

## Prevalence of Monogenetic Trematodal Diseases in Some freshwater fishes at Kafr El-Sheikh Governorate

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**Abstract:** Eight hundred specimens of freshwater fishes (*Oreochromis niloticus* and *Clarias gariepinus*) (100 fish per season/ species) were investigated for seasonal incidence of monogenean trematodes (*Dactylogyrus sp.* and *Gyrodactylus sp.*). The clinical signs of most examined fishes showed dark or pale body coloration, scales detachment in *O. niloticus*, emaciation in *C. gariepinus*, excessive amounts of mucous on the external body surface, ascites, scattered hemorrhagic patches and ulcerative areas in different parts of the skin as well as severe congestion of the gills. *Dactylogyrus sp.* have the highest rate of the infestation (17.5% and 34%) in *O. niloticus* and *C. gariepinus*, respectively. While, *Gyrodactylus sp.* infection was found to be a mixed infection in all cases with *Dactylogyrus sp.*; showing a prevalence rate (0.25% and 2.25%) in *O. niloticus* and *C. gariepinus*, respectively. The seasonal prevalence for monogenean infestation was the highest in summer season (39% and 63%), followed by spring (14% and 41%), autumn (13% and 27%) and winter (4% and 5%) among *O. niloticus* and *C. gariepinus*, respectively. In addition, some hematological investigations and histopathological alterations of the infested freshwater fishes were recorded.

**Keywords:** *Clarias gariepinus*, monogenea, *Oreochromis niloticus*, Prevalence.

### 1. Introduction

Aquatic species are considered one of the most important sources of animal proteins; the world can rely on it to compensate the shortage in high quality protein due to the rapid increase of human population (Abd El-Aziz, 2002). Kafr El-Sheikh Governorate has the highest fish production rate; 599,698 MT from either governmental; 3146 MT or private ( owned, leased or temporarily) fish farms; 196,000, 194,20 and 280,000 MT, respectively, also the production from rice fields and cages were 1894 and 100,000 MT, respectively (GAFRD, 2015).

Increasing intensification of fish and lack of health management measures lead to many disease problems in fish and about 80% of fish diseases are parasitic (Eissa, 2002). In Egypt, the optimum warm weather, enable the outburst of parasites spread; causing worse effects on fish (Eissa et al., 2013). Generally, fish parasites result in economic losses not only due to mortalities, tissue damage and growth reduction, but also due to high expenses of drug treatment (El-Asely et al., 2015).

Monogeneans are a group of parasitic flatworms; commonly found on fishes and lower aquatic invertebrates (Reed et al., 2012). Monogenean worms are ectoparasites that are known to be infectious to a wide variety of fish. (Lari and Pyle, 2017). These worms are characterized by having a posterior attachment organ known as the haptor or opisthaptor

(Gusev, 1985). Monogeneans can be divided into two major groups, the monopisthocotyleans which have hook-like organs on their haptors to attach to their host, and the polyopisthocotyleans which use clamp-like structures for attachment (Reed et al., 2012). Monogeneans have a direct life cycle. The larva is usually a small ciliated oncomiracidium, which hatches from the egg and swims to locate and infect another host (MonoDb, 2015). Monogenean pathogenicity is due to their attachment organs, gland secretion and feeding strategy (Buchmann and Bresciani, 2006). Other information on pathogenicity is demonstrated by Noga (2010) and Reed et al. (2012).

The member species of Monogenoidea, a class of phylum Platyhelminthes, are primarily ectoparasitic usually infecting the gills and/or external surfaces of freshwater and marine fishes. *Dactylogyrus* is one of the largest genera of the monogenoidea with more than 900 species described to date (Gibson et al., 1996). More than 95% species of *Dactylogyrus* are parasites of the cyprinid fish (Gibson et al., 1996 and Šimková et al., 2007) and are known to be significant pathogens, producing chronic debility, poor development and growth, impaired respiration, mass mortality of infested hosts and significant economic losses in aquaculture (Bauer, 1951; Paperna, 1963; Woo et al., 2002; Reed et al. 2009; Jiang et al. 2013; Tu et al., 2015; Wangchu et al., 2016 and

**Chaudhary et al., 2017**). These monogeneans are the world's most common gill parasites of freshwater fishes (**Woo, 2006**).

