

Effects of Delayed First Feeding on the Development of the Digestive Tract and Skeletal Muscles of Nile Tilapia, *Oreochromis niloticus* L.

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INTRODUCTION

- During the last half century, fish farmers throughout the tropical and semi-tropical world have begun farming tilapia.
- Today, more than 90% of all commercially farmed tilapia are Nile tilapia, *Oreochromis niloticus* L. (Popma and Masser, 1999).

INTRODUCTION

- **Second to milkfish, tilapia now ranks as the most important cultured fish in the Philippines (Guerrero, 1994).**
- **Tilapia farming is not only associated with its potential as a source of food, but also as an attractive source of investment (Bimbao and Smith, 1988).**

INTRODUCTION

In a **NATURAL ENVIRONMENT** where predators abound, female tilapias delay the release of their fry for protection.

In **COMMERCIAL HATCHERIES**, delay in first feeding occurs when farmers fail to notice that the fry had already totally absorbed their yolk and are ready to receive exogenous food.



OBJECTIVES OF THE STUDY

- To trace the development of the digestive tract and skeletal muscles of Nile tilapia, *O. niloticus* L. from 0-150 days post-feeding when first feeding is delayed

SIGNIFICANCE OF THE STUDY

- **essential to the understanding of whether the development of certain organs in Nile tilapia larvae is most vulnerable during starvation**
- **allows tilapia farmers to decide the cost effectiveness of rearing previously starved fry up to their marketable size**

MATERIALS AND METHODS

Egg incubation and rearing of larvae – 1 month



Preliminary testing – 10 days



Starvation and feeding experiments – 5 months



Histology



Light Microscopy

Electron Microscopy

(SEM and TEM)



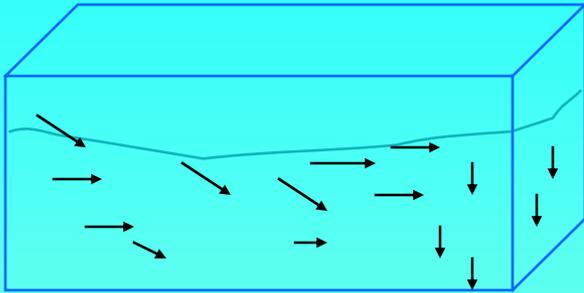
Statistical analysis

BREEDING AND REARING OF FRY OR LARVAE

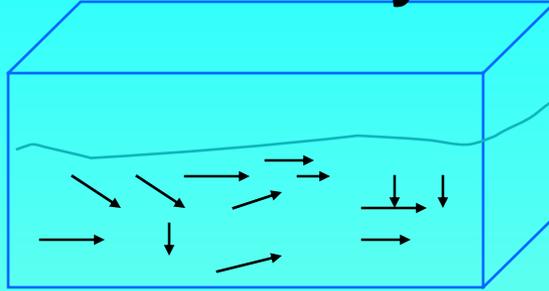


PRE-TRIAL EXPERIMENT

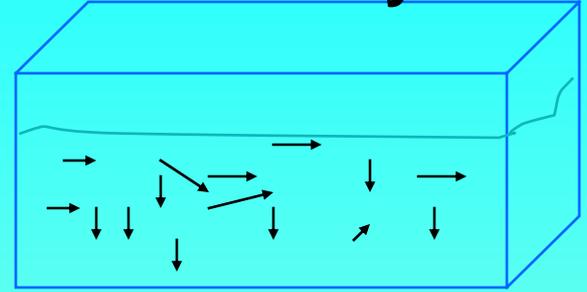
200 fry



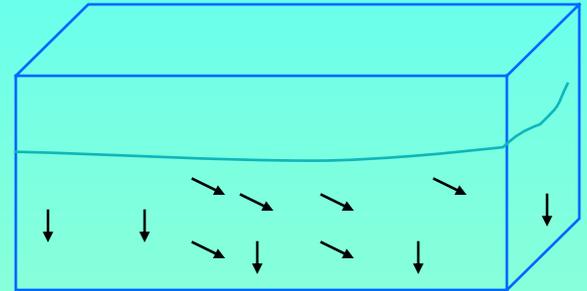
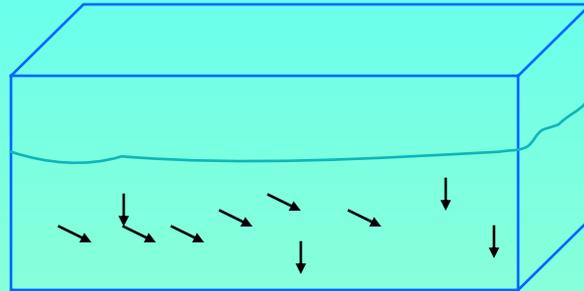
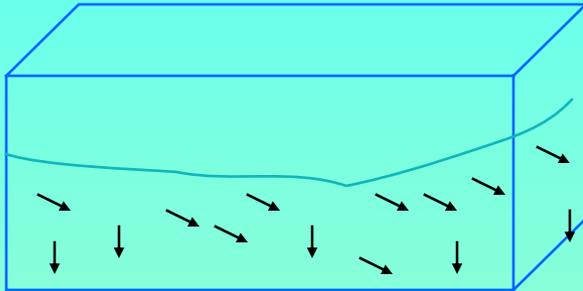
200 fry



200 fry



1st batch – one parental source



2nd batch – one parental source



Starved until total mortality was observed

Actual Experimentation

ACTUAL EXPERIMENTATION



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SAMPLING

	T1 (control)	T2-starved 2 days	T3-starved 4 days	T4-starved 6 days	T5-starved 8 days
Day 0	5	5	5	5	5
Day 2	5	5	5	5	5
Day 4		5 samples – histology			5
Day 6		3 samples– SEM			5
Day 8		3 samples– TEM			5
Day 20					5
Day 30		3 samples - histochemistry			5
Day 60	5	5	5	5	5
Day 90	5	5	5	5	5
Day 120	5	5	5	5	5
Day 150	14	14	14	14	14

LIGHT MICROSCOPY

PURPOSE: to trace the development of the skeletal muscles and organs of the digestive tract from day 0 (day of hatching) to day 150 (adult marketable size).



EMPHASIS

TISSUE

Stomach – height of mucosal fold

Height of muscularis layer (μm)

Anterior and posterior intestine - height of mucosal fold

Height of muscularis layer (μm)

