# Length and Weight (LW) Related Variations on Haematological Parameters of Hetropneustes Fossilis (Bloch.)

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## Abstract

The paper mainly described the haematological values of a fresh water catfish *Hetropneustes fossilis* (Bloch.) in relation to its length and weight which are the prime basic factor of the differences in blood parameters of any living creature including fishes. The hematological parameters showed significant differences with increase in length and weight of the fishes. Differential leucocytes count did not showed any significant differences except macrophages and differential erythrocytes count showed significant differences only in macrocytes. All three groups (Length and weight) of fishes showed significant positive correlations for total erythrocytes count, hemoglobin, mean corpuscular hemoglobin and erythrocyte sedimentation rate while negative correlation was noted for mean corpuscular hemoglobin; mean cell volume. Among differential leukocytes count only neutrophil showed positive correlation, while other parameters did not show any significant correlation. In differential erythrocyte count only microcytes showed significant positive correlation.


## Introduction

A major part of the world's food is being supplied from fishery sources, thus it is important to protect the proliferation of fishes [1]. Sheikh and Ahmed [2] recognized that the physiological assessments of hematological parameters are species specific and age dependent. Hematocrit, erythrocytes count and hemoglobin concentrations are the most readily resolved hematological parameters in fisheries [3]. Joshi and Tandon [13] found that with the increasing length and weight of fish *Clarius btrachus* there is a respective increase in the hematological parameters upto a certain age, after which the values become almost constant. Shrama and Joshi [5] examined haematological studies on a cyprinoid hillstream fish *Nemacheilus rupicola* and had found comparatively higher haematological values than most other fresh water teleost of the same size. Shripriya et al. [6] investigated the role of direct relationship between the body weight and length, haematocrit and haematological parameters of Indian fresh water Eel *Anguila bicolor* (McClelanand) and discussed that length and weight were increased with increasing age of the fish. This paper mainly described the haematological values of a fresh water catfish in relation to its length and weight.

## Materials And Methods

The Fresh water fishes were brought alive from the Pheet Bazar of BHEL Haridwar in laboratory in the plastic bags and transferred to the aquarium and maintained under laboratory condition for the further study. The fish were acclimation of at least 24- 40 hrs. To conduct various observations from blood samples fishes were handled, anaesthetized, blood drawn, and processed for making studies followed the method describe by Tandon and Joshi [8] and smear were prepared following the methods of Tandon and Joshi [8]. Blood was collected from the live fishes from the caudal peduncle described by Hicky [7]. Thin blood smear were air dried and fixed in absolute methanol slides were stained with Giemsa stain. The first 100 red and white blood corpuscles examined on each slide were counted and listed. The statistical analysis was done with the help of SPSS tool (version-20) and MS excel.

The study was carried to find out the length and weight related variations in haematological parameters of *H. fossilis* observed per month of the year (December 2014 to November 2016). The length weight (LW) based variation were assessed on the basis of length groups i.e. Group I (7-17 cm), Group II (17.1-20 cm) and Group III (20.1-30 cm) having 137 fishes in each length group. The average values of body weight were 16.6±0.49, 26.88±0.18 and 30.36±0.23 g for Group I, II and III, respectively. The average values of body length were 14.06±0.19, 18.87±0.65 and 21.31±0.12 cm for Group I, II and III, respectively (Fig. 1a). The mean changes in haematological parameters of *H. fossilis* in relation to different length groups during two years of study period are given in the present study.

## Results And Discussion

**Total erythrocyte count (TEC)** The significant ($p<0.05$) mean values of TEC were $1.96\pm0.03\times 10^6/mm^3$ for Group I, $2.37\pm0.02 \times 10^6/mm^3$for Group II and $2.47\pm0.02 \times 10^6/mm^3$for Group III (Table 1; Fig. 1b).

**Haemoglobin (Hb)** The values of Hb content showed significant ($p<0.05$) differences between groups. The values were 10.75±0.11, 12.07±0.07 and 12.39±0.10 g % for Group I, II and III, respectively (Table 1; Fig. 1b).

**Total leukocyte count (TLC)** TLCwas 18.60±0.12, 18.04±0.92 and $18.07\pm0.09 \times 10^3/mm^3$ for Group I, II and III respectively (Table 1; Fig. 1b).The values of TLC significant at ($p<0.05$) level.

**Packed cell volume (PCV)** The significant ($p<0.05$) mean values of PCV were 33.04±0.21 % for Group I, 32.10±0.16 % for Group II and 32.40±0.20 % for Group III (Table 1; Fig. 1b).