Species of *Gyrodactylus* (Monogenea) are viviparous ectoparasites, living on the body surfaces (i.e. body trunk, fins, gills, eyes, buccal cavity and olfactory chamber) of fish (**Cone & Cusack 1988; Bakke et al., 2007**). Having a direct life cycle and high reproductive rates, *Gyrodactylus sp.* can significantly infect both captive and wild fish (**Cone & Odense 1984**).

*Gyrodactylus sp.* strongly attach themselves to fish gills, fins and skin. Their movement from one location to another on fish cause integumental breaks as well as mechanical injuries to the epithelium (**Cone and Odense, 1984**). These injuries are effective entrance routes consequently; enhance the transmission competence of pathogens making fish with *Gyrodactylus* more susceptible to secondary bacterial infections (**Shoemaker et al., 2008**).

The present study was carried out to investigate the seasonal incidence of monogenetic trematodal infestation among freshwater fishes (*Oreochromis niloticus* and *C. gariepinus*) as well as, evaluation of some haematological, serum biochemical parameters, immunological indices and histopathological changes induced by the detected parasites.

## 2. Materials and Methods

### Fish samples:

A total number of 800 examined cultured freshwater fishes; 400 *Oreochromis niloticus* and 400 *Clarias gariepinus* with different weights and sizes; were collected alive from different freshwater fish farms in Kafr El-Sheikh governorate along the four seasons of the year 2016. The collected fishes were transferred alive to the wet lab., Fish Diseases and Management Department, Faculty of Veterinary Medicine, Kafr El-Sheikh University, Egypt (**Hetrick, 1983 and Langdon & Jones, 2002**). Collected samples were held in well-prepared glass aquaria supplied with sufficient amounts of dechlorinated water with continuous aeration (**Innes, 1966**).

### Clinical examination:

The alive collected fishes were subjected to full clinical examination for the changes in colour and any clinical abnormalities on the external body surface (skin, gills, eye and mouth), just immediately after picking up from glass aquaria, and for any external gross lesions like wounds, hemorrhages, ulcers, slimness or eroded skin, according to the method described by **Lucky (1977); Austin & Austin (1987); Woo (1995) and Noga (2010)**.

### Parasitological Examination:

The parasitological examination was carried out for the detection and identification of the monogenean

ectoparasites on the skin and gills of the collected samples of *Oreochromis niloticus* and *Clarias gariepinus*.

Tissue scrapings from skin and gill filaments were obtained by scraping either the skin and/or the outer layer of gill filaments and spread with a drop of normal saline and covered with a clean cover slip (Wet mount preparation) and examined microscopically (**Lucky, 1977**).

Monogenean ectoparasites were collected using binocular dissecting microscope with a small pipette and transferred into small petri-dish and cleared several times with water to remove the attached debris and mucus. The worms were then left in refrigerator at 4 °C for complete relaxation, fixed in 5 % formalin for permanent preparation, washed carefully in water to get rid of formalin traces and finally stained with acetocarmine for 5-10 minutes. Specimens were passed through ascending grades of ethyl alcohol (30, 50, 70, 90 % and absolute) for dehydration and then cleared in clove oil, xylene and mounted in Canada Balsam (**Pritchard and Kruse, 1982**).

### Haematological investigations:

Fresh blood samples were collected without anticoagulant from the caudal posterior blood vessels. The needle is run, quite deep, as much as possible through a middle line just behind the anal fin in a dorso-cranial direction till striking the vertebrae. By drawing the needle gently backward, blood is usually sucked into the syringe.

Blood samples were divided into two parts; one part was collected in heparinized micro-hematocrite tubes for hematological studies and the other part was centrifuged post collection at 3000 rpm for 10 minutes to separate serum for serum biochemical analysis.

