ALLOMETRIC GROWTH COEFFICIENTS AND BIOCHEMICAL INDICATORS OF 'CONDITION' IN AIR BREATHING FISHES

DISSERTATION SUBMITTED IN PARTIAL FULFILMENT FOR THE AWARD OF THE DEGREE OF MASTER OF PHILOSOPHY IN ZOOLOGY

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DIVISION OF ICHTHYOLOGY AND FISHERIES DEPARTMENT OF ZOOLOGY ALIGARH MUSLIM UNIVERSITY ALIGARH 1981
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ALIGARH MUSLIM UNIVERSITY
ALIGARH, U. P. INDIA

This is to certify that the dissertation entitled
's, allometric growth coefficients and biochemical indicators
of condition in air breathing fishes' has been completed
under my supervision by Y. M. M. A. H. A.

The matter
embodied here is original and has been independently
pursued by the candidate. It reports some interesting
observations and is a distinct addition to the existing
knowledge on the subject.

I permit the candidate to submit the dissertation
in partial fulfilment for the award of the degree of
Master of Philosophy in Zoology of the Aligarh Muslim
University, Aligarh.

Jaleel Mustafa
Lecturer.
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I would like to express my deep sense of gratitude and indebtedness to Dr. Saleem Mustafa under whose supervision this work was completed and to Prof. Hawab Hasan Khan, Head of the Department, for providing laboratory facilities.

Thanks are also due to my laboratory colleagues for their cooperation.
GENERAL INTRODUCTION
Studies have been carried out by several workers on the dynamics of change in the weight of internal organs of fishes in relation to starvation, ration size, food composition, etc. (Fontaine and Halsey, 1953; Chang and Idler, 1960; Crouch and Cournot, 1965; Antsyshkina et al., 1971; Tyler and Dunn, 1976; Yakovelva et al., 1976; Seidinger and Crawford, 1977; Buckley, 1979a, b; Shama, 1980; Mustafa and Kittal, 1981). These investigations have revealed differences in the nature of response of different organs to the various factors. There seems a general unanimity about liver as the most dynamic visceral organ of the body. Its weight undergoes significant fluctuations depending upon the degree of depletion and accumulation of biochemical reserves under various life processes. Quantitative biochemical and weight changes of an organ in preference to others emphasize the need for indepth study of the internal organs of fish. This will enable the revelation of the relative metabolic stability of vital organ(s) and selection of the organ most sensitive to dietary and other factors. Field biologists and fisheries management scientists require a reliable index to determine the condition of fishes resulting from physical and chemical changes in their environment especially due to pollutants.
and to monitor the effects of short-term changes in supply of food and its consumption by fishes. In addition to liver weight, nucleic acid ratios have emerged as a sensitive parameter. This has been amply indicated by Satomi and Ishida (1976); Bulow et al. (1978); Satomi (1978); Buckley (1979a, b); Buckley (1980); Ohama (1980). These authors have outlined the potential of their findings and emphasised the need to carry out more research on those and allied lines.

In the present dissertation the data have been compiled in two chapters. Chapter I contains information on the allometric growth relationships of internal organs (heart, brain, liver) with the body in two species of air breathing freshwater fishes: Heteropneustes fossilis (Lloch), the common catfish, and Channa punctata Bloch, the common pond murrel. Chapter II deals with the quantitative relationship of Hx, DHA and specific gravity with the living 'condition' in Heteropneustes fossilis (Bloch).
CHAPTER - I
Introduction

The process of allometric growth and scaling arise from the interaction of a pair of simple geometric and functional relationships but these relationships are often seen to lack explanation (Coulé, 1975). A survey of literature of embryology, growth studies, biological dimensional analysis, physiological ecology, evolutionary ecology, paleontology, systematics, locomotor mechanics and energetics reveals a wide range of scaling relationships which are hitherto unexplained (Sweet, 1980). Obviously, interpretation of allometric relations of various internal organs with a functional bias assumes considerable importance. This has been attempted here.