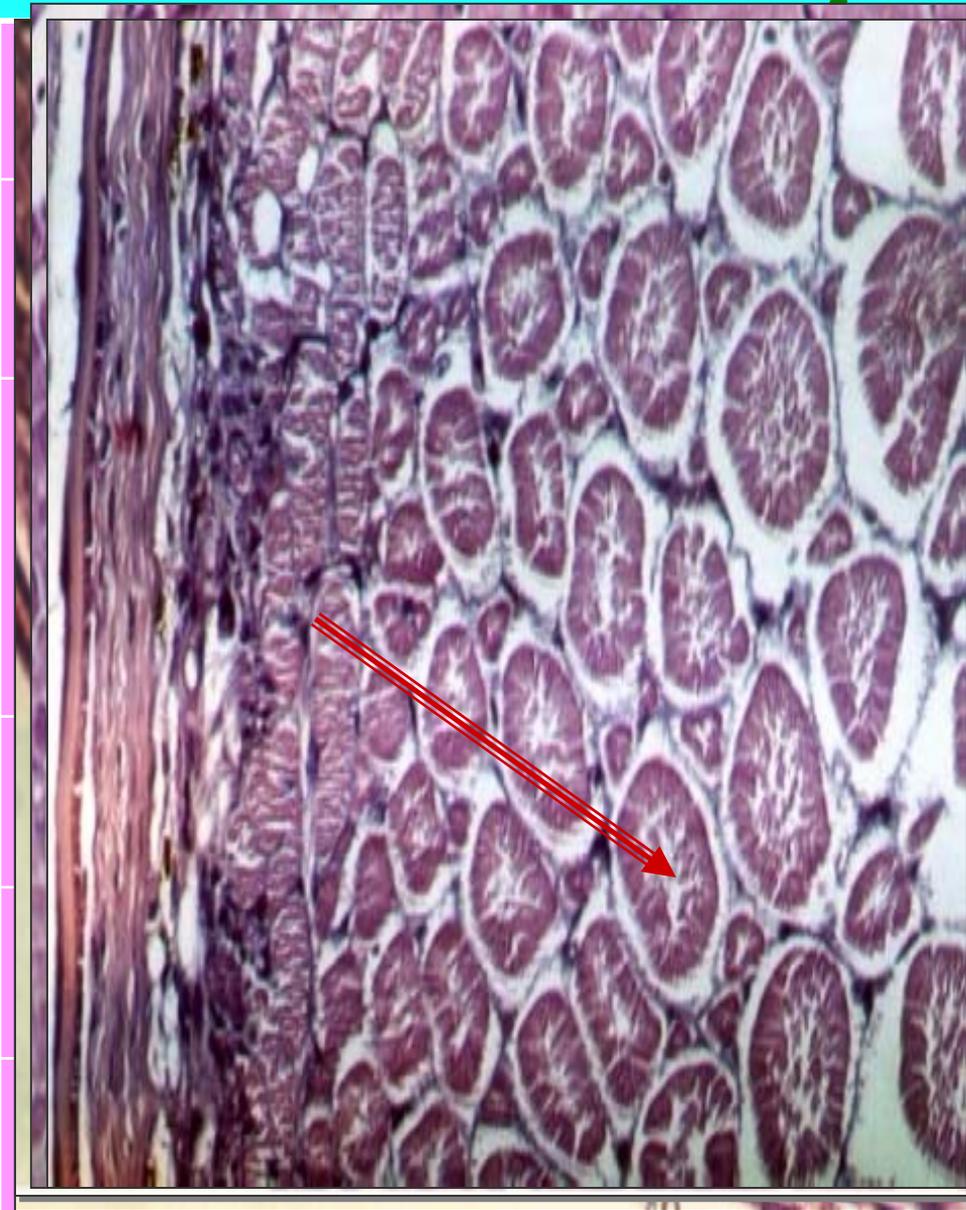
Number of goblet cells

Liver – diameter of hepatic portal vein (μm)

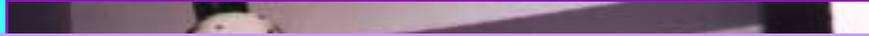
Skeletal Muscles – diameter of muscle fibers (μm)

Esophagus

Pancreas



TRANSMISSION ELECTRON MICROSCOPY



PURPOSE: to trace the development of the cells in the anterior intestine of the adult marketable fish (day 150).



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SCANNING ELECTRON

PURPOSE: to examine the surface features of the anterior intestine of the adult marketable fish (day 150).



STATISTICAL ANALYSIS

- Data were presented as means \pm standard error.
- Means were tested using Analysis of Variance (ANOVA) followed by Duncan's Multiple Range Test (DMRT).
- Analyses were performed using Statistical Analysis Software (SAS).

RESULTS AND DISCUSSION

RESULTS

Figure 1. Comparison of the effect of delayed first feeding on the weight (grams) of 0-150 dph unstarved and starved *O. niloticus*.

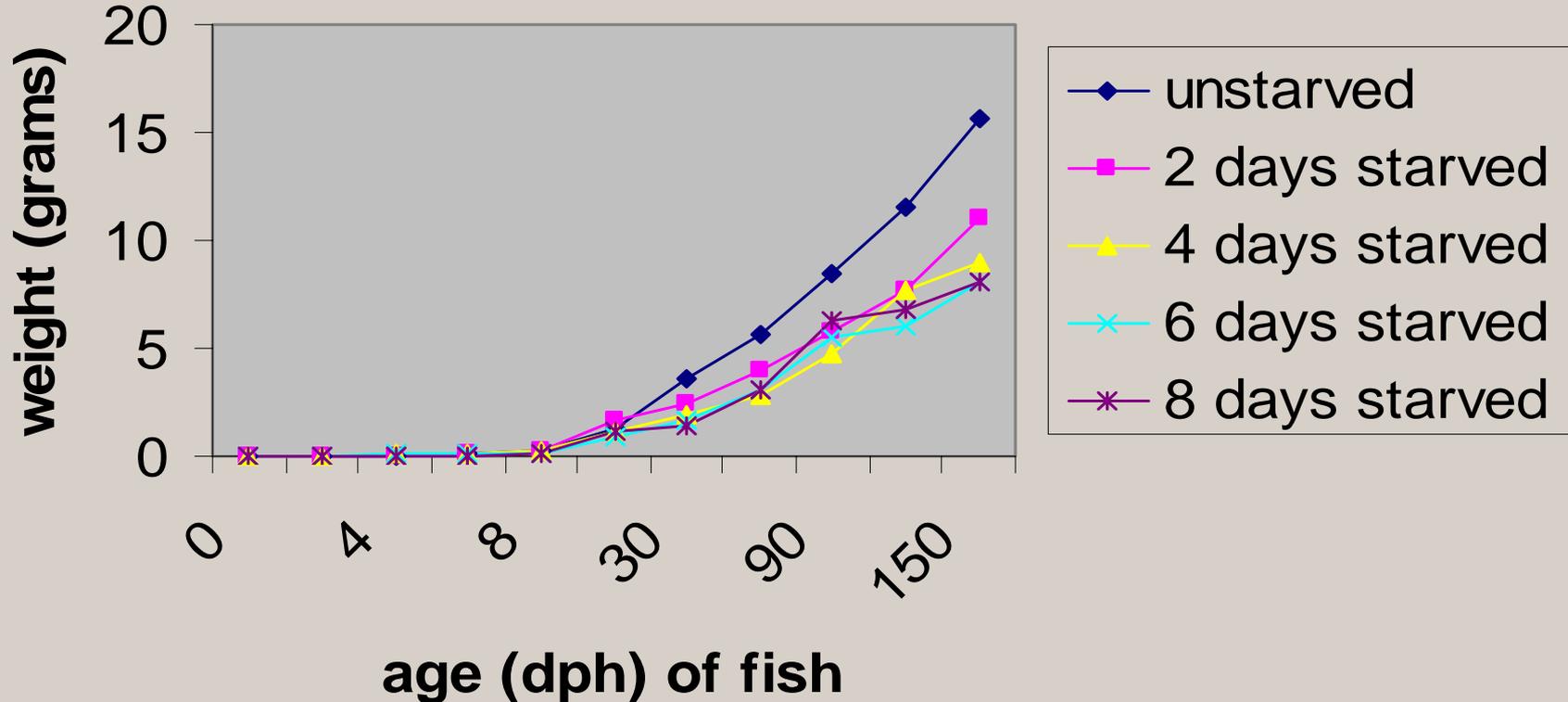


Figure 2. Comparison of the effect of delayed first feeding on the length (centimeters) of 0-150 dph unstarved and starved *O. niloticus* samples.

