity effect on thermal resistance by statistical analysis.

Analysis of data

As referred to already the method of analysis and terminology adopted in the present study are those described by Brett(1952) except in a few specific cases as indicated. The primary analysis of the thermal and salinity tolerance and the effect of salinity on thermal resistance data involved probit analysis. Then the processed data were subjected to regression analysis to differentiate the resistance pattern with respect to different factors. To distinguish the significance of resistance among the species the primary data of all the experiments were subjected to analysis of variance. The lethal levels of temperature and salinity which might cause 50% mortality among the population were calculated by the method of Miller and Tainter (1944) and these values of temperature alone were used further to define zones of resistance and tolerance qulitatively and quantitatively.

Different combinations of lethal temperature and salinity employed for the assessment of salinity effect on thermal resistance among *R. corsula*. Places (x) markes indicate the specific lethal temperature and salinity combinations in which tests have been made

Accli. temp. (°C)	Test Temp. (°C)	Test salinity ‰									
		25	20	18	15	12	10	7	5	3	F.W.
35	37, 39, 41	-	-	-	x	-	-	x	-	-	x
30	37, 39, 41	-	-	-	x	-	-	x	-	-	x
25	39	-	x	-	x	x	x	x	x	x	x
20	37	x	x	x	x	x	x	x	x	x	x

F.W = Fresh water

a. Probit analysis

When the resistance times of individual fish of a sample i.e. the cumulative time to death, are plotted on an arithmetic graph, sigmoid curves could be fitted through the plots (Fig.4). These mortality curves can be converted into simple straight lines as it has been demonstrated by Bliss (1935, 1937), when plotted on a probability chart in which the axes have been converted into probability units as one variable (Y-axis) and logarithm of time as the other variable (X-axis). From these levels 50% mortality time can be read (Fig.5). This 50% mortality time is usually referred to as 'median resistance time' or 'median lethal time' (TLM 50) which has been shown to be same as the 'geometric mean time' in most cases (Fry et al., 1946). Hence, the median resistance time is the time within which a population will suffer 50% mortality at the given lethal temperature for the particular acclimation level. Thus a series of median resistance times are estimated for every group of animals acclimated to a temperature or salinity and exposed to various lethal levels. The validity of relation and the normality of distribution which makes it feasible for further statistical processing have been much emphasized by Fry et al., (1946), Hart, (1947, 1949) and Brett (1952).

b. Regression analysis

A series of median resistance times are estimated for the group of animals acclimated to a temperature and exposed to different lethal levels. When all these median resistance times are plotted on a semi-logarithmic grid against corresponding levels of temperature, straight, sloping lines can be fitted through plots of the logarithm of median resistance time against test temperature by 'least square' method (Fig.6). These regression lines can be expected to picturize the resistance of the species concerned at respective state of acclimation and thus facilitates a comparative study.

c. Incipient lethal level

The incipient lethal levels are the doses at which 50% of individuals of a sample exposed will die or conversely dose at which 50% of the population can be expected to survive for an indefinite time of exposure (Brett, 1952). The evaluation of these incipient lethal levels by experimentation needs that the tests should be conducted at closer intervals and the levels which cause 0% to 100% mortality are alone taken for analysis. Safety dosage calculation method advanced by Miller and Tainter (1944) was employed in which the probit and logarithmic units were used. Lethal levels are shown by a boundary line (broken line) (Fig.6) which cuts the lower ends of all the regression lines and there is a distinct break in the logarithmic plot. The observation of this break (Fry *et al*., 1946; Brett, 1952) and its changing nature with acclimation is most significant and constitutes the main difference in present experimental procedure from other investigations (Kennedy *et al*., 1974a and b; Feldmeth *et al*., 1974; Holland *et al*., 1974; Hidu *et al*., 1974; Raynold and Thompson, 1974; Andronikov, 1975; Wallis, 1975). This boundary demarcates the 'zone of tolerance' from the 'zone of resistance'.

d. Zones of resistance and tolerance

As mentioned already the upper and lower incipient lethal temperatures are further used to demarcate the biokinetic range of the species concerned by plotting incipient lethal temperatures against respective acclimation temperatures on an arithmetic graph. This results in the formation of a trapezium (Fry *et al*., 1942; Brett, 1952; Ananthakrishnan and Kutty, 1976). This trapezium has an additional construction line running at an angle of 45° to both the axes. This line traverses through all points at which measurements along each axis have equal values (Fig.41). In the upper and lower temperature regions the lethal and acclimation temperatures meet this 45° line at two points separately providing finally an ultimate upper and lower lethal temperatures (Fry et al., 1946; Brett, 1952). Beyond these ultimate lethal temperatures the extension of thermal tolerance becomes impossible whatever may be the state of acclimation and duration of experiment. Thus the area bounded by the incipient lethal temperatures in the shape of a polygon is known as the 'zone of tolerance' and beyond which lies the 'zone of resistance'. The area of the zone of tolerance can be expressed in degree centigrade squared (°C^2) units (Fry <u>et</u> <u>al</u>., 1942) and this can be expected to show quantitatively the thermal tolerance of any species investigated by these procedures.

c. <u>Analysis of variance</u>

This method of analysis have been used to differentiate the sources of specific variations from those resulting from sample variability coupled with interaction of acclimation, lethal temperatures and resistance time. Since almost the same acclimation and lethal temperatures were employed for each species, it was possible to consider the data for the following relations: i.e.

1 species x lethals, for each acclimation temperature
2 species x acclimations, for each lethal temperature
3 lethal x acclimations, for each species

The following method of compilation to elucidate the significance of resolved components from a measure of the unresoved variability (error) by F test was taken mainly from Brett(1952) and Snedecor (1967)

x = log median resistance time

Tx = total median resistance time (log)

S = sum of all items

G = Grand total

df = degrees of freedom = (n-1), (k-1)

C = correction factor = G^2/nk

Model Table (after Brett, 1952)

		1	2	3	....	n	
L*	1	Tx1	......	......	....	Txn	S_1
e t h	2	⋮					S_2
a l s	3	⋮					S_3
A** c c l i m a t	⋮	⋮					⋮
i o n s	k	Txk					Sk
		S_1	S_2	S_3	....	Sn	G

*Lethals = Lethal test temperatures

**Acclimations = Acclimation temperatures

Sum of Squares

1 Between species $= \overset{n}{\underset{1}{S}} \left[\overset{k}{\underset{1}{S}} (Tx)^2 \quad k \right] - c$

2 Between lethals $= \overset{k}{\underset{1}{S}} \left[\overset{n}{\underset{1}{S}} (Tx)^2 \quad n \right] - c$

3 Error $= (4) - (1) + (2)$

4 Total $= \overset{nk}{\underset{1}{S}} (Tx)^2 - C$

Orthogonal comparison is a second step in the analysis of variance in which the sum of squares are broken down into proportionate amount of variation pertaining to single degree of freedom and in turn subject to 'F' test with the error term of concerned sum of square.

Table of multipliers used for orthogonal comparison between each species using total log resistance time for all lethals and all acclimation per species

Species	1	2	3	4	5
Multipliers	1	1	0	-1	-1
	1	1	-4	1	1
	1	-1	0	1	-1
	1	-1	0	-1	1

Apart from orthogonal comparison, as a further step non-orthogonal comparisons have also been done as per the Table shown below:

Table of multipliers used for non-orthogonal comparison between each species using total log resistance times for lethals and acclimations of 25 , 30 and 35°C

Species	1	2	3	4	5
Multipliers	1	-1	0	0	0
	1	1	-2	0	0
	0	0	-2	1	1
	0	0	0	1	-1

The analysis of variance, orthogonal and non-orthogonal comparisons are further explained with due results of present work in discussions.

Results

Results for the three series of experiments as explained in 'Materials and methods' are presented separately hereunder in the following order:

I Thermal tolerance
II Salinity tolerance
III Salinity effect on thermal resistance

I Thermal tolerance

Results of the thermal tolerance of the five species studied are presented as upper and lower thermal tolerance separately for the respective species.

a) Upper thermal tolerance:

i. Mrigal:

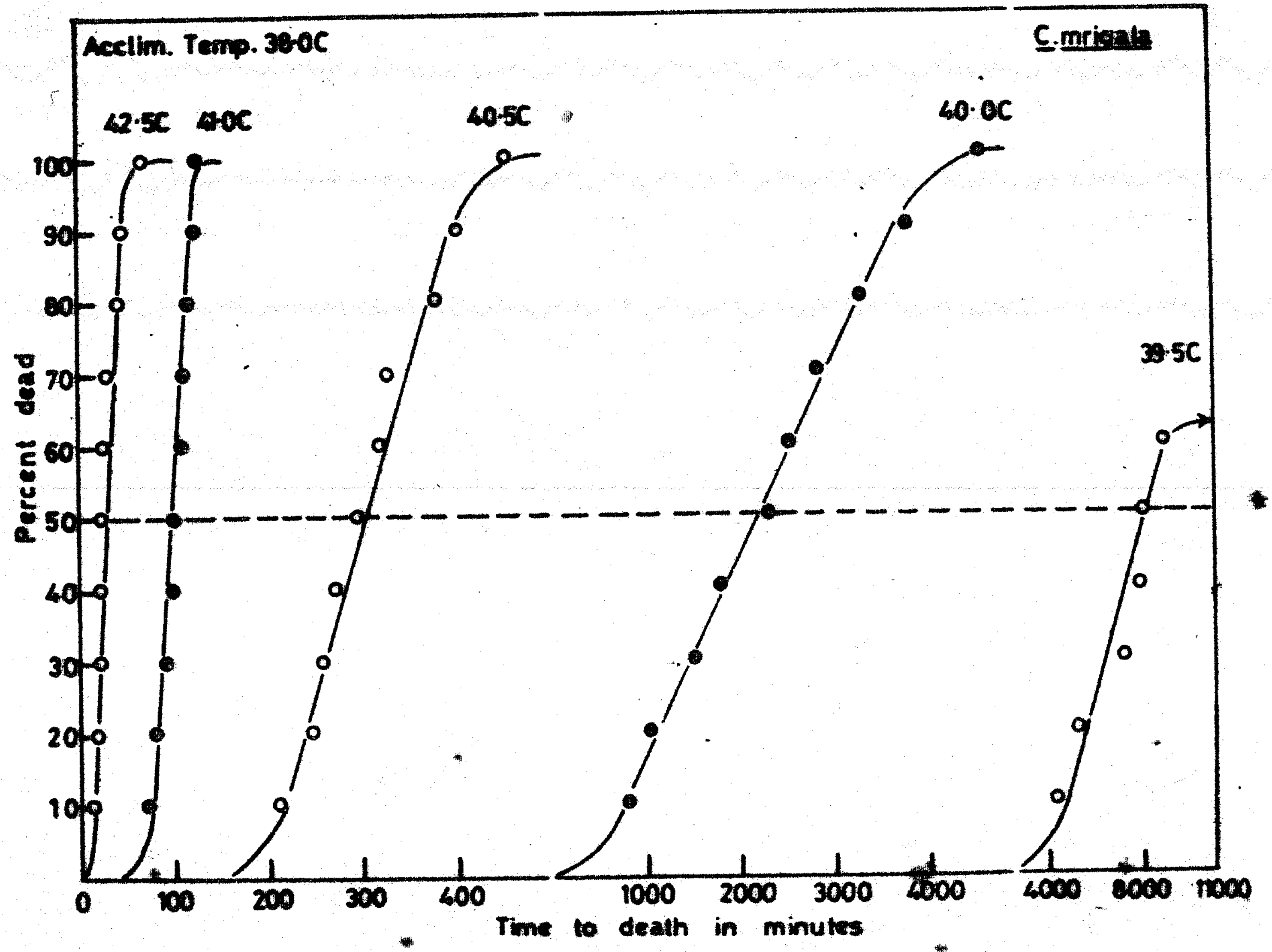
The raw data obtained on time to death in minutes of mrigal acclimated to six different temperatures (15, 20, 25, 30, 35 and 38°C) and exposed to various high lethal temperatures (from 35.0 to 42.5°C) with other relevant details such as length and weight, are presented in appendices Aa1 to Aa6. The analysis of these data is done according to the methods described by Fry et al., (1946) and Brett (1952) as already referred to under 'Methods'.

As an example of the data obtained, resistance times (time to death in minutes) of individual fish in lots (acclimated to 38°C and tested to various high lethal temperatures, namely, 39.0, 39.5, 40.0, 40.5, 41.0, 42.0 and 42.5°C) are plotted against percent fish dead, in Fig.4. As explained under 'Methods', such arithmetic plot curves fitted through the points, obtained for each lot of fish are generally sigmoid in shape, just as it was obtained by earlier workers in similar studies (Fry et al., 1942; Brett, 1952; Ananthakrishnan and Kutty, 1976). It is also seen in Fig.4 that these sigmoid curves shift towards the time axis as the high lethal temperature becomes less. The mortality was 100% in 42.5, 41.0, 40.5 and 40.0°C, but in 39.5°C the

Fig.4

Times to death at different high lethaltest temperatures among mrigal fry acclimated to 38°C.

Plotted on arithmetic axes


Acclim. Temp. 38·0C
C. mrigala
42·5C
41·0C
40·5C
40·0C
39·5C
Percent dead
10
20
30
40
50
60
70
80
90
100
0
100
200
300
400
1000
2000
3000
4000
4000
8000
11000
Time to death in minutes

mortality was only 60% as indicated by the arrow terminating the shorter curve. Intersections of the horizontal broken line with the curves in Fig.4 indicate times to 50% mortality for the respective test temperatures.

The probit-logarithmic transformation (vide 'Methods') of the data in Fig.4 is shown in Fig.5. Times to 50% mortality, referred to as 'median resistance time' or 'median lethal time' (TLM 50) are taken from these curves for further analysis. The geometric mean values estimated for the same data (closed circles with ± one SD in Fig.5) indicate that there is no significant difference between these values and median resistance times, as also shown by earlier workers (Fry *et al.*, 1946; Brett, 1952).

Median resistance times for other acclimation levels obtained similarly for various high lethal temperatures in mrigal are collectively presented in Table V. It is seen from Table V that for each acclimation, with an increase in test temperatures the median resistance time decreases. It is also seen that the median resistance times are characteristic of each acclimation temperature. However, with an increase in acclimation temperature the median resistance time also increases. These observations are in consonance with the findings of earlier workers on other fishes (Fry *et al.*, 1946; Brett, 1952).

Fig.5

Times to death at different high lethaltest temperatures among mrigal fry acclimated to 38°C. Plotted on probit x logarithmic axes. Calculated geometric mean resistance times (closed circles, horizontal bars indicated one S.D) coincide with the median resistance times (at probit 5.0)

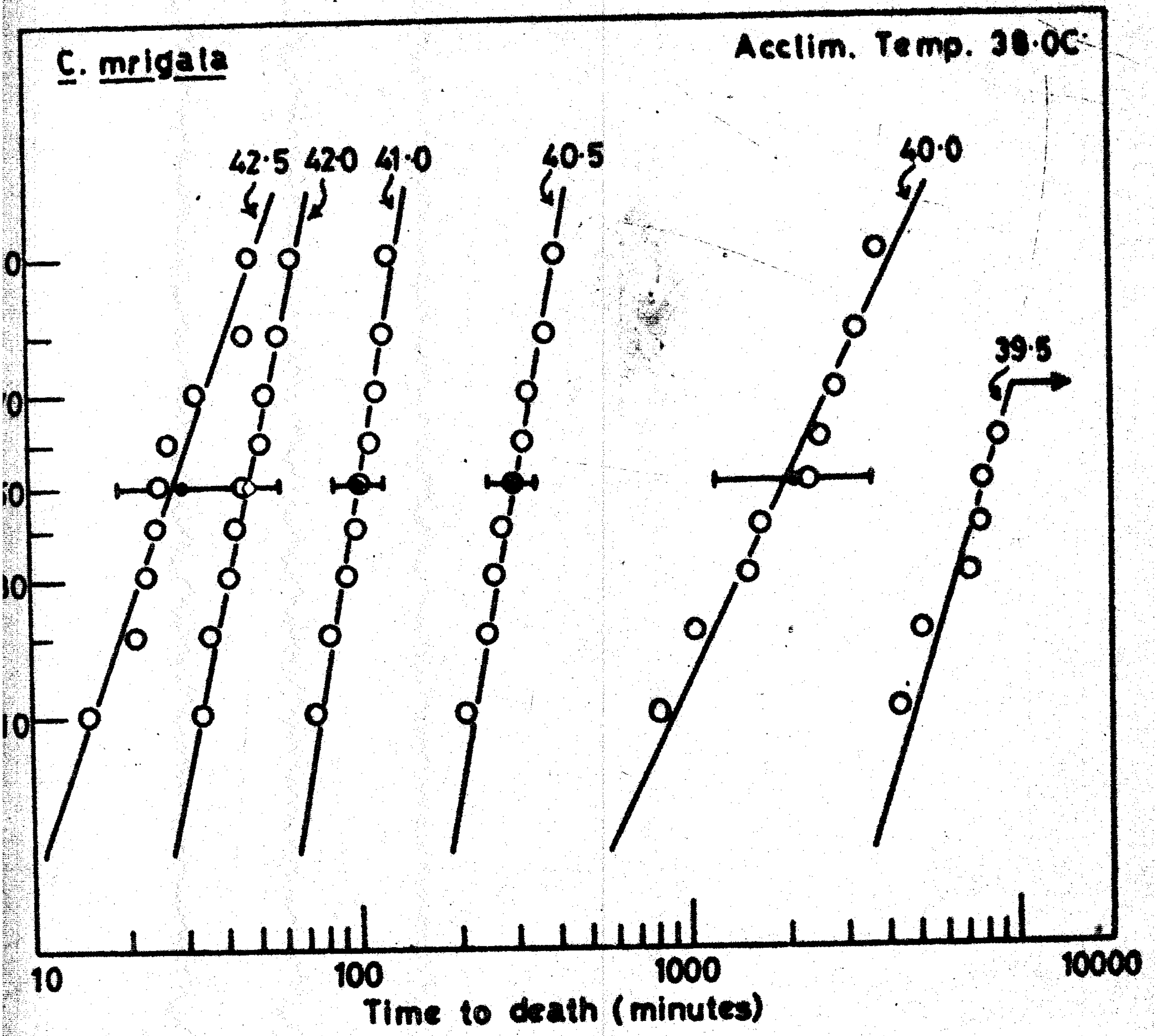

C. mrigala
Acclim. Temp. 38·0C
42·5
42·0
41·0
40·5
40·0
39·5
0
70
0
0
10
10
100
1000
10000
Time to death (minutes)

Table V

Median resistance times to high lethal temperatures of mrigal (Cirrhinus mrigala) fry acclimated to various temperatures

Acclimation Temp. (°C)	Lethal temperatures in °C and median resistance times in minutes													
	42.5	42.0	41.0	40.5	40.0	39.5	39.0	38.5	38.0	37.5	37.0	36.0	35.5	35.0
38	28	47	100	298	2000	8000	-	-	-	-	-	-	-	-
35	-	36	66	120	255	6500	-	-	-	-	-	-	-	-
30	-	-	21	64	115	250	5800	-	-	-	-	-	-	-
25	-	-	-	-	-	11	34	48	-	-	-	-	-	-
20	-	-	-	-	-	-	-	-	15	24	51	-	-	-
15	-	-	-	-	-	-	-	-	-	-	-	15	33	110

The data on median resistance times of mrigal are plotted on arithmetic axes against respective high lethal temperatures for each acclimation, in Fig.6. The curves fitted through the plots indicate that with the decrease in high lethal temperature the median resistance time increases until the incipient lethal level is reached, as indicated by the point where the curve becomes parallel to time axis. It is also seen in this figure that the space between the parallel lines of each acclimation on the time axis is progressisvely reduced with the increase in acclimation temperature. This gradual reduction suggests that the difference in incipient lethal temperature becomes progressively less at the higher acclimation temperatures. The data presented in Fig.6. are shown on a semilogarithmic graph in Fig.7 and regression lines have been fitted through the plots. These regression lines are straight and negatively sloping and demonstrate the characterstic resistance response of this species to high lethal temperatures for different acclimation temperatures. These regression lines have been terminated by a broken line at the lower ends, beyond which the lines are extended as parallels to the x axis. The flexion points of these curves denote that at temperatures below that indicated by the broken line, there will not occur 50% mortality in the samples

Fig.6

Median resistance times to high temperatures among mrigal fry acclimated to temperatures indicated. Plotted on arithmetic axes

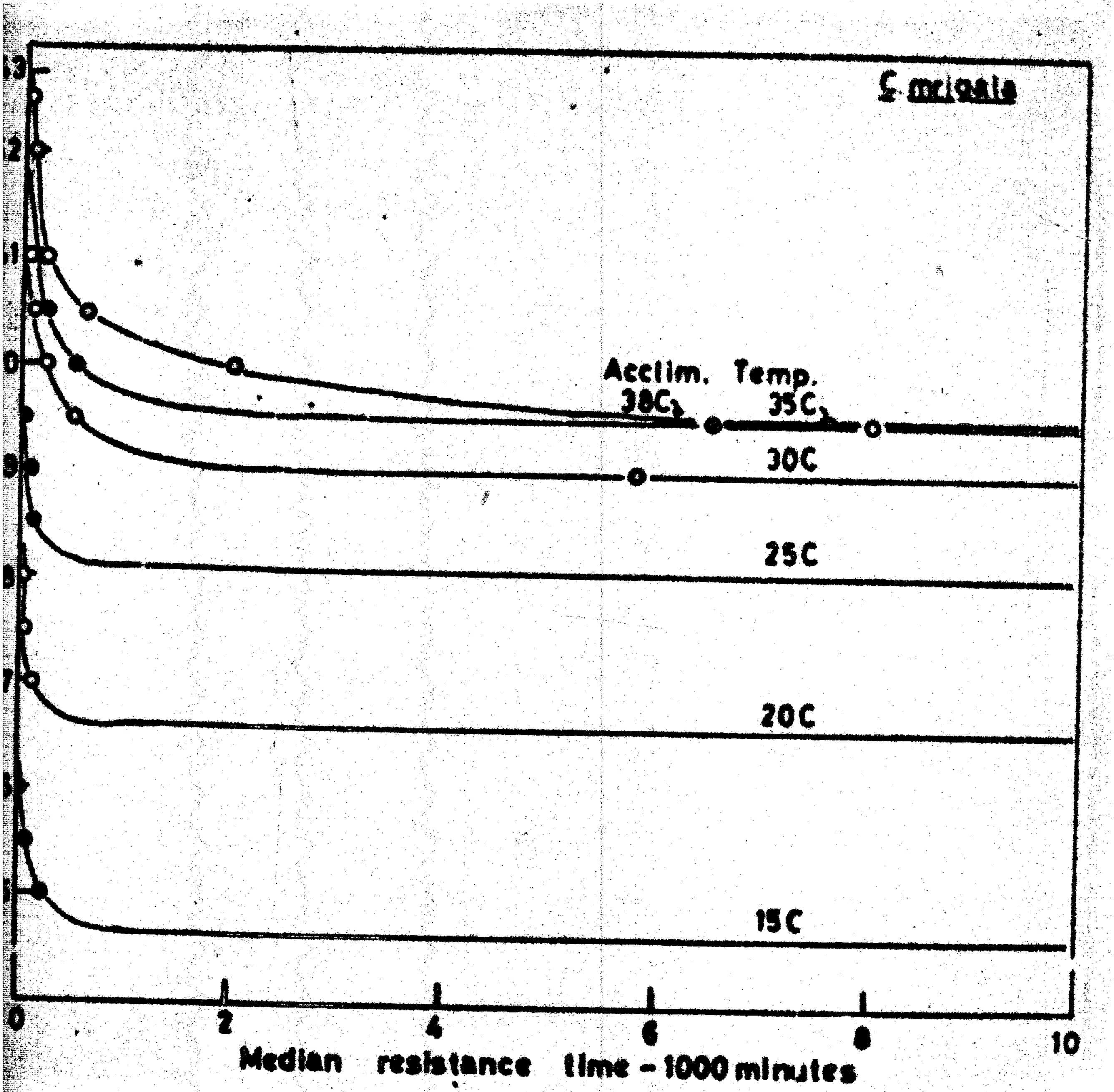
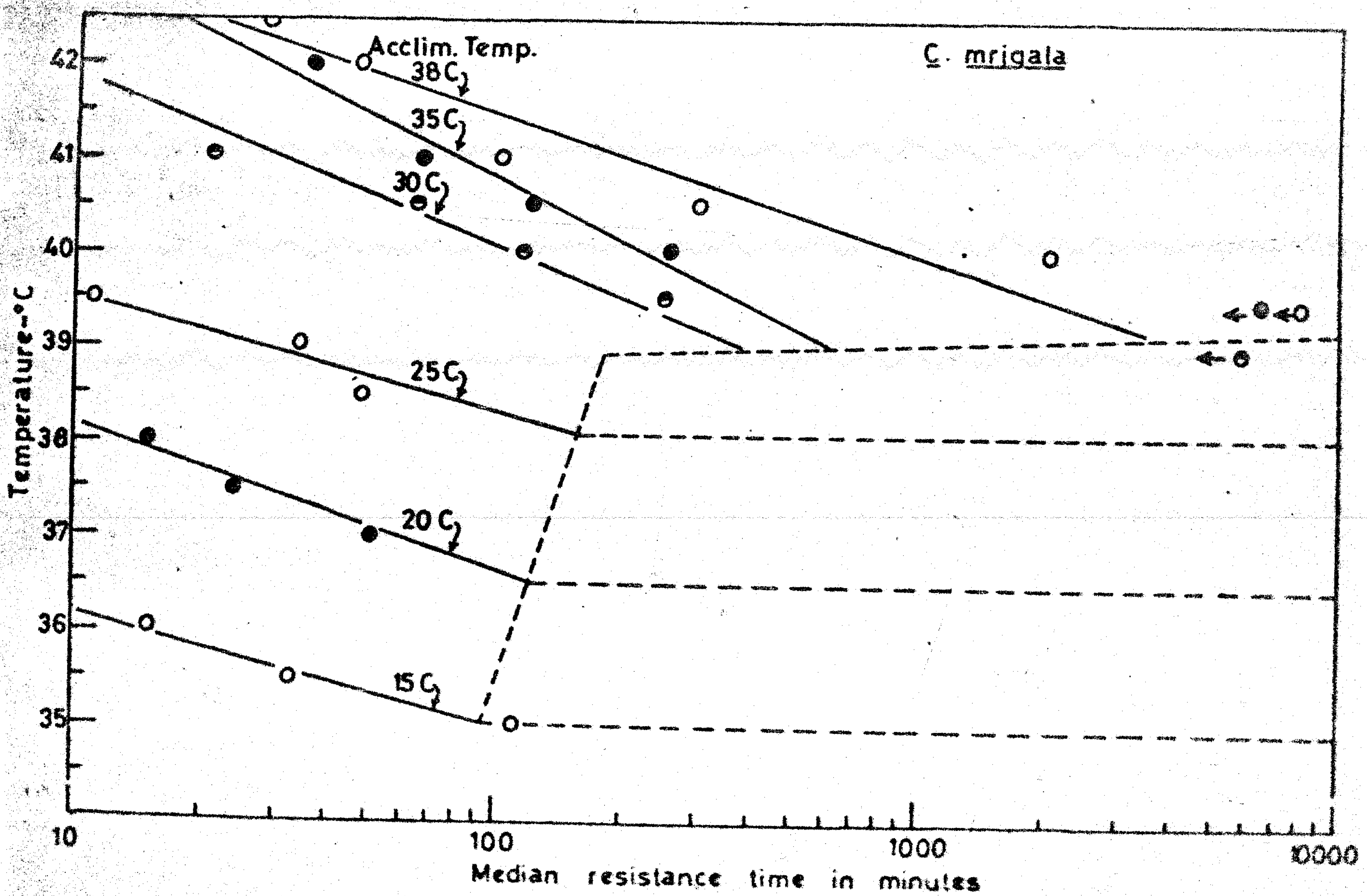

C. mrigala
Acclim. Temp.
38C
35C
30C
25C
20C
15C
0
2
4
6
8
10
Median resistance time – 1000 minutes

Fig.7

Median resistance times to high temperatures among mrigal fry acclimated to temperatures indicated. Plotted on arithmetic logarithmic axes (see text for further explanation)


Acclim. Temp.
38 C
35 C
30 C
25 C
20 C
15 C
C. mrigala
Temperature-°C
42
41
40
39
38
37
36
35
10
100
1000
10000
Median resistance time in minutes

experimented for any longer period of time. This boundary line differntiates the zone of tolerance from the zone of resistance. Further, the positive slope of the boundary line indicates the change in incipient lethal level with respect to increase in acclimation temperature from 15 to 25°C. Above 25°C there is no marked change in incipient lethal level, as indicated by the upper boundary line. The equations for the regression lines fitted in Fig.7 are presented in Table VI. The three points with arrows next to them (Fig.7) pertaining to 30, 35 and 38°C acclimations were not taken into account for the regression analysis so as to get a better fitting.

None of the fish sufferred death among the samples of 38°C acclimated group, when exposed to 39°C and below within 10,000 minutes exposure time. Whereas 100% mortality was recorded at 40°C and above (see appendix Aa1). Hence the incipient lethal temperature lies somewhere in between 39 and 40°C for 38°C acclimated group of mrigal. The value estimated as per the method of Miller and Tainter (1944) was 39.5°C. The incipient lethal temperatures thus calculated for all the acclimation temperatures of the five species presently studied are presented in Table VII. Upper incipient lethal temperatures for mrigal are 35.0, 38.3, 38.8 and 39.3 for acclimations 15, 20, 25, 30 and 35°C respectively.

Table VI

Formulae for the regression lines describing the thermal resistance of *C. mrigala* acclimated and tested to various temperatures. (x is the temperature in degree C; y is log time in minutes)

Acclimation Temp. (°C)	Upper	Lower
38.0	$y = 30.4009 - 0.6850\ x$	$y = -7.7035 + 0.5728\ x$
35.0	$y = 20.0362 - 0.4421\ x$	$y = -3.6248 + 0.3647\ x$
30.0	$y = 32.3620 - 0.7569\ x$	$y = -3.2519 + 0.3251\ x$
25.0	$y = 26.3780 - 0.6400\ x$	$y = -10.7861 + 1.0263\ x$
20.0	$y = 21.3526 - 0.5315\ x$	$y = -14.7500 + 1.6986\ x$
15.0	$y = 32.2968 - 0.8653\ x$	$y = -9.1884 + 1.3110\ x$

Table VII

Upper incipient lethal temperatures of fry of carps and fingerlings of freshwater mullet acclimated to various temperatures. Estimates have been made from the resistance times and from temperatures causing no mortality for exposures of 10,000 minutes, based on the raw data presented in appendices, by the method of Miller and Tainter (1944)

Acclimation Temp. (°C)	Upper lethal temperatures (°C) ± one SD				
	C. mrigala	*L. rohita*	*L. fimbriatus*	*C. carpio*	*R. corsula*
38	39.5 ± 0.3	39.0 ± 0.2	38.9 ± 0.2	-	-
35	39.3 ± 0.1	39.0 ± 0.3	38.8 ± 0.1	38.5 ± 0.3	36.5 ± 0.3
30	38.8 ± 0.1	38.5 ± 0.3	38.8 ± 0.1	37.6 ± 0.4	36.2 ± 0.4
25	38.3 ± 0.1	38.0 ± 0.3	38.1 ± 0.2	36.5 ± 0.3	36.0 ± 0.3
20	36.5 ± 0.3	35.8 ± 0.4	37.2 ± 0.4	35.3 ± 0.4	34.1 ± 0.4
15	35.0 ± 0.3	35.0 ± 0.3	35.0 ± 0.3	34.3 ± 0.4	32.4 ± 0.1

Fig. [redacted]

Tim death at different high lethaltest temperatur ng rohu fry acclimated to 20°C. Plotted on pr logarithmic axes

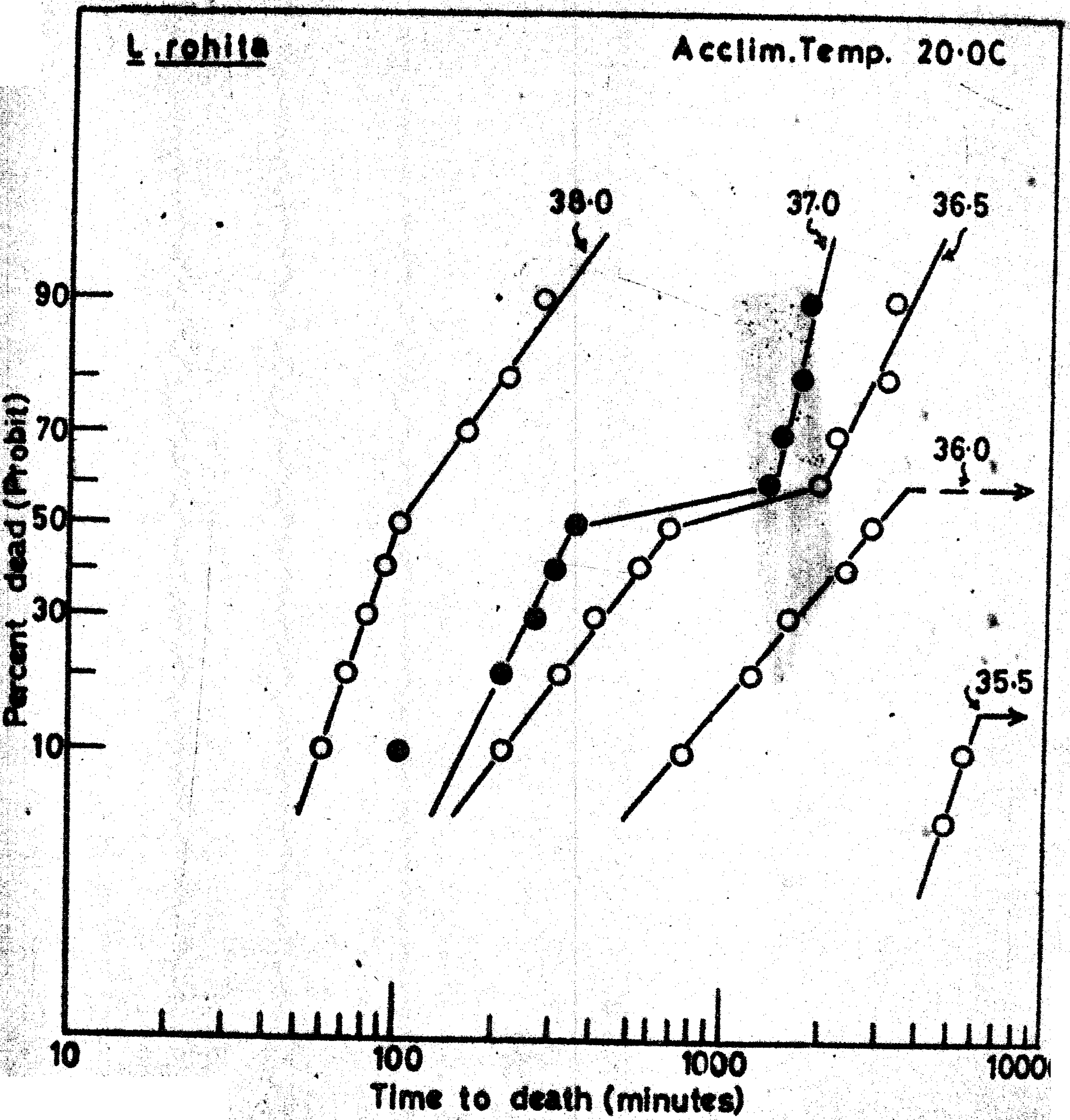

L. rohita
Acclim.Temp. 20·0C
38·0
37·0
36·5
36·0
35·5
90
70
50
30
10
Percent dead (Probit)
10
100
1000
1000
Time to death (minutes)

Fig.9

Median resistance times to high temperatures among rohu fry acclimated to temperatures indicated. Plotted on arithmetic x logarithmic axes

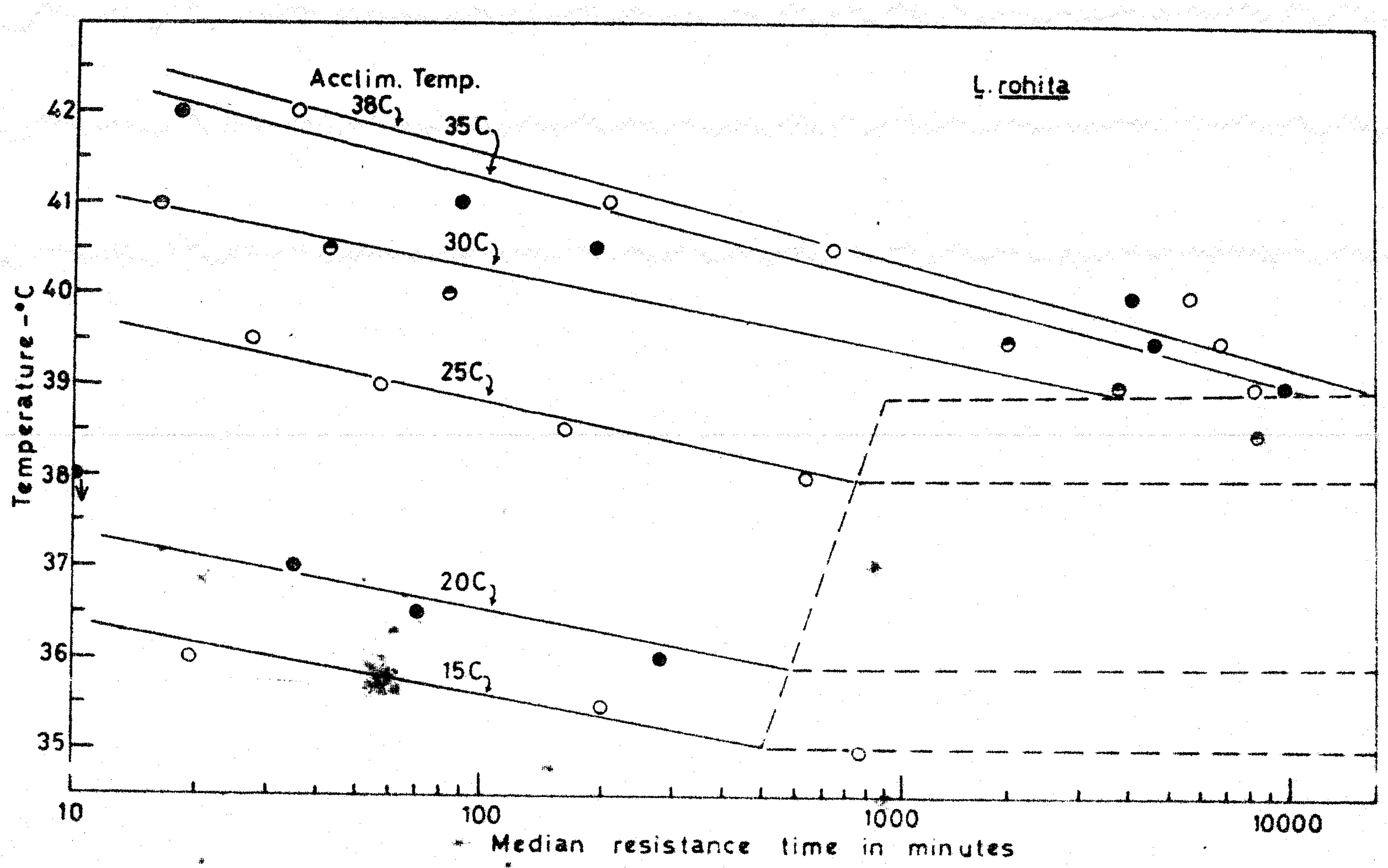

Acclim. Temp.
38C
35C
30C
25C
20C
15C
L. rohita
Temperature -°C
42
41
40
39
38
37
36
35
10
100
1000
10000
Median resistance time in minutes

Table VIII

Median resistance times to high lethal temperatures of rohu (Labeo rohita) fry acclimated to various temperatures

Acclimation Temp. (°C)	Lethal temperatures in °C and median resistance times in minutes													
	43	42.0	41.0	40.5	40.0	39.5	39.0	38.5	38.0	37.0	36.5	36.0	35.5	35.0
38	-	35	205	720	5500	6550	8000	-	-	-	-	-	-	-
35	5	18	88	190	3950	4500	9500	-	-	-	-	-	-	-
30	-	-	16	42	83	2000	3650	8100	-	-	-	-	-	-
25	-	-	-	-	-	27	56	160	630	-	-	-	-	-
20	-	-	-	-	-	-	-	-	10	35	70	280	-	-
15	-	-	-	-	-	-	-	-	-	-	-	19	200	860

Table IX

Formulae for the regression lines describing the thermal resistance of <u>L. rohita</u> acclimated and tested to various temperatures (x is the temperature in °C; y is the log time in minutes)

Acclimation (°C)	Upper	Lower
38.0	$y = 37.8767 - 0.8640\ x$	-
35.0	$y = 38.8168 - 0.8923\ x$	$y = -2.7487 + 0.3011\ x$
30.0	$y = 49.6543 - 1.1841\ x$	$y = -9.7023 + 0.8004\ x$
25.0	$y = 37.3826 - 0.9119\ x$	$y = -6.3330 + 0.7063\ x$
20.0	$y = 34.9087 - 0.9031\ x$	$y = -2.3524 + 0.4572\ x$
15.0	$y = 60.9488 - 1.6557\ x$	$y = -16.2558 + 2.2410\ x$

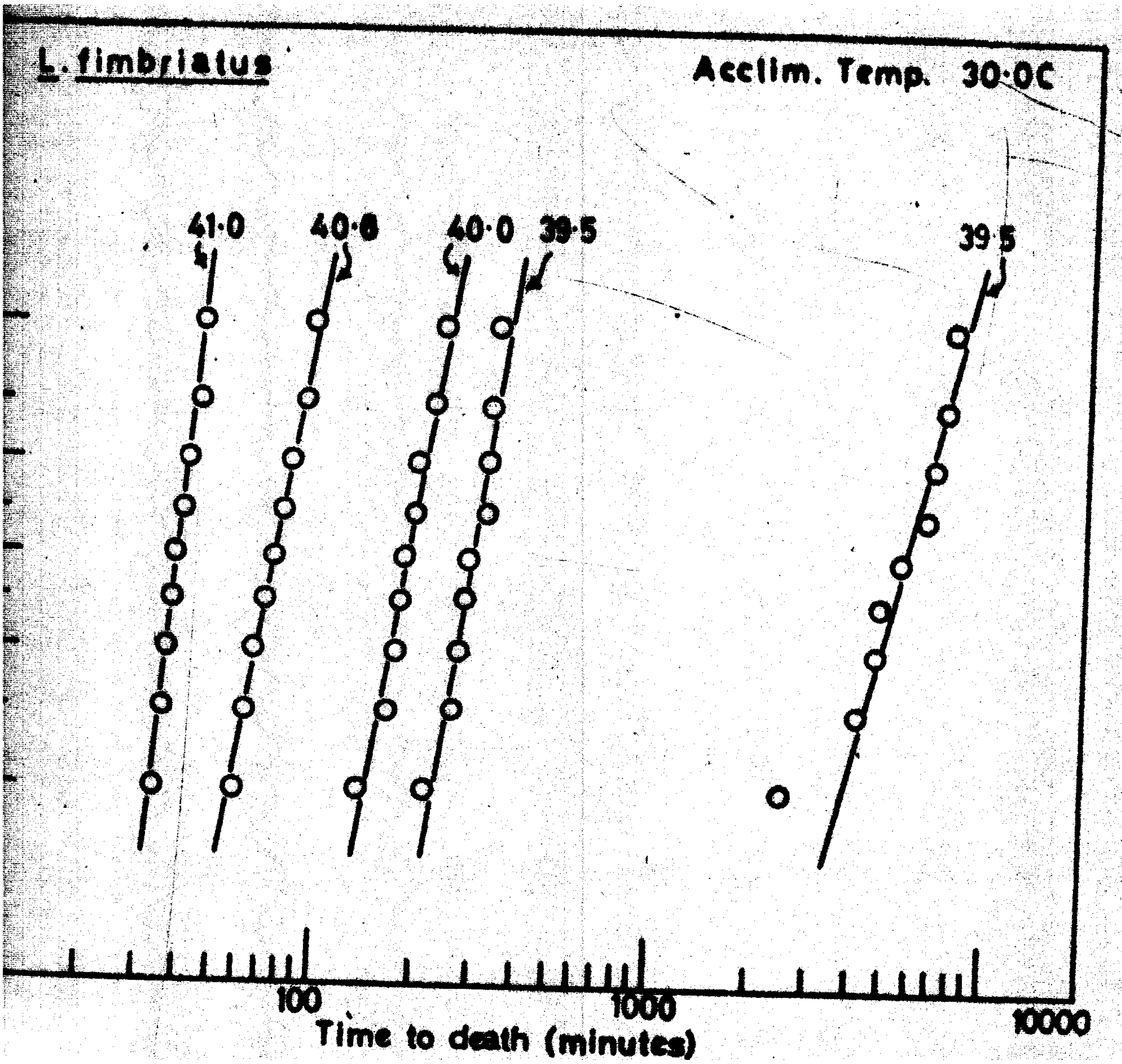
Mortality of rohu acclimated to 15, 20, 25, 30, 35 and 38°C was nil or partial (ie., below 50%) when tested at 34.5, 35.5, 37.0, 38.0 and 38.5°C respectively (see also appendices from Ab1 to Ab6). From these results it appears that the upper incipients lethal temperatures lie somewhere just above these test temperatures. Upper incipient lethal temperatures estimated, as in the case of mrigal, for this species are 35.0, 35.8, 38.0, 38.5, 39.0 and 39.0°C for the acclimations 15, 20, 25, 30, 35 and 38°C respectively (Table VII).

iii. Fringe-lipped carp:

As already shown for the previous two species, the raw data on time to death of individual fringe-lipped carp acclimated to six different temperatures (from 15 to 38°C) and tested to various high lethal temperatures, with other relevant details are presented in appendices from Ac1 to Ac6. As an example of the raw data obtained, the data of 30°C acclimated group are presented in Fig.10 on probit chart. The probit curves pertaining to all the high lethal temperatures displayed in Fig.10 are straight and simple. The mortality among the samples exposed to temperatures above 39.5°C was 100% within exposure time of 400 minutes; whereas at 39.0°C mortality began only after 2000 minutes

Fig.10

Times to death at different high lethaltest temperatures among fringe-lipped carp fry acclimated to 30°C. Plotted on probit x logarithmic axes


L. fimbriatus
Acclim. Temp. 30·0C
41·0
40·6
40·0
39·5
39·5
100
1000
10000
Time to death (minutes)

to reach 100% death among the sample. This long lapse of time in the mortality pattern within an interval of 0.5°C (i.e., between 39.0 and 39.5°C) in this species may suggest two factors acting on two different loci to bring about the mortality (see also 'mixed lethal effect' under 'Discussion').

The median resistance times obtained from various probability curves pertaining to the lethal temperatures of the six acclimation series are presented in Table X. The median resistance times summarised in Table X are plotted on a semi-logarithmic graph in Fig.11 and regression lines have been fitted through the plots for each acclimation level. The formulae for the regression lines describing the thermal resistance of fringe-lipped carp are given in Table XI. The general features of this figure are same as indicated for mrigal (see Fig.7).

The upper incipient lethal temperatures estimated from the raw data for this species are 35, 37.2, 38.1, 38.8, 38.8 and 38.9°C for the acclimation series 15, 20, 25, 30, 35 and 38°C respectively (Table VII). There is no marked change in incipient lethal temperatures above 30°C acclimation levels.

Table X

Median resistance times to high lethal temperatures of fringe-lipped carp (*Labeo fimbriatus*) fry acclimated to various temperatures

Acclimation Temp.(°C	Lethal temperatures in °C and median resistance times in minutes											
	42.0	41.0	40.5	40.0	39.5	39.0	38.5	38.0	37.5	36.0	35.5	35.0
33	46	290	1200	3800	5700	7200	-	-	-	-	-	-
35	27	78	290	3100	5300	6200	-	-	-	-	-	-
30	-	39	75	190	290	5650	-	-	-	-	-	-
25	-	-	-	14	42	120	275	-	-	-	-	-
20	-	-	-	-	-	-	8	17	27	-	-	-
15	-	-	-	-	-	-	-	-	-	8	22	44

Fig.11

Median resistance times to high temperatures among fringe-lipped carp fry acclimated to temperatures indicated. Plotted on arithmetic x logarithmic axes

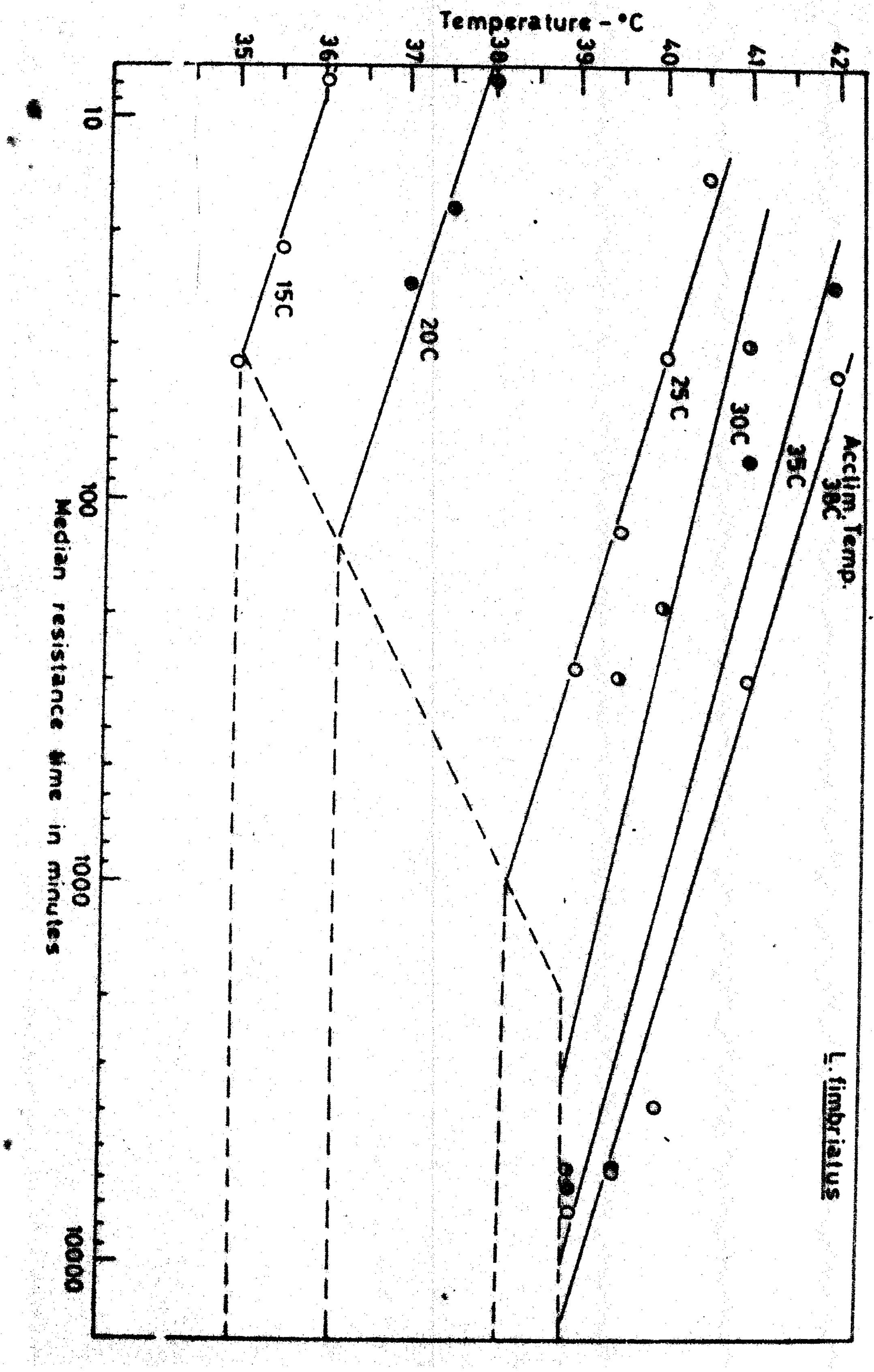

Temperature - °C
42
41
40
39
38
37
36
35
10
100
1000
10000
Median resistance time in minutes
15C
20C
25C
30C
35C
Acclim. Temp.
38C
L. fimbriatus

Table XI

Formulae for the regression lines describing the thermal resistance of *L. fimbriatus* acclimated and tested to various temperatures. (x is the temperature in °C; y is the log time in minutes)

Acclimation (°C)	Upper	Lower
38.0	$y = 34.4858 - 0.7790\,x$	$y = -3.7621 + 0.3382\,x$
35.0	$y = 39.2119 - 0.9028\,x$	$y = -7.0896 + 0.5632\,x$
30.0	$y = 41.6639 - 0.9818\,x$	$y = -24.2474 + 1.6655\,x$
25.0	$y = 35.8556 - 0.8671\,x$	$y = -19.6636 + 1.6138\,x$
20.0	$y = 21.2637 - 0.5283\,x$	$y = -15.6490 + 1.5544\,x$
15.0	$y = 27.5805 - 0.7404\,x$	$y = -6.2552 + 0.9253\,x$

vi. Common carp:

Times to death of individual common carp acclimated to 15, 20, 25, 30 and 35°C and tested at different high lethal temperatures ranging from 34.0 to 42.0°C are presented as raw data in appendices from Ad1 to Ad5, with other relevant details. The data of 25°C acclimated common carp are presented on probit chart in Fig.12, as an example of the raw data obtained. The probit curves pertaining to temperatures above 38°C are simple; whereas the curves of 37.0 and 37.5°C are split-probits as in the case of rohu (Fig.8). Unlike in the previous three species, the general shift of these probit curves towards the x axis is more in this species, indicating the low rate of mortification. As it is seen from Fig.12 the mortality was 100, 60 and 0% at 37.0, 36.5 and 36.0°C respectively.

Median resistance times obtained from a series of such probit curves as shown in Fig.12, for the five acclimation temperatures are summarised in Table XII. The semi-logarithmic plot of these median resistance times are shown in Fig.13 with respective regression lines for each acclimation temperature. In contrast to the previous three species the broken boundary line which cuts the lower ends of these regression lines indicates that incipient lethal temperature continues

Fig.12

Times to death at different high lethaltest temperatures among common carp fry acclimated to 25°C. Plotted on probit x logarithmic axes

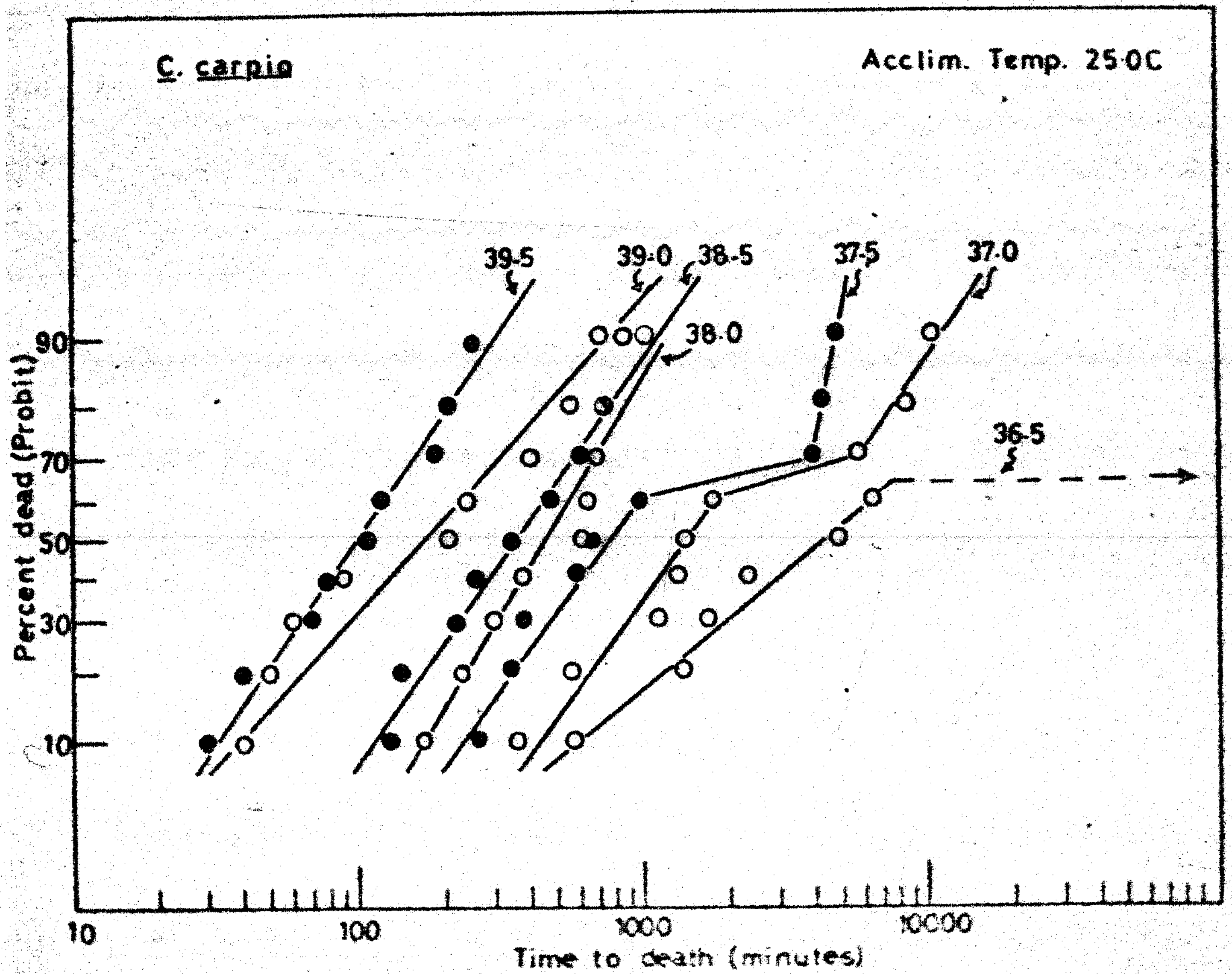

C. carpio
Acclim. Temp. 25·0C
Percent dead (Probit)
10
30
50
70
90
Time to death (minutes)
10
100
1000
10000
39·5
39·0
38·5
38·0
37·5
37·0
36·5

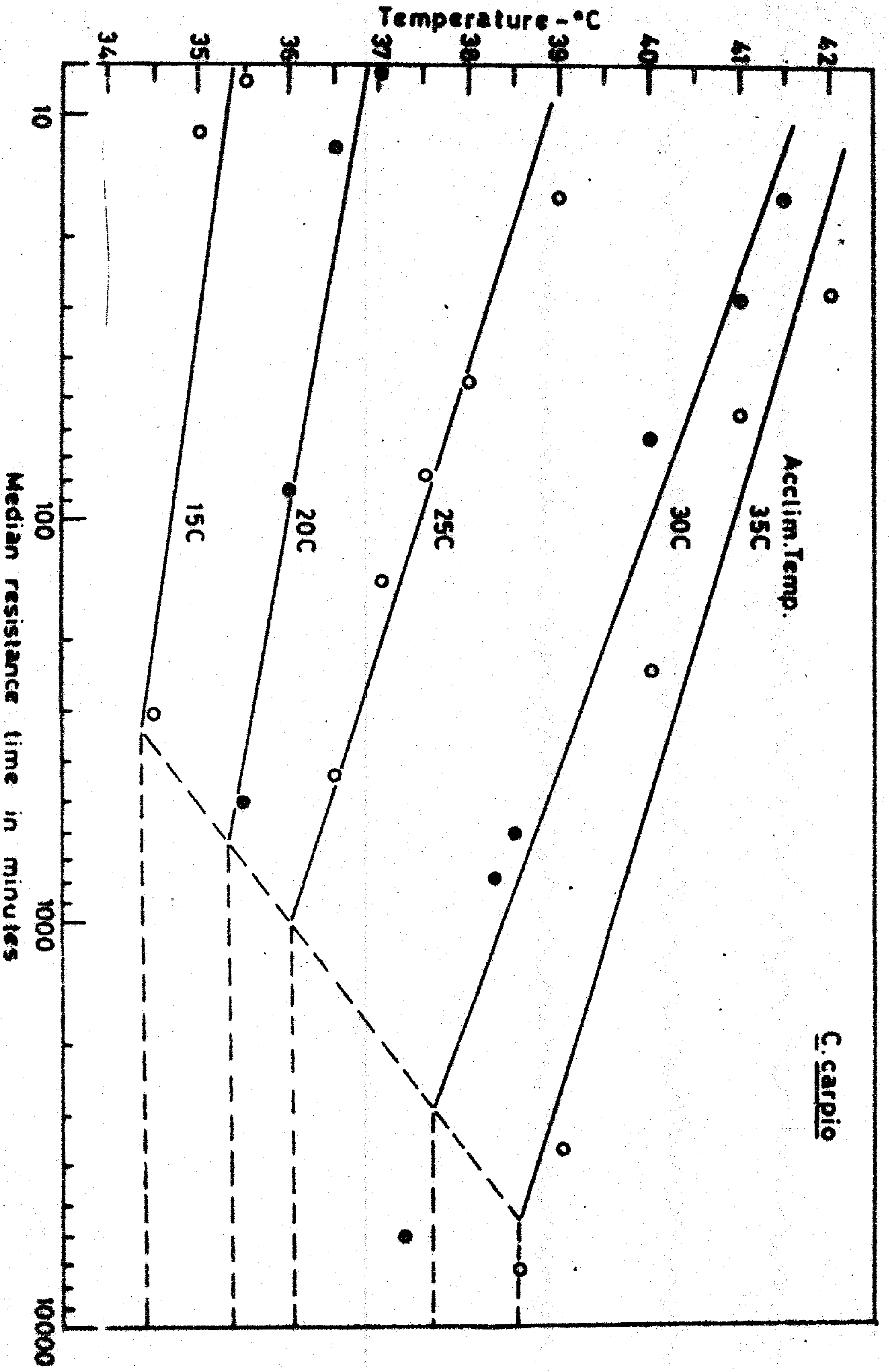
Table XII

Median resistance times to high lethal temperatures of common carp (*Cyprinus carpio* var *communis*) fry acclimated to various temperatures

Acclimation	Lethal temperatures in °C and median resistance times in minutes															
Temp.(°C)	42.0	41.5	41.0	40.5	40.0	39.5	39.0	38.5	38.0	37.5	37.0	36.5	36.0	35.5	35.0	34
35	28	-	56	92	240	2000	3625	7200	-	-	-	-	-	-	-	
30	-	16	29	52	64	350	610	780	6000	-	-	-	-	-	-	
25	-	-	-	-	-	10	16	35	46	78	142	430	-	-	-	
20	-	-	-	-	-	-	-	-	-	-	8	12	85	500	-	
15	-	-	-	-	-	-	-	-	-	-	-	-	-	8	11	30

Fig.13

Median resistance times to high temperatures among common carp fry acclimated to temperatures indicated. Plotted on arithmetic x logarithmic axes


C. carpio
Temperature - °C
42
41
40
39
38
37
36
35
34
Acclim.Temp.
35C
30C
25C
20C
15C
10
100
1000
10000
Median resistance time in minutes

to increase with the increase in acclimation temperature in this species. This phenomenon is unique to this species as in the case of goldfish (Fry _et al._, 1942) and absent in other four species presently studied. Table XIII constitutes the formulae for the regression lines describing the thermal resistance of this species.