The erythrocytes, leukocytes and hemoglobin concentration were determined according to the method described by **Stoskopf (1993)**. For differential leucocytic count, blood films were prepared and stained according **Lucky (1977)**. The percentage and absolute value for each type of cells were calculated according to **Schalm (1986)**.

### Blood serum biochemical analysis:

Serum total proteins were determined colorimetrically according to the method described by **Peters et al. (1982)**. Serum albumin was estimated colorimetrically according to **Peters (1970)**, however, globulins content was calculated mathematically as described by **Doumas and Biggs (1972)**. Aspartate aminotransferase (AST) and alanine aminotransferase (ALT) activities in serum were determined according to **Reitman and Frankel, (1957)**. The serum alkaline phosphatase (ALP) was determined colorimetrically according to the method described by **Kind & King (1954)**. Creatinine value was determined according to **Henry (1974)**. Glucose level (mg/100 ml) was

determined according to Kaplan *et al.* (1984).

**Immunological studies:**

**Determination of phagocytic activity (PA) and phagocytic index (PI):**

Phagocytic activity was determined according to Kawahara *et al.* (1991).

**Statistical analysis**

Statistical analysis was performed using SPSS software version 16.0, Chicago, IL. Significant difference was determined at a probability level of ( $P < 0.05$ ).

**Histopathological Examination:**

Tissue specimens from the skin and gills of the infested fish samples were taken. Specimens were fixed immediately in 10% buffered neutral formalin, dehydrated and embedded in paraffin wax. Paraffin blocks were sectioned at 4-5  $\mu$ m thickness and stained with Hematoxylin & Eosin (H & E) and examined under light microscope (Leica) using X200 and X400 magnification power according to Bancroft and Gamble (2007).

**3. Results**

The present work was applied to investigate the seasonal incidence of monogenetic trematods (*Dactylogyrus sp.* & *Gyrodactylus sp.*) infestation among naturally infested freshwater fishes (*Oreochromis niloticus* & *Clarias gariepinus*) in Kafr El- Sheikh governorate.

**Incidences of fish monogenea among different seasons:-**

Parasitological examination of 400 *Oreochromis niloticus* and 400 *Clarias gariepinus* revealed an incidence of 17.5% and 34% respectively in different seasons.

The seasonal prevalence for monogenean infestation among *Oreochromis niloticus* and *Clarias gariepinus* was the highest in summer season (39% and 63%, respectively), followed by spring (14% and 41%, respectively), autumn (13% and 27%, respectively) and winter (4% and 5%, respectively) as shown in table (2 and 3).

**Clinical examination:**

The external gross lesions of the examined *Oreochromis niloticus* revealed dark or pale body coloration, detachment of scales in some areas of the body, excessive amounts of mucous on the external body surface, scattered hemorrhagic patches and ulcerative areas in different parts of the skin (Fig. 1) as well as severe congestion on the isthmus region and over the two gill covers (Fig. 2).

However, the infested *C. gariepinus* showed emaciation, dark or pale body coloration, excessive amounts of mucous secretion on the external body surface, erosion of the skin with some small wounds (Fig. 3), hemorrhagic ulcers especially on the ventral

abdomen (Fig. 4), and severe congestion of the gills (Fig. 5).

**Parasitological examination:**

Microscopic smears taken from skin, gills, eye and mouth of examined *O. niloticus* and *C.gariepinus* revealed the presence of both *Dactylogyrus sp.* over skin and gills, and *Gyrodactylus sp.* over the skin.

*Dactylogyrus sp.* have the highest rate of the infestation (17.5% and 34%) in *O. niloticus* and *C.gariepinus*, respectively. *Gyrodactylus sp.* infection was found to be a mixed infection in all cases with *Dactylogyrus sp.*; showing a prevalence rate (0.25% and 2.25%) in *O. niloticus* and *C.gariepinus*, respectively as shown in table (1-3).