It is well known that depletion of nutrient reserves of internal organs of fish or their accumulation therein under different conditions of life results in noticeable changes in the weights of the concerned organs. Intensity of feeding and cycle of sexual maturation are two of the factors known for their profound influence on biochemical
constituents of fishes. An overwhelming number of studies point out that liver is the most vulnerable site for the turnover of the nutrients (Fontaine and Ratey, 1953; Chang and Idler, 1960; Bulow, 1971; Hustaja, 1976; Chams, 1980, Hustaja and Kittel, 1981).

Vital organs like heart and brain maintain remarkable stability; virtually all of their constituents remain intact for unimpaired functioning, even under extremely stressful condition (Hustaja, 1976) which do not spare intestine, liver, kidney, muscle, etc. Study of the condition of internal organs as evaluated through their weight is, therefore, of considerable importance in understanding the physiological response of fish body to intrinsic and extrinsic factors. The present report based on two air breathing teleosts *Heteropneustes fossilis* and *Channa punctatus* attempts to furnish information on these aspects and also on interspecific differences.

**MATERIALS AND METHODS**

Live specimens of *Heteropneustes fossilis* (total length 18-34 cm., body weight 34-279 g) and *Channa punctatus* (total length 10.5-24.5 cm; body weight 14-170 g) were procured from local ponds at Allgarh (lat. 27°34'36"N, long 78°4'26"E) and reared in aquaria. Water in aquaria was
changed daily. During the experimental period the fishes were fed to satisfy level with frozen minced meat. After recording the total length and body weight of fish the internal organs (heart, brain, liver) were dissected out, placed on blotting paper to remove the adhering fluid. Heart was emptied of its contained blood. Gall bladder was detached from the liver and discarded. Weights of internal organs were recorded on a sensitive electric balance. The cardio-somatic index, cephalo-somatic index and hepato-somatic index, termed in short as heart ratio, brain ratio, and liver ratio, respectively, were calculated by the following formula:

\[
\frac{\text{set weight of the concerned organ, } g}{\text{set weight of the fish (-gonad weight)} g} \times 100
\]

Jeidinger and Crawford (1977) have emphasized the importance of deleting gonad weight from intact body weight in determining the ratios of the internal organs and also suggested the exclusion of gall bladder before recording the liver weight. Only female specimens were selected for study since males were not available in adequate numbers.

The relationships of the weight and ratios of the various organs with the total length and body weight of fish specimens were expressed by the following standard
Figure 1. Allometric relations of some internal organs (heart, brain, liver) of *Istherophamus fossilis* (left side) and *Channa punctatus* (right side).
Table - I

<table>
<thead>
<tr>
<th>Total Body Length, cm</th>
<th>Total Body Weight, g</th>
<th>Weight of Internal Organs, g</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Heart</td>
<td>Brain</td>
</tr>
<tr>
<td></td>
<td>(24)</td>
<td>(24)</td>
</tr>
<tr>
<td>Heteropneustes fossilis</td>
<td>16-34</td>
<td>34-279</td>
</tr>
<tr>
<td></td>
<td>(19)</td>
<td>(23)</td>
</tr>
<tr>
<td>Channa punctatus</td>
<td>10-20</td>
<td>14-119</td>
</tr>
<tr>
<td></td>
<td>(19)</td>
<td>(22)</td>
</tr>
</tbody>
</table>

Values in parentheses indicate number of observations.