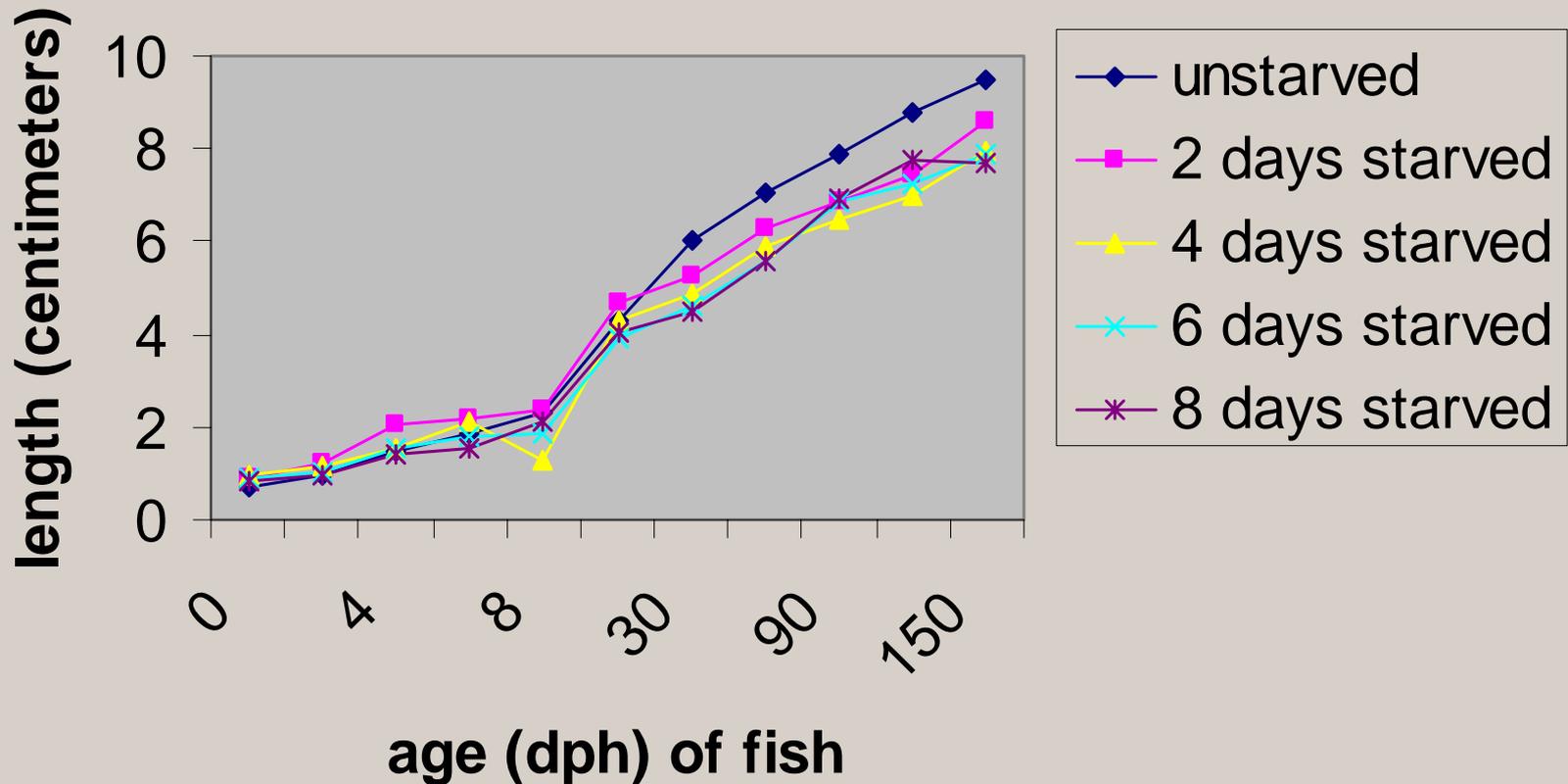
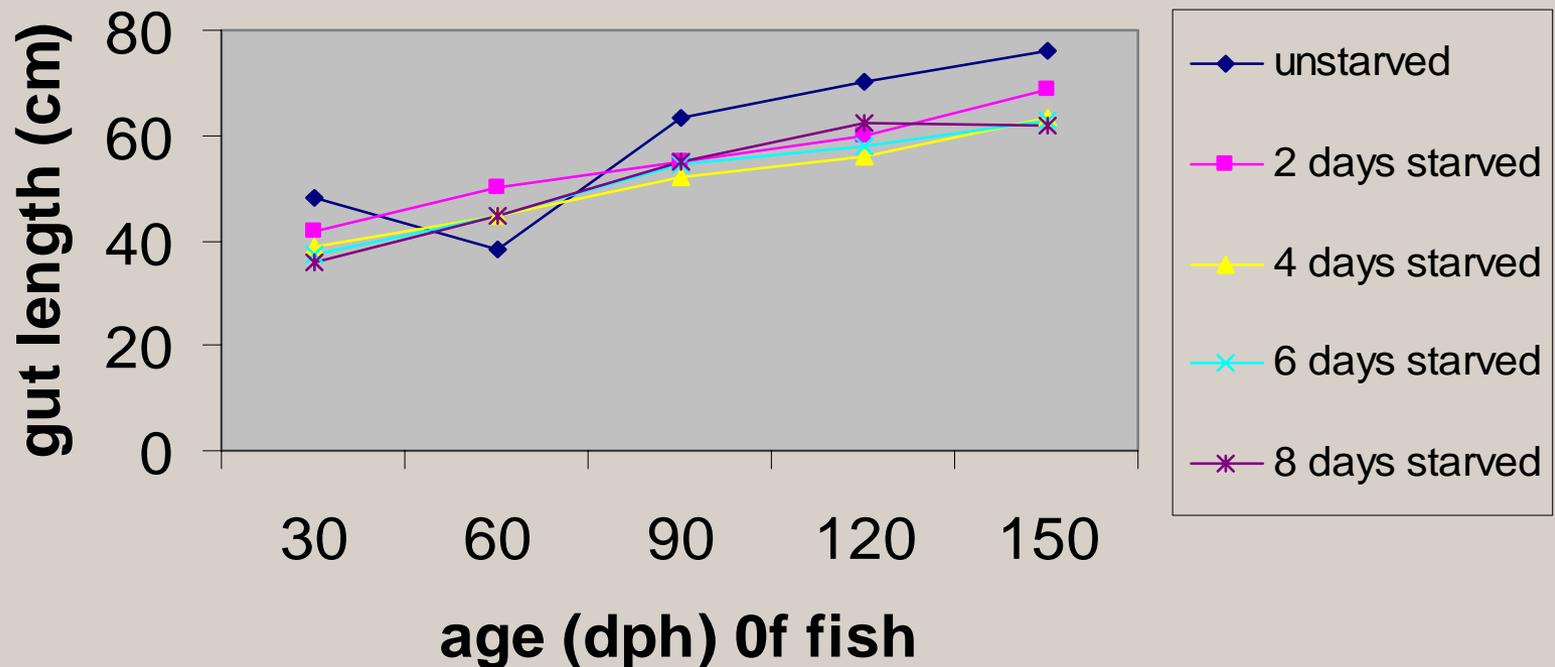


Figure 3. Comparison of the effect of delayed first feeding on the gut length (centimeters) of 30-150 dph unstarved and starved *O. niloticus* samples.



STARVATION

- ❖ To deprive the Nile tilapia fry of exogenous food, notwithstanding their readiness, is a form of STARVATION, which delayed their growth and development for lack of basic nutrients.
- ❖ This deprivation is a type of STRESS that organisms often encounter in nature (www.biol.unt.edu, 2004).
- ❖ Many pathological changes occur in a starved animal: decrease in size and weight; atrophy in the musculature; digestive tract becomes empty.

METHODS USED TO STUDY STARVATION IN FISH

- ◆ **Morphological, histological or biochemical**
- ◆ **Morphological measurements have the advantages of ease and practicality of application as an indicator of starvation (Dou *et al.*, 2002).**
- ◆ **For this study, however, it was helpful to also use histological examination of tissues during starvation to validate the practicality and applicability of the morphological indicators used.**

Fig. 4. Cross-section of the anterior abdominal cavity of 6 days post-hatch (dph) Nile Tilapia, 100X.

