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สนับสนุนโดย

สำนักงานกองทุนสนับสนุนการวิจัยและสำนักงานคณะกรรมการอุดมศึกษา

**Risk Assessment of Endocrine and Reproductive Physiology in Fish Collected from
Different Ecological Reservoirs in Khon Kaen Province
(Bung Kaen Na Korn and Bung Kang Num Ton)**

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Abstract

Environmental contamination becomes a major problem worldwide due to ubiquitous chemicals from agricultural, industrial, and household discharge as well as natural products. Using many animal models and various techniques, researchers have revealed several mechanisms of actions that have been postulated to impair normal physiological function, however, is discrepancy between laboratory and field study is still exist. This study was designed to investigate if there are any indicators implying that reproductive potential of fish collected from the so-called “Bung Kaen Na Korn” (KNK) which used to be collecting area for waste water and it is still vulnerable to be exposed to household discharge. In addition, KNK has a history of high mortality of fish in the reservoirs a few times. Bung Kang Num Ton (KNT) was used to be a controlled site since situate in a much less dense community without any evidence of high mortality. Nile tilapia (*Oreochromis niloticus*) and Silver barb (*Puntius gonionotus*) were chosen as animal model. Plasma steroids, eg. testosterone, 11keto-testosterone and 17 β -estradiol, triiodotyronine and tetraiodotyronine were measured. Gonadosomatic index (GSI) and Liversomatic index (LSI) were also recorded as well as morphological and histological of the liver. Selected heavy metals were also analyzed. Testosterone and 17 β -estradiol concentrations were highly varied while 11keto-testosterone concentration in the male of tilapia was consistently higher than silver barb. There were no differences of T3 and T4 concentration as well as for GSI and LSI indices. Selected heavy metals include Pb, Cd, Zn, Ni, and Cu bioconcentrations in a pooled liver and gonad were similar between two reservoirs, 0.006-0.011 mg/g dry weight of Pb, and 0.002-0.003, 0.074-0.085, 0.011-0.013 and 0.005-0.044 mg/g dry weight, respectively. In addition, there was no evidence of pesticides bioaccumulation except for 4,4 DDD which found in pooled liver tissue in very small amount, but it was not detected in pooled water sample and pooled sediment sample collected from both lakes.

Evidence of blood congestion found only in tilapia liver coincide with histological evidence revealed sign of cell death, vacuole and disappearance of hepatic lobule and hepatic cell shrinkage was not correlated with selected environmental parameters in this study.

In summary, there is no clear evidence suggesting an impaired reproductive performance based on our primary results. Evidence of selected heavy metal bioconcentration suggesting a vulnerability of consumer for metal toxicity. Even though those heavy metal concentration is not too high, a possibility of biomagnification is still exist. These pose interesting scenario for further investigation. Furthermore, cause of observed blood congestion and abnormal cell morphology remain to be identified.

1. Introduction

Environmental contamination reflects predominately from mishandling of domestic, industrial and agricultural wastes and can have chronic or lethal effects on aquatic organisms (Foster, 1995; Kelly et al., 1991; Anderson et al., 1998). Lethal effects occur within a short time after exposure, usually under a day or two in contrast to chronic when effects are more subtle. In the past few years, effects of environmental chemicals at sub-lethal concentration have been widely investigated and many mechanism of actions have been proposed include hormone mimicking action, enzyme induction, enzyme inhibition, and interruption on various physiological processes of many organs. These usually have been related to alteration on growth performance, reproductive system and endocrine system are often reportedly in many species eg. lake trout, frog (Jurgella et al, 2006, Mosconi et al., 2005). Discrepancy was also found between a field observation and a laboratory testing. Interpretation of those results is even more difficult with confounding factors involved include species differences, developmental stages.

Disruption of hormone eg steroid and thyroid hormones are often used as early alarming to environmental contamination (Leatherland, 1994; Crain et al., 1997; Guillette et al., 1999). These hormones particularly testosterone, 11keto-testosterone and 17 β -estradiol play important roles in reproductive system of fish. Although thyroid hormone play important roles in growth and thermoregulation, its also play integral roles in many physiological processes include reproductive system (Cook, 1996).

In this study, Bung Kaen Na Korn (KNK), an approximate 661,600 m² water body, was selected due to its location and being a collecting site for household discharge for a certain period of time. There was evidence of sudden high mortality occurred in

the past few years. Recently, this reservoir has been renovated to be a recreational area for the city. Bung Kang Na Korn, is situated in the city of Khon Kaen. It locates in between a dense community, and has long been a collecting area for run off and discharge from the community. Since 1985 a draining system was built around the reservoir to protect a direct discharge. However, it is believed that leaching of water from surrounding area is still existence. Bung Kang Num Ton (KNT), an approximate 8,000,000 m² water body, locates at a southwest of the city of Khon Kaen about 8 km. This latter reservoir is selected as a control. Tilapia and silver barb are common species found in these two reservoirs thus were selected as a representative of fish species.

The purpose of this study was to investigate whether there is evidence of a reproductive impairment in fish collected from the two reservoirs. Several indices related to endocrine and reproductive physiology include plasma steroids (testosterone, 11-ketotestosterone and 17 β -estradiol), thyroid hormone, Gonado-somatic index (GSI), Liver-somatic index (LSI) were used in this study. In addition, selected heavy metals in fish tissue were also determined. These are to provide early warning indices (if any) and necessary basic information for further study, management, policy and regulation.

2. Materials and methods

2.1 Animals

Tilapia (*Oreochromis niloticus*) and silver barb (*Puntius gonionotus*) were chosen for this study due to their widely distribution and economic important. Adult tilapia (n = 87) and silver barb (n = 85) were captured between August, 2005 and July 2006 from Bung Kaen Na Korn (n = 117) and Bung Kang Num Ton (n = 55). Fish were captured by cast net, hook, trap and gill net at three seasons, September-October, 2005,

January-February, 2006 and May-June, 2006 with sample sizes of tilapia of 10, 24 and 53, and silver barb, 20, 39 and 26, respectively. After captured, fish were transported to aerated tanks at Department of Fisheries, Faculty of Agriculture, Khon Kaen University where they were held until samples, generally within five hours of capture. Weight and length were recorded individually prior to blood sample and sex was also determined.

In the second year (August 2006-July 2007), tilapia and silver barb were captured for the lakes using similar time as stated in previous paragraph. Necessary animal tissues were sampled at Department of Fishery, Faculty of Agriculture, Khon Kaen University for further analysis.

2.2 Chemical

Radioimmunoassay kits for testosterone and 17β -estradiol were purchased from CIS bio international, France. 11keto-testosterone enzyme immunoassay kit was purchased from Caymen chemical Ann Arbor, MI. Triiodotyronine (T3) and tetraiodotyronine (T4) kits were purchased from MP Biomedicals, LLC, Ohio. Additional hormone analysis was conducted at the radioimmunoassay laboratory, Faculty of Medicin, Khon Kaen University.

2.3 Heavy metal and pesticides analysis

Lead, cadmium, zinc, nickeled and copper were measured by atomic absorption spectroscopy (Absorption Spectrophotometer: Analyst 100, Perkin-Elmer, USA) in a laboratory at Department of Environmental Engineering, Kasetsart University, Bangkok, Thailand according to the procedures described in Perkin-Elmer Corporation (1996).

In the second year, heavy metal analysis from liver tissues, water and sediment was analyzed at the Laboratory Center for Food and Agriculture Products Co., Ltd

(LCFA), Khon Kaen branch using similar parameters stated in previous paragraph (AOAC, 2000). In addition, pesticide analysis (organophosphates and organochlorines; OPs and OCs, respectively) was also conducted at LCFA. Gass chromatography screening method was applied using 23 and 18 standard for OPs and OCs, respectively (APHA, AWWA, WEF, 2005).

2.4 Blood sample

Blood samples were taken from the caudal vein using heparinized syringes follow by centrifugation (10,000 rpm for 10 minutes at 4°C) (Centrifuge 2500, Harikul, Thailand) to separate plasma fraction. Samples were stored at –20°C until analyzed.

2.5 Liver and gonad samples

Liver and gonad tissues were collected following dissection. Morphology of the liver and gonad was observed and their weight was recorded. The tissues were then preserved in neutral formalin for further histological analysis using hematoxylin and eosin staining. Liver and gonadal somatic indices, LSI and GSI respectively, were calculated according to equation 1 and 2, respectively.

$$\text{LSI} = (\text{liver weight} / \text{body weight}) \times 100 \quad (1)$$

$$\text{GSI} = (\text{gonad weight} / \text{body weight}) \times 100 \quad (2)$$

2.6 Water quality

Water quality measurements included temperature, pH, and dissolved oxygen (DO) were made with calibrated meters.

2.7 Statistical analysis

In most case, data in this annual report is presented using mean and standard deviation (SD). A thorough statistical analysis will be applied on a completion of the additional analysis.