From appendices Ad1 to Ad5 it can be noted that the mortality among the lots drawn from 15, 20, 25, 30 and 35°C acclimations and exposed to 34.0, 35.0, 36.0, 37.5 and 38.0°C respectively was partial and below 50%. Based on these raw data, the estimated upper incipient lethal temperatures are 34.3, 35.3, 36.5, 37.6 and 38.5°C for the acclimation 15, 20, 25, 30 and 35°C respectively (Table VII).

v. Freshwater mullet:

Appendices from Ae1 to Ae5 constitute the raw data obtained on times to death of individual freshwater mullet acclimated to five different temperatures (from 15 to 35°C) and exposed to various high lethal temperatures from 32 to 42°C, along with other relevant details. As an example of the results obtained on mullet, the data pertaining to 35°C acclimation are graphically shown in Fig.14 on a probability graph as done **for** other four species. Two features are evident from

Table XIII

Formulae for the regression lines describing the thermal resistance of *C. carpio* acclimated and tested to various temperatures (x is the temperature in °C; y is the log time in minutes)

Acclimation (°C)	Upper	Lower
35.0	$y = 33.4430 - 0.7695\ x$	$y = -0.7389 + 0.1609\ x$
30.0	$y = 29.8243 - 0.6931\ x$	$y = -4.5324 + 0.6528\ x$
25.0	$y = 21.1184 - 0.5103\ x$	$y = -1.4218 + 0.4934\ x$
20.0	$y = 46.8744 - 1.2475\ x$	$y = -4.5058 + 1.1800\ x$
15.0	$y = 56.8112 - 1.5810\ x$	–

Fig.14

Times to death at different high lethaltest temperatures among freshwater mullet fingerlings acclimated to 30°C. Plotted on probit x logarithmic axes

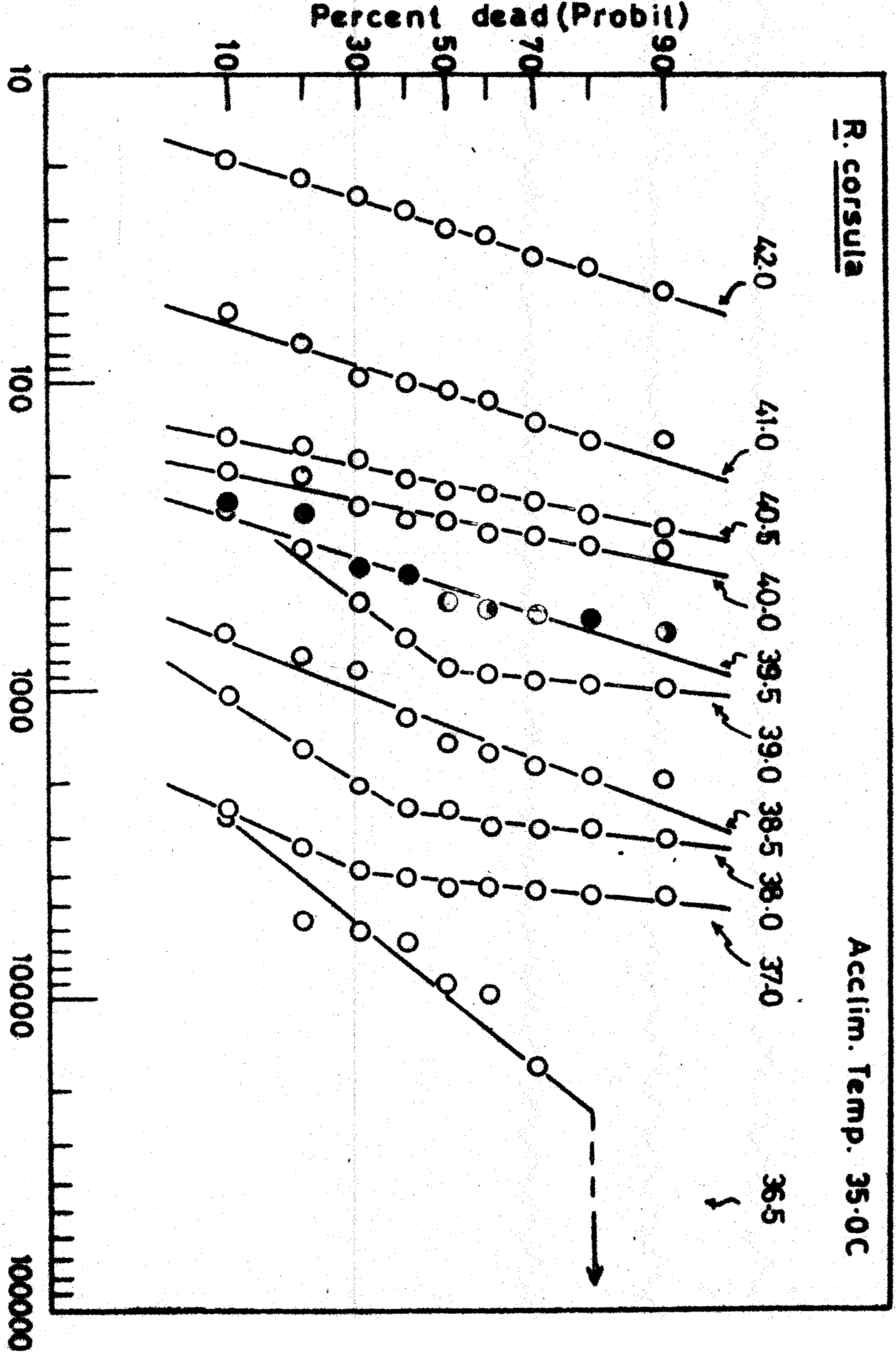

R. corsula
Acclim. Temp. 35·0C
Percent dead (Probit)
10
30
50
70
90
Time to death (minutes)
10
100
1000
10000
100000
42·0
41·0
40·5
40·0
39·5
39·0
38·5
38·0
37·0
36·5

Fig.14 i.e., the group of mullet acclimated to 35°C resisted a wide range of temperature from 36.5 to 42°C and mortality was present continuously even after one week time of exposure (approximately 10,000 minutes) at 36.5°C (see also 'Exposure time' under 'Discussion'). This phenomenon indicates that this species has relatively higher resistance to high lethal temperatures than the rest of four species in the present study.

Median resistance times estimated from the probit curves of the high lethal temperatures from 32 to 42°C for the five acclimations are summarised in Table XIV. The graphical illustration of these data from Table XIV, on a semilogarithmic grid is shown in Fig.15. The regression lines fitted through the plots for the five acclimation series indicate that the zone of resistance for this species is larger than that of other four species (see also 'Relative tolerance and resistance' under 'Discussion'). The formulae for the regression lines describing the resistance pattern of this species are given in Table XV.

The upper incipient lethal temperatures estimated, as in the case of previous four species, for this species are 32.4, 34.1, 36.0, 36.2 and 36.5°C for the acclimations 15, 20, 25, 30 and 35°C respectively (Table VII).

Table XIV

Median resistance times to high lethal temperatures of freshwater mullet (<u>Rhinomugil corsula</u>) fingerlings acclimated to various temperatures

Acclimation Temp. (°C)	Lethal temperatures in °C and median resistance times in minutes														
	42)	41.0	40.5	40.0	39.5	39.0	38.5	38.0	37.0	36.5	36.0	35.0	34.0	33.0	32.5
35	32	108	210	278	460	850	1330	2575	4150	10000	-	-	-	-	-
30	-	39	64	103	320	650	1550	2000	4750	8300	-	-	-	-	-
25	-	-	-	45	94	178	790	1575	2800	6600	8100	-	-	-	-
20	-	-	-	-	-	--	-	8	390	-	2000	3000	7000	-	-
15	-	-	-	-	-	-	-	-	-	-	150	600	2550	3750	66

Fig.15

Median resistance times to high temperatures among freshwater mullet fingerlings acclimated to temperatures indicated. Plotted on arithmetic x logarithmic axes

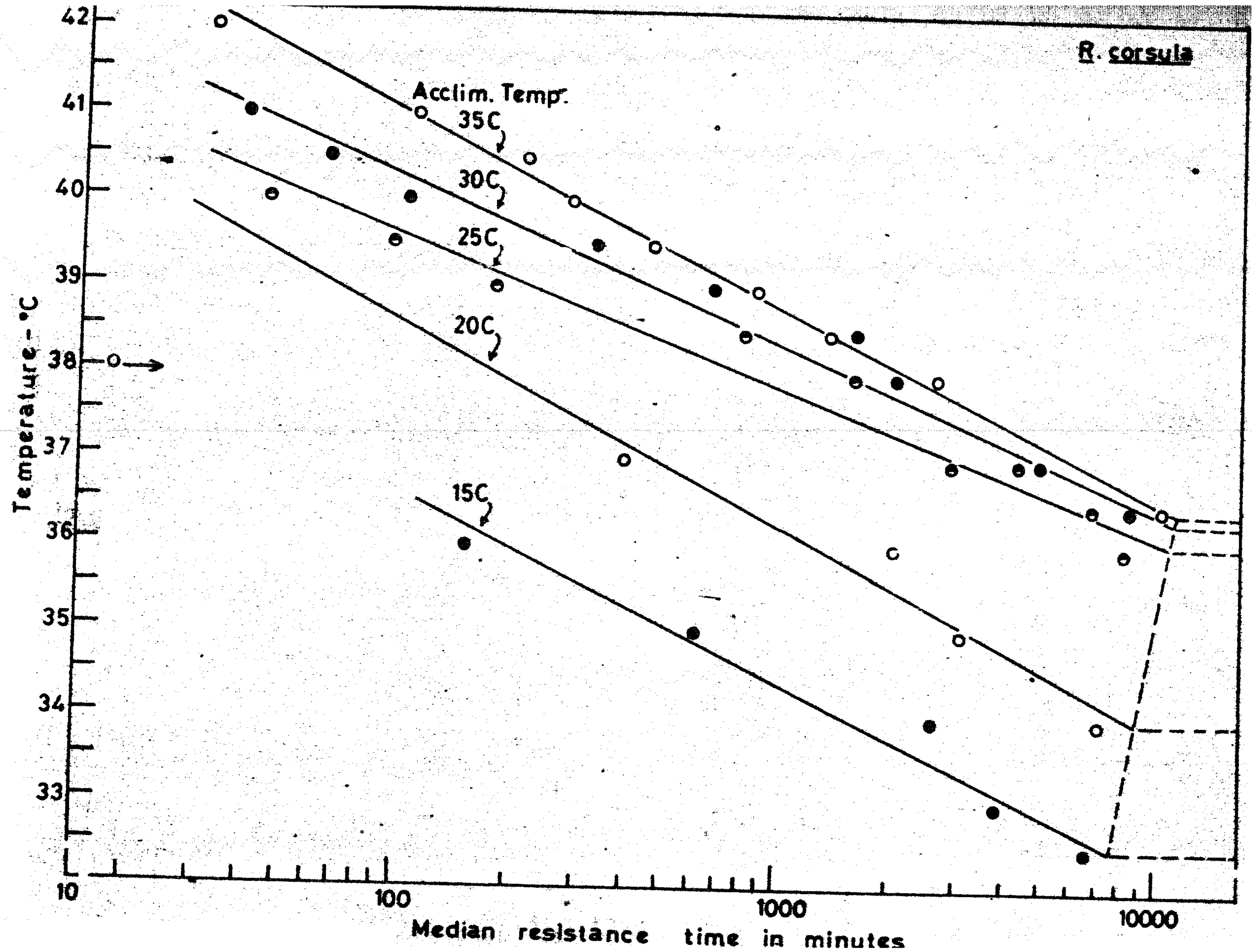

R. corsula
Acclim. Temp.
35C
30C
25C
20C
15C
Temperature - °C
42
41
40
39
38
37
36
35
34
33
10
100
1000
10000
Median resistance time in minutes

Table XV

Formulae for the regression lines describing the thermal resistance of R. corsula acclimated and tested to various temperatures (x is the temperature in °C; y is the log time in minutes)

Acclimation Temp. (°C)	Upper	Lower
35.0	$y = 19.8178 - 0.4340\ x$	$y = -0.0125 + 0.1804\ x$
30.0	$y = 23.6483 - 0.5372\ x$	$y = -1.4160 + 0.3280\ x$
25.0	$y = 24.8173 - 0.5760\ x$	$y = -0.9090 + 0.3434\ x$
20.0	$y = 17.2835 - 0.3938\ x$	$y = -1.5533 + 0.4614\ x$
15.0	$y = 18.7382 - 0.4571\ x$	$y = -0.3002 + 0.3854\ x$

Fig.16

Times to death at various low lethal test temperatures among mrigal fry acclimated to 38°C. Plotted on probit x logarithmic axes

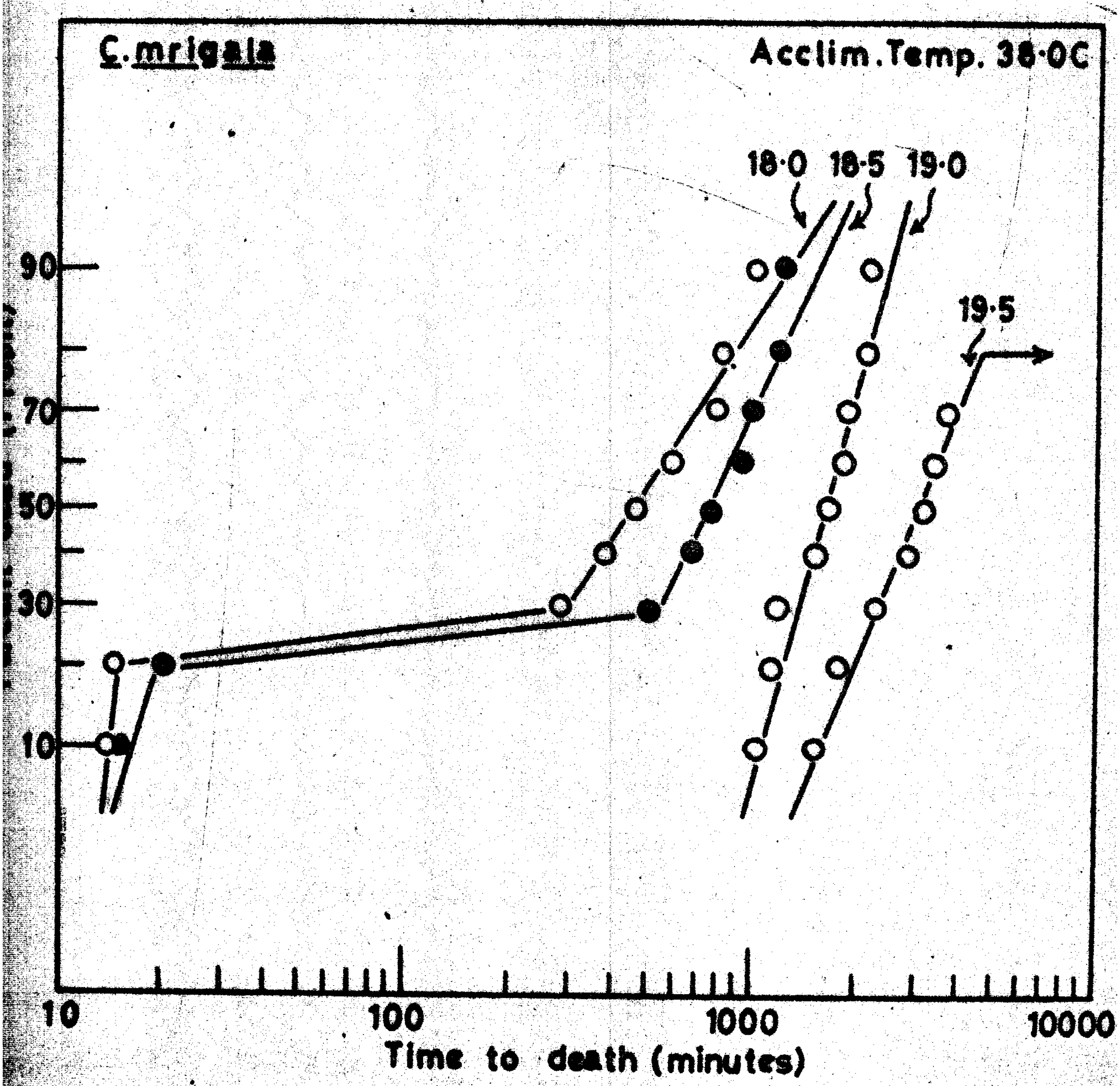

C. mrigala
Acclim. Temp. 38·0C
18·0
18·5
19·0
19·5
90
70
50
30
10
10
100
1000
10000
Time to death (minutes)

Fig.16) also differ to a considerable extent from median resistance times obtained from split probits (18.0 and 18.5°C). There is no difference among these two values for 19°C test which has a straight probit curve. However, in the present study only median resistance times have been taken into account for further analysis of the data for one important reason, that the median resistance times can be obtained for lethal levels where the mortality is partial i.e. 50% and above, but below 100% (Brett, 1952) (see also 'Methods').

The median resistance times, thus obtained by probit analysis for low lethal temperatures of the six acclimations for mrigal are summarised in Table XVI. It is seen from this Table that the median resistance times of fish exposed to low temperatures increase with rise in low lethal temperatures and decrease with increase in acclimation temperature. This phenomenon is reverse of that obtained on fish tested at high temperatures and is in agreement with observations on other species as well by earlier workers (Fry _et_ _al_., 1942, 1946; Hart, 1947; Brett, 1952).

The data on median resistance times from Table XVI are plotted on an arithmetic graph in Fig.17, as it is shown for high lethal temperatures for this species (Fig.6). The

Table XVI

Median resistance times to low lethal temperatures of mrigal,

Acclimation Temp(°C)	Lethal temperatures in °C and								
	8.0	8.5	9.0	9.5	10.0	10.5	11.0	12.5	13.0
38	-	-	-	-	-	-	-	-	-
35	-	-	-	-	-	-	-	-	-
30	-	-	-	-	-	-	-	-	-
25	-	-	-	-	-	-	-	125	300
20	-	-	-	10	790	1450	3800	-	-
15	18	110	370	-	-	-	-	-	-

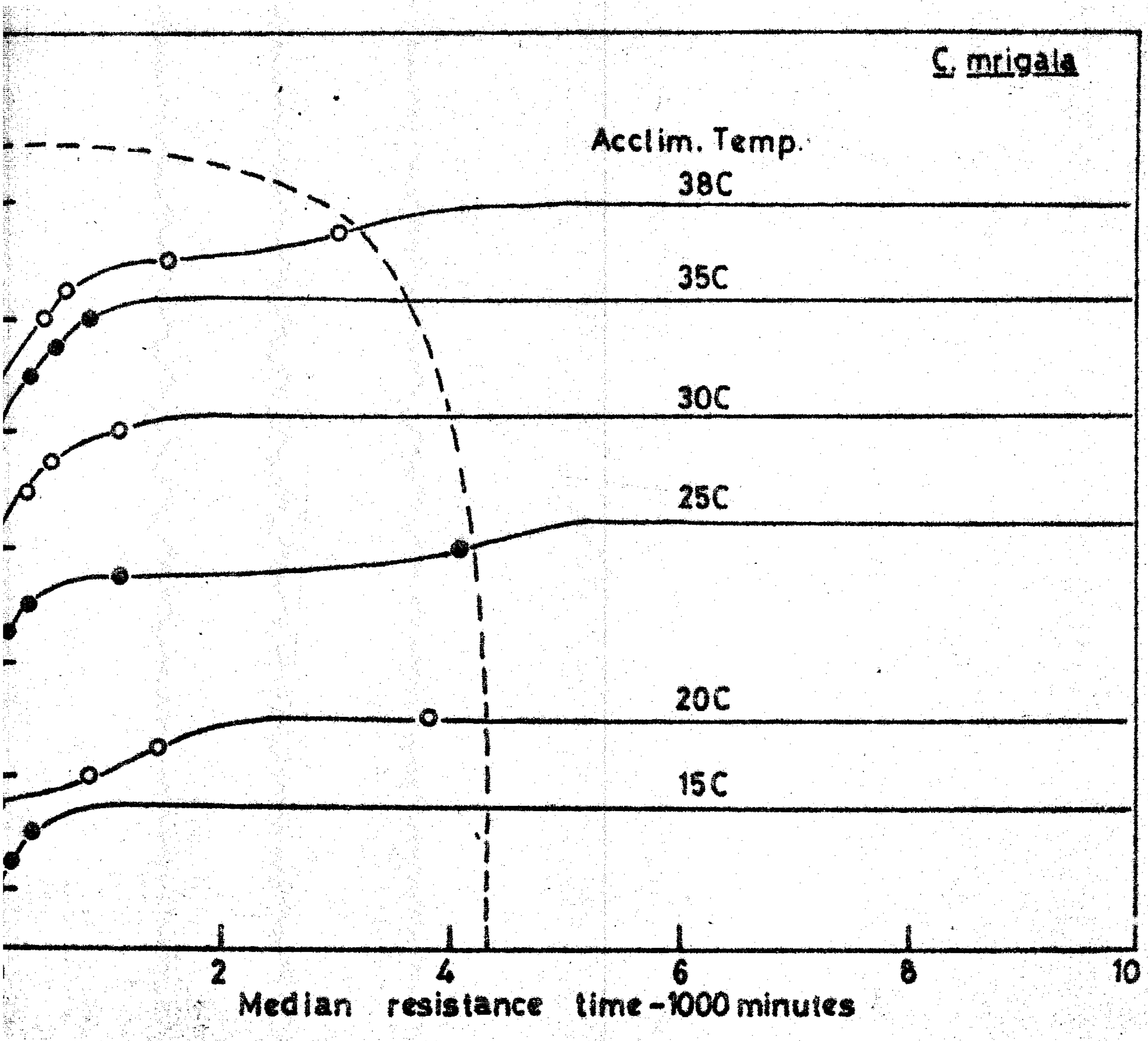
Table XVI

inus mrigala) fry acclimated to different temperatures

n resistance times in minutes									
14.0	15.0	15.5	16.0	17.0	17.5	18.0	18.5	19.0	19.5
-	-	-	-	-	-	450	660	1550	3050
-	-	-	-	380	560	880	-	-	-
-	330	520	1125	-	-	-	-	-	-
4100	-	-	-	-	-	-	-	-	-
-	-	-	-	-	-	-	-	-	-
-	-	-	-	-	-	-	-	-	-

Fig.17

Median resistance times to low temperatures among mrigal fry acclimated to temperatures indicated. Plotted on arithmetic axes (see also text for further explanation)


C. mrigala
Acclim. Temp.
38C
35C
30C
25C
20C
15C
2
4
6
8
10
Median resistance time-1000 minutes

lines drawn through all the points are not as smooth as they are in high lethal temperatures. The broken boundary line in this figure roughly divides the zones of tolerance and resistance. The same data are presented on a semi-logarithmic graph in Fig.18 and fitted with regression lines through the plots of respective acclimation series. The zones of tolerance and resistance are differentiated here by a negatively sloping broken boundary line, connecting dots. The formulae for the regression lines describing the thermal resistance at low temperatures are presented in Table VI for this species. The positive regression lines in Fig.18 are terminated at the points indicating respective incipient lethal temperatures as explained for high lethal tests (Fig.7).

The lower incipient lethal temperatures have been estimated, as it was done for high lethal temperature, by the method of Miller and Tainter (1944) and the values are presented in Table XVII. The lower incipient lethal temperatures for this species are 9.1, 11.1, 14.0, 16.4, 18.4 and 19.5°C for the acclimations 15, 20, 25, 30, 35 and 38°C respectively. These values increase with the increase in acclimation temperature. Hence, there is a reduction in low temperature tolerance with the increase in acclimation temperature.

Fig.18

Median resistance times to low temperatures among mrigal fry acclimated to temperatures indicated. Plotted on arithmetic x logarithmic axes (see text for further explanation)

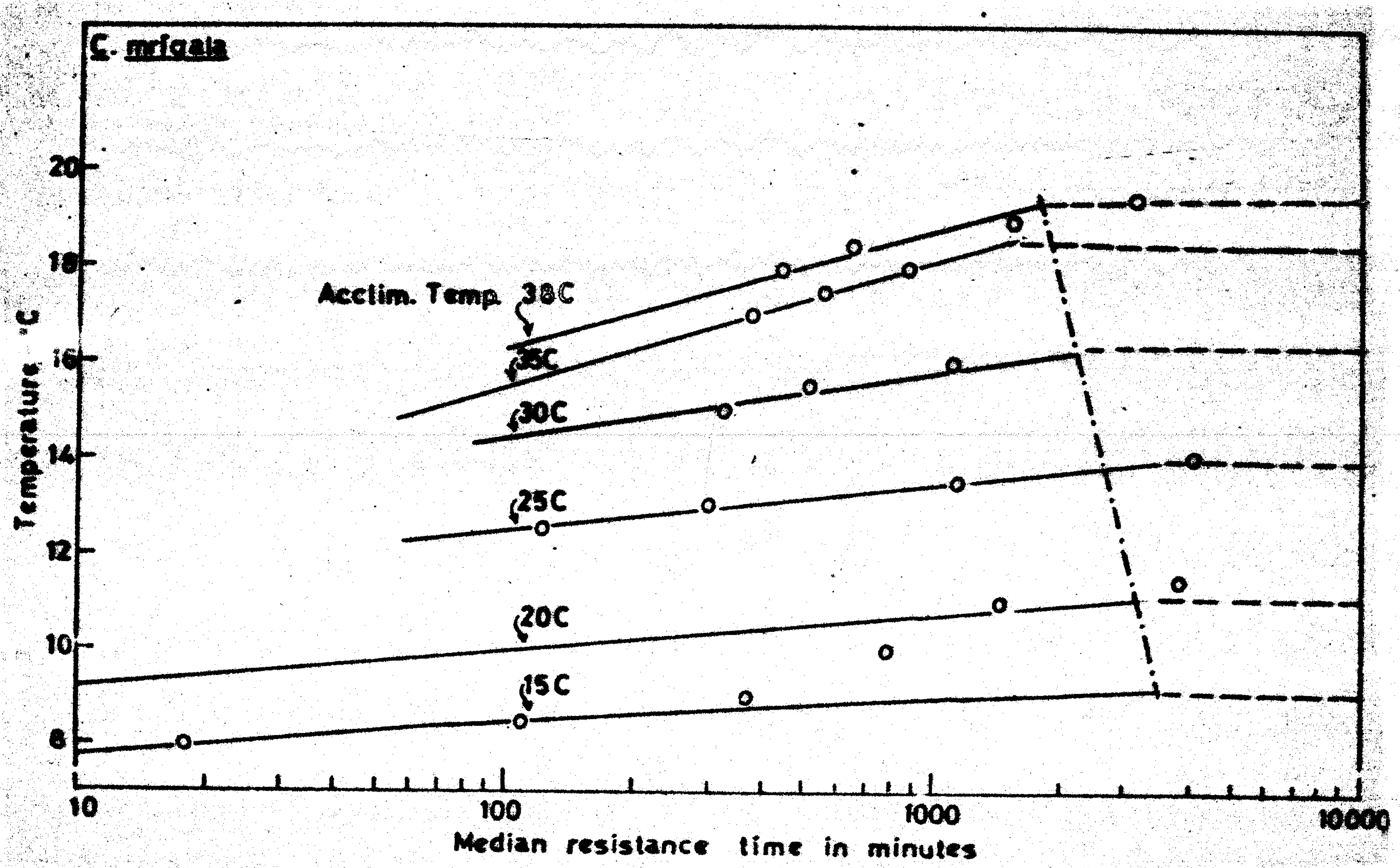

C. mrigala
Acclim. Temp. 38C
35C
30C
25C
20C
15C
Temperature °C
8
10
12
14
16
18
20
10
100
1000
10000
Median resistance time in minutes

Table XVII

Lower incipient lethal temperatures of young (fry) carps and fingerlings of freshwater mullet acclimated to various temperatures. Estimates have been made from the resistance times and from temperatures causing no mortality for exposures of 10,000 min., based on the raw data presented in appendices, by the method of Miller and Tainter (1944)

Acclimation Temp. (°C)	Lower lethal temperatures (°C)				
	C. mrigala	*L. rohita*	*L. fimbriatus*	*C. carpio*	*R. corsula*
38	19.5 ± 0.3	-	20.7 ± 0.3	-	-
35	18.4 ± 0.2	19.5 ± 0.3	18.6 ± 0.3	16.0 ± 0.5	19.5 ± 0.3
30	16.4 ± 0.2	16.6 ± 0.3	17.0 ± 0.3	12.7 ± 0.4	15.8 ± 0.5
25	14.0 ± 0.3	14.1 ± 0.3	14.8 ± 0.2	10.1 ± 0.3	13.2 ± 0.1
20	11.1 ± 0.3	11.3 ± 0.4	12.8 ± 0.1	7.0 ± 0.3	11.5 ± 0.3
15	9.1 ± 0.2	9.1 ± 0.3	10.6 ± 0.2	-	10.5 ± 0.3

This phenomenon is uniformly present in all the species presently studied and has also been reported for other species by earlier workers (Fry _et al._, 1942, 1946; Hart, 1947; Doudoroff, 1945; Brett, 1952).

ii. Rohu:

Times to death of rohu acclimated to five different temperatures (from 15 to 35°C) and exposed to various low lethal temperatures ranging from 8.0to 19.5°C are given in appendices from Bb1 to Bb5 along with other relevant details. As an example of the raw data obtained, the data from appendix Bb4 pertaining to 20°C acclimated group of rohu are presented on probit chart in Fig.19. The probit curves of all the low lethal temperatures are split-probits.

The data on median resistance times obtained from low lethal temperatures of the five acclimations, by probability analysis are presented in Table XVIII. These data (from Table XVIII) are plotted on an arithmetic graph in Fig.20 and the lines drawn connecting all the points show the complex low temperature response of this species as well. Fig.21 contains the semilogarithmic plots of the same data from Fig.20 and the regression lines fitted through the plots of each acclimation series also describe the low temperature response

Fig.19

Times to death at various low lethaltest temperatures among rohu fry acclimated to 20°C. Plotted on probit x logarithmic axes

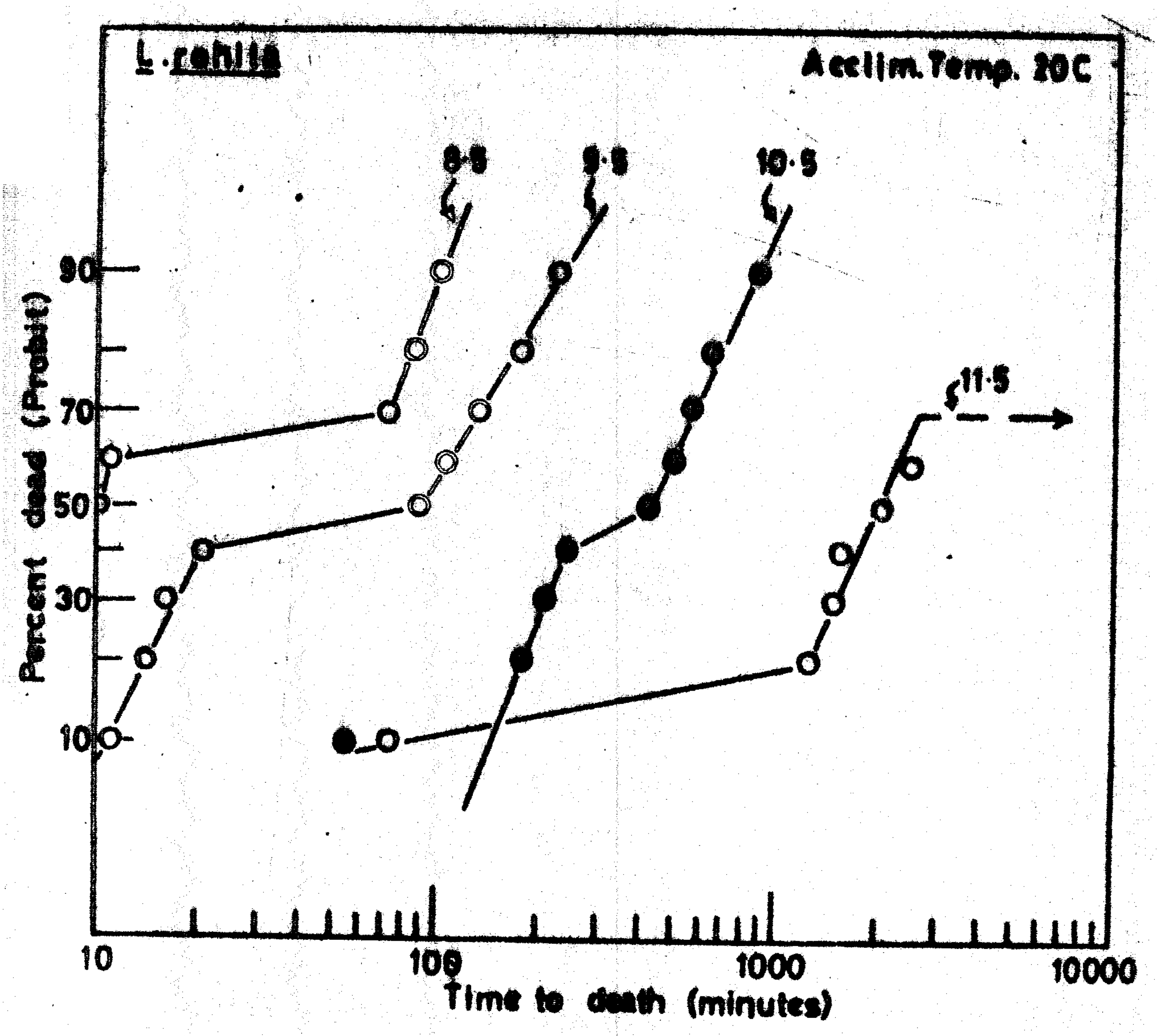

L. rohita
Acclim. Temp. 20C
8·5
9·5
10·5
11·5
90
70
50
30
10
Percent dead (Probit)
10
100
1000
10000
Time to death (minutes)

Table XVIII

Median resistance times to low lethal temperatures of Rohu (*Labeo rohita*) fry acclimated to different temperatures

Acclimation Temp. (°C)	Lethal temperatures in °C and median resistance times in minutes															
	8.0	8.5	9.0	9.5	10.5	11.5	12.5	13.0	13.5	14.0	15.5	16.0	16.5	17.0	18.0	19
35	-	-	-	-	-	-	-	-	-	-	-	-	-	250	410	10
30	-	-	-	-	-	-	-	-	-	-	570	1000	3600	-	-	
25	-	-	-	-	-	-	420	630	1550	3100	-	-	-	-	-	
20	-	10	-	90	420	2000	-	-	-	-	-	-	-	-	-	
15	30	190	4800	-	-	-	-	-	-	-	-	-	-	-	-	

Fig.20

Median resistance times to low temperatures among rohu fry acclimated to temperatures indicated. Plotted on arithmetic axes

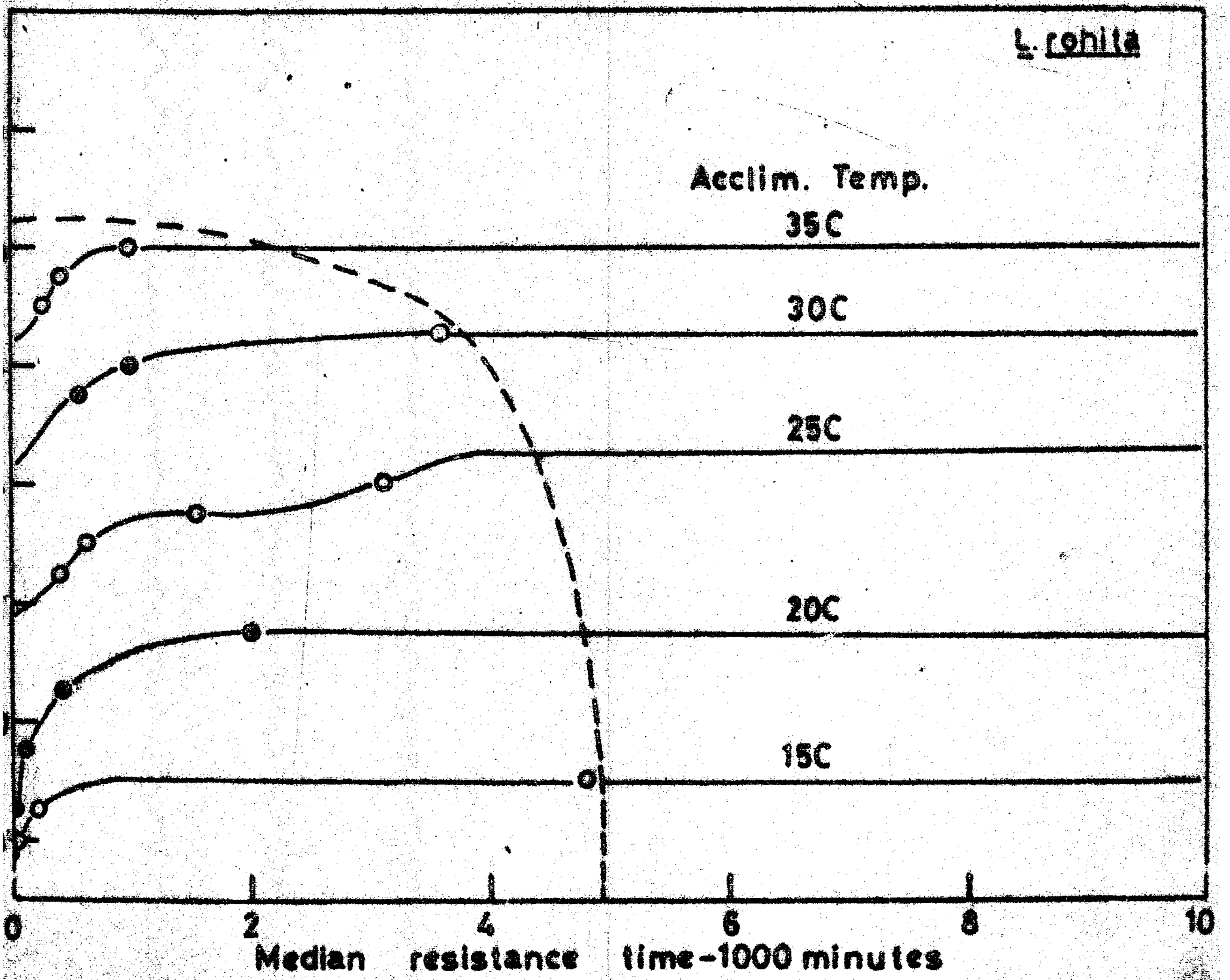
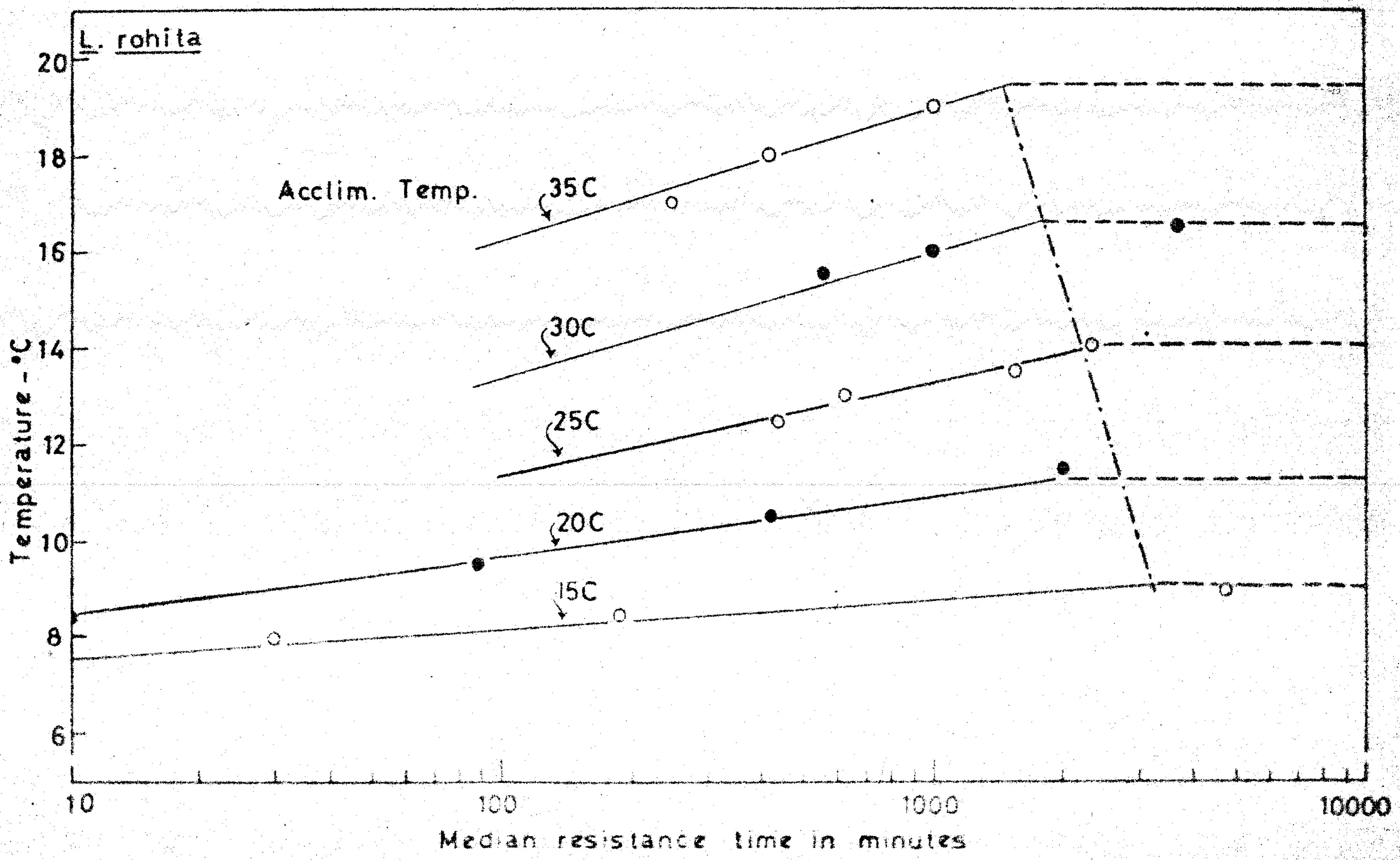

L. rohita
Acclim. Temp.
35C
30C
25C
20C
15C
0
2
4
6
8
10
Median resistance time-1000 minutes

Fig.21

Median resistance times to low temperatures among rohu fry acclimated to temperatures indicated. Plotted on arithmetic x logarithmic axes


L. rohita
Acclim. Temp.
35C
30C
25C
20C
15C
Temperature - °C
20
18
16
14
12
10
8
6
10
100
1000
10000
Median resistance time in minutes

of this species. The equations for these regression lines are presented in Table IX.

The lower incipient lethal temperatures estimated from the raw data for the five acclimation series are given in Table XVII for this species and the values are 9.1, 11.3, 14.1, 16.6 and 19.5°C for the acclimations 15, 20, 25, 30 and 35°C respectively.

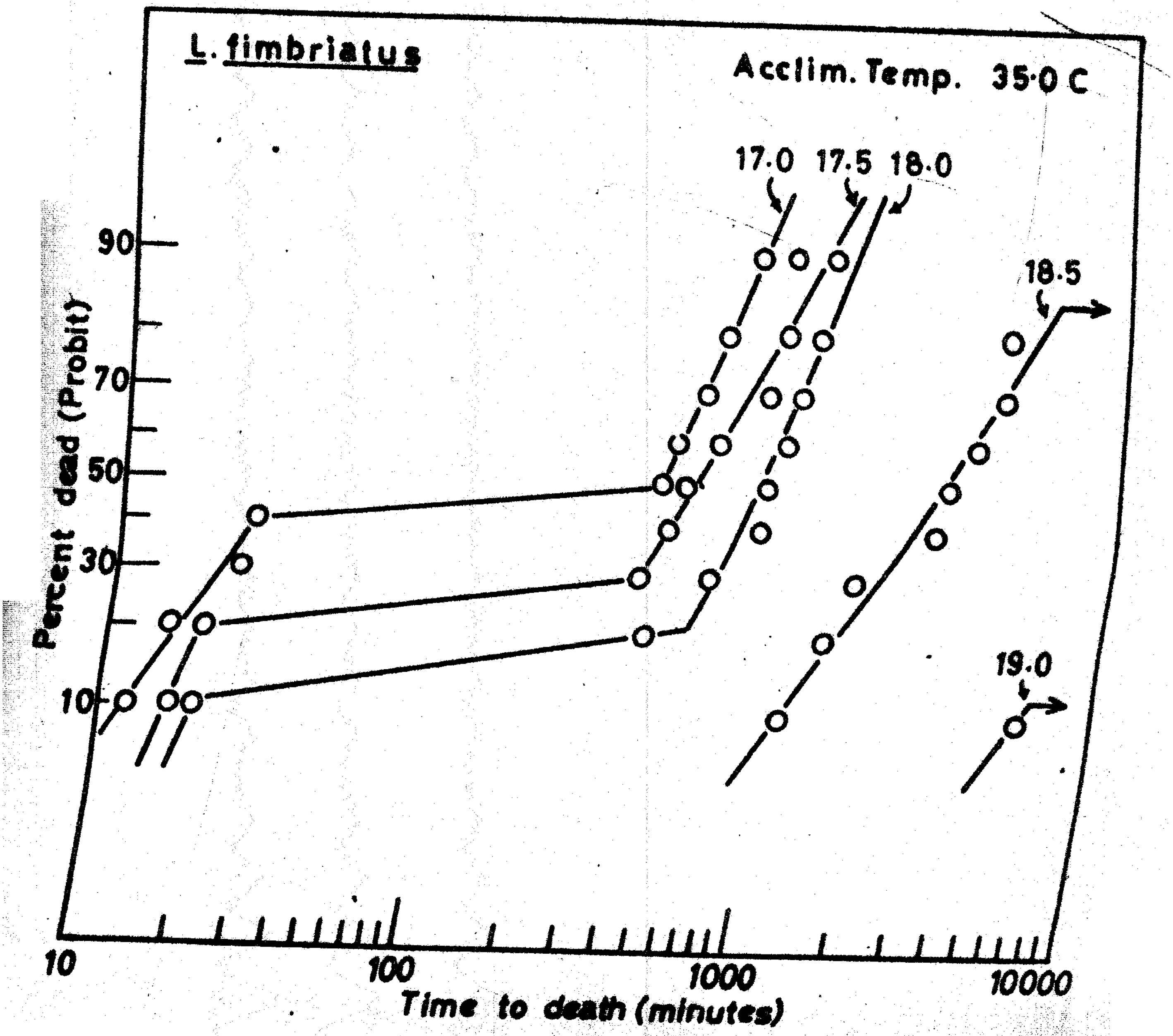
iii. <u>Fringe-lipped carp</u>:

Appendices from Bc1 to Bc6 contain the raw data on times to death of fringe-lipped carp acclimated to 15, 20, 25, 30, 35 and 38°C and exposed in lots to various low lethal temperatures ranging from 9.5 to 21.5°C with other relevant details. As an example of this data, the time to death of fringe-lipped carp acclimated to 35°C (Appendix Bc2) and exposed to 17.0, 17.5, 18.0, 18.5 and 19.0°C in lots are shown in Fig.22 on a probability graph. Except 18.5°C, the probit curves of all other test temperatures are split- probits.

Median resistance times estimated from the lots exposed to various low lethal temperatures from the six different acclimations, by probability analysis are summarised in Table **XIX**. The low temperature resistance features of these median

Fig.21

Median resistance times to low temperatures among rohu fry acclimated to temperatures indicated. Plotted on arithmetic x logarithmic axes



L. fimbriatus
Acclim. Temp. 35·0 C
17.0
17.5
18.0
18.5
19.0
90
70
50
30
10
Percent dead (Probit)
10
100
1000
10000
Time to death (minutes)

Table XIX

Median resistance times to low lethal temperatures of fringe

temperatures

Acclimation Temp(°C)	Lethal temperatures in °C and									
	9.5	10.0	10.5	11.0	11.5	12.0	12.5	13.0	13.5	14.
38	-	-	-	-	-	-	-	-	-	-
35	-	-	-	-	-	-	-	-	-	-
30	-	-	-	-	-	-	-	-	-	-
25	-	-	-	-	-	-	-	11	410	69
20	-	-	-	13	600	1180	3150	-	-	-
15	380	810	3200	-	-	-	-	-	-	-

Table XIX

arp, (*Labeo fimbriatus*) fry acclimated to different

resistance times in minutes								
15.5	16.0	16.5	17.0	17.5	18.0	18.5	19.5	21.0
-	-	-	-	165	-	270	620	2400
-	-	-	380	510	780	2800	-	-
15	1300	1900	5000	-	-	-	-	-
-	-	-	-	-	-	-	-	-
-	-	-	-	-	-	-	-	-
-	-	-	-	-	-	-	-	-

resistance times are the same as explained for mrigal and rohu. The data on median resistance times are graphically presented in Fig.23 on arithmetic axes and also in Fig.24 on a semilograthmic graph as already shown for mrigal and rohu. The lines drawn through the plots for each acclimation series describe the low temperature resistance of this species. The broken boundary line in these figures divede the zones of tolerance and resistance. The formulae for the regression lines fitted in Fig.24 are given in TableXI.

The lower incipient lethal temperatures estimated from the raw data (Appendices Bc1 to Bc6) for this species are given in Table XVII and the values are 10.6, 12.8, 14.8, 17.0, 18.6 and 20.7°C for the acclimations 15, 20, 25, 30, 35 and 38°C respectively.

iv. Common carp:

The raw data on time to death of common carp acclimated to 20, 25, 30 and 35°C and tested to various low lethal temperatures ranging from 6.0 to 16.5°C are presented in appendices from Bd1 to Bd4 with other relevant details. As an example, the data of 30°C acclimated common carp are shown on probit chart in Fig.25. The mortality is 100% at 11.5 and 12.0°C with split-probit curves; whereas 60 and 30% at 12.5 and 13.0°C respectively. The median resistance

Fig.23

Median resistance times to low temperatures among fringe-lipped carp fry acclimated to temperatures indicated. Plotted on arithmetic axes

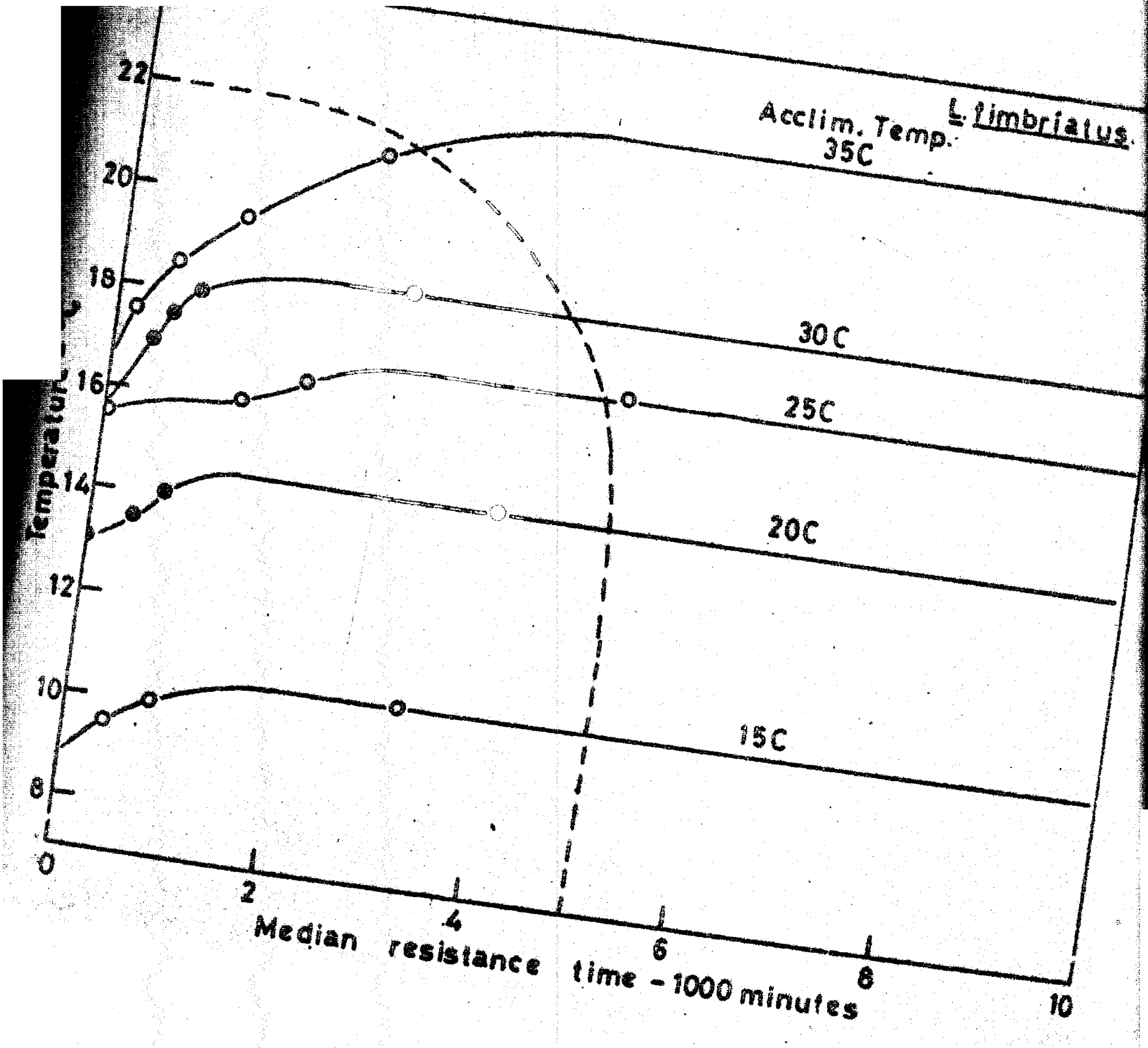

L. fimbriatus.
Acclim. Temp.
35C
30C
25C
20C
15C
Temperature C
22
20
18
16
14
12
10
8
0
2
4
6
8
10
Median resistance time – 1000 minutes

Fig.24

Median resistance times to low temperatures among fringe-lipped carp fry acclimated to temperatures indicated. Plotted on arithmetic x logarithmic axes

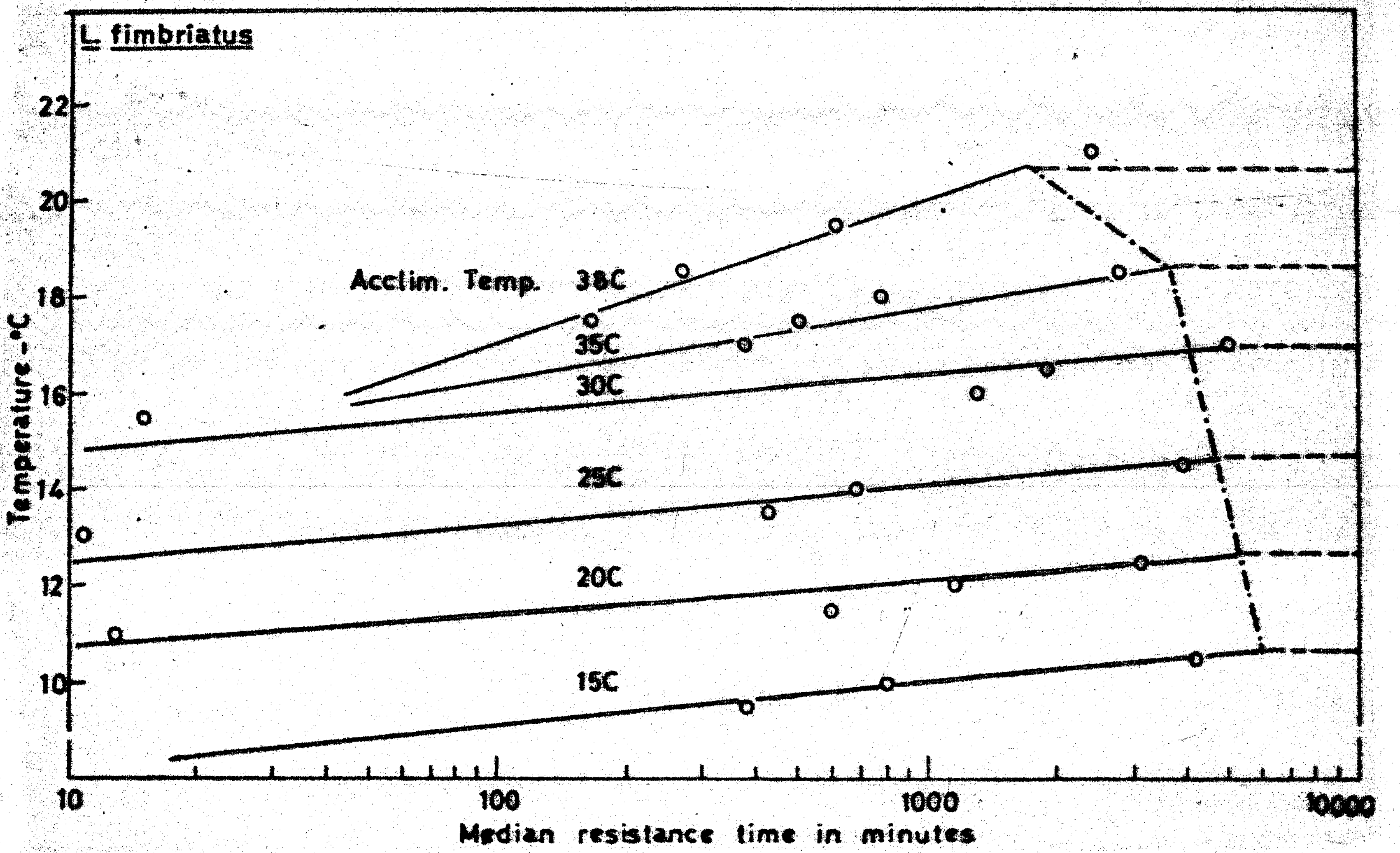
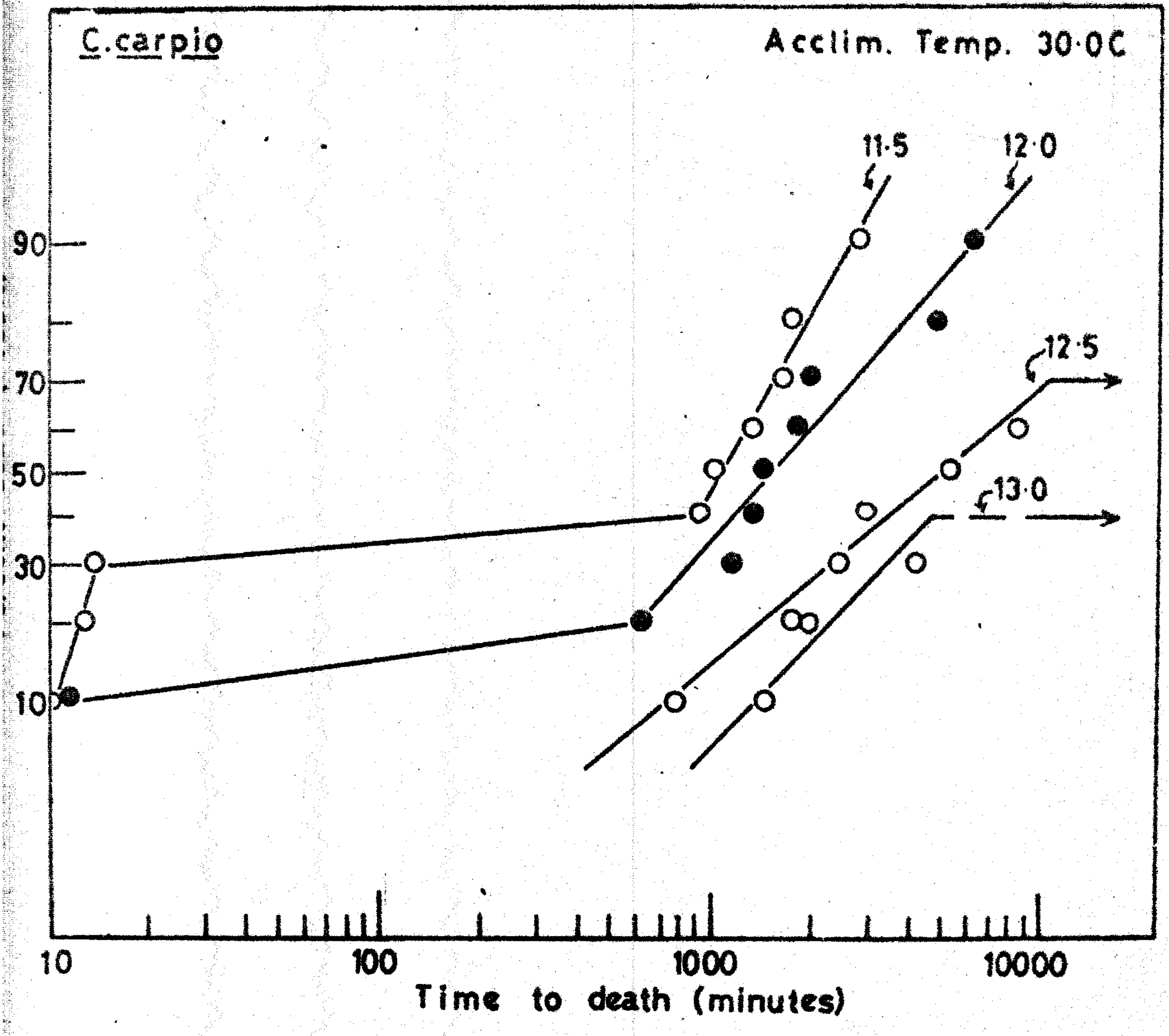

L. fimbriatus
Acclim. Temp.
38C
35C
30C
25C
20C
15C
Temperature -°C
22
20
18
16
14
12
10
10
100
1000
10000
Median resistance time in minutes

Fig.25

Times to death at various low lethaltest temperatures among common carp fry acclimated to 30°C. Plotted on probit x logarithmic axes


C.carpio
Acclim. Temp. 30·0C
11·5
12·0
12·5
13·0
90
70
50
30
10
10
100
1000
10000
Time to death (minutes)

times obtained by probability analysis, as already done for other species, are summarised in Table XX. These data indicate that the thermal tolerance of this species to low lethal temperature is higher than the other four species presently studied; for this species resists the low lethal temperature over 370 minutes at 6°C (Table XX) when the sample is exposed from 20°C acclimated group. Though this species could be acclimated to temperatures, below 20°C, experiments could not be performed at temperatures lower than 6°C due to the difficulty in maintaining lower temperatures for a longer duration of time under the limitation of the local technique (Kasim et al., 1977) (see also 'Methods'), but the available data suggest that this limitation has not affected the results severely (see also 'Discussion').