Table - II

<table>
<thead>
<tr>
<th>Total Body Length, cm</th>
<th>Total Body Weight, g</th>
<th>Ratios of Internal Organs</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Heart</td>
<td>Brain</td>
</tr>
<tr>
<td></td>
<td>(24)</td>
<td>(24)</td>
</tr>
<tr>
<td>Heteropneustes fossilis</td>
<td>16-34</td>
<td>34-279</td>
</tr>
<tr>
<td></td>
<td>(19)</td>
<td>(23)</td>
</tr>
<tr>
<td>Channa punctatus</td>
<td>10-20</td>
<td>14-119</td>
</tr>
<tr>
<td></td>
<td>(19)</td>
<td>(22)</td>
</tr>
</tbody>
</table>

Values in parentheses indicate number of observations.
Table - II

Relationships of weights and ratios of some internal organs with body length and weight in *Heteropneustes fossilis* and *Channa punctatus*

<table>
<thead>
<tr>
<th>Species of fish</th>
<th>No. of observations</th>
<th>Parameters</th>
<th>Regression equation</th>
<th>Correlation coefficient significance</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Heteropneustes fossilis</em></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>24</td>
<td>Heart weight, (Y(g)/) Body weight, (A(g))</td>
<td>Log (Y = -1.947 + 0.685 \log A)</td>
<td>0.519, (p &lt; 0.001)</td>
<td></td>
</tr>
<tr>
<td>24</td>
<td>Brain weight, (Y(g)/) Body weight, (A(g))</td>
<td>Log (Y = 0.627 + 0.139 \log A)</td>
<td>0.357, (p &lt; 0.001)</td>
<td></td>
</tr>
<tr>
<td>24</td>
<td>Liver weight, (Y(g)/) Body weight, (A(g))</td>
<td>Log (Y = -1.731 + 0.973 \log A)</td>
<td>0.981, (p &lt; 0.001)</td>
<td></td>
</tr>
<tr>
<td>17</td>
<td>Heart weight, (Y(g)/) Body length, (A(Cm))</td>
<td>Log (Y = 10.026 + 3.178 \log A)</td>
<td>0.476, (p &lt; 0.001)</td>
<td></td>
</tr>
<tr>
<td>17</td>
<td>Brain weight, (Y(g)/) Body length, (A(Cm))</td>
<td>Log (Y = 1.756 + 0.564 \log A)</td>
<td>0.956, (p &lt; 0.001)</td>
<td></td>
</tr>
<tr>
<td>17</td>
<td>Liver weight, (Y(g)/) Body length, (A(Cm))</td>
<td>Log (Y = 10.892 + 3.624 \log A)</td>
<td>0.967, (p &lt; 0.001)</td>
<td></td>
</tr>
<tr>
<td>19</td>
<td>Heart ratio, (Y/) Body weight, (A(g))</td>
<td>Log (Y = 1.297 - 0.112 \log A)</td>
<td>0.567, (p &lt; 0.001)</td>
<td></td>
</tr>
<tr>
<td>23</td>
<td>Brain ratio, (Y/) Body weight, (A(g))</td>
<td>Log (Y = 4.254 - 0.751 \log A)</td>
<td>0.976, (p &lt; 0.001)</td>
<td></td>
</tr>
<tr>
<td>22</td>
<td>Liver ratio, (Y/) Body weight, (A(g))</td>
<td>Log (Y = 1.903 + 0.060 \log A)</td>
<td>0.521, (p &lt; 0.001)</td>
<td></td>
</tr>
<tr>
<td>15</td>
<td>Heart ratio, (Y/) Body length, (A(Cm))</td>
<td>Log (Y = 3.066 - 0.654 \log A)</td>
<td>0.775, (p &lt; 0.001)</td>
<td></td>
</tr>
<tr>
<td>21</td>
<td>Brain ratio, (Y/) Body length, (A(Cm))</td>
<td>Log (Y = 0.515 - 2.555 \log A)</td>
<td>0.942, (p &lt; 0.001)</td>
<td></td>
</tr>
<tr>
<td>21</td>
<td>Liver ratio, (Y/) Body length, (A(Cm))</td>
<td>Log (Y = 1.311 + 0.267 \log A)</td>
<td>0.573, (p &lt; 0.001)</td>
<td></td>
</tr>
</tbody>
</table>

Table contd. on next page
### Table II Contd.