3. Results

3.1 Hormone levels

Steroid hormones includes testosterone, 11keto-testosterone and 17 β -estradiol are measured together with thyroid hormone (T3 and T4). Due to a shipment mishap, the May-June, 2005 samples were destroyed. Testosterone concentration from sample collected from KNK and KNT between January-February, 2006 and May-June, 2006 showed considerable variation (Figure 1). Mean testosterone concentration of male tilapia collected from KNK is higher than male tilapia from KNT. Similarly, mean testosterone concentration of silver barb collected from KNK is higher than KNT, but testosterone concentration of silver barb is several fold lower than tilapia. For 11keto-testosterone, mean concentration was between 7-9 ng/ml for most groups (Figure 2). In case of 17 β -estradiol concentration, there was no clear suggestion of differences between species or sites (Figure 3) with the mean concentration ranging between 1.1-1.6 ng/ml. Thyroid hormone concentration showed considerable high variation between sites and time of sampling, while the concentration of T4 is consistently several fold higher than the concentration of T3 for both tilapia and silver barb collected from the two reservoirs (Figure 4).

3.2 GSI, LSI

There was high variation of GSI and LSI for the two fish species and collecting sites. In addition, there was no trend of differences among the predetermined sampling times (Figure 9-14)

Interestingly there was evidence of blood congestion in most of the tilapia liver, which was ranged from 45-100% of tilapia collected from KNK. In contrast, no blood congestion evidence was found in silver barb collected either from KNK or KNT sites.

3.3 Heavy metals and Water quality

Heavy metal concentration was not on the original proposal, but it was eventually included due to the history of KNK which used to be a collecting area for household discharge. The concentration of selected heavy metals includes Pb, Cd, Zn, Ni and Cu. Each of the heavy metals was present in the pooled liver and gonad samples from both reservoirs, although in low concentrations (Table 1). Primary result showed that the concentration of selected heavy metals indicated above is relatively low. In addition, the heavy metals found from two collection sites and species of fish was found to be similar.

In terms of water quality indices, water temperature of both reservoirs was generally low during January-February, 2006, 26.4-29.8 °C at KNK and 22.5-27.5 at KNT sites (Table 2 and 3). Dissolved oxygen and pH in both reservoirs were similar and above 5 mg O₂ /L and between 7-8, respectively.

3.4 Pesticide analysis

Based on organophosphate and organochlorine pesticides screening using GC (Table 4) compared with 23 organophosphate and 18 organochlorine standards,

surprisingly it was found that only 4,4 DDD was detected in fish liver tissue from both lakes (KNK and KNT).

3.5 Morphological and histological of liver tissue

Histological study (hematoxylin and eosin staining) using liver that show blood congestion (45-100% of tilapia collected from KNK), it was found that an abnormal liver tissue (B) from fish liver showing external blood congestion reveal evidence of cell death, vacuole and disappearance of hepatic lobule which differ from a normal liver tissue (A) (Figure 16)

4. Discussion

High variation of steroid hormone content particularly for testosterone and 17β -estradiol could be partially explained by a small sample size. Being a multiple spawning species, steroid hormones produced by tilapia will render more variation due to its developmental stages rather than seasonal (Hines et al., 1999). Additional release of tilapia could also be accounted for those variations (communication). However, previous explain could not be used for the variation of steroid produced by silver barb since it is generally accepted as seasonal spawning. The selected sampling period may not properly fit the spawning season of fish used in this study. It is generally accepted that 11keto-testosterone is probably a key gonadal steroid of the male fish. A similar concentration of 11keto-testosterone between KNK and KNT reservoirs suggested that biosynthesis of 11keto-testosterone is normal. A concentration of 11keto-testosterone produced by tilapia is consistently higher than silver barb could be due to species differences.

For thyroid hormone concentration, although T4 concentration showed a considerable high variation, its concentration is consistently higher than T3 concentration. These are particularly true for both species and reservoirs. The higher level of T4 is consistent with a normal physiological level of T4 in most vertebrates where T4 serves as less potent thyroid hormone, while T3 is generally accepted to be a potent thyroid hormone in most vertebrates (Leatherland, 1994). Concentration of T3 and T4 is commonly used to indicate some effects of environmental chemicals includes organochlorines, polycyclic aromatic hydrocarbon (Leatherland, 1994; Klasson-Wehler et al., 1998). Earlier studies suggested that deiodinase, an enzyme converts T4 to T3, is vulnerable to environmental chemicals (Leatherland, 1994; Boas et al., 2006). Disruption of this enzyme could result to alteration of T3 concentration in plasma of vertebrates. This could lead to modification of many physiological events since T3 plays integral roles of many physiological processes. However, based on the result of T3 and T4 concentration in this study, there is no evidence to suggest any alteration of thyroid hormone concentration.

GSI is commonly used to indicate a developmental stage of reproductive system in aquatic animals. Based on a result from this study, in most cases median GSI are well below 5% except for silver barb collected from KNT reservoir during September-October, 2005 (Figure 9, 10, and 11). The rise of GSI was probably due seasonal. The missing values and small sample size problem in this study make it difficult for data interpretation. Similarly, LSI from this study showed high variation, and in most cases they were consistently lower than 5%. Regardless to the high variation of GSI and LSI, observed blood congestion is strongly correlated to tilapia and histological study

showed the abnormal cell morphology which strongly correlated with the blood congestion samples as well.

Selected heavy metal accumulation measured from a pooled liver and gonad samples found in this study suggested that level of accumulation is relatively low. Surprisingly, heavy metal bioaccumulation found in the sample collected from KNK which used to be a collecting area of household discharge from a community was not different from KNT. In addition, selected pesticides screening using GC showed only 4,4 DDD which relatively non toxic metabolite of DDT. It is not know if this resulted from sensitivity of detecting unit, sampling location or number of samples from the lake.

5. Conclusion

The results of this study suggested that gonadal steroids, testosterone, 11keto-testosterone, and 17 β -estradiol, and thyroid hormones, T3 and T4, concentrations in fish collected from KNK and KNT reservoirs were similar. These suggested that a reproductive performance of fish collected from a suspected household discharge contamination KNK was not affected. However, the bioaccumulation of heavy metals may suggest vulnerability to heavy metal toxicity if exposure is being continued. Negative effects from this study could be due to parameters used may not be sensitive or there is no effect of suspected contamination. However, the results provide valuable data for further study.

Acknowledgement

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Figure 1. A comparison of plasma testosterone concentration of male Nile tilapia and silver barb collected from KNK and KNT between Jan-Feb, 06

- 1) Male Nile tilapia collected from KNK (n = 8)
- 2) Male silver barb collected from KNK (n = 8)
- 3) Male Nile tilapia collected from KNT (n = 4)
- 4) Male silver barb collected from KNT (n = 0)

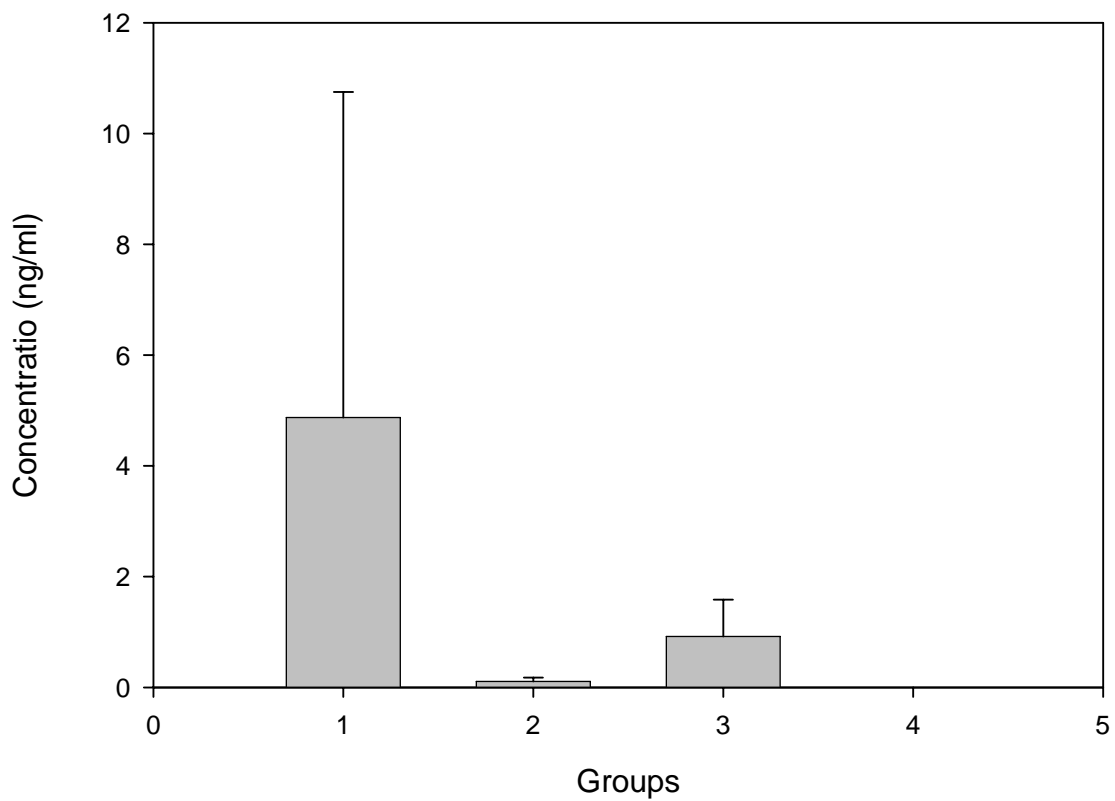


Figure 2. A comparison of plasma testosterone concentration of male Nile tilapia and silver barb collected from KNK and KNT between May-Jun, 06