Further, the data on median resistance times from Table XX are presented graphically in Fig.26 on arithmetic graph and in Fig.27 on semilogarithmic graph. As it is seen from these figures the general features are found to be same as in the case of previous species. The regression lines fitted through the plots describe the lower thermal resistance of this species and the broken boundary line differentiates the zones of thermal tolerance and resistance at the

Table X X

Median resistance times to low lethal temperatures of common carp (*Cyprinus carpio*) fry acclimated to different temperatures

Acclimation Temp. (°C)	Lethal temperatures in °C and median resistance times in minutes														
	6.0	6.5	7.0	8.5	9.0	9.5	10.0	11.5	12.0	12.5	13.0	13.5	14.0	15.0	16.0
35	-	-	-	-	-	-	-	-	-	530	700	830	1000	1280	1950
30	-	-	-	-	-	-	-	1090	1500	4900	-	-	-	-	-
25	-	-	-	560	1075	2050	3000	-	-	-	-	-	-	-	-
20	370	1500	5600	-	-	-	-	-	-	-	-	-	-	-	-

Fig.26

Median resistance times to low temperatures among common carp fry acclimated to temperatures indicated. Plotted on arithmetic axes

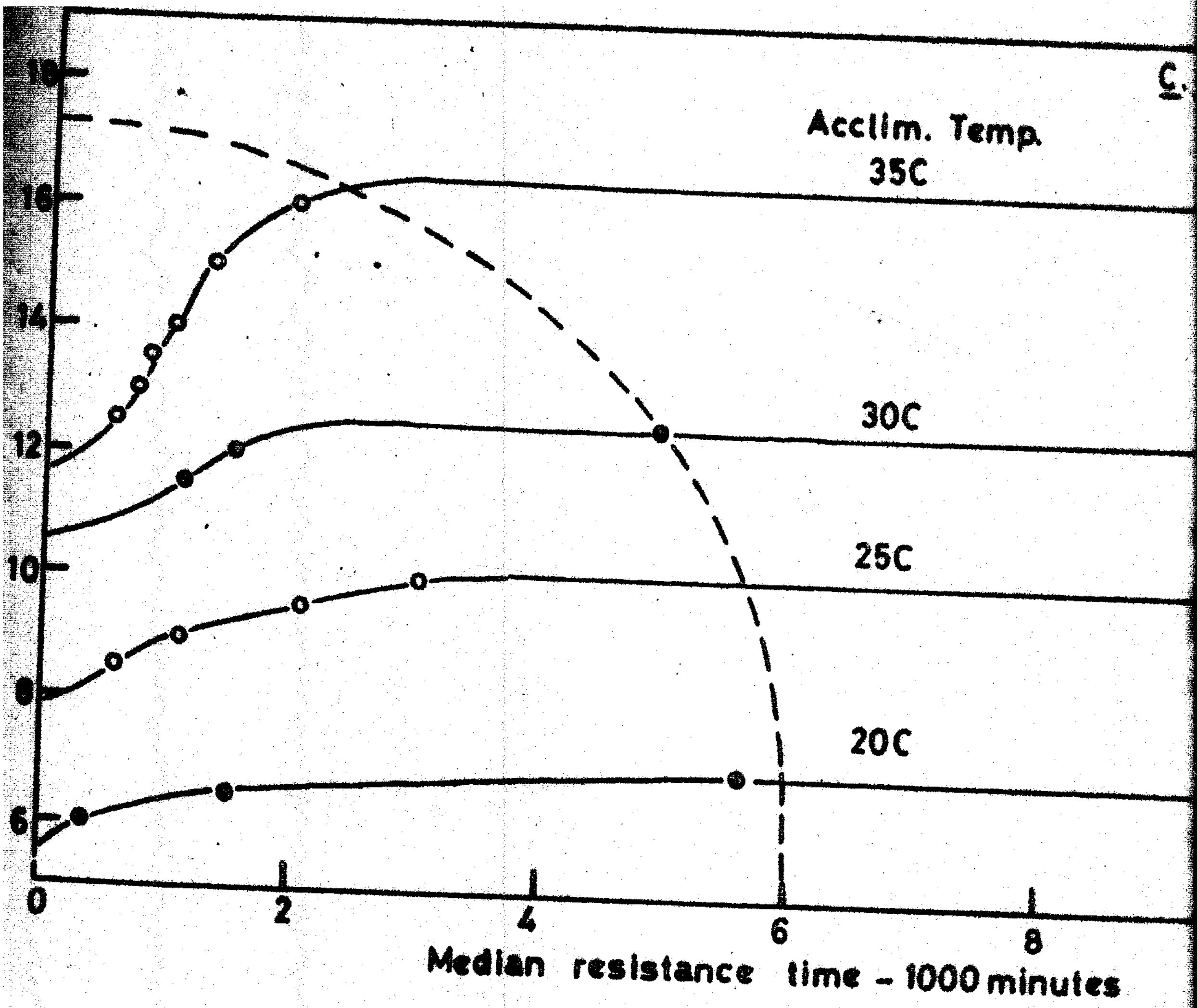
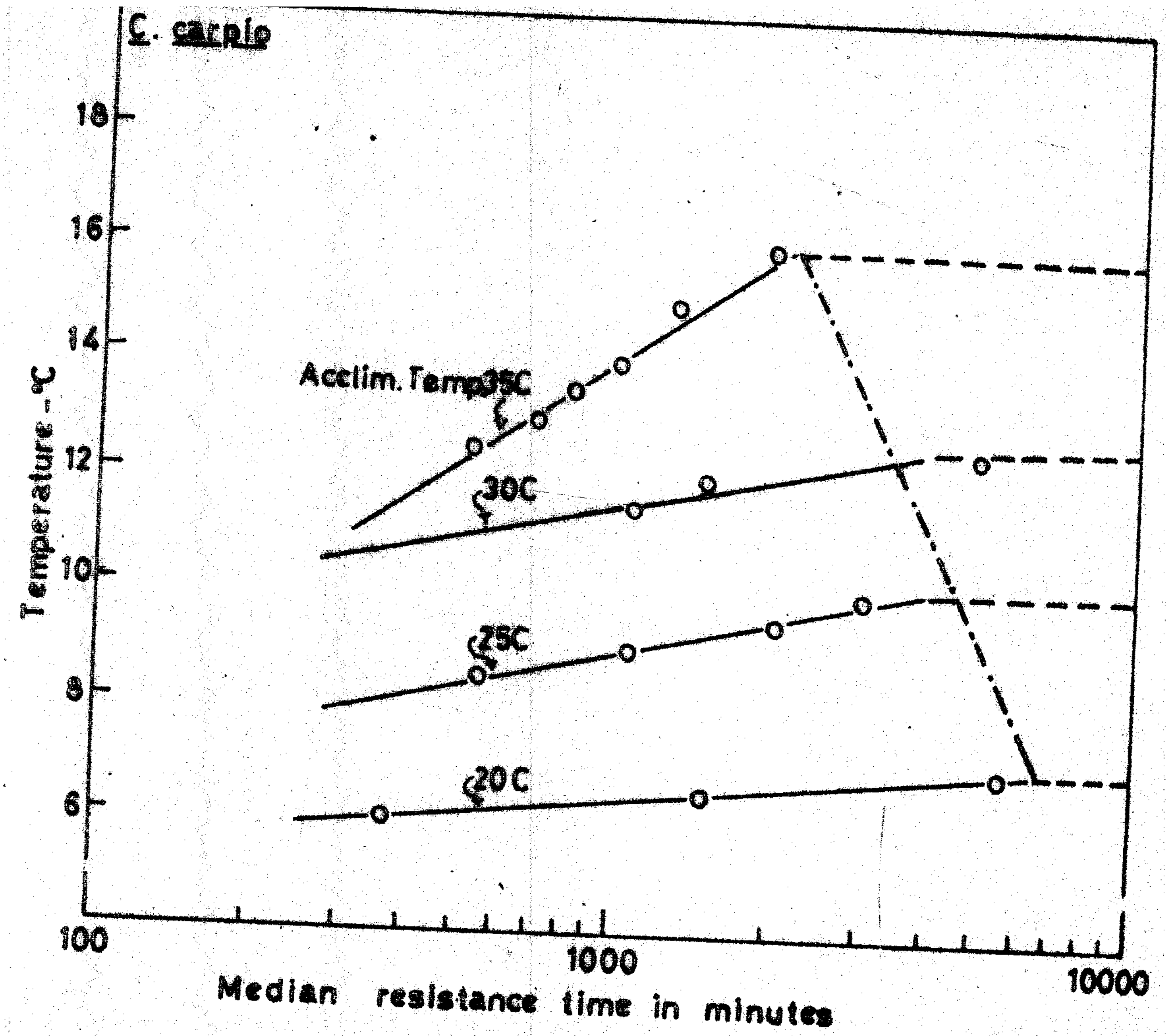

C.
Acclim. Temp.
35C
30C
25C
20C
18
16
14
12
10
8
6
0
2
4
6
8
Median resistance time - 1000 minutes

Fig.27

Median resistance times to low temperatures among common carp fry acclimated to temperatures indicated. Plotted on arithmetic x logarithmic axes


C. carpio
Acclim. Temp 35C
30C
25C
20C
Temperature -°C
18
16
14
12
10
8
6
100
1000
10000
Median resistance time in minutes

incipient lethal level. The lower incipient lethal temperatures calculated from the raw data (from the appendices Bd1 to Bd4) are 7.0, 10.1, 12.7 and 16.0°C for the acclimations 20, 25, 30 and 35°C respectively (Table XVII). The formulae for the regression lines (Fig.27) describing the thermal resistance to low temperatures are given in Table XIII.

v. Freshwater mullet:

Times to death in minutes of freshwater mullet acclimated to 15, 20, 25, 30 and 35°C and exposed to various low lethal temperatures ranging from 8.5 to 19.5°C are presented as raw data in appendices from Be1 to Be5, with other relevant details such as length and weight. As an example of these data, the times to death of 15°C acclimated freshwater mullet are shown in Fig.28 on a probit graph. The time-mortality curves fitted to 8.5 and 9.0°C in this figure are split-probits, whereas the other three curves pertaining to 9.5, 10.0 and 10.5°C are simple and straight.

The median resistance times estimated from various low lethal temperature exposures for the five acclimations by probability analysis are summarised in Table XXI. The general features of these data are same as in the case of

Fig.28

Times to death at various low lethaltest temperatures among freshwater mullet fingerlings acclimated to 15°C. Plotted on probit x logarithmic axes

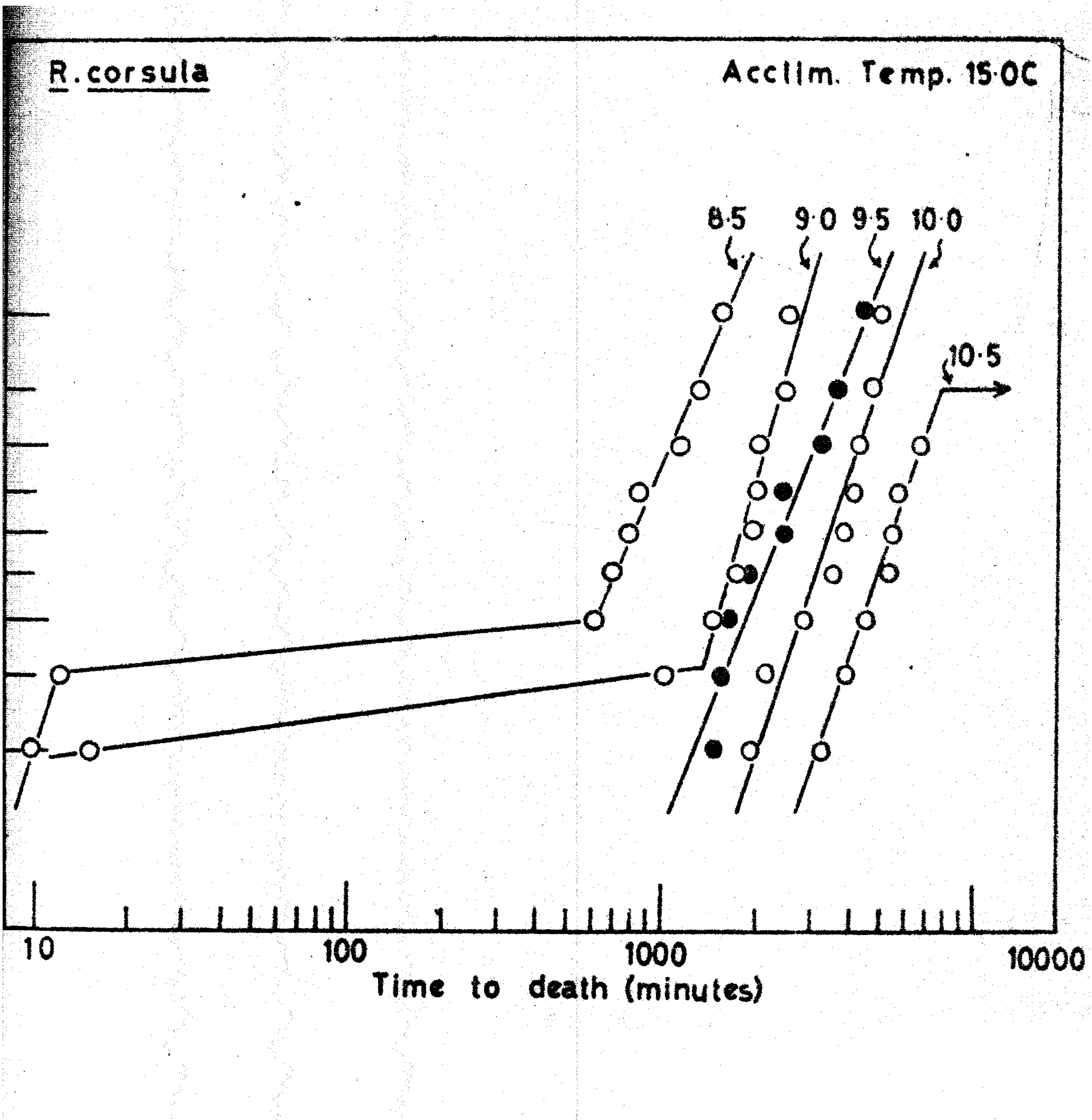

R. corsula
Acclim. Temp. 15·0C
8·5
9·0
9·5
10·0
10·5
10
100
1000
10000
Time to death (minutes)

Table XXI

Median resistance times to low lethal temperatures of

to different temperatures

Acclimation	Lethal temperatures in °C and							
Temp.(°C)	8.5	9.0	9.5	10.0	10.5	11.0	11.5	12.5
35	-	-	-	-	-	-	-	-
30	-	-	-	-	-	-	-	550
25	-	-	-	-	680	-	800	2300
20	-	-	-	1300	1625	3350	6000	-
15	810	1800	2375	3425	5400	-	-	-

Table XXI

eshwater mullet, (<u>Rhinomugil</u> <u>corsula</u>) fingerlings acclimated

dian resistance times to minutes									
.0	13.5	14.5	15.5	16.0	16.5	17.5	18.5	19.0	19.5
-	-	530	-	-	840	960	1530	2400	5800
-	810	2325	5200	6400	-	-	-	-	-
0	-	-	-	-	-	-	-	-	-
.	-	-	-	-	-	-	-	-	-
.	-	-	-	-	-	-	-	-	-

previous four species. The graphical presentation of these median resistance times is made in Fig.29 on arithmetic axes and on semilogarithmic axes in Fig.30. Formulae for the regression lines describing the thermal resistance of this species in Fig.30 are given in Table XV. The estimated lower incipient lethal temperatures for this species are 10.5, 11.5, 13.2, 15.8 and 19.5°C for the acclimations 15, 20, 25, 30 and 35°C respectively (Table XVII).

II Salinity tolerance:

The study of salinity tolerance has been carried out in five species, namely, mrigal, fringe-lipped carp, common carp, freshwater mullet and the cichlid fish *T. mossambica* (Table I).

i. Freshwater mullet:

The raw data on times to death in minutes of freshwater mullet exposed to various lethal salinities at different temperatures are presented in appendices from Ce1 to Ce5, with other relevant details. As an example, a probit plot of these data is presented in Fig.31; where the times to death of lots exposed to various salinities (40.0, 35.0,

Fig.29

Median resistance times to various low temperatures among freshwater mullet fingerlings acclimated to temperatures indicated. Plotted on arithmetic axes

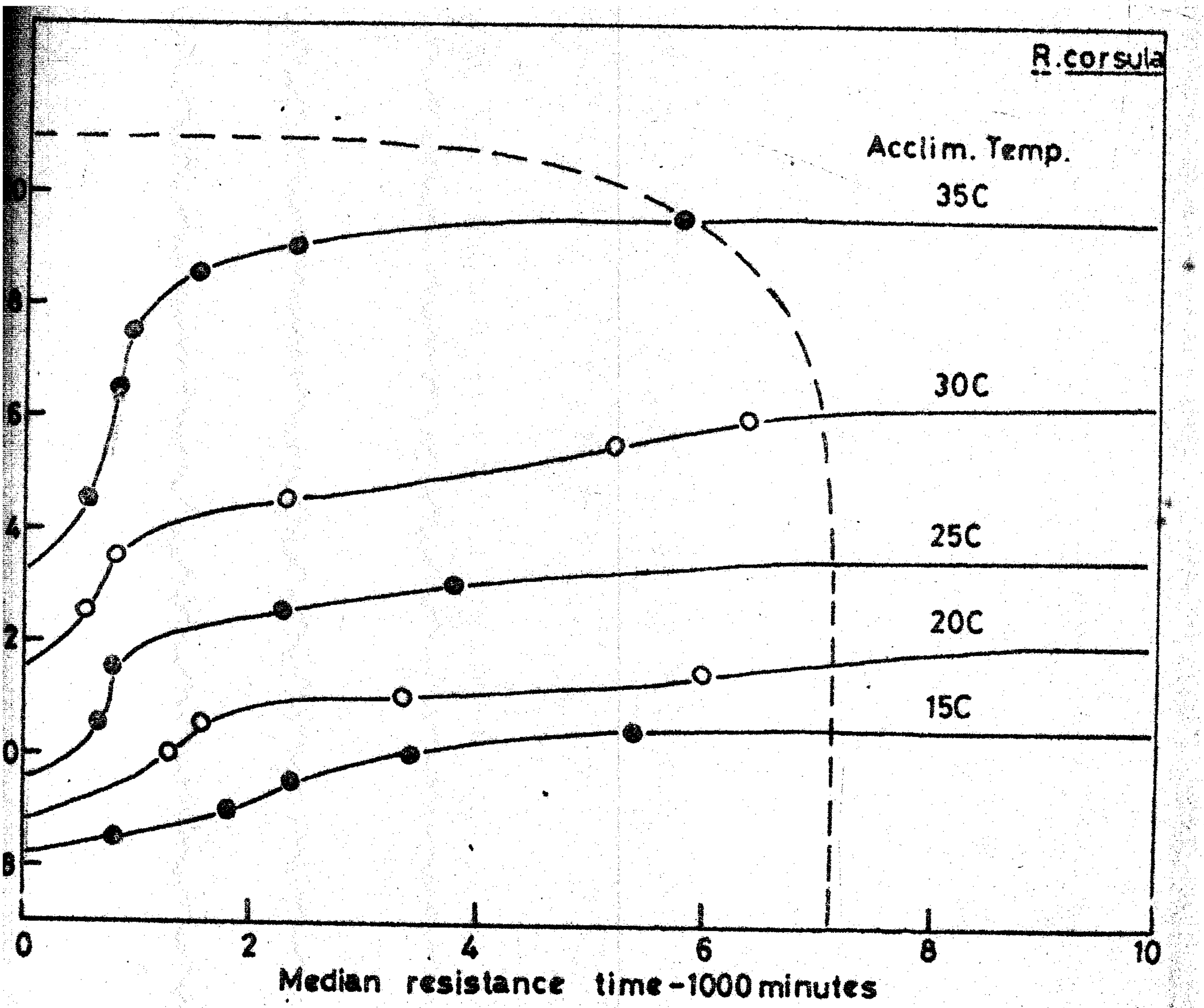

R. corsula
Acclim. Temp.
35C
30C
25C
20C
15C
0
2
4
6
8
10
Median resistance time - 1000 minutes

Fig.30

Median resistance times to various low temperatures among freshwater mullet fingerlings acclimated to temperatures indicated. Plotted on arithmetic x logarithmic axes

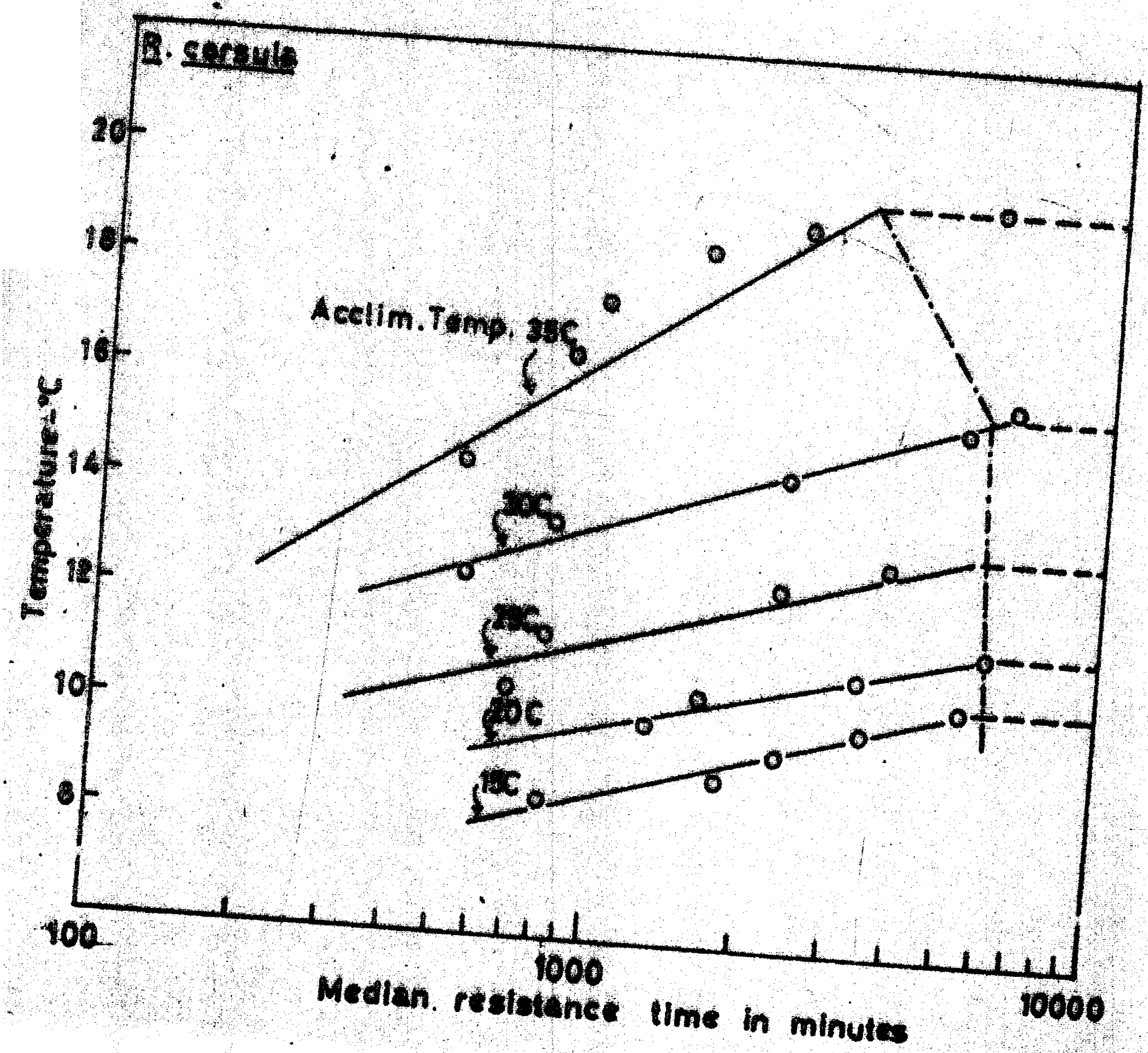
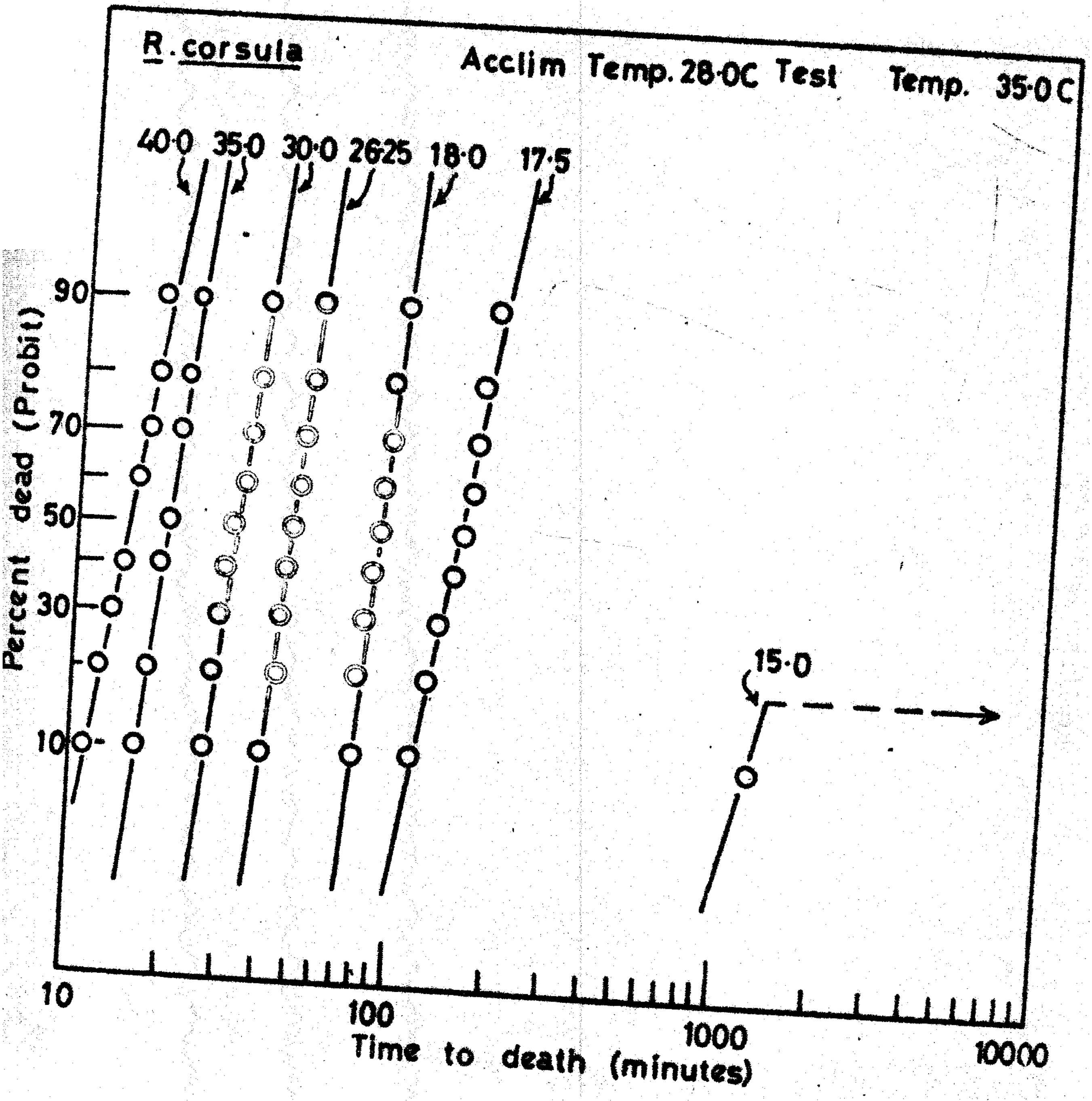

R. cersule
20
18
16
14
12
10
8
Temperature - °C
Acclim. Temp. 35C
30C
25C
20C
15C
100
1000
10000
Median. resistance time in minutes

Fig.31

Times to death in various lethal salinities at 35°C among freshwater mullet fingerlings acclimated to 28°C. Plotted on probit x logarithmic axes


R. corsula
Acclim Temp. 28·0C Test Temp. 35·0C
40·0
35·0
30·0
26·25
18·0
17·5
15·0
Percent dead (Probit)
90
70
50
30
10
10
100
1000
10000
Time to death (minutes)

26.25, 18.0, 17.5, 15.0 and 12.5‰ (S) at 35°C are treated as it was done for thermal tolerance (see also 'Methods'). The time mortality curves fitted through the plots are exclusively straight and simple, devoid of split-probits in any of these curves. The median resistance time with respect to each lethal salinity could be read from these curves as in the case of thermal death curves. The mortality in 12.5 and 15.0‰ (S) was 0% and 10% respectively, at 35°C and 100% mortality was recorded in 17.5‰ (S) and above (Fig.31).

The data obtained on median resistance times pertaining to different lethal salinity levels at various temperatures from 17.5 to 35.0°C are summarised in Table XXII. It is seen that the median resistance times decrease with an increase in lethal salinity level and temperature (within tolerance range).

The data from Table XXII are presented on a semilogarithmic graph in Fig.32, where the median resistance times and their respective lethal salinity levels have been plotted. The regression lines fitted through these plots describe the salinity resistance of this species at different temperatures. The two points with arrows next to them, pertaining to 20 and 17.5°C were not considered for the regression

Table XXII

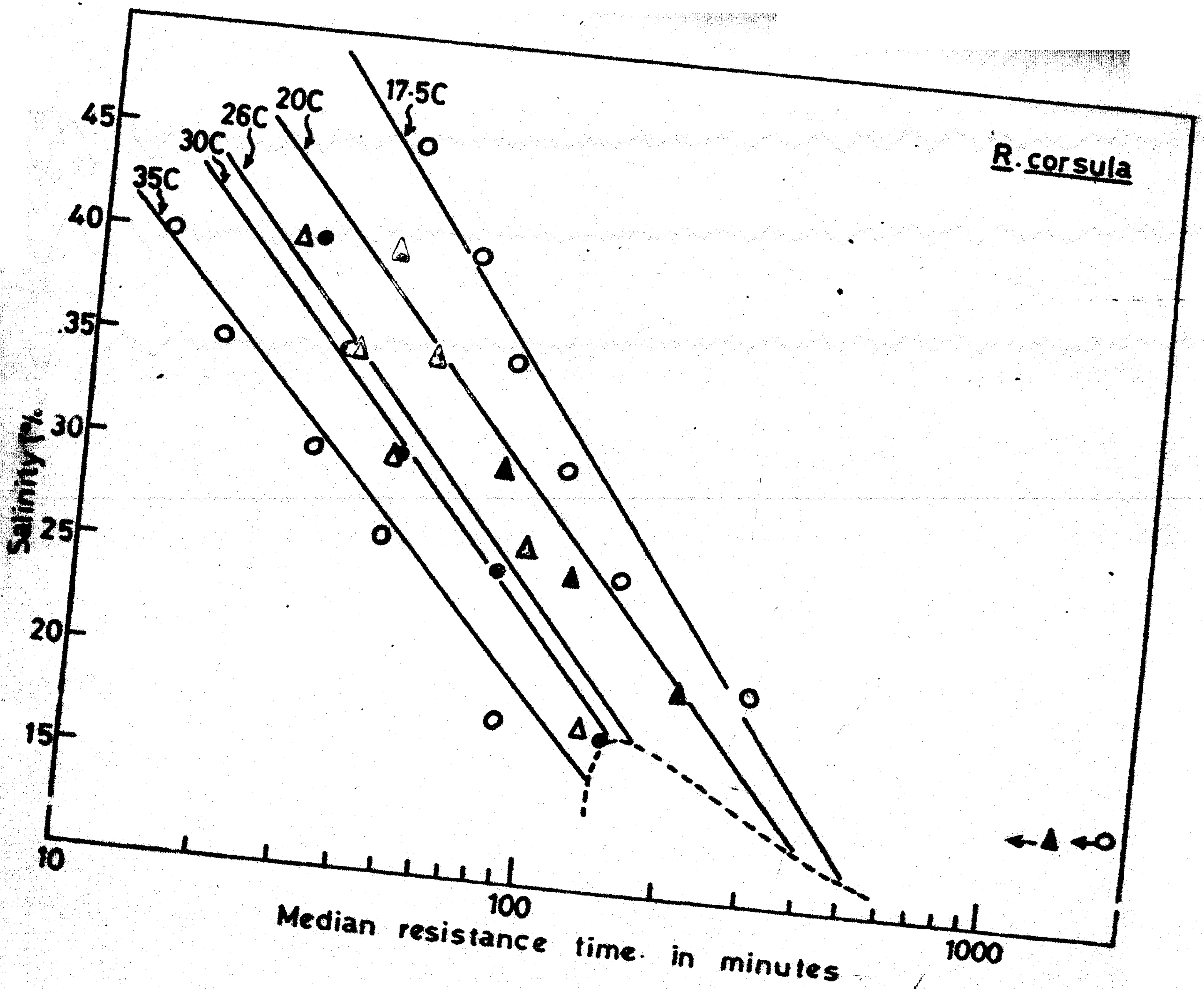
Median resistance times to death among fingerlings of <u>R</u>. <u>corsula</u> acclimated to freshwater at 28°C ± 1°C and exposed to different salinities and temperatures

Test Temp. (°C)	Median resistance times in minutes and salinity in ‰											
	45.00	40.0	35.0	30.0	26.25	25.0	20.0	18.0	17.5	15.0	12.5	10.0
35.0	-	14	19	31	45	-	-	85	148	ND	ND	ND
30.0	-	27	37	46	92	-	-	130	ND	ND	ND	ND
26.0	-	29	35	46	-	80	280	ND	ND	ND	ND	ND
20.0	-	43	54	80	-	116	210	-	-	1400	ND	ND
17.5	47	65	82	108	-	150	300	-	-	1800	ND	ND

ND = No death

Fig.32

Median resistance times to various lethal salinities at test temperatures indicated among freshwater mullet acclimated to ambient temperature 28°C. Plotted on arithmetic x logarithmic axes (see text for explanation)


R. corsula
17.5C
20C
26C
30C
35C
45
40
35
30
25
20
15
10
Salinity ‰
100
1000
Median resistance time in minutes

analysis so as to get a better fitting. Formulae for the regression lines are given in Table XXIII. It can be noted from this figure that the increase in test temperature reduces the salinity resistance of this species.

As in the case of thermal tolerance study, the incipient lethal salinities have been calculated from the raw data (appendices from Ce1 to Ce5) as per the method of Miller and Tainter (1944) and summarised in Table XXIV. The incipient lethal salinities are 12.3, 12.3, 17.3, 17.1 and 15.4‰ (S) at 17.5, 20.0, 26.0, 30.0 and 35.0°C respectively for the freshwater mullet. These values are also shown graphically in Fig.32 by a broken boundary line which terminates the regression lines at the lower ends and this line differentiates the zone of tolerance from the lethal zone.

ii. Other species:

Results of salinity tolerance study of mrigal, fringe-lipped carp, common carp and *T. mossambica* are presented hereunder. Raw data on times to death in minutes of the above mentioned four species, acclimated to freshwater at 30°C and tested to various salinities at 30°C are presented in appendices Ca1, Cc1, Cd1 and Cf1 respectively, along with other relevant details. The raw data of fringe-lipped carp

Table XXIII

Formulae for the regression lines describing the salinity resistance of *R. corsula* tested to different salinities and temperatures. (x is the salinity in parts per thousand; y is the log time in min)

Test Temperature (°C)	Formula
35.0	$y = 2.7807 - 0.0417\ x$
30.0	$y = 2.7599 - 0.0336\ x$
26.0	$y = 3.2145 - 0.0468\ x$
20.0	$y = 3.6465 - 0.0547\ x$
17.5	$y = 3.5590 - 0.0454\ x$

Table XXIV

Upper incipient lethal salinity with ± one SD for fingerlings of freshwater mullet, R. *corsula* acclimated to fresh water at 28°C ± 1°C and exposed to various lethal salinities at different temperatures

Temperature °C	Upper incipient lethal salinity ‰ (S)
35.0	15.35 ± 1.2
30.0	17.90 ± 0.7
26.0	17.32 ± 1.3
20.0	12.25 ± 1.3
17.5	12.25 ± 1.3

acclimated to 5‰ (S) at 30°C and tested at the same temperature to different salinities, are presented in Appendix Cc2. As an example of these data, the time to death values from Appendix Cc1 and Cc2 of fringe-lipped carp are presented on a probit chart in Fig.33. The time-mortality curves of both freshwater (open circles) and salt water (closed circles) acclimated fish are again straight and simple devoid of any break in shape as in the case of freshwater mullet. The median resistance times can be obtained from these curves as shown in previous cases. The positive time-mortality curves tend to shift towards the time axis as the lethal salinity level decreases suggesting the increasing salinity resistance of this species.

The median resistance times obtained by probability analysis are summarised in Table XXV for all the species including freshwater mullet (30°C data from Table XXI). The data indicated by asterisk marks are the values for saltwater acclimated (5‰ (S) fringe-lipped carp. It is seen from these data, among the five species mrigal is the least resistant to salinity, whereas the freshwater mullet and Tilapia mossambica are the highly resistant species and the other two species are intermediate. The saltwater acclimated fringe-lipped carp show enhanced salinity resistance, but there seems to be no change in tolerance level. The data

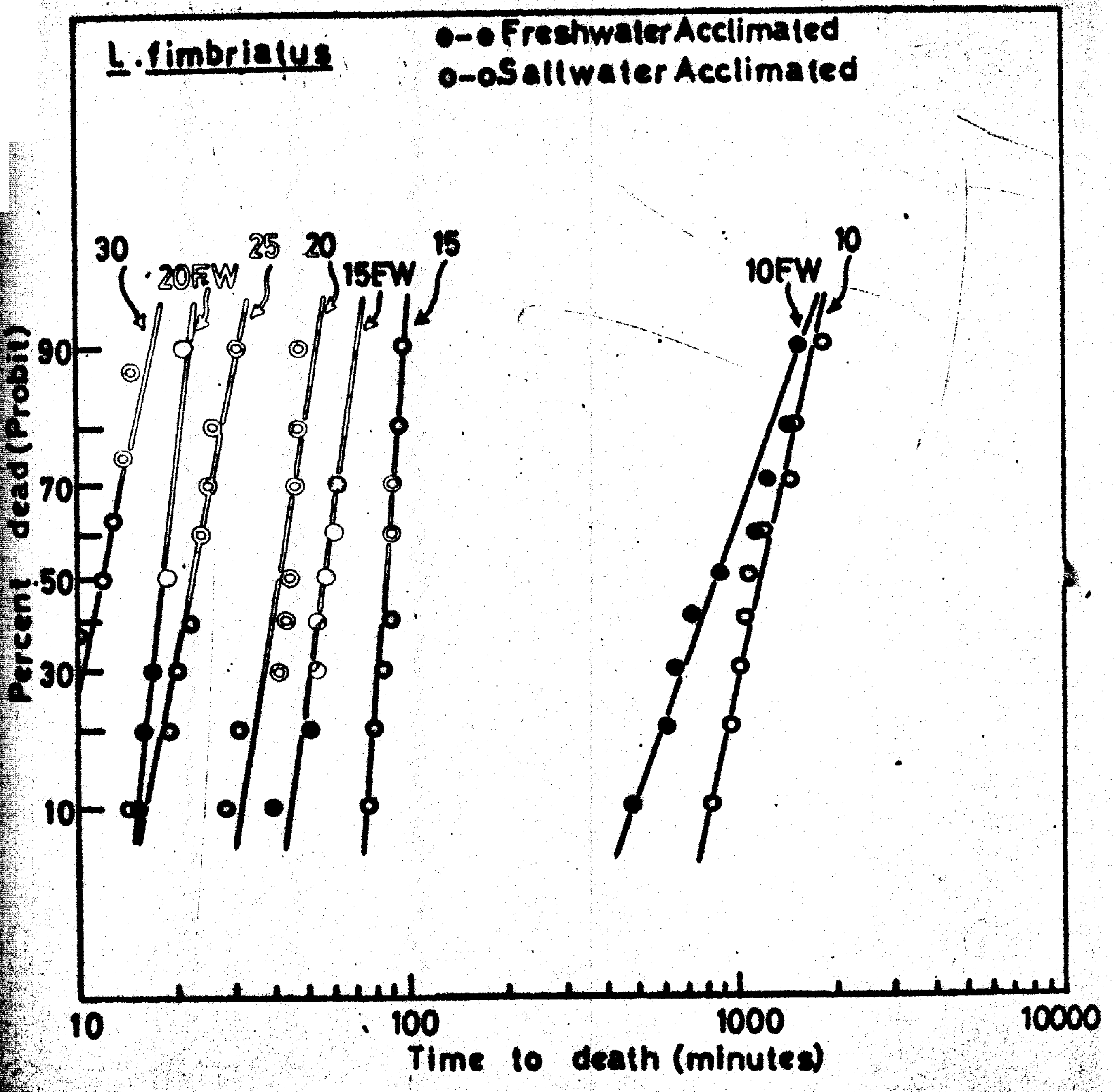

L. fimbriatus
●–● Freshwater Acclimated
o–o Saltwater Acclimated
30
20FW
25
20
15FW
15
10FW
10
90
70
50
30
10
Percent dead (Probit)
10
100
1000
10000
Time to death (minutes)

Table XXV

Median resistance times to death among fry of carps and T. mossambica acclimated to freshwater at 30°C ± 1°C and tested to various salinities at 30°C ± 1°C. Data on R. corsula taken from Table XXI

Species	Median resistance times in minutes and salinity in ‰								
	35.00	30.00	26.25	25.00	20.00	18.00	15.00	10.00	5.00
C. mrigala	-	-	-	-	14	-	35	270	3650
L. fimbriatus	-	-	-	-	19	-	57	870	ND
	-	-	-	23*	43*	-	88*	1160*	ND
C. carpio	-	8	-	13	18	-	31	1475	ND
T. mossambica	48	53	-	95	3350	-	5000	10200	ND
R. corsula	37	46	92	-	-	130	ND	ND	ND

ND = No death

*Acclimated to 5‰ (S) at 30°C ± 1°C

on median resistance times in Table XXV are graphically presented in Fig.34 on a semilogarithmic graph and regression lines have been fitted through the plots of individual species. The formulae for the regression lines describing the salinity resistance of carps and *T.mossambica* are shown in Table XXVa. The short, steep, negatively sloped curve of fresh water mullet indicates that this species has a lower resistance than *T. mossambica*. Though, *Tilapia* has a higher resistance and its tolerance level is about 7‰ (S) which is lower than that of freshwater mullet. Two different lines are shown for fringe-lipped carp, one (continuous line) for freshwater acclimated group and the other (broken line connecting dots) for saltwater acclimated group. The space between these two lines indicates the gain in salinity resistance by this species due to saltwater acclimation. However, there is no change in tolerance level. The incipient lethal salinity levels calculated are 3.54, 7.07, 8.13 and 7.07 ‰ (S) for mrigal, fringe-lipped carp, common carp and *Tilapia mossambica*. The incipient lethal value for the salinity acclimated fringe-lipped carp is also 7.07‰ (S).

Fig.34

Median resistance times to different lethal salinities among the species indicated at ambient temperatures 30°C. Plotted on arithmetic x logarithmic axes. F.W. fresh water acclimation

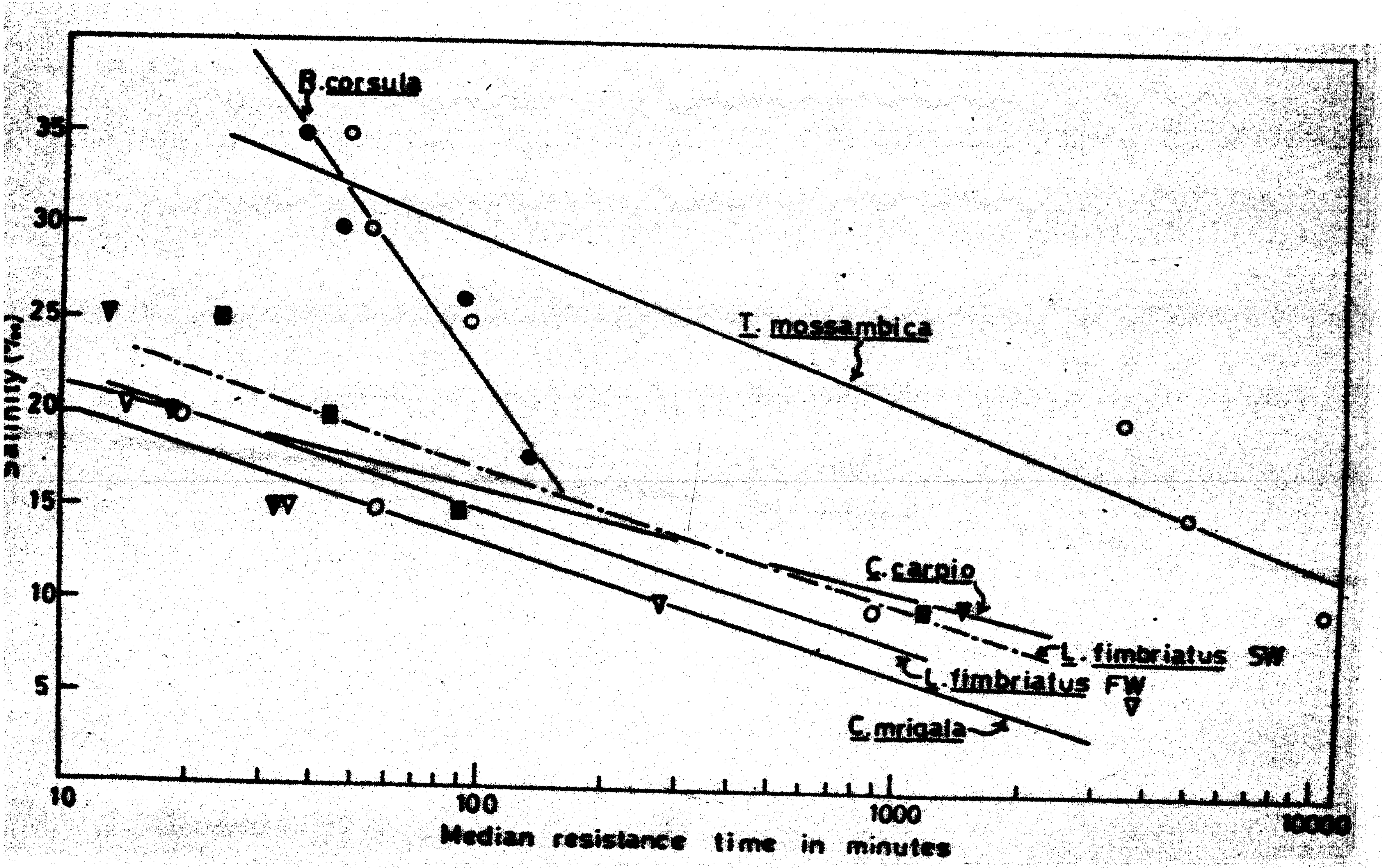

Salinity (‰)
35
30
25
20
15
10
5
10
100
1000
10000
Median resistance time in minutes
B. corsula
T. mossambica
C. carpio
L. fimbriatus SW
L. fimbriatus FW
C. mrigala

Table XXVa

Formulae for the regression lines describing the salinity resistance of <u>C</u>. <u>mrigala</u>, <u>L</u>. <u>fimbriatus</u>, <u>C</u>. <u>carpio</u> and <u>T</u>. <u>mossambica</u> tested to various salinity concentrations at 30 ± 1°C; (x is the salinity in parts per thousand; y is the log time in minutes)

Species	Formulae
C. mrigala	$y = 3.6352 - 0.1285\ x$
L. fimbriatus	$y = 4.4825 - 0.1661\ x$
	$^{*}y = 3.6470 - 0.0915\ x$
C. carpio	$y = 4.1838 - 0.1387\ x$
T. mossambica	$y = 5.4628 - 0.1176\ x$

*Formula for salinity acclimated (5‰ (S) *L. fimbriatus*

II Salinity effect on thermal resistance

The study of salinity effect on thermal resistance has been carried out in freshwater mullet alone (Table I). The raw data obtained on time to death in minutes of freshwater mullet acclimated to 20, 25, 30 and 35°C and tested at different salinities (from freshwater to 25‰(S) at 37, 39 and 41°C (lethal temperatures) are presented in appendices from Ce6 to Ce13, with other relevant details. As already done in the preceeding cases these data are treated by probability analysis. As an example, the data of 20°C acclimation groups, exposed in lots of various salinities (freshwater, 3, 5, 7, 10, 12, 15, 18, 20 and 25 ‰(S) at 37°C (lethal temperature) are presented in Fig.35 on a probit chart. It can be seen from Fig.35 that the median resistance time of fish exposed to 3, 5, 7, 10 and 12 ‰(S) are higher than that of freshwater group (salinity being non-lethal) and for those salinities above 12 ‰(S) are lower than that of the freshwater group.

The median resistance times obtained from the time-mortality curves pertaining to different lethal levels of salinity and temperatures of all the four acclimation levels (20, 25, 30 and 35°C) are summarised in Table XXVI. The data of 30 and 35°C acclimations were obtained from tests

Fig.35

Times to death in fresh and different salt water media at lethaltest temperature 37°C among fresh-water mullet fingerlings acclimated to 20°C. Plotted on probit x logarithmic axes

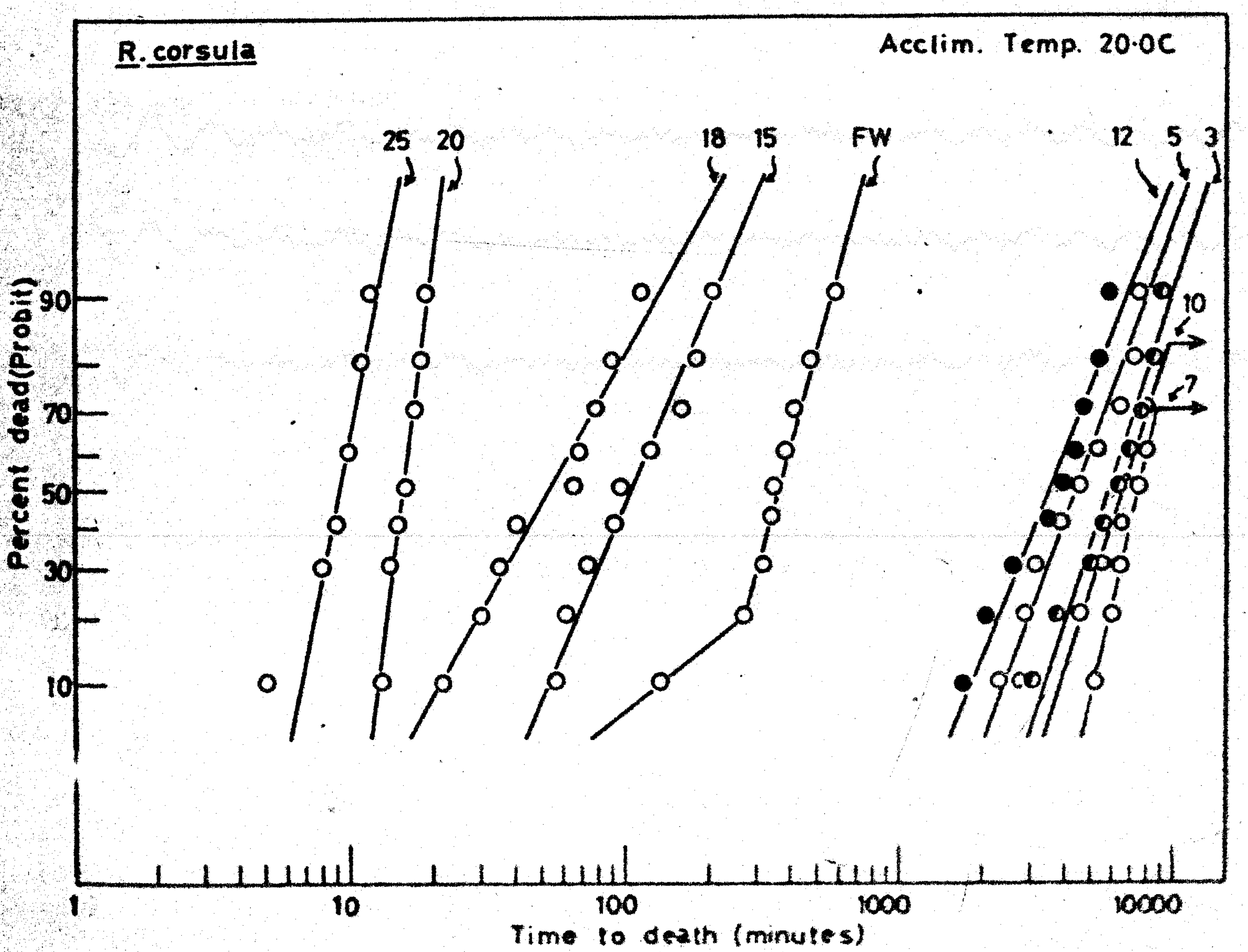

R. corsula
Acclim. Temp. 20·0C
25
20
18
15
FW
12
5
3
10
7
Percent dead(Probit)
10
30
50
70
90
Time to death (minutes)
1
10
100
1000
10000

Table XXVI

Median resistance times to death among fingerlings of freshwater mullet R. corsula acclimated and tested at different lethal temperature and salinity concentrations

Acclimation Temp. (°C)	Test Temp. (°C)	Median resistance times in minutes and salinity in ‰									
		25	20	18	15	12	10	7	5	3	Fresh water
35	41	-	-	-	46	-	-	158	-	-	104*
	39	-	-	-	700	-	-	930	-	-	840*
	37	-	-	-	2380	-	-	4800	-	-	4250*
30	41	-	-	-	31	-	-	53	-	-	38*
	39	-	-	-	385	-	-	1475	-	-	640*
	37	-	-	-	1880	-	-	5950	-	-	3000*
25	39	-	14	-	90	470	780	870	570	660	175*
20	37	9	16	53	105	3350	6680	7600	4500	6000	360*

*Fresh water data taken from thermal tolerance study in freshwater, 7 and 15 °/₀° (S) at three different lethal temperatures i.e. 37, 39 and 41 °C. The median resistance times increase as the salinity decreases down to 7 °/₀° (S) and afterwards decreases until fresh water level is reached. This indicates that the highest resistance to lethal temperature is exhibited by this species at 7 °/₀° (S).

The data on median resistance times from Table XXVI for acclimations 20, 25 and 30°C are graphically shown in Fig.36, 37 and 38 respectively on arithmetic graphs. The curves fitted through the plots are bimodel, having peaks at 5 and 7 °/₀° (S). In Fig.38, the curves are partially shown by broken lines from freshwater level to 7 °/₀° (S) to indicate the bimodel nature which might have been obvious if tests could have also been carried out at 5 °/₀° (S) as in the case of 20 and 25°C acclimations. These three figures show the salinities from 3 to 12 °/₀° (S) being close to isotonic level enhance the resistance capacity and salinities above 15 °/₀° (S) being lethal counter act with lethal temperature to speed up mortification. Bimodel resistance response at 5 and 7 °/₀° (S) is explained in detail under 'Discussion'.

The data on median resistance times from 30 and 35°C acclimations also are presented on semilogarithmic graph in

Fig.36

Median resistance times to lethaltest temperature 37°C in fresh and salt water media among freshwater mullet fingerlings acclimated to 20°C. Plotted on arithmetic axes

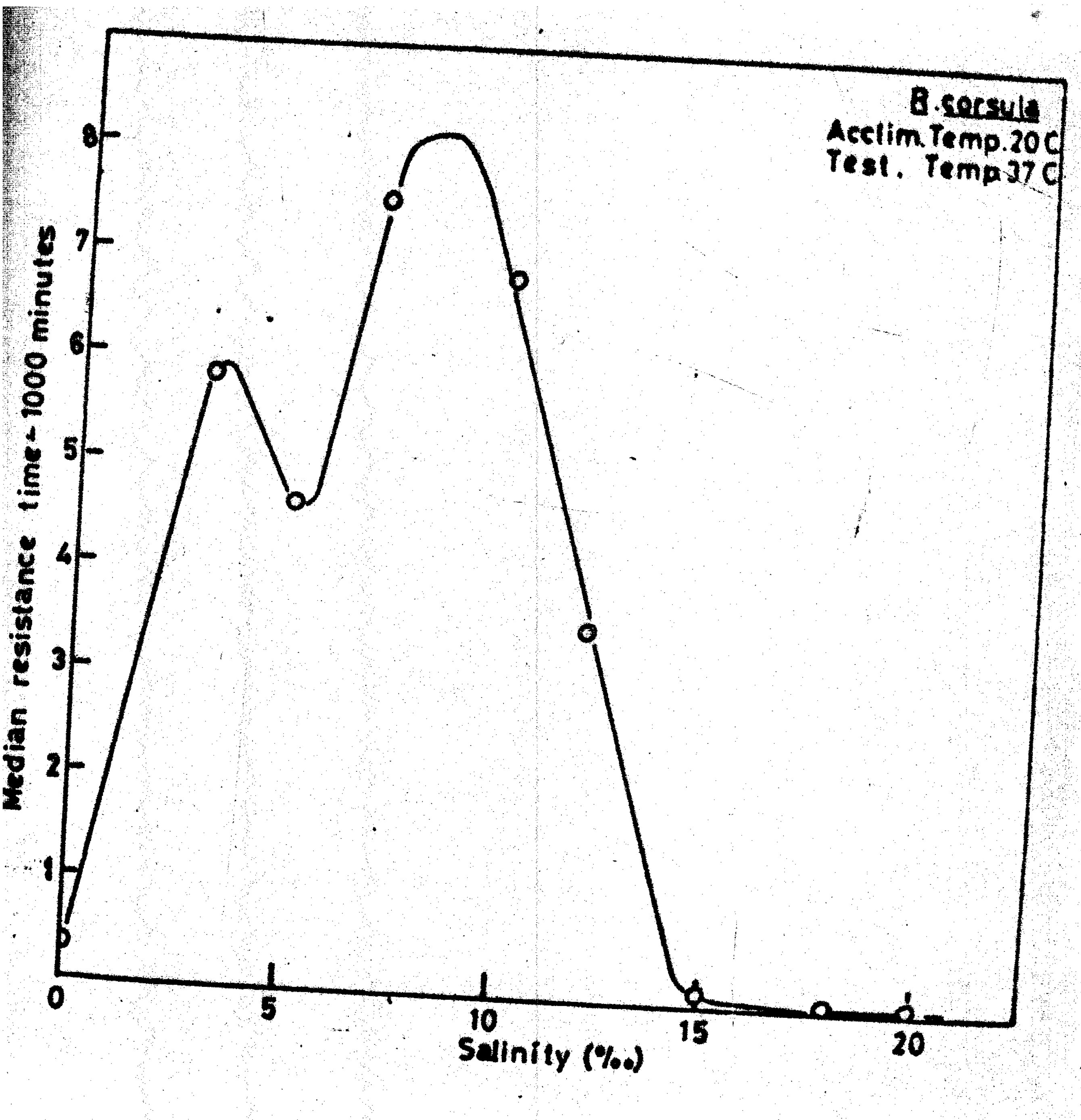

B. corsula
Acclim. Temp. 20 C
Test. Temp 37 C
Median resistance time - 1000 minutes
8
7
6
5
4
3
2
1
0
5
10
15
20
Salinity (‰)

Fig.37

Median resistance times to lethaltest temperature 39°C in fresh and salt water media among freshwater mullet acclimated to 25°C. Plotted on arithmetic axes

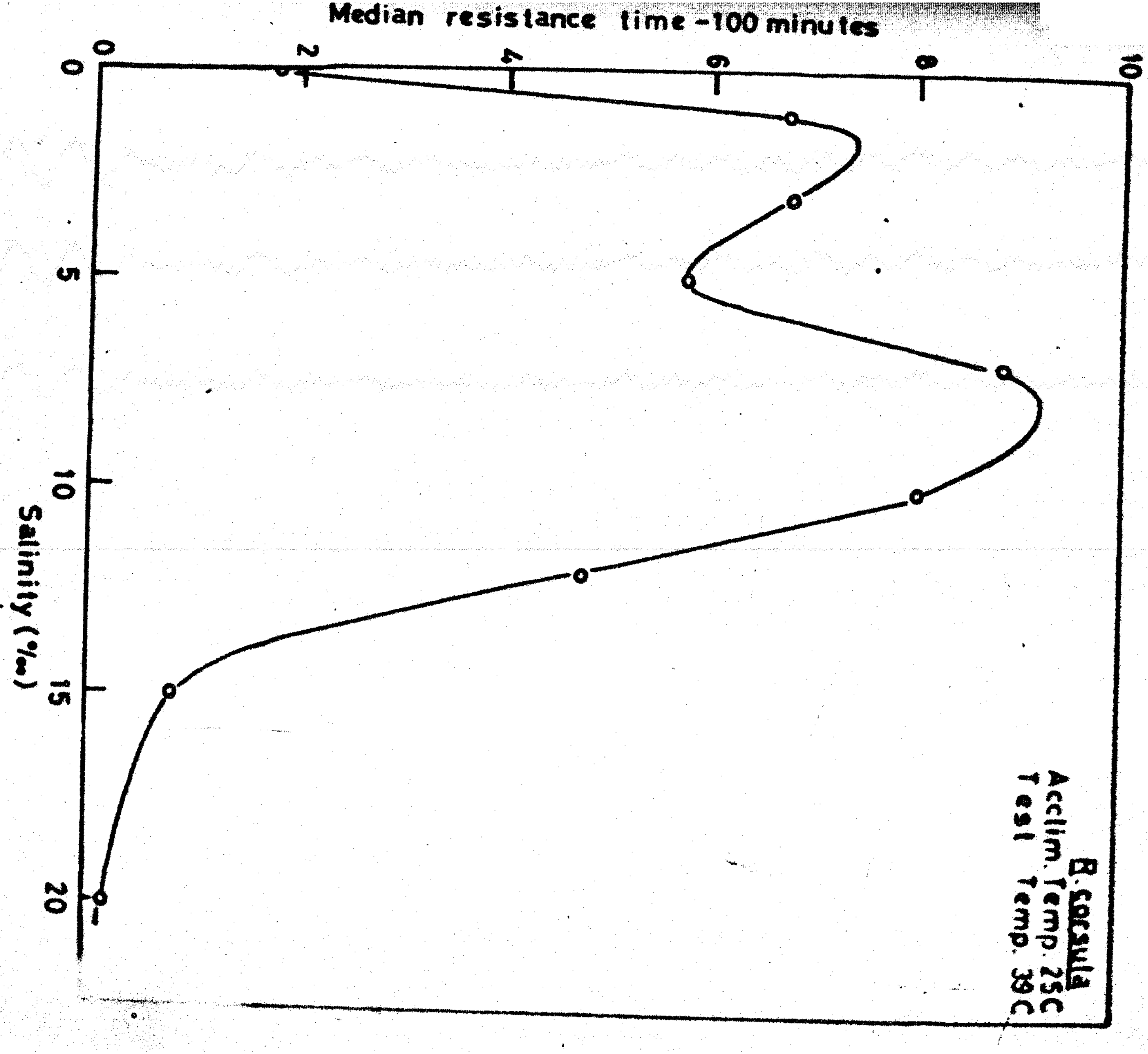

Median resistance time -100 minutes
0
2
4
6
8
10
0
5
10
15
20
Salinity (‰)
B. corsula
Acclim. Temp. 25C
Test Temp. 39C

Fig.38

Median resistance times to different high lethal temperatures in different salinities among the freshwater mullet acclimated to 30°C. Plotted on arithmetic axes

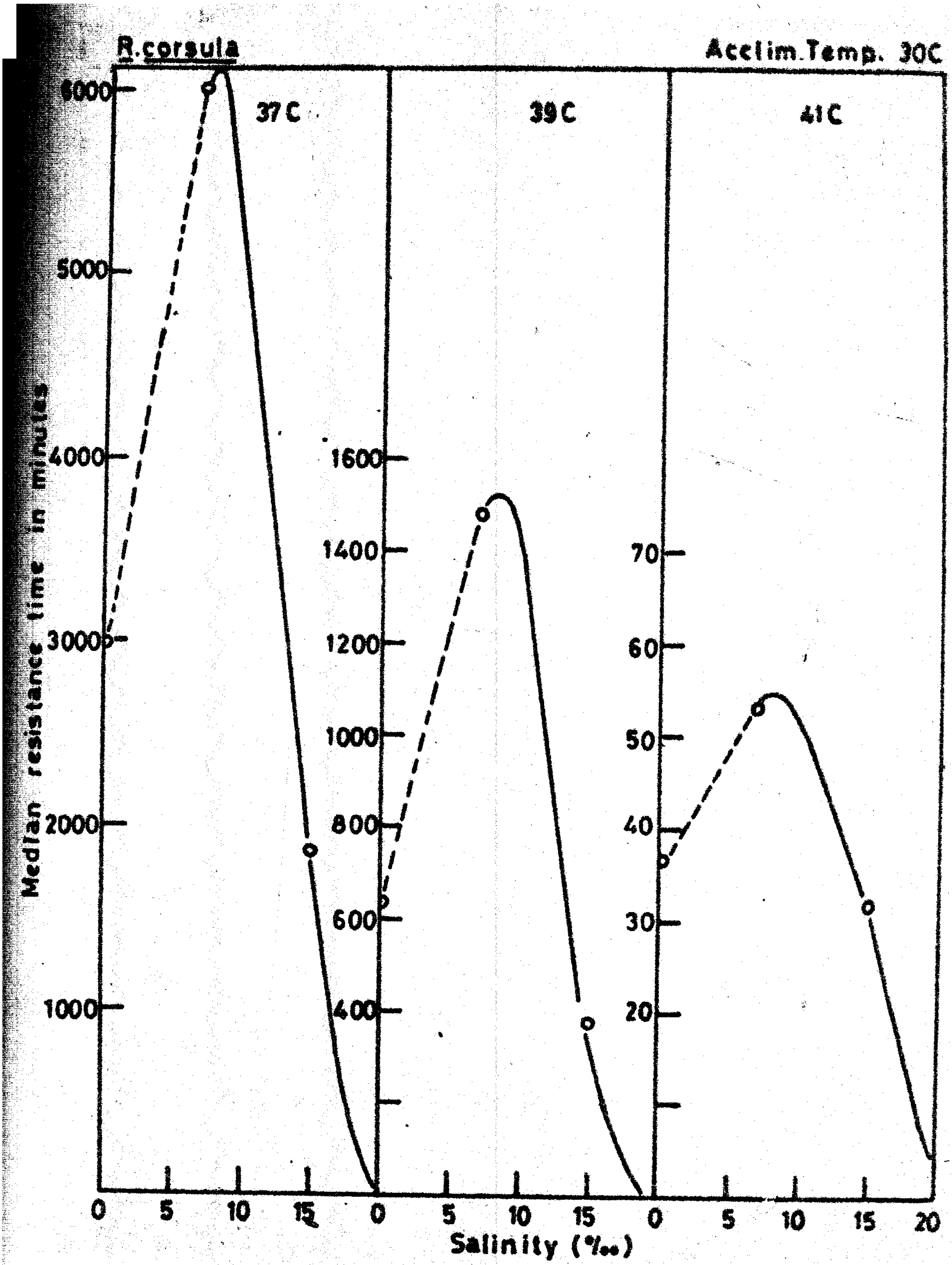

R.corsula
Acclim.Temp. 30C
37C
39C
41C
Median resistance time in minutes
6000
5000
4000
3000
2000
1000
1600
1400
1200
1000
800
600
400
70
60
50
40
30
20
0
5
10
15
0
5
10
15
0
5
10
15
20
Salinity (‰)

Fig.39 where two distinct observations can be made. First the resistance is high at 7 $^{0}/_{00}$ (S) at all lethal temperatures of both the acclimations. Secondly, the effect of thermal acclimation is different at the lethal temperatures. At 41°C, the samples from 35°C acclimated group exhibit higher resistance at all the three salinity levels (fresh water, 7 and 15 $^{0}/_{00}$ (S). Whereas at 37 and 39°C the resistance of the 35°C acclimated samples is lower at 7 $^{0}/_{00}$ (S) than the 30°C acclimated group. 35°C is very close to the upper incipient lethal temperature of this species i.e., 36.5°C (Table VII) and hence the acclimation at this temperature has not effectively enhanced the resistance of this species at lethal temperatures below sharp lethal levels (i.e. 39 and 37°C). In Fig.40, the same data on median resistance times shown in Fig.39 are plotted against temperature levels on a semilogarithmic graph and regression lines are fitted. The continuous lines belong to 35°C acclimation and broken lines belong to 30°C. The curves pertaining to 15 and 7 $^{0}/_{00}$ (S) of both acclimations fall respectively on the left and right side of the freshwater curve, which again indicate the higher resistance at 7 $^{0}/_{00}$ (S) The effect of thermal acclimation can be noted from the shifting of the 30°C acclimation curves (freshwater and 7 $^{0}/_{00}$ (S) towards the right at the lower ends in Fig.40. The formulae for these regression lines are given in Table XXVII.