**Red blood cell indices** The variations in the values of red cell indices significantly ($p<0.05$) varied within different length groups. The mean values of MCH were 55.95±0.69 pg for Group I, 51.47±0.55 pg for Group II and 50.46±0.39 pg for Group III. The mean value of MCHC was 32.85±0.39, 37.80±0.22 and 38.44±0.36 (g %) for Group I, II and III, respectively, while the mean values of MCV were 175.28±3.10, 137.80±1.78 and 132.78±1.31 $\mu m^3$ for Group I, II and III, respectively (Table 1; Fig. 1d).

**Erythrocyte Sedimentation Rate (ESR)** The significant ($p<0.05$) variations in the mean values of ESR were 6.28±0.05, 6.81±0.10 and 7.23±0.12 mm for Group I, II and III respectively (Table 1; Fig. 1c).

**Blood Glucose (G)** The insignificant ($p>0.05$) mean values of G were 34.46±0.40, 34.84±0.52 and 34.11±0.63 mg/dl for Group I, II and III respectively (Table: 1; Fig. 1c).

**Correlation between haematological parameters and length weight of *H fossilis*** All three groups having different length showed significant positive correlations TEC(r=0.542); Hb (r=0.500); MCHC (r =0.485) and ESR (r =0.327), while negative correlation were noted for MCH(r =-0.313); MCV(r =-0.534) in relation to different length groups of the fishes. Body weight was positively correlated with length (r =0.947); TEC (r=0.630); Hb (r =0.584); MCHC (r =0.322), while negatively correlated with MCH(r =-0.388) and MCV(r =-0.678). Standard length showed positive correlation with TEC(r =0.598); Hb(r =0.752) and ESR(r =0.583), while negative correlation with MCH (r =-0.347) and MCV(r =-0.617). TEC was found positively correlated with Hb (r =0.752) and MCHC (r =0.583), while negatively correlated with MCH (r =-0.761) and MCV(r =-0.898). TLC established positive correlation with MCV(r =0.337). Hb was found positively correlated with MCHC(r =0.712), while negatively correlated with MCV(r =-0.610).PCV was found negatively correlated with MCHC (r =-0.320). MCH was positively correlated with MCV (r =0.783). MCHC showed negative correlation with MCV(r =-0.677), (Table 4).

# Differential leukocyte count (DLC)

**Lymphocyte (L) and Small Lymphocytes (SL)**The insignificant ($p>0.05$) mean values of L were 14.38±0.21 % for Group I, 14.77±0.19 % for Group II and 14.86±0.25 % for Group III. (Table 2; Fig. 1e). The mean values of SL were 14.77±0.20, 14.72±0.20 and 14.43±0.21% for Group I, II and III respectively (Table 2; Fig. 1e).

**Neutrophil (N)** The significant ($p<0.05$) mean values of N were 4.01±0.15, 4.35±0.14 and 4.58±0.16 % for Group I, II and III respectively (Table 2; Fig. 1e).

**Eosinophil (E)** The insignificant ($p>0.05$) mean values of E were 4.18±0.14 for Group I, 3.77±0.14 for Group II and 3.75±0.12 % for Group III (Table 2; Fig. 1e).

**Monocyte (M)** The mean insignificant ($p>0.05$) values of M were 12.50±0.15, 12.88±0.15 and 12.63±0.15 % for Group I, II and III respectively (Table 2; Fig. 1e).

**Thrombocyte (T)** The insignificant ($p>0.05$) mean values of T were 19.34±0.21, 18.94±0.19 and 19.10±0.17 % for Group I, II and III respectively (Table 2; Fig. 1e).

**Basophil (B)** The insignificant ($p>0.05$) mean values of B were 3.62±0.12 % for Group I, 3.65±0.12 % for Group II and 3.66±0.12 % for Group III (Table 2; Fig. 1f).

**Hemoblast (H) and Small Hemoblast (SH)** The insignificant ($p>0.05$) mean values of H cells were 13.02±0.17 % for Group I, 13.12±0.15 % for Group II and 12.66±0.15 % for Group III. The mean values of SH were 13.01±0.16, 12.84±0.14 and 12.88±0.17 % for Group I, II and III respectively (Table 2; Fig. 1f).

**Macrophage (MP)** The significant ($p<0.05$) mean percentages of MA were 1.15±0.10, 0.93±0.09 and 1.36±0.11 % for Group I, II and III respectively (Table 2; Fig. 1f).