- 1) Male Nile tilapia collected from KNK (n = 6)
- 2) Male silver barb collected from KNK (n = 5)
- 3) Male Nile tilapia collected from KNT (n = 6)
- 4) Male silver barb collected from KNT (n = 0)

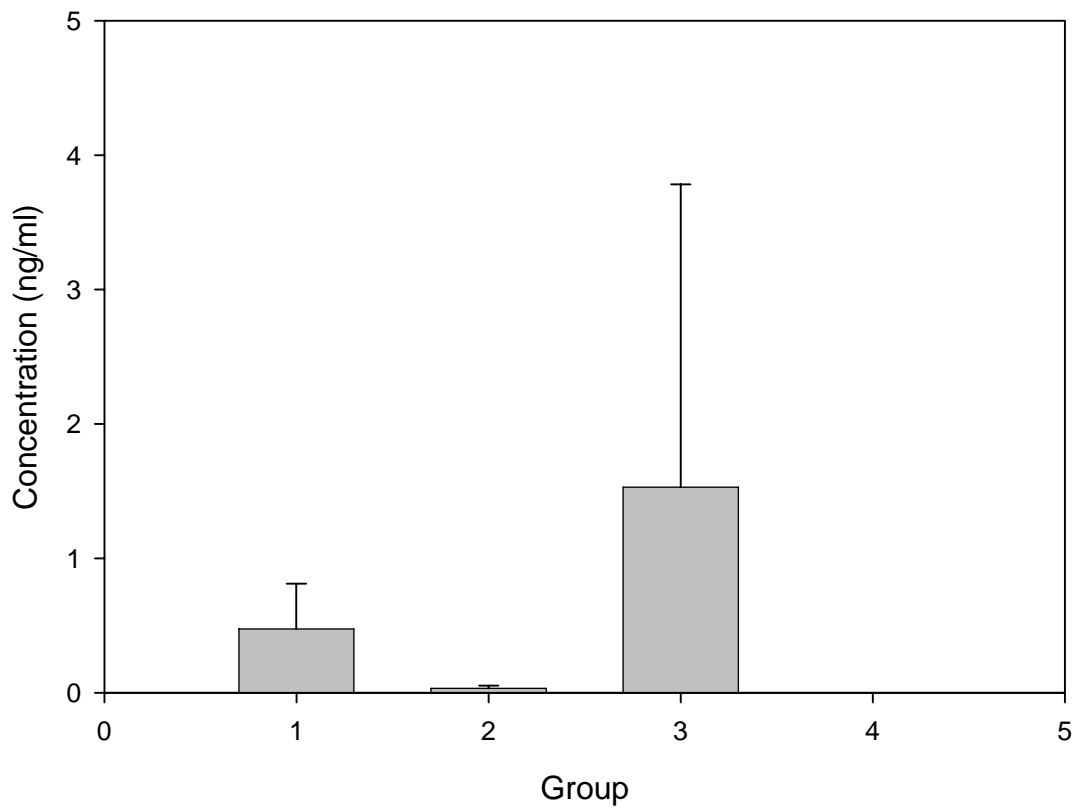


Figure 3. A comparison of plasma 11keto-testosterone concentration of male Nile tilapia and silver barb collected from KNK and KNT between Jan-Feb, 06

- 1) Male Nile tilapia collected from KNK (n = 5)
- 2) Male silver barb collected from KNK (n = 4)
- 3) Male Nile tilapia collected from KNT (n = 1)
- 4) Male silver barb collected from KNT (n = 0)

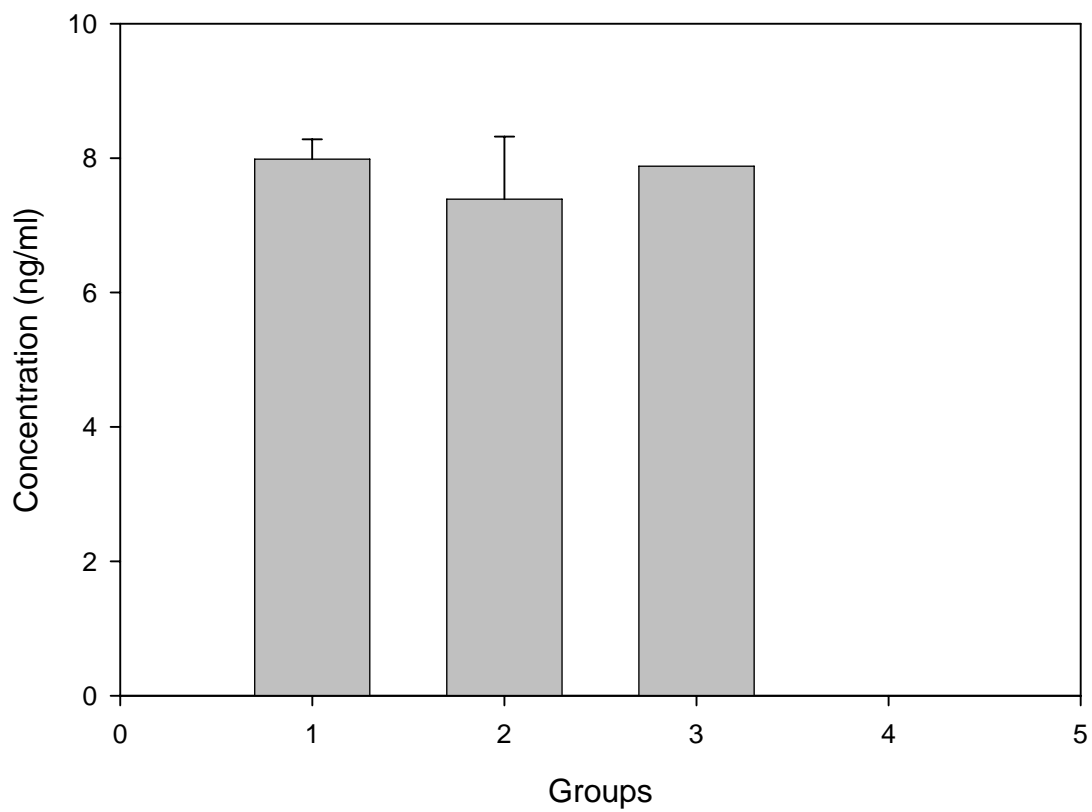


Figure 4. A comparison of plasma 11keto-testosterone concentration of male Nile tilapia and silver barb collected from KNK and KNT between May-Jun, 06

- 1) Male Nile tilapia collected from KNK (n = 6)
- 2) Male silver barb collected from KNK (n = 3)
- 3) Male Nile tilapia collected from KNT (n = 6)
- 4) Male silver barb collected from KNT (n = 4)