<table>
<thead>
<tr>
<th>Species of fish</th>
<th>No. of observations</th>
<th>Parameters</th>
<th>Regression equation</th>
<th>Correlation coefficient</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Heart weight, Y (g)/</td>
<td>Log $Y = -0.357 + 0.810 \log X$</td>
<td>0.90</td>
<td>$&lt; 0.001$</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Body weight, $X$ (g)</td>
<td>Log $Y = -0.213 + 0.337 \log X$</td>
<td>0.955</td>
<td>$&lt; 0.001$</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Brain weight, Y (g)/</td>
<td>Log $Y = 1.687 + 0.536 \log X$</td>
<td>0.962</td>
<td>$&lt; 0.001$</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Liver weight, X (g)/</td>
<td>Log $Y = -7.906 + 2.680 \log X$</td>
<td>0.990</td>
<td>$&lt; 0.001$</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Body length, $X$ (Cm)</td>
<td>Log $Y = -2.314 + 1.055 \log X$</td>
<td>0.850</td>
<td>$&lt; 0.001$</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Brain weight, Y (g)/</td>
<td>Log $Y = -7.436 + 2.880 \log X$</td>
<td>0.956</td>
<td>$&lt; 0.001$</td>
</tr>
<tr>
<td>Channa punctatus</td>
<td></td>
<td>Heart ratio, Y/Body weight, $X$ (g)</td>
<td>Log $Y = 1.694 - 0.192 \log X$</td>
<td>-0.739</td>
<td>$&lt; 0.001$</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Brain ratio, Y/Body weight, $X$ (g)</td>
<td>Log $Y = 3.657 - 0.671 \log X$</td>
<td>-0.576</td>
<td>$&lt; 0.001$</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Liver ratio, Y/Body weight, $X$ (g)</td>
<td>Log $Y = -1.724 + 1.091 \log X$</td>
<td>0.582</td>
<td>$&lt; 0.001$</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Heart ratio, Y/Body length, $X$ (Cm)</td>
<td>Log $Y = 1.654 - 0.201 \log X$</td>
<td>-0.551</td>
<td>$&lt; 0.001$</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Brain ratio, Y/Body length, $X$ (Cm)</td>
<td>Log $Y = 2.084 - 2.102 \log X$</td>
<td>-0.987</td>
<td>$&lt; 0.001$</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Liver ratio, Y/Body length, $X$ (Cm)</td>
<td>Log $Y = 1.500 + 0.196 \log X$</td>
<td>0.423</td>
<td>$&lt; 0.005$</td>
</tr>
</tbody>
</table>
regression equation:

\[ \log y = \log a + b \log A \]

where \( i \) was the weight (g)/ratio of the organ,
\( A \) was the total length(cm)/body weight (g)
of the fish,
\( \log a \) was the intercept of the regression line,
b was the slope of the regression line.

\( \log a \) and \( b \) were evaluated by the standard method
of least squares (Coulson, 1952).

In this work, the values used for plotting in Fig. 1
as well as deriving the regression equations are in \( \log_{10} \),
obtained by multiplication of the arithmetic values of the
various measurements of the body and internal organs by 100.