**Correlation between DLC count and length weight of *H fossilis*** Among different leukocytes only N (r=0.125) showed positive correlation, while other parameters did not show any significant correlation with respect to different length groups (Table 5).

# Differential Erythrocyte Count (DEC)

**Erythrocyte (ER)** The insignificant (p>0.05) percentages of ER were 86.63±0.24, 86.77±0.21 and 86.04±0.25 % for Group I, II and III respectively (Table 3; Fig. 1g).

**Erythroblast (EB)** The insignificant (p>0.05) mean values of erythroblast cells were 10.31±0.13 % for Group I, 10.26±0.13 % for Group II and 10.26±0.14 %for Group III (Table 3; Fig.1g).

**Macrocyte (MA)** The insignificant (p>0.05) mean values of macrocytes were 0.78±±0.07, 0.57±0.06 and 0.90±0.07 % for Group I, II and III respectively (Table 3; Fig. 1h).

**Microcyte (MI)** The insignificant (p>0.05) mean values of microcytes were 1.18±0.09, 1.22±0.10 and 1.48±0.13 % for Group I, II and III respectively (Table 3; Fig. 1h).

**Bizarre Form (BZ)** The insignificant (p>0.05) mean values of bizarre forms were 1.13±0.10 % for Group I, 1.16±0.10 % for Group II and 1.36±0.11 %for Group III (Table 3; Fig. 1h).

**Correlation between DEC count and length weight of *H fossilis*** Only MI (r =0.103) showed significant positive correlation with different length groups while all other parameters did not established any correlation in respect of length. ER was found negatively correlated with ER (r =-0.567); MA (r =-0.375); microcytes (r =-0.605) and BZ (r =-0.592). Moreover microcytes showed positive correlation with BZ (r =0.305) (Table 6).

The present study corroborate with the findings of Purnarn and Freel [11] who suggested the cells of active, tropical fishes were more densely packed with Hb due to an increased number of red blood cells than less active species. Ahmedet al. [9] notedRBC and Hb contents tend to increase with length and age of the fishes. Jawad et al. [14] was found the correlation between PCV and physiological parameters of the Indian shad, *Tenulosa ilisha* (Family: Clupeidae) and concluded that Ht values illustrated a quadratic relationship of fish size (lenght). Joshi [12] reported that in *Wallago attu,*TEC,TLC and Hb increased up to a certain length and thengradually fell. Goel and Gupta [15] reported MCV, MCH and MCHC values for *Heteropneustes fossilis.* The same aspect of red cell indices was investigated by Prasad et al.[16] in some fresh water fishes. Study on *Clarias batrachus*of different length groups demonstrate that blood values increase as the fish grow, but later than attaining a maximum level, most of the values tend to nearly stabilize, while a few tend to fall Joshi and Tandon [17]. Das [10] determined that the number of blood cells and Hb concentration tends to increase with length and age, showing positive correlation. Joshi [12] reported some blood values of *Wallago attu*, the fresh water shark of varying length groups and showed that blood values increases, as the fish increasing in length and weight. But after attaining a maximal level, most of the values become nearly constant while few show a declining trend. On contrary with the present findings, Khanam and Latifa [18] reported DLC of *Heteropneustes fossilis*increased with increase of length and reach at peak (146 $\times 10^3 mm^3$) in 16 to 18 cm length group and lowest count (142 $\times 10^3 mm^3$) was observed in 10 -12 cm length group. Tavares-Dias [19] assessed weight-length and blood parameters of farmed *Cichlan temensis* Humboldt, (Cichlidae) in central Amazon and concluded that RBC, total T and DLC, PCV, Hb, MCV, MCHC, L, M and N had shown intra-specific variation.

## Conclusion

In the present study, the haematological values were increased with both body weight and length, but to a certain weight/length group after which a slight fall in their value was observed. All parameters except RBC/WBC ratio, PCV, ESR and MCV decreased with increasing length of the fish. Yousefzadeh and Khara [20] investigated the haematological and biochemical parameters of *Capoeta capoeta* with respect to age had not shown any significant changes. Ejraei et al. [21] assessed the changes in hematological of *Ctenopharyngodon idella* (Val.) with regard to age, sex, and hormonal treatment in sixty specimens of *C. idella.* Numerous factors were reported to influence the haemoglobin contents of the blood of fishes due to length and weight by Radzinskaya [22] in loach (*Misqurnus fossilis*). It may be conclude that fish becomes less active and its metabolic rate is affected as it grows older.