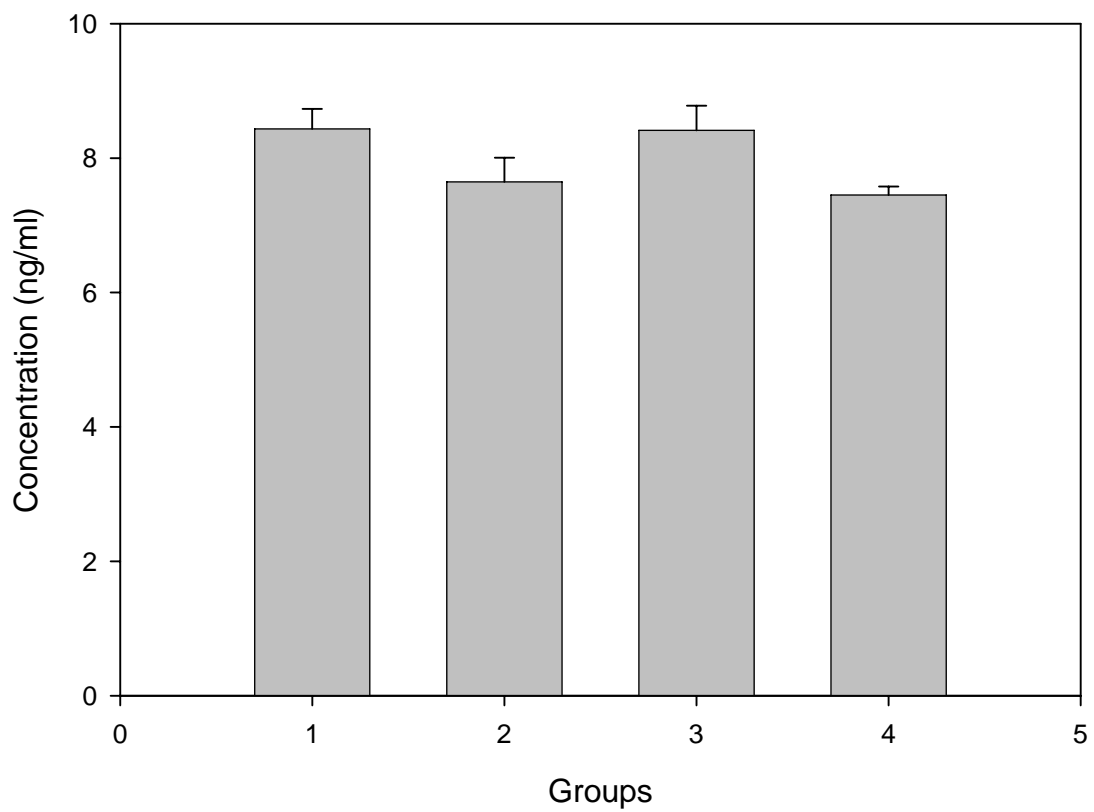


Figure 5. A comparison of plasma 17β -estradiol concentration of female Nile tilapia and silver barb collected from KNK and KNT between Jan-Feb, 06

- 1) Female Nile tilapia collected from KNK (n = 5)
- 2) Female silver barb collected from KNK (n = 7)
- 3) Female Nile tilapia collected from KNT (n = 6)
- 4) Female silver barb collected from KNT (n = 0)

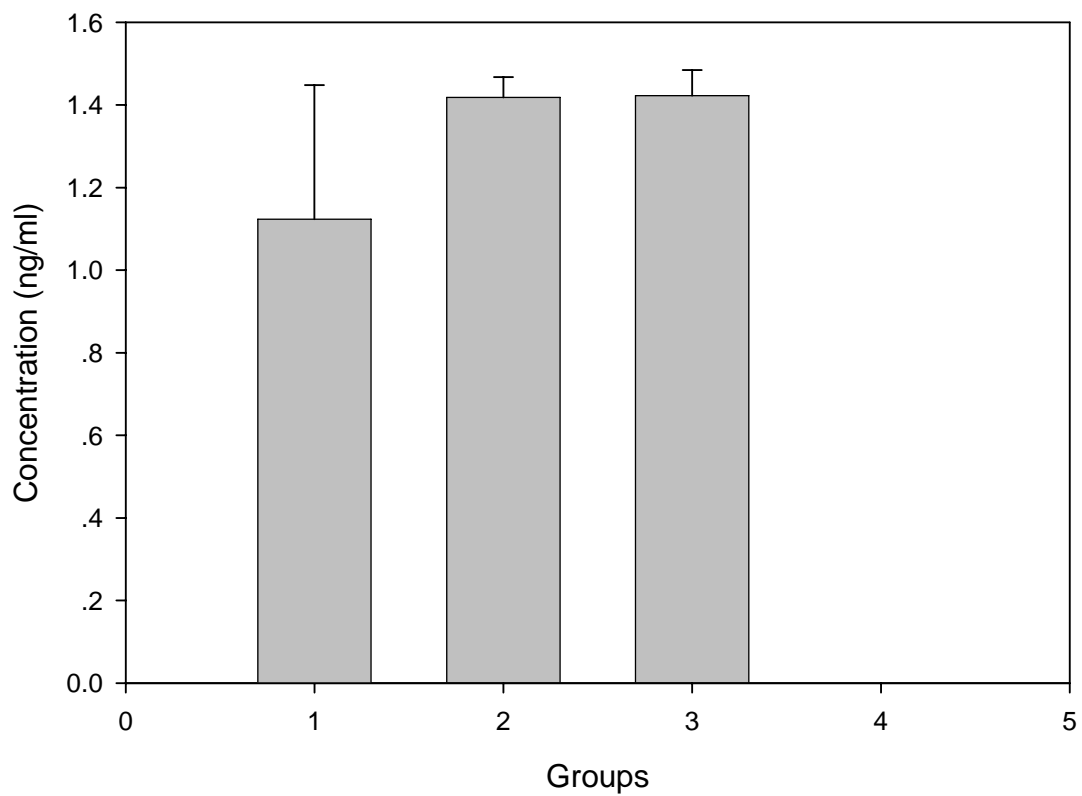


Figure 6. A comparison of plasma 17β -estradiol concentration of female Nile tilapia and silver barb collected from KNK and KNT between Jan-Feb, 06

- 1) Female Nile tilapia collected from KNK (n = 6)
- 2) Female silver barb collected from KNK (n = 6)
- 3) Female Nile tilapia collected from KNT (n = 6)
- 4) Female silver barb collected from KNT (n = 0)

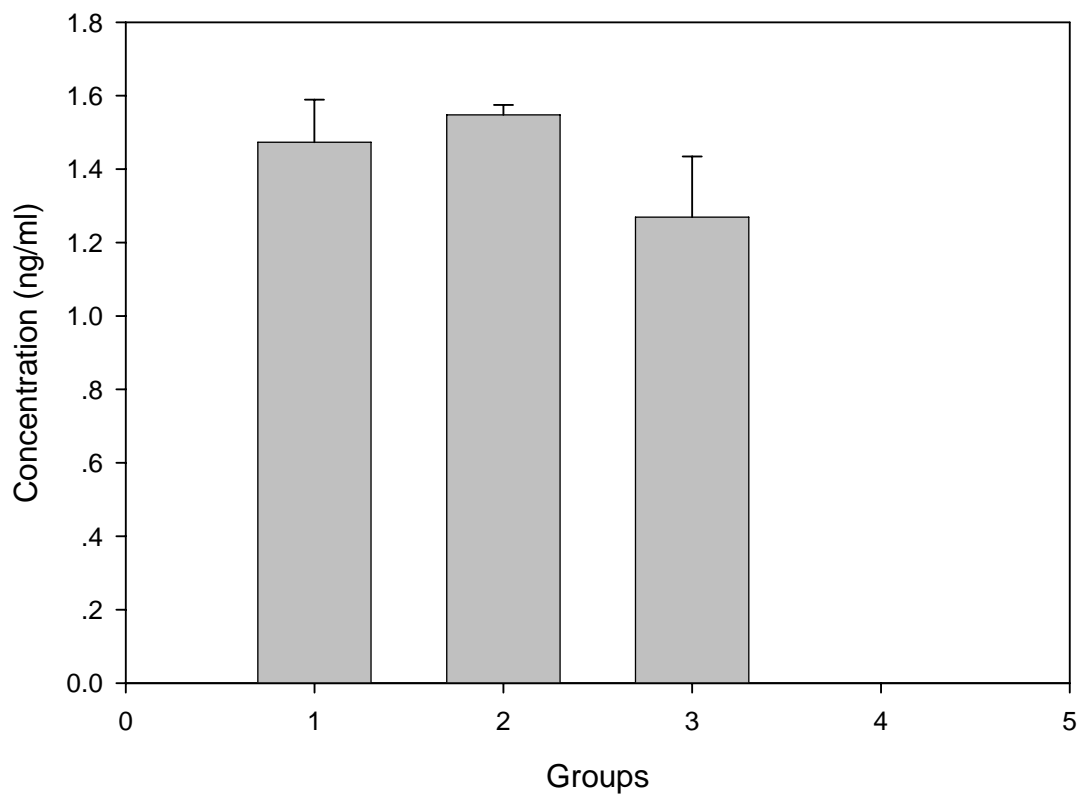


Figure 7. A comparison of plasma triiodothyronine (T3) and tetraiodothyronine (T4) concentrations of Nile tilapia and silver barb collected from KNK and KNT between Jan-Feb, 06

- 1) T3 concentration of Nile tilapia collected from KNK (n = 8)
- 2) T4 concentration of Nile tilapia collected from KNK (n = 8)
- 3) T3 concentration of silver barb collected from KNK (n = 11)
- 4) T4 concentration of silver barb collected from KNK (n = 11)
- 5) T3 concentration of Nile tilapia collected from KNT (n = 8)
- 6) T4 concentration of Nile tilapia collected from KNT (n = 8)
- 7) T3 concentration of silver barb collected from KNT (n = 0)
- 8) T4 concentration of silver barb collected from KNT (n = 0)