The adult worms isolated from the skin of infested *Oreochromis niloticus*; were flat with two elliptical projections at its anterior end. The posterior end (haptor) has two pairs of anchors and a number of marginal hooklets (Fig. 6) Such adult worms are related to the phylum *Platyhelminthes*, class *Trematoda*, order *Mongenea*, family *Gyrodactylidae* and genus *Gyrodactylus cichilidae*.

However, the adult worms isolated from the skin of infested catfish; were flat and elliptical in shape and provided with one pair of projection at its anterior pole. It can be distinguished from other monogeneans by the absence of eye spots and the occurrence of the embryos in the mid-region of the body (Viviparous monogenean). Posterior end has the organ of fixation, opisthaptor, which is guarded with a number of marginal hooklets and a central one pair of hooks. Such adult worms are related to the phylum *Platyhelminthes*, class *Trematoda*, order *Mongenea*, family *Gyrodactylidae* and genus *Gyrodactylus claridii* (Fig. 7 and 9 A).

On the other side, adult worms isolated from the gills and skin of infested *Oreochromis niloticus*; were flat and elliptical in shape. Their anterior end (prohaptor) was divided into four cephalic lobed heads, with sticky and adhesive organs (cephalic glands), in addition to four black eye spots. The posterior end, appeared a dome shape and composed of one pairs of connecting bars (V-shaped) and seven pairs of small marginal hooklets. The intestinal limbs were connected, the ovary located in front to testes. Such adult worms are related to the phylum *Platyhelminthes*, class *Trematoda*, order *Mongenea* family *Dactylogyridae* and genus *Dactylogyrus cichilidae*.

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pairs of small marginal hooklets. The intestinal limbs were connected, the ovary located in front to testes. Such adult worms are related to the phylum *Platyhelminthes*, class *Trematoda*, order *Mongenea* family *Dactylogyridae* and genus *Dactylogyrus claridii* (Fig. 8 and 9 B).

#### Hematological investigations of naturally infected fishes:

Different blood parameters were illustrated in table (4).

In *Oreochromis niloticus*, the results revealed a non-significant increase in RBCs count, lymphocyte and basophil count when compared with values of normal fish samples, significant increase in eosinophil count, non significant decrease in WBCs count PCV% value, neutrophil and monocyte count as well as significant decrease in Hb amount. However, in *Clarias gariepinus*, the results revealed a non-significant increase in RBCs count, PCV% value, neutrophil and basophil count when compared with values of normal fish samples, non-significant decrease in WBCs count, lymphocyte and monocyte count as well as significant decrease in Hb amount and eosinophil count.

#### Serum Biochemical parameters of naturally infected fishes:

Some serum biochemical parameters of the liver functions of diseased and normal fish were displayed in table (5).

In *Oreochromis niloticus*, the results revealed a non-significant increase in albumin, globulin and alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP), creatinine and glucose level with liver and kidney function tests], non-significant decrease in total serum protein and A/G ratio. However, in *Clarias*

*gariepinus*, the results revealed a non-significant increase in creatinine, significant increase in globulin, non-significant decrease in total serum protein and alkaline phosphatase (ALP) level and significant decrease in albumin, A/G ratio, alanine aminotransferase (ALT), aspartate aminotransferase (AST) and glucose level.

#### Phagocytic activity and phagocytic index of naturally infected fishes:

Results of phagocytic activity and index are summarized in table (6). Both phagocyte activity and Phagocyte index was increased in both *Oreochromis niloticus* and *Clarias gariepinus*.

#### Histopathological findings

The skin layer of normal catfish was consisted from stratified squamous epithelial layers contained many alarm cells, which is large eosinophilic cell and the basal layer contained melanocytic abundant cells. The epithelial layer rest on connective tissue layer which is rich in lymphatic and vascular vessels.