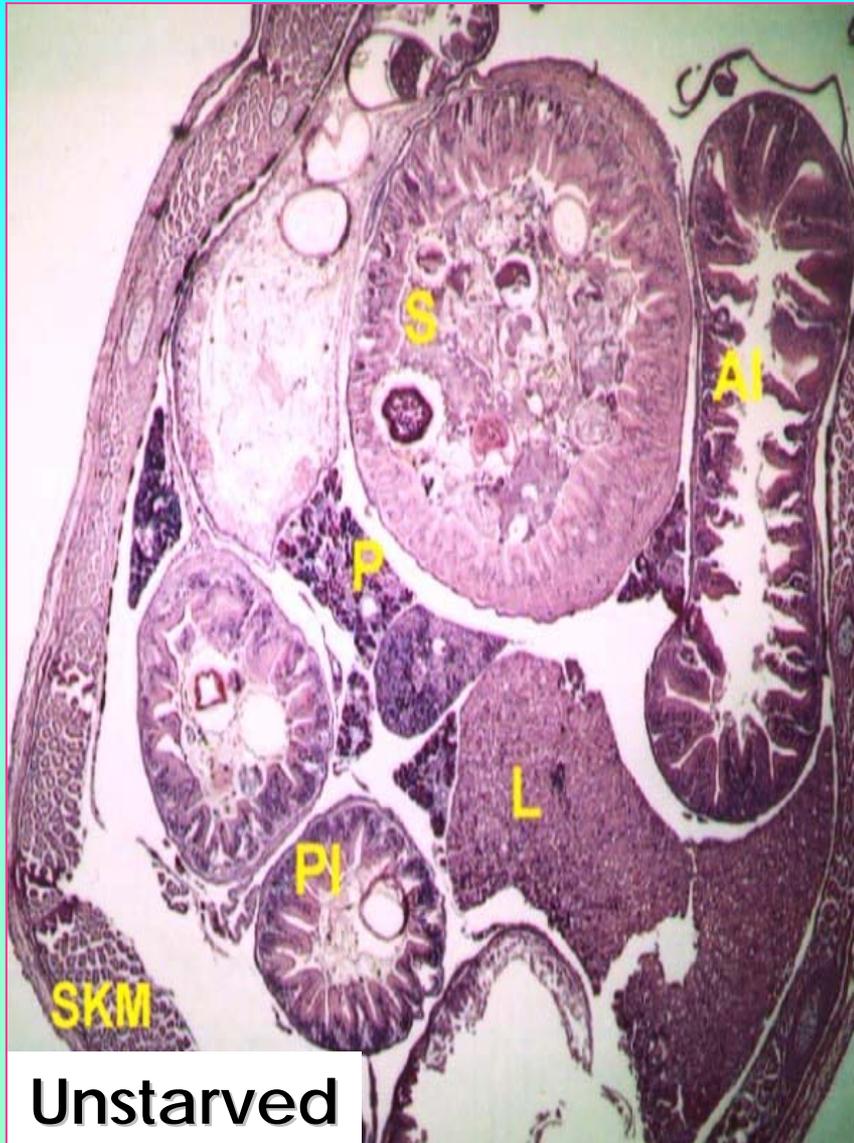


Fig. 5. Cross-section of the esophagus, stomach, and intestine of an 8 dph starved Nile Tilapia, 100X.



Fig. 6. Cross-section of the anterior intestine of 6 dph Nile Tilapia, X1000.



Fig. 7. Cross-section of the anterior intestine of the 120 dph Nile tilapia, X1000.