Fig.39

Median resistance times to different lethaltest temperatures in various salinities among freshwater mullet fingerlings acclimated to 30 and 35°C. Plotted on arithmetic x logarithmic axes

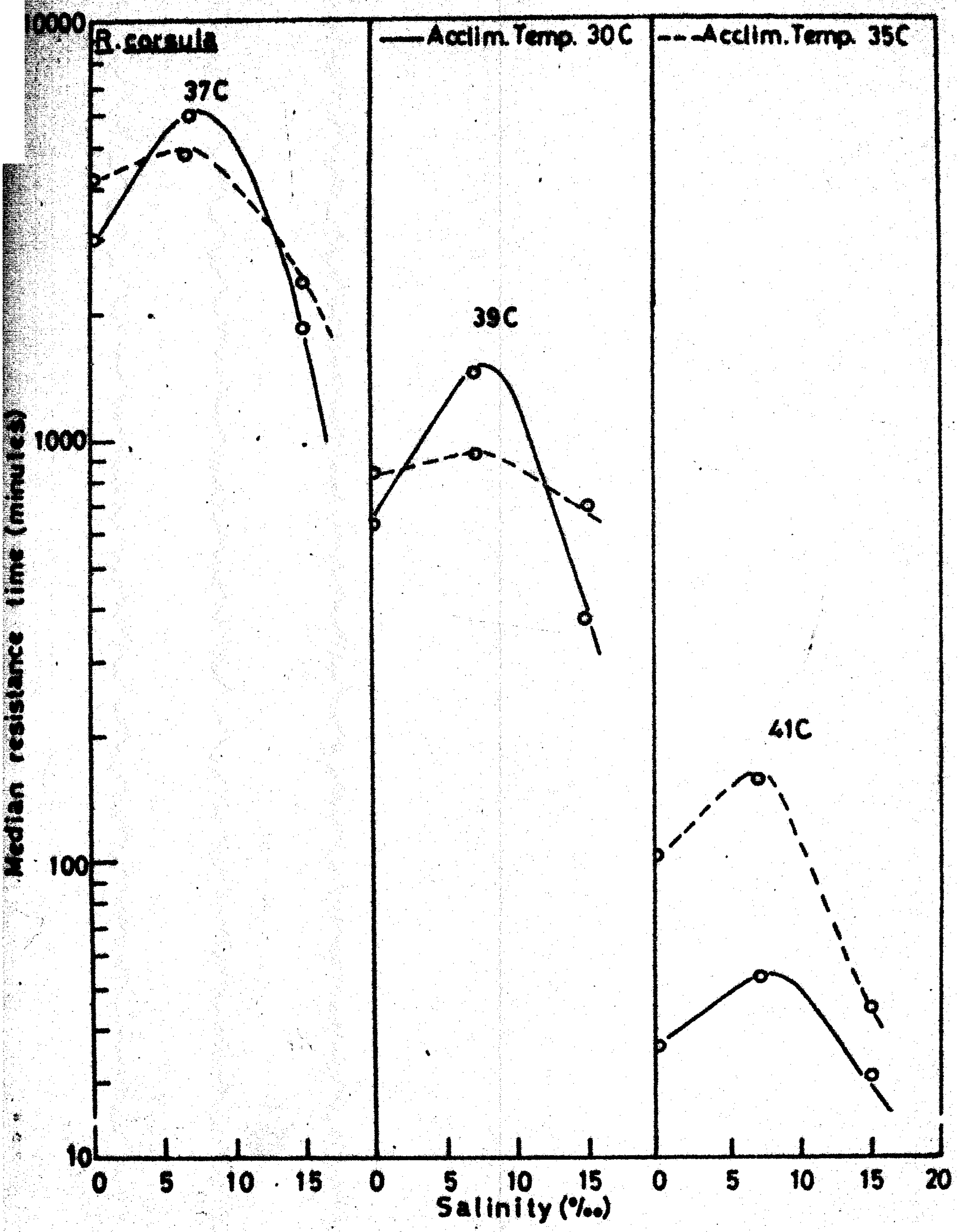

R. corsula
—— Acclim. Temp. 30C
- - - Acclim. Temp. 35C
37C
39C
41C
10000
1000
100
10
Median resistance time (minutes)
0
5
10
15
0
5
10
15
0
5
10
15
20
Salinity (‰)

Fig.40

Median resistance times to various lethaltest temperatures indicated in different salinities among freshwater mullet acclimated to 30 and 35°C. Plotted on arithmetic x logarithmic axes

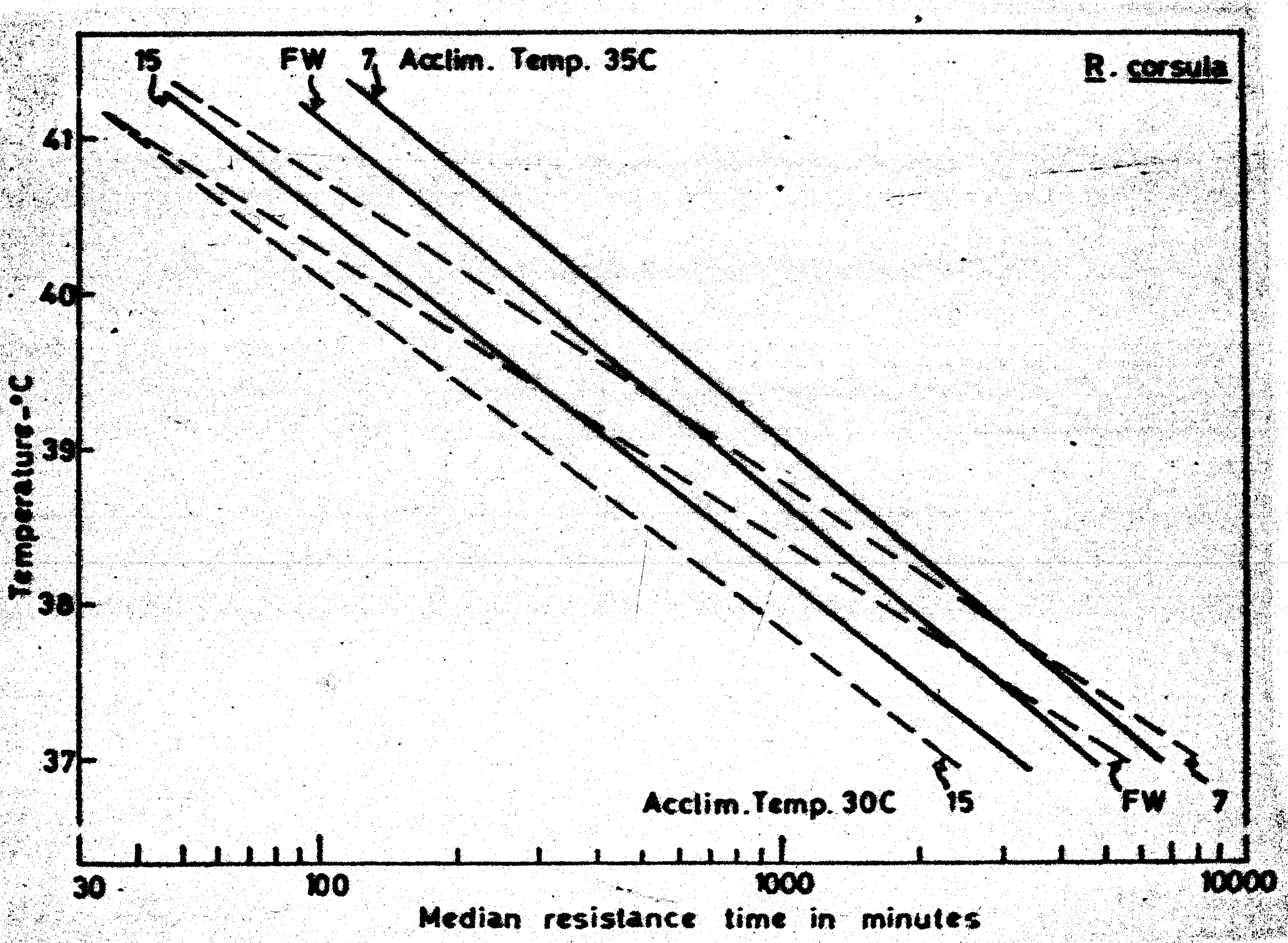

15
FW
7 Acclim. Temp. 35C
R. corsula
Temperature-°C
41
40
39
38
37
Acclim.Temp.30C
15
FW
7
30
100
1000
10000
Median resistance time in minutes

Table XXVII

Formulae for the regression lines describing the thermal resistance at different salinities of <u>R. corsula</u> acclimated to 30 and 35°C (x is the temperature in °C; y is the log time in min)

Salinity (‰)	Acclimation temperature	
	~~tion~~ 30°C	35°C
15.0	$y = 20.0480 - 0.4510\,x$	$y = 19.2866 - 0.4253\,x$
7.0	$y = 22.7231 - 0.5085\,x$	$y = 18.4927 - 0.3972\,x$
F.W	$y = 23.6483 - 0.5372\,x$	$y = 19.8178 - 0.4340\,x$

F.W = Fresh water

Discussion

Thermal Tolerance:

Zones of thermal tolerance and resistance:

Temperature response of organisms, as dealt with in the earlier text herein, is divisible into tolerance and resistance; quantified values of these two physiological parameters have been used to describe the thermal characteristics of several fishes by various workers (Fry *et al.*, 1942, 1946; Doudoroff, 1942, 1945; Brett, 1944, 1952; Hart, 1952; Charlon, 1968; Ananthakrishnan and Kutty, 1976). Similar descriptions are presented herein for the five species presently studied.

In an illustration of the plots of data on tolerance and resistance of individual species the zones of tolerance and resistance can be marked out. The zone of thermal tolerance (see also 'Materials and Methods') is bounded by upper and lower incipient lethal temperatures for the widest possible range of temperature acclimation and terminated by ultimate lethal temperatures (Fry _et_ _al_., 1942, 1946; Brett, 1944, 1952; Hart, 1947, 1949, 1952; Cocking, 1959; Pandian, 1967; Ananthakrishnan and Kutty, 1976). The zone of resistance is the area surrounding the zone of tolerance, bounded by the upper and lower incipient lethal temperature of immediate death as the outer boundary for both high and low temperatures (Coutant, 1970; Charlon, 1968; Charlon _et_ _al_., 1970). Further, the zones of resistance and tolerance can be summarily divided into 'upper' and 'lower' zones of resistance and tolerance with the aid of isotherm-line which is constructed at 45° angle for both the axes (Mc Erlean _et_ _al_., 1969) as has also been shown in the present study.

The zones of tolerance and resistance for mrigal, rohu, fringe-lipped carp, common carp and freshwater mullet based on present data are shown in Figs.41, 42, 43, 44 and 45 respectively. For a general discussion of the common

Fig.41

Zones of thermal tolerance and resistance for mrigal fry in fresh water (see text for furthe explanation)

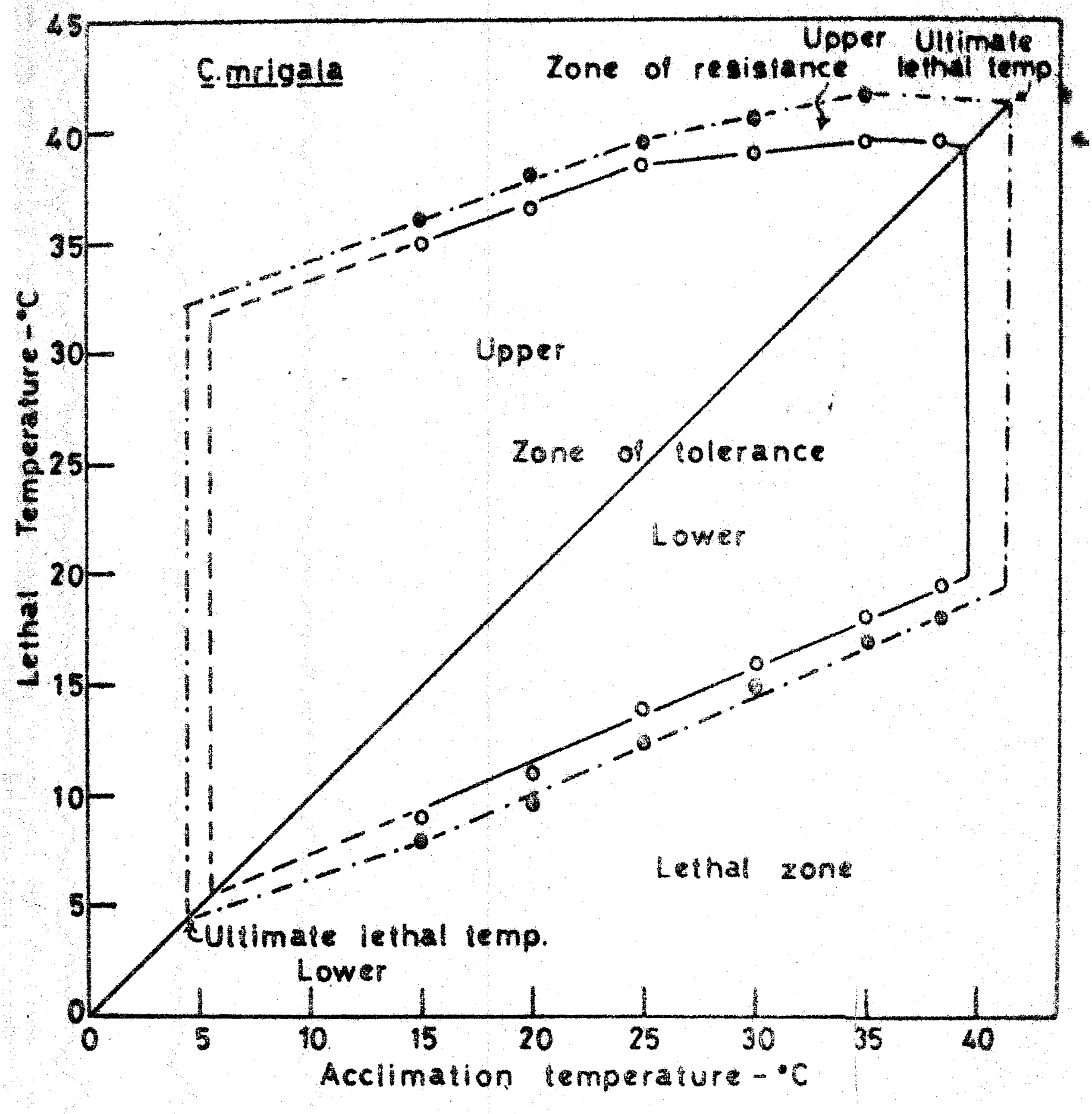

C. mrigala
Upper Ultimate lethal temp.
Zone of resistance
Upper
Zone of tolerance
Lower
Lethal zone
Ultimate lethal temp.
Lower
Lethal Temperature - °C
Acclimation temperature - °C
0
5
10
15
20
25
30
35
40
45

Fig.42

Zones of thermal tolerance and resistance for rohu fry in fresh water

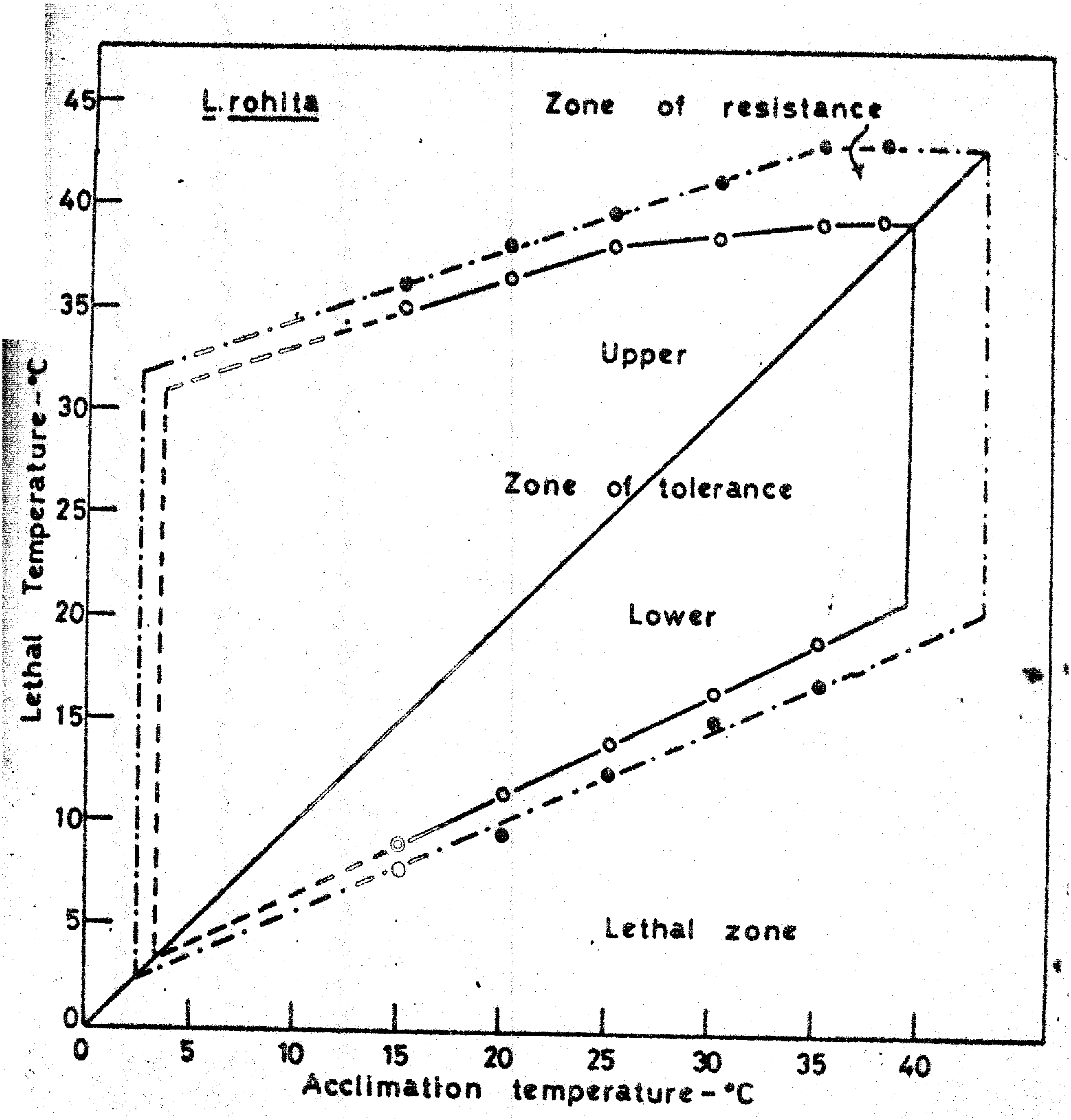

L.rohita
Zone of resistance
Upper
Zone of tolerance
Lower
Lethal zone
Lethal Temperature - °C
Acclimation temperature - °C
0
5
10
15
20
25
30
35
40
45

Fig.43

Zones of thermal tolerance and resistance for fringe-lipped carp fry in fresh water

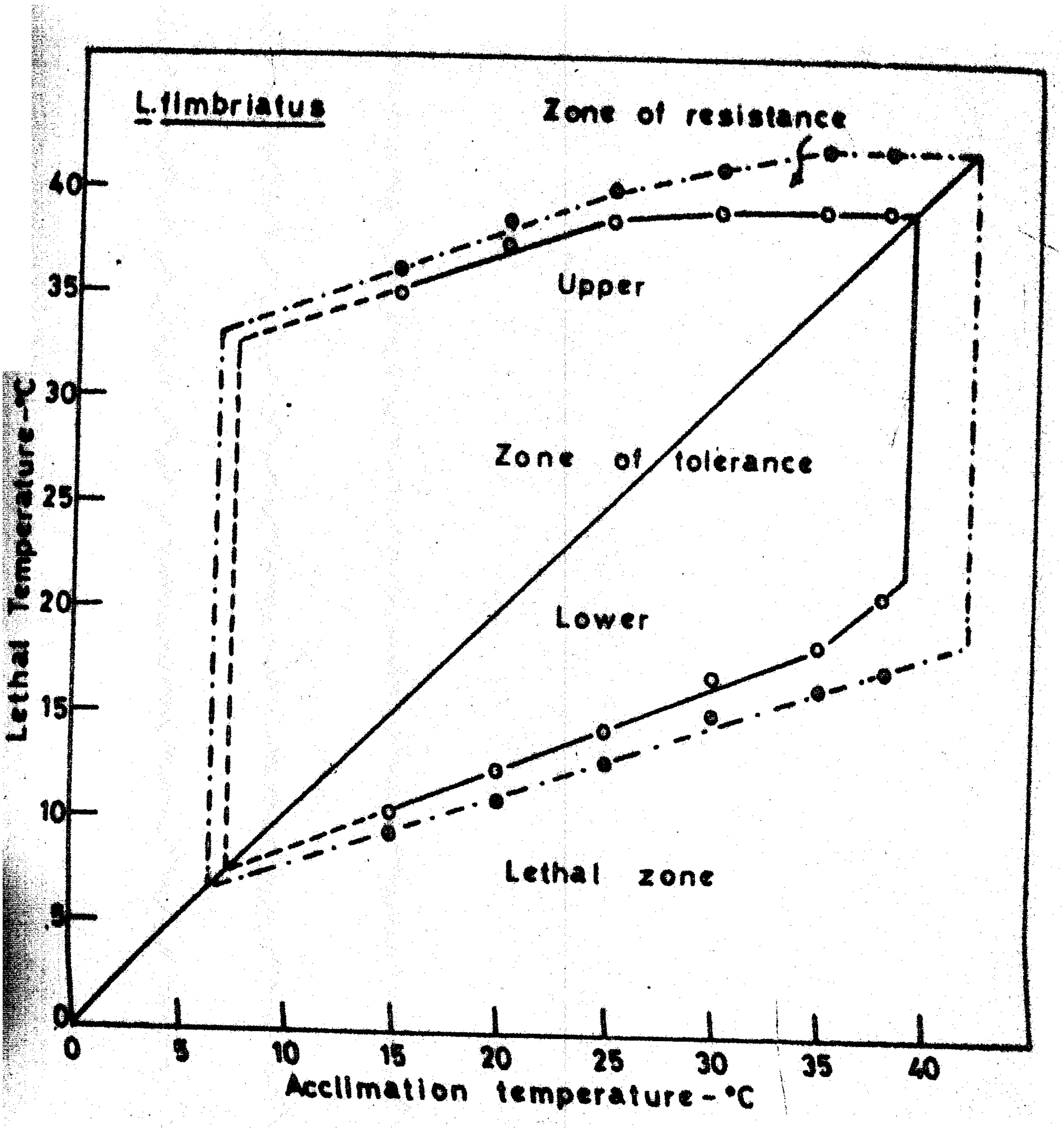

L. fimbriatus
Zone of resistance
Upper
Zone of tolerance
Lower
Lethal zone
Lethal Temperature - °C
Acclimation temperature - °C
0
5
10
15
20
25
30
35
40

Fig.44

Zones of thermal tolerance and resistance for common carp fry in fresh water

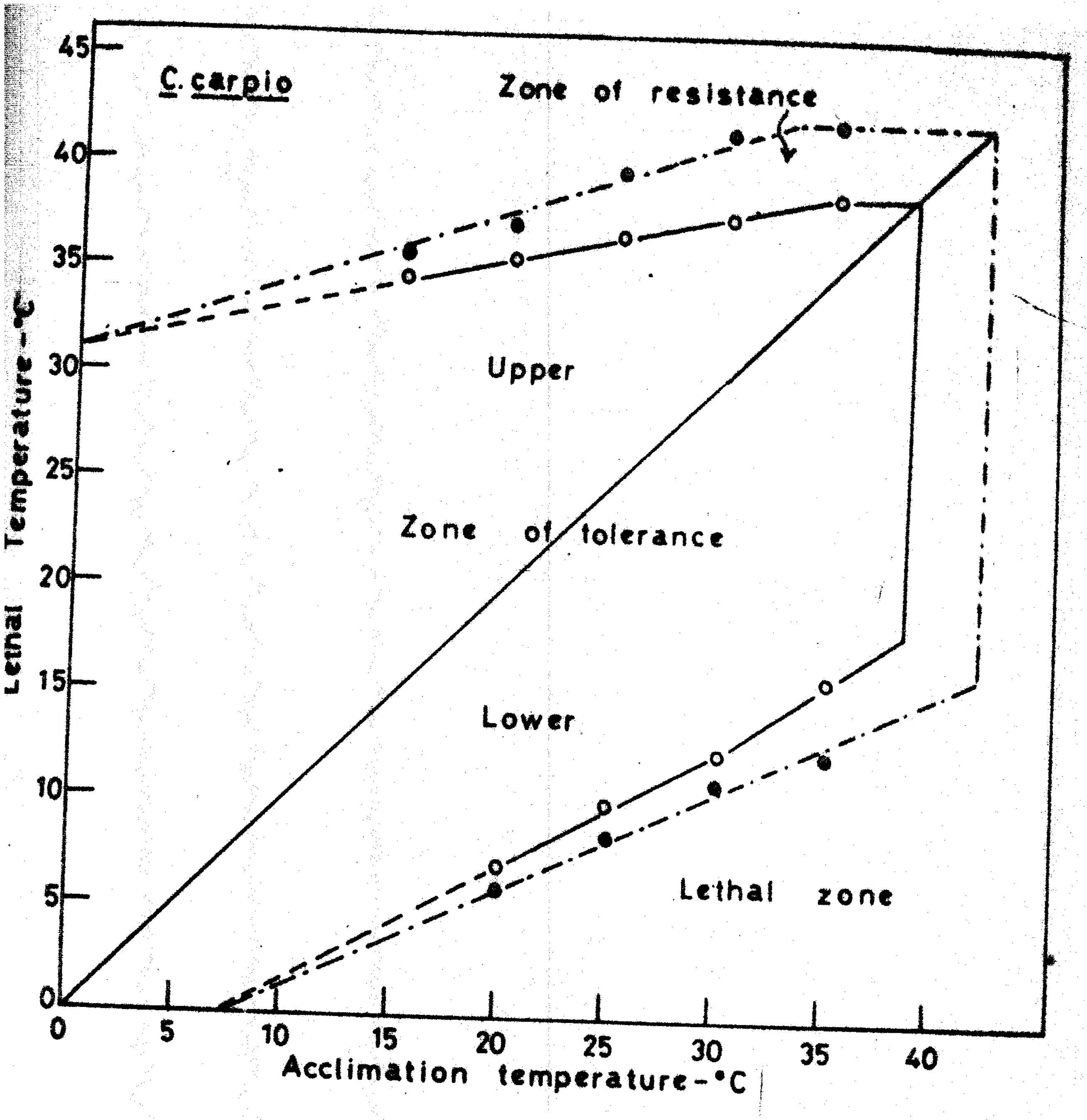
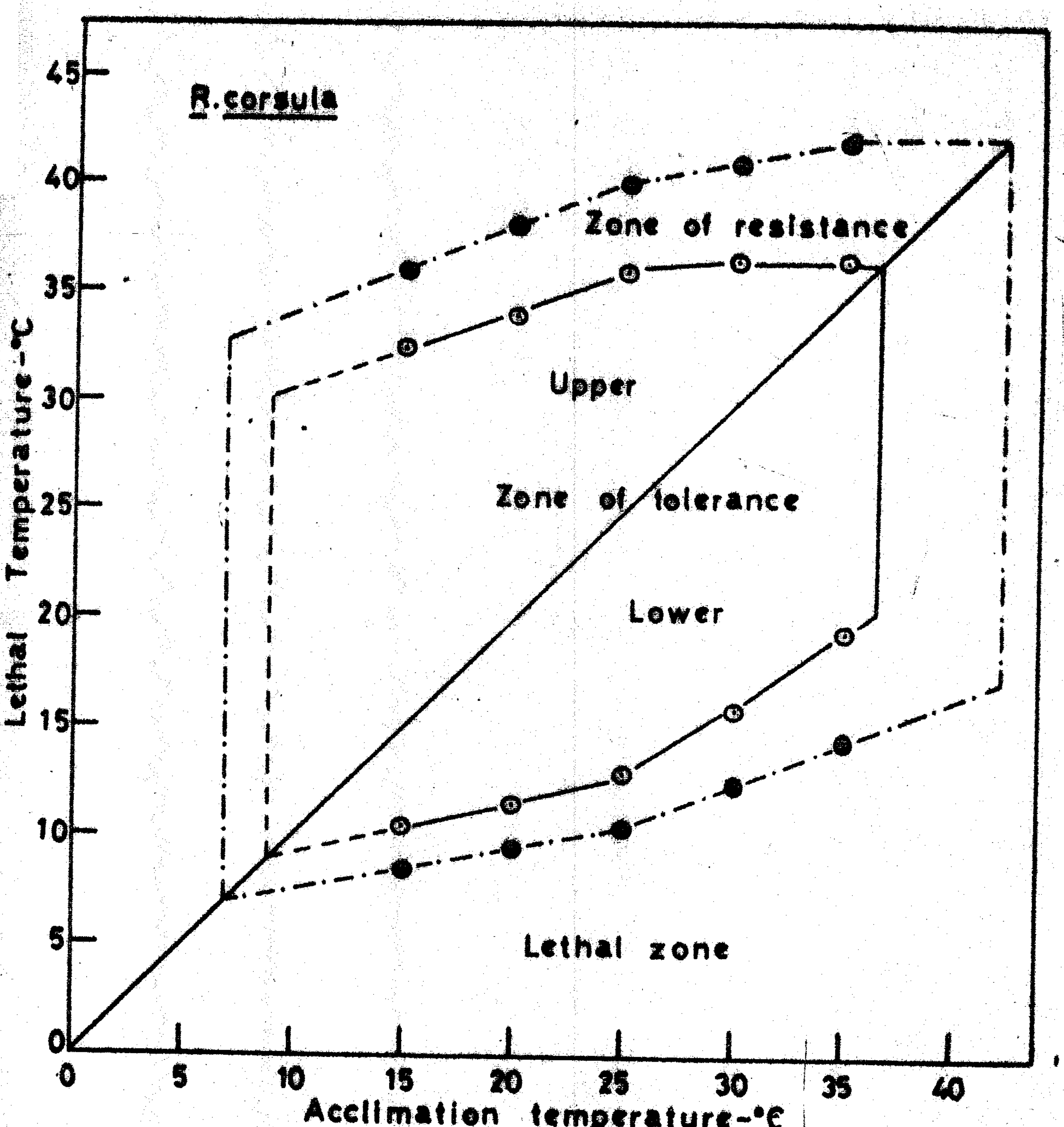

C. carpio
Zone of resistance
Upper
Zone of tolerance
Lower
Lethal zone
Lethal Temperature -°C
Acclimation temperature -°C
0
5
10
15
20
25
30
35
40
45

Fig.45

Zones of thermal tolerance and resistance for freshwater mullet fingerlings in fresh water


R. corsula
Zone of resistance
Upper
Zone of tolerance
Lower
Lethal zone
Lethal Temperature-°C
Acclimation temperature-°C
0
5
10
15
20
25
30
35
40
45

features of these illustrations, Fig.41 of mrigal can be taken as an example. The polygon which lies inside, bounded by continuous line connecting open circles, is the area of tolerance. Since acclimation was not done below 20°C for common carp (Fig.44) and 15°C for other four species (Figs.41, 42, 43 and 45) for low temperature experiments (see also 'Methods'), the tolerance area at low temperatures below 20 and 15°C acclimations for common carp and other species respectively is shown by extrapolation as indicated by broken line. It is felt that this extrapolation is acceptable to describe the zone of tolerance of the species adequately, since the lower incipient lethal temperatures of all the acclimations almost fall in a straight and sloping line, though there could be variations from this pattern. Further, it appears from the general nature of the plots shown in Figs.41 to 45, that the limitation in technique, as pointed out earlier, has not affected the expression of the zone of tolerance in all the five species.

The outer polygon which surrounds the zone of tolerance, indicated by the broken line connecting dots, describes the zone of resistance. It is realised that all area beyond the tolerance boundary depicts the resistance zone of the organisms, but beyond a point it would be difficult to maintain an observable time component. In view of this the boundary for the resistance area in this study was fixed as the curve passing through temperatures which cause immediate death to the fish exposed as observed from the experiments. Adoption of this approach of limiting the resistance area is in agreement with the practice accepted by earlier workers, Charlon (1968), Charlon et al., (1970) and Coutant (1970). As pointed out earlier, the 45° isotherm-line which runs diagonally through these polygons in Figs.41 - 45 cuts the zones of resistance and tolerance into 'upper' and 'lower' zones of resistance and tolerance of the species concerned (Charlon, 1968; Charlon et al., 1970; McErlean et al., 1969). The areas of the zones of thermal tolerance and resistance along with the thermal triangles (upper and lower thermal tolerance areas) in °C^2 are given in Table XXVIII for all the five species. The total areas of tolerance are 812, 850, 731, 1075 and 569°C^2 for mrigal, rohu, fringe-lipped carp, common carp and freshwater mullet respectively. For the same order of species the respective upper zones of tolerance are 481.0, 525.0, 434.0, 612.5 and 316.0°C^2 and the respective lower zones of tolerance are 331.0, 325.0, 294.0, 462.5 and 253.0°C^2. The total area of the zones of resistance are 152, 247, 197, 225 and 391°C^2 for mrigal, rohu, fringe-

Table XXVIII

Zones of thermal resistance, tolerance and triangles for carps and freshwater mullet; the difference (excess area) in thermal triangles due to truncation (see text) in zones of tolerance is also given (all units in $^{\circ}C^2$)

Species	Zone of resistance			Zone of tolerance			Zone of triangles		Difference
	Total	Upper	Lower	Total	Upper	Lower	Upper	Lower	
<u>C</u>. <u>mrigala</u>	152	64.5	87.5	812	481.0	331.0	500	331	19
<u>L</u>. <u>rohita</u>	247	116.0	131.0	850	525.0	325.0	537	325	12
<u>L</u>. <u>fimbriatus</u>	197	75.0	122.0	731	437.0	294.0	462	281	12
<u>C</u>. <u>carpio</u>	225	94.0	131.0	1075	612.5	462.5	-	-	-
<u>R</u>. <u>corsula</u>	391	181.0	210.0	569	316.0	253.0	331	263	25

lipped carp, common carp and freshwater mullet respectively. The upper zones of resistance are 64.5, 116.0, 75.0, 94.0 and 181.0°C^2 and the lower zones of resistance are 87.5, 131.0, 122.0, 131.0 and 210.0°C^2 for mrigal, rohu, fringe-lipped carp, common carp and freshwater mullet respectively.

These values suggest that the lower zones of resistance areas are larger than the areas of upper zones of resistance among the species studied. Whereas in the case of the upper and lower zones of tolerance this phenomenon is reversed. This inverse relation between the zones of resistance and tolerance is further discussed under 'Relative tolerance and resistance' (page 69).

The upper and lower ultimate lethal temperatures obtained from the thermal tolerance polygons (Figs.41 - 45, see also 'Methods') of all the five species are given in Table XXIX. The upper ultimate lethal temperatures (see 'Methods') are 39.4, 39.0, 39.8, 38.5 and 36.3°C for mrigal, rohu, fringe-lipped carp, common carp and freshwater mullet respectively and the respective lower ultimate lethal temperatures for the same order of species are 5.5, 3.5, 7.5, 0.0 and 9.0°C. Figure 44, describing the zone of thermal tolerance for common carp is unique among the species

Table XXIX

Upper and lower ultimate lethal temperatures derived from the thermal tolerance polygons among the five species investigated

Species	Ultimate lethal temperature	
	Upper	Lower
Cirrhinus mrigala	39.4	5.5
Labeo rohita	39.0	3.5
L. fimbriatus	39.8	7.5
Cyprinus carpio	38.5	0.0
Rhinomugil corsula	36.3	9.0

tested; where the extrapolated broken line at low temperature reaches 0°C even at 7°C acclimation level. As pointed out by Fry (1971), in many freshwater species the ultimate lower lethal temperature is indeterminate, since the fish can still be active at the freezing point of water. In the case of common carp, the formation of ice in the outer medium, or the formation of ice crystals in body fluids below 0°C may limit its existence. Hence, the lower ultimate lethal temperature for common carp can be taken as 0°C as in the case of goldfish (Fry et al., 1942) and spring and sockeye salmon (Brett, 1952). Thermal death at this point needs further exploration covering different aspects such as the freezing of blood (Pott and Morris, 1968).

Thermal triangles:

Thermal tolerance polygons obtained by plotting acclimation temperatures on X axis and the incipient lethal temperatures on Y axis are irregular geometric figures. Although the 'isotherm-line', constructed at 45° in the polygon, differentiates the upper and lower zones of thermal tolerance, it is understood that both are barely separable from each other. Presumably, the upper thermal tolerance area could be estimated by measuring the area bounded by the upper incipient lethal temperature plots, the Y axis and the isotherm-line and the rest of the area being the lower thermal tolerance area (Table XXVIII). However, these areas are usually irregular geometric figures and computation of these two would still require recourse to planimetry or other methods (McErlean et al., 1969).

McErlean et al., (1969) have given an alternate method for constructing the zone of upper thermal triangle with the available limited data. This involves the determination of the 'difference values' between the incipient lethal values and acclimation values for different acclimation temperatures of the species concerned and plotting such series of 'difference values' against the respective acclimation temperatures on Y and X axes respectively, as shown in Fig.46 for mrigal. Now, this results in a regular triangle and the validity of this method depends upon the linearity of the 'difference values' obtained. This kind of upper and lower thermal triangles are shown in Figs.47, 48, 49 for rohu, fringe-lipped carp and freshwater mullet respectively. In the case of common carp the triangles are not given, the reasons for omitting common carp are indicated separately below. The areas of upper thermal triangles are 500, 537, 462 and 331°C^2 and lower thermal

Fig.46

Upper and lower thermal tolerance triangles for mrigal in fresh water (see text for further explanation)

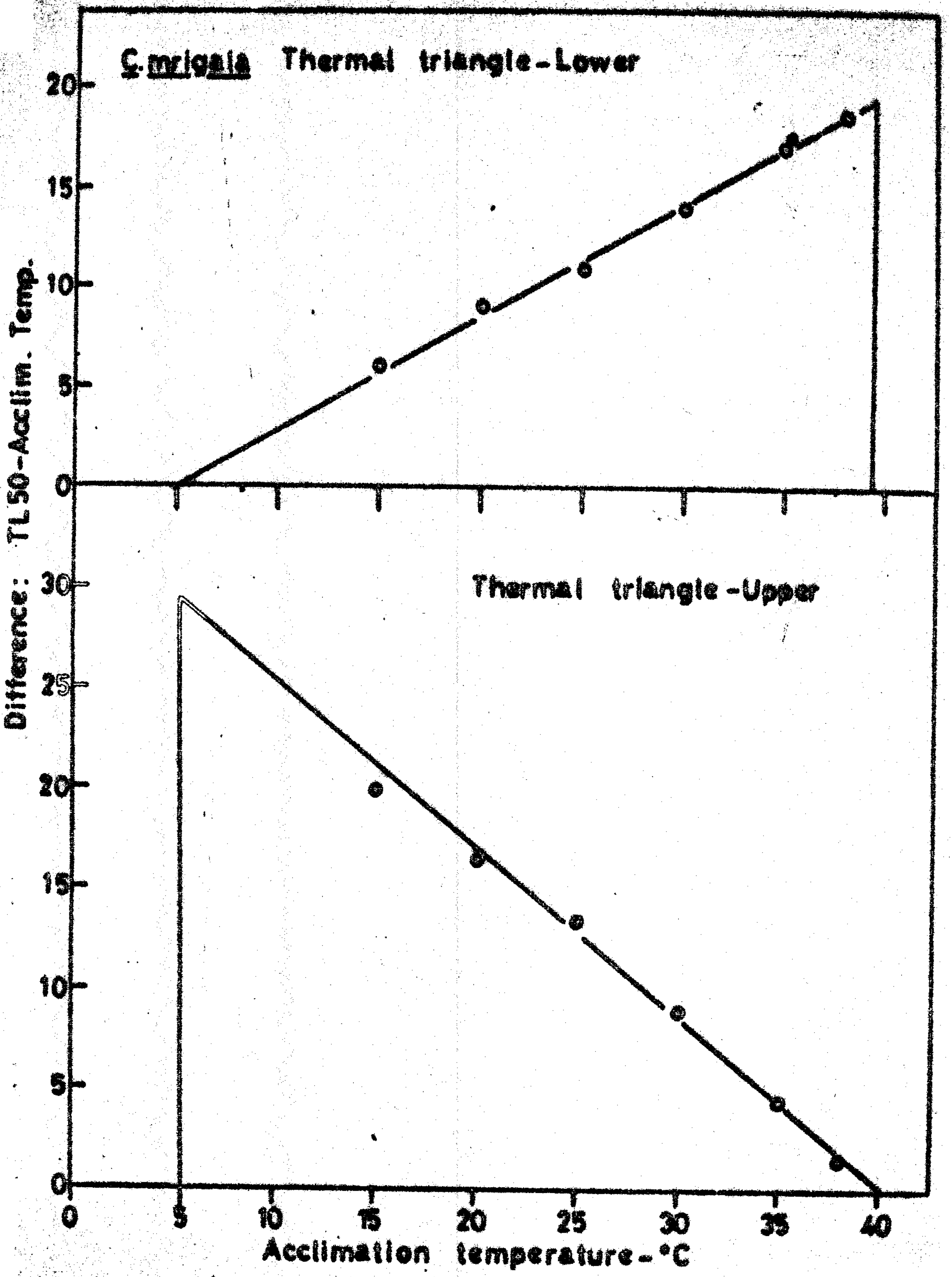

C. mrigala Thermal triangle-Lower
Thermal triangle-Upper
Difference: TL50-Acclim. Temp.
Acclimation temperature-°C
0
5
10
15
20
25
30
0
5
10
15
20
25
30
35
40

Fig.47

Upper and lower thermal tolerance triangles for rohu in fresh water

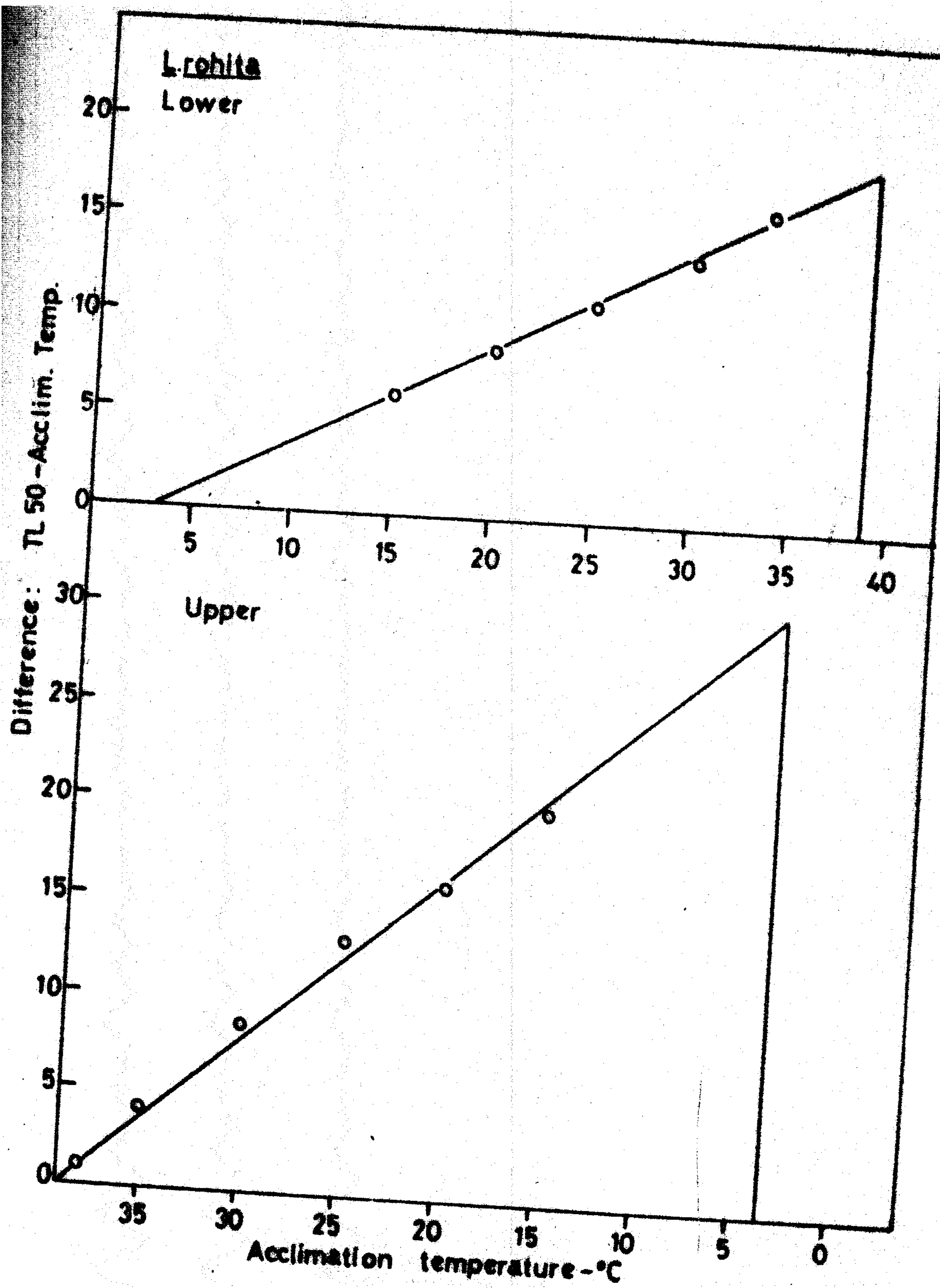

L. rohita
Lower
Upper
Difference: TL 50 - Acclim. Temp.
Acclimation temperature - °C
0
5
10
15
20
25
30
0
5
10
15
20
25
30
35
40
35
30
25
20
15
10
5
0

Fig.48

Upper and lower thermal tolerance triangles for fringe-lipped carp in fresh water

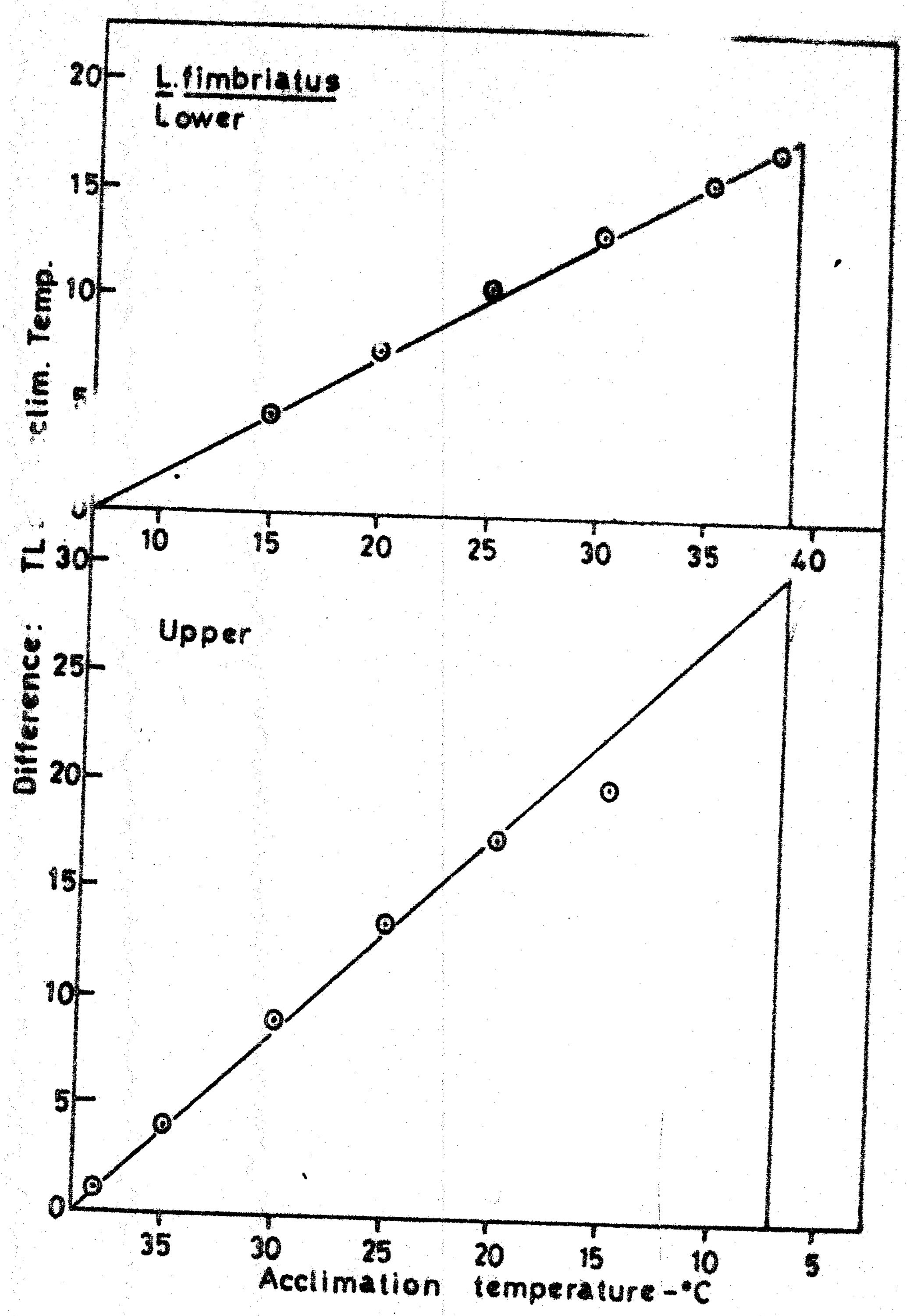
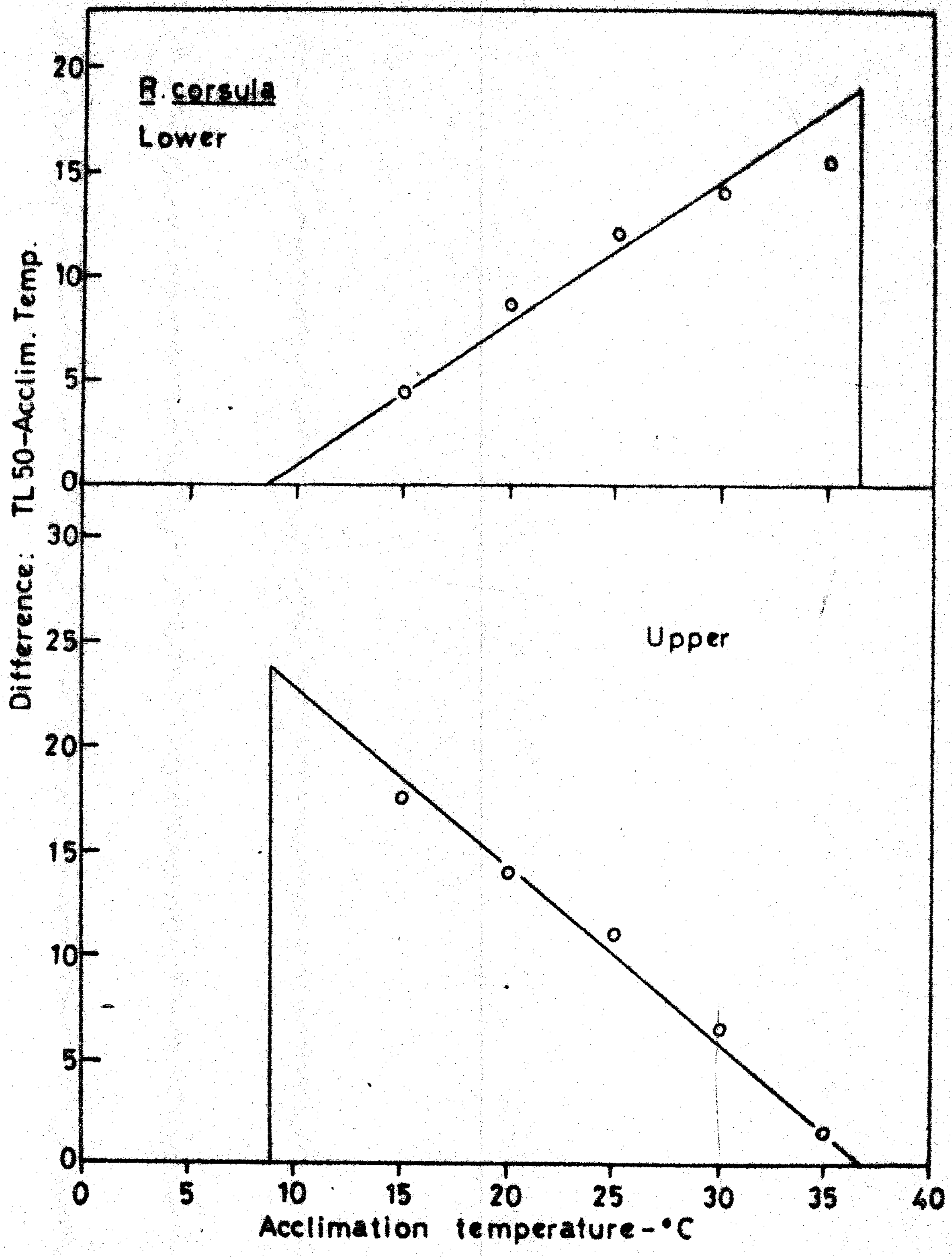

L. fimbriatus
Lower
Upper
Difference: TL clim. Temp.
20
15
10
5
0
30
25
20
15
10
5
0
10
15
20
25
30
35
40
35
30
25
20
15
10
5
Acclimation temperature -°C

Fig.49

Upper and lower thermal tolerance triangles for freshwater mullet in fresh water


R. corsula
Lower
Upper
Difference: TL 50-Acclim. Temp.
Acclimation temperature-°C
0
5
10
15
20
25
30
35
40

triangles are 331, 321, 281 and 263°C^2 for mrigal, rohu, fringe-lipped carp and freshwater mullet respectively (Table XXVIII). The areas of both upper and lower thermal tolerance are expressed as two independent values by these triangles and in addition, it is also possible to show the tolerance areas below and above ambient testing level. Further, it also permits comparison of upper and lower thermal tolerances among the species.

A possible limitation of this procedure (thermal triangles) is the over estimation of tolerance areas for species which have a 'plateau' and a 'floor' (Brett, 1952) in the upper and lower tharmal tolerance levels respectively. In the present study, the polygon of common carp has a 'floor' in the lower thermal tolerance level, which does not permit the construction of thermal triangles accurately. Likewise, the other four species have a 'plateau' in upper thermal tolerance area due to intolerance to high lethal temperatures among these species. The 'plateau' in these polygons is formed by the truncation of the linear extrapolation of the incipient lethal temperature values (Brett, 1952; McErlean _et_ _al_., 1969; Fry, 1971 and Pers.comm.,). In these cases the difference values between the incipient lethal temperatures and acclimation temperatures are smaller for higher acclimation levels than those for lower acclimations. The projection of the hypotenuese to the X axis forms an acute triangle at acclimation levels clearly outside the usual tolerance limit. Hence, the truncation shown in the polygons is not accounted in the extrapolation of thermal triangles. This phenomenon is inevitable to construct the triangles and the resultant differences (i.e., the excess area in the triangles) due to this phenomenon are also shown in Table XXVIII for all the four species. The excess areas indicated are narrow and inspite of this limitation it would appear that the thermal triangle is a useful concept especially in view the facility it gives in separating upper and lower thermal zones.

Relative tolerance and resistance:

The data on the area of zones of upper and lower thermal tolerance and resistance in $°C^2$ (Table XXVIII) can be further made use of by a study of relative tolerance and resistance of the species. The value can be expressed in percentage. In this concept the total area of tolerance or resistance is considered as 100% and the fractions of this total area are the upper and lower thermal tolerance or resistance. Thus the formulae used are :

1 $$\text{Relative tolerance (upper or lower)} = \frac{\text{Area of tolerance (upper or lower)}}{\text{Area of total tolerance}} \times 100$$

2 $$\text{Relative resistance (upper or lower)} = \frac{\text{Area of resistance (upper or lower)}}{\text{Area of total resistance}} \times 100$$

These values obtained on relative (percent) tolerance and resistance could be expected to show the difference in thermal tolerance and resistance areas among the species in a uniform scale. The upper and lower relative (percent) tolerance and resistance for the species presently studied are given in Table XXX. The values for upper relative tolerance are 59, 62, 60, 57 and 55.0 and those for lower relative tolerance are 41, 38, 40, 43 and 45.0 for mrigal, rohu, fringe-lipped carp, common carp and freshwater mullet respectively. The values of upper relative resistance are 42, 47, 38, 42 and 46 and those for lower relative resistance are 58, 53, 62, 58 and 54 for mrigal, rohu, fringe-lipped carp, common carp and freshwater mullet respectively.

The relative resistance for upper and lower temperatures indicates that the relative lower resistance is higher than the upper resistance uniformly for all the five species; whereas, in tolerance, the upper relative tolerance is highe

Table XXX

Upper and lower relative thermal tolerance and resistance (in %) for the five species presently studied and the upper and lower relative thermal tolerance (in %) for a tropic, a subtropic and six temperate species (see text for further explanation)

Species	Relative tolerance		Relative resistance		Author and year
	Upper	Lower	Upper	Lower	
Ophiocephalus punctatus	49	51	-	-	Ananthakrishnan and Kutty, 1976
Carassius auratus	48	52	-	-	Fry *et al.*, 1942
Ameiurus nebulosus	49	51	-	-	Brett, 1944
Rutilus rutilus	44	56	-	-	Cocking, 1959
Oncorhynchus nerka	54	46	-	-	Brett, 1952
O. tshawytscha	52	48	-	-	Brett, 1952
O. keta	55	45	-	-	Brett, 1952
O. kisutch	51	49	-	-	Brett, 1952
Cirrhinus mrigala	59	41	42	58	Present study
Labeo rohita	62	38	47	53	Present study
L. fimbriatus	60	40	38	62	Present study
Cyprinus carpio	57	43	42	58	Present study
Rhinomugil corsula	55	45	46	54	Present study

than the lower. This phenomenon indicates an inverse relation between tolerance and resistance (see also 'zones of tolerance and resistance' under 'Discussion'). This phenomenon, applied to total tolerance and resistance, is manifested markedly by the freshwater mullet, which has a comparatively smaller tolerance area (569°C^2) and a wider resis tance area (391°C^2); whereas in the case of carps, the zone of tolerance is much larger than the zone of resistance, as highly exhibited by common carp (1075 and 225°C^2 respectivel see also Table XXVIII). A similar parallel was drawn by Fry *et al*., (1942, 1946) for goldfish, *Carassius auratus* and Hart (1952) for Bullhead, *Ameiurus nebulosus*.