## Declarations

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# Significance' statement

Blood is considered as an essential component of life of vertebrates and invertebrates. The hematocrit, erythrocytes count and hemoglobin concentrations are the most momentous hematological parameters within the field and hatchery conditions. In this concern this study plays a significant role in the field of fisheries and aquaculture.

# Conflict of interest

The authors declare that they have no competing interests

# Ethical approval

This article does not contain any studies with human participants or animals performed by any of the authors.

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# Tables

**Table 1**

**Haematological Parameters of different length group of *H. fossilis*.**

| Variables | Group I (7cm-17cm) | Group II (17cm-20cm) | Group III (20cm-30cm) | P-value |
|---|---|---|---|---|
| TEC × $10^6$/$mm^3$) | 1.96±0.03 | 2.37±0.02 | 2.47±0.02 | 0.000* |
| TLC × $10^3$/$mm^3$ | 18.60±0.12 | 18.04±0.92 | 18.07±0.09 | 0.000* |
| Hb (g%) | 10.75±0.11 | 12.07±0.07 | 12.39±0.10 | 0.000* |
| PCV (%) | 33.04±0.21 | 32.10±0.16 | 32.40±0.20 | 0.002* |
| MCH (pg) | 55.95±0.69 | 51.47±0.55 | 50.46±0.39 | 0.000* |
| MCHC (g%) | 32.85±0.39 | 37.80±0.22 | 38.44±0.36 | 0.000* |
| MCV ($\mu m^{3)}$ | 175.28±3.10 | 137.80±1.78 | 132.78±1.31 | 0.000* |
| ESR (mm) | 6.28±0.05 | 6.81±0.10 | 7.23±0.12 | 0.000* |
| Glucose (mg/dl) | 34.46±0.40 | 34.84±0.52 | 34.11±0.63 | 0.658 |
| **(All the values are expressed as mean ± SE of 137 observations. The mean difference is significant at the 0.05 level.*)** | | | | |

**Table 2**

**Differential Leukocytes Count (DLC) of different length group of *H. fossilis.***

| Variables | Group I (7cm-17cm) | Group II (17cm20cm) | Group III (20cm-30cm) | P-value |
|---|---|---|---|---|
| Large Lymphocytes (%) | 14.38±0.21 | 14.77±0.19 | 14.86±0.25 | 0.218 |
| Small Lymphocytes (%) | 14.77±0.20 | 14.72±0.20 | 14.43±0.21 | 0.394 |
| Neutrophils (%) | 4.01±0.15 | 4.35±0.14 | 4.58±0.16 | 0.036 |
| Eosinophils (%) | 4.18±0.14 | 3.77±0.14 | 3.75±0.12 | 0.135 |
| Monocytes (%) | 12.50±0.15 | 12.88±0.15 | 12.63±0.15 | 0.419 |
| Thrombocytes (%) | 19.34±0.21 | 18.94±0.19 | 19.10±0.17 | 0.675 |
| Basophils (%) | 3.62±0.12 | 3.65±0.12 | 3.66±0.12 | 0.952 |
| Hemoblast (%) | 13.02±0.17 | 13.12±0.15 | 12.66±0.15 | 0.109 |
| Small Hemoblast (%) | 13.01±0.16 | 12.84±0.14 | 12.88±0.17 | 0.344 |
| Macrophage (%) | 1.15±0.10 | 0.93±0.09 | 1.36±0.11 | 0.010* |
| **(All the values are expressed as mean ± SE of 137 observations. The mean difference is significant at the 0.05 level.*)** | | | | |

**Table 3**

**Differential Erythrocyte Count (DEC) of different length group of *H. fossilis***

| Variables | Group I (7cm-17cm) | Group II (17cm20cm) | Group III (20cm-30cm) | P-value |
|---|---|---|---|---|
| Erythrocytes (%) | 86.63±0.24 | 86.77±0.21 | 86.04±0.25 | 0.070 |
| Erythroblast (%) | 10.31±0.13 | 10.26±0.13 | 10.26±0.14 | 0.997 |
| Macrocytes (%) | 0.78±±0.07 | 0.57±0.06 | 0.90±0.07 | 0.003* |
| Microcytes (%) | 1.18±0.09 | 1.22±0.10 | 1.48±0.13 | 0.100 |
| Bizarre Forms (%) | 1.13±0.10 | 1.16±0.10 | 1.36±0.11 | 0.279 |