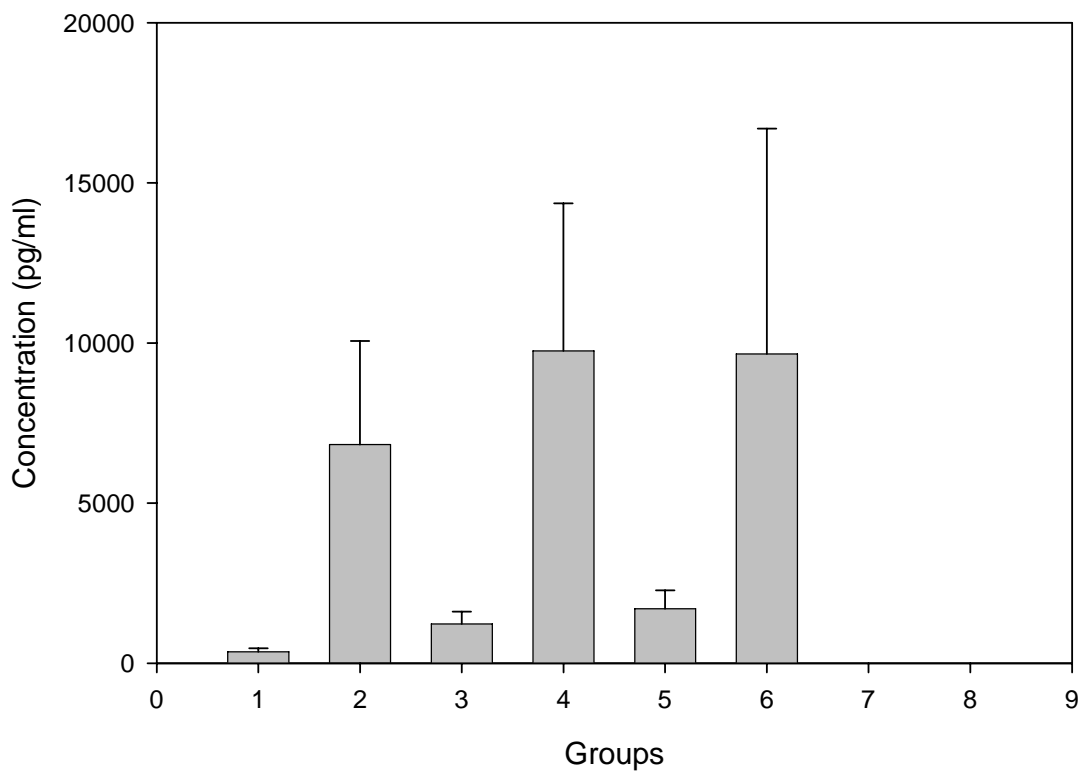


Figure 8. A comparison of plasma triiodothyronine (T3) and tetraiodothyronine (T4) concentrations of Nile tilapia and silver barb collected from KNK and KNT between May-Jun, 06

- 1) T3 concentration of Nile tilapia collected from KNK (n = 13)
- 2) T4 concentration of Nile tilapia collected from KNK (n = 13)
- 3) T3 concentration of silver barb collected from KNK (n = 16)
- 4) T4 concentration of silver barb collected from KNK (n = 16)
- 5) T3 concentration of Nile tilapia collected from KNT (n = 7)
- 6) T4 concentration of Nile tilapia collected from KNT (n = 7)
- 7) T3 concentration of silver barb collected from KNT (n = 7)
- 8) T4 concentration of silver barb collected from KNT (n = 7)

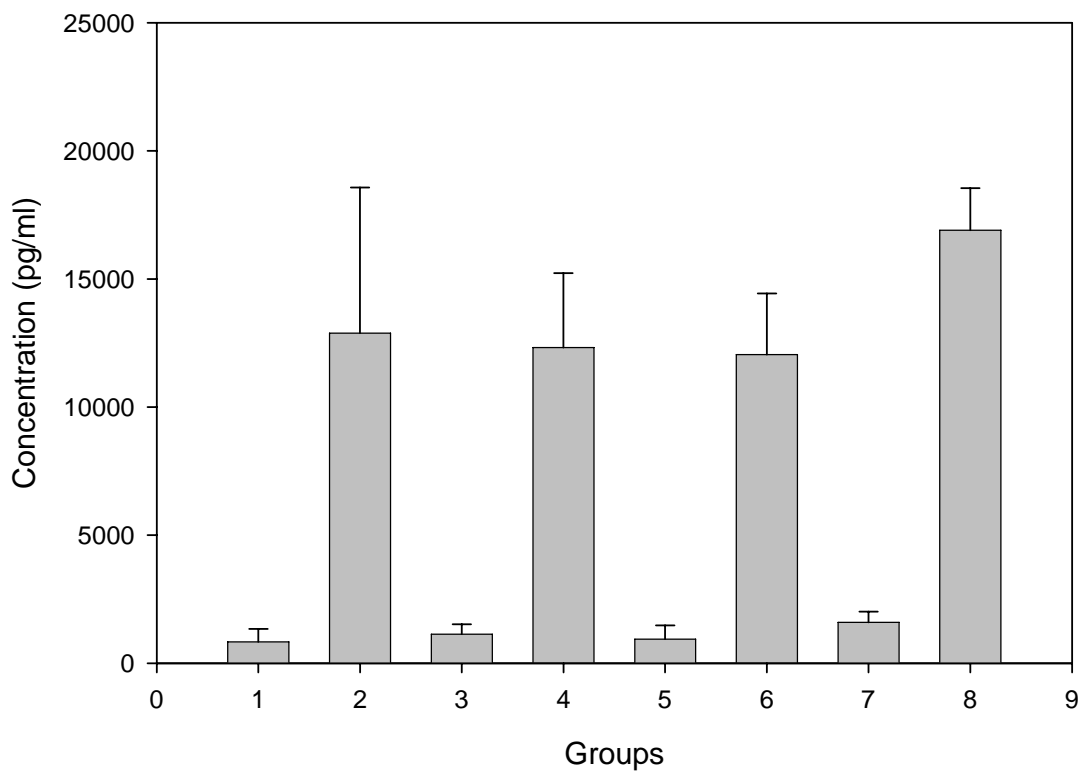


Figure 9. A comparison of gonado-somatic index (GSI) of Nile tilapia and silver barb collected from KNK and KNT between Sep-Oct, 05. Horizontal line on the bar represents the median

- 1) GSI of male Nile tilapia collected from KNK (n = 6)
- 2) GSI of female Nile tilapia collected from KNK (n = 0)
- 3) GSI of male silver barb collected from KNK (n = 6)
- 4) GSI of female silver barb collected from KNK (n = 0)
- 5) GSI of male Nile tilapia collected from KNT (n = 0)
- 6) GSI of female Nile tilapia collected from KNT (n = 0)
- 7) GSI of male silver barb collected from KNT (n = 3)
- 8) GSI of female silver barb collected from KNT (n = 13)

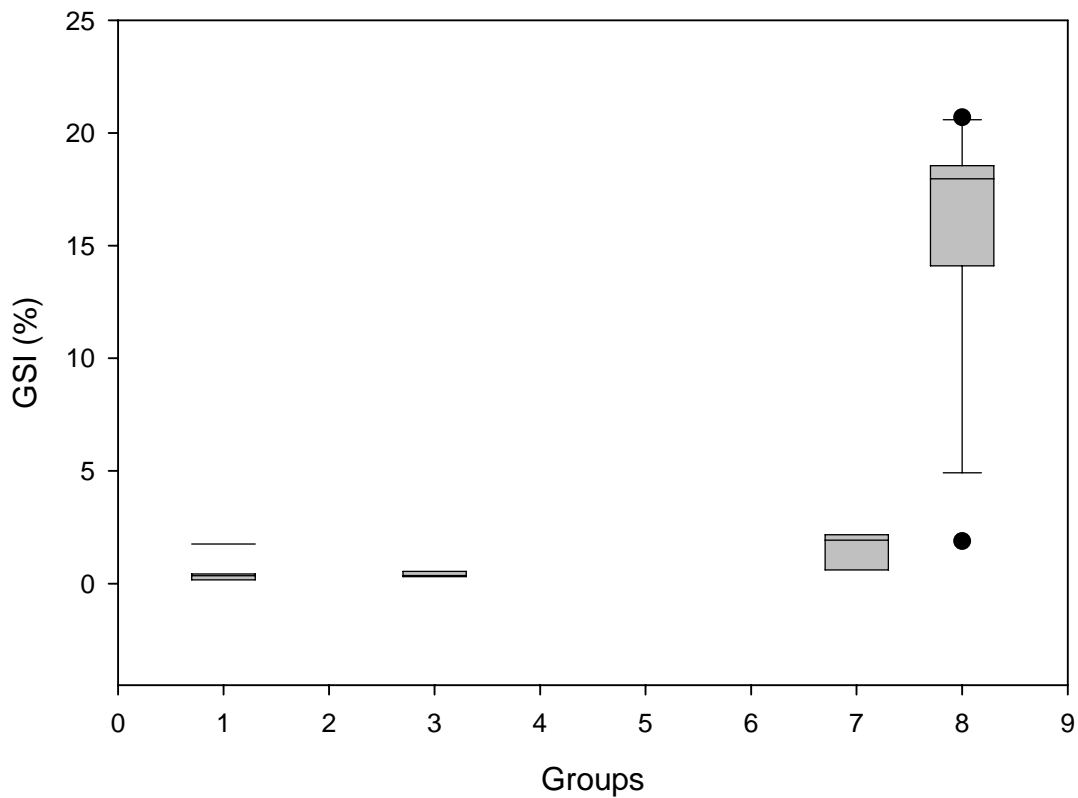


Figure 10. A comparison of gonado-somatic index (GSI) of Nile tilapia and silver barb collected from KNK and KNT between Jan-Feb, 06. Horizontal line on the bar represents the median