Among the very few tropical species subjected to the study of thermal response (Allanson and Noble, 1964; Kutty and Murugapoopathy, 1969; Ananthakrishnan and Kutty, 1974, 1976; Ananthakrishnan and Sreenivasan, 1977), the complete thermal requirement of the species to the level of 'zone of thermal tolerance' was not worked out until the work on *Ophiocephalus punctatus* by Ananthakrishnan and Kutty (1976). This air breathing fish has a tolerance area of 410°C^2 which is far less than that obtained for carps and freshwater mullet in the present study. This information available on tropical species indicates that the eurythermal carps, namely, mrigal, rohu, fringe-lipped carp and common carp, have higher tolerance area than the stenothermal freshwater mullet and the air breather O. punctatus. Further, this limited comparison among the tropical species is extended to include subtropical and temperate species also, not only due to the greater availability of literature on these species, but also to have further understanding on differential responses of a wider spectrum of fish species.

The 'percent tolerance' calculation used for the five species presently studied is applied for six temperate, a tropical and a subtropical species by measuring their upper and lower area of tolerance with the aid of planimeter and their 'relative thermal tolerance' (in %) are also given in Table XXX. The first four species Ophiocephalus punctatus, Carassius auratus, Ameiurus nebulosus and Rutilus rutilus have relative lower thermal tolerance higher than the relative upper thermal tolerance. But the remaining four species of Pacific salmon and five species presently studied have their relative upper thermal tolerance higher than the lower thermal tolerance.

As mentioned by Brett (1952), the freezing point of water limited the minimum acclimation to 0°C for Pacific salmons which inturn limited the area of tolerance and this is so for the subtropical species like goldfish (Fry et al., 1942) and common carp (present work) as well (Table XXIX). However, all the tropical species studied so far have minimum acclimations well above 0°C (Ananthakrishnan and Kutty, 1976; present work); since intolerance to low temperature results in higher low incipient lethal temperatures. As pointed out by Brett (1952), the relative low values of lower thermal tolerance among the Pacific salmons are due to their characteristic intolerance to low temperatures. This is true for all the species which have relatively low values of lower thermal tolerance area, irrespectively whether tropical, subtropical or even temperate. Whereas, this phenomenon is reversed among the species which have relatively higher values of lower thermal tolerance, as exhibited by Carassius auratus, Ameiurus nebulosus, Rutilus rutilus and Ophiocephalus punctatus. It is seen from these data that the geographic nativity does not seem to have any direct relacion to the relative upper and lower thermal tolerance among the species, as also indicated by the data on subtropical goldfish and common carp which have an inverse relation of upper and lower tolerance levels. In general, apart from these comparisons, each species has characteristic individuality with respect to upper and lower thermal tolerance which is species specific and can be considered as genetically controlled.

Proportionate gain or loss in tolerance:

Brett (1944, 1946) and various workers, in particular Doudoroff (1942) and Cocking (1959) have studied the rate at which fish adjust their lethal temperature in relation to a change in acclimation temperature. Following this principle, the proportionate gain in high temperature tolerance and loss in low temperature tolerance, can be calculated, with respect to every increase in acclimation temperature, by dividing the difference in high and low lethal levels due to acclimation, with the difference in respective acclimation levels The discussion of the present data has the limitation of low temperature acclimation as already referred to, but it is felt that this limitation does not seriously affect the trends in results discussed herein. Data on mrigal are discussed further, as an example, for computation.

The lowest and the highest temperatures for which mrigal were acclimated, are 15 and 38°C respectively and the difference or increase in acclimation temperature is 23°C (from 15 to 38°C). The respective high and low incipient lethal temperatures for the acclimations 38 and 15°C are 39.5 and 35.0°C and the difference in incipient lethal level is 4.5°C. Thus, the proportionate gain in high temperature is $= \frac{4.5}{23} =$ 0.19°C/1°C for mrigal. This value may be treated as an index of the rate of increase in tolerance for an elevation of 1°C in acclimation level with respect to high temperature. For low temperature the rate of loss in tolerance for every 1°C increase in acclimation is 0.46°C (i.e.,10.48/23).

The values on proportionate gain or loss in tolerance for all the five species presently studied are summarised in Table XXXI. The proportionate gain in tolerance are 0.19, 0.17, 0.17, 0.21 and 0.21°C/1°C for mrigal, rohu, fringe-lipped carp, common carp and freshwater mullet respectively and the proportionate loss in tolerance are 0.46, 0.52, 0.44, 0.60 and 0.45°C/1°C for the same order of species. As indicated by Fry (1971), typically the upper incipient lethal temperature changes approximately 1°C for a 3°C change in acclimation temperature for several species (Brett, 1944, 1946; Doudoroff, 1942). The lower incipient lethal shows a somewhat greater response, usually shifting

Table XXXI

Proportionate gain or loss in tolerance per degree centigrade raise in acclimation temperature among the four species of carp fry and fingerlings of freshwater mullet acclimated and tested to various temperatures

Species	Increase in Acclimation Temp. (°C)		Increase in tolerance (°C)	Decrease in tolerance (°C)	Proportionate gain in tolerance (°C/1°C)	Proportionate loss in tolerance (°C/1°C)
Temperature	High	Low	High	Low	High	Low
C. mrigala	23	23	4.52	10.48	0.19	0.46
L. rohita	23	20	3.99	10.46	0.17	0.52
L. fimbriatus	23	23	3.96	10.09	0.17	0.44
C. carpio	20	15	4.20	9.03	0.21	0.60
R. corsula	20	20	4.25	9.00	0.21	0.45

1°C for about 2°C change in acclimation temperature. The present data (Table XXXI) indicate that the change in lower incipient lethal temperatures apparently fall in line with above rate i.e. 1°C change for every 2°C change in acclimation level; whereas the upper incipient lethals approximately show a change of 1°C for about 5°C increase in acclimation temperature. Further, these values indicate that common carp exhibits the highest proportion of gain and loss in high and low temperatures and the values are 0.21 and 0.6°C/1°C respectively; whereas fringe-lipped carp has the lowest proportion of gain and loss i.e. 0.17 and 0.44°C/1°C respectively. The difference in proportionate gain in tolerance among the five species is not marked in high temperature; whereas in low temperature the difference among the species in proportionate loss is comparatively high. As already mentioned, the highest proportion of loss is exhibited by common carp followed by rohu and the lowest value by fringe-lipped carp.

Thus in agreement with earlier workers (Brett, 1944, 1946; Fry, 1971) present results also show that the proportionate loss at low temperature is higher than the proportionate gain at high temperatures uniformly among the five species studied. This higher response at low temperature is due to high intolerance to low temperature when compared to their better tolerance to high temperature among the species. As it is pointed out by Brett (1946) in goldfish, the upper lethal temperature is adjusted within a day when they are shifted from 20 to 28°C, while the shift from 24 to 16°C requires over 2 weeks for the reciprocal adjustment in Pimphales promelas (Brett, 1944). Further, as discussed by Fry (1971), it is not likely that these differences are the result of different species being used by the worker. Thus it appears that all the species have an innate, characteristic intolerance towards low temperatures and hence the consequent high proportionate values in low temperature response.

Comparison of thermal tolerance:

As pointed out earlier (see also 'Methods') the upper and lower incipient lethal values are the measure of tolerance level of the species concerned, since, these data are the basic requisite for the construction of thermal polygon which defines thermal tolerance graphically. Further, these data can be subjected to statistical analyses (see also 'Data analysis' under 'Methods') and the products of these analyses may be expected to define the significance of thermal tolerance among the species studied. In the presen to high intolerance to low temperature when compared to their better tolerance to high temperature among the species. As it is pointed out by Brett (1946) in goldfish, the upper lethal temperature is adjusted within a day when they are shifted from 20 to 28°C, while the shift from 24 to 16°C requires over 2 weeks for the reciprocal adjustment in *Pimphales promelas* (Brett, 1944). Further, as discussed by Fry (1971), it is not likely that these differences are the result of different species being used by the worker. Thus it appears that all the species have an innate, characteristic intolerance towards low temperatures and hence the consequent high proportionate values in low temperature response.

Comparison of thermal tolerance:

As pointed out earlier (see also 'Methods') the upper and lower incipient lethal values are the measure of tolerance level of the species concerned, since, these data are the basic requisite for the construction of thermal polygon which defines thermal tolerance graphically. Further, these data can be subjected to statistical analyses (see also 'Data analysis' under 'Methods') and the products of these analyses may be expected to define the significance of thermal tolerance among the species studied. In the present work, the upper and lower incipient lethal levels of all the five species have been subjected to analysis of variance and the results are summarised in Table XXXII. The values of 'F' ratio from this Table indicate a highly significant difference ($P < 0.01$) in tolerance among the species at both the treatments i.e., between species and between acclimations. Tables XXXIII and XXXIV constitute the breakdown of significant difference in upper and lower incipient lethal temperatures respectively, between species to single degree of freedom and a comparison for significance with respective error terms and degrees of freedom. The 'F' ratio values obtained by this orthogonal comparison, on the incipient lethals indicate a highly significant difference among the species at all the four orders of comparisons. This high significance ($P < 0.01$) may be taken as one of the important index to conclude that the tolerance to high and low temperatures is species specific.

The estimated area of thermal tolerance for the five species studied indicate that the common carp has the largest tolerance area of 1075°C^2 which is close to that of goldfish with an area of 1220°C^2 (Fry _et al._, 1942). In the case of other three carps, mrigal, rohu, and fringe-lipped carp the areas of tolerance are comparatively larger (812, 850

Table XXXII

Results from analysis of variance of incipient lethal temperatures of different acclimation for the five species

Incipient lethal Temperature	Species	Sum of squares	Degrees of freedom	Variance	Value of F.ratio	Value for F=0.05	Value for F=0.01
Upper	Between species (C.m.,L.r.,L.f.,C.c.,R.c.,)	23.27	4	5.817	54.88**	3.01	4.77
	Between acclimations (15, 20, 25, 30, 35°C)	57.22	4	14.304	134.94**	3.01	4.77
	Species x acclimations (error)	1.70	16	0.106			
	Total	82.19	24				
Lower	Between species (C.m.,L.r.,L.f.,C.c.,R.c.,)	48.94	4	12.24	36.34**	3.26	5.41
	Between acclimations (20, 25, 30, 35°C)	161.98	3	53.993	160.38**	3.49	5.95
	Species x acclimations (error)	4.04	12	0.337			
	Total	214.96	19				

** Highly significant

C.m. = *Cirrhinus mrigala*; L.r. = *Labeo rohita*;
L.f. = *Labeo fimbriatus* ; C.c. = *Cyprimus carpio*;
R.c. = *Rhinomugil corsula*

Table XXXIII

Comparison between each species of carps and freshwater mullet acclimated and tested to various temperatures. This constitutes the breakdown of significant difference in upper incipient lethal temperatures between species to single degree of freedom and a comparison for significance with the error term, 0.106 with 16 degrees of freedom recorded in Table XXXII

	Comparison (Orthogonal)	Variance	F. ratio	F (P = 0.05)	F (P = 0.01)
1	(*C.mrigala*-*L.rohita*)-(*C.carpio*-*R.corsula*)	14.11	133.13**	4.49	8.53
2	(*C.mrigala*-*L.rohita* - *C.carpio*-*R.corsula*) - (4 *L.fimbriatus*)	4.00	37.73**	4.49	8.53
3	(*C.mrigala* *C.carpio*)-(*L.rohita*-*R.corsula*)	3.70	34.88**	4.49	8.53
4	(*C.mrigala*-*R.corsula*)-(*L.rohita*-*C.carpio*)	1.46	13.75**	4.49	8.53

**Highly significant

Table XXXIV

Comparison between each species of carps and freshwater mullet for lower incipient lethal temperatures. This constitutes the results of the treatment as shown in Table XXXIII with comparison with error term, 0.337 with 12 degrees of freedom recorded in Table XXXII

	Comparison (Orthogonal)	Variance	F. ratio	F (P = 0.05)	F (P = 0.01)
1	(*C.mrigala*-*L.rohita*)-(*C.carpio*-*R.corsula*)	15.21	45.17**	4.75	9.33
2	(*C.mrigala*-*L.rohita*)-(*C.carpio*-*R.corsula*) - (4 *L.fimbriatus*)	8.19	24.33**	4.75	9.33
3	(*C.mrigala*-*C.carpio*)-(*L.rohita*-*R.corsula*)	15.62	46.39**	4.75	9.33
4	(*C.mrigala*-*R.corsula*)-(*L.rohita*-*C.carpio*)	9.92	29.47**	4.75	9.33

** Highly significant

and 731°C^2 respectively) than that of the freshwater mullet (569°C^2) but smaller than that of the common carp.

Zones of thermal tolerance have been worked out by various workers more for temperate and subtropical species (Fry *et al.*, 1942, 1946; Brett, 1944, 1952; Hart, 1947, 1952; Cocking, 1959; Hoff and Westman, 1966; Somero and DeVries, 1967) than for tropical ones (Ananthakrishnan and Kutty, 1976). Among these workers Hoff and Westmann (1966) recorded an area of 715°C^2 for *Menidia menidia*, a temperate species from Atlantic coast where the natural water temperature ranged from 0 to 32.5°C. Whereas Somero and DeVries (1967) have recorded an area of only 100°C^2 for *Trematonus* sp. from polar region (Antarctic) where the natural water temperature ranged from -3 to 5°C only. Among these two, the temperate *Menidia menidia* has been designated as 'Eurythermal' and the Antarctic *Trematomus* sp. as 'Stenothermal', since the former has comparatively wider tolerance area than the latter (Brett, 1970). In the present study, as already mentioned, the freshwater mullet has the lowest area of tolerance (569°C^2) which is close to that of *Ophiocephalus punctatus* (Ananthakrishnan and Kutty, 1976) with an area of 410°C^2 and the carps have comparatively larger area of tolerance. As per the above mentioned distinction, the carps can be termed as 'eurythermal' and the freshwater mullet as 'stenothermal' with respect to thermal tolerance.

Comparison of thermal resistance:

The source of variability among the population of experimental animals can roughly be divided into controlled and uncontrolled, as pointed out by Brett (1952). The controlled variables are acclimation temperature, lethal temperature and diet. The uncontrolled variables are sex, size and age. The experimental animals were fry and fingerlings and it was impossible to determine the sex readily. Sex difference on thermal tolerance has rarely been demonstrated even in mature fish (Hart, 1949), but Gibson (1954) has indicated a possible sex factor, but possibly in the early life stage in which the present tests are made sex may not have influenced the results.

Size difference has usually been associated with age difference and these influences were not analysed. Lack of any influence of size has been reported in the majority of experimental studies where high lethal temperatures have been determined (Tsukuda, 1960; Timet, 1963; Lewis, 1965). However, Brawn (1960) noted larger individuals being more susceptible than smaller ones and Spass (1960) an increase in the high lethal temperatures of 3 species of Salmonidae. Brett (1952) reported a greater susceptibility at low temperature among the smaller fish, Pacific salmon (<u>Oncorhynchus</u>) and not for high temperature. In the present work a preliminary random survey by scatter diagram analysis suggested a possible absence of size effect within the size range studied among all the five species.

The uncontrolled variables apart from those mentioned earlier are grouped as 'error' components. The smaller the error, the greater the chance for detecting the differences which might otherwise be masked by contaminating effects. Under this condition, the method of analysis of variance and the experimental design greatly help in making a comparison among the species (Yates, 1937; Fisher, 1945, 1948; Snedecor, 1967 and Brett, 1952). Since, almost the same acclimation and lethal test temperatures were used for each species, it is possible to consider the data for the following relations and the differences exhibited thereby, as indicated below:

1 Species x lethals for three levels of acclimations i.e., 25, 30 and 35°C. The results from three analyses of varianc of resistance times (logarithms) to various high lethal temperatures among carps and freshwater mullet are summa-

rised in Table XXXV. The results indicate a highly significant response between lethals ($P < 0.01$). At a lower level of acclimation for a comparable treatment of species the difference is more distinct. Whereas in between species the difference is not significant at higher acclimation levels i.e., 30 and 35°C ($P < 0.05$). On the other-hand, the difference between species and lethals at 25°C acclimation is highly significant ($P < 0.01$). Figure 50 displays the comparative thermal resistance curves to high lethal temperatures for carps and freshwater mullet acclimated to 25 and 35°C, which can be expected to illustrate the above mentioned relation. A further analysis to determine which species are contributing to the difference is considered later.

2 Species x acclimations for two levels of lethal temperatures. The results from analysis of variance for all the five species acclimated to 25, 30 and 35°C and tested at 39.0 and 39.5°C are presented in Table XXXVI. It can be inferred from these results that there is no significant difference between species at 39.0 and 39.5°C, but there is significant difference ($P < 0.05$) between acclimations at 39.0°C and a highly significant difference ($P < 0.01$) at 39.5°C.

Table XXXV

Results from analysis of variance of resistance times (logarithms) to various high temperatures among carps and freshwater mullet acclimated to different temperatures

Accli. Temp.	Comparison	Sum of squares	Degrees of freedom	Variance	Value of F.ratio	Value for F=0.05	Value for F=0.01
35°C	Between species (C.m., L.r., L.f., C.c., R.c.,)	53.7057	4	13.4264	1.46	3.01	4.77
	Between lethals (39.5, 40.0, 41.0, 40.5, 42.0°C)	1161.6670	4	290.4168	31.53**	3.01	4.77
	Species x lethals (error)	147.4800	16	9.2100			
	Total	1362.8527	24				

contd...

Table XXXV contd . . .

	Between species (C.m., L.r., L.f., C.c., R.c.,)	35.1888	4	8.7972	1.22	3.01	4.77
30°C	Between lethals (39.0, 39.5, 40.0, 40.5, 41.0°C)	1119.6028	4	279.9007	38.68**	3.01	4.77
	Species x lethals (error)	115.7698	16	7.2356			
	Total	1270.5614	24				
	Between species (C.m., L.r., C.c., L.f., R.c.,)	230.3245	4	57.5811	56.40**	3.84	7.01
25°C	Between lethals (38.5, 39.0, 39.5°C)	152.6827	2	76.3414	74.77**	4.46	8.65
	Species x lethals (error)	8.2399	8	1.0210			
	Total	391.2471	14				

** Highly significant

C.m. = *Cirrhinus mrigala*; L.r. = *Labeo rohita*;
L.f. = *Labeo fimbriatus* ; C.c. = *Cyprinus carpio*;
R.c. = *Rhinomugil corsula*

Fig. 50

Comparative median resistance times to high temperatures among the fishes acclimated to 25 and 30°C. Plotted on arithmetic x logarithmic axes

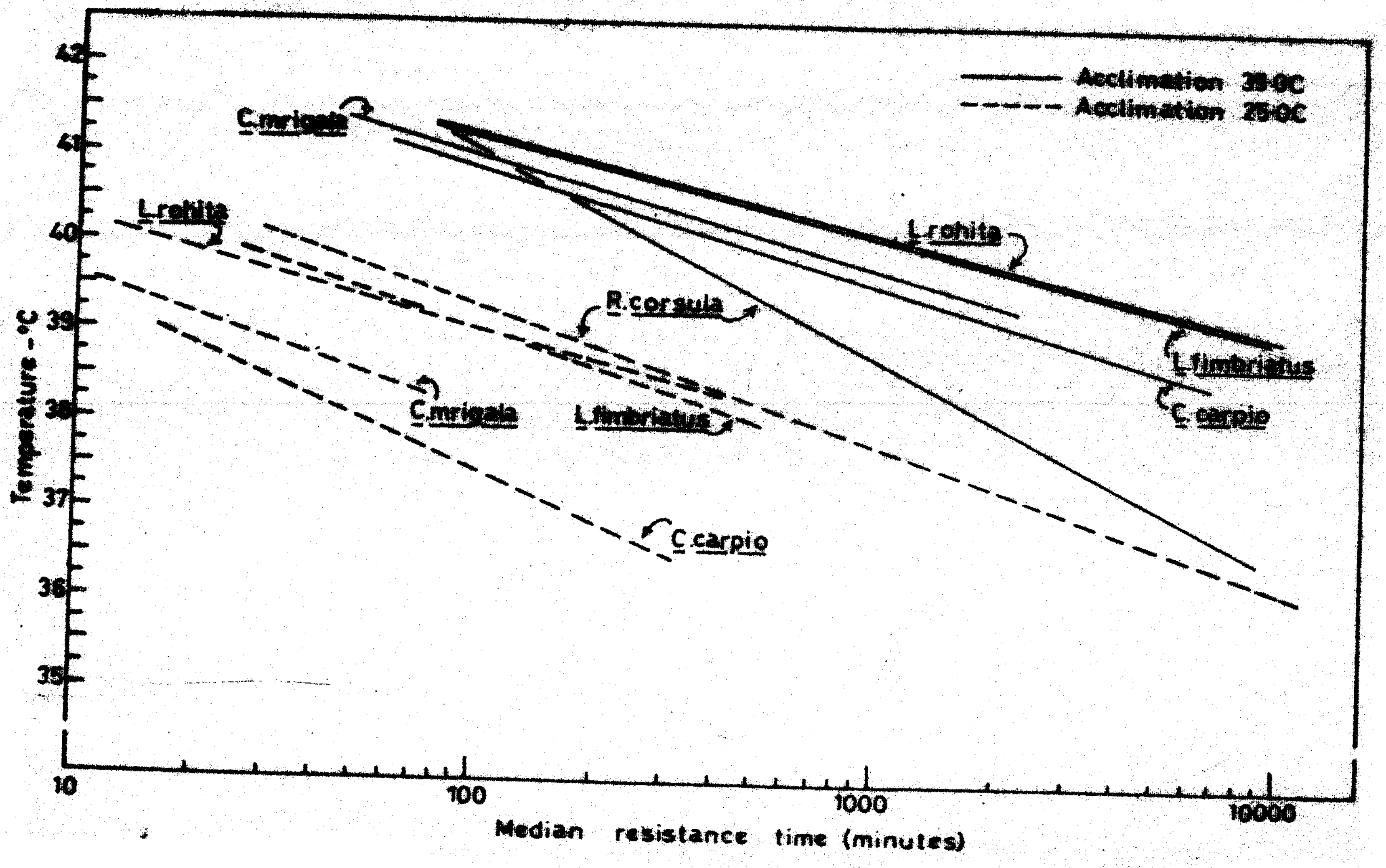

Acclimation 35·0C
Acclimation 25·0C
C.mrigala
L.rohita
R.corsula
C.mrigala
L.fimbriatus
C.carpio
L.rohita
L.fimbriatus
C.carpio
Temperature -°C
42
41
40
39
38
37
36
35
10
100
1000
10000
Median resistance time (minutes)

Table XXXVI
Results from analysis of variance of resistance times (logarithms) for carps and freshwater mullet acclimated to 25, 30 and 35°C and tested at 39.5 and 39.0°C

Lethal Temp.	Comparison	Sum of squares	Degrees of freedom	Variance	Value of F. ratio	Value for F. 0.05	Value for F = 0.01
39.5°C	Between species (C.m.,L.r.,L.f.,C.c.,R.c.,)	80.3310	4	20.0828	2.31	3.84	7.01
	Between acclimations (25, 30, 35°C)	1168.2314	2	584.1157	67.16**	4.46	8.65
	Species x acclimations (error)	69.5836	8	8.6980			
	Total	1318.1460	14				
39.0°C	Between species (L.f.,C.c.,R.c.,)	81.1475	2	40.5738	1.98	6.94	18.00
	Between acclimations (25, 30, 35°C)	394.6495	2	197.3248	9.65*	6.94	18.00
	Species x acclimations (error)	81.8327	4	20.4582			
	Total	557.6297	8				

* Significant
** Highly significant

C.m. = _Cirrhinus mrigala_; L.r. = _Labeo rohita_;
L.f. = _Labeo fimbriatus_ ; C.c. = _Cyprinus carpio_;
R.c. = _Rhinomugil corsula_

3 Lethals x acclimations for all the five species studied. The results of all these five analyses of variance are summarised in Table XXXVII. Briefly, these results confirm what has already been stated. Temperatures of both acclimation and lethal levels have a highly significant effect on resistance in 'each' of these species. The former two Tables (Tables XXXV and XXXVI) are only applicable to the 'group' as a whole. All the five species are highly significant between acclimations and lethals except for one species, fringe-lipped carp which does not show ($P < 0.05$) the same degree of significance between acclimations as the others ($P < 0.01$).

In the second order of comparison, the total variance, i.e., sum of squares (see also 'Data analysis' under 'Methods') pertaining to four degrees of freedom from Table XXXV have been broken down to allot a particular amount of variation to each degree of freedom. These values may be considered to assist in each independent comparison (orthogonal comparison). As in the case of earlier studies (Brett, 1952), the difference in temperature response among the species studied resulted in the use of different lethal limits of temperatures to a considerable extent to connect the cases from non-lethal to fairly rapid lethality and this factor

Table XXXVII

Results from analysis of variance of resistance times (logarithms) to various test temperatures at different acclimations for four species of carps and fres mullet

Species	Comparison	Sum of squares	Degrees of freedom	Variance	Value of F.ratio	Value for F=0.05
C.mrigala	Between lethals (39.5, 40.0, 40.5, 41.0°C)	463.1686	3	154.3895	23.35**	4.76
	Between acclimations (30, 35, 38°C)	213.4926	2	106.7463	16.14**	5.14
	Lethals x acclimations (error)	39.6727	6	6.6121		
	Total	716.3339	11			
L.rohita	Between lethals (39.5, 40.0, 40.5, 41.0°C)	578.4127	3	192.8042	24.61**	4.76
	Between acclimations (30, 35, 38°C)	269.4494	2	134.7247	29.47**	5.14
	Lethals x acclimations (error)	47.0106	6	7.8351		

Table XXXVII contd

L. fimbriatus	Between lethals (39.0,39.5,40.0,40.5,41.0°C)	521.6700	4	130.4175	9.63**	3.84	7.01
	Between acclimations (30, 35, 38°C)	200.2870	2	100.1435	7.39*	4.46	8.65
	Lethals x acclimations (error)	108.3870	8	13.5484			
	Total	830.3440	14				
C. carpio	Between lethals (38.5, 39.0, 39.5°C)	45.0033	2	22.5017	315.04**	6.94	18.00
	Between acclimations (25, 30, 35°C)	784.7293	2	392.3647	5493.38**	6.94	18.00
	Lethals x acclimations (error)	0.2854	4	0.0714			
	Total	830.0180	8				
R. corsula	Between lethals (37.0,38.0,38.5,39.0,39.5, 40.0°C)	482.1060	5	96.4212	41.58**	3.33	5.64
	Between acclimations (25, 30, 35°C)	50.9400	2	25.4700	10.98**	4.10	7.56
	Lethals x acclimations (error)	231.8980	10	2.3190			
	Total	764.9440	17				

* Significant

** Highly significant

reduced these values for uniform comparison below a feasible quantum in present work.

The results obtained by these analyses concerning species x lethals for acclimation 25, 30 and 35°C are summarised in Tables XXXVIII, XXXIX and XL respectively for orthogonal comparison. The values, for non-orthogonal comparison, obtained from variance analyses are summarised in Tables XXXVIIIa, XXXIXa and XLa for acclimations 25, 30 and 35°C respectively. The results obtained on orthogonal and non-orthogonal comparison from these six Tables permit to make the following conclusions:

a. No significant difference in response to upper lethal temperatures exists between all the five species at any level of comparison for higher acclimations of 30 and 35°C (Tables XXXIX, XXXIXa, XL and XLa).

b. There are significant differences in response among the species at 25°C acclimation level (Tables XXXVIII and XXXVIIIa). The orthogonal comparison shown in Table XXXVIII indicates a highly significant difference ($P < 0.01$) as shown by the four different comparative combinations among the five species studied. In the non-orthogonal comparison (Table XXXVIIIa), mrigal exhibits a moderately significant

Table XXXVIII

Comparison between each species of carps and freshwater mullet acclimated to 25°C and subjected to various lethal test temperatures from 38.5 to 39.5°C. This constitutes a breakdown of the significant difference between species to single degree of freedom and a comparison for significance with the error tèrm 1.021, with 8 degrees of freedom, recorded in Table XXXV

	Comparison (Orthogonal)	Variance	F. ratio	F (P = 0.05)	F (P = 0.01)
1	(C.mrigala-L.rohita)-(C.c rpio-R.corsula)	13.82	13.53**	5.32	11.26
2	(C.mrigala-L.rohita - C.carpio-R.corsula) (4 L.fimbriatus)	26.77	26.22**	5.32	11.26
3	(C.mrigala-C.carpio)-(L.rohita-R.corsula)	149.31	146.24**	5.32	11.26
4	(C.mrigala-R.corsula)-(L.rohita-C.carpio)	40.52	39.69**	5.32	11.26

**Highly significant

able XXXVIIIa

omparison between each species of carps and freshwater mullet acclimated to 25°C and subjected to various high lethal temperatures. This constitute a reakdown of the significant difference between species to single degree of reedom and a comparison for significance with the error term 1.021 with 8 egrees of freedom recorded in Table XXXV

	Comparison (non-orthogonal)	Variance	F. ratio	F (P = 0.05)	F (P = 0.01)
1	(*C.mrigala* - *L.rohita*)	10.2785	10.06*	5.32	11.26
2	(*C.mrigala* - *L.rohita*) - (2 *L. fimbriatus*)	23.3062	22.83**	5.32	11.26
3	(*C. carpio* - *R. corsula*) - (2 *L. fimbriatus*)	6.1343	6.01*	5.32	11.26
4	(*C.carpio* - *R.corsula*)	103.6203	101.49**	5.32	11.26

* Significant

** Highly significant

Table XXXIX

Comparison between each species of carps and freshwater mullet acclimated to 30°C and subjected to various lethal test temperatures from 39 to 41°C. This constitutes a breakdown of the significant difference between species to single degree of freedom and a comparison for significance with the error term 7.2356 with 16 degrees of freedom recorded in Table XXXV

	Comparison (Orthogonal)	Variance	F. ratio	F (P = 0.05)	F (P = 0.01)
1	(*C.mrigala*-*L.rohita*)-(*C.carpio*-*R.corsula*)	16.81	2.32	4.49	8.53
2	(*C.mrigala*-*L.rohita*.- *C.carpio*-*R.corsula*) (4 *L. fimbriatus*)	16.57	2.29	4.49	8.53
3	(*C.mrigala*-*C.carpio*)-(*L.rohita*-*R.corsula*)	01.79	0.25	4.49	8.53
4	(*C.mrigala*-*R.corsula*)-(*L.rohita*-*C.carpio*)	00.02	0.00	4.49	8.53

Table XXXIXa

Comparison between each species of carps and freshwater mullet acclimated to 30°C and subjected to various high lethal temperatures. This constitutes a breakdown of the significant difference between species to single degree of freedom and a comparison for significance with the error term 7.2356 with 16 degrees of freedom recorded in Table XXXV

	Comparison (non-orthogonal)	Variance	F. ratio	F (P = 0.05)	F (P = 0.01)
1	(*C.mrigala* - *L.rohita*)	1.1064	0.1529	4.49	8.53
2	(*C.mrigala* - *L.rohita*) - (2 *L. fimbriatus*)	4.1688	0.5762	4.49	8.53
3	(*C.carpio* - *R.corsula*) - (2 *L. fimbriatus*)	29.0439	4.0140	4.49	8.53
4	(*C.carpio* - *R. corsula*)	0.7080	0.0979	4.49	8.53

Table XL

Comparison between each species of carps and freshwater mullet acclimated to 35°C and subjected to various lethal test temperatures from 39.5 to 42°C. This constitutes a breakdown of the significant difference between species to single degree of freedom and a comparison for significance with the error term 9.21 with 16 degrees of freedom recorded in Table XXXV

	Comparison (orthogonal)	Variance	F. ratio	F (P = 0.05)	F (P = 0.01)
1	(*C.mrigala*-*L.rohita*)-(*C.carpio*-*R.corsula*)	32.39	3.52	4.49	8.53
2	(*C.mrigala*-*L.rohita* - *C.carpio*-*R.corsula*)- (4 *L.fimbriatus*)	19.12	2.08	4.49	8.53
3	(*C.mrigala*-*C.carpio*)-(*L.rohita*-*R.corsula*)	1.81	0.20	4.49	8.53
4	(*C.mrigala*-*R.corsula*)-(*L.rohita*-*C.carpio*)	0.38	0.04	4.49	8.53

Table XLa

Comparisons between each species of carps and freshwater mullet acclimated to 35°C and subjected to various high lethal temperatures. This constitutes a breakdown of the significant difference between species to single degree of freedom and a comparison for significance with the error term 9.21 with 16 degrees of freedom recorded in Table XXXV

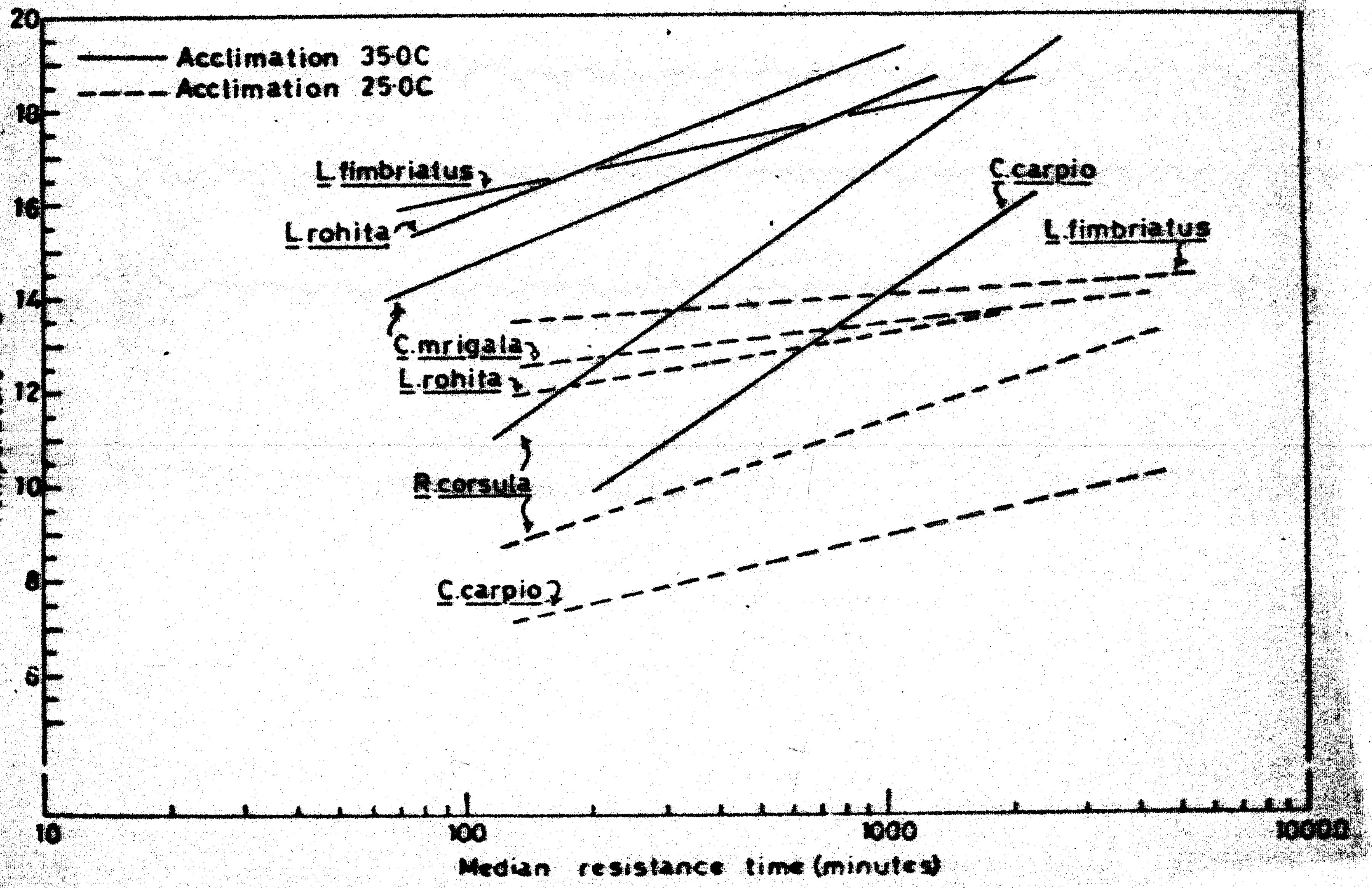
	Comparison (non-orthogonal)	Variance	F. ratio	F (P = 0.05)	F (P = 0.01)
1	(*C.mrigala* - *L.rohita*)	1.9278	0.2093	4.49	8.53
2	(*C.mrigala* - *L.rohita*) - (2 *L.fimbriatus*)	2.7829	0.3022	4.49	8.53
3	(*C.carpio* - *R.corsula*) - (2 *L.fimbriatus*)	39.8820	4.3300	4.49	8.53
4	(*C.carpio* - *R.corsula*)	0.2651	0.0288	4.49	8.53

difference ($P < 0.05$) in response from rohu; whereas common carp shows a highly significant difference ($P < 0.01$) from freshwater mullet. Further, the common carp and freshwater mullet exhibit a barely significant difference in response from fringe-lipped carp ($P < 0.05$); but mrigal and rohu show a highly significant difference ($P < 0.01$) from fringe-lipped carp. As already indicated, the regression curves pertaining to resistance response of all the species, for 25 (broken lines) and 35°C acclimations (continuous lines) shown in Fig.50 may be expected to illustrate the above mentioned points.

As for the resistance times to low lethal temperatures no systematic analysis of the data is possible due to the lack of consistency in response to low temperatures, as has also been observed in the case of Pacific salmons (Brett, 1952). The differences in low temperature response among the species resulted in the use of different lethal levels of low temperatures, to link the cases from non-lethal to fairly sharp lethality. This factor formed further limitation for the analysis of the data on low temperature. The regression curves pertaining to resistance response of all the species acclimated to 25 (broken lines) and 35°C (continuous lines) are collectively given in Fig.51 and this graphic presentation may give a general idea in place of

Fig.51

Comparative median resistance times to low lethal temperatures among the species shown, acclimated to 25 and 30°C. Plotted on arithmetic x logarithmic axes



Acclimation 35·0C
Acclimation 25·0C
L. fimbriatus
L. rohita
C. mrigala
L. rohita
R. corsula
C. carpio
C. carpio
L. fimbriatus
20
18
16
14
12
10
8
6
10
100
1000
10000
Median resistance time (minutes)

the statistical analysis in understanding the differential response of the species studied to a certain extent. It is seen from this figure, that the curves of the three tropical carps, mrigal, rohu and fringe-lipped carp lie close to each other and fall above the curves of freshwater mullet and common carp at both the acclimation levels. The curves of common carp occupy the base and just above this lie the curves of freshwater mullet. Apparently, there does not seem to exist a differential response at low temperatures, as in the case of high temperatures (see Fig 50 and Table XXXV). Further, it is seen from Fig.51 the resistance curves of common carp and freshwater mullet look distinctly different from the other three tropical carps. These differences in the response curves of all the species studied may be taken as the index of their differential response to low lethal temperatures.

Exposure time and lethal temperatures:

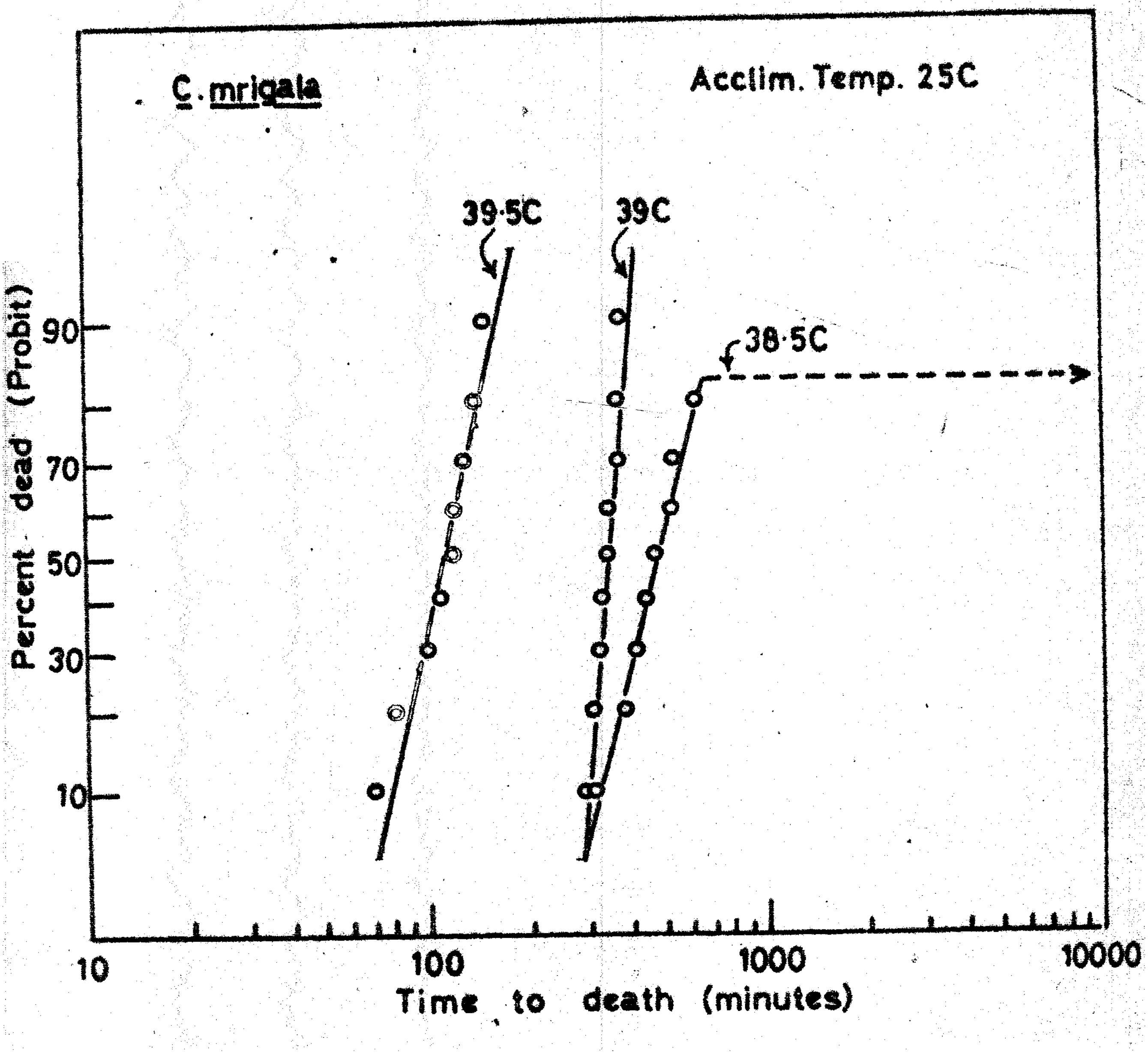
Various reports on functional responses to extremes of temperatures have been summarised by Heilbrunn (1943), Altman and Dittmer (1966), Charlon (1968), Brett (1970) and Fry (1971). Among them, Heilbrunn, as early as in 1943, pointed out the importance of the time factor when measuring heat or cold death. Subsequently, Brett (1952) indicated that the measure of threshold level of temperature below or above which an organism either lives or dies alone will not explai the thermal relation vividly, whereas the outcome of both temperature and exposure time gives a clear insight into this field.

As the lethal level of temperature is lowered in high temperature or increased in low temperature there is a characteristic rate of mortification (Fry et al., 1946) which may be influenced to a considerable extent by acclimation. This rate of mortification becomes zero at a particular threshold level if the experiment is continued for a longer time duration with the view to consider the time factor. Mortality, due to temperature as a primary cause, may not be present beyond this threshold level.

In the present work, as pointed out earlier (see 'Methods') a period of 10,000 min in high temperature experiments and 8,500 min in low temperature levels were found to be sufficient to indicate the threshold levels where the mortality would be zero. As already mentioned in 'Methods' various workers have used different exposure times. Initially, Doudoroff (1945) raised the question whether a 14 hr perio is of sufficient length to give a measure of thermal tolerance. Subsequently, Fry et al., (1946) have discussed

Fig.52

Times to death at various high lethaltest temperatures among mrigal fry acclimated to 25°C. Plotted on probit x logarithmic axes


C. mrigala
Acclim. Temp. 25C
39·5C
39C
38·5C
Percent dead (Probit)
90
70
50
30
10
10
100
1000
10000
Time to death (minutes)

the time of exposure (in the appendix of their paper) with the data on goldfish (Fry, Brett and Clawson, 1942) and have concluded that the 14 hr period was ample. Brett (1944) shortened this period to 12 hrs, but subsequently he has conducted experiments for Pacific salmons as long as seven days (10,000 min) for high temperatures and 5,000 min for low temperatures (Brett, 1952). Allen and Strawn (1968) accept heat death as being complete by 20,000 min although the fish were apparently not able to live indefinitely beyond that period since their food intake could not meet their maintenance requirements. However, the mortality among the sample due to temperature as the primar cause ceases after a particular period of resistance time in many animals.

In the present study, the resistance response of carps acclimated to high temperatures (i.e., 30, 35 and 38°C) and tested to high lethal temperatures indicates the presence of mortality among the samples as long as 10,000 min. This can be seen from Fig. 52 shown as an example, which presents the data of mrigal acclimated to 30°C and tested to various high lethal temperatures (see also Appendices from Aa1 to Ad5). Whereas for lower acclimations (i.e., 15, 20 and 25°C) the data indicate the absence of mortality among the lots even before 1000 min though the experiments were continued upto 10,000 min and Fig.53 presented as an example illustrates this response with the data of mrigal acclimated to 25°C (see also Appendices from Aa1 to Ad5). In the case of freshwater mullet, unlike in carps, the response is almost uniform, which is indicated by the presence of mortality upto 10,000 min irrespective of the acclimation level. Figures 54 and 55 pertaining to 25 and 30°C acclimated mullet respectively illustrate this phenomenon (see also Appendices from Ae1 to Ae5). The former phenomenon exhibited by the carps results in the entry into the zone of tolerance at lower acclimation temperatures and in a lower exposure time and has been reported to be present among *Girella nigricans* and *Fundulus parvipinnis* (Doudoroff, 1942, 1945), *Salvellinus fontinalis* and *Carassius auratus* (Fry *et al.*, 1946, 1942). As obvious from above phenomenon, the effective duration of the resistance time at a given lethal temperature depends on the thermal history of the fish; a precise knowledge of which is most essential for understanding thermal performance of fish.

The range of temperature within which no more death are likely to occur could be inferred from the figures of zones of thermal tolerance for the five species studied (see

Fig.53

Times to death at various high lethaltest temperatures among mrigal fry acclimated to 30°C. Plotted on probit x logarithmic axes

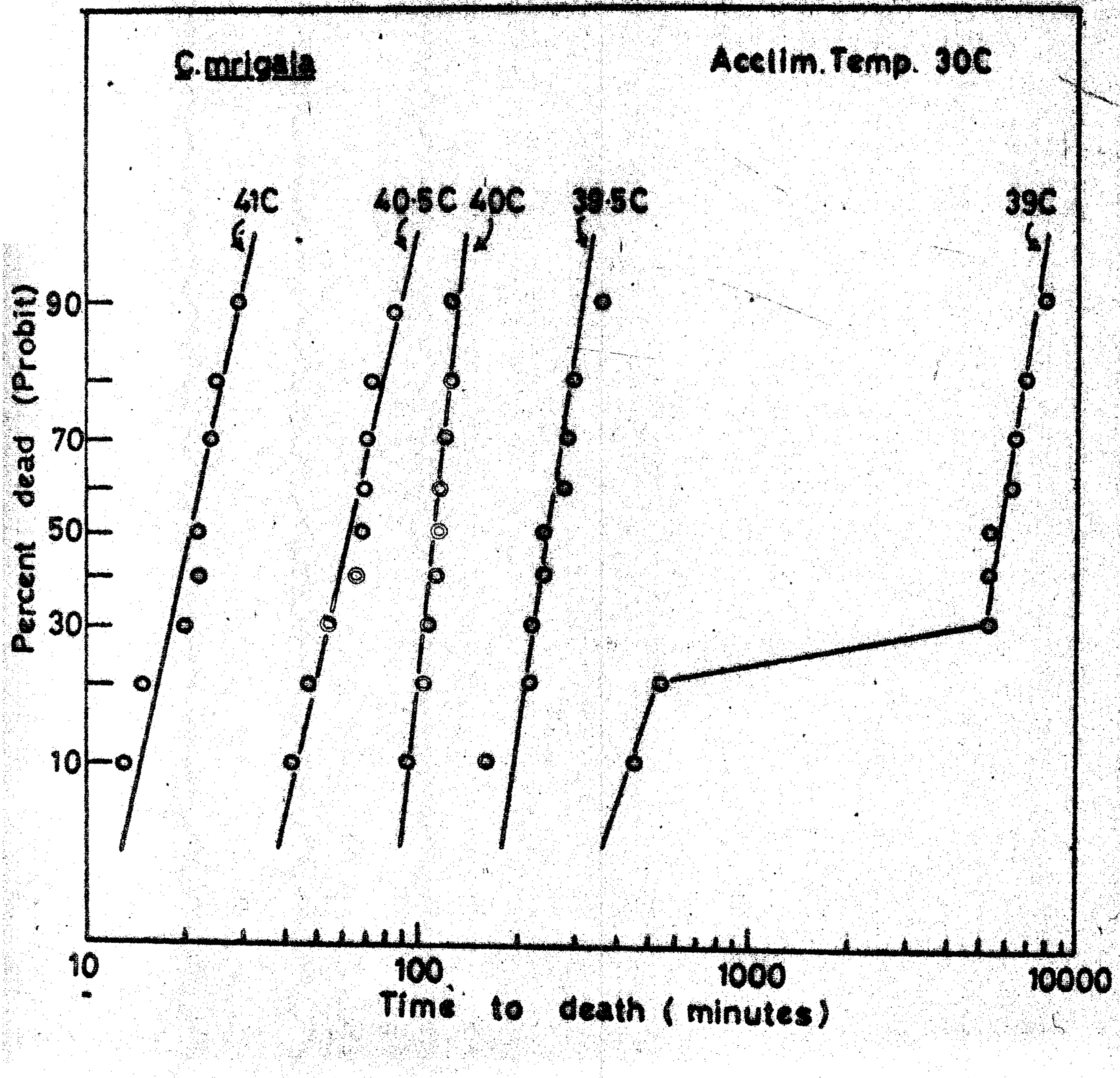

C. mrigala
Acclim. Temp. 30C
41C
40·5C
40C
39·5C
39C
Percent dead (Probit)
90
70
50
30
10
10
100
1000
10000
Time to death (minutes)

Fig.54

Times to death at various high lethaltest temperatures among freshwater mullet fingerlings acclimated to 30°C. Plotted on probit x logarithmic axes

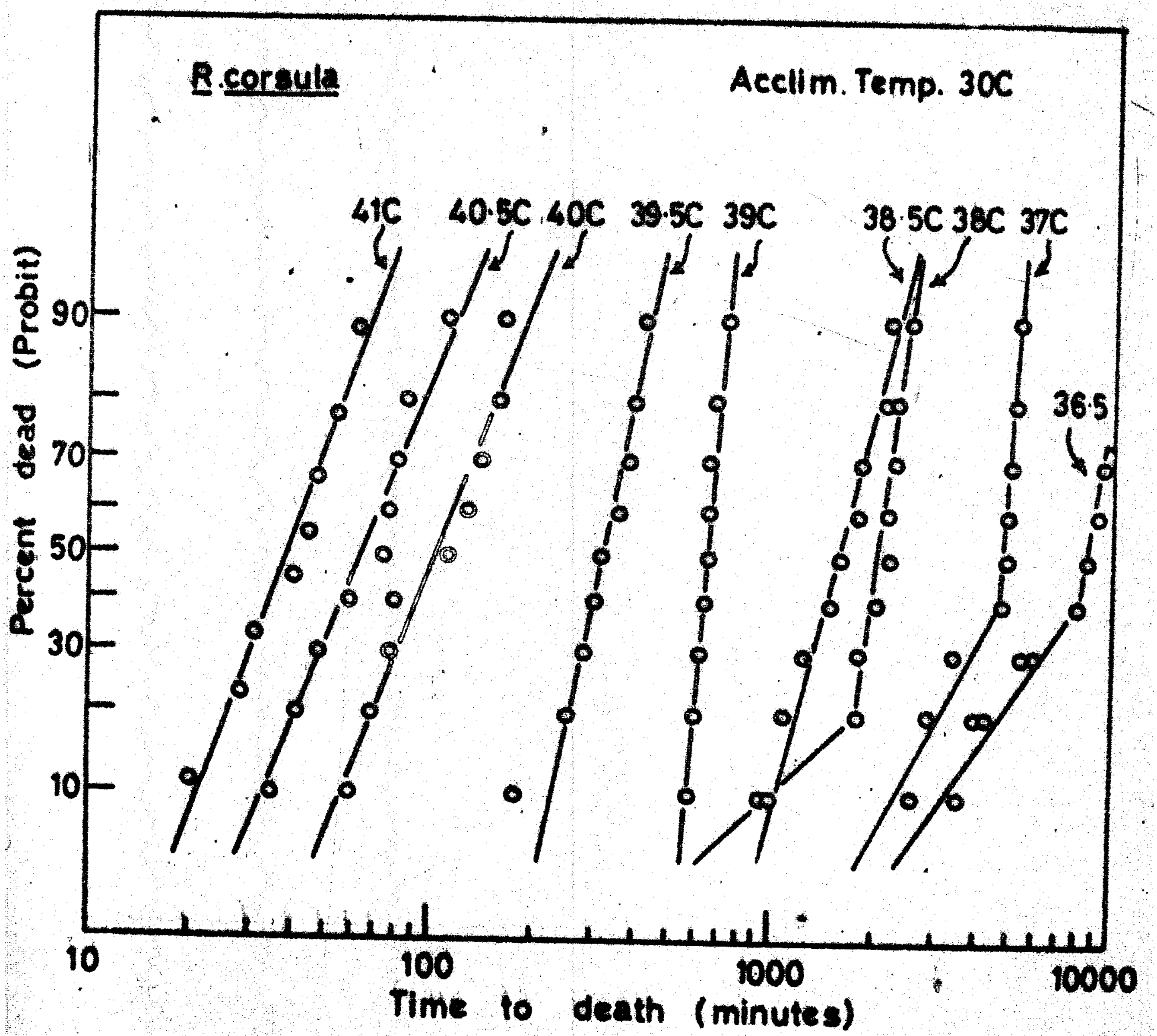
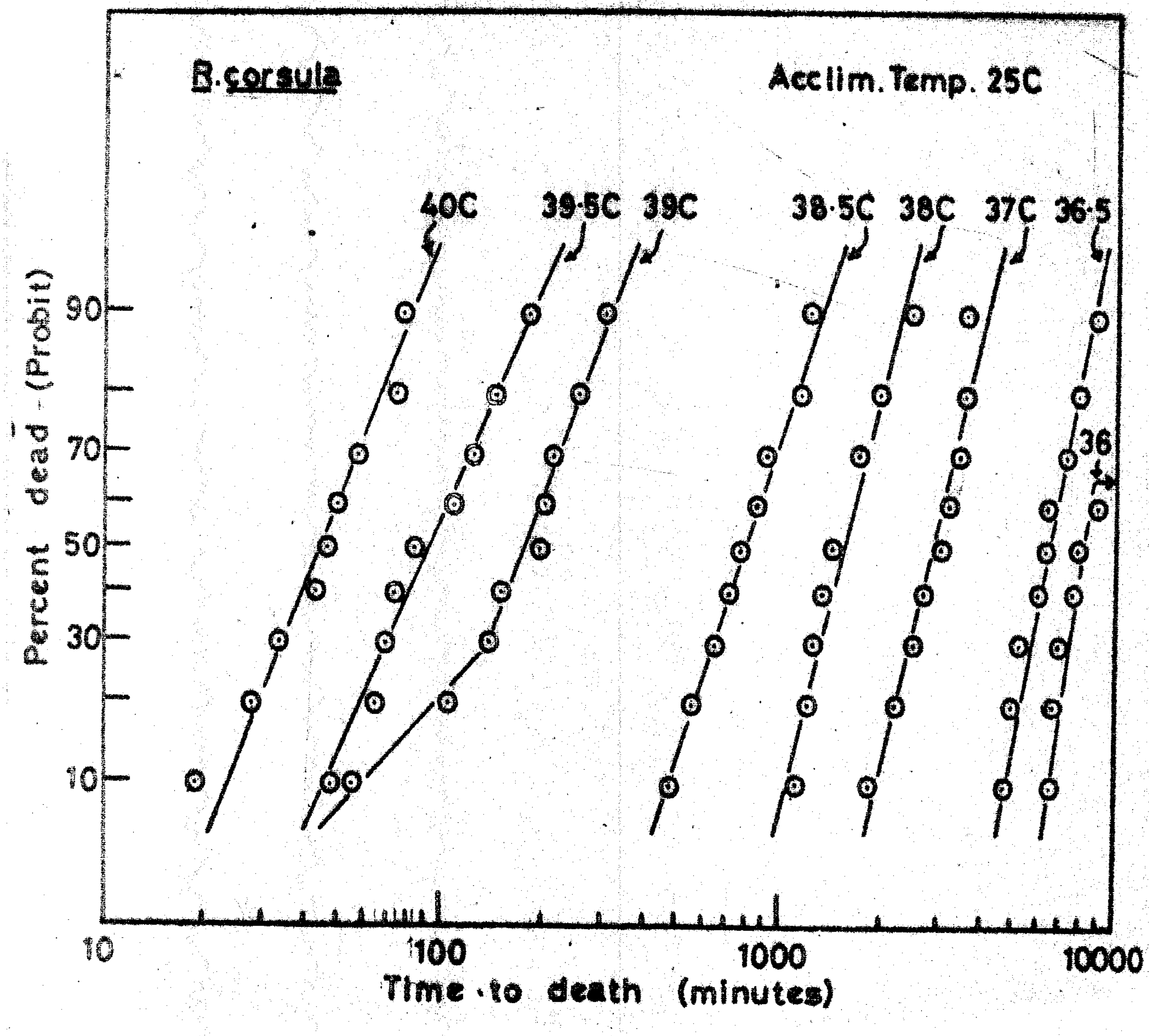

R.corsula
Acclim. Temp. 30C
41C
40·5C
40C
39·5C
39C
38·5C
38C
37C
36·5
Percent dead (Probit)
90
70
50
30
10
10
100
1000
10000
Time to death (minutes)

Fig.55

Times to death at various high lethaltest temperatures among freshwater mullet fingerlings acclimated to 25°C. Plotted on probit x logarithmic axes


R.corsula
Acclim. Temp. 25C
40C
39·5C
39C
38·5C
38C
37C
36·5
36
Percent dead - (Probit)
90
70
50
30
10
10
100
1000
10000
Time to death (minutes)

Figs. 41 to 45). As an example, among mrigal, temperatures between 20 to 32°C are unlikely to cause death, no matter what the acclimation, nor what the exposure time. To determine the incipient lethal temperatures for mrigal the exposure time should sufficiently be longer when tests are performed above 32 and below 20°C. This applies also for other species with their respective tolerance limits. On the other hand, if the tests would have not been continued beyond one day (i.e., 24 hrs) for both high and low lethal levels a different series of incipient lethal temperatures would have bound the zone of tolerance. This would result in a larger area of tolerance due to the addition of a considerable area from the zone of resistance (Fry, 1971). Brett (1970) in his review has presented a table of values for adjusting the incipient lethal temperatures to a 72 hr exposure time to ensure a uniform comparison among the studies. Thus the significance of exposure time in determining the incipient lethal level is emphasised.

Mixed lethal effect:

The sigmoid curves obtained on the raw data (time to death) of most of the high temperature are not found to occur when the data of almost all low temperature experiments are treated similarly (see also 'Method' and 'Results'). Further analysis of the same data on probit chart clearly indicates the presence of two distinct rates of mortification, an initial rapid and a delayed one among all the five species studied. This phenomenon of 'mixed lethal effect' has already been reported by earlier workers also for Girella nigricans (Doudoroff, 1942) and for Pacific salmons (Brett, 1952) and has been referred to as 'split-probit' or 'statistical heterogeneity' (Fry, 1971).

Doudoroff (1945) distinguished this phenomenon for cold death as 'primary chill coma' and 'secondary chill coma'. Further he stated that the initial shock was not manifest until several seconds after transfer to the low temperatures and was not apparently due to stimulation of cutaneous sense organs, but was produced only when the low temperature had penetrated internally, probably to the central nervous system. In the present study also, when exposed to low temperature the fish were found to sense the chillness by the act of wild swimming around the lethal bath only after a considerable time lapse, unlike in high lethal temperatures, where the response was immediate. Accordingly, some of the earlier workers report that in a few species this initial shock was more delayed in larger specimens than in smaller ones (Brett, 1952; Brawn, 1960; Spass, 1960). As already mentioned under 'comparison of thermal resistance' size effect would not have interfered in the results obtained among the species studied.

Though this phenomenon of split-probit was prevalent in all the species, the data of 25 and 30°C acclimated fringe-lipped carp are presented as an example, in Figs.56 and 57 respectively. It is seen in Fig.56, the sample exposed to 13°C suffered very rapid mortality, whereas at 13.5, 14.0 and 14.5°C the mortality among the lots was split into rapid and delayed ones and above 15°C mortality was completely absent. The split-probit curves can be seen in Fig.57 for 15.5, 16.0 and 16.5°C exposures, but at 17°C the mortality is regular However, this kind of split-probits occur at a given critical temperature or between samples at slightly different temperatures. As pointed out by Brett (1952), the time to 50% mortality is consequently affected by the percent occurrence of 'primary' rapid death individuals within the sample.