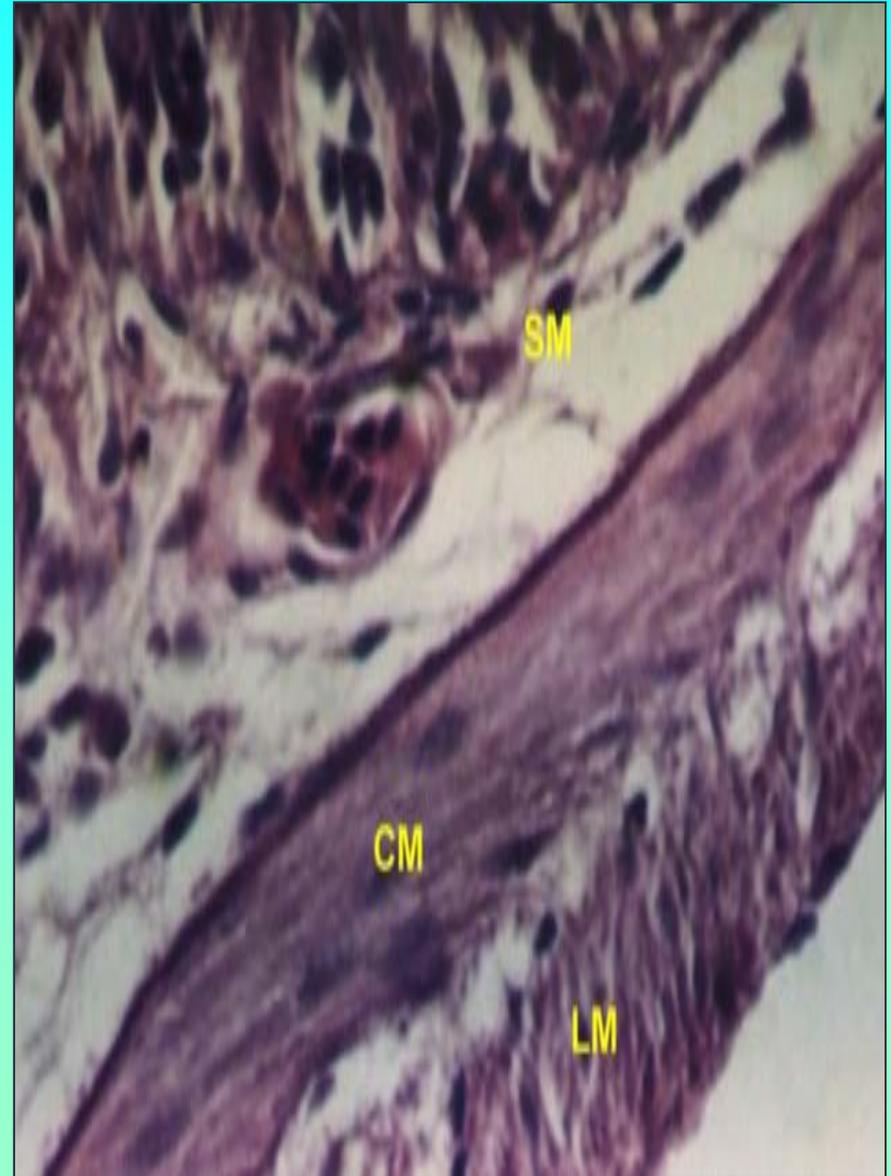
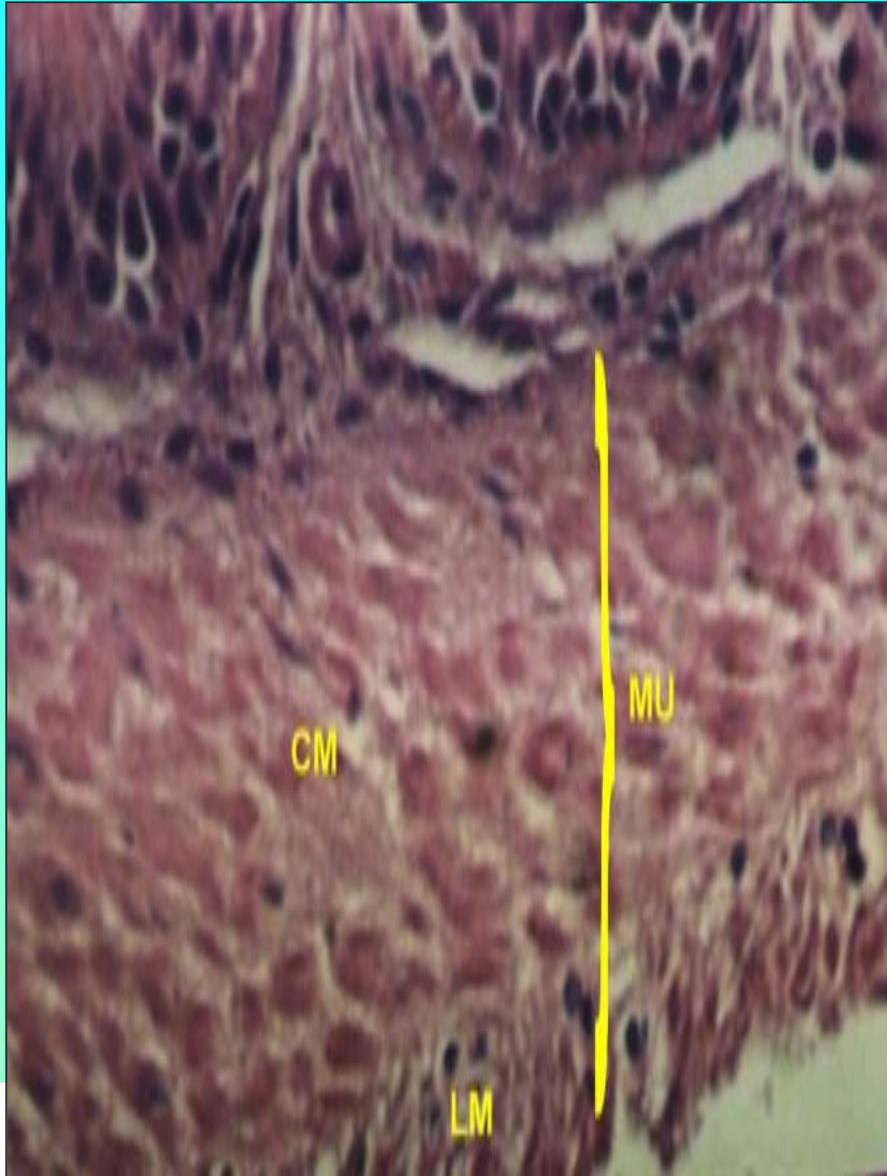
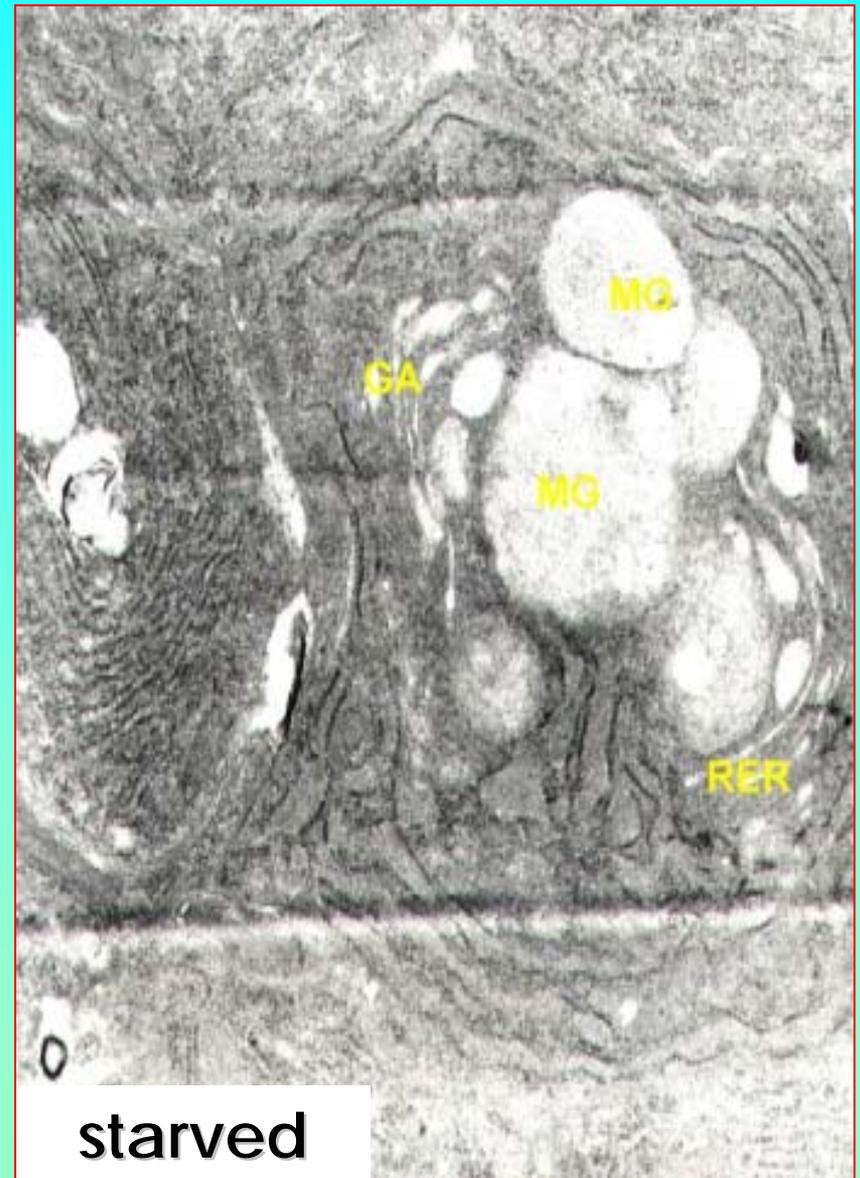
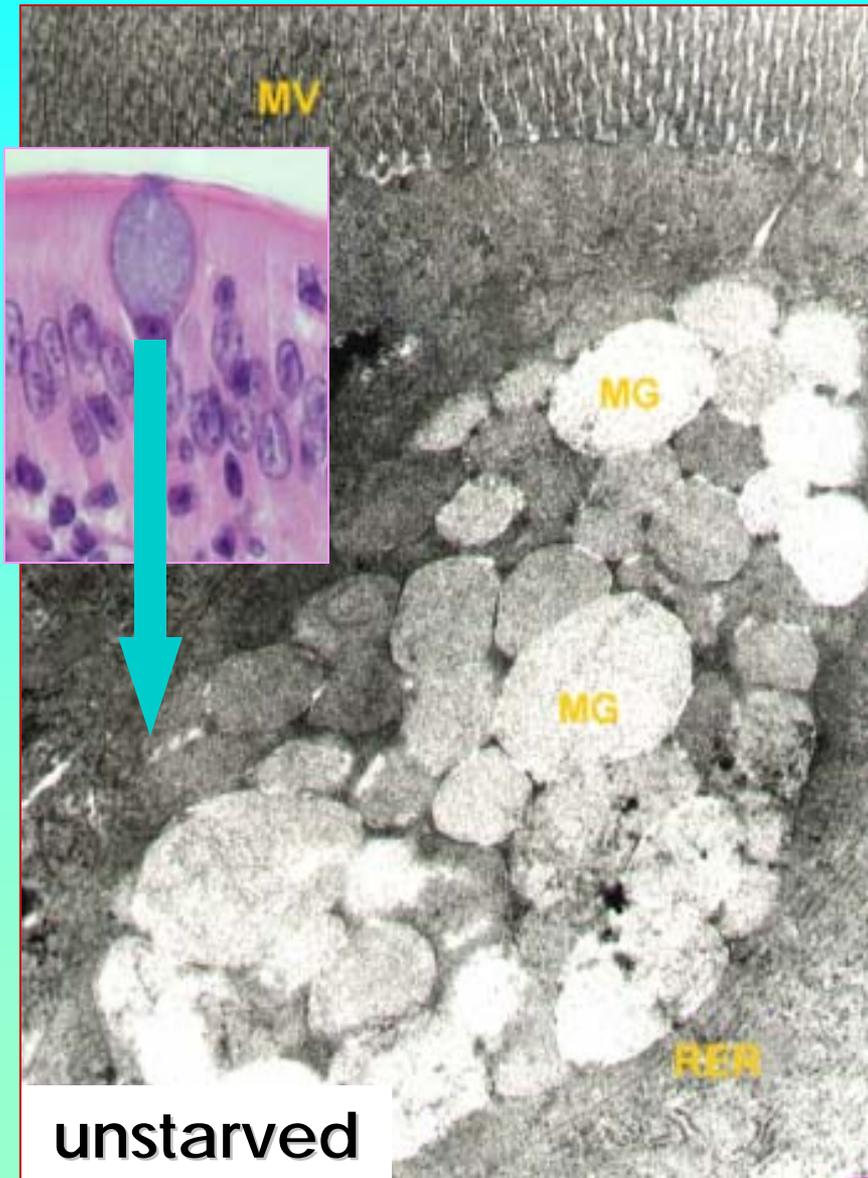
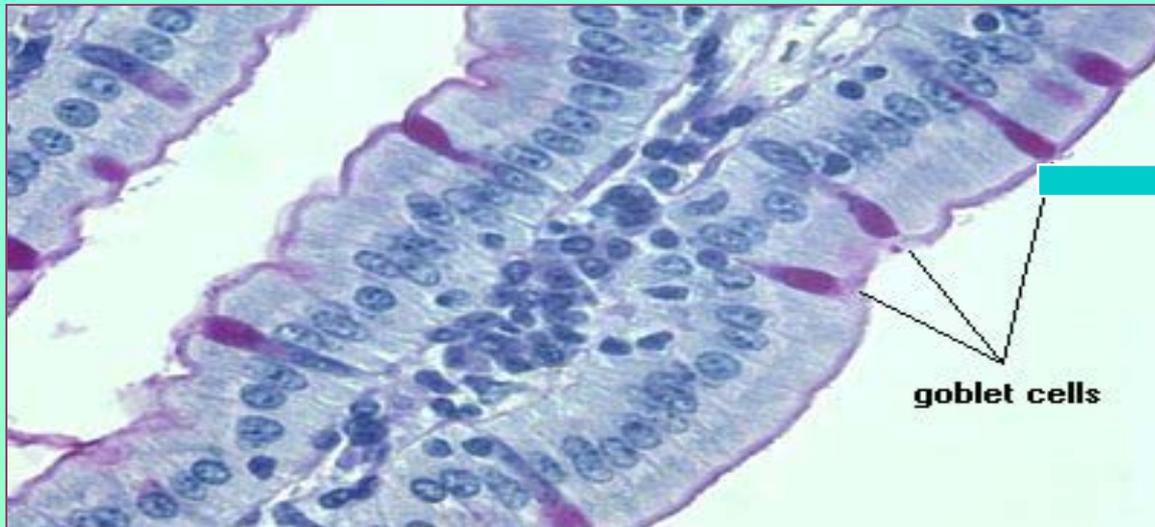