- 1) GSI of male Nile tilapia collected from KNK (n = 8)
- 2) GSI of female Nile tilapia collected from KNK (n = 5)
- 3) GSI of male silver barb collected from KNK (n = 24)
- 4) GSI of female silver barb collected from KNK (n = 17)
- 5) GSI of male Nile tilapia collected from KNT (n = 5)
- 6) GSI of female Nile tilapia collected from KNT (n = 10)
- 7) GSI of male silver barb collected from KNT (n = 0)
- 8) GSI of female silver barb collected from KNT (n = 0)

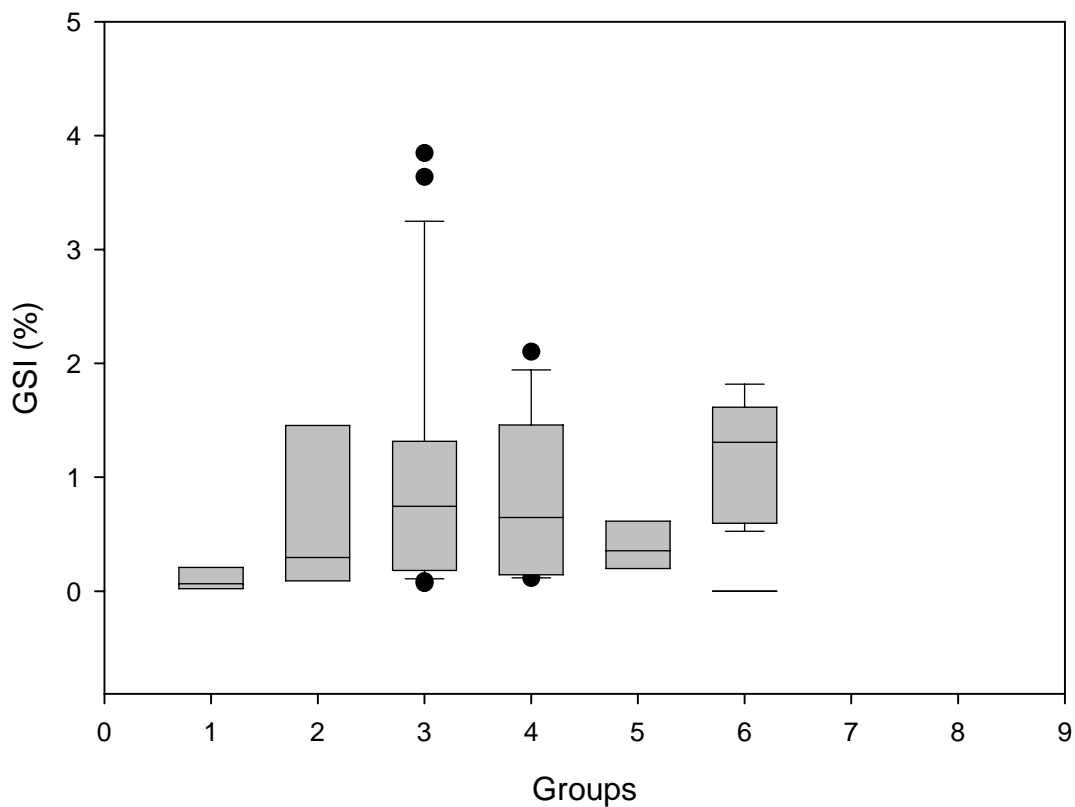


Figure 11. A comparison of gonado-somatic index (GSI) of Nile tilapia and silver barb collected from KNK and KNT between May-Jun, 06. Horizontal line on the bar represents the median

- 1) GSI of male Nile tilapia collected from KNK (n = 12)
- 2) GSI of female Nile tilapia collected from KNK (n = 17)
- 3) GSI of male silver barb collected from KNK (n = 11)
- 4) GSI of female silver barb collected from KNK (n = 17)
- 5) GSI of male Nile tilapia collected from KNT (n = 15)
- 6) GSI of female Nile tilapia collected from KNT (n = 13)
- 7) GSI of male silver barb collected from KNT (n = 0)
- 8) GSI of female silver barb collected from KNT (n = 0)

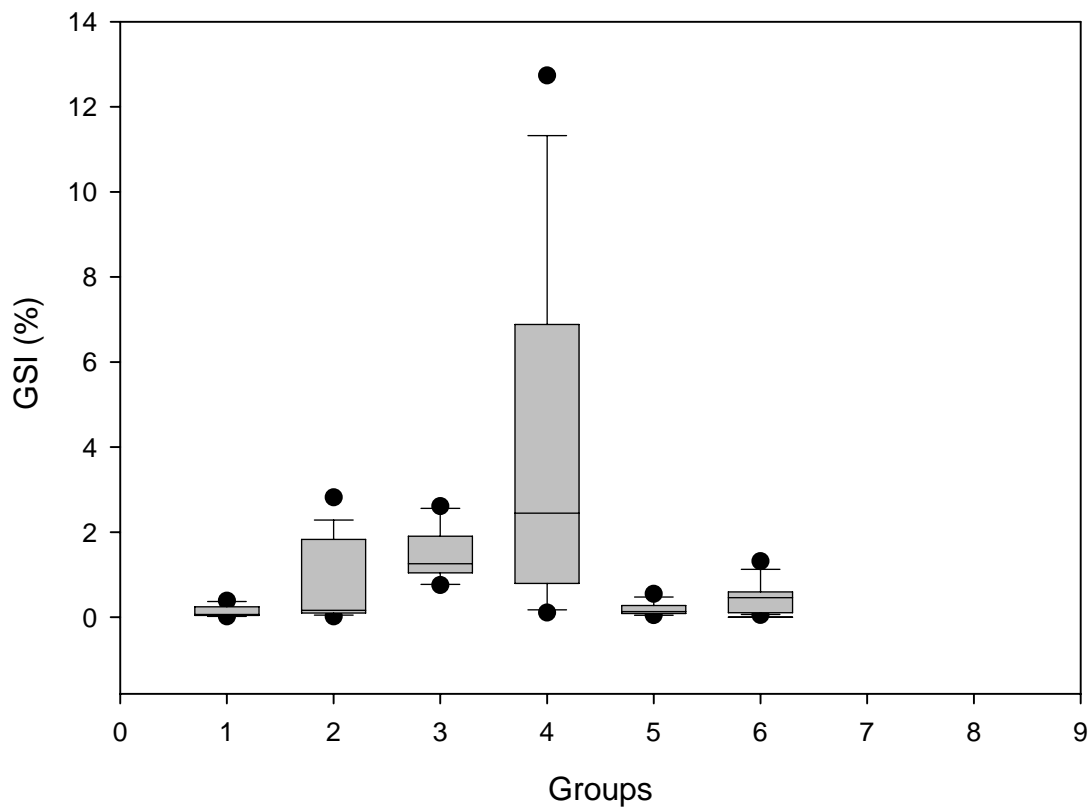


Figure 12. A comparison of Liver-somatic index (LSI) of Nile tilapia and silver barb collected from KNK and KNT between Sep-Oct, 06. Horizontal line on the bar represents the median

- 1) LSI of male Nile tilapia collected from KNK (n = 6)
- 2) LSI of female Nile tilapia collected from KNK (n = 0)
- 3) LSI of male silver barb collected from KNK (n = 6)
- 4) LSI of female silver barb collected from KNK (n = 0)
- 5) LSI of male Nile tilapia collected from KNT (n = 0)
- 6) LSI of female Nile tilapia collected from KNT (n = 0)
- 7) LSI of male silver barb collected from KNT (n = 4)
- 8) LSI of female silver barb collected from KNT (n = 13)