(All the values are expressed as mean ± SE of 137 observations. The mean difference is significant at the 0.05 level.*)

**Table 4**

**Correlations Matrix of Length-weight related variation in the haematological parameters of *H. fossilis***

| L.Wt. | Group | BW | BL | TEC | TLC | Hb | PCV | MCH | MCHC | MCV | ESR | G |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Group | 1 | | | | | | | | | | | |
| BW | 0.802** | 1 | | | | | | | | | | |
| BL | 0.870** | 0.947** | 1 | | | | | | | | | |
| TEC | 0.542** | 0.630** | 0.598** | 1 | | | | | | | | |
| TLC | -0.169 | -0.294** | -0.251** | -0.249** | 1 | | | | | | | |
| Hb | 0.500 | 0.584** | 0.569** | 0.752** | -0.108* | 1 | | | | | | |
| PCV | -0.118 | -0.204** | -0.145** | 0.123* | 0.237** | 0.281** | 1 | | | | | |
| MCH | 0.313 | -0.388** | -0.347** | -0.761** | 0.291** | -0.193** | 0.123* | 1 | | | | |
| MCHC | 0.485** | 0.607** | 0.547** | 0.583** | -0.224** | 0.712** | -0.320** | -0.238** | 1 | | | |
| MCV | -0.534** | -0.678** | -0.617** | -0.898** | 0.337** | -0.610** | 0.259** | 0.783** | -0.677** | 1 | | |
| ESR | 0.327** | 0.322** | 0.343** | 0.150** | -0.076 | 0.038 | -0.186** | -0.176** | 0.142** | -0.194** | 1 | |
| G | -0.014 | -0.069 | -0.056 | 0.159** | 0.068 | 0.130** | 0.162** | -0.124* | 0.009 | -0.083 | -0.296** | 1.00 |

**. Correlation is significant at the 0.01 level (2-tailed).
*. Correlation is significant at the 0.05 level (2-tailed).

**Table 5**

**Correlations Matrix of Length-weight related variation in the DLC Count of *H.fossilis***

| | Group | LL | SL | N | E | M | T | B | H | SH | MP |
|---|---|---|---|---|---|---|---|---|---|---|---|
| Group | 1 | | | | | | | | | | |
| LL | 0.081 | 1 | | | | | | | | | |
| SL | -0.066 | -0.239** | 1 | | | | | | | | |
| N | 0.125* | -0.073 | -0.123* | 1 | | | | | | | |
| E | -0.093 | -0.193** | -0.199** | -0.202** | 1 | | | | | | |
| M | 0.012 | -0.217** | -0.137** | 0.000 | -0.037 | 1 | | | | | |
| T | -0.020 | -0.206** | -0.036 | -0.306** | -0.002 | -0.221** | 1 | | | | |
| B | 0.002 | -0.138** | -0.177** | 0.054 | 0.053 | -0.007 | -0.219** | 1 | | | |
| H | -0.072 | -0.012 | -0.200** | -0.242** | -0.023 | -0.006 | -0.046 | -0.105* | 1 | | |
| SH | -0.047 | -0.175** | -0.048 | 0.050 | -0.151** | -0.099* | -0.151** | -0.036 | -0.213** | 1 | |
| MP | 0.072 | -0.040 | -0.182** | -0.023 | 0.061 | -0.065 | -0.014 | -0.062 | -0.137** | -0.083 | 1.00 |

**. Correlation is significant at the 0.01 level (2-tailed).
*. Correlation is significant at the 0.05 level (2-tailed).

Table 6

Correlations Matrix of Length-weight related variation in the EC Count of *H.fossilis*

| | Group | ER | EB | MA | MI | BZ |
|---|---|---|---|---|---|---|
| Group | 1 | | | | | |
| ER | -0.096 | 1 | | | | |
| EB | -0.002 | -0.567** | 1 | | | |
| MA | 0.063 | -0.375** | 0.081 | 1 | | |
| MI | 0.103* | -0.605** | 0.033 | 0.082 | 1 | |
| BZ | 0.074 | -0.592** | 0.027 | 0.058 | 0.305** | 1.00 |