Fig. 8. Transmission electron micrograph of the anterior intestine of the 150 dph Nile Tilapia, X21,600.



GOBLET CELLS

- ◆ The talent of goblet cells is to secrete mucus.
- ◆ **Functions of Mucus Cells:**
 - protection against shear stress and chemical damage
 - trapping and elimination of particulate matter and microorganisms.



GOBLET CELLS

- ◆ The abundant mucous granules seen in the unstarved fish may not be due to a higher activity in terms of functions mentioned; instead, it may be attributed to the abundance of raw materials present for its formation.

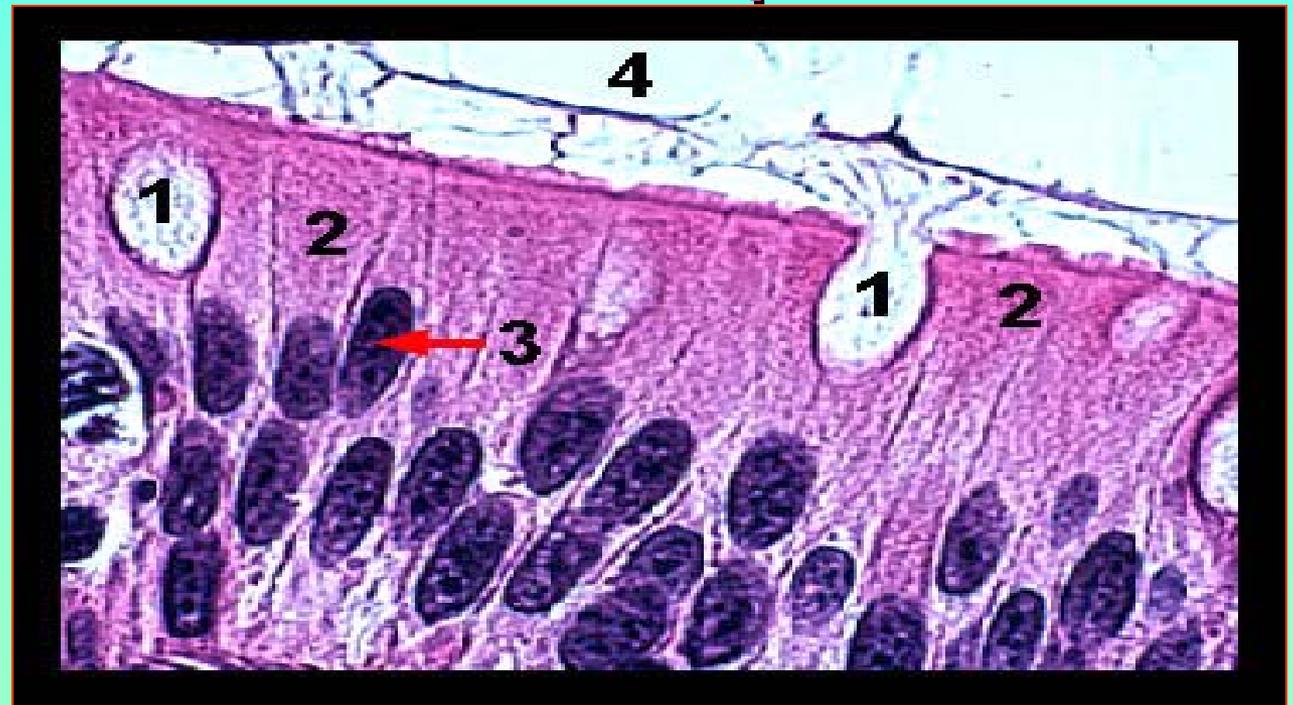


Fig. 9. Scanning electron micrographs of the anterior intestine of 150 dph Nile Tilapia, X3000.