While the affected fish showed erosion and desquamation of the stratified squamous layer with marked congestion of subcutaneous blood vessels with depletion of the melanotic cells (Fig. 10 A), with formation of variable skin ulcerations (Fig. 10 B). The subcutaneous tissue revealed marked vascular congestion (Fig. 10 C) and haemorrhage (Fig. 10 D). As well as, there was marked inflammatory cells infiltration mostly eosinophilic granular cells associated with oedema (Fig. 11 A). The infiltration mostly perilymphatic and perivascular (Fig. 11 B).

The gills showed marked erosive lesions of the lining epithelium of the gill filament with blunt-ended lamellae. Fusion of secondary gill lamellae associated with severe degree of leucocytic infiltration could be also observed (Fig. 11 C).

**Table ( 1): Total prevalence of monogenetic trematodes among investigated *Oreochromis niloticus* and *Clarias gariepinus*:**

Fish species	Total No. of examined fish	Positive infested cases	% of infections
<i>Oreochromis niloticus</i>	400	70	17.5
<i>Clarias gariepinus</i>	400	136	34

**Table (2): Seasonal prevalence of monogenetic trematodes among the examined *Oreochromis niloticus*:**

Season	Total no. of exam in ed fish	Positive infested cases		<i>Dactylogyrus sp.</i>		<i>Gyrodactylus sp.</i>		Mxed <i>Dactylogyrus sp.</i> + <i>Gyrodactylus sp.</i>	
		No.	%	No.	%	No.	%	No.	%
Winter	100	4	4%	4	4%	-	-	-	-
Spring	100	14	14%	14	14%	-	-	-	-
Summer	100	39	39%	39	39%	1	1%	1	1%
Autum n	100	13	13%	13	13%	-	-	-	-
Total	400	70	17.5%	70	17.5%	1	0.25%	1	0.25%

**Table (3): Seasonal prevalence of monogenetic trematodes in examined *Clarias gariepinus*:**

Season	Total no. of examined fish	Positive infested cases		<i>Dactylogyrus</i> sp.		<i>Gyrodactylus</i> sp.		Mixed <i>Dactylogyrus</i> sp. + <i>Gyrodactylus</i> sp.	
		No.	%	No.	%	No.	%	No.	%
Winter	100	5	5%	5	5%	-	-	-	-
Spring	100	41	41%	41	41%	2	2%	2	2%
Summer	100	63	63%	63	63%	-	-	-	-
Autumn	100	27	27%	27	27%	7	7%	7	7%
<b>Total</b>	<b>400</b>	<b>136</b>	<b>34%</b>	<b>136</b>	<b>34%</b>	<b>9</b>	<b>2.25%</b>	<b>9</b>	<b>2.25%</b>

**Table (4): Hematological investigations of naturally infected fishes:**

Fish Blood Parameters	<i>Oreochromis niloticus</i>		<i>Clarias gariepinus</i>	
	Infected	Non infected	Infected	Non infected
RBCs	1.8±0.100	1.76±0.06	1.67±0.07	1.62±0.05
HB	7.418±0.49 <sup>D</sup>	9.278±08 <sup>a</sup>	6.63±0.15 <sup>D</sup>	9.83±0.86 <sup>a</sup>
PCV%	26.43±1.76	29.73±0.97	29.76±0.80	28.90±0.6
WBCs	24.62±1.27	26.58±1.72	22.71±1.2	24.78±1.1
Lymphocyte	65.53±0.97	64.95±0.76	62.32±1.73	62.85±0.97
Neutrophil	23.83±0.68	25.28±1.21	27.04±1.5	26.04±0.89
Eosinophil	6.39±0.52 <sup>a</sup>	4.91±0.44 <sup>D</sup>	4.9±0.77	5.8±0.79
Basophil	2.66±0.66	2.271±0.36	3.20±0.57	2.71±0.63
Monocyte	1.76±0.37	2.71±0.55	1.7±0.66	2.6±0.67