RESULTS AND DISCUSSION

Weight of the heart, brain and liver seem to be fairly
correlated with both weight and length of the body in
*Heteropneustes fossilis* and *Channa punctatus* (Table IA, II;
Fig. 1). The value of slope in the regression analysis of
these relations (Table II) revealed that weight of the heart
varied 0.68 times the body weight and 3.17 times the body
length in *Heteropneustes fossilis*, whereas in *Channa punctatus*
heart weight was 0.81 times and 2.66 times the body weight
and body length, respectively. Other workers have also found heart-body weight relationship with a slope appreciably less than 1.0. Foupa and Castadal (1969) reported a slope of 0.74 for poikilotherms, 0.69 for birds and 0.65 for mammals. Claridge and Potter (1974) worked out the respective slopes for different stages of the life cycle of lampreys as 1.03 – 1.04 (*Isopyrus marinus*); 0.97 – 1.19 (*Lampetra fluviatilis*); 0.92 – 1.15 (*Lampetra planeri*). Higher heart ratio in *Channa punctatus* (0.109) compared to *Heteropneustes fossilis* (0.073) is no doubt due to difference in the size and weight of heart relative to body in these species; heart weight proportionate to body weight must evidently be greater in the catfish. Presumably the nature of myocardial organisation may be the basis. The muscle fibres in the myocardium may be more densely packed in *Channa punctatus* whereas in the heart of *Heteropneustes fossilis* the arrangement of fibres may be loose. The compactness of heart muscle is known to increase the efficiency of this organ, enabling it to increase its metabolic activity (Claridge and Potter, 1974). Findings of Hill and Potter (1970), and Rogers (1972) substantiate this theory. Wrams and Taylor (1965), and Foupa et al. (1970) went to the extent of making strong correlation between heart ratio and activity of fish.
Increase in the heart weight with length and weight of body signifies that myocardium thickens and becomes more dense in structure during the growth process. However, the progressive decline in the heart ratio with increase in body length and weight (Table Ia and Fig. 1) is almost certainly due to increase in body weight at a rate faster than that of the heart.

Brain weight scales in proportion to the 0.139 and 0.337 power of the body weight in Heteropneustes fossilis and Channa punctatus, respectively, and 0.664 power of body length in Heteropneustes fossilis and 1.055 in Channa punctatus (Table II). The slope of the regression relationship in these parameters suggests that fish of fairly large range in length and weight have approximating brain weight. Thus exponentially declining curves were obtained when brain ratio was plotted against increasing body length and weight (Table II; Fig. 1) revealing that relative growth of brain in weight lags farther behind the progression in length and weight of body of the fish Landry (1960) also observed almost no change in the brain size of several ectotherms over the range of 0-100 gms of body weight, but noted dramatic enlargement of brain size in endotherms.

The slope of logarithmic relationships between liver weight and body weight was close to 1.0 (Table II).
(precisely 0.97 in *Heteropneustes fossilis* and 0.93 in *Channa punctatus*). This closeness of the value to unity suggests that the rate of gains in liver weight approximates the growth in body weight, and emphasizes a tendency of liver weight to scale isometrically with respect to intact body weight. Linearly close progression of liver ratio with the body length and weight strengthens this fact (Table II, Fig. 1). This tendency, however, falls short of obliterating the allometry on howsoever small scale it exists.

Hence the regression line involving the liver ratio runs nearly but not completely parallel to the abscissa (Fig. 1). When liver weight is considered in relation to growth in length, allometric coefficient of 3.02 (*Heteropneustes fossilis*) and 2.08 (*Channa punctatus*) were obtained (Table II), showing that this organ adds weight a little less than four times the body length in catfish and a little less than three times the body length in the murrel. Inter-specific differences evidently exist in the allometric coefficient for liver weight/body weight and liver weight/body length, with higher coefficient in *Heteropneustes fossilis* emphasizing that gains in weight or length of body of this fish accompany proportionately greater increase in liver weight. Liver weight increases chiefly by the accumulation of glycogen and fat (Phillips *et al.*, 1960;
Wangaard et al., 1967; scarf and randall, 1971; holt, 1971; edward et al., 1972; winton et al., 1972; shuman, 1974; ratland et al., 1976; jensen, 1975). With growth of the body more and more quantities of these nutrients are stored in the liver. This is advantageous since fishes become more fecund as they grow and the increasing quantities of organic constituents of the liver are mobilised and supplied towards gonad buildup, while keeping the metabolic integrity of the liver intact. It is thus interesting to note that liver weight and fecundity both increase two-to-three times the rate of increase in body length of the fishes. Yasmin and rayyam, (1963), dhargava (1971) and shott (1977) have documented these fecundity—body length relations in the fishes.