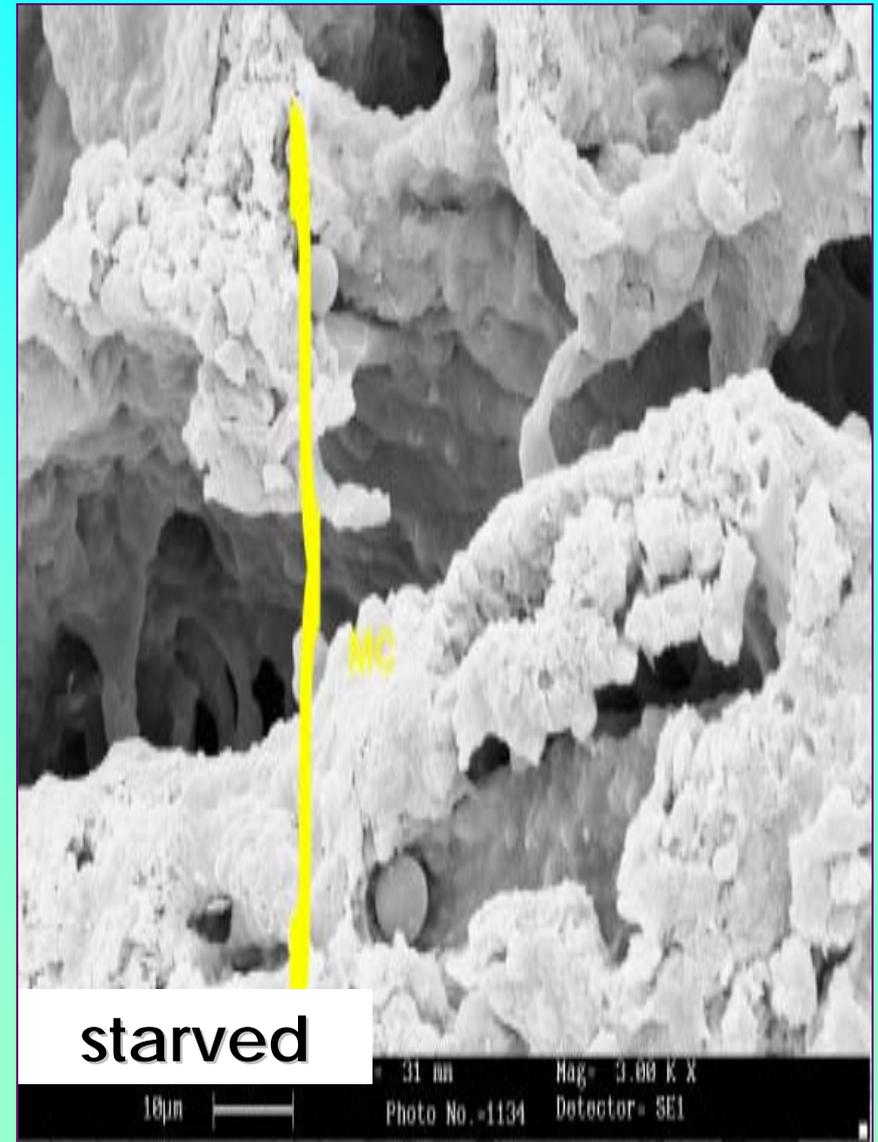
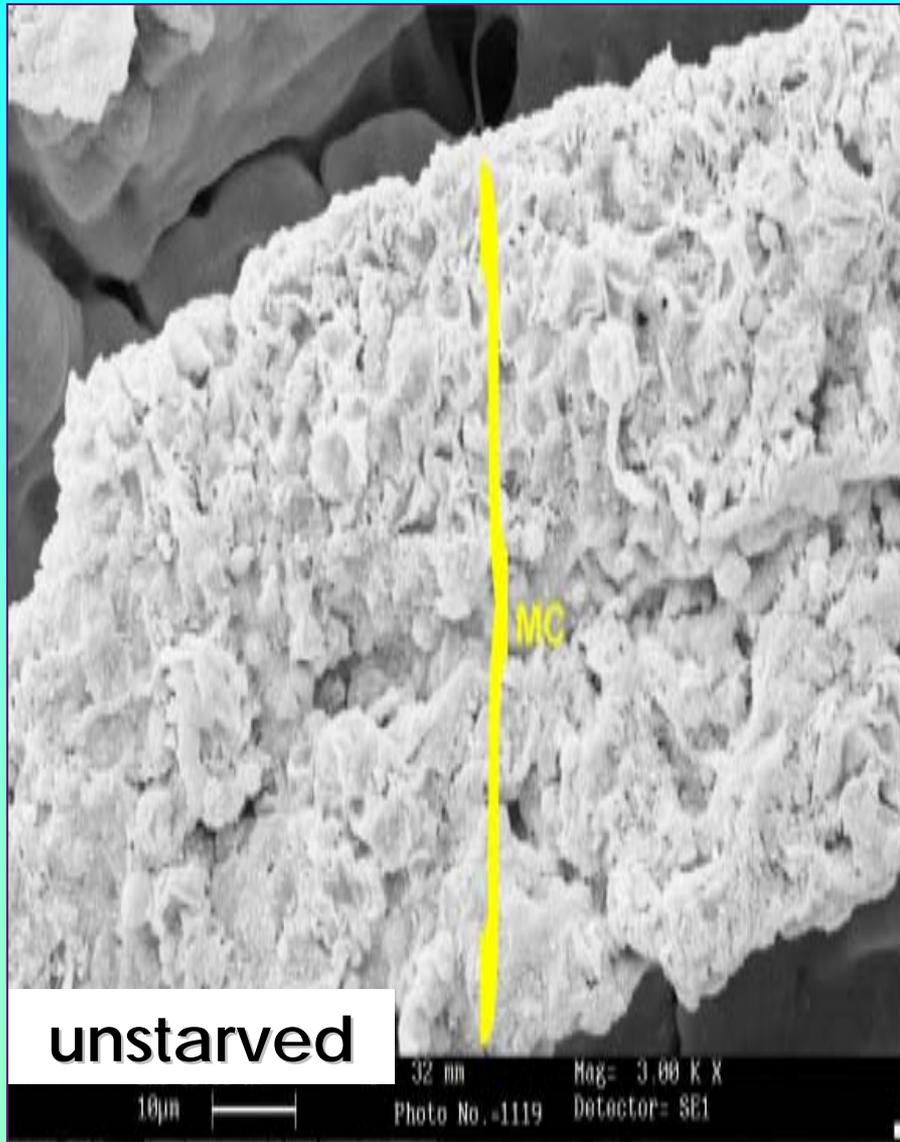


Fig. 10. Cross-section of the posterior intestine of 6 dph Nile Tilapia, X1000.