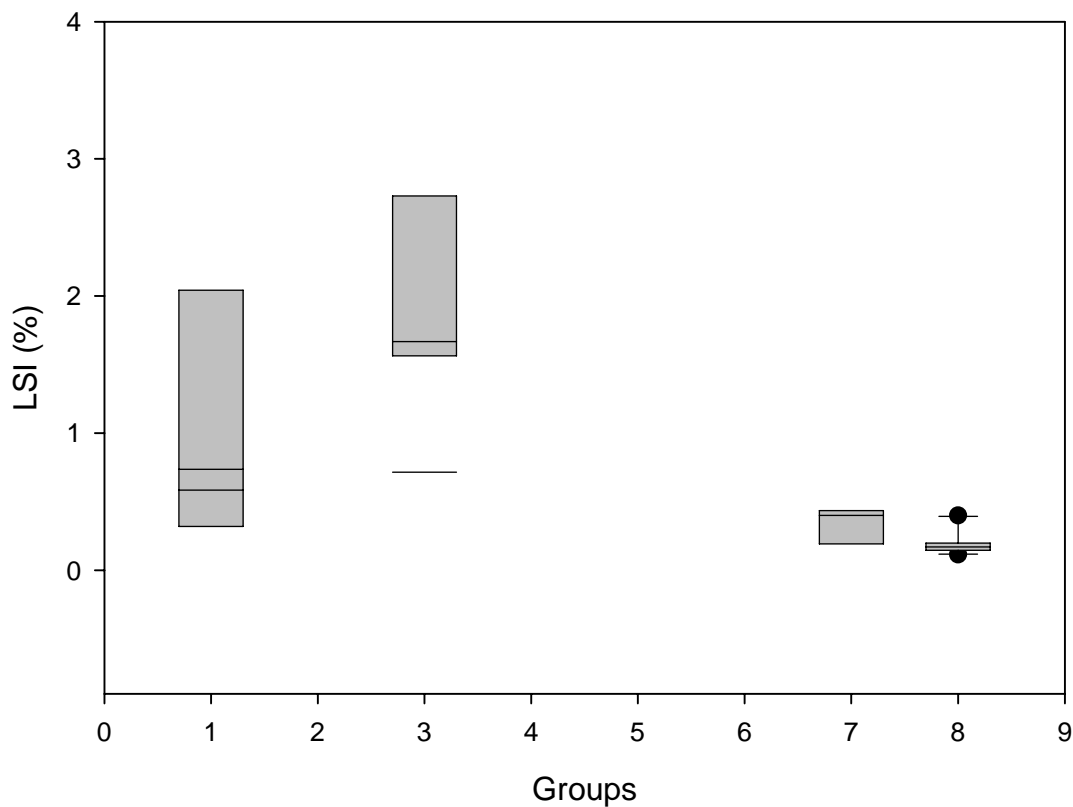


Figure 13. A comparison of Liver-somatic index (LSI) of Nile tilapia and silver barb collected from KNK and KNT between Jan-Feb, 06. Horizontal line on the bar represents the median

- 1) LSI of male Nile tilapia collected from KNK (n = 8)
- 2) LSI of female Nile tilapia collected from KNK (n = 5)
- 3) LSI of male silver barb collected from KNK (n = 24)
- 4) LSI of female silver barb collected from KNK (n = 17)
- 5) LSI of male Nile tilapia collected from KNT (n = 5)
- 6) LSI of female Nile tilapia collected from KNT (n = 10)
- 7) LSI of male silver barb collected from KNT (n = 0)
- 8) LSI of female silver barb collected from KNT (n = 0)

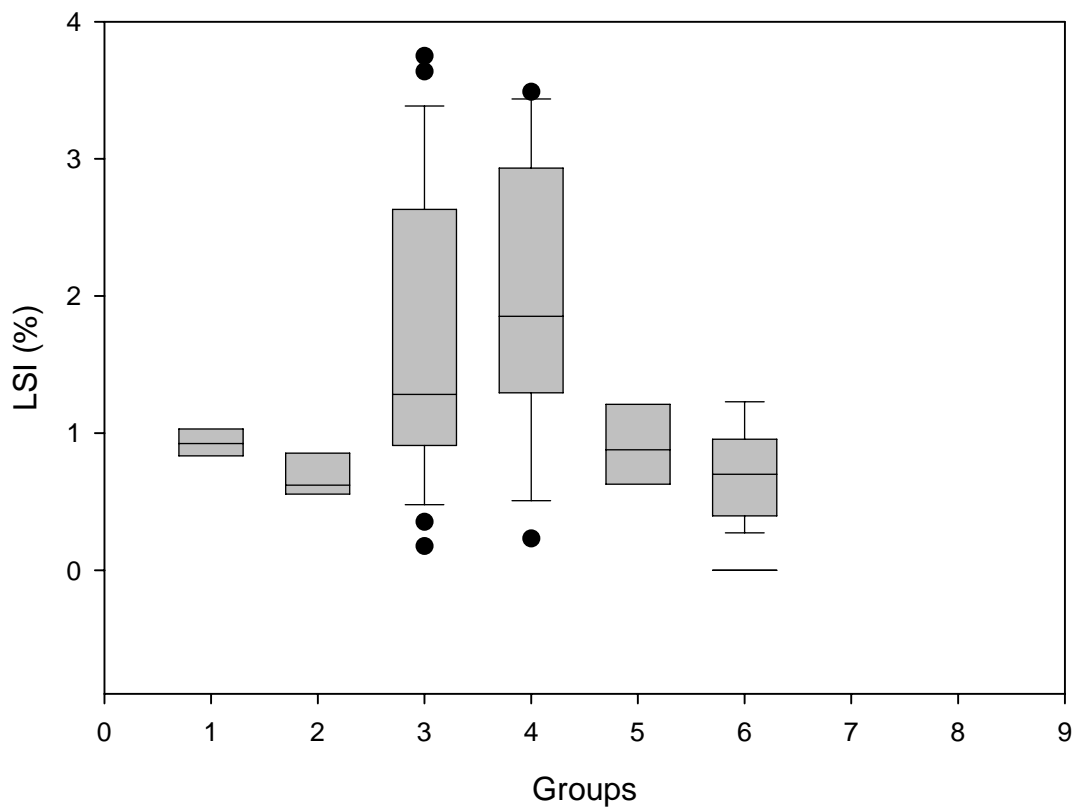


Figure 14. A comparison of Liver-somatic index (LSI) of Nile tilapia and silver barb collected from KNK and KNT between May-Jun, 06. Horizontal line on the bar represents the median

- 1) LSI of male Nile tilapia collected from KNK (n = 12)
- 2) LSI of female Nile tilapia collected from KNK (n = 17)
- 3) LSI of male silver barb collected from KNK (n = 11)
- 4) LSI of female silver barb collected from KNK (n = 17)
- 5) LSI of male Nile tilapia collected from KNT (n = 15)
- 6) LSI of female Nile tilapia collected from KNT (n = 13)
- 7) LSI of male silver barb collected from KNT (n = 0)
- 8) LSI of female silver barb collected from KNT (n = 0)

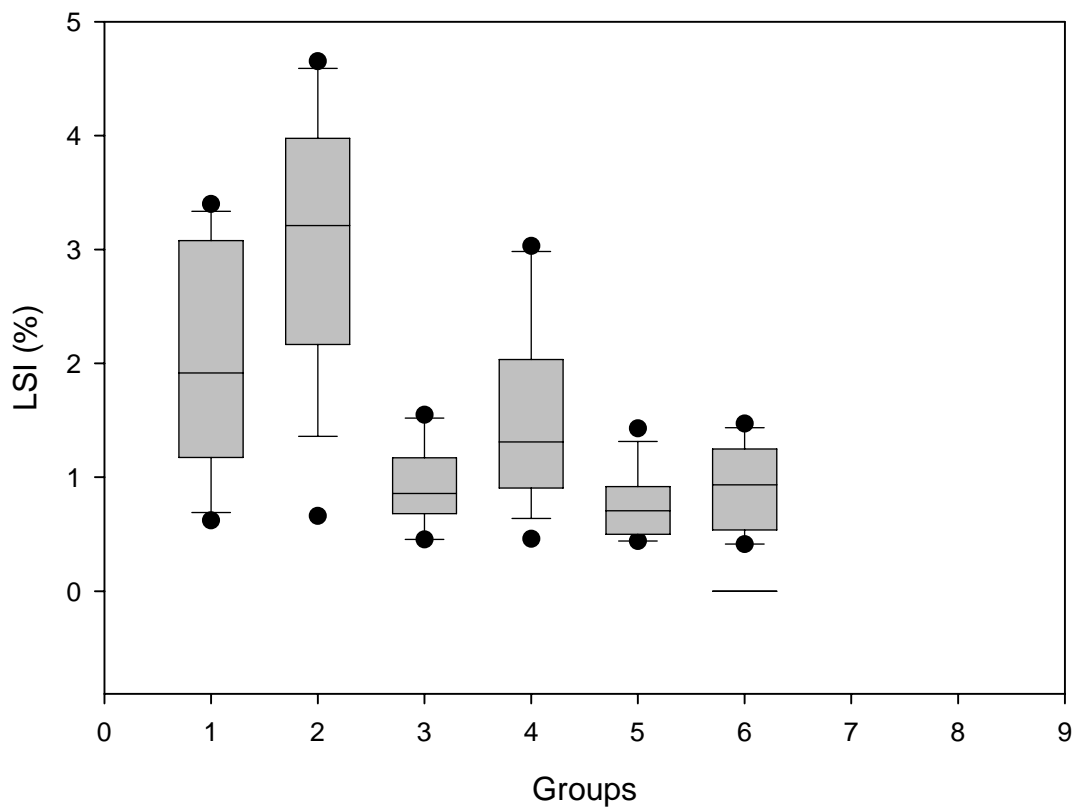


Figure 15 Observed blood congestion on intact liver from tilapia sample.