Several workers have reported fluctuations in liver/body weight ratios and correlated them with gonadal maturation and feeding intensity, etc. (saud and cross, 1966; antyskina et al., 1971; tyler and wunn, 1976). According to jensen (1979) liver/body weight ratio could not be used a true index of fish condition especially its nutritional status, due mainly to considerable difference in the response of liver and condition to various ecological and biological factors. The same author pointed out the utility of biochemical analysis in furnishing data important
in evaluation of the physiological condition of the body.

SUMMARY

Good correlations existed between the weight of heart, brain, liver each and weight as well as length of the body in teleosts, *Heteropneustes fossilis* and *Channa punctatus*. Relationship of cardio-somatic, cephalo-somatic, and hepato-somatic indices with the body length and weight were also worked out. The three investigated organs were found to scale allometrically with the body in varying degrees. Nature of these relations were discussed. Inter-specific differences observed in various parameters outlined have been interpreted.
In fishery biology investigations determination of the condition coefficient is basically meant to indicate the relative heaviness of fishes. This coefficient is, however, a quantitative measure of the deviation of case of an individual from average case for given length, and fails to exactly reveal the underlying qualitative changes in the body which can truly be indicator of the physiological or nutritional status of fish. Efforts have been made to employ certain biochemical constituents of significance as indices of condition or the general well being of fish. Caulton and Lursell (1977) reviewed literature wherein lipid was regarded as an important reserve contributing to condition, and 'fat indices' were derived as correlates of condition. In a recent communication Mustafa and Jafri (1981) emphasised the importance of fat and glycogen levels as indicators of living condition in catfish *Heteropneustes fossilis*. Some other constituents used for the purpose include protein and ...
the fact that growth is mainly a function of protein biosynthesis. Love (1962) used protein concentration as an index of condition in cod Gadus morhua. Based on the assumption that 

\[ \text{growth} \rightarrow \text{protein} \rightarrow \text{condition} \]

is chiefly responsible for protein synthesis in the body which accomplishes the growth. Mustafa (1979) pointed out the utility of these organic constituents in explaining condition in murrel Channa punctatus. The present study was followed up to find out the relation of AM and BM concentrations in flesh and specific gravity of the tissue with the condition index (fillet condition factor) in catfish Heteropneustes fossilis (Bloch).

MATERIALS AND METHODS

Live specimens of the catfish of the size 25 ± 0.68 cm (mean ± S.D.) and body weight 105.3 ± 8.62 g (mean ± S.D.) were collected from local ponds at Aligarh (Lat. 27°34'30"N, Long. 78°4'26"E) and transferred to glass aquaria. They were fed to satiety by providing chopped meat at the rate of 3% of the body weight per day. Unused food was siphoned off. Since males were not available in adequate numbers, only female specimens formed the basis of present study. At the time of investigation fish were taken out of aquaria and decapitated. Fillets were removed and weighed in a sensitive electric balance. Fillet condition factor (C)
was calculated according to the equation suggested by Stikhin (1967):

\[ V = \frac{E}{3} \times 1000 \]

where, \( E \) and \( L \) were weight (g) and length (cm) of fillet.

Specific gravity of white trunk muscle was determined as the ratio of the mass of the tissue/mass of an equal volume of distilled water. Sample of white muscle for analysis of \( \text{NH}_4 \) and \( \text{DHA} \) was removed from the epaxial portion of trunk, below the place of origin of dorsal fin.