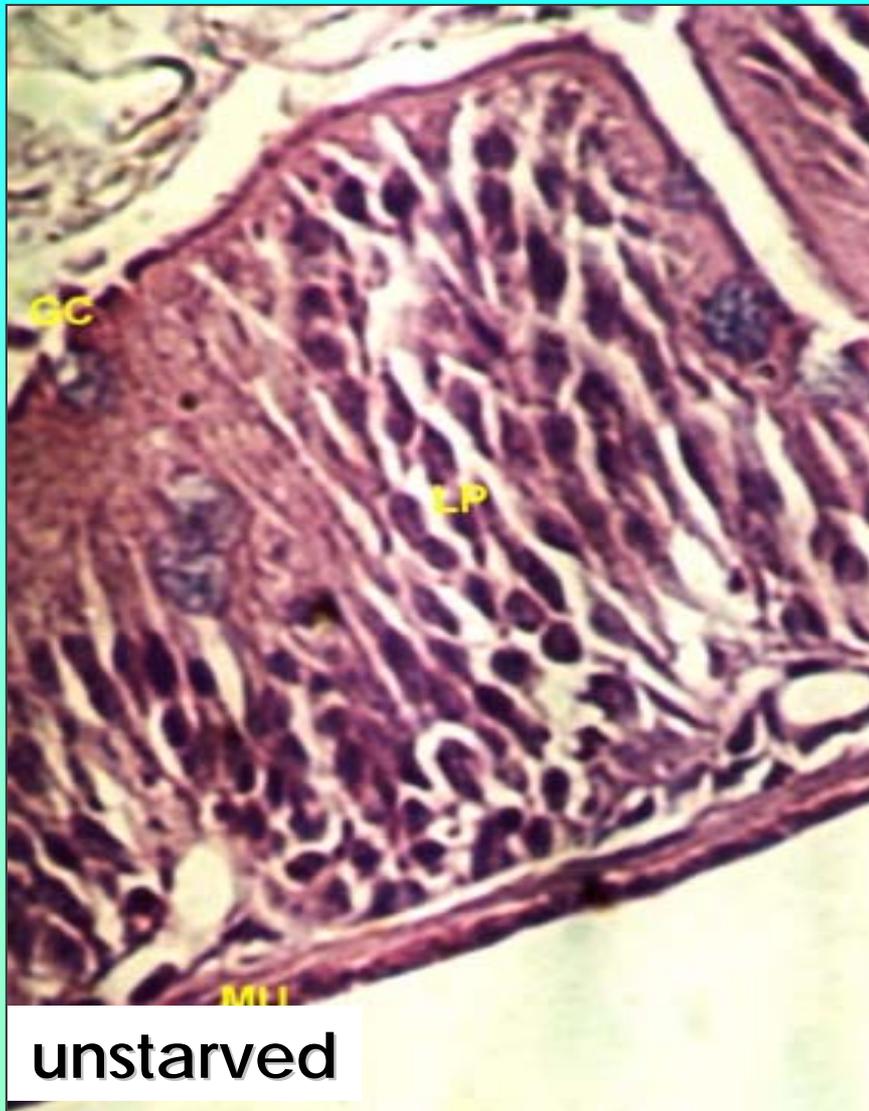
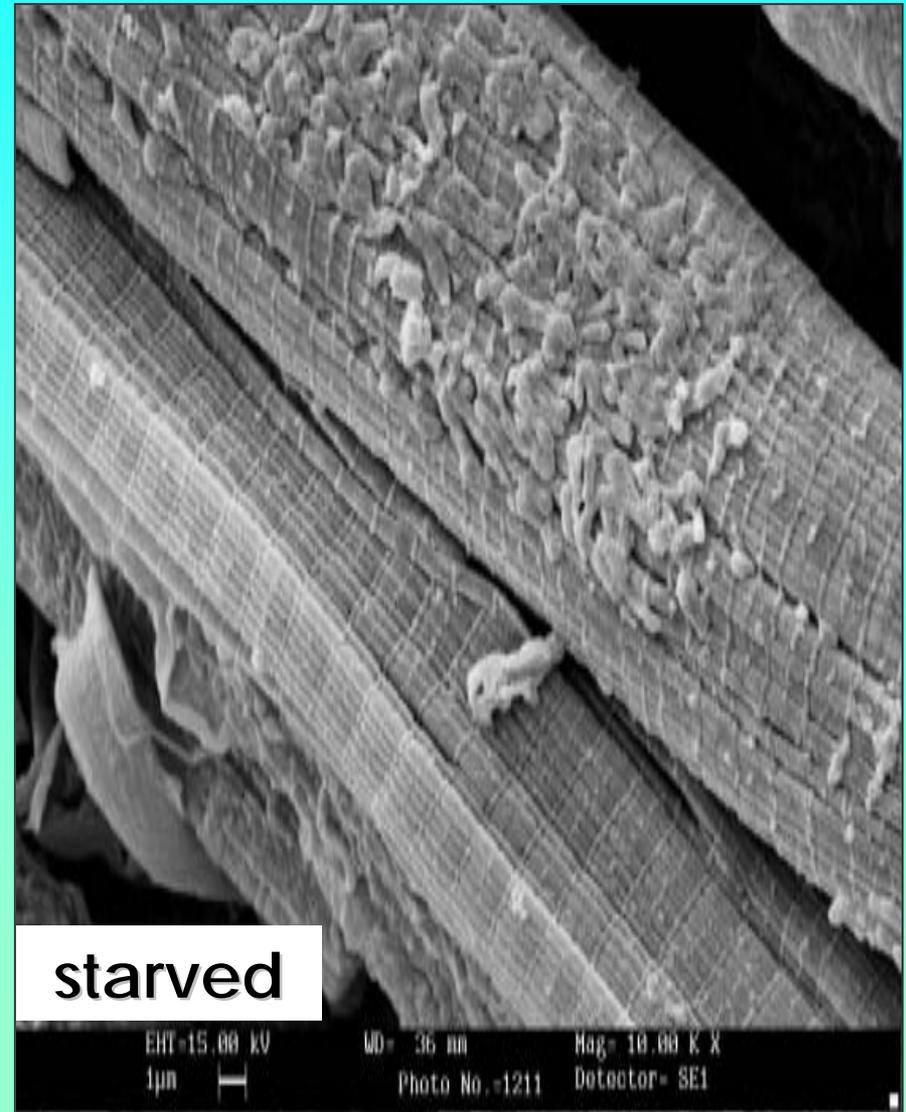
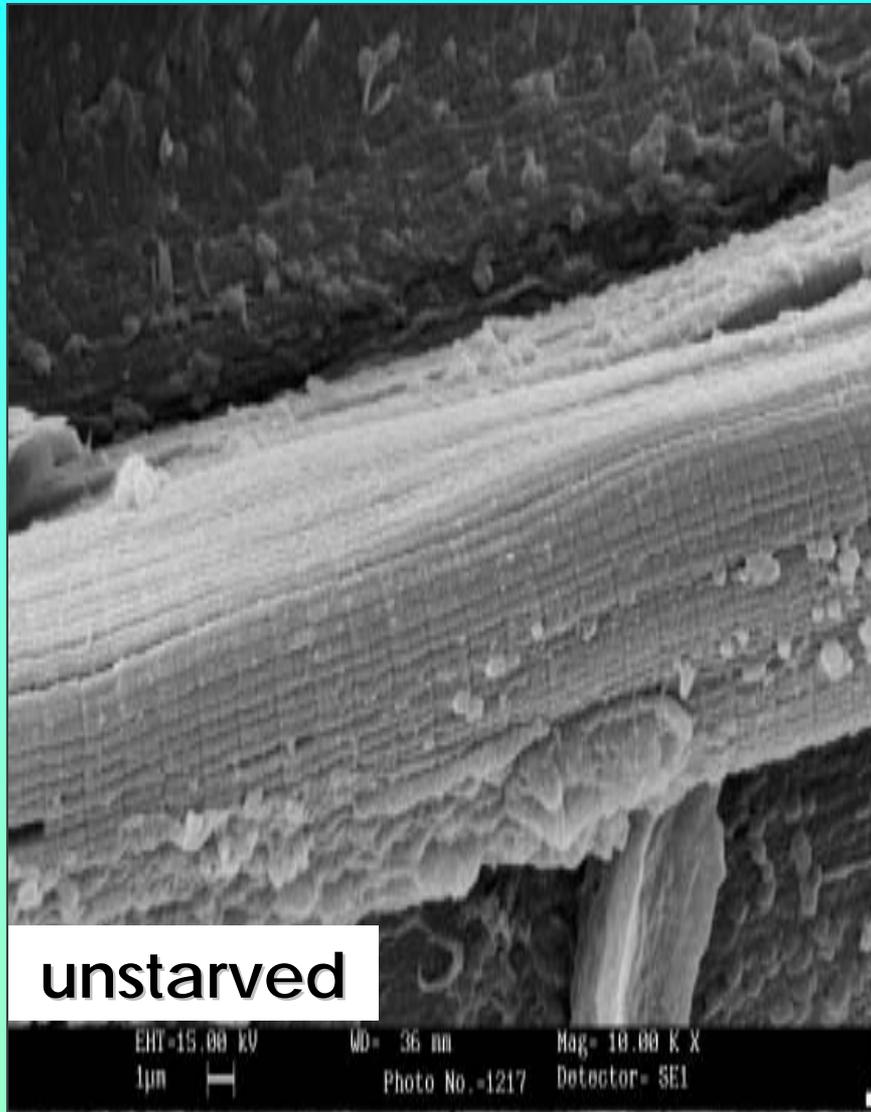


Fig. 13. Cross-section of the skeletal muscle of the 120 dph Nile Tilapia, X1000.



Fig. 14. Scanning electron micrograph of the longitudinal section of the 150 dph Nile Tilapia, X10,000.



What then might have caused the significant differences in total weight and length between unstarved and starved fish?

Recent findings:

Delayed development or smaller size of the following in starved fish were observed:

- 1. Kidney and testis (Badulis, R.J., A.A. Herrera and M.D. Fabillo, 2004)**
- 2. Cardiovascular system (Cruz, J., A.A. Herrera and M.D. Fabillo, 2004)**
- 3. Immune system (Tiongco, A.T., A.A. Herrera and M.D. Fabillo, 2004)**
- 4. Skin and skeletal system – thinner adipose tissue in hypodermis (Somejo, M., A.A. Herrera and M.D. Fabillo, 2004)**

CONCLUSIONS

- ✚ Delay in initial feeding has inhibitory effect on the total length and weight of the 30 dph until mature marketable fish (150 dph). The most statistically significant result was observed in the fish starved for 8 days before initial feeding.

CONCLUSIONS

- ✚ **Histological measurements used for this study, did not show any statistically significant difference between the unstarved and starved fish, except the abundance of goblet cells in the unstarved fish which was higher than those found in the starved fish aged 90-150 dph.**

CONCLUSIONS

- ✚ **Skeletal muscle TEM and SEM analyses showed that there were no observable differences between unstarved and starved fish.**

RECOMMENDATIONS

- It may be useful to study several hormones, such as growth and thyroid hormones, in relation to delay in first feeding.

Thank you!