Pitkow (1960) attributed primary chill coma to the failure of the respiratory centre and Doudoroff (1942, 1945) suggested an osmoregulatory problem as an accessory lethal factor at the secondary chill coma point. Brett (1952) who exposed sockeye solmon to slightly isosmotic medium

Fig.56

Times to death at various low lethaltest temperatures among fringe-lipped carp fry acclimated to 25°C. Plotted on probit x logarithmic axes

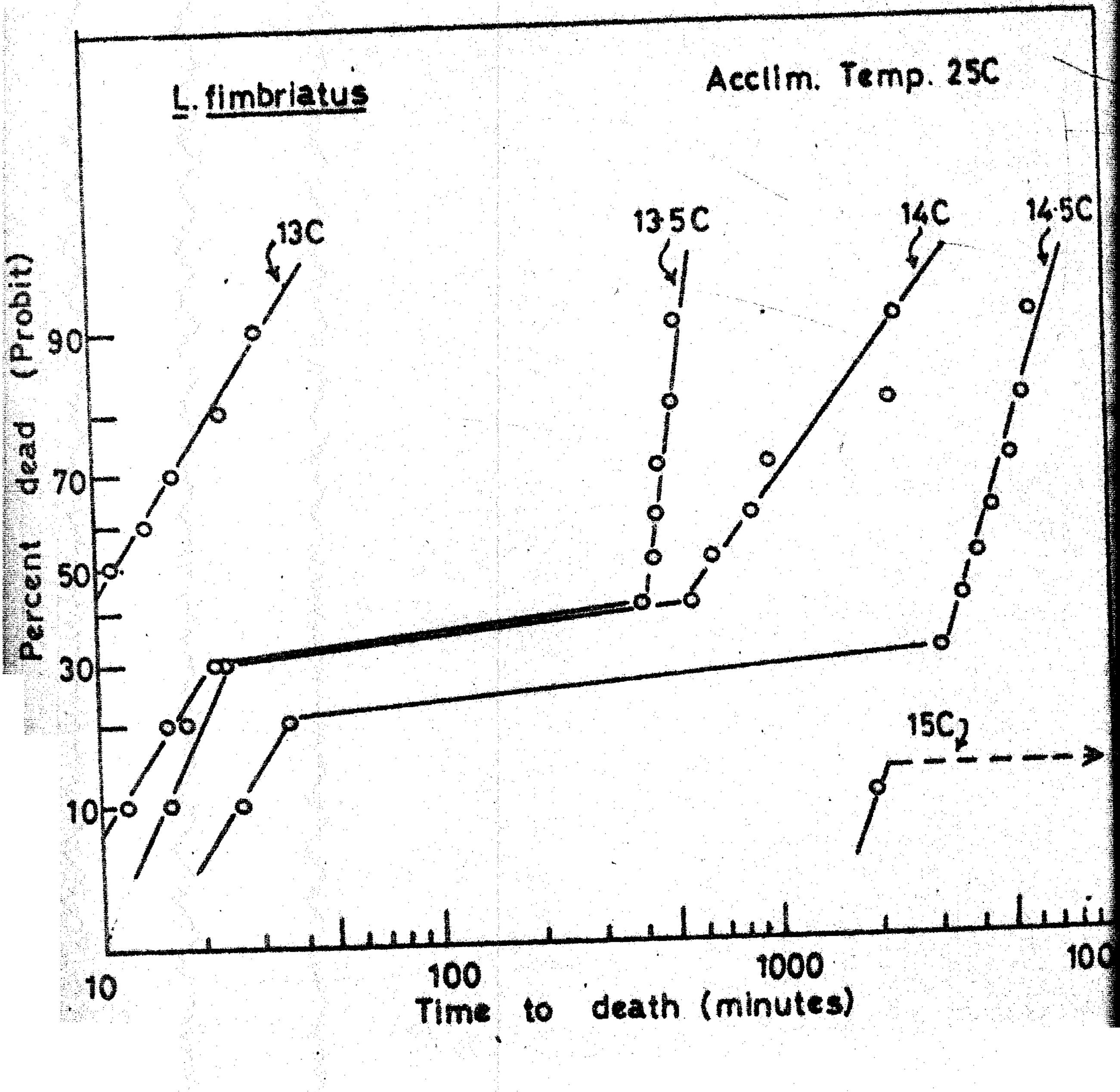
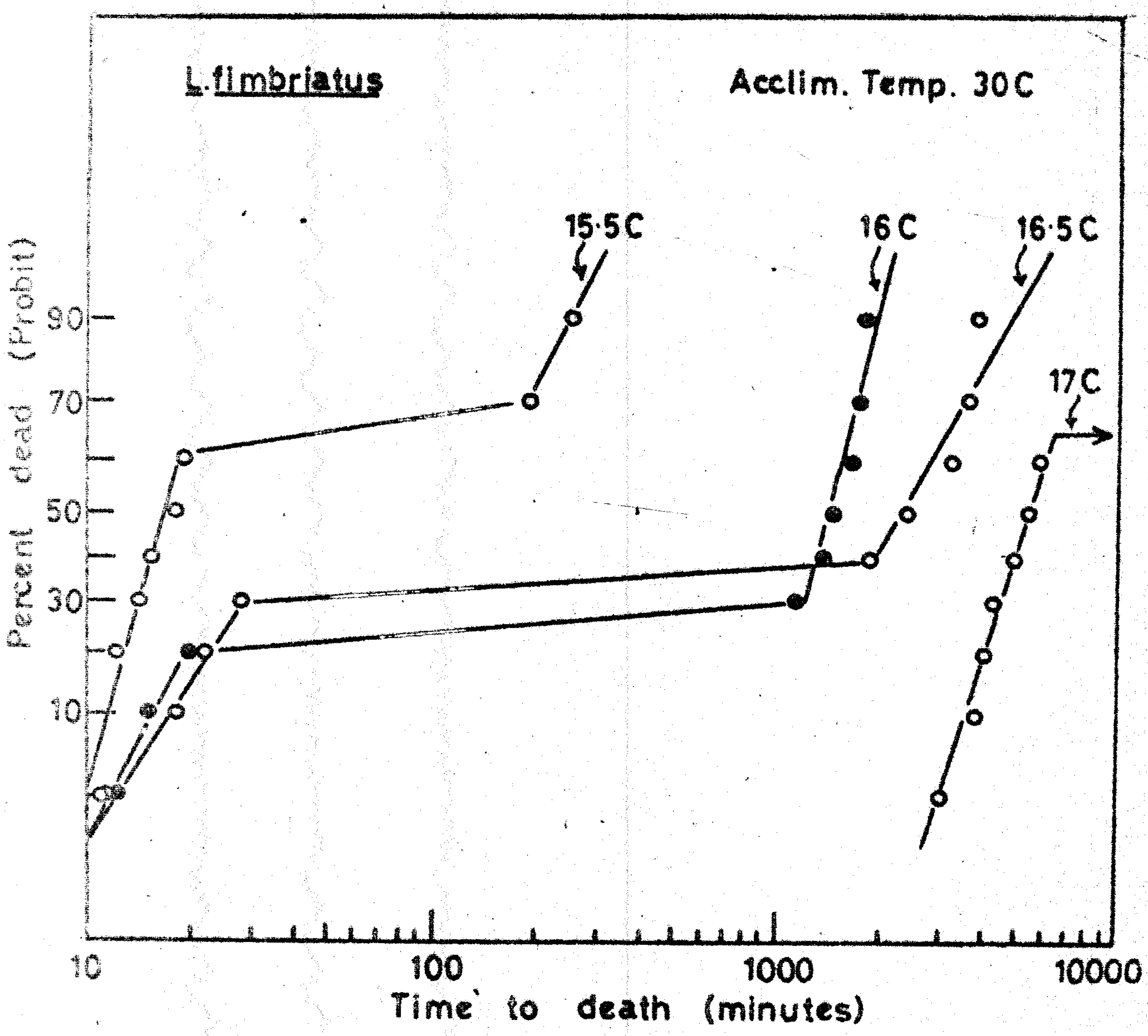

L. fimbriatus
Acclim. Temp. 25C
13C
13·5C
14C
14·5C
15C
Percent dead (Probit)
90
70
50
30
10
10
100
1000
100
Time to death (minutes)

Fig.57

Times to death at various low lethaltest temperatures among fringe-lipped carp fry acclimated to 30°C. Plotted on probit x logarithmic axes

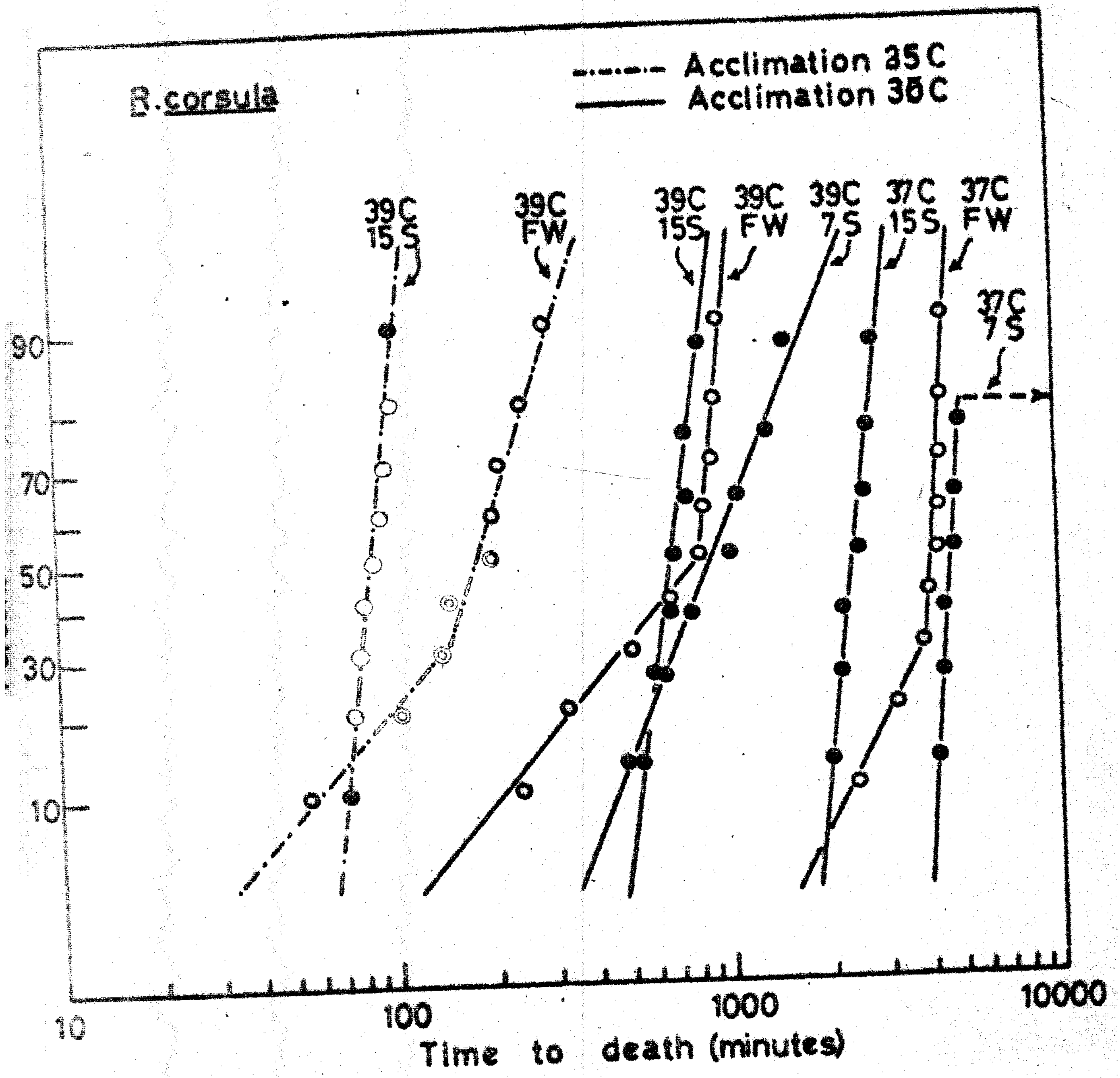

L. fimbriatus
Acclim. Temp. 30C
15·5C
16C
16·5C
17C
Percent dead (Probit)
90
70
50
30
10
10
100
1000
10000
Time to death (minutes)

(9.9 ‰ (S) at 0.2, 0.7 and 3.2°C also supports the above statement by suggesting osmoregulation as a third factor in the event of cold death; the other two factors being, one, a very rapid agent usually effective within 60 min of exposure, a second, not so rapid in action.

Fry (1971) refers to this kind of statistical heterogeneity at high lethal temperatures by Arai *et al*., (1963) in *Poecilia reticulata*, by Tyler (1966) in *C. neogenus* and *C. eos* and by Brett (1952) in *O. keta*. The same kind of statistical discontinuity has been recorded in freshwater mullet and carps in the present study. As an example of the present results, the heterogeneity in thermal response among freshwater mullet is shown in Fig.58, where the curves through open circles and closed circles belong to freshwater and salt water exposures respectively at 37 and 39°C for 25 (broken line connecting dots) and 35°C (continuous line) acclimations. The curves through closed circles either belong to 7 ot 15 ‰ (S) exposures, as indicated in figure. These probits of tests in freshwater are broken or rather bent by an early death of few individuals among the samples. Whereas all the curves of salt water exposures are simple and straight. This result is suggestive of an osmoregulatory stress coupled with thermal shock.

Fig.58

Times to death at 37 and 39°C in fresh and salt water media indicated among freshwater mullet acclimated to 25 and 35°C. Plotted on probit x logarithmic axes


R. corsula
Acclimation 35C
Acclimation 30C
39C 15 S
39C FW
39C 15S
39C FW
39C 7 S
37C 15 S
37C FW
37C 7 S
10
30
50
70
90
10
100
1000
10000
Time to death (minutes)

Salinity Tolerance:

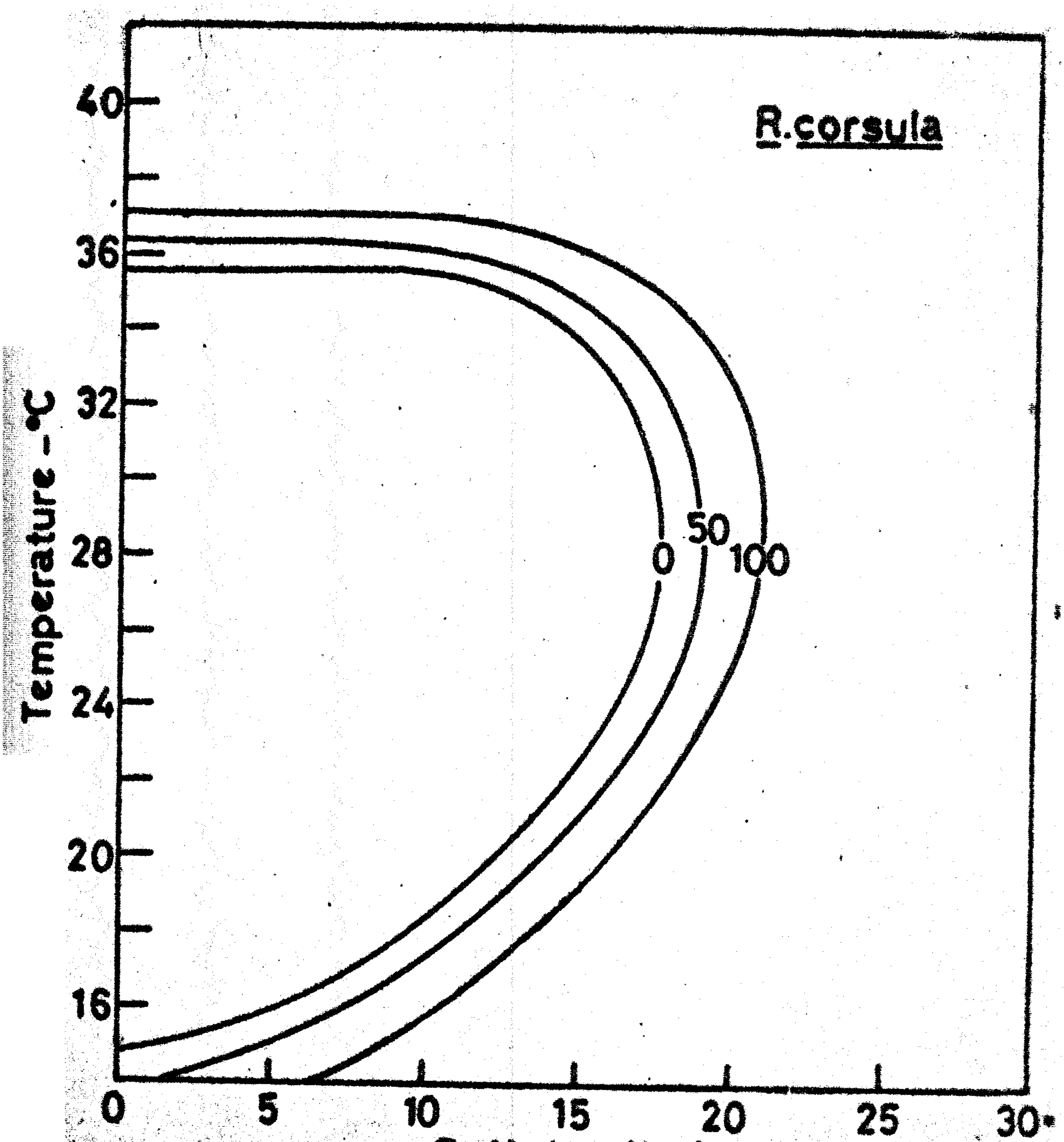
Salinity tolerance of *Rhinomugil corsula*

The results obtained from salinity tolerance experiments on *R. corsula* indicate the incipient lethal salinity levels (below which the mortality will not reach 50%) of 15.35, 17.09, 17.32, 12.25 and 12.25 ‰ (S) at 35, 30, 26, 20 and 17.5°C respectively (Table XXIII). Temperatures above 36.5°C and below 16°C being lethal for 30°C acclimation (see also 'Thermal tolerance'), salinity tolerance tests have been carried out by exposing samples from 28°C, fresh water acclimated group within tolerance range from 17.5 to 35°C and the respective incipient lethal salinities at the thermal extremes are 12.5 and 15.35 ‰ (S). The highest value of 17.32 ‰ (S) has been obtained at 26°C.

In Fig.59 incipient lethal salinities for *R. corsula* are plotted against concerned test temperatures. Resistance

Fig.59

Estimation of relative mortality of freshwater mullet in different salinity and temperature combinations, based on the data of salinity tolerance and salinity effect on thermal resistance


R.corsula
40
36
32
28
24
20
16
Temperature -°C
0
50
100
0
5
10
15
20
25
30
Salinity (‰)

ble XLI

esults from analysis of variance of resistance times (logarithms) for freshwater
ullet acclimated to 28 ± 1°C and tested at various salinities and temperatures

ccli. emp.	Comparison	Sum of squares	Degrees of freedom	Variance	Value of F ratio	Value for F = 0.05	Value for F = 0.01
8°C	Between salinities (18.0, 26.25, 30.0, 35.0, 40‰ (S)	70.6850	4	17.6713	114.82**	6.39	15.98
	Between temperatures (30, 35°C)	15.2384	1	15.2384	99.01**	7.71	21.20
	Salinities x temperatures (error)	0.6156	4	0.1539			
	Total	86.5390	9				
8°C	Between salinities (20, 25, 30, 35, 40‰ (S)	114.7656	4	28.6914	40.20**	3.84	7.01
	Between temperatures (17.5, 20.0, 26.0°C)	19.6915	2	9.8458	13.79**	4.46	8.65
	Salinities x temperatures (error)	5.7099	8	0.7137			
	Total	140.1670	14				

** Highly significant

The error terms are very low (i.e., 0.1539 and 0.7137), which indicate that the interference of the uncontrolled factors is very low.

In the present study on salinity tolerance the fish were not acclimated to specific conditions of test temperatures and salinities, except at the ambient condition of acclimation at the single temperature (28°C) in freshwater. Under this condition salinity concentrations below 12.25 $^{0}/_{00}$ (S) are not at all lethal at any temperature within the thermal tolerance limit, in an acute exposure from freshwater. Further, studies on this aspect by acclimating this species to different salinities and temperature can only indicate the complete salinity tolerance of the species. Even the limited results obtained here from acute tests demonstrate the euryhaline adaptability of this species, apparently owing to the iono-osmoregulatory mechanism of this species. This may be another instance of physiological versatility, where the same regulatory mechanism, involving different structures like gills, kidney, gut etc., functions with equal facility in either media fresh and salt waters, through a reversal in its physiological polarity (Parvatheswararao, 1970).

Salinity tolerance- other species:

The data obtained on incipient lethal salinity from the comparative study of salinity tolerance on three carps, namely *C. mrigala*, *L. fimbriatus* and *C. carpio* and cichlid fish *Tilapia mossambica* are 3.54, 7.07, 8.13 and 7.07 ‰ (S) respectively at 30°C. As in the case of *R. corsula*, acclimations were not done at specific temperature and salinity in these species also and exposures were made abruptly from 30°C fresh water except for one group of *L. fimbriatus*, which were acclimated to 5 ‰ (S) at 30°C (see also 'Methods'). Among the carps, as it is seen from these data, *C. mrigala* is the least tolerant to salinity with incipient lethal value of 3.54 ‰ (S); whereas the *C. carpio* is the most tolerant, *L. fimbriatus* being intermediate in tolerance. But the euryhaline *Tilapia mossambica* appears to be comparatively less tolerant when compared to *C. carpio* which has the highest value of incipient lethal salinity (i.e. 8.13 ‰ (S). (*T. mossambica* tested were hatchlings while the others were fry and fingerlings). On the other hand the data on the median resistance times (Table XXIV) demonstrate that *T. mossambica* has much higher value than for those of the carps, including *C. carpio*. This results show that *T. mossambica* is more resistant than the carps at all lethal salinities tested. Further, *T. mossambica* hatchlings were able to resist salinity even upto 35 ‰ (S) whereas carps suffer immediate death above 25 ‰ (S) (Table XXIV). This inverse relation in resistance and tolerance indicated by their differential response towards salinity among the euryhaline *T. mossambica* and stenohaline *C. carpio* might be due to the size difference of the fish tested i.e., *T. mossambica* size was only 0.9 cm and *C. carpio* was about 2.7 cm, but not less than 1.5 cm (Table I). Hence, the osmoregulatory mechanism would have not developed so well in the hatchlings of *T. mossambica* as in the larger and older carp *C. carpio*. This is supported by the observation of Liu (1942, 1944) on the development of chloride cells in paradise fish transferred from fresh water to salt water.

Wikgren (1953) has pointed out and excessive loss of ion among carps at low temperatures and Morris (1960) also supports this finding based on his work on lamprey. This appears to be true in the case of *R. corsula* also, for the salinity becomes progressively lethal for this species at low temperatures. Thus it appears, that these species presently studied would also be more susceptible at low temperatures; but as already mentioned, salinity-temperature interaction on these species has not been done at temperatures below 30°C. Further, as suggested already a full picture of salinity tolerance and resistance of the mullet could be obtained only with information with performance of this species acclimated and tested under different salinity and temperature levels.

Comparison among the five species:

The results obtained on incipient lethal salinities of all the five species demonstrate that the euryhaline R. corsula has the highest value of 17.09 %° (S) at 30°C and the rest of the species have comparatively much lower values. As far as T. mossambica is concerned, the adults of this species have been observed to survive in total sea water but the incipient lethal salinity of hatchlings of this species, in the present work, is only 7.07 %° (S). Among the carps the salt water acclimated L. fimbriatus does not display any change in its tolerance level, since the incipient lethal salinity values for fresh and salt water acclimated groups being the same i.e. 7.07 %° (S). But the data on median resistance times (Table XXIV) show an increase in resistance due to salinity acclimation. This result suggests salinity acclimations among these stenohaline carps may not markedly increase their salinity tolerance, as it can be expected among the euryhaline species. Thus, as obvious the stenohaline carps have comparatively low tolerance and adaptability to salinity.

Salinity effect on thermal resistance:

Displacement of lethal temperature of fish induced by the imposition of osmoregulatory stress have been reported by a number of workers, following the initial investigation by Loeb and Wasteneys (1912). The results of present work on *R. corsula* acclimated to four different temperatures (20, 25, 30 and 35°C) and tested at various potentially lethal temperatures in different saline media show a complex response. The data obtained on the exposures above that of fresh water to 12 ‰ (S) show an increasing thermal resistance when the fresh water exposure is taken as a standard (Table XXVI). As the data on incipient lethal salinity (Table XXIII) indicate, salinity levels above 15.34 ‰ (S) being lethal at high temperature (35°C), these salinities reduce resistance of this species i.e., the adverse salinity interacts with lethal temperature to reduce the resistance time. These above mentioned features can be seen from Figs.36 and 37 which present the data for *R. corsula* acclimated to 20 and 25°C and exposed to various salinities (fresh water 3, 5, 7, 10, 12, 15, 18 and 20 ‰ (S) at lethal temperatures (37 and 39°C). As seen from these figures the highest resistance is observed at 7 ‰ (S) at both the acclimations and lethal temperatures. Curiously, as it appears, there is a depression at 5 ‰ (S) levels (Figs. 36 and 37; Table XXIV. A possibility for an artifact is ruled out, since the result has been obtained uniformly for both the acclimations i.e., 20 and 25°C. This unique result can be attributed to the following reasons individually or collectively, as mentioned hereunder.

Khlebovich (1968) points out there is sharp change in electrical properties of live forms at ambient salinity of 5 ‰ (S) when exposed to a series of ambient salinity changes. Khlebovich (1968) further suggests a pronounced change in the ion ratio, when marine and fresh water is mixed. The critical salinity has been shown to be about 5 ‰ (S) (in the Kara, White, Baltic, Azov, Black and probably, Caspian seas and British Columbia). In addition a close connection is suggested between mixed water hydrochemistry and the existence of an ecophysiological barrier. This barrier divides at salinities of 5 to 8 ‰ (S), the two basic types of aquatic animals into marine and fresh water representatives. Moreover, the aquatic fauna are least in number and density at 5 ‰ (S) (Khlebovich, 1962). These phenomena pointed out by Khlebovich,(1962, 1968) pertaining to the critical salinity of 5 ‰ (S) may be expected to explain the depression in resistance at 5 ‰ (S) among *R. corsula*. Secondly, this work has been done on immature fingerlings of *R. corsula* and it is suspected that the osmoregulatory mechanism might have not been developed so well as to cope with the osmotic imbalance as pointed out earlier for *T. mossambica* hatchlings (see 'salinity tolerance'). Job (1959) also suggests a possible difference in osmoregulatory mechanism with respect to size difference (i.e., age) in his work on the measurement of lethal oxygen at different salinities in *Plotosus anguilaris*. Finally, *R. corsula* is brackish water in origin, capable of survival and reproduction in fresh water (Pakrasi and Alikunhi, 1952; Narayanan, 1974). This change in ecological environment may also be considered a possible reason for this kind of unique response at 5 ‰ (S). Subsequent studies on the measure of lethal temperatures instead of resistance times, for different factorials of temperature acclimations may provide additional information in this regard (Garside and Chin-yun-kee, 1972; Garside and Jordan, 1968; Garside, per.comm.,).

As reported by Craigie (1963) and Alabaster (1967) for salmonids, salinities close to isosmotic level (i.e. as already pointed out, non-lethal salinity from 3 to 12 ‰ (S) also favour thermal resistance to considerable extent, but the highest resistance was observed in 7 ‰ (S) at all lethal (test) temperatures in all temperature acclimations among R. corsula. This is obviously due to the isosmotic effect at this salinity and the energy conserved from the diminished osmotic stress might have been diverted to maintain equilibrium (homeostasis) at high (lethal) temperatures. On the other hand, salinities above and below 7 ‰ (S) being hyper and hyposmotic respectively to almost all teleost, have hastened thermal death due to added effect of increased osmotic stress. Garside and Jordan (1968) obtained an elevation of 6°C in lethal temperature in isosmotic medium, compared to fish and sea water test values in Fundulus heteroclitus and Fundulus diaphanus. Garside and Chin-Yun-Kee (1972) have also reported the same trend for F. heteroclitus acclimating this species to different salinity and temperature combinations. Further, Rao (1968, 1971) recorded the lower metabolic rate in isosmotic salinity of 7.5 ‰ (S) for Salmo gairdneri. The results of present work fall in line with above reports and agree with the observations of Gibson (1954), Arai et al., (1963) and Strawn and Dunn (1967).

The results obtained on 'F' ratio from analysis of variance of resistance times for 35°C acclimation alone are shown in Table XLII. There is just significant difference in response of this species towards salinity and temperature interaction, between salinities (P 0.05); whereas there exists a highly significant difference between temperatures (P 0.01). The error term is very low (0.8795) for 4 degrees of freedom and this indicates the least interference of the uncontrolled variables in this work.

When the overall work on thermal tolerance and salinity effect on thermal resistance of this species is considered, it may be stated, in agreement with Garside and Chin-Yun-Kee (1972), that a simple statement of lethal temperature with respect to thermal acclimation alone is not sufficient. Atleast for euryhaline species like R. corsula, reference to salinity of the lethal medium should be made to make the statement meaningful.

Table XLII

Results from analysis of variance of resistance times (logarithms) for freshwater mullet acclimated to 35°C and tested at different salinities and temperatures

Accli. Temp.	Comparison	Sum of squares	Degrees of freedom	Variance	Value of F ratio	Value for F = 0.05	Value for F = 0.01
35°C	Between salinities (F.W., 7, 15‰ (S)	28.0748	2	14.0374	15.96*	6.94	18.00
	Between temperatures (37, 39, 41°C)	586.5528	2	293.2764	333.46**	6.94	18.00
	Salinities x temperatures (error)	3.5179	4	0.8795			
	Total	618.1455	8				

* Significant

** Highly significant

General Discussion and Conclusions

In the present study thermal tolerance and resistance of mrigal rohu, fringe-lipped carp, common carp and freshwater mullet have been described. In addition salinity tolerance and resistance of freshwater mullet, mrigal, fringe-lipped carp, common carp and Tilapia mossambica acclimated to fresh water (except L. fimbriatus exposed to 5 °/°° (S) and natural ambient temperature (28 - 30°C), have been studied. Observations have also been made on survival of freshwater mullet acclimated to several temperatures in freshwater and exposed to various salinity and temperature combinations.

These observations indicate that the thermal and salinity responses studied are species specific. It is likely that these differences are genetically determined and might also reflect the environmental conditions prevailing in the specific geographical zones from which they have originated. Among the species studied for thermal tolerance the common carp which is subtropical in origin, has the highest thermal tolerance (1075°C^2) and its capacity to live under different thermal conditions is proved by its present worldwide distribution as a culturable species. The Indian major carps mrigal and rohu, have more or less similar thermal tolerance (812 and 850°C^2 respectively) and their original distribution was restricted to North Indian rivers originating from Himalayas. The fringe-lipped carp, which is more tropical in distribution, naturally occur in east flowing rivers of South India, have a lesser thermal tolerance (731°C^2) as compared with the former two Indian carps.

L. fimbriatus has relatively higher low incipient lethal temperatures (see also text, Table VII). It is tempting to correlate the greater low temperature tolerance with the natural lower temperature to which mrigal and rohu would be exposed in the higher latitudes (as compared with Labeo fimbriatus), in which they inhabit.

The mullet, R. corsula originating from the estuaries of the North Indian rivers is euryhaline like the other members of mugilidae, but unlike other mullets this is capable of survival and reproduction in freshwater (Pakrasi and Alikunhi 1952; Narayanan, 1974). The thermal tolerance of this species is least (569°C^2) among the species studied. The narrowness of the thermal tolerance range of this species would indeed be genetical, but this could have been determined by the lesser fluctuations in temperature which might occur in estuaries, as compared with that of inland waters.

Eventhough it may be difficult to correlate the upper and lower thermal tolerance to specific geographical latitudes, a broad generalisation might be applicable. For example, the polar Trematomus sp. has a very narrow thermal tolerance (100°C^2) and the temperate species especially the salmonids have thermal tolerance intermediate between those of the subtropical/tropical and polar species.

As referred to earlier there appears to be an inverse correlation between thermal tolerance and resistance (Table XX The stenothermal mullet has a thermal tolerance of 569°C^2 while the estimated resistance is 391°C^2; whereas the corre ponding thermal tolerance and resistance for the eurytherma common carp are 1075 and 225°C^2 respectively.

The present observation on salinity tolerance and resistance are limited in view of the fact that the acclimations to various salinities were not made. For purposes of comparisons of salinity tolerance the data obtained fr single level of acclimation i.e., fresh water and acutely exposed to various lethal salinities can be made use of. Thus it is seen that euryhaline mullet has the highest incipient lethal salinity among the species studied. As well known the carps as a group have low salinity toleran the lowest incipient lethal salinity (3.54 ‰ (S) being f mrigal. *Tilapia mossambica*, which is well known to toler both fresh and total sea water was tested in the present study as hatchlings only (less than 1 cm) and as such the capacity for salinity tolerance was found to be limited, i.e., less than that of mullet. Apparently the capacity for salinity tolerance for *T. mossambica* would be increa sing with incresing size and age of the fish.

Tests on the effect of temperature on salinity tolerance reveal that the salinity tolerance of the mullet decreases towards both the upper and lower thermal extremes (optimum range being 26 to 30°C). The reduction in salinity tolerance being more marked at low temperature suggesting a breakdown of iono-osmoregulatory mechanism more easily at the low temperature extremes than at high. Salinity greatly influences the thermal resistance of R. corsula and the highest resistance to temperature is at isosmotic medium of 7 ‰ (S) and 5 ‰ (S) is observed to be critical, which adversely affect thermal resistance. Salinity levels above that of fresh water to 12 ‰ (S) being non-lethal, within the thermal tolerance limits, enhance the thermal resistance, whereas salinities above 12 ‰ (S) being lethal, reduce the thermal resistance acutely by the interaction with lethal temperature. In the single case of salinity acclimation (5 ‰ (S) among L. fimbriatus, salinity tolerance was not enhanced when compared with that of freshwater acclimation. But a clear increase in resistance to lethal salinity was observed.

As evident, present study is mainly concentrated around thermal tolerance of a few selected freshwater fishes and to the extent possible descriptions of thermal tolerance and resistance have been given. As made out earlier, certain limitations of technique did not enable the study to be conducted at still lower temperatures than those presently done. Estimation of thermal tolerance over this narrow region of low temperature was made by extrapolation and it is felt that this treatment does not seriously affect estimates made. Observations on salinity and temperature effect on survival of fishes, presently confined to single level of acclimation, could yield much more information if the acclimations were extended to several other salinity levels, within the tolerance range. Thermal tolerance area, presently described for fishes acclimated and tested in freshwater, and resistance have been studied at certain salinity and temperature combinations only in the case of R. corsula. It would be worthwhile knowing how thermal tolerance changes for various salinity acclimations and exposures so that one can have a full picture of the effect of temperature and salinity on the survival of the species concerned.

It is hoped that the results of the present study will be of help in chosing suitable aquaculture practices especially with reference to controlling ambient temperature during culture and transportation of fish. At this time of ever increasing demand for energy, commissioning of more number of thermal plants would lead to discharges of heated effluents to the natural waters, leading to an imbalance in the aquatic ecosystem. The values of thermal tolerance and resistance obtained for the various fresh water fishes in the present study may be of use in evolving proper measures for environmental protection.

Summary

1 Young of four species of carps (average 4 cm), namely, mrigal (*Cirrhinus mrigala*), rohu (*Labeo rohita*), fringe-lipped carp (*Labeo fimbriatus*) and common carp (*Cyprinus carpio*), a freshwater mullet (*Rhinomugil corsula*) and a cichlid fish (*Tilapia mossambica*) were used in the present study on the 'Ecophysiology fish fry and fingerlings with special reference to temperature tolerance'. Among the three species of experiments conducted namely, 'thermal tolerance', 'salinity tolerance' and 'salinity effect on thermal resistance', thermal tolerance study was carried out on all species except *Tilapia*, the salinity tolerance study was made on all species except rohu

and the study of salinity effect on thermal resistance was carried out only on freshwater mullet.

2 The fish were acclimated to 15, 20, 25, 30, 35 and 38°C and resistance times were determined at intervals of 0.5°C for both high and low temperatures. Upper lethal temperatures were calculated for exposures of 10,000 min (7 days) and lower lethal temperatures for exposures of 8,500 min (6 days). Salinity acclimation was restricted to single level at ambient temperatures 28 - 30°C in fresh water except fringe-lipped carp, which was acclimated both in fresh and salt water (5 ‰ (S). Resistance times were obtained on salinity exposures from 10 to 45 ‰ (S) at 17.5, 20.0, 26.0, 30.0 and 35.0°C for R. corsula; for other species exposures were made from 2.5 to 35 ‰ (S) at 30°C. For salinity effect on thermal resistance exposures were made in salinities above that of freshwater to 20 ‰ (S) at test temperatures (37, 39 and 41°C) from acclimations 20, 25, 30 and 35°C.

3 The raw data obtained on resistance time from all these three series of experiments were first subjected to 'probability analysis' and median resistance times were obtained. The usual sigmoid time-mortality

curves became irregular often for low temperature data. The treatment of the same data on 'probit charts' resulted in 'split-probits' which separated the two different rates of mortification, reported by earlier workers as 'mixed lethal effect', 'statistical heterogeneity' and 'split-probit' (Brett, 1952; Doudoroff, 1942, 1945, 1952; Fry, 1971). The data on thermal tolerance were treated, further, according to regression analysis (differentiate resistance response). The significant difference in tolerance and resistance were detected by the analysis of variance ('F' test). The incipient lethal values were estimated by the method of Miller and Tainter (1944). Zone of thermal tolerance and resistance were obtained from the incipient lethal temperatures.

4 The median resistance times for high temperatures decrease with an increase in lethal temperature but increase with increase in acclimation temperature. This phenomenon is reversed for low temperature. Median resistance time obtained on salinity tolerance show decreasing trend with increase in lethal salinity and test temperature. The data on salinity effect on thermal resistance show a complex feature. The median resistance times increase with the increase in salinity above that of fresh water to 7 7 ‰ (S) with a depression at 5 ‰ (S) and decrease in salinities over 7 ‰ (S) at all lethal (test) temperatures and thermal acclimations.

5 The graphically constructed zones of thermal tolerance and resistance are given in °C^2 units. The total area of tolerance and resistance are further divided into 'upper' and 'lower' zones and thermal triangles, with the aid of 45° isotherm-line and by the method of McErlean et al., (1969) respectively. The upper zone of thermal tolerance is larger than the lower in all the species studied; whereas this phenomenon is reversed in upper and lower zones of thermal tolerance. The widest area of thermal tolerance was exhibited by common carp (1075°C^2) and the smallest by freshwater mullet. The other three species, namely, mrigal, rohu and fringe-lipped carp being intermediate (812, 850 and 731°C^2 respectively), but comparatively wider than that of freshwater mullet. Hence, the carps are 'eurythermal' and the freshwater mullet is 'stenothermal'. Thermal triangles are not given for common carp and the given triangles for other species overestimate the thermal tolerance, due to two limitations. The formation of 'Plateau' and 'floor' in thermal polygons due to intolerance to high and low temperatures respectively could not be shown in thermal triangles because of the projection of the hypotenues beyond usual acclimation and tolerance limit.

6 The data on upper and low thermal tolerance and resistance were further analysed by the study of 'relative tolerance and resistance', in which the total area of tolerance or resistance is considered as 100% and the fractions there of constitute the upper and lower thermal tolerance or resistance in percent. As mentioned earlier, the phenomenon of inverse relation between tolerance and resistance is obvious from the data on 'relative tolerance and resistance'. This phenomenon is more obvious in freshwater mullet, when the total area on tolerance and resistance is considered. Though freshwater mullet has a low tolerance area (569°C^2), the resistance area is relatively higher (391°C^2) than that of carps (as an example, 1075 and 225°C^2 for tolerance and resistance respectively for common carp). There is marked difference in 'relative upper and lower thermal tolerance' among various species. *O.punctatus* *C.auratus*, *A.nebulosus* and *R.rutilus* have relative lower thermal tolerance higher than the relative upper thermal tolerance. Whereas the Pacific salmons and the five species presently studied have relative lower thermal tolerance lower than the upper. Hence, the original geographic distribution does not have any direct relation to the relative upper and lower thermal tolerance.

7 Proportionate change in tolerance was estimated by dividing the total difference in incipient lethal temperatures by the difference in temperature acclimations concerned. The rate of change in upper incipient lethal temperature is 1°C for about 5°C increase in acclimation temperature. The rate of change in lower incipient lethal temperature is 1°C for about 2°C change in acclimation temperature. The common carp displays the highest rate (0.21°C/1°C and 0.6°C/1°C for high and low temperatures respectively). The lowest rate is exhibited by fringe-lipped carp (0.17°C/1°C and 0.44°C/1°C for high and low temperatures respectively). Briefly, the rate of change in high temperature is much lower than the rate in low temperature when compared with the data on other species (Brett, 1944; Doudoroff, 1945).

8 The values obtained on analysis of variance of the upper and lower incipient lethal temperatures indicate a highly significant difference in tolerance between species and between acclimations ($P < 0.01$). Further, the four different combinations in 'Orthogonal comparison' also show a highly significant difference in response among the species. Thus, the thermal tolerance response is species-specific and genetically controlled.

9 The following conclusions were made from the analysis of variance of resistance times in high lethal temperatures.

i No significant difference in resistance to high temperatures exists among the species at any level of treatment for higher acclimations of 30 and 35°C

ii There is significant difference in resistance among these species at 25°C acclimation level

iii Mrigal shows a moderately significant difference in resistance ($P < 0.05$)

iv Common carp exhibits a high significant difference from freshwater mullet ($P < 0.01$)

v Common carp and freshwater mullet show a barely significant difference in resistance from fringe-lipped carp ($P < 0.05$)

vi Mrigal and rohu show a highly significant difference in response from fringe-lipped carp ($P < 0.01$)

10 The resistance times to low lethal temperatures could not be subjected to systematic analysis owing to the lack of consistency in response, as has also been observed in other fishes by earlier workers. Difference in response to low temperature among

the species necessitated in the use of different lethal levels to link the cases of non-lethal to fairlyrapid lethal level. This factor formed additional limitation on further analysis of the data on low temperature.

11 The study on salinity tolerance on R.corsula indicate that salinities below 12.25 ‰ (S) are not lethal at any temperature within tolerance limit in an acute exposure from freshwater. The optimum temperature from 26 to 30°C favours salinity tolerance. Low temperature is more detrimental than high, though this species have higher resistance times at low temperature. Tilapia resists salinity better than the carps; but has incipient lethal salinity lower than common carp and freshwater mullet. The low incipient lethal salinity for Tilapia (7.07 ‰ (S) is apparently owing to the use of hatchlings, the osmoregulatory mechanism might have not developed so well as in the case of older carps and freshwater mullet. Among the carps, common carp is highly tolerant to salinity (8.13 ‰ (S), mrigal is the least tolerant (3.54 ‰ (S) and fringe-lipped carp is intermediate (7.07 ‰ (S) . Salinity acclimation in fringe-lipped carp enhances the resistance to salinity, but there is no change in tolerance level.

12 Salinities from that of freshwater to 12 ‰ (S) being non-lethal, enhance the thermal resistance of freshwater mullet and the highest resistance was observed at 7‰ (S) (isotonic medium). At salinities below and above 7 ‰ (S) comparatively lower values were obtained than that of 7 ‰ (S). There is depression in thermal resistance in 5 ‰ (S), this is apparently due to the change in the electrical properties at cellular level in this species (Khlebovich, 1962, 1968).

It is hoped that the present study will be of help in aquaculture practices especially with reference to controlling ambient temperatures during culture and transportation of fish. The values of thermal tolerance and resistance obtained for the various freshwater fishes in the present study may be of use in evolving proper measures for environmental protection.

ferences

Ahokas,R.A. and Duerr,F.G. 1975. Salinity tolerance and extracellular osmoregulation in two species of euryhaline teleosts, *Culaea inconstans* and *Fundulus diaphanus*. Comp. Biochem. Physiol. Vol.52A:445-448

Alabaster,J.S. 1967. The survival of salmon (*Salmo salar* L.) and sea trout (*S. trutta* L.) in fresh and saline water at high temperatures. Water Res. I: 717 - 730

Allanson,B.R. and Noble,R.G. 1964. The tolerance of *Tilapia mossambica* (Peters) to high temperature. Trans. Am. Fish. Soc. *93*: 323 - 332

Allen,K.O. and Strawn,K. 1968. Heat tolerance of channel cat fish *Ictalurus punctatus*. Proc. 21st Con . Southeast Assoc. Game Fish Comm. 1967. pp. 399 - 411

Altman,P.L. and Dittmer,D.S. (Eds.). 1966. Environmental Biology. Fedn. Proc. Fedn. Am. Socs. exp. Biol. 25: 1 - 694

Ananthakrishnan,K.R. and Kutty,M.N. 1974. Mortality and breathing rate at high ambient temperatures in the cichlid fish, *Tilapia mossambica* (Peters). Indian J. Exp. Biol. 12; 55 - 59

Ananthakrishnan,K.R. and Kutty,M.N. 1976. Temperature tolerance of the air-breathing fish *Ophiocephalus punctatus*. Comp. Physiol. Ecol. 1: 18 - 22

Ananthakrishnan,K.R. and Srinivasan,K. 1977. Effect of dilutions of sea water on the upper lethal temperature of the cichlid fish, *Tilapia mossambica*. Comp. Physiol. and Ecol. Vol.2, No.3: 154 - 157

Andronikov, 1975. Heat resistance of gametes of marine invertebrates in relation to temperature conditions under which the species exist. Mar. Biol. 30: 1-11

Arai,M.N., Cox,E.T. and Fry,F.E.J. 1963. An effect of dilutions of sea water on the lethal temperature of the guppy. Can. J. Zool. **41**: 1011 - 1015

Armitage,K.B. and Olund,L.J. 1962. Salt tolerance of the brook stickleback. Am. Midl. Nat. **68**: 274 - 277

Bapat,S.V. 1970. The Bombay duck, *Harpodon nehereus* (Hamilton). Bull. Cent. Mar. Fish. Res. Inst. (21): 66p

Bardach,J.E., Ryther,J.H. and McLarney,W.O. 1972. Aquaculture - The farming and husbandry of freshwater and marine organisms. Wiley Interscience, London

Battle,H.I. 1929. Effects of extreme temperatures and salinities on the development of *Enchelyopus cimbrius*(L.). Contr. Can. Biol. Fish. **4**: 109 - 191

Becker,C.D. 1973. Columbia river thermal effects study: reactor affluent problems. J. Water Poll. Cont. Fed. Vol.45, No.5: 850 - 869

Binet,L. and Morin,G. 1934. Action de la chaleur sur les poissons. Jour. de Physiol. et de Pathol. Gen. **32**, 272 - 379

Black,E.C. 1952. Upper lethal temperatures of some British Columbia fresh water fishes. J. Fish. Res. Bd. Can. 10, 4: 196 - 210

Black,V.S. 1957. Excretion and osmoregulation. In: Physiology of fishes. Vol.I, (Ed.): M.E.Brown. Academic Press. Inc., New York. p. 163 - 206

Black,V.S. 1962. In: 'The Physiology of Fishes'. Vol.I. Page 163. (Ed.) M.E.Brown. Acad. Press. New York

Blaxter,J.H.S. 1960. The effect of extremes of temperature on herring larvae. J. mar. biol. Ass. U.K. 39: 605 - 608

Blaxter,J.H.S. and Holliday,F.G.T. 1963. The behaviour and physiology of herring and other clupeids. Adv. mar. Biol. 1: 261 - 393

Beamish,F.W.H. 1964. Respiration of fishes with special emphasis on standard oxygen consumption. II. Influence of weight and temperature on respiration of several species. Can. J. Zool. 42: 176 - 188

Bliss,C.I. 1935. The calculation of the dosage-mortality curve. Ann. Appl. Biol. 22: 134 - 167

Bliss,C.I. 1937. Calculation of the time-mortality curve. Ann. Appl. Biol. 24: 815 - 852

Bovee,E.C. 1949. Studies on the thermal death of Hyalella azteca saussure. Biol. Bull. 96: 123 - 128

Brawn,V.M. 1960. Temperature tolerance of unacclimated herring (Clupea harengus L.). J. Fish. Res. Bd. Can. 17: 721 - 723

Brett,J.R. 1944. Some lethal temperature relations of Algonquin park fishes. Univ. Toronto Stud. Biol. Ser. 52. Publ. Ont. Fish Res. Lab. 63

Brett,J.R. 1946. Rate of gain of heat-tolerance in gold fish (Carassius auratus). Univ. Toronto Studies Biol. Ser. 53: 1 - 28

Brett,J.R. 1952. Temperature tolerance in young Pacific salmon, genus Oncorhynchus. J. Fish. Res. Bd. Can. 9(6): 265 - 323

Brett,J.R. 1964. The respiratory metabolism and swimming performance of young sockeye salmon. J. Fish. Res. Bd. Can. 21: 1183 - 1226

Brett, J.R. 1970. Temperature: animals-fishes. In Marine Ecology. Vol. 1: Environmental factors, Part I (Ed.), O.Kinne. Wiley Interscience, London, pp. 515 - 560

Brown, J.H. and Feldmeth, C.R. 1971. Evolution in constant and fluctuating environments: Thermal tolerances of desert pup fish (Cyprinodon). Evolution, Vol.25, No.2, 390 - 398

Burrows, R.E. 1964. Effects of accumulated excretory products on hatchery reared salmonids. Bureau of Sport Fish and Wild Life. Res. Rep. No 66, 12p

Cairns, J.Jr. 1972. Coping with heated waste discharges from steam-electric power plants. Bioscience, Vol.22, No.7: 411 - 420

Charlon, N. 1968. Resistance due poisson-chat (<u>Ameiurus nebulosus</u> L.) a de brusques variations de temperature. Bull. Fran. De. Pis. Nos. 230 et 231

Charlon, N., Barbier, B. and Bonnet, L. 1970. Resistance de la truite arc-en-ciel (<u>Salmo gairdneri</u> Richardson) a des variations brusques de temperature. Ann. Hydrobiol. <u>1</u>(1): 73 - 89

Cockling,A.W. 1959. The effect of high temperature on roach (Rutilus rutilus) J. Exp. Biol. 36:217-226

Coutant,C.C. 1970. Biological aspects of thermal pollution I.Entrainment and discharge canal effects. CRC Critical Reviews in environmental control. 1(3): 341 - 382

Craigie,D.E. 1963. An effect of water hardness in the thermal resistance of the rainbow trout, Salmo gairdneri Richardson. Can. J. Zool. 41: 825 - 830

Doudoroff,P. 1942. The resistance and acclimatization of marine fishes to temperature changes. 1. Experiments with Girella nigricans (Ayres). Biol. Bull. 83: 219 - 244

Doudoroff,P. 1945. The resistance and acclimatization of marine fishes to temperature changes 2. Expements with Fundulus and Atherinops. Biol. Bull 88(2). 194 - 206

Edney,E.B. 1964. Acclimation to temperature in terrestrial isopods. I. Lethal temperatures. Physiol. Zool. 37: 364 - 377

Fahmy,F.K. 1972. Heterogenous acclimation of fish to temperature. Can. J. Zool. 50(7): 1035 - 1037

Fahmy,F.K. 1973. Effect of partial exposure to lethal temperature on heterogenously acclimated fish. Can. J. Zool. 51(12): 1249 - 1255

'Feldmeth,C.R., and Waggoner,J.P. III. 1972. Field measurement of tolerance to extreme hypersalinity in the California, *Fundulus parvipinnis*. Copeia, No.3: 592 - 594

Feldmeth,C.R., Stone,E.A. and Brown,J.H. 1974. An increased scope for thermal tolerance upon acclimation pupfish (*Cyprinodon*) to cycling temperatures. J. Comp. Physiol. 89: 39 - 44

·Fisher,R.A. 1945. The design of experiments, 4th ed. Edinburgh: Oliver and Boyd

Fisher,R.A. 1958. 'Statistical methods for research workers', 13th ed. Oliver and Boyd, Edinburgh and London

Forrester,C.R. and Alderdice,D.F. 1966. Effects of salinity and temperature on embryonic development of the Pacific cod (*Godus macrocephalus*). J. Fish

Fry, F.E.J., Brett, J.R. and Clauson, G.H. 1942. Lethal limits of temperature for young goldfish. Rev. Can. de Biol. 1: 50 - 56

Fry, F.E.J., Hart, J.S. and Walker, K.F. 1946. Lethal temperature relations for a sample of young speckled trout, Salvelinus fontinalis. Univ. Toronto Stud. Biol. Ser. No.54 (Publ. Ont. Fish Res. Lab. No.66) 9 - 35

Fry, F.E.J. 1947. Effects of the environment on animal activity. Univ. Toronto. Stud. Biol. Ser. No.55, (Pub. Ont. Fish Res. Lab No.68) 62 pp

Fry, F.E.J. and Hart, J.S. 1948. The relation of temperature to oxygen consumption in the goldfish. Biol. Bull. 94: 66 - 67

Fry, F.E.J. 1957. The lethal temperature as a tool in taxonomy. Annee Biol. 33: 205 - 219. (also in: Colloques Un. Int. Sci. Biol. 24, 1958)

Fry, F.E.J. 1971. The effect of environmental factors on the physiology of fish. In: Fish Physiology, Vol.VI, (Eds) W.S.Hoar and D.J.Randall, Academic Press Inc. (London) Ltd. pp. 1 - 98

Garside,E.T. and Jordan,C.M. 1968. Upper lethal temperature at various levels of salinity in the euryhaline cyprinidonts Fundulus heteroclitus and F. diaphanus. J. Fish. Res. Bd. Can. 25:2717-2720

Garside,E.T. and Chin-Yun-Kee,Z.K. 1972. Influence of osmotic stress on upper lethal temperatures in the cyprinodontid fish Fundulus heteroclitus (L.) Can. J. Zool. 50: 787 - 791

Gibson,M.B. 1954. Upper lethal temperature relations of the guppy, Lebistes reticulatus. Can. J. Zool. 36: 393 - 407

Hart,J.S. 1947. Lethal temperature relations of certain fish of the Toronto region. Trans. Roy. Soc. Can. 41(3): 57 - 71

Hart,J.S. 1949. Geographic variations of some physiological and morphological characteristics in fish. Ph.D Thesis, Dept. Zool. Univ. Toronto. 175 pp

Hart,J.S. 1952. Geographic variations of some physiological and morphological characters in certain fresh water fish. Univ. Toronto Biol. Ser. 60: 1 - 79

Hathaway,E.S. 1927. Quantitative study of the changes produced by acclimation on the tolerance of high temperature by fishes and amphibians. Bull. U.S. Bur. Fish. 43(2): 169 - 192

Heath,W.G. 1967. Ecological significance of temperature tolerance in Gulf of California shore fishes. J. Ariz. Acad. Sci. 43: 172 - 178

Heilbrunn,L.V. 1943. An outline of general physiology 2nd (Ed) Philadelphia. W.B.Saunders, 748 pp

Hickling,C.F. 1971. Fish Culture. 2nd (Ed). London. Faber and Faber

Hidu,H., Roosenburg,W.H., Drobeck,K G., McErlean,A.J. and Mihursky,J.A. 1974. Thermal tolerance of oyster larvae, *Crassostrea virginica* Gmelin, as related to power plant operation. Proc. Nat. shellfisheries Assoc. Vol.64: 102 - 110

Hoff,J.G. and Westman,J.R. 1966. The temperature tolerance of three species of marine fishes. J. Mar. Res. (Sears Found Marine Res.). 24:131 - 140

Holland,W.E., Smith,M.H., Gibbons,J.W. and Brown,D.H.1974. Thermal tolerance of fish from a reservoir receiving heated effluent from a nuclear reservoir.

Kennedy,V.S., Roosenburg,W.H., Zion,H.H. and Castagna,M. 1974a. Temperature-time relationship for survival of embryos and larvae of *Mulinia lateralis* (Mollusca: Bivalvia). Mar. Biol. 24: 137 - 145

Kennedy,V.S., Roosenburg,W.H., Castagna,M. and Mihursky,J.A. 1974b. *Mercenaria mercenaria* (Mollusca:Bivalvia). Temperature relationships for survival of embryos and larvae. Fish.Bull. Vol.72: 1160 - 1166

Khlebovich,V.V. 1962. Peculiarities of the aquatic fauna composition in relation to the salinity of the medium (Russ). Zh. Obshch. Biol 23(2): 90 - 97

Khlebovich,V.V. 1968. Some peculiar features of the hydrochemical regime and the fauna of mesohaline waters. Mar. Biol. 2: 47 - 49

Kawamoto,N.Y. 1961. The influence of excretory substances of fishes on their own growth. Progressive Fish Culturist. 23: 70 - 75

Kutty,M.N. 1966. Some studies on the respiratory quotient in gold fish and rainbow trout. Ph.D Thesis. Univ. Toronto. Toronto. Ontario. vii + 104b (Nat. Lib. Can. Canadian Theses on microfilm No.646)

Kutty,M.N. 1968. Respiratory quotient in gold fish and rainbow trout. J. Fish. Res. Bd. Can. 25:1689-1728

Kutty,M.N. and Murugapoopathy,G. 1969. Low lethal temperatures of the fishes *Liza macrolepis* (Smith) and *Tilapia mossambica* (Peters). Cur. Sci. 38(7): 171 - 172

Kutty,M.N. and Sukumaran,N. 1975. Influence of upper and lower temperature extremes on the swimming performance of *Tilapia mossambica*. Trans. Am. Fish. Soc. Vol.104, No.4: 755 - 761

Kutty,M.N., Narayanan,M. and Sukumaran,N. 1976. Use of a biological filter for removal of ammonia in small aquaria. J. Mad. Univ. Vol.V. 61 - 65

Lewis,R.M. 1965. The effect of minimum temperatures on the survival of larval Atlantic menhaden (*Brevoortia tyrannus*). Trans. Am. Fish. Soc. 94: 409 - 412

Lewis,R.M. 1966. Effect of salinity and temperature on survival and development of larval Atlantic menhaden (*Brevoortia tyrannus*). Trans. Am. Fish. Soc. 95: 423 - 425

Liu,C.K. 1942. Sinensina, 13: 17 - 20

Liu,C.K. 1944. Nature, London, 153:252

Lloyd,R. 1961. The toxicity of ammonia to rainbow trout (Salmo gairdneri, Richardson). Water waste treat. J. 8: 278 - 279

Loeb,J. and Wasteneys,H. 1912. On the adaptation of fish (Fundulus) to higher temperatures. J. Exp. Zool. 12: 543 - 557

Mackay,W.C. 1974. Effect of temperature on osmotic and ionic regulation in gold fish, Carassius auratus L. J. Comp. Physiol. 88: 1 - 19

Maurel,E. and Lagriffe. 1899. Action comparee de la charleur et du froid sur poissons. C.R. Soc. Biol. 51: 915 - 918

McErlean,A.J., Mihursky,J.A. and Brinkley,H.J. 1969. Determination of upper temperature tolerance triangles for aquatic organisms. Chesapeake Science, Vol.10, No.3 and 4, 293 - 296

McLeese,D.W. 1956. Effects of temperature, salinity and oxygen on the survival of the American lobster J. Fish. Res. Bd. Can. 13: 247 - 272

Miller,L.C. and Tainter,M.L. 1944. Estimation of ED50 and its error by means of logarithmic-probit graph paper. Proc. Soc. Exp. Biol. and Med. 57: 261 - 4

Morris,R. 1960. General problems of osmoregulation with special reference to cyclostomes. Symp. Zool. Soc. London. 1: 1 - 16

Morris,R.W. 1967. High respiratory quotients of two species of bony fishes. Physiol. Zool. 40:409-423

Motwani,M.P., Jayaram,K.C. and Sehgal,K.L. 1962. Fish and fisheries of Brahmaputra river system, Assam 1. Fish fauna with observations on their zoogeographical significance. Trop. Ecol. 3(1-2): 17 - 43

Narayanan,M. 1974. Studies on the biology of the mullet, *Rhinomugil corsula* (Hamilton). Ph.D Thesis, Mad. Univ. Madurai. India.

Nelson,J.S. 1968. Salinity tolerance of brook sticklebacks, *Culaea inconstans*, fresh water nine spine stickle backs, *Pungitius pungitius*, and freshwater four spine stickle backs, *Apeltes quadracus* Can. J. Zool. Vol.46: 663 - 667

Otto, R.G. and O'Hara Rice, J. 1977. Response of a freshwater sculpin (*Cottus cognatus gracilis*) to temperature. Trans. Am. Fish. Soc. Vol.106(1):89-94

Pakrasi, B. and Alikunhi, K.H. 1952. On the development of the fresh water grey mullet, *Mugil corsula* (Hamilton). J. Zool. Soc. Ind. 4(2): 123-140

Pandian, AL P. 1967. Studies on the effects of temperature on the activity of some poikilotherms. Ph.D Thesis submitted to Univ. of Madras , India

Panikkar, N.K., and Tampi, P.R.S. 1954. On the mouth breeding cichlid *Tilapia mossambica* Peters. Ind. J. Fish. 1(1 and 2): 217 - 230

Parvatheswararao, V. 1970. Adaptation to osmotic stress in fishes. Indian Biologist. Vol.2, No.1:16 - 36

Pitt, T.K., Garside, E.T. and Hepburn, R.L. 1956. Temperature selection of the carp (*Cyprinus carpio* Linn) Can. J. Zool. 34: 555 - 557

Pitkow, R.B. 1960. Cold death in the guppy. Biol. Bull 119: 231 - 245

Potts, W.T. and Parry, G. 1964. Osmotic and Ionic Regulation in Animals. Macmillan, New York.

Potts,W.T.W. and Evans,D.H. 1967. Sodium and chloride balance in the killifish Fundulus heteroclitus. Biol. Bull. mar. Biol. Lab. Woods Hole. 133: 411-425

Potts,D.C. and Morris,R.W. 1968. Some body fluid characteristics of the Antarctic fish, Trematomus bernacchii. Mar. Biol. 1: 269 - 276

Prosser,C.E. and Brown,F.A.Jr. 1965. Comparative Animal Physiology. Saunders, Philadelphia,

Prosser,C.L., Mackay,W. and Kato,K. 1970. Osmotic and ionic concentrations in some Alaskan fish and goldfish from different temperatures. Physiol. Zool. Vol.43, No.2: 81 - 89

Raj,B.S. 1954. The problem of the apparent discontinuous distribution of Harpodon nehereus (Hamilton) Proc. Ind. Acad. Sci. 40: 58 - 68

Rao,G.M.M. 1968. Oxygen consumption of rainbow trout (Salmo gairdneri) in relation to activity and salinity. Can. J. Zool. 46: 781 - 786

Rao,G.M.M. 1969. Effect of activity, salinity and temperature on plasma concentration of rainbow trout. Can. J. Zool. 47: 131 - 134

Rao,G.M.M. 1971. Influence of activity and salinity on the weight-dependent oxygen consumption of the rainbow trout Salmo gairdneri. Int. J. on Life in oceans and coastal waters, Vol.8, No.3: 205-212

Regnard,P. 1895. Action des tres basses temperatures sur les animaux aquatiques. C.R.Soc. Biol. 2: 652 - 653

Reynolds,W.W. and Thomson,D.A. 1974. Temperature and salinity tolerance of young Gulf of California Grunion, Leuresthes sardina (Atheriniformes: Atharinidae). J. Mar. Res. Vol.32(1): 37 - 45

Reynolds,W.W. 1975. Salinity tolerance of the tidepool shrimp Palaemon ritteri Holms. Comp. Biochem. Physiol. Vol.52A: 665 - 667

Saeki,A. 1958. Studies on fish culture in filtered closed circulation aquaria. Bull. Jap. Soc. Sci. Fish. 23: 684 - 695 (Def. Res. Bd. Trans by E.R.Hope, 777. J. 15p. 1964)

Schlieper,C., Blasing,J. and Halsband,E. 1952. Experimentelle Veranderungeu der temperatur toleranz bei stenothermen und eurythermen Wassertieren. Zool. Anz. 149: 163 - 169

Rao,G.M.M. 1971. Influence of activity and salinity on the weight-dependent oxygen consumption of the rainbow trout *Salmo gairdneri*. Int. J. on Life in oceans and coastal waters, Vol.8, No.3: 205-212

Regnard,P. 1895. Action des tres basses temperatures sur les animaux aquatiques. C.R.Soc. Biol. *2*: 652 - 653

Reynolds,W.W. and Thomson,D.A. 1974. Temperature and salinity tolerance of young Gulf of California Grunion, *Leuresthes sardina* (Atheriniformes: Atharinidae). J. Mar. Res. Vol.*32*(1): 37 - 45

Reynolds,W.W. 1975. Salinity tolerance of the tidepool shrimp *Palaemon ritteri* Holms. Comp. Biochem. Physiol. Vol.52A: 665 - 667

Saeki,A. 1958. Studies on fish culture in filtered closed circulation aquaria. Bull. Jap. Soc. Sci. Fish. *23*: 684 - 695 (Def. Res. Bd. Trans by E.R.Hope, 777. J. 15p. 1964)

Schlieper,C., Blasing,J. and Halsband,E. 1952. Experimentelle Veranderungeu der temperatur toleranz bei stenothermen und eurythermen Wassertieren. Zool. Anz. *149*: 163 - 169

Snedecor,G.W. and Cochran,W.G. 1967. Statistical methods. 6th Edn. Oxford and IBH. Publishing Co., New Delhi

Somero,G.N. and Vries,A.L. DE. 1967. Temperature tolerance of some Antarctic fishes. Science N.Y. 156:257-258

Spaas,J.T. 1960. Contribution to the comparative physiology and genetics of the European salmonidae. 3. Temperature resistance at different ages. Hydrobiologia, 15: 78 - 88

Strawn,K. and Dunn,J.E. 1967. Resistance of Texas salt and freshwater marsh fishes to heat death at various salinities. Tex. J. Sci. 19: 57 - 76

Sumner,F.B. and Doudoroff,P. 1938. Some experiments upon temperature acclimatization and respiratory metabolism in fishes. Biol. Bull. 74: 403 - 429

Timet,D. 1963. Studies on heat resistance in marine fishes. I. Upper lethal limits in different species of the Adriatic littoral. Thalassia jugosl. 2: 5 - 17

Tsukuda,H. 1960. Heat and cold tolerances in relation to body size in the guppy (*Lebistes reticulatus*) J. Inst. Polytech. Osaka Cy Univ. (Ser D) 11: 55 - 62

Tyler,A.V. 1966. Some lethal temperature relations of two minnows of the genus Chrosomus. Can. J. Zool. 44: 349 - 364

Vernberg,F.J. and Vernberg,W.B. 1971. In: 'Chemical Zoology'. Vol.VI. Part B. p.347 (Eds) M Florkin and B.T.Scheer, Academic Press, New York.