**. Correlation is significant at the 0.01 level (2-tailed).
*. Correlation is significant at the 0.05 level (2-tailed).

## Figures

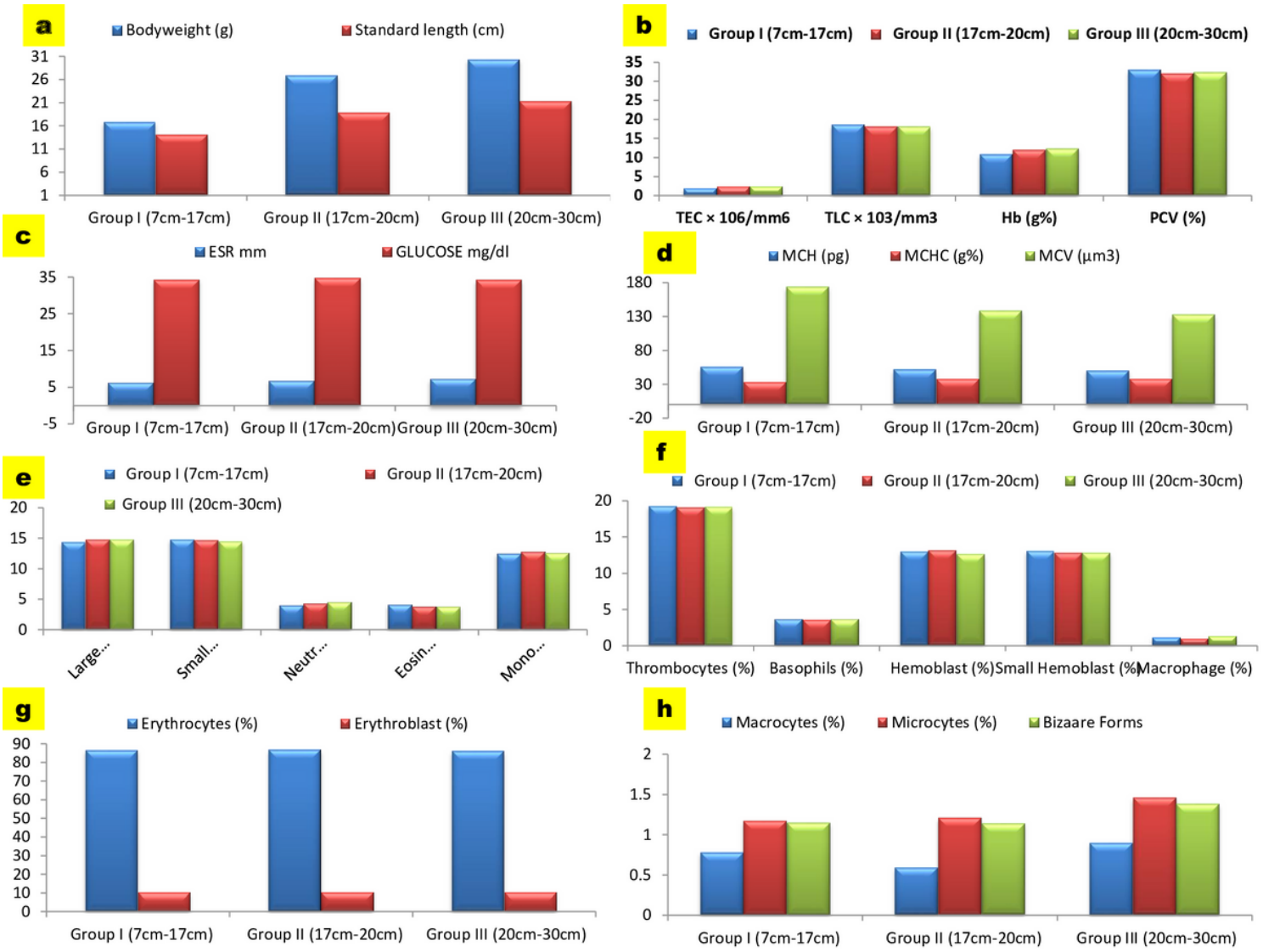


**Figure 1**

a: Values between Bodyweight and Standard length in different groups of H fossilis during 2014-16 b: Values of TEC, TLC, Hb and PCV in different groups of H fossilis during 2014-16 c: Values of ESR and Glucose in different groups of H fossilis during 2014-16 d: Values of red cell indices (MCH, MCHC and MCV) in different groups of H fossilis during 2014-16 e: Variation in DLC (LL, SL, N, E and M) in different groups of H fossilis during 2014-16 f: Variation in DLC (T, B, H, SH and MP) in different groups of H fossilis during 2014-16 g: Variation between Erythrocytes and Erythroblast in different groups of H fossilis during 2014-16 h: Values of Macrocytes, Microcytes and Bizarre forms in different groups of H fossilis during 2014-16