**Table (5): Serum Biochemical investigations of naturally infected fishes:**

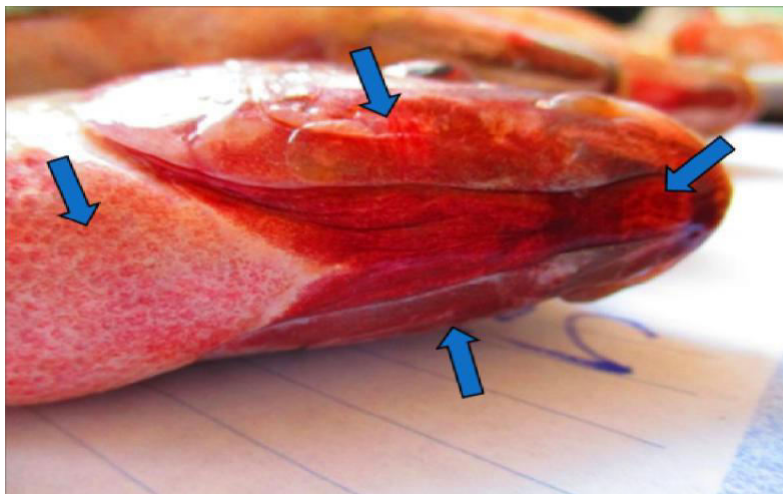
Fish Blood Parameters	<i>Oreochromis niloticus</i>		<i>Clarias gariepinus</i>	
	Infected	Non infected	Infected	Non infected
Total Protein	5.07±0.5	5.21±0.3	3.97±0.07 <sup>D</sup>	5.83±0.06 <sup>a</sup>
Albumin	3.87±0.60	3.65±0.7	1.93±0.23 <sup>D</sup>	4.95±0.06 <sup>a</sup>
Globulin	1.42±0.2	1.33±0.2	2.037±0.22 <sup>a</sup>	0.87±0.02 <sup>D</sup>
A/G	4.84±1.29	4.95±1.18	1.39±0.47 <sup>D</sup>	5.70±0.18 <sup>a</sup>
GOT	110.9±3.4 <sup>a</sup>	101±8.1 <sup>D</sup>	114±3.3 <sup>D</sup>	134.9±5.17 <sup>a</sup>
GPT	70.82±1.9 <sup>a</sup>	65.78±2.9 <sup>D</sup>	50.15±6.17 <sup>D</sup>	71.6±3.03 <sup>a</sup>
ALP	19.85±2.5 <sup>a</sup>	13.28±0.25 <sup>D</sup>	26.11±0.73	24.84±1.26
Creatinin	1.38±0.03	1.29±0.06	1.25±0.04	1.18±0.07
Glucose	112.4±3.39	111.8±4.17	83.67±4.9 <sup>D</sup>	118.2±1.06 <sup>a</sup>

**Table (6): Phagocytic activity of naturally infected fishes:**

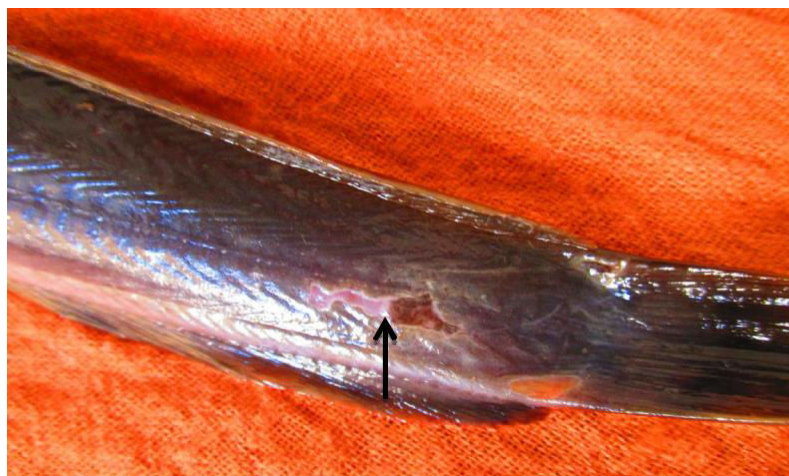
Fish Parameters	<i>Oreochromis niloticus</i>		<i>Clarias gariepinus</i>	
	Infected	Non-infected	Infected	Non-infected
Phagocyte	36.43±0.42	35.56±0.37	39.12±0.29 <sup>a</sup>	36.56±0.48 <sup>D</sup>
Phagocyte Index	1.61±0.05	1.46±0.06	1.93±0.07 <sup>a</sup>	1.45±0.04 <sup>D</sup>