Vernon,H.M. 1899. The death temperature of certain marine organisms. J. Physiol. 25: 136

Wallis, 1975. Thermal tolerance of Mytilus edulis of Eastern Australia. Mar. Biol. 30: 183 - 191

Wikgren,B.J. 1953. Osmotic regulation in some aquatic animals with particular respect to temperature Acta Zool. Fenn. 71: 1 - 93

Yates,F. 1937. The design and analysis of factorial experiments. Harpenden : Imperial Bureau of Soil Science.

Tyler, A.V. 1966. Some lethal temperature relations of two minnows of the genus Chrosomus. Can. J. Zool. 44: 349 - 364

Vernberg, F.J. and Vernberg, W.B. 1971. In: 'Chemical Zoology'. Vol.VI. Part B. p.347 (Eds) M Florkin and B.T.Scheer, Academic Press, New York.

Vernon, H.M. 1899. The death temperature of certain marine organisms. J. Physiol. 25: 136

Wallis, 1975. Thermal tolerance of Mytilus edulis of Eastern Australia. Mar. Biol. 30: 183 - 191

Wikgren, B.J. 1953. Osmotic regulation in some aquatic animals with particular respect to temperature Acta Zool. Fenn. 71: 1 - 93

Yates, F. 1937. The design and analysis of factorial experiments. Harpenden : Imperial Bureau of Soil Science.

APPENDICES

Appendices explanation

The appendices are divisible into 3 major categor A, B and C, as explained below:

A = Experiments on high lethal temperatures
B = Experiments on low lethal temperatures
C = Experiments on salinity tolerance and salinity effect on thermal resistance

Each of these higher categories are further divided into divisions, a, b, c, d, e and f, denoting the species of fish studied, namely, *Cirrhinus mrigala*, *Labeo rohita*, *Labeo fimbriatus*, *Cyprinus carpio*, *Rhinomugil corsula* and *Tilapia mossambica* respectively.

The divisions are subsequently split into a1, a2, a3 and so on, each denoting particular acclimation temperature or salinity as the case may be.

To give an example, Aa1 denotes data obtained from tests at high lethal temperatures on mrigal acclimated to 38°C.

Appendix Aa1

Time to death (min), total length (cm) and weight (mg) of individual fish mrigal (Cirrhinus mrigala) acclimated to 38°C and tested at different high temperatures

Temp.(°C) 42.5			42.0			41.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
15	4.2	570	34	4.2	610	73	4.2	620
21	4.3	695	36	4.0	555	82	4.3	670
23	4.4	705	41	4.0	540	93	4.1	565
24	4.5	750	43	4.1	585	100	4.4	740
25	4.6	815	46	4.6	710	102	4.4	725
27	4.0	595	51	4.4	730	110	4.6	805
33	4.2	610	53	4.5	685	113	4.5	880
47	4.6	830	59	4.2	670	118	4.8	1010
49	4.5	740	64	4.3	585	126	4.6	805
69	4.5	690	67	4.1	590	127	4.6	855

contd...

Appendix Aa1 contd...

40.5			40.0			39.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
213	4.3	760	804	4.4	615	4385	3.9	530
248	4.3	720	1030	4.2	575	5182	4.4	715
259	4.6	875	1512	4.6	780	7272	4.2	585
273	4.5	710	1774	4.0	590	7832	4.5	690
298	4.0	605	2308	4.2	605	7997	4.9	845
321	4.3	685	2505	4.6	785	8712	4.8	815
330	4.5	835	2833	4.3	695	No death		
361	4.5	845	3294	4.5	740			
403	4.5	695	3799	4.5	705			
454	4.4	705	4566	4.6	835			

n (min), total length (cm) and weight (mg) of individual fish mrigal, rigala) acclimated to 35°C and tested at different high temperatures

42.0		41.0			40.5		
otal ength	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
4.0	530	58	4.1	565	85	4.1	565
4.2	570	61	4.3	630	92	4.2	585
4.0	505	62	4.1	580	95	4.0	505
4.0	530	64	5.0	1205	107	3.9	485
4.1	590	67	4.5	815	126	4.1	580
4.0	570	71	4.0	510	134	4.0	565
3.8	525	73	4.0	595	140	4.0	530
4.6	805	74	4.1	620	147	3.9	525
4.3	700	77	4.3	710	153	4.4	715
4.6	795	82	4.4	725	162	4.6	810

Appendix Aa2 contd...

40.0			39.5			39.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
166	4.4	790	4072	4.9	1018	No death		
187	4.6	880	4732	4.2	495			
231	4.5	880	5605	4.8	780			
243	4.8	1030	6539	4.2	470			
254	4.5	835	6833	4.7	750			
460	4.4	740	7272	4.4	580			
676	4.6	855	7402	4.6	730			
1754	4.7	925	8637	4.5	690			
2615	4.3	655	8754	4.5	710			
5086	4.6	805	9021	4.4	650			

Appendix Aa3

Time to death (min), total length (cm) and weight (mg) of individual mrigal, *Cirrhinus mrigala* acclimated to 30°C and tested at different high lethal temperatures in fresh water

Temp.(°C) 41.0			40.5			40.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
13	3.7	415	42	4.1	695	94	4.2	710
15	4.3	665	47	4.2	710	103	4.5	885
20	4.1	615	54	4.5	865	107	4.2	670
22	3.4	290	66	4.2	670	113	4.6	940
23	3.8	460	69	4.6	910	115	3.9	565
24	4.1	585	70	4.2	745	117	4.3	745
24	4.3	620	71	4.1	650	121	4.1	650
25	4.3	660	74	3.9	520	126	3.8	535
29	4.0	550	86	3.8	550	128	3.9	595
30	4.7	810	95	4.0	505	131	4.0	565

contd...

Appendix **Aa**3 contd...

Temp.(°C) 39.5			39.0			38.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
161	4.3	760	451	4.2	545	No death		
217	4.4	720	538	4.4	650			
223	4.6	880	5262	4.3	590			
238	4.5	710	5287	4.0	505			
240	4.0	610	5302	4.2	540			
273	4.2	650	6258	4.4	630			
282	4.6	835	6412	3.8	350			
296	4.6	875	6972	4.2	560			
353	4.5	695	7927	3.9	355			
1351	4.4	710	8279	4.0	490			

Appendix Aa5

Time to death (min), total length (cm) and weight (mg) of individual mrigal, *Cirrhinus mrigala* acclimated to 20°C and tested at different high lethal temperatures in fresh water

Temp.(°C) 38.0			37.5			37.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
4	4.2	630	6	3.8	430	25	4.4	760
5	4.9	960	17	4.6	835	31	4.4	770
13	4.1	665	19	4.6	845	38	4.0	595
14	4.7	880	23	4.8	930	46	4.7	860
15	4.4	705	25	4.1	575	53	4.2	740
16	4.8	960	26	4.3	625	56	4.3	770
17	4.2	600	30	4.4	755	70	3.9	560
18	4.0	600	31	4.6	850	80	4.3	775
19	5.1	1280	34	4.2	635	83	4.1	590
20	4.3	610	77	3.9	500	91	4.2	655

Temp.(°C) 36.5

Time to death	Total length	Weight
47	4.9	1050
52	4.2	715
53	4.6	990
56	4.5	740
No death		

Appendix Aa6

Time to death (min), total length (cm) and weight (mg) of individual mrigal *Cirrhinus mrigala* acclimated to 15°C and tested at different high lethal temperatures in fresh water

Temp.(°C) 36.0		
Time to death	Total length	Weight
7	4.0	520
11	4.7	805
13	4.0	535
14	4.6	740
15	4.4	705
18	4.2	595
19	4.8	950
20	4.0	580
22	5.2	1285
23	4.2	580

35.5		
Time to death	Total length	Weight
13	4.5	780
22	3.8	430
27	4.6	810
33	4.8	925
35	4.1	560
44	4.2	595
46	4.3	625
47	4.4	705
54	4.6	850
76	4.0	570

35.0		
Time to death	Total length	Weight
50	4.1	575
57	4.7	780
70	3.9	510
85	5.1	1095
101	4.3	690
145	4.4	785
No death		

34.5		
Time to death	Total length	Weight
No death		

Appendix Ab1

Time to death (min), total length (cm) and weight (mg) of individual rohita *Labeo rohita* acclimated to 38°C and tested at different high lethal temperatures in fresh water

Temp.(°C) 42.0			41.0			40.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
13	4.6	920	79	4.4	1030	428	4.2	855
22	4.2	870	107	5.4	1960	455	4.4	1025
31	7.3	3600	151	4.6	1265	600	5.3	1920
35	5.4	1405	187	5.1	1495	723	4.5	1105
42	6.2	1250	227	4.5	1120	779	5.1	1490
51	4.5	950	291	5.0	1220	817	4.5	1000
65	4.4	910	301	4.1	845	853	4.2	890
73	5.2	1155	317	5.2	1320	892	5.2	1400
			453	6.0	2155	1121	5.3	1870
						1266	5.9	1985

Appendix Ab1 contd . . .

Temp.(°C) 40.0

Time to death	Total length	Weight
3590	4.5	905
3822	4.2	890
4497	7.3	3020
4653	5.4	1280
5897	4.7	950
6404	4.4	720
6928	5.2	985
7305	5.2	1030
8416	4.8	920
8566	5.4	1010

39.5

Time to death	Total length	Weight
3810	4.5	1050
4379	4.2	895
5467	5.2	1185
5668	5.3	1450
6986	4.6	905
7571	4.4	720
8235	5.3	1050
9210	5.3	1250
9601	4.8	985
9943	4.9	1005

39.0

Time to death	Total length	Weight
4209	4.1	980
4618	4.4	1080
6326	5.4	1725
6890	4.5	1005
8517	5.0	1185
8836	4.1	825
No death		

Temp.(°C) 38.5

Time to death	Total length	Weight
None died		

Appendix Ab2

Time to death (min), total length (cm) and weight (mg) of individual rohu, *Labeo rohita* acclimated to 35°C and tested at different high lethal temperatures in fresh water

Temp.(°C) 43.0			42.0			41.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
1	3.1	320	10	3.3	350	54	4.1	650
3	3.0	310	12	3.2	280	76	3.7	420
4	3.4	380	15	3.3	310	92	3.8	540
5	3.3	345	17	3.8	480	94	3.2	340
5	3.7	395	20	3.5	360	96	3.0	305
6	2.9	275	22	3.9	460	100	3.3	355
7	4.2	475	23	2.4	140	104	3.0	270
8	3.9	445	24	3.0	250	106	2.9	245
10	3.8	510	26	2.4	135	109	3.2	315
14	3.2	295	29	4.5	830	119	3.0	255

contd...

Appendix Ab2 contd...

Temp.(°C) 40.5			40.0			39.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
127	2.4	140	3129	2.3	120	2209	3.2	305
158	3.5	325	3292	3.6	345	2778	3.3	350
176	2.9	245	3417	2.9	245	3349	3.2	280
181	4.3	750	3583	4.3	990	3570	3.3	310
201	3.8	380	4214	3.8	380	4552	3.8	475
208	3.1	285	4550	3.2	280	5724	3.4	360
214	2.9	220	4857	3.0	220	6846	3.9	460
223	3.4	315	5425	3.4	320	7280	2.4	165
226	3.4	320	5473	3.5	395	7543	3.0	250
256	3.5	395				8492	2.4	135

contd...

Appendix Ab2 contd...

Temp.(°C) 40.5			40.0			39.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
127	2.4	140	3129	2.3	120	2209	3.2	305
158	3.5	325	3292	3.6	345	2778	3.3	350
176	2.9	245	3417	2.9	245	3349	3.2	280
181	4.3	750	3583	4.3	990	3570	3.3	310
201	3.8	380	4214	3.8	380	4552	3.8	475
208	3.1	285	4550	3.2	280	5724	3.4	360
214	2.9	220	4857	3.0	220	6846	3.9	460
223	3.4	315	5425	3.4	320	7280	2.4	165
226	3.4	320	5473	3.5	395	7543	3.0	250
256	3.5	395				8492	2.4	135

contd...

Appendix Ab2 contd . . .

Temp.(°C) 39.0			38.5		
Time to death	Total length	Weight	Time to death	Total length	Weight
3868	2.1	75	None died		
6119	3.2	215			
6942	3.0	205			
7792	3.2	200			
8428	3.9	470			
No death					

Appendix Ab3

Time to death (min), total length (cm) and weight (mg) of individual rohu, *Labeo rohita* acclimated to 30°C and tested at different high lethal temperatures in fresh water

Temp.(°C)	41.0		40.5			40.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
12	2.9	270	29	2.8	265	49	2.1	155
13	3.5	460	33	3.5	456	65	2.9	190
14	3.0	280	36	3.1	395	77	3.2	295
15	3.2	350	38	3.0	290	80	3.3	370
16	3.2	310	40	3.2	352	89	3.4	385
17	3.1	350	44	3.1	310	95	2.7	185
19	2.9	260	48	3.1	350	101	2.9	245
19	3.0	275	56	2.9	275	109	2.9	230
20	2.9	255	59	2.9	250	115	3.1	260
20	2.8	260	61	2.8	260	138	3.4	360

contd...

Appendix Ab3 contd...

Temp.(°C) 39.5		
Time to death	Total length	Weight
760	3.7	520
813	3.9	560
1886	4.4	720
1923	4.8	590
1970	3.9	540
1996	4.2	590
2082	3.5	450
2203	4.2	570
2238	4.5	790
2300	3.2	395

39.0		
Time to death	Total length	Weight
2402	2.4	175
2592	3.4	325
2734	2.8	245
3024	3.9	540
3637	3.8	410
4354	2.7	290
4586	3.0	250
5062	3.3	340
5349	3.4	390
5682	3.4	360

38.5		
Time to death	Total length	Weight
4615	2.5	175
5088	3.0	255
6195	3.0	235
7184	3.7	450
8302	3.4	395
8786	3.8	465
No death		

38.0		
Time to death	Total length	Weight
None died		

Appendix Ab4

Time to death (min) total length (cm)&weight (mg) of individual rohu, *Labeo rohita* acclimated to 25°C and tested at different high lethal temperatures in fresh water

Temp.(°C) 39.5			39.0			38.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
5	3.1	320	13	2.5	110	18	5.0	1120
11	3.3	390	15	2.7	180	79	3.2	295
19	2.9	290	22	3.3	360	97	3.3	370
24	3.4	420	50	3.5	400	123	4.4	720
27	2.8	260	72	3.5	425	169	4.8	590
31	3.0	305	97	3.0	280	202	3.9	540
42	3.3	395	109	4.0	610	277	3.8	410
50	3.2	340	142	3.9	570	318	3.0	235
67	2.7	240	173	3.7	530	352	3.7	450
72	2.5	170	229	3.9	595	375	3.9	510

contd...

Appendix Ab4 contd...

Temp.(°C)	38.0			37.5	
Time to death	Total length	Weight	Time to death	Total length	Weight
188	3.0	305	None died		
220	3.3	395			
431	3.0	310			
555	2.8	260			
626	2.9	290			
697	3.1	315			
940	2.9	295			
No death					

Appendix Ab5

Time to death (min), total length (cm) and weight (mg) of individual rohu *Labeo rohita* acclimated to 20°C and tested at different high lethal temperatures in fresh water

Temp.(°C) 38.0			37.0			36.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
6	4.4	820	10	3.8	890	21	3.9	685
7	3.5	455	21	3.9	610	31	4.7	1100
8	3.7	540	26	2.7	690	40	3.7	625
9	3.3	390	30	2.6	555	57	4.0	780
10	4.0	700	34	3.6	500	66	3.0	340
10	5.2	1420	134	5.2	1560	192	3.5	570
16	3.3	400	141	3.5	475	216	4.5	1615
21	4.3	850	157	3.7	620	307	4.5	1705
27	3.5	445	180	3.5	510	322	3.6	675
45	4.5	890	192	3.8	710	340	4.3	1220

contd...

Appendix Ab5 contd . . .

Temp.(°C) 36.0			35.5			35.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
75	3.0	365	478	3.7	540	None died		
120	3.2	470	531	3.9	620			
157	4.9	1290	No death					
233	3.7	625						
281	4.1	720						
No death								

Appendix Ab6

Time to death (min), total length (cm) and weight (mg) of individual rohu *Labeo rohita* acclimated to 15°C and tested at different high lethal temperatures in fresh water

Temp.(°C, 36.0

Time to death	Total length	Weight
10	4.9	1100
11	4.5	945
13	4.6	980
16	3.6	535
21	5.0	1210
23	5.6	1750
24	5.7	1725
32	5.2	1510
36	3.9	695
53	3.6	470

35.5

Time to death	Total length	Weight
14	4.0	710
16	4.2	805
21	4.9	1090
110	3.4	620
182	4.3	795
239	4.6	920
283	5.2	1395
318	3.9	695
334	4.1	720
407	3.6	470

35.0

Time to death	Total length	Weight
35	4.0	730
66	4.4	825
445	5.2	1590
649	3.2	620
836	4.6	920
No death		

Temp.(°C) 34.5

Time to death	Total length	Weight
None died		

Appendix Ac1

Time to death (min), total length (cm) and weight (mg) of individual fringe-lipped carp *Labeo fimbriatus* acclimated to 38°C and tested at different high lethal temperatures in fresh water

Temp.(°C)	42.0			41.0			40.5	
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
39	3.6	590	236	3.5	550	762	2.9	275
41	3.7	640	254	3.6	545	924	3.4	470
42	3.4	475	273	3.7	590	1128	3.2	400
44	3.4	500	281	3.8	635	1159	3.9	610
46	2.7	270	284	3.0	310	1211	2.8	210
47	3.3	520	288	3.3	430	1258	3.3	390
50	3.2	410	314	2.7	210	1446	3.5	520
51	3.3	470	326	3.8	675	1467	3.8	595
52	3.5	560	336	3.5	535	1704	2.7	245
55	3.4	495	533	5.0	1180	2039	4.1	740

contd . . .

Appendix Ac1 contd...

Temp.(°C) 40.0

Time to death	Total length	Weight
1240	3.1	300
1745	3.0	285
2787	4.2	830
3345	3.1	380
3803	3.1	375
4051	3.5	540
4181	3.6	495
4379	3.4	415
4713	3.5	475
4885	3.6	480

39.5

Time to death	Total length	Weight
4547	3.1	295
4853	2.9	245
5267	3.2	270
5822	2.9	260
5894	2.5	190
6222	2.7	200
6318	2.4	170
6594	2.8	210
6673	3.1	270
7008	3.2	275

39.0

Time to death	Total length	Weight
5366	2.9	245
6023	3.2	270
6709	2.9	260
7179	2.5	190
7615	2.7	200
7964	2.4	170
8460	2.8	210
8913	3.1	270
9440	3.3	295
No death		

38.5

Time to death	Total length	Weight
No death		

Appendix Ac2 contd...

Temp.(°C) 40.0

Time to death	Total length	Weight
2061	3.3	320
2122	2.9	290
2624	3.2	315
2825	2.8	245
3411	3.4	360
3482	3.1	310
3592	3.0	295
4189	3.2	300
4494	3.3	295
4833	3.4	350

39.5

Time to death	Total length	Weight
3433	3.1	320
4600	2.9	290
4829	3.3	315
5055	3.2	320
5877	2.8	245
6252	3.4	360
6652	3.1	310
7170	3.2	300
7516	3.3	295

39.0

Time to death	Total length	Weight
3719	3.2	345
4493	3.0	310
4961	3.1	310
6065	3.2	315
6910	2.9	300
7395	2.4	195
7890	3.0	285
8703	3.0	305
9013	3.3	300
9188	3.2	295

38.5

Time to death	Total length	Weight
None died		

Appendix Ac3

Time to death (min) total length (cm) and weight (mg) of individual fringe-lipped carp, Labeo fimbriatus acclimated to 30°C and tested at different high lethal temperatures in fresh water

Temp.(°C) 41.0			40.5			40.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
33	1.7	36	58	2.1	70	134	1.6	30
35	2.0	72	62	1.8	60	164	1.8	60
36	1.8	46	65	1.6	40	174	2.0	65
37	1.6	33	70	1.8	49	178	1.8	50
38	1.5	33	74	1.9	65	187	1.8	49
40	1.7	40	79	1.8	45	195	1.9	60
41	1.6	36	83	1.5	30	197	1.	60
44	1.7	41	90	1.8	50	220	1.5	30
45	1.9	69	95	1.7	45	231	1.8	47
57	1.9	62	107	1.6	30	341	1.8	48

contd...

Appendix Ac3 contd . . .

Temp.(°C) 39.5			39.0			38.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
213	2.8	275	2447	2.3	115	None died		
255	3.0	290	4141	2.5	150			
265	2.7	260	4659	2.6	160			
272	2.5	225	4828	3.0	265			
280	2.2	195	5893	2.9	240			
320	2.8	265	6477	2.7	205			
322	2.9	270	6914	3.1	205			
325	2.3	220	7358	3.0	195			
335	2.5	230	7680	2.8	175			
376	2.4	235	8070	2.8	190			

Appendix Ac4

Time to death (min), total length (cm) and weight (mg) of individual fringe-lipped carp, <u>Labeo fimbriatus</u> acclimated to 25°C and tested at different high lethal temperatures in fresh water

Temp.(°C) 40.0			39.5			39.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
9	2.7	150	17	3.4	350	32	2.8	275
11	2.3	110	19	3.0	245	67	3.0	295
12	2.7	170	40	3.0	275	80	2.7	260
13	2.6	170	44	2.8	210	111	2.5	225
14	2.3	95	46	2.8	240	116	2.7	265
15	3.1	275	49	3.3	290	137	2.3	220
16	2.1	70	50	3.1	320	191	2.9	270
17	2.1	80	51	2.6	200	234	3.1	310
19	2.2	85	53	3.3	380	243	2.9	295
21	2.5	135	59	2.8	225	292	2.6	265

contd...

Appendix Ac4 contd...

Temp.(°C) 38.5			38.0			37.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
164	2.2	120	1382	2.6	240	None died		
188	2.4	140	No death					
195	2.5	190						
231	3.0	255						
258	2.8	250						
310	2.6	185						
351	2.6	190						
448	3.1	335						
461	3.0	295						
2365	3.2	340						

Appendix Ac5

Time to death (min), total length (cm) and weight (mg) of individual fringe lipped carp, Labeo fimbriatus acclimated to 20°C and tested at different high lethal temperatures in fresh water

Temp.(°C)	38.5		38.0			37.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
4	2.6	145	4	2.1	120	5	2.2	115
4	2.5	140	5	2.4	140	7	2.2	120
5	2.4	140	6	2.5	195	23	2.5	150
6	2.5	195	6	3.0	260	25	2.7	240
8	3.0	255	17	2.7	245	27	2.8	245
10	2.8	245	21	2.8	255	29	3.2	335
11	2.6	205	22	2.6	195	30	3.1	330
12	2.9	310	25	3.1	330	39	2.7	200
15	2.9	305	34	3.0	305	51	2.7	220
17	3.2	350	38	3.2	345	No death		

contd...

Appendix Ac5 contd...

Temp.(°C) 37.0			36.5		
Time to death	Total length	Weight	Time to death	Total length	Weight
13	3.3	365	No death		
98	3.5	495			
1827	3.2	325			
No death					

pendix Ac6

me to death (min), total length (cm) and weight (mg) of individual fringe-lipped
rp *Labeo fimbriatus* acclimated to 15°C and tested at different high lethal tempe-
tures in fresh water

mp.(°C)	36.5		36.0			35.5		
me to eath	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
4	2.2	118	5	2.1	110	11	3.4	350
5	2.1	110	6	2.4	140	12	3.0	255
6	2.5	160	8	2.5	195	13	3.0	270
6	2.2	125	8	3.0	255	23	2.8	215
7	2.7	245	9	2.8	250	27	2.7	240
8	2.8	250	10	2.6	185	29	3.3	295
8	2.6	210	10	**2.6**	**185**	31	3.1	315
9	3.2	350	11	3.1	345	39	2.6	205
9	3.1	330	12	3.0	295	43	3.2	345
10	2.7	200	14	3.2	324	57	2.8	235

ontd...

Appendix Ac6 contd...

Temp.(°C) 35.0			34.5		
Time to death	Total length	Weight	Time to death	Total length	Weight
19	2.1	160	No death		
21	2.2	195			
34	2.4	190			
38	2.0	190			
44	2.9	280			
183	2.7	210			
218	2.4	205			
No death					

Appendix Ad1

Time to death (min), total length (cm) and weight (mg) of individual common carp, *Cyprinus carpio* (Bankok strain) acclimated to 35°C and tested at different high lethal temperatures in fresh water

Temp. (°C)	42.0			41.0			40.5	
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
22	2.7	210	50	2.9	330	70	2.3	145
24	3.0	295	51	2.8	260	76	2.6	205
27	2.7	225	52	3.0	370	82	2.4	150
28	3.2	340	54	2.6	203	88	2.4	155
30	3.1	335	55	2.6	175	93	2.6	235
31	2.5	190	57	3.0	344	97	2.5	240
32	2.8	240	58	2.5	198	98	2.4	182
32	2.9	265	61	3.0	291	101	2.3	140
34	3.1	400	65	2.7	276	106	2.5	175
35	2.7	240	67	3.2	419	113	2.6	180

contd...

Appendix Ad1 contd...

Temp.(°C) 40.0			39.5			39.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
81	2.2	128	1066	2.9	325	1412	2.5	195
98	2.6	188	1234	2.8	260	2020	2.7	230
179	2.3	146	1803	3.0	365	3070	3.0	310
216	2.3	153	1908	2.7	205	3410	2.9	255
260	2.6	237	1987	2.6	180	3857	2.6	205
292	2.6	240	2084	3.0	340	4236	2.7	275
446	2.5	182	2121	2.5	200	4584	2.5	205
491	2.2	133	2290	3.0	295	4609	3.0	290
536	2.5	175	2416	2.7	275	4857	2.7	250
691	2.6	178	2932	3.2	425	5006	3.1	376

contd...

Appendix **Ad1** contd...

Temp.(°C) 38.5			38.0		
Time to death	Total length	Weight	Time to death	Total length	Weight
3553	2.1	105	No death		
4187	2.0	95			
5142	2.4	100			
5827	2.2	122			
7383	2.1	112			
9381	2.2	103			
No death					

Appendix Ad2

Time to death (min), total length (cm) and weight (mg) of individual common carp, *Cyprinus carpio* (Bankok strain) acclimated to 30°C and tested at different high lethal temperatures in fresh water

Temp(°C)	41.5		41.0			40.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
7	2.7	170	22	2.9	250	28	2.2	105
11	2.7	280	24	3.1	315	41	1.9	65
14	2.6	175	28	2.7	225	46	2.6	155
15	2.8	255	29	3.3	395	50	2.1	80
16	2.8	235	30	3.1	350	52	2.15	90
17	2.9	270	31	3.4	475	55	2.5	140
18	2.[illegible]	275	31	3.1	405	57	2.5	160
19	2.7	230	32	3.1	340	64	2.35	115
19	2.4	140	34	3.1	400	71	2.55	220
20	2.7	195	34	2.7	215	75	2.3	125

contd...

Appendix Ad2 contd...

Temp.(ºC) 40.0			39.5			39.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
50	2.9	270	199	2.9	255	288	2.9	270
59	2.7	250	236	3.1	320	387	2.1	115
61	2.7	270	307	2.7	225	432	2.7	270
62	3.0	395	326	3.3	390	464	3.0	380
63	2.8	310	347	3.1	350	669	2.8	310
64	2.8	290	369	3.2	395	694	2.7	260
65	2.8	250	469	3.1	350	943	2.8	250
78	2.7	290	492	3.1	345	1032	2.6	235
83	3.1	395	554	3.1	400	1208	3.1	395
98	2.8	315	577	2.9	265	1250	2.5	215

contd...

Appendix Ad2 contd . . .

Temp.(°C)	38.5	
Time to death	Total length	Weight
403	2.6	165
489	2.7	295
575	2.1	178
744	2.1	110
800	3.0	235
933	2.9	250
2279	2.8	230
2756	2.4	115
3041	2.7	280
3843	2.5	215

	38.0	
Time to death	Total length	Weight
3751	2.8	245
4166	2.8	230
4252	2.6	190
4695	2.7	215
5542	2.3	125
7407	2.5	230
8652	2.4	170
No death		

	37.5	
Time to death	Total length	Weight
No death		

	37.0	
No death		

Appendix Ad3

Time to death (min), total length (cm) and weight (mg) of individual common carp *Cyprinus carpio* (Bankok strain) acclimated to 25°C and tested at different high lethal temperatures in fresh water

Temp.(°C) 39.5			39.0			38.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
3	1.8	85	4	2.1	120	13	1.9	90
4	1.7	42	5	2.0	110	14	2.0	105
7	1.8	90	6	1.9	70	22	2.2	125
8	2.0	118	9	1.9	80	26	2.0	110
11	1.9	75	21	1.6	60	35	1.9	90
12	1.9	90	24	1.8	80	48	2.2	128
19	2.0	110	41	1.8	85	62	1.9	85
21	2.0	115	56	2.1	120	74	1.5	50
25	2.5	125	71	2.2	160	105	1.7	60
32	1.7	55	73	1.8	95	160	1.8	85

contd...

Appendix Ad3 contd...

Temp.(°C) 38.0			37.5			37.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
17	1.8	75	26	1.7	69	36	1.8	80
23	1.9	93	34	1.8	68	55	1.6	39
30	2.1	112	38	1.0	49	110	1.9	90
38	1.9	90	58	1.9	92	132	2.0	118
61	1.9	91	67	1.9	73	142	1.9	70
64	2.2	123	99	2.1	118	177	2.0	110
69	1.9	83	404	2.0	97	577	2.0	112
76	1.6	50	433	2.1	120	755	2.6	127
86	1.7	56	474	1.8	98	1041	1.7	56
87	1.8	77	540	1.9	80	1980	1.9	65

contd...

Appendix Ad3 contd...

Temp.(°C)	36.5		36.0		
Time to death	Total length	Weight	Time to death	Total length	Weight
57	1.7	70	No death		
135	2.0	95			
168	1.9	140			
232	2.2	89			
475	1.9	92			
642	2.1	120			
None died					

Appendix Ad4

Time to death (min), total length (cm) and weight (mg) of individual common carp *Cyprinus carpio* (Bankok strain) acclimated to 20°C and tested at different high lethal temperatures in fresh water

Temp.(°C) 37.0			36.5			36.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
5	2.4	120	6	2.6	155	9	2.4	115
6	2.2	105	7	2.9	245	16	2.0	95
7	2.9	235	8	2.9	205	17	2.9	228
7	2.4	105	9	2.7	210	32	2.5	125
8	2.6	175	13	2.3	145	154	2.1	85
9	2.6	185	14	2.6	180	179	2.7	185
11	2.1	72	16	2.5	195	328	2.6	175
17	2.7	175	48	2.0	90	401	2.5	180
68	2.5	180	55	2.2	130	433	2.1	75
107	2.1	70	119	2.3	140	662	2.6	125

contd...

Appendix Ad4 contd . . .

Temp.(°C) 35.5			35.0		
Time to death	Total length	Weight	Time to death	Total length	Weight
16	2.7	205	61	2.2	90
19	2.7	185	95	2.3	145
21	2.4	155	107	2.6	162
150	2.6	150	No death		
526	2.7	210			
No death					

Appendix Ad5

Time to death (min), total length (cm) and weight (mg) of individual common carp *Cyprinus carpio* (Bankok strain) acclimated to 15°C and tested at different high lethal temperatures in fresh water

Temp.(°C) 35.5			35.0			34.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
3	2.7	185	7	2.5	145	22	2.4	105
4	2.7	205	8	2.7	225	67	2.2	125
5	2.4	155	10	2.9	205	80	2.3	115
7	2.3	170	11	2.6	205	268	2.4	175
13	2.9	325	13	2.4	155	313	2.9	285
24	2.8	220	200	2.5	175	337	2.7	175
31	2.7	260	212	2.4	185	416	2.5	185
46	3.1	355	234	2.0	90	No death		
58	2.8	255	247	2.1	120			

contd . . .

Appendix Ad5 contd...

Temp.(°C) 34.0			33.5		
Time to death	Total length	Weight	Time to death	Total length	Weight
157	2.3	115	No death		
234	2.9	235			
No death					

Appendix Ae1

Time to death (min), total length (cm) and weight (mg) of individual freshwater mullet, <u>Rhinomugil corsula</u> acclimated to 35°C and tested at different high lethal temperatures in fresh water

Temp.(°C) 42.0			41.0			40.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
19	12.3	14250	60	12.3	14550	149	11.5	10850
22	11.2	10550	76	11.6	12050	160	10.7	10800
25	11.5	11900	96	11.2	10000	178	11.8	11750
28	11.0	10050	100	11.1	10500	202	12.2	13950
32	10.8	10100	108	10.9	9900	230	10.4	7850
34	11.6	11750	116	12.5	16100	232	11.6	12100
40	12.5	15800	135	10.8	9800	241	12.0	13500
43	10.9	10200	152	9.9	7300	269	11.1	10900
52	11.8	12250	153	11.4	10900	299	11.4	10850
60	9.8	8550	175	9.0	5700	304	10.9	10400

contd...

Appendix Ae1 contd...

Temp.(°C) 40.0			39.5			39.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
193	10.5	7300	244	11.2	10700	241	10.6	9200
200	10.8	7900	266	10.9	8100	337	10.4	8900
253	12.7	14900	405	10.7	9350	524	10.3	9000
281	11.4	10300	418	11.5	10800	691	11.6	11700
284	11.5	10200	535	12.3	14050	850	11.5	10400
309	10.5	7800	557	11.6	10750	885	12.1	13900
317	12.5	14200	563	10.9	10100	925	11.4	13300
340	10.8	9700	587	12.0	13100	957	11.4	13400
348	13.5	19200	653	11.4	10750	998	10.3	10400
389	12.1	13000	776	12.1	12500	1032	9.6	7000

contd...

Appendix Ae1 contd...

Temp.(°C) 38.5			38.0			37.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
668	11.2	11900	1020	11.1	9800	2423	10.9	10500
774	11.6	13200	1528	11.7	12100	3211	10.0	7200
858	11.4	11950	2023	10.6	11400	3868	12.1	13600
1228	10.5	9500	2410	10.9	10000	4035	11.7	10380
1443	10.2	9350	2475	10.3	7100	4360	10.9	10060
1557	9.4	7500	2732	11.5	11300	4385	10.8	10040
1783	11.3	11100	2803	11.7	11200	4480	9.9	7600
1910	9.2	6900	2823	12.9	14700	4615	10.4	9100
1959	12.2	14950	3074	9.6	6200	4648	11.0	10090
2182	11.3	11700	3201	10.6	9000	4650	9.05	6040

contd...

Appendix Ae1 contd...

Temp.(°C) 36.5			36.0		
Time to death	Total length	Weight	Time to death	Total length	Weight
2475	11.1	10600	None died		
5738	11.3	10500			
6007	9.7	7900			
6431	10.2	8500			
9040	8.4	4650			
10040	9.9	6270			
11657	8.7	5200			
No death					

Appendix A.2

Time to death (min), total length (cm) and weight (mg) of individual freshwater mullet, *Rhinomugil corsula* acclimated to 30°C and tested at different high lethal temperatures in fresh water

Temp.(°C) 41.0			40.5			40.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
20	8.6	4600	35	10.7	9200	69	10.1	7800
28	12.7	16650	41	11.0	8950	78	10.7	9750
31	11.4	12550	48	10.5	8400	86	9.4	6400
40	11.6	13700	59	12.0	12650	89	11.6	11900
44	9.9	7500	73	9.8	6900	120	10.9	10600
47	10.3	8700	76	12.2	13700	127	11.1	13200
53	12.5	16150	80	10.9	10350	140	12.6	16300
61	10.5	8950	85	11.0	9100	159	11.9	14800
67	10.4	8600	111	10.4	7700	161	9.9	7750
			131	10.4	8300	171	11.5	12600

contd . . .

Appendix A.2 contd . . .

Temp.(°C) 39.5			39.0			38.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
178	10.8	9300	578	11.3	12050	933	10.5	9000
252	10.9	8800	598	11.5	13200	1076	10.4	8800
285	10.4	8350	617	11.5	12300	1220	10.5	9260
302	12.1	12750	631	10.2	9400	1463	10.3	9000
313	9.8	6700	642	9.1	6400	1593	11.2	10500
353	11.9	13450	647	9.4	7050	1767	11.5	10400
378	11.0	10400	649	9.0	7000	1803	11.4	13300
394	11.1	9050	676	9.2	6850	2068	11.3	13400
412	10.4	7850	721	12.3	15900	2161	10.2	10100
511	10.5	8300	749	11.3	11800	2487	9.5	6900

contd . . .

Appendix Ae2 contd...

Temp.(°C) 38.0			37.0			36.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
997	12.6	19350	2552	11.0	6500	3400	11.4	12800
1738	12.6	16150	2865	10.0	5800	4208	11.5	12500
1762	10.9	9500	3395	11.8	9500	5360	12.5	20600
1970	11.5	11200	4600	10.5	8500	7773	11.5	13100
2151	11.8	13000	4712	11.2	6900	8233	11.6	12900
2161	10.4	8750	4780	11.5	9500	8905	10.9	11300
2217	8.4	6400	4874	10.7	8500	9187	11.2	12500
2371	11.0	11300	4982	11.8	9200	No death		
2435	10.6	10600	5158	11.5	9100			
3124	7.5	4100	5445	10.9	6200			

contd...

Appendix A.2 contd....

Temp.(°C) 36.0			35.5		
Time to death	Total length	Weight	Time to death	Total length	Weight
3417	9.8	6850	None died		
3867	10.1	7900			
5520	10.2	7580			
No death					

Appendix Ae3

Time to death (min), total length (cm) and weight of individual freshwater mullet, *Rhinomugil corsula* acclimated to 25°C and tested at different high lethal temperatures in fresh water

Temp(°C) 40.0			39.5			39.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
19	6.7	1900	49	6.7	1850	55	10.4	8550
28	6.3	1450	65	6.4	1400	105	11.2	12050
34	7.5	2825	70	7.8	2790	140	10.2	9900
43	7.2	2370	74	7.2	2300	150	10.0	8700
47	7.6	3115	84	7.8	3415	199	12.8	16900
50	8.7	4530	110	8.5	4000	201	12.8	16900
59	8.0	3520	124	8.6	3500	216	10.0	8450
74	7.5	3000	144	7.2	3140	255	12.0	13700
78	6.4	1875	181	6.5	1985	302	13.8	22400
93	7.4	2875	191	7.4	2860	335	11.5	13800

contd:..

Appendix Ae3 contd...

Temp(°C) 38.5			38.0			37.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
489	8.6	4600	1129	6.9	1800	1835	10.8	9300
558	12.6	16650	1205	7.1	2100	2262	10.9	8800
654	11.4	12500	1277	7.1	2000	2509	10.5	8400
719	11.6	13700	1343	7.0	1800	2684	12.2	13000
762	9.9	7500	1476	6.9	2700	3020	9.8	6700
862	10.3	8700	1476	7.8	3600	3183	12.0	13500
925	12.5	16150	1729	7.4	2500	3353	11.0	10500
1159	10.5	8950	1970	7.2	2100	3494	11.1	9100
1231	10.4	8600	2467	7.9	2900	3543	10.5	7700
1354	11.3	11800	2858	7.8	3500	3696	10.5	8400

contd...

Appendix Ae3 contd...

Temp (°C) 36.5			36.0			35.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
4695	7.3	2450	6591	11.5	12750	None died		
4972	6.9	2100	6717	12.5	20600			
5204	7.2	2300	6973	11.5	13100			
5956	6.9	1950	7669	11.6	12300			
6428	6.4	1700	7966	10.9	11200			
6775	7.3	2100	9146	11.1	12500			
7282	7.8	3250	10726	11.4	12500			
7643	7.6	2780	No death					
8934	7.5	3950						
9521	7.5	3500						

Appendix Ae4

Time to death (min), total length (cm) and weight (mg) of individual freshwater mullet, *Rhinomugil corsula* acclimated to 20°C and tested at different high lethal temperatures in fresh water

Temp(°C) 38.0			37.0			36.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
1	12.2	10000	133	10.1	9825	539	10.0	7400
2	11.6	8000	271	11.3	13075	1332	8.5	3400
4	11.2	7700	323	11.5	14150	1789	11.0	8500
5	9.5	3700	344	10.7	9850	2154	11.2	8100
7	11.5	9000	357	10.8	9175	2701	11.6	7800
122	10.2	5300	385	11.0	11150	2953	12.8	12600
159	11.6	8500	515	9.8	8950	3206	11.7	9200
162	9.7	5000	577	10.8	12400	3587	11.4	8700
166	8 2	3000	688	9.7	8600			
310	13.0	12000	892	9.7	8350			

contd...

Appendix Ae4 contd . . .

Temp(°C) 35.0			34.0			33.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
2086	10.7	8100	3561	12.0	11800	None died		
2201	11.6	8700	3753	11.5	9600			
2506	10.2	6000	4930	11.0	5500			
2751	10.5	8000	6729	11.7	9000			
3471	10.8	7400	9291	10.5	9500			
3886	10.4	6200	10289	11.2	8400			
3988	10.0	6400	No death					
4197	10.2	6500						

Appendix Ae5

Time to death (min), total length (cm) and weight (mg) of individual freshwater mullet, *Rhinomugil corsula* acclimated to 15°C and tested at different high lethal temperatures in fresh water

Temp(°C)	36.0			35.0			34.0	
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
11	6.7	1900	123	5.8	1100	388	5.7	1315
13	6.3	1455	309	6.1	1350	1607	6.5	1705
17	7.5	2820	345	6.2	1530	1737	6.9	1980
128	7.2	2380	509	8.7	3960	1878	6.2	1580
167	7.6	3125	641	8.3	3630	2791	7.0	2345
223	8.7	4550	678	5.8	1290	3192	8.7	4535
237	8.0	3525	980	7.8	2800	3351	7.9	3405
273	7.5	3010	1264	7.1	2150	3826	9.4	2900
341	6.5	1950	1305	7.0	1915	4168	6.5	1950
442	7.5	2870	1950	8.2	3250	4758	7.5	2810

contd...

Appendix Ae5 contd . . .

Temp(°C) 33.0			32.5			32.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
1457	6.6	1825	2461	7.4	3050	None died		
2022	6.4	1720	2974	8.3	4725			
2146	7.0	1895	5621	7.6	3690			
3793	6.2	1530	5912	8.7	5520			
4165	8.7	3915	6849	8.1	3410			
4406	6.9	2060	7187	8.6	5050			
4657	7.0	2015	7856	7.2	2600			
6296	7.3	2090	8731	6.5	1290			
7107	8.0	3205	9638	7.6	2595			
7404	8.2	3310	10068	7.4	2800			

Appendix Ba1

Time to death (min), total length (cm) and weight (mg) of individual mrigal, *Cirrhinus mrigala* acclimated to 38°C and tested at various low lethal temperatures in fresh water

Temp.(°C) 18.0			18.5			19.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
14	4.9	985	15	4.0	525	1061	4.6	875
16	4.6	810	21	3.8	440	1152	4.8	1020
287	4.6	745	518	4.3	705	1171	4.8	1010
383	4.2	590	686	4.8	890	1549	4.5	910
477	4.7	925	740	4.6	765	1685	4.7	990
594	4.7	1005	877	4.7	810	1842	4.1	630
792	4.4	760	1009	4.7	835	1892	4.2	645
829	4.5	780	1205	4.1	550	2163	4.5	845
1025	4.1	710	1222	4.1	540	2194	4.4	740
1096	4.5	805	1413	4.9	1005	2644	4.2	630

contd...

Appendix Ba1 contd...

Temp.(°C) 19.5			20.0		
Time to death	Total length	Weight	Time to death	Total length	Weight
1515	4.2	575	No death		
1783	4.4	695			
2309	4.0	590			
2858	4.2	605			
3146	4.6	780			
3397	4.3	695			
3624	4.1	515			
No death					

Appendix Ba2

Time to death (min), total length (cm) and weight (mg) of individual mrigal *Cirrhinus mrigala* acclimated to 35°C and tested at various low lethal temperatures in fresh water

Temp.(°C) 17.0			17.5			18.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
9	5.1	1310	335	4.2	620	560	4.8	1040
15	4.6	1015	385	3.7	430	596	4.5	810
19	4.6	920	418	4.3	710	612	4.7	995
251	4.2	870	437	4.3	730	681	4.8	1010
399	4.6	1130	586	4.6	800	1057	4.3	730
418	4.6	1075	743	4.8	805	1138	4.5	830
564	4.4	830	761	4.7	830	1165	4.1	635
642	4.2	810	838	4.1	550	1205	4.2	640
670	4.1	770	889	4.0	505	1251	4.5	845
718	4.6	980	1055	5.2	1200	1337	4.4	760

contd...

Appendix Ba2 contd . . .

Temp.(°C) 18.5			19.0		
Time to death	Total length	Weight	Time to death	Total length	Weight
2472	4.7	925	No death		
2554	4.6	870			
No death					

Appendix Ba3 contd . . .

Temp.(°C) 16.5			17.0		
Time to death	Total length	Weight	Time to death	Total length	Weight
22	3.8	540	No death		
1496	4.2	575			
No death					

Appendix Ba4

Time to death (min), total length (cm) and weight (mg) of individual mrigal *Cirrhinus mrigala* acclimated to 25°C and tested at various low lethal temperatures in fresh water

Temp.(°C) 12.5			13.0			13.5			14.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
12	4.1	540	11	3.8	415	737	4.2	700	2023	4.9	700
14	3.95	505	15	3.7	400	920	3.9	500	2293	4.2	600
15	4.0	545	17	4.3	615	1073	3.8	460	2551	3.7	440
18	4.6	800	21	4.1	540	1098	3.9	510	3617	3.9	540
125	4.2	560	297	3.1	250	1168	4.2	670	4522	3.5	375
151	4.1	555	315	3.8	490	1185	3.8	440	4746	4.0	578
187	4.0	540	442	4.0	500	1355	3.9	500	No death		
215	3.7	470	583	3.9	550	1480	4.0	485			
263	4.0	530	713	4.5	745	1539	4.7	805			
292	4.3	630	989	4.5	810	1892	4.1	500			

Temp.(°C) 14.5		
Time to death	Total length	Weight
No death		

Appendix Ba5

Time to death (min), total length (cm) and weight (mg) of individual mrigal, *Cirrhinus mrigala* acclimated to 20°C and tested at different low lethal temperatures in fresh water

Temp.(°C) 9.5			10.0			10.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
7	3.7	410	10	3.5	365	13	4.2	595
8	3.9	450	12	3.9	425	804	4.8	980
9	4.2	600	17	4.2	570	1129	4.1	560
10	4.2	580	637	4.3	605	1546	4.2	650
10	4.2	600	932	4.2	545	1628	4.0	550
11	4.3	665	953	4.6	795	1731	4.0	540
13	4.2	550	1162	4.4	705	1961	4.35	760
14	4.8	895	1406	4.8	890	2032	3.9	530
864	4.4	790	1902	4.3	650	2682	4.1	560
1093	4.3	650	2099	4.1	620	2771	3.8	510

contd...

Appendix Ba5 contd...

Temp.(°C) 11.0			11.5		
Time to death	Total length	Weight	Time to death	Total length	Weight
1008	4.0	502	No death		
2165	4.1	515			
3109	4.7	805			
3481	4.1	560			
3700	4.8	900			
4179	4.2	580			
4692	4.0	540			
5363	4.3	725			
No death					

Appendix Ba6

Time to death (min), total length (cm) and weight (mg) of individual mrigal, *Cirrhinus mrigala* acclimated to 15°C and tested at various low lethal temperatures in fresh water

Temp.(°C) 8.0

Time to death	Total length	Weight
5	4.0	575
8	3.9	470
12	4.7	895
15	4.5	725
16	4.6	790
97	4.6	760
113	4.1	485
136	4.5	780
140	4.2	605
153	4.3	680

8.5

Time to death	Total length	Weight
7	4.1	680
9	3.8	480
15	4.9	1020
17	4.5	800
122	4.8	920
145	4.6	820
167	4.0	480
171	4.5	825
173	4.2	625
189	4.4	795

9.0

Time to death	Total length	Weight
8	4.4	800
15	4.1	720
16	4.0	610
335	4.5	830
354	4.4	820
390	3.8	565
549	3.9	580
743	4.4	750
No death		

Temp.(°C) 9.5

Time to death	Total length	Weight
No death		

Appendix Bb1

Time to death (min), total length (cm) and weight of individual rohu *Labeo rohita* acclimated to 35°C and tested to various low lethal temperatures in fresh water

Temp.(°C) 17.0			18.0			19.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
13	2.3	130	148	4.0	525	491	3.9	475
145	2.3	145	317	3.7	490	578	3.7	420
180	2.7	205	339	2.5	130	746	3.0	295
225	3.0	285	373	3.3	325	997	3.2	325
264	3.8	570	408	4.2	670	903	4.2	645
316	3.3	390	435	3.8	620	1110	3.7	595
355	3.2	335	487	4.5	900	1438	4.3	745
387	3.7	510	523	4.4	1050	1843	4.2	890
438	3.5	420	592	4.2	850	2060	4.5	850
446	3.8	620	669	4.5	1150	2303	3.9	650

contd...

Appendix Bb1 contd...

Temp.(°C) 19.5			20.0		
Time to death	Total length	Weight	Time to death	Total length	Weight
642	3.9	695	None died		
850	5.2	1515			
1093	4.0	670			
1636	4.4	930			
No death					

Appendix Bb2

Time to death (min), total length (cm) and weight (mg) of individual rohu, *Labeo rohita* acclimated to 30°C and tested to various low lethal temperatures in fresh water

Temp.(°C) 15.5

Time to death	Total length	Weight
7	3.5	450
228	3.6	490
375	3.9	620
479	3.9	655
612	4.1	670
837	3.9	600
982	4.2	800
1120	3.8	630
1202	3.0	285
1271	3.3	395

16.0

Time to death	Total length	Weight
564	2,6	1205
641	2.9	180
704	3.0	260
913	3.3	370
1099	3.4	380
1170	2.7	180
1301	2.7	180
1450	2.9	230
1540	3.0	290
1785	3.3	395

16.5

Time to death	Total length	Weight
1332	3.5	350
1596	3.2	305
2399	2.8	245
3094	2.9	270
3554	3.6	465
4512	3.7	495
No death		

Temp.(°C) 17.0

Time to death	Total length	Weight
1460	2.7	180
No death		

Appendix Bb3

Time to death (min), total length (cm) and weight (mg) of individual rohu, *Labeo rohita* acclimated to 25°C and tested at various low lethal temperatures in fresh water

Temp.(°C) 12.5			13.0			13.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
21	3.6	395	189	3.1	260	1073	2.6	210
294	3.2	360	300	3.4	370	1167	3.0	290
328	3.3	380	469	2.6	200	1242	3.2	340
372	3.1	360	556	3.0	290	1507	4.1	710
451	3.4	455	640	3.1	300	1615	4.3	860
535	3.3	410	737	3.2	360	1692	4.0	650
578	4.1	735	814	3.3	370	1770	4.5	910
685	3.6	505	970	3.4	395	1945	4.2	870
832	3.4	480	1203	3.5	410	2158	4.4	915
			1396	3.9	550	2593	5.0	1420

contd...

Appendix Bb3 contd...

Temp.(°C) 14.0			14.5		
Time to death	Total length	Weight	Time to death	Total length	Weight
1483	3.0	295	1689	3.4	470
1837	4.1	675	No death		
1991	4.0	650			
2037	4.2	720			
3572	4.3	860			
No death					

Appendix Bb4

Time to death (min), total length (cm) and weight (mg) of individual rohu, *Labeo rohita* acclimated to 20°C and tested at various low lethal temperatures in fresh water

Temp.(°C) 8.5			9.5			10.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
6	3.6	510	11	3.7	520	55	3.95	750
8	4.1	675	14	3.8	585	180	4.0	750
9	4.8	1010	15	4.5	680	209	4.3	1080
10	4.2	810	21	3.8	590	242	4.1	810
11	4.4	825	90	4.2	790	424	4.5	1190
72	4.8	1145	108	3.5	510	500	4.5	1140
87	4.5	1005	132	3.5	545	552	4.3	1105
104	4.8	880	178	3.7	590	643	4.6	1235
110	4.7	995	222	3.9	655	894	4.2	930
			250	3.6	570	1023	4.5	1265

contd...

Appendix Bb4 contd . . .

Temp. (°C) 11.5			12.0		
Time to death	Total length	Weight	Time to death	Total length	Weight
74	4.4	900	No death		
1281	3.75	605			
1480	3.8	620			
1565	3.3	375			
2079	3.7	610			
2533	4.3	895			
No death					

Appendix Bb5

Time to death (min), total length (cm) and weight (mg) of individual rohu, *Labeo rohita* acclimated to 15°C and tested at various low lethal temperatures in fresh water

Temp.(°C) 8.0

Time to death	Total length	Weight
11	2.9	235
19	2.7	190
21	3.0	280
22	3.0	260
31	3.5	430
33	3.5	460
185	3.0	275
230	2.5	195
363	3.1	295
521	2.8	235

8.5

Time to death	Total length	Weight
29	5.0	1120
44	4.8	925
85	4.9	1100
134	4.3	730
176	5.6	1780
1991	5.8	1740
2539	6.8	2690
3014	5.5	1690
4012	5.7	1785

9.0

Time to death	Total length	Weight
30	4.6	920
80	4.2	720
3370	4.8	925
4460	5.0	1120
5125	4.3	730
5590	5.8	1740
6214	6.8	2645
No death		

Temp.(°C) 9.5

Time to death	Total length	Weight
2963	2.9	285
No death		

Appendix Bc1

Time to death (min), total length (cm) and weight (mg) of individual fringe lipped carp *Labeo fimbriatus* acclimated to 38°C and tested at various low lethal temperatures in fresh water

Temp.(°C) 17.5			18.5			19.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
7	2.7	160	85	4.3	930	422	4.0	820
9	3.0	295	120	4.5	1100	443	5.1	1800
13	3.75	570	206	4.3	1050	490	4.5	1210
15	3.3	405	235	4.3	915	547	4.0	850
165	4.2	870	293	4.2	800	592	3.1	400
199	3.8	710	311	4.7	1270	680	3.7	595
255	3.6	510	348	4.5	1130	777	3.7	640
272	4.3	930	375	4.5	1070	807	4.0	870
307	6.3	1830	430	4.3	990	974	4.7	1330
414	4.4	990	488	4.4	1030	1007	3.9	760

contd . . .

Appendix Bc1 contd...

Temp.(°C)	21.0		21.5		
Time to death	Total length	Weight	Time to death	Total length	Weight
847	5.0	1470	No death		
885	5.0	1440			
1057	4.3	1030			
1232	4.1	880			
4429	4.2	805			
No death					

Appendix Bc2

Time to death (min), total length (cm) and weight (mg) of individual fringe-lipped carp, *Labeo fimbriatus* acclimated to 35°C and tested at various low lethal temperatures in fresh water

Temp. (°C) 17.0			17.5			18.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
12	2.7	245	16	2.8	240	19	3.6	440
15	3.1	310	19	3.0	295	407	4.4	960
23	3.3	385	360	3.2	325	598	4.5	1020
25	2.9	305	430	2.9	290	704	4.0	800
404	3.1	325	474	3.1	320	755	4.0	780
441	3.0	295	590	3.0	310	940	4.7	1420
533	2.8	295	825	2.8	295	1037	4.3	1030
602	3.2	345	923	3.1	325	1135	4.5	1100
740	3.3	380	960	3.3	395	1250	4.3	940
814	3.4	470	1083	3.5	490	1327	5.0	1500

contd . . .

Appendix Bc2 contd . . .

Temp.(°C) 18.5			19.0		
Time to death	Total length	Weight	Time to death	Total length	Weight
1101	4.1	625	5821	4.2	925
1408	3.8	595	No death		
1619	4.1	680			
2748	3.2	345			
2965	4.0	790			
3554	4.2	815			
4242	3.5	440			
4309	3.9	645			
No death					

Appendix Bc3

Time to death (min), total length (cm) and weight (mg) of individual fringe-lipped carp, *Labeo fimbriatus* acclimated to 30°C and tested at various low lethal temperatures in fresh water

Temp.(°C) 15.5			16.0			16.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
11	2.9	290	12	2.5	180	11	2.0	98
12	2.3	120	15	3.0	325	18	2.8	235
12	2.4	140	20	2.7	235	22	2.8	240
15	3.1	320	1117	2.6	220	28	2.7	210
16	2.7	215	1374	3.5	580	1878	2.8	255
18	2.9	310	1462	3.3	425	2397	2.7	200
19	2.8	240	1624	3.0	330	3151	2.9	265
191	3.8	745	1723	2.9	295	3458	2.8	250
254	3.4	530	1770	2.8	275	3792	2.9	285
323	2.5	190	2076	2.7	200	4438	3.3	425

contd . . .

Appendix Bc3 contd . . .

Temp.(°C) 17.0			17.5		
Time to death	Total length	Weight	Time to death	Total length	Weight
3013	2.9	285	No death		
3823	2.5	180			
4005	3.0	355			
4325	3.1	340			
4947	3.0	325			
5502	2.8	255			
5954	3.1	350			
No death					

Appendix Bc4

Time to death (min), total length (cm) and weight (mg) of individual fringe-lipped carp *Labeo fimbriatus* acclimated to 25°C and tested at various low lethal temperatures in fresh water

Temp.(°C) 13.0			13.5			14.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
3	3.0	225	12	2.9	220	16	2.3	160
6	2.9	235	16	2.7	180	18	2.5	170
8	2.4	120	22	2.5	145	24	2.3	110
9	2.7	190	409	3.1	330	557	3.2	340
11	2.3	120	438	3.1	370	644	3.0	300
14	2.6	180	450	2.9	295	863	2.8	225
17	2.6	195	459	3.5	455	977	2.7	210
24	2.6	205	501	2.6	220	2202	3.0	290
31	2.7	215	524	3.0	300	2334	2.6	185
54	3.1	315	719	3.5	460	2827	3.2	340

contd...

Appendix Bc4 contd...

Temp.(°C) 14.5			15.0		
Time to death	Total length	Weight	Time to death	Total length	Weight
26	2.4	170	1923	2.9	285
37	2.6	190	No death		
3057	2.9	290			
3562	3.1	310			
4049	2.5	380			
4488	3.3	385			
5041	2.8	180			
5538	2.7	180			
5993	3.0	280			
6033	3.1	300			

Appendix Bc5

Time to death (min), total length (cm) and weight (mg) of individual fringe-lipped carp, *Labeo fimbriatus* acclimated to 20°C and tested at various low lethal temperatures in fresh water

Temp. (°C)	11.0		11.5			12.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
6	2.5	165	11	2.6	180	15	2.4	125
7	2.6	175	13	2.7	205	18	2.5	135
9	3.0	270	14	2.8	205	768	2.7	220
11	2.8	270	17	3.2	295	989	2.4	145
14	3.1	230	604	2.7	220	1084	2.6	200
18	3.5	385	685	2.7	235	1692	2.7	235
732	3.0	380	931	2.4	145	1880	2.5	165
1098	3.0	380	1074	3.1	360	2079	3.1	325
1365	3.3	330	1248	3.2	400	2134	3.0	320
			1720	2.6	200	2597	2.6	215

contd...

Appendix Bc5 contd...

Temp.(°C) 12.5			13.0		
Time to death	Total length	Weight	Time to death	Total length	Weight
902	2.2	110	No death		
1363	3.1	390			
2295	2.7	210			
2555	2.9	280			
3229	2.5	180			
3808	2.7	230			
4120	2.4	130			
5088	2.8	180			
5333	2.5	260			
6312	3.1	315			

Appendix Bc6

Time to death (min), total length (cm) and weight (mg) of individual fringe-lipped carp, Labeo fimbriatus acclimated to 15°C and tested at various low lethal temperatures in fresh water

Temp.(°C) 9.5

Time to death	Total length	Weight
9	3.9	690
11	3.1	365
15	3.2	380
359	3.3	395
400	3.2	365
413	3.1	325
491	3.6	630
543	3.6	620
619	3.2	400
710	3.4	535

10.0

Time to death	Total length	Weight
359	2.4	150
422	2.9	280
516	2.3	170
831	3.7	575
887	2.9	280
1080	3.0	295
1316	3.7	575
1351	3.4	480
1571	3.2	345
2357	3.1	310

10.5

Time to death	Total length	Weight
1109	2.3	135
1638	3.1	385
2392	2.9	285
2697	2.7	265
3184	2.4	170
4567	2.5	180
4743	2.7	240
5780	2.8	290
No death		

Temp.(°C) 11.0

Time to death	Total death	Weight
No death		

Appendix Bd1

Time to death (min), total length (cm) and weight (mg) of individual common carp *Cyprinus carpio* (Bankok strain) acclima ted to 35°C and tested at various low lethal temperatures in fresh water

Temp.(°C 12.5			13.0			13.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
4	2.4	230	159	2.5	185	186	2.4	195
23	2.5	280	238	2.8	280	312	2.7	275
472	2.6	280	636	2.4	230	667	2.5	245
517	2.6	275	674	2.5	240	763	2.8	295
544	2.8	360	709	2.7	340	807	3.0	390
564	3.1	580	764	2.4	250	910	3.2	515
623	2.8	350	814	2.5	290	1134	2.7	275
641	3.0	500	842	3.1	440	1200	2.8	320
684	3.2	510	860	2.7	285	1295	2.7	265
816	3.1	460	912	3.7	950	1666	3.1	430

contd...