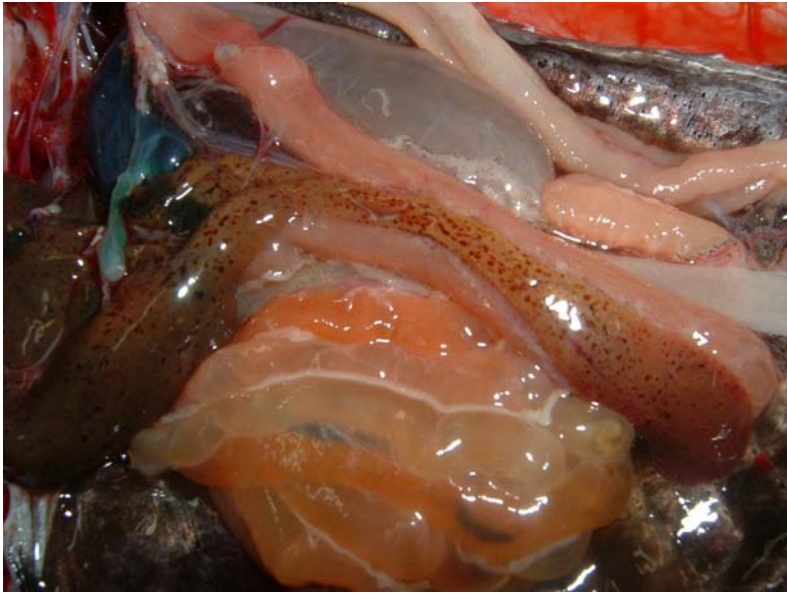
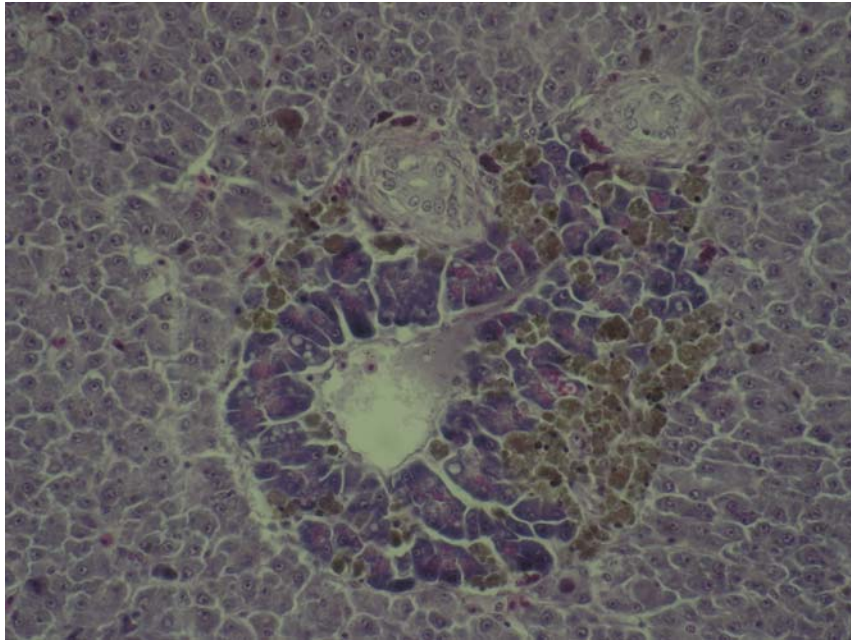


Figure 16 Histology of normal liver tissue (A) and abnormal liver tissue (B) from fish liver showing external blood congestion with cell death, vacuole and disappearance of hepatic lobule

(A)



(B)

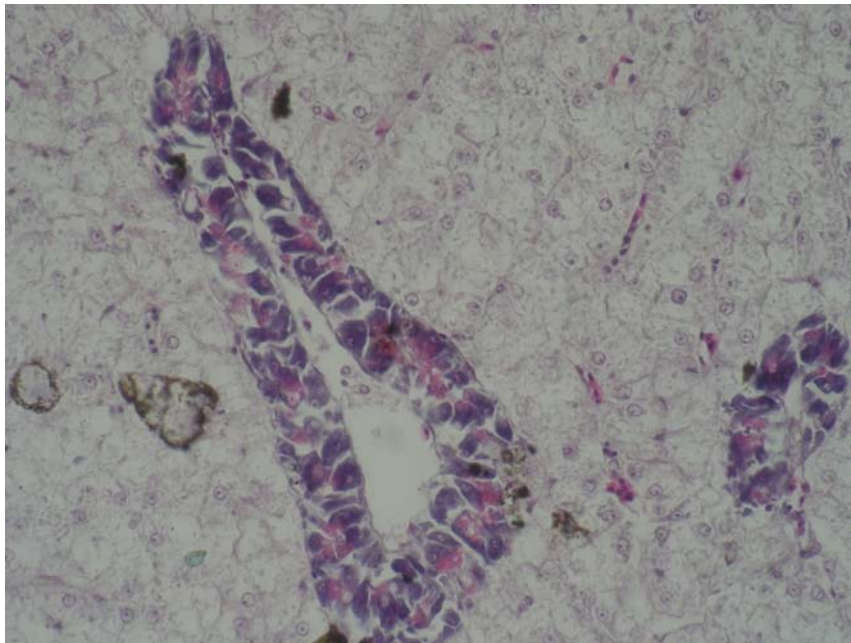


Table 1. Heavy metal accumulation in pooled sample of liver and gonad tissues from selected fish

Source	Species	Heavy Metal (mg/g dry weight)				
		Pb	Cd	Zn	Ni	Cu
KNK	Tilapia	0.006	0.003	0.074	0.013	0.044
	Barb	0.002	0.002	0.079	0.011	0.005
KNT	Tilapia	0.011	0.003	0.085	0.013	0.044
	Barb	-	-	-	-	-

Table 2. Illustration of water quality parameters in Bung Kaen Na Korn

Parameters Time	Temperature (°C)	pH	Dissolved Oxygen (mg/l)
Sept-Oct, 05	28.5-31.0	7.23-8.02	6.50-7.16
Jan-Feb, 06	26.4-29.8	7.46-8.34	5.98-7.56
May-June, 06	30.7-33.1	7.54-8.12	5.34-6.78

Table 3. Illustration of water quality parameters in Bung Kang Num Ton

Parameters Time	Temperature (°C)	pH	Dissolved Oxygen (mg/l)
Sept-Oct, 05	28.40-28.54	6.88-8.11	6.56-7.08
Jan-Feb, 06	22.50-27.57	7.44-7.86	6.53-7.12
May-June, 06	30.22-32.00	7.26-7.90	6.20-7.31

Table 4 Organophosphate and organochlorine standards used for pesticide screening
 (-: no detection).

Item	Detected Unit (mg/L or mg/kg)					
	Liver tissue		Water		Sediment	
	KNK	KNT	KNK	KNT	KNK	KNT
Organophosphates						
Methamidophos	-	-	-	-	-	-
Mevinphos	-	-	-	-	-	-
Diazinon	-	-	-	-	-	-
Dicrotophos	-	-	-	-	-	-
Monocrotophos	-	-	-	-	-	-
Dimethoate	-	-	-	-	-	-
Malathion	-	-	-	-	-	-
Fenitrothion	-	-	-	-	-	-
Prothiophos	-	-	-	-	-	-
Pirimiphos-methyl	-	-	-	-	-	-
Chlorpyrifos	-	-	-	-	-	-
Pirimiphos-ethyl	-	-	-	-	-	-
Parathion-methyl	-	-	-	-	-	-
Profenofos	-	-	-	-	-	-
Triazophos	-	-	-	-	-	-
Dichlorvos	-	-	-	-	-	-
Omethoate	-	-	-	-	-	-
Parathion-ethyl	-	-	-	-	-	-
Methidathion	-	-	-	-	-	-
Ethion	-	-	-	-	-	-
EPN	-	-	-	-	-	-
Phosalone	-	-	-	-	-	-
Azinphos-ethyl	-	-	-	-	-	-
Organochlorines						
Alpha-HCH	-	-	-	-	-	-
Gamma-HCH	-	-	-	-	-	-
Beta-HCH	-	-	-	-	-	-
Heptachlor	-	-	-	-	-	-
Aldrin	-	-	-	-	-	-
Heptachlor epoxide	-	-	-	-	-	-
2,4 DDE	-	-	-	-	-	-
4,4 DDE	-	-	-	-	-	-
Alpha-endosulfan	-	-	-	-	-	-
2,4 DDD	-	-	-	-	-	-
Deildrin	-	-	-	-	-	-
2,4 DDT	-	-	-	-	-	-
Endrin	-	-	-	-	-	-
4,4 DDD	0.09	0.09	-	-	-	-
Beta-endosulfan	-	-	-	-	-	-
4,4 DDT	-	-	-	-	-	-
Endosulfansulfate	-	-	-	-	-	-
Dicofol	-	-	-	-	-	-