Dry fat free tissue was obtained according to the technique of Webb and Levy (1955). Known weight of the sample was homogenised in distilled water and treated with 2 volumes of 10%, trichloro acetic acid (TCA). The contents were centrifuged at 5000-6000 rpm for 15 minutes. Supernatant containing the acid soluble substance was discarded and the process repeated. The pellet was washed repeatedly with ethanol to remove the lipoidal substances. The tissue residue was then treated several times with solvent ether and traces of the solvent were removed by putting the tissue remains in thermostat. The entire process ultimately yielded a white fat-free dry material in powdered form which was used for \( \text{NH}_4 \) and \( \text{DHA} \) bioassays. \( \text{DHA} \) was extracted and estimated following the method of Schneider (1957).
Figure 1. Standard curve of Eq.
100 mg of the sample obtained by the procedure described above was suspended in 2.0 ml of 1N potassium hydroxide and incubated for 20 hrs. at 37°C. After incubation 0.4 ml of 6N hydrochloric acid and 2.0 ml of 5% HCl were added. The contents were mixed, centrifuged at 3000-4000 rpm for 10 minutes and filtered. NH₃ was estimated in the filtrate through the orcinol reaction. Orninol was purified by boiling in benzene, decolorized with charcoal and recrystallized with hexane. A known weight of purified orcinol was dissolved in concentrated hydrochloric acid containing ferric chloride (0.5%) to a concentration of 1%. This orcinol reagent was mixed with equal volume of diluted DNA extract and the mixture was heated in boiling water bath for about 30 minutes. Contents were cooled to room temperature and the intensity of color developed was read at 660 nm wave length after setting the instrument to zero density with the blank. Blank was prepared by substituting the DNA extract with distilled water and processing the contents in a similar way. Values were read off against a calibration curve (fig. 1) relating optical density to micrograms of DNA, with purified yeast RNA serving as the standard.

For extraction of DNA, 100 mg of the dry, fat-free sample was suspended in 5.0 ml of 5% HCl. The content was
Figure 2. Standard curve of D14.
Figure 3: Relations of specific gravity, and concentrations of LHA and DHA with fillet condition factor in *Heteropneustes fossilis*.
Table I
Specific gravity, concentrations of n-3 and DHA in white muscle of *Heteropneustes fossilis* of different fillet condition factors

<table>
<thead>
<tr>
<th>Total length (cm)</th>
<th>Body weight (g)</th>
<th>Fillet condition factor</th>
<th>Specific gravity</th>
<th>DHA (μg/100 mg)</th>
<th>EPA (μg/100 mg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>15.0</td>
<td>34.0</td>
<td>1.926</td>
<td>1.052</td>
<td>653.79</td>
<td>399.5</td>
</tr>
<tr>
<td>18.5</td>
<td>36.0</td>
<td>1.937</td>
<td>1.111</td>
<td>635.90</td>
<td>279.6</td>
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heated in boiling waterbath for 30 minutes and then cooled to room temperature in running tap water. Loss of volume during boiling was compensated by the required amounts of 5°C. The sample was filtered and quantitated in the filtrate according to the technique of Ashwell (1957). To a 0.5 ml of dib extract were added 0.05 ml of 5% solution of cystein in water, 5.0 ml of 70% sulphuric acid and contents were mixed and allowed to stand at room temperature for 15 minutes for color development. The colour was measured at 490 nm wave length against the blank and compared with known values plotted in a standard curve (Fig. 2). The standard curve was prepared by taking highly polymerised calf thymus DNA. LHA and DNA values were expressed as μg/100 mg on dry fat-free weight basis.

The relationships between different parameters (LHA, DNA, specific gravity, each with condition) were evaluated through the standard regression equation (Couldey, 1952).

RESULTS AND DISCUSSION

Results of this study show that concentration of LHA increases with fillet condition factor while that of the DNA decreases exponentially (Table 1; Fig. 3). The relation being more of a curvilinear nature, the method of graphic...
approximation of the two parameters gives a good visual conceptions of the existing relationship but the empirical derivation of the value of one parameter for a given value of the other through the regression equation \( \text{log } Y/100 \text{ mg} = 15.467 + 255.280 \times \) is just not pertinent and fair in as much as the slope is not straight and the curve can be divided into segments each of which is nearly rectilinear, having its own value. A single value of slope(b) as derived in equation will not accurately apply to any of the different slopes. Discrepancy is, therefore, obtained between the values of log for particular condition found out through graph and the ones calculated through the equation. To indicate the amount of difference, we selected four points along different regions of the curve to work out EMP concentration correlating to condition. Comparison of this data with the empirically determined values is given as under:

<table>
<thead>
<tr>
<th>Inlet condition factor</th>
<th>AsA concentration (μg/100 mg)</th>
<th>Graphically evaluated</th>
<th>Empirically evaluated</th>
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<tr>
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<td>540</td>
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The comparison is enough to point out that formulation of curvilinear relation between $\ln d$ and fillet condition factor in the form of one and only one equation is not justifiable as the $\ln d$ concentration tends to be over-estimated if calculated on its basis. In an earlier communication Lustafa (1979) reported a linear relation between $\ln d$ and fillet condition factor in a murrel Channa punctatus and evolved an equation fairly descriptive of this relation. However, Shams (1980) working on catfish Clarias batrachus suggested caution in seeking out statistical relations of universal importance on the pattern of regression models. Increase in $\ln d$ with fillet condition factor also points to the role of this nucleic acid in robustness of the fish in view of its involvement in protein biosynthesis and hence growth. Work of Lustafa (1979) on the relationship of $\ln d$ and protein with condition and strikingly close relationship between these two components strengthens this view. Considerable data is also available to prove the correlation between $\ln d$ turnover and growth rate of fish (Bulow, 1970, 1971; Haines, 1973; Bulow, 1974; Lustafa and Safri, 1977; Buckley, 1978b; Lustafa, 1979; Buckley, 1980; Shams, 1980; Lustafa and Mittal, 1981).

During gain in weight for a given length, the nutrients, chiefly protein, evidently accumulate in the cell cytoplasm, and change the specific gravity of the tissue in the direction
of a definitive increase (Table I; Fig. 3). The formula that describes the relation of specific gravity with fillet condition factor is:

\[ \text{specific gravity} = 0.870 + 0.109 \cdot C \]

Muscle tissue obtained from a fish of higher fillet condition has higher specific gravity. It must not, however, be overlooked that alteration in specific gravity may be a common attribute of many cellular components like fats, water, ash, in addition to protein. Undoubtedly the influence of protein overrides. In any case it is this variation in the specific gravity of fish tissues that limits the applicability of cube law in fishery investigations involving length-weight relationships and condition of fish.

Decline of DNA concentration with increase in the fillet condition factor (Table I; Fig. 3) as seen in the present study, does not contradict the universally accepted view vis-à-vis metabolic stability of this genetic material. This apparent decrease is in amount/unit weight of tissue and not DNA content/cell. The regression equation establishing the relationship between the two parameter is:

\[ \text{DNA (µg/100 mg)} = 568.189 - 170.116 \cdot C \]

Since quantitative increase in the fillet weight and
condition is a function of accumulation of biochemical constituents in cells in the form of cytoplasmic inclusions evidenced by rise in specific gravity of muscle tissue. A given volume of tissue becomes heavier than equivalent volume of sample excised from a fish of low condition and whose cytoplasmic reserves are in smaller quantity and specific gravity lower. A smaller number of cells of larger weight can contribute to a unit weight of sample compared to larger number of cells of lower weights. Cell content, which is related to the number of cells in a tissue (Notchfis, 1955; Leslie, 1955; Dulow, 1970; Jafri and Lustafa, 1970; Lustafa, 1977b; Lustafa and Jafri, 1970; Lustafa and Mittal, 1981) is greater in tissue sample from a depleted fish (poor condition) than the one obtained from robust specimens (good condition) where Ws seems to be 'diluted' by piling up of substantial quantities of several nutrients in the cells.

SUMMARY

A study was conducted to determine the relation of 
\( \text{As} \) and \( \text{W} \) concentrations in body tissue with 'condition' in catfish *Heteropneustes fossilis*. \( \text{As} \) was curvilinearly related to condition while \( \text{W} \) maintained a reciprocal relation. Biochemical basis of changes in condition and their effect on specific gravity of the tissue has been interpreted. The said relationships have been formulated.


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Austafa, O. and Safr, ... 1978: Some biochemical constituents and the calorific values of different regions of the body musculature of pond murrel *Channa punctatiss* (Boh.). *Fish. Technol.* 15: 57-59.


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