Appendix Bd1 contd...

Temp.(°C) 14.0

Time to death	Total length	Weight
248	2.3	255
415	2.6	230
819	2.6	260
860	2.7	265
1007	2.7	310
1115	2.6	250
1202	3.0	455
1294	2.9	350
1505	3.1	480
1560	3.0	400

15.0

Time to death	Total length	Weight
386	2.4	210
521	2.5	275
1011	2.6	265
1194	2.6	250
1308	2.7	345
1339	3.0	460
1490	3.0	450
1651	3.1	430
1780	3.1	480
1877	2.9	385

16.0

Time to death	Total length	Weight
515	2.9	380
594	2.8	220
1298	2.7	260
1656	3.1	250
2005	3.1	380
2667	2.8	255
2787	2.4	150
No death		

17.0

Time to death	Total length	Weight
1901	3.4	395
2443	3.2	320
No death		

Appendix Bd2

Time to death (min), total length (cm) and weight (mg) of individual common carp, *Cyprinus carpio* (Bankok strain) acclimated to 30°C and tested at various low lethal temperatures in fresh water

Temp.(°C) 11.5			12.0			12.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
8	2.3	112	14	2.5	180	787	2.5	175
16	2.5	165	625	2.4	175	1778	2.2	80
22	2.4	145	1182	2.0	80	2437	2.5	170
923	2.3	115	1317	2.3	160	2952	2.1	85
1022	2.4	172	1445	2.4	155	5302	2.3	135
1327	2.6	190	1823	2.9	270	8582	2.6	200
1645	2.4	165	2038	2.6	200	No death		
1762	2.6	185	4862	2.1	105			
2833	2.4	185	6209	2.5	185			
4171	2.3	145	7379	2.7	245			

contd...

Appendix Bd2 contd . . .

Temp.(°C) 13.0			13.5		
Time to death	Total length	Weight	Time to death	Total length	Weight
1455	2.0	85	No death		
1926	2.4	130			
4135	2.6	130			
No death					

Appendix Bd3

Time to death (min), total length (cm) and weight (mg) of individual common carp, *Cyprinus carpio* (Bankok strain) acclimated to 25°C and tested at various low lethal temperatures in fresh water

Temp.(°C) 8.5			9.0			9.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
12	2.7	245	490	2.7	256	711	2.7	290
16	2.4	190	580	2.4	195	806	2.7	290
18	2.6	365	748	2.7	355	1035	2.9	400
513	2.6	275	983	2.6	265	1840	2.8	360
587	2.7	315	1118	2.6	320	2050	3.2	560
625	2.8	350	1260	2.8	350	2455	3.2	450
647	3.1	490	1408	2.8	405	3732	2.7	290
849	2.6	235	1458	2.8	315	4583	2.9	385
1144	2.8	340	1681	3.1	450			
1193	2.9	385	3017	3.2	510			

contd...

Appendix Bd3 contd . . .

Temp.(°C) 10.0		
Time to death	Total length	Weight
1167	2.7	290
1366	2.7	305
1760	2.9	305
2728	3.1	490
2867	2.6	235
3709	2.8	340
No death		

10.5		
Time to death	Total length	Weight
1211	3.2	470
No death		

Appendix Bd4

Time to death (min), total length (cm) and weight (mg) of individual common carp, *Cyprinus carpio* (Bankok strain) acclimated to 20°C and tested at different low lethal temperatures in fresh water

Temp.(°C) 6.0			6.5			7.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
13	2.4	105	54	2.6	250	184	3.1	340
19	2.9	185	242	2.4	195	341	2.2	95
23	3.1	315	1078	2.7	290	3046	2.9	265
347	2.9	270	1244	2.5	265	4363	2.5	225
378	2.5	240	1527	2.6	320	5489	2.9	280
449	2.5	236	1797	2.8	325	No death		
459	2.7	290	2000	2.8	350			
493	3.1	425	2998	2.7	330			
536	2.7	325	3150	3.1	450			
561	2.9	340	3490	2.9	345			

Temp.(°C) 7.5		
Time to death	Total length	Weight
No death		

Appendix Be1

Time to death (min), total length (cm) and weight (mg) of individual freshwater mullet *Rhinomugil corsula* acclimated to 35°C and tested at various low lethal temperatures in fresh water

Temp.(°C) 14.5			16.5			17.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
254	7.0	1900	536	6.8	2800	598	8.5	3700
353	8.1	4300	698	7.9	3700	794	8.3	4200
403	7.0	2000	761	7.6	2700	857	7.1	2900
448	7.4	2800	794	7.2	2700	884	7.7	3900
528	7.5	3200	837	6.5	2200	928	8.7	5100
577	8.1	4500	888	7.7	3400	982	8.2	4100
621	6.9	2000	910	7.5	3200	1054	8.8	5000
672	7.2	2300	936	6.9	2800	1146	8.5	4100
728	7.6	2900	1091	7.1	2700	1221	8.3	4000
742	7.8	3200	1107	7.6	3100	1242	8.5	4300

contd...

Appendix Be1 contd...

Temp.(°C) 18.5

Time to death	Total length	Weight
706	8.7	4350
998	8.2	3575
1282	8.7	5050
1431	8.6	4425
1542	8.2	4120
1668	8.6	4575
1780	9.1	5650
2090	8.5	5000
2315	8.7	4810
2451	8.4	4320

19.0

Time to death	Total length	Weight
879	6.9	1850
1326	8.1	4375
1550	7.0	2050
2143	7.3	2625
2395	7.5	3210
2573	8.0	4300
2859	7.2	2300
3361	7.6	2950
3566	7.4	2800
3719	7.8	3250

19.5

Time to death	Total length	Weight
2389	8.4	7000
4528	8.0	4300
4773	8.7	5800
5286	7.6	3100
5926	7.3	2900
6250	8.1	4515
6484	7.9	3450

No death

21.0

Time to death	Total length	Weight

None died

Appendix Be2

Time to death (min) total length (cm) and weight (mg) of individual freshwater mullet *Rhinomugil corsula* acclimated to 30°C and tested at various low lethal temperatures in fresh water

Temp.(°C) 12.5			13.5			14.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
12	7.9	3600	473	6.4	1900	619	9.9	8400
18	8.5	4000	558	7.9	3500	749	9.3	6700
426	10.1	8800	638	10.1	8100	2564	8.8	5800
516	10.5	7500	703	8.0	3600	2840	10.2	8700
616	10.6	8100	795	8.7	5400	3157	10.6	10000
671	6.2	3500	824	8.3	5000	3439	10.6	10600
681	7.8	4000	889	8.5	4900	3767	5.5	6000
751	8.1	5500	1241	7.9	4500	3857	10.3	8600
826	8.1	5300	1662	8.5	5000	4193	10.5	9900
877	8.2	4500	2635	- *	- *	4250	9.8	6900

contd . . .

* Length and weight not recorded

Appendix Be2 contd . . .

Temp(°C) 15.5			16.0			16.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
1785	9.7	8000	2169	6.9	2530	No death		
2889	8.5	6500	3675	8.0	3720			
3591	9.0	6500	4206	7.5	2710			
4100	9.2	6200	5112	7.3	2700			
4713	8.7	6000	6512	6.5	2275			
6016	10.6	10600	7488	7.8	3490			
7086	9.5	6100	No death					
7424	8.1	5500						
7878	10.5	9900						
No death								

Appendix Be3

Time to death (min), total length (cm) and weight (mg) of individual freshwater mullet *Rhinomugil corsula* acclimated to 25°C and tested at various low lethal temperatures in fresh water

Temp.(°C) 10.5			11.5			12.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
185	7.5	3700	442	7.9	3300	1369	11.1	13200
361	8.5	5800	551	8.1	3700	1871	8.8	6100
435	7.8	4060	687	8.0	4200	2298	11.0	11100
601	8.7	5800	737	8.4	4200	2368	12.3	14650
702	8.2	3500	825	5.5	4500	2687	9.3	6925
759	8.3	3500	907	8.2	4200	2696	12.2	15650
865	8.6	5000	1009	8.3	4300	3123	9.4	7350
908	8.4	4900	1106	8.3	3900	3552	12.2	17350
963	8.5	5000	1140	7.9	3300			
1035	8.5	5100	1203	10.4	10200			

contd...

Appendix Be3 contd . . .

Temp.(°C) 13.0			13.5		
Time to death	Total length	Weight	Time to death	Total length	Weight
2941	11.0	11900	None died		
3161	9.0	6600			
3276	12.3	15500			
3467	10.7	10030			
3556	11.2	10800			
3655	10.5	10020			
4046	12.8	18700			
4892	12.9	17025			
6039	11.5	11950			
6652	12.0	14050			

Appendix Be4

Time to death (min), total lengh (cm) and weight (mg) of individual freshwater mullet *Rhinomugil corsula* acclimated to 20°C and tested at various low lethal temperatures in fresh water

Temp.(°C) 9.5			10.0			10.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
9	7.0	1915	10	10.2	7600	600	7.5	3900
12	8.1	4010	669	10.8	10100	940	9.3	5800
542	6.9	1895	1154	11.1	10100	1334	10.6	10100
632	7.4	2800	1245	11.7	14200	1475	10.3	9300
667	7.5	3185	1354	10.8	10100	1490	8.7	4800
820	8.0	3990	1439	10.6	10400	1695	8.1	5500
874	6.9	2010	1519	9.9	8500	1915	10.8	9500
1007	7.2	2295	1549	9.7	8400	2520	12.8	16500
1110	7.5	2755	1647	8.7	8100	2529	12.7	16000
1456	7.8	3200	2352	9.9	8600	2995	12.5	15900

contd...

Appendix Be4 contd...

Temp.(°C) 11.0			11.5			12.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
1468	7.4	3050	4549	9.8	8500	None died		
1691	8.4	4900	4991	12.0	14400			
2958	7.7	3850	5032	9.5	7250			
3116	8.7	5520	5090	9.7	6750			
3192	8.2	3570	6028	11.35	12100			
3645	8.1	3400	6484	12.1	14600			
4145	8.6	5050	6687	10.4	8700			
4195	8.4	4850	7140	10.4	10100			
4227	8.5	5000	No death					
4534	8.5	5150						

Appendix Be5

Time to death (min), total length (cm) and weight (mg) of individual freshwater mullet *Rhinomugil corsula* acclimated to 15°C and tested at various low lethal temperatures in fresh water

Temp.(°C) 8.5			9.0			9.5		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
9	7.4	3050	15	7.3	2890	1480	6.9	1805
12	8.3	4750	1030	8.2	4510	1551	8.1	4005
618	7.6	3720	1455	7.5	3520	1635	6.8	1800
702	8.6	5210	1705	8.7	5515	1920	7.3	2650
784	8.1	3295	1910	8.0	3090	2379		
852	8.0	3290	1992	7.9	3010	2447	7.8	3750
1175	8.4	4775	2025	8.2	4325	3210	8.2	4325
1316	8.3	4590	2522	8.3	4310	3694	8.3	4495
1585	8.5	5000	2561	7.9	3540	4379	7.9	3510
1683	8.4	4920	2703	6.8	1895	4477	8.4	4920

contd...

Appendix Be5 contd...

Temp.(°C) 10.0			10.5			11.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
1915	5.7	1310	3255	7.0	1910	None died		
2147	6.5	1895	3850	8.2	4210			
2850	6.8	1830	4504	6.9	1895			
3515	6.2	1595	5240	- *	- *			
3802	7.4	2985	5311	7.4	2810			
4189	6.8	2010	5797	7.9	3990			
4345	7.0	1995	6720	6.9	2010			
4725	7.4	2995	No death					
5074	8.1	3580						
5286	8.2	3225						

* Length and weight were not recorded

Appendix Ca1

Time to death (min), total length (cm) and weight (mg) of individual mrigal, *Cirrhinus mrigala* acclimated to 30°C in fresh water and tested to different lethal salinities at 30°C

Salinity (‰) 20.0			15.0			10.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
9	1.2	9	22	1.2	17	213	1.2	18
10	1.2	10	26	1.3	19	220	1.2	20
11	1.3	11	28	1.2	16	229	1.1	15
11	1.4	16	30	1.3	18	235	1.2	16
11	1.2	10	37	1.2	15	248	1.1	15
16	1.4	19	41	1.3	21	313	1.1	16
18	1.2	10	43	1.2	14	324	1.1	19
19	1.4	21	44	1.4	21	474	1.2	23
20	1.3	17	50	1.3	18	478	1.2	21
21	1.3	17	50	1.4	26			

contd . . .

Appendix Ca1 contd...

Salinity (‰) 5.0			2.5		
Time to death	Total length	Weight	Time to death	Total length	Weight
1469	1.3	10	None died		
2416	1.4	15			
2524	1.3	12			
3122	1.3	15			
3388	1.3	17			
4471	1.2	10			
5839	1.2	10			
5865	1.2	13			
8078	1.3	15			
8949	1.4	18			

Appendix Cc1

Time to death (min), total length (cm) and weight (mg) of individual fringe-lipped carp, *Labeo fimbriatus* acclimated to 30°C in fresh water and tested to different lethal salinities at 30°C

Salinity (‰) 20.0			15.0			10.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
15	1.5	25	39	1.3	18	477	1.3	17
16	1.7	40	51	1.8	37	613	1.7	28
17	1.6	33	53	1.5	23	644	1.9	41
19	1.8	43	54	1.5	21	736	1.8	32
19	1.4	20	57	1.8	38	1007	1.6	30
21	1.5	30	61	1.6	29	1205	1.6	31
21	1.7	40	62	1.5	22	1502	1.5	33
21	1.6	34	64	1.6	33	1527	1.7	36
21	1.7	43	64	1.7	39	1566	1.5	26
22	1.8	50	64	1.5	24	1582	1.5	28

5.0		
Time to death	Total length	Weight
None died		

Appendix Ce2

Time to death (min), total length (cm) and weight (mg) of individual fringe-lipped carp, Labeo fimbriatus acclimated to 30°C in salt water (5‰ (s) and tested to different lethal salinities at 30°C

Salinity (‰) 30.0			25.0			20.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
7	1.8	30	14	1.8	30	28	1.8	45
8	1.9	40	19	1.7	30	31	1.8	40
10	1.7	37	20	1.9	41	41	1.9	50
12	1.8	38	22	1.8	37	43	2.0	65
13	1.8	40	24	1.8	45	44	1.8	48
14	2.0	65	24	2.0	50	46	1.8	45
14	1.7	30	25	1.9	50	46	2.0	70
16	1.8	44	26	1.7	41	47	1.9	63
			31	1.9	55	48	1.8	43
			32	2.3	95	50	1.9	68

contd...

Appendix Cc2 contd...

15.0			10.0		
Time to death	Total length	Weight	Time to death	Total length	Weight
76	1.5	20	824	2.0	83
79	1.5	21	968	1.9	50
84	1.9	60	1015	1.9	52
89	1.8	40	1049	1.8	45
90	1.8	41	1096	1.8	35
90	2.2	93	1207	2.0	55
91	1.8	30	1460	1.8	45
96	1.8	32	1506	1.8	47
97	1.9	67	1828	1.8	50
97	1.7	45	2073	2.0	67

Appendix Cd1

Time to death (min), total length (cm) and weight (mg) of individual common carp, *Cyprinus carpio* acclimated to 30°C in fresh water and tested at different lethal salinities at 30°C

Salinity (‰) 30.0			25.0			20.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
6	2.4	115	10	2.0	70	10	2.5	141
7	2.2	80	11	2.4	141	15	2.0	87
7	2.1	90	12	2.1	100	16	2.1	100
8	2.1	70	12	2.5	150	17	2.1	83
8	2.0	69	13	2.0	100	18	2.3	130
9	2.1	80	14	2.1	90	19	2.5	185
10	2.2	98	14	1.9	70	20	2.4	137
10	2.4	120	15	1.9	80	21	2.5	140
11	2.0	80	15	2.1	98	22	2.5	187
12	2.0	91	15	2.2	102	22	2.2	99

contd...

Appendix Cd1 contd...

Salinity (‰) 15.0			10.0			5.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
22	2.3	98	236	2.1	68	None died		
28	2.0	70	740	2.5	120			
29	2.2	105	957	2.4	115			
30	2.3	112	1247	2.2	75			
32	2,0	65	1444	2.3	89			
33	2.2	81	1941	2.9	190			
34	2.3	100	2010	2.5	162			
35	2.5	132	5607	2.6	205			
37	2.3	120	6172	2.5	180			
39	2.1	92						

Appendix Ce1

Time to death (min), total length (cm) and weight (mg) of individual freshwater mullet, *Rhinomugil corsula* acclimated to 28°C in freshwater and tested to different lethal salinities at 35°C

Salinity (‰) 40.0			35.0			30.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
11	6.2	1370	16	6.4	2125	26	6.1	1490
12	6.3	1820	17	5.0	1055	27	7.0	2450
13	5.7	1140	18	5.8	1580	28	9.0	5120
14	5.7	1160	18	6.1	1850	29	6.6	1980
15	6.6	2020	19	6.1	1895	30	7.2	2725
15	6.2	1660	20	8.0	4010	32	6.3	1840
16	6.6	2050	20	7.2	3010	33	6.4	1895
17	6.9	2380	21	4.9	960	35	8.1	3410
17	7.2	2800	22	6.8	2635	36	9.5	6270
18	7.2	2440	23	6.7	2865	38	7.7	3540

contd...

Appendix Ce1 contd . . .

Salinity (‰) 26.25

Time to death	Total length	Weight
39	6.3	1930
42	5.0	950
43	4.9	905
44	5.9	1700
46	6.0	1630
48	5.1	1050
49	4.8	905
50	6.1	1800
52	6.1	1750
66	6.8	2570

18.0

Time to death	Total length	Weight
75	5.5	1020
76	5.6	1075
79	5.8	1365
81	6.1	1500
85	8.3	4070
86	7.3	2660
88	6.9	2350
89	7.7	3070
95	8.3	3830
102	7.8	3110

17.5

Time to death	Total length	Weight
111	6.4	2010
122	7.0	2875
133	7.3	3145
147	5.8	1530
153	6.3	2065
160	5.7	1440
166	6.4	1935
168	6.7	2830
180	5.4	1105
196	7.0	2800

15.0

Time to death	Total length	Weight
1225 No death	- *	- *

* Length, weight not recorded

Appendix Ce2

Time to death (min), total length (cm) and weight (mg) of individual freshwater mullet, *Rhinomugil corsula* acclimated to 28°C in fresh water and tested to different lethal salinities at 30°C

Salinity (‰) 40.0			35.0			30.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
24	5.1	885	29	3.9	445	39	6.9	2520
25	5.3	1040	32	5.3	1090	40	6.3	1660
25	5.8	1220	34	5.2	1060	42	6.7	2000
26	5.4	1080	35	6.6	2080	44	6.2	1480
26	4.9	815	39	6.5	1965	46	6.5	1895
27	6.7	1860	40	7.5	3060	47	6.1	1445
28	5.8	1110	41	7.9	2600	49	7.2	2600
29	6.0	1530	42	6.0	1790	50	7.9	3430
30	5.6	1270	43	7.2	2840	54	7.0	2465
31	8.0	3500	45	7.6	3815	55	7.5	2990

contd...

Appendix Ce2 contd...

Salinity (‰) 26.25

Time to death	Total length	Weight
77	5.5	1390
79	6.4	1780
82	5.9	1575
84	7.1	2615
89	7.8	3725
91	7.7	3595
109	6.5	2215
112	8.2	3970
114	7.7	3870
120	7.1	2715

18.0

Time to death	Total length	Weight
95	5.0	620
101	5.1	820
116	5.9	1320
123	5.4	1060
130	5.7	1205
138	7.1	2570
142	5.9	1220
171	7.1	2640
181	6.6	2120
216	6.9	2415

17.5

Time to death	Total length	Weight
208	4.1	- *
No death		

*Weight not recorded

15.0

Time to death	Total length	Weight
None died		

Appendix Ce7

Time to death (min), total length (cm) and weight (mg) of individual freshwater mullet, *Rhinomugil corsula* acclimated to 28°C in fresh water and tested to various lethal salinities at 26°C

Salinity (°/₀₀) 40.0			35.0			30.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
23	5.6	1050	28	6.3	1620	39	7.1	2350
24	7.1	2700	30	7.5	2995	40	6.9	2010
25	5.5	1110	32	8.1	3540	41	7.2	2560
26	5.8	1440	33	5.9	1300	42	6.8	1990
30	6.5	1850	35	6.8	2145	45	7.0	2550
32	6.8	2025	36	7.1	2580	47	6.5	1815
33	7.9	3410	37	-*	-*	49	7.5	2950
34	8.3	4280	39	7.9	3470	53	6.7	2150
36	7.9	3550	40	8.3	3890	54	8.3	4255
37	7.5	2990	41	10.0	7100	59	9.0	5360

contd . . .

* Length and weight not recorded

Appendix Ce3 contd....

Salinity (‰) 25.0			20.0			15.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
66	8.9	4800	259	8.3	3810	No death		
73	8.4	3940	263	7.4	2750			
75	9.2	5330	267	7.0	2335			
82	8.0	3000	271	7.5	3010			
83	7.7	3240	281	7.5	2910			
86	8.3	3700	292	7.5	2990			
87	7.9	3410	295	7.2	2520			
91	7.2	2100	303	8.0	3120			
95	7.7	3140	308	8.4	4200			
97	7.6	3050	311	8.3	3950			

Appendix Ce4

Time to death (min), total length (cm) and weight (mg) of individual freshwater mullet, *Rhinomugil corsula* acclimated to 28°C in fresh water and tested to various lethal salinities at 20°C

Salinity (‰) 40.0			35.0			30.0		
Time to death	Total length	Weight	Time to death	Total length	weight	Time to death	Total length	Weight
37	6.0	1200	45	5.9	1180	71	5.4	1090
39	6.2	1450	50	5.4	1150	72	6.2	1450
40	5.9	1320	53	5.3	930	74	5.1	870
41	6.1	1530	55			77	6.1	1495
42	6.5	1995	56	6.0	1300	79	5.8	1300
44	5.4	1150	58	5.9	1320	83	5.4	1180
47	6.2	1610	60	6.2	1610	89	6.1	1520
50	5.8	1440	61	7.2	2700	91	5.8	1440
51	6.4	2185	63	6.4	1840	92	8.2	4150
53	8.4	3860	66	7.8	3060	94	6.9	2180

contd...

Appendix Ce4 contd...

Salinity (‰) 25.0		
Time to death	Total length	Weight
107	5.4	1085
109	5.3	1095
111	5.9	1350
112	8.5	4060
113	5.8	1310
116	6.5	1870
120	5.6	1220
122	7.2	2700
126	7.3	2790
128	9.3	6090

	20.0	
Time to death	Total length	Weight
158	5.5	1020
162	5.4	1090
177	5.4	1500
192	6.5	1680
208	7.4	2720
232	5.8	1510
245	8.5	4060
254	8.2	4100
265	7.3	2810
270	7.8	3150

	15.0	
Time to death	Total length	Weight
1041	6.4	1870
1189	6.2	1715
1250	6.5	2020
1312	8.4	4045
1355	6.7	2305
1462	7.1	2780
1591	7.4	2800
1689	7.7	3455
1723	6.9	2295
1838	8.2	4005

	12.5	
Time to death	Total length	Weight
None died		

Appendix Ce5

Time to death (min), total length (cm) and weight (mg) of individual freshwater mullet, Rhinomugil corsula acclimated to 28°C in fresh water and tested to various lethal salinities at 17.5°C

Salinity (‰) 45.0			40.0			35.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
36	7.3	2800	50	5.5	995	67	5.6	1310
40	7.2	2680	55	6.6	2020	72	5.9	1500
43	7.0	2405	57	6.4	1850	74	5.2	1020
46	7.3	3020	61	8.9	5150	77	5.5	1310
48	8.1	3620	64	7.2	2705	82	5.1	920
49	7.9	3390	65	7.6	3060	87	5.4	1180
51	8.8	4820	69	6.5	2160	89	4.8	785
53	7.4	3240	71	7.8	3390	92	5.2	1060
55	6.8	2240	75	7.5	3105	98	5.8	1390
56	7.5	3195	79	8.3	4390	102	6.6	2220

contd...

Appendix Ce5 contd...

Salinity (‰) 30.0			25.0			20.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
91	7.0	2455	129	4.3	560	256	5.3	980
96	9.1	5390	135	7.2	2955	267	9.3	2400
100	8.2	4195	140	6.8	2180	280	6.7	1860
107	11.1	9790	151	6.5	2060	291	6.4	1550
109	6.8	2180	153	6.5	1985	306	7.4	2520
112	6.6	2020	159	6.9	2385	323	7.2	2375
118	6.4	1870	170	7.3	2660	327	7.3	2590
121	7.8	3370	174	8.0	4175	338	6.9	2050
125	8.4	4365	179	6.5	2220	343	8.3	4010
128	7.2	2705	208	7.7	3375	361	9.1	5200

contd...

Appendix Ce5 contd...

Salinity (‰) 15.0			12.5		
Time to death	Total length	Weight	Time to death	Total length	Weight
1227	6.2	1690	None died		
1706	6.5	2010			
1752	6.8	2190			
1790	8.5	4580			
1821	7.1	2780			
1867	7.5	2900			
1890	7.9	3600			
1931	6.9	2295			
1974	8.7	4700			
1993	8.2	3995			

Appendix Ce7

Time to death (min), total length (cm) and weight (mg) of individual freshwater mullet *Rhinomugil corsula* acclimated to 35°C in fresh water and tested to different salinities at 39°C

Salinity (‰)	*Fresh water			7.0			15.0	
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
241	10.6	9200	492	13.3	18000	549	9.6	7600
337	10.4	8900	659	13.5	18800	635	8.2	3700
524	10.3	9000	798	12.8	12900	670	11.7	10900
691	11.6	11700	1051	14.7	23900	715	11.8	10200
850	11.5	10400	1100	12.2	12200	770	11.5	11400
885	12.1	13900	1353	11.7	11100	797	7.9	3700
925	11.4	13300	1522	12.2	10000	860	11.2	8900
957	11.4	13400	1682	11.9	9900	893	11.9	9300
998	10.3	10400						
1032	9.6	7000						

*Data taken from Appendix Ae1

Appendix Ce8

Time to death (min), total length (cm) and weight (mg) of individual freshwater mullet Rhinomugil corsula acclimated to 35°C in fresh water and tested to different salinities at 37°C

Salinity (‰) *Fresh water			7.0			15.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
2423	10.9	10500	4312	11.4	8700	2052	9.6	6700
3211	10.0	7200	4429	11.0	7900	2180	10.7	8000
3868	12.1	13600	4575	11.7	9200	2262	11.4	9500
4035	11.7	10380	4890	11.0	8300	2494	10.2	5700
4360	10.9	10060	4978	12.4	12200	2651	11.0	7700
4385	10.8	10040	5032	12.7	12200	2746	12.7	12500
4480	9.9	7600	No death			2823	11.0	8000
4615	10.4	9100				3019	10.4	7500
4648	11.0	10090						
4650	9.05	6040						

*Data reproduced from Appendix Ae1

Appendix Ce9

Time to death (min), total length (cm) and weight (mg) of individual fresh water mullet *Rhinomugil corsula* acclimated to 30°C in fresh water and tested to different salinities at 41°C

Salinity (‰) *Fresh water			7.0			15.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
20	8.6	4600	35	10.9	10010	20	10.6	9650
28	12.7	16650	42	11.6	11350	26	10.5	8750
31	11.4	12550	47	9.5	6600	29	13.1	17100
40	11.6	13700	55	9.9	7650	30	11.1	10450
44	9.9	7500	57	10.2	9050	31	11.5	12400
47	10.3	8700	60	12.4	14100	32	9.9	7400
53	12.5	16150	61	10.2	8700	33	10.6	9000
61	10.5	8950	63	10.2	8100	34	10.3	9300
67	10.4	8600	67	10.4	11400	35	11.2	10700
			68	11.4	12150	36	10.7	9000

*Data reproduced from Appendix Ae2

Appendix Ce11

Time to death (min), total length (cm) and weight (mg) of individual freshwater mullet, Rhinomugil corsula acclimated to 30°C in fresh water and tested to different salinities at 37°C

Salinity (‰) *Fresh water			7.0			15.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
2552	11.0	6500	5450	10.8	6700	1766	10.0	7000
2865	10.0	5800	5623	9.9	5100	1788	10.8	9500
3395	11.8	9500	5716	10.9	7300	1824	10.4	7900
4600	10.5	8500	5779	10.5	6300	1862	10.3	7800
4712	11.2	6900	5875	11.0	7400	1905	10.3	7800
4780	11.5	9500	5967	10.3	6800	1933	10.8	9600
4874	10.7	8500	6095	10.2	6900	1980	10.2	6800
4982	11.8	9200	6192	10.5	7200	2023	10.3	7500
5158	11.5	9100	6320	10.2	6900	2067	11.0	8300
5445	10.9	6200				2130	10.3	7400

*Data reproduced from Appendix Ae2

Appendix Ce12

Time to death (min), total length (cm) and weight (mg) of individual freshwater mullet, *Rhinomugil corsula* acclimated to 25°C in fresh water and tested to different salinities at 39°C

Salinity (‰) 20.0			15.0			12.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
9	6.2	1370	75	5.5	1020	217	7.0	1725
10	6.2	1455	78	5.6	1075	390	6.7	1910
12	5.7	1120	80	5.8	1365	467	8.0	3720
12	6.3	1795	84	6.1	1500	488	8.2	3925
13	6.3	1810	91	8.3	4070	500	7.5	3010
14	5.6	1050	95	7.2	2575	506	8.5	4240
16	6.6	1160	101	6.8	2125	515	9.1	5310
18	6.3	1670	103	7.5	2905	527	8.3	3900
22	6.7	2095	104	8.2	3700	534	7.9	3840
23	7.2	2800	107	7.9	3290	556	7.2	2245

contd . . .

Appendix Ce12 contd . . .

Salinity (‰) 10.0			7.0			5.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
619	6.3	1795	531	11.5	10800	416	10.5	8350
672	5.0	925	582	10.5	6300	455	8.3	3655
707	4.9	905	692	11.6	12000	505	9.6	6210
738	5.8	1700	766	11.0	9000	524	6.5	1870
772	6.1	1785	787	11.5	8800	612	11.0	11200
817	5.4	1295	919	10.2	6000	666	6.4	1950
839	6.1	1800	1174	11.2	8700	725	12.1	11500
916	5.9	1750				774	6.5	2230
943	6.1	1925				809	9.9	7590
964	6.8	2570				856	11.3	11180

contd . . .

Appendix Ce12 contd...

Salinity (‰) 3.0			* Fresh water		
Time to death	Total length	Weight	Time to death	Total length	Weight
463	10.9	10910	55	10.4	8550
497	10.5	8400	105	11.2	12050
565	8.9	5010	140	10.2	9900
675	9.6	6345	150	10.0	8700
725	8.3	3600	199	12.8	16900
742	11.4	12180	201	12.8	16900
777	9.4	7125	216	10.0	8450
823	8.5	4010	255	12.0	13700
851	10.2	8110	302	13.8	22400
867	11.2	10900	335	11.5	13800

*Data reproduced from Appendix Ae3

Appendix Ce13

Time to death (min), total length (cm) and weight (mg) of individual freshwater mullet, *Rhinomugil corsula* acclimated to 20°C in fresh water and tested to different salinities at 37°C

Salinity (‰) 25.0			20.0			18.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
5	5.6	1110	13	6.4	1720	56	6.7	1900
5	6.4	1695	13	7.0	1895	61	6.3	1450
8	6.8	1830	14	6.2	1530	73	7.5	2820
9	6.2	1580	15	8.7	3960	91	7.2	2320
9	7.0	2350	16	8.3	3630	96	7.6	3110
10	6.8	2040	16	6.5	1800	123	8.7	4530
10	7.0	1815	17	5.8	1290	162	8.0	3520
11	7.4	2930	18	7.8	2800	181	7.5	3050
12	8.1	3580	19	7.1	2150	210	6.5	1950
12	8.2	3220	20	7.0	1910	233	7.5	2870

contd...

Appendix Ce13 contd...

Salinity (‰) 18.0			12.0			10.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
22	6.5	1508	1651	6.7	2310	2810	5.6	1700
30	6.9	1781	2021	- *	- *	4659	7.2	1970
35	8.3	3930	2571	6.4	1640	5625	6.2	1080
41	9.0	4600	3602	5.9	1560	6198	5.4	890
65	7.1	2400	4338	6.5	2105	6507	7.1	1750
69	6.4	1420	4648	6.6	2417	7012	7.1	1860
78	6.7	1710	4871	7.1	2670	8225	6.9	1595
91	8.1	3525	5435	8.1	3695	8685	7.0	1670
115	7.0	2325	5900	6.9	2155			
127	6.5	1610	6411	7.2	2780			

contd...

* Length and weight not recorded

Appendix Ce13 contd...

Salinity (‰) 7.0			5.0			3.0			*Fresh water		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
5232	6.0	960	2324	6.6	2770	3076	7.4	2815	133	10.1	9825
6028	6.0	1140	2941	6.4	1575	3790	8.2	4029	271	11.3	13075
6540	5.8	1070	3195	5.7	1250	5021	7.1	2190	323	11.5	14150
6584	6.2	1630	3905	6.7	2455	5906	6.9	1985	344	10.7	9850
7856	11.2	9200	4522	6.5	1920	6423	6.5	1845	357	10.8	9175
8007	8.7	2550	5477	6.4	1710	7133	6.4	1811	385	11.0	11150
			6585	7.1	2075	8135	7.2	2070	515	9.8	8950
			7427	8.2	3725	8894	6.8	1630	572	10.8	12400
			7759	6.9	1980	9438	6.5	1470	688	9.7	8600
			8699	7.2	2120				892	9.7	8350

* Data reproduced from Appendix Ae4

Appendix Cf1

Time to death (min), total length (cm) and weight (mg) of individual cichlid fish *Tilapia mossambica* acclimated to 30°C in fresh water and tested at different lethal salinities at 30°C

Salinity (‰) 35.0			30.0			25.0		
Time to death	Total length	Weight	Time to death	Total length	Weight	Time to death	Total length	Weight
22	0.7	5	40	0.7	6	61	0.8	8
35	0.7	6	48	0.8	7	77	0.8	9
37	0.8	8	50	0.8	8	86	0.9	9
46	0.7	6	51	0.7	6	93	0.8	8
48	0.8	6	54	0.7	6	95	0.8	7
50	0.7	6	56	0.7	5	98	0.8	7
51	0.7	9	57	0.8	8	100	0.8	8
53	0.7	8	61	0.7	7	114	0.9	9
55	0.7	7	64	0.8	7	122	0.8	7
56	0.7	7	69	0.7	6	130	0.9	8

contd...

Appendix Cf1 contd...

Salinity (‰) 20.0

Time to death	Total length	Weight
402	0.7	7
954	0.7	7
3134	0.7	8
3333	0.7	7
3445	0.8	8
3475	0.8	8
3547	0.8	8
3552	0.8	8
5673	0.9	9
5708	0.9	9

15.0

Time to death	Total length	Weight
3515	0.9	9
4227	0.8	8
4487	0.6	7
4889	0.7	8
4980	0.7	7
5107	0.8	9
5727	0.7	7
6139	0.8	9
6349	0.8	8
7177	0.8	8

10.0

Time to death	Total length	Weight
9115	0.8	8
9674	0.9	9
9864	0.7	7
10157	0.8	8
10366	0.8	8
10375	0.8	8
10397	0.7	8
10421	0.9	9
10690	0.8	9
10866	0.8	8

5.0

Time to death	Total length	Weight
None died		

Reprinted from J. Madurai Univ. Vol. VI - I, Page (54 - 55)

A CONSTANT LOW TEMPERATURE RECIRCULATING WATER BATH

H. MOHAMAD KASIM, M. N. KUTTY & J. A. BASIL
School of Biological Sciences, Madurai University, Madurai-625021.

(Received for publication 26th March, '77)

Introduction :

This paper deals with the fabrication of a constant recirculating low temperature water bath with mostly indigenous materials, for long term experiments.

Fabrication materials :

The main components of the present fabrication are the Electronic Relay (Electric Control Equipment Co., Madurai), Water cooler (Blue star, Calcutta), Immersion pump (Little Giant Pump Co., Oklahoma, U. S. A.) or any Indian make immersion pump (Jyoti, Baroda) can be used, contact thermometer (Jumo, U. S. A.) Instead a *Toulene contact thermometer (Fig. 2) can be used, two water containers of 70 lit. capacity to serve as water bath and a sump, and two aeraters with air diffuser stones to mix the water and to maintain the water near air saturation as well.

Electrical connections :

In this set-up the electrical connections of water cooler and pump are connected together to the relay (H1, H2 & E) The contact thermometer which is fixed in the water bath is connected to the relay (T1, T2) and the relay (N, P, E) in turn is connected to the main (Fig. 1). The two aeraters (220/230 V) have been connected to the main separately.

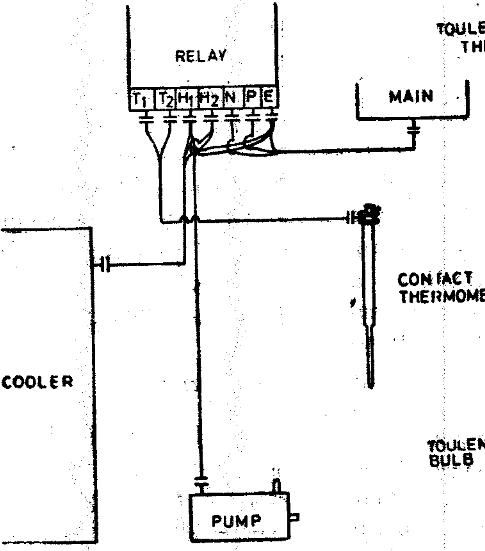


Fig. 1

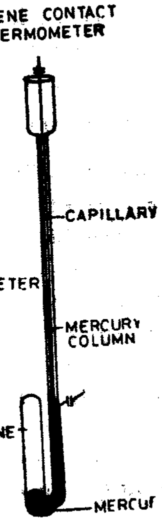


Fig. 2

---

* The toulene thermosensor is made up of glass (Fig. 2). It has a lower vertical bulb filled with toulene and a 'J' shaped capillary filled with mercury. There are two electrical contacts, one at the middle of the mercury column with permanent connection to the mercury and the other through the upper opening of the thermosensor by a wire This second contact serves to close and open the electrical circuit.

Water connections :

The immersion pump is placed in the sump which is situated at the ground level beside the water cooler. The outlet of the pump is connected to the inlet of the water cooler and in turn the outlet of the cooler is connected to the upper low temperature water bath which is placed at a height of 3 feet. The overflow of this water bath is drained into the lower reservoir. Thus a continuous recirculating system is attained by making all connections by polythene tubings. All the four air diffuser stones of the two aeraters are secured on all the four corners of the rectangular water bath (Fig 3)

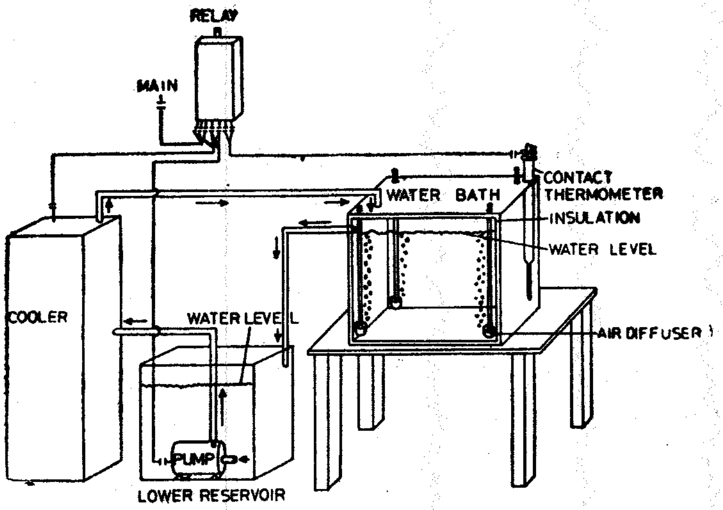


Fig. 3

Operation and Temperature setting :

To start with, the lower reservoir and the upper water bath are filled with water. The relay is switched on and the contact thermometer is adjusted by rotating the knob anticlockwise to fix the temperature at a desired lower level i. e., below room temperature down to 6°C. Relay sets the cooler and pump to work simultaneously and the water is pumped up through the cooler, from the lower reservoir into the water bath and the overflow of this bath drains back into the lower reservoir. By manipulating the screw cock of the cooler, the volume of output of the cooler can be reduced; this in turn will reduce the temperature of the outflowing water from the cooler. This reduction in flow rate will increase the cooling efficiency of the cooler and the temperature of the outflowing water can be brought down to 5°C. The aeraters help in preventing thermal stratification and in maintaining the O_2 content in the water near air saturation. The water in the bath can be 'boost cooled' by adding a few ice cubes so as to reduce the time of initial cooling. Temperature fluctuation in the water bath will be ± 0.05°C or even less when the 'Jumo' contact thermometer is used and ± 0.1°C when 'Toulene' contact thermosensor with a capillary of 1mm diameter is used. Both the water tanks are insulated with thermocole which reduces the thermal conduction through radiation to a great extent. When the required temperature is reached the contact thermometer "cuts off" the cooler and pump simultaneously through the electronic relay. When the temperature in the bath rises due to thermal dissipation the thermal sensor 'puts on" the 'Cooler-pump' unit to work automatically to bring down the temperature. Thus an alternate, automatic 'switch on and off, mechanism is maintained to keep a constant temperature. This set-up can function indefinitely unless it is intervened by inevitable disturbances like current failure and mechanical disorders.

Application :

This bath will be of immense use in numerous assay systems in the field of biological sciences, especially when dealing with aquatic organisms. Universities, post-graduate institutions, research organisations and industries which have insufficient funds for the purchase of foreign makes, can use this unit for various aspects like determination of viscosity. Also this is useful for continuous recirculation, at constant temperature through refractometers, polarimeters etc. Finally this water bath unit replaces the need for a ready-made, costly commercial product.

Reprinted from J. Madurai Univ. Vol. VI - I, Page (86 - 92)

SOME OBSERVATIONS ON THE ANATOMY OF LITTORINA SCABRA (LINNAEUS)

H. MOHAMAD KASIM
School of Biological Sciences, Madurai University, Madurai-625021

(Received for publication 14th March 1977)

ABSTRACT

Work on tropical Littorinids are confined to limited fields. Anatomical studies of tropical species are rather scanty and relatively recent. In this work, in order to get information about anatomical details of the tropical form, **Littorina scabra** *and its reproductive and nervous systems have been studied and also some aspects of spawning.*

This is a viviparous species. There is a prominent penial gland with pigmentation. The covering gland is absent in the female reproductive system and the egg capsules are devoid of outer covering. There is a brood pouch in the mantle cavity and the development of the eggs are completed within and then the veligers are expelled into water. In the nervous system the pedal commissures are very short or rather fused together and the cerebral commissure is short and not distinct. The nerve from the pedal ganglia to the propodium and metapodium are very prominent; otherwise eac hmay bear a ganglion. This work brings about the differences between the subtropical and temperate and tropical species.

Introduction :

Representatives of the Prosobranch family Littorinidae are distributed throughout the world. The Littorinids are chiefly confined to the marine intertidal rocky shore. Some of them are well ~~penetrated~~ into the estuaries to a considerable extent. The Littorinids of the subtropical and temperate regions are subjected to extensive studies. Chiefly the morphology and anatomy of the above forms have been worked out by *Bouvier* (1887), *Johansson* (1939), *Linke* (1933, 1935), *Warner* (1950), *Thomas* (1952) and *Fretter* and *Graham* (1962). Especially an intensive study has been made on the life history, breeding and spawning of Littorinids of all regions by *Tattersall* (1920), *Sewell* (1924). *Linke* (1935), *Abe* (1936, '39) *Lebour* (1945), *Habe* (1955), *Kojima* (1957) and *Rose Water* (1963).

Works on tropical Littorinids are limited. Anatomical studies of the tropical species are rather scanty and relatively recent. The present work was carried out in order to gather details about the anatomy of the tropical forms and to study the differences between the subtropical or temperate forms and the tropical species.

Materials and Methods :

Habits and Habitat: In the present study specimens of Littorina Scabra (Linnaeus) were collected from the Vellar estuary jetty region They were found attached to the cement and wooden pillars of the jetty and also to a certain extent on the stones which are laid behind the jetty. The specimens are generally distributed within the high and low tide limits. Following are the hydrographical conditions of the jetty region. Salinity of the surface water varied from 16.5%to29.6%(s),whereas the bottom salinity varied from 20.55% to 32.94%.(s) Temperature varies between 25.2°C to 28.4°C. The surface water oxygen content fluctuates from 3.2 ml/L to 6.17 ml/L and the bottom water oxygen content from 2.69 ml/L to 4.7 ml/L. These animal feed mainly on the algae which grows on the surface of the cement and wooden poles and stones. Invariably the specimens of *Littorina scabra* are associated with Balanus and Neritids in the same area. Sometimes Balanus attaches to the shell of Littorina itself.

All the collected living specimens were kept in a glass aquarium for the studies and the water of the glass aquarium was renewed everyday with fresh estuarine water. The terminology of the soft parts is primarily that of Fretter and Graham (1962). Before conducting the dissections the snails were narcotised with menthol and after complete narcotisation (about 20 to 30 hours) the specimens were fixed in Formal acetic alcohol. Dissections were carried out under binocular dissection microscope with a magnification 10 x 6.3 or 10 x 10. Live specimens were also dissected out for the measurements of various parts of the different functional systems. The specimens used for the study of nervous system were fixed in Bouin before commencing the dissection. All the measurements were made with a micrometer eye piece and camera lucida drawings were made.

Observations :

(*a*) *Morphology of the Shell* : The shell (Fig. 1) is moderately large, generally attaining a lenth of 1″ or 1.2″, though it is by no means uncommon to find much smaller specimens measuring half an inch in length or even less; living side by side with larger specimens. The whorls are inflated and the spire is considerably elevated with sharp, pointed apex. The body whorl, as already remarked is more or less sharply angular, and being marked below this by a slightly raised sharp spiral ridge. The surface of the whorls are spirally grooved throughout and these grooves are uniform and fine in arrangement. The colour of the shell is rather variable; it is generally of a dark mottled brownish colour, the darker brown spots usually tend to be aggregated to form transpirally elongated oblique patches. which are sometimes well marked only near the sutures and the angular part of the body whorl, but sometimes extend throughout the surface.

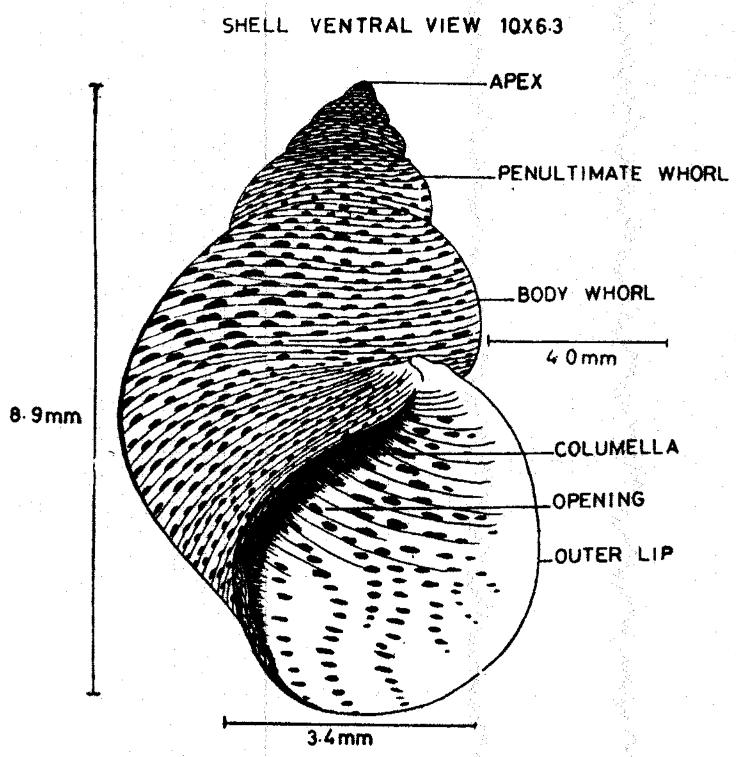


Fig. 1.

(b) Anatomy :

(i) Male Reproductive System : In males the testis (Fig. 2.) is a large branching organ which lies in the upper part of the visceral mass. It is commonly greyish brown in colour. The testicular duct runs forward in the mantle cavity and on its course it is straight at first, then becomes more convoluted as the cavity is approached where this part is used as a seminal vesicle (Fig. 2) for the storage of spermatazoa. It appears here as a chalk white tube and on reaching the posterior region of the mantle cavity it enlarges to form the prostate gland (Fig. 2). The prostate gland is of an open type with a thick fold and a groove. This gland opens anteriorly at the inner end of the mantle cavity as the malepore. From the male opening a ciliated sperm groove (Fig. 2) runs forward on the floor of the mantle cavity, on to the left side of the head and to the base of the penis (Fig. 2). The penis is a long and flattened structure with a white colour. On the right side of the penis lies a glandular mass with heavy pigmentation known as the penial gland (Fig. 2).

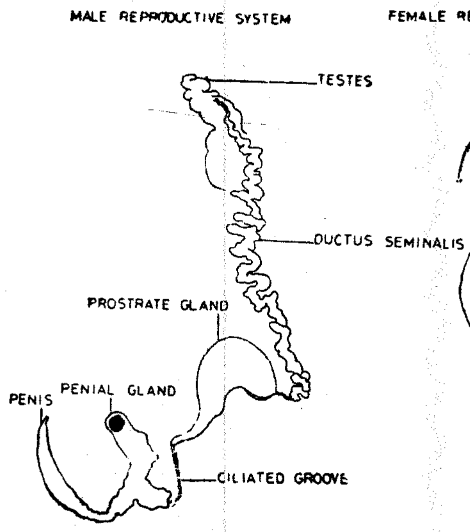


Fig. 2

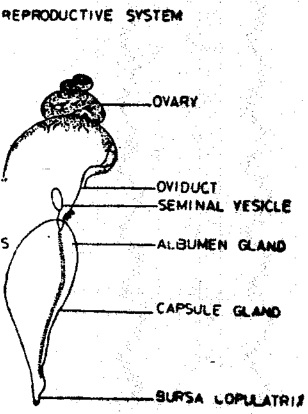


Fig. 3

ii) Female reproductive system : The ovary (Fig. 3) lies in the visceral hump in a similar position to that occupied by the testes in males. It is obviously conspicuous by its yellow structure. The oviduct (Fig. 3) from the ovary runs forward along the columellar side of the visceral hump. At one point along this part of the duct a side branch opens to the pericardial cavity and this is the gonopericardial duct (Fig. 3). The oviduct runs further to the posterior region of the mantle cavity and enlarges to form the pallial oviduct. This pallial oviduct traverses the whole length of the mantle cavity on the right side running parallel with the rectum and opens by means of a gonopore. The pallial oviduct is differentiated into a posterior albumen gland (Fig. 3) and an anterior capsule gland (Fig. 3). The left anterior region of the pallial oviduct, by the side of the female pore carries blind sac known as the Bursa copulatrix (Fig 3); from which a channel runs to the posterior region of the pallial oviduct and opens to a small blind tubule, the receptaculam seminis. The anterior part of the pallial oviduct is modified to form a broodpouch (Fig. 5) in the mantle cavity in which the embryos are retained until their development is completed. In the female there is a structure occupying a corresponding portion of the penis in the male on the right side of the head in the form of an unpigmented glandular tract running down the side towards the foot. This is the ovipositor, which carries the veliger (Fig. 6) out of the mantle cavity into the surrounding water.

iii) Nervous system : The cerebral ganglia are lying dorsal to the buccal mass one on either side of the oesophagus. Each cerebral ganglion is connected to the other by a very short cerebral commissure which passes dorsal to the anterior oesophagus. Each is connected to the pleural, pedal, and buccal ganglia by connectives namely the cerebropleural connective the cerebropedal connective and the cerebrobuccal connective (Fig. 4). In addition each cerebral ganglion gives off five nerves which innervate the snout, ventral lip, the tentacles, the eye and the fibres running in the cerebropedal connective, and the statocyst.

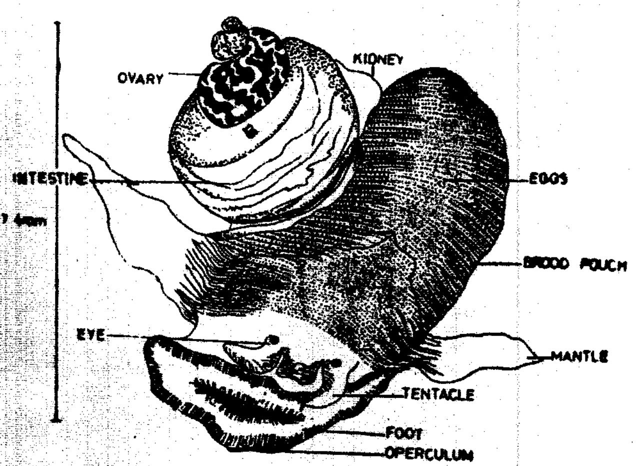


Fig. 5

The pleural ganglia lie close to the cerebrals but slightly moved anterior and ventral. Each gives off three connections to other ganglia, (a) the cerebropleural, (b) the pleuropedal and (c) the pleuroparietal connectives. The right pleural ganglion gives rise to a connective which runs, dorsal to the oesophagus, to supraoesophageal ganglion lying on the left, likewise the left pleural ganglion gives rise to a connective which runs, ventral to the oesophagus, to the suboesophageal, ganglion lying on the right side. There is no zygoneury in *Littorina scabra*. The right pleural ganglion gives off nerves which innervate the right mantle edge. The left pleural ganglion innervates a corresponding region on the left. Here is a connection between the main nerve into the mantle cavity from oesophageal ganglia and pallial nerves from the pleural ganglia. This condition establishes a sort of secondary pleuro-oesophageal connections on both the sides which is known as dialyneury.

Pedal ganglia: From each pedal ganglion arise three nerve connections to other ganglia; a) a very short almost fused pedal commissure, b) a much large thick pleuropedal connective and c) a cerebropedal connective. Each in turn gives rise to nerves to various parts of the foot. These innervations usually in groups from lobe-like expansions of the ganglion. The labial ganglion and the labial commissure have been lost, only the buccal ganglia survive in the innervation of the anterior gut walls.

Buccal ganglia: The buccal ganglia are placed lateral to the gut at the point where oesophagus

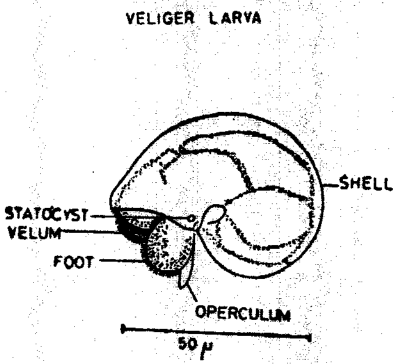


Fig. 6

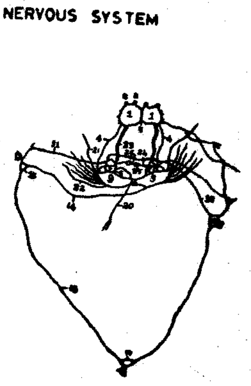


Fig 4

and radular sac originate from the buccal cavity. Each is the source of two connections to other ganglion; a) the buccal commissure and b) the cerebrobuccal connective (Fig. 4). The buccal nerves were not traced out. From the supraoesophageal, connectives run forward to the right pleural and posteriorly to the visceral ganglia; giveing off pallial, branchial and osphradial nerves. The suboesophageal ganglion is connected to the left pleural and the visceral gangalion and gives off nerves to the mantle, the anus and the reproductive system.

Visceral ganglia: These are represented by a double structure more or less fused well, lying below the integument forming the floor of the mantle cavity almost at its innermost end. Two main nerves originate from the larger ganglion; one innervates the heart and the excretory organ, while the other is a genital nerve which innervates the female and male pores according to the sex respectively.

Spawning and Development :

The spawning behaviour in *Littorina scabra* was observed in the laboratory only for a month i. e., April. Spawning activity is probably continuous throughout the year, for the availability of young immature stock is continuous with little variation in numbers. In *Littorina scabra* the embryo develops within the mantle cavity. So there is no planktonic egg capsule, and also no outer membrane is added to the egg capsules. Copulation was not observed in the laboratory and only to a limited extent the development of this species has been investigated.

Fertilisation is internal. The egg is enclosed in a covering which is devoid of an outer capsule. These eggs are of different stages in development and are embedded in a gelatinous mass within the mantle cavity, arranged in rows. The broodpouch which has a series of grooved bottom, holds the eggs. The rows of eggs which are nearer to the ovipositor are the earlier cleavage stages and the rows further posterior in the mantle cavity are yet to develop into the cleavage stage. The eggs of different stages of development in the mantle cavity suggests that the eggs were spawned into the mantle cavities on different days.

Discussion :

Littorina scabra resembles *Littorina sexatilis*, a viviparous British species in having a broodpouch in the mantle cavity. The anatomy of the reproductive and nervous system fits well into the general organisation of *Littorina littorea* (Fretter and Graham, 1962). However, *Littorina scabra* differs significantly from *Littorina littorea* in the following points: Table I to (B)

1) *Littorina scabra* is a viviparous species.

2) The presence of a prominent penial gland with a pigmentation.

3) The presence of a brood-pouch in the mantle cavity and the expulsion of veligers into water in the case of female.

4) The absence of covering gland since the egg capsules are devoid of outer covering.

5) Unlike that of *Littorina littorea* the pedal commissure is very very short or rather fused together in *Littorina scabra*.

6) The cerebral commissure is short and not distinct

7) The nerve from the pedal ganglia to the propodium and metapodium are very prominent or otherwise each may bear a ganglion.

TABLE 1.

No.	Particulars	Average length (mm)	Average width (mm)
1	Ductus seminalis	33.2	.57
2	Prostrate gland	6.3	2.1
3	Sperm ciliated groove	3.6	
4	Penial gland	3.6	
5	Penis	6.0	

TABLE 2.

No.	Particulars	Average length (mm)	Average width (mm)
1	Oviduct	3.7	
2	Seminal vesicle	1	
3	Albumen and Capsule gland	9.7	3.6

Average length and width of different parts of male and female reproductive systems of Littorina scabra.

TABLE 3.

No.	Particulars	Average length (mm)	Average width (mm)
1	Right cerebral ganglin	1.0	.29
2	Left cerebral ganglion	1.0	.29
3	Cerebral commissure	.57	
4	Supra oesophageal ganglion		.29
5	Supra oesophageal nerve	2 3	
6	Sub oesophageal ganglion		.14
7	Sub oesophageal nerve	2.6	
8	Left visceral loop	5.4	
9	Right visceral loop	4.1	
10	Right pedal ganglion		.57
11	Left pedal ganglion		.72
12	Cerebro pedal connective	1.9	
13	Pleural ganglion Right		.14
14	Pleural ganglion left		.29

Average length and width of various parts of nervous system of Littorina scabra.

There would appear to exist a connection between the state of development of the gonad and the state of development of the secondary sexual organs in *Littorina littorea* and *Littorina sexatilis* i. e., during the breeding season when the gonad is ripe, the penis, prostate and pallial oviduct are at maximal size (Fretter, 1962). For *Littorina scabra* the observations were made only during the maximal breeding season and so the condition of the gonad and the secondary sexual organs during nonbreeding season has not been observed. In a number of mesogastropods the vas-deferens anterior to the renal section is an open groove (Fretter and Graham 1962).

In this *Littorina scabra* resembles the British species *Littorina littorea*, *Littorina littoralis* and *Littorina sexatilis*. As suggested by Linke (1933) the seminal receptacle is placed at the inner end of the pallia, oviduct in *Littorina scabra*. Spawning and development has not been studied completely, only the veligers were observed in the laboratory.

Acknowledgement

I am chiefly indebted to Dr. R. Natarajan Director, C. A. S. in Marine Biology, Porto Novo for all the facilities and encouragement. My heartful thanks to Mr. Govindan and Mr. Kasinathan, who helped me very much by giving valuable suggestions wherever necessary. And my thanks to Mr. Venkataramanujam who helped in the identification of the gastropod.

Explanation for Figure 4.

1 Pedal ganglion
2 Propodial nerve
3 Metapodial nerve
4 Cerebropedal connective
5 Pedal commissure
6 Cerebropleural connective
7 Left pleural ganglion
8 Right pleural ganglion
9 Cerebral ganglion
10 Cerebral commissure
11 Pallial nerves
12 Sub Oesophageal nerve
13 Sub Oesophageal ganglion
14 Supra Oesophageal nerve
15 Supra Oesophageal ganglion
16 Visceral loop
17 Visceral ganglion
20 Nerve to columellar muscle
21 Tentacular nerve
22 Optic nerve
23 Pleuropedal connective
24 Cerebrobuccal connective
25 Buccal ganglion
26 Buccal commissure

References :

Abe, N. 1936 Ecological observation of Melarhaphe (Littorinopsis) scabra (Linnaeus) mainly on its locomotion. Ecol. Rev, Sendai 2, 35-42

———— 1939 Ecological observation on Melarhaphe Littorinopsis scabra (Linnaeus) inhabiting the mangrove-tree. Palao trop. biol. Stn stud. 1, 391-435.

Bouvier, E. 1887 System nerveux, morphologic generate et classification des gastropodes prosobranchs. Ann Sci. Natur. Zool, Ser. 7, Vol. 3, 1-510.

Fretter, V., & A. Graham. 1962 British prosobranch molluscs Ray Soc., London. 755 pp. Barthlomew Press, Dorking, U.K.

Habe, T. 1953 Eggs and larvae of Japanese gastropods. Publ. Seto Marine Biol. Lab. 3.

Johansson, J. 1939 Anatomische studien über die Gastropoden tamilien Rissodae und Littorinidae. Zool. Bidr Uppsala Vol. 18, pp. 289-296

Kojima, Y. 1957 On the breeding of periwinkbe Littorivaga brevicula (Philippi). Bull. biol. Stn Asamushi 8 : 59-62.

Lebour, M. V., 1945 The eggs and larvae of some prosobranchs from Bermuda. Proc. Zool. Soc. Lond. 114 : 262 -289.

Linke, O. 1933 Morphologic und physiologic des genital apparates der Nordscelittorines. Wiss. Meeresunters, Abt. Helgoland 19 : 3-52.

———— 1935 Der Laich Von Littorina (Melarhaphe) neritoids L Zool. Anz 112: 37-62.

Rosewater, J. 1963 **Problems of species analogues in world Littorinidae. Rep. Am. Malac. U, Bull. 30 : 3-6.**

Sewell, **R. B. S.** 1924 **Observations on growth in certain molluscs and on changes correlated with growth in the radula of Pyrazus palushis. Rec. Indian Mus 26 : 529-548.**

Tattersal, W. M. 1920 **Notes on the breeding habits and Life history of periwinkle Sci. Invest Fish. Br. Jre 1 : 1-11.**

Thomas, S. S. 1952 **The mollusca of Kurusadi Island Bulletin of Madras Govt. Vol. 1 (64, 65).**

Warner 1950 **External anatomy and morphology of the alimentary tract of Littorina irrorata. Journ. Tennessy Acad. Sc., 25.**