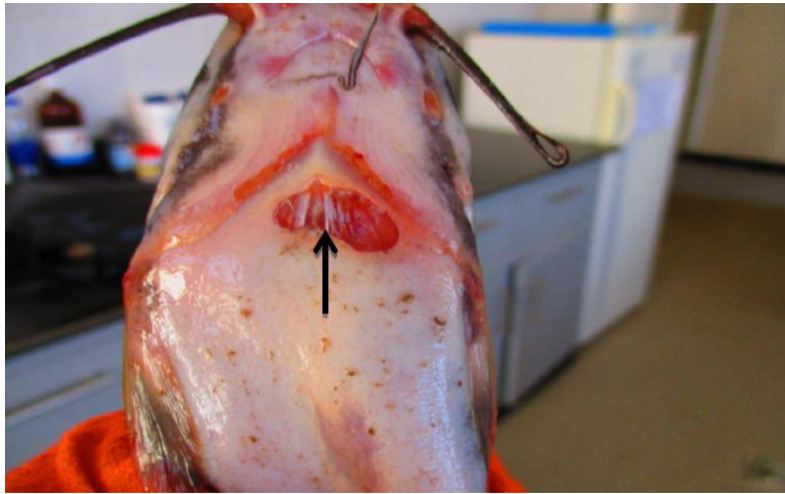
**Figure (1):** Skin of *Oreochromis niloticus* infected with *Gyrodactylus cichilidae* showing scattered ulcerative areas (arrows) in different parts of the skin.



**Figure (2):** *Oreochromis niloticus* infected with *Dactylogyrus cichilidae*, showing severe congestion on the isthmus region and over the two gill covers (Block arrows).



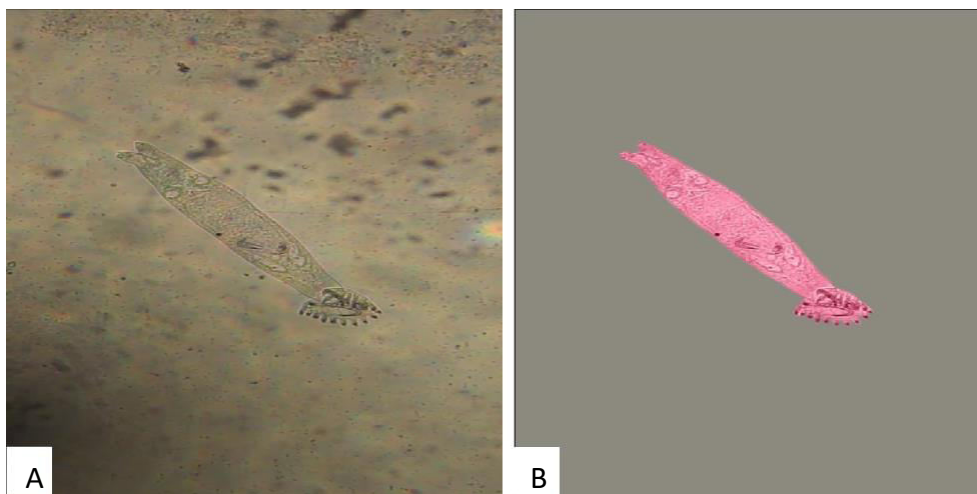
**Figure (3):** Skin of *Clarias gariepinus* infected with *Gyrodactylus claridae* showing erosion of the skin with small wound (arrow).



**Figure (4):** Ventral abdomen of *Clarias gariepinus* infected with *Dactylogyus claridae* showing hemorrhagic ulcer (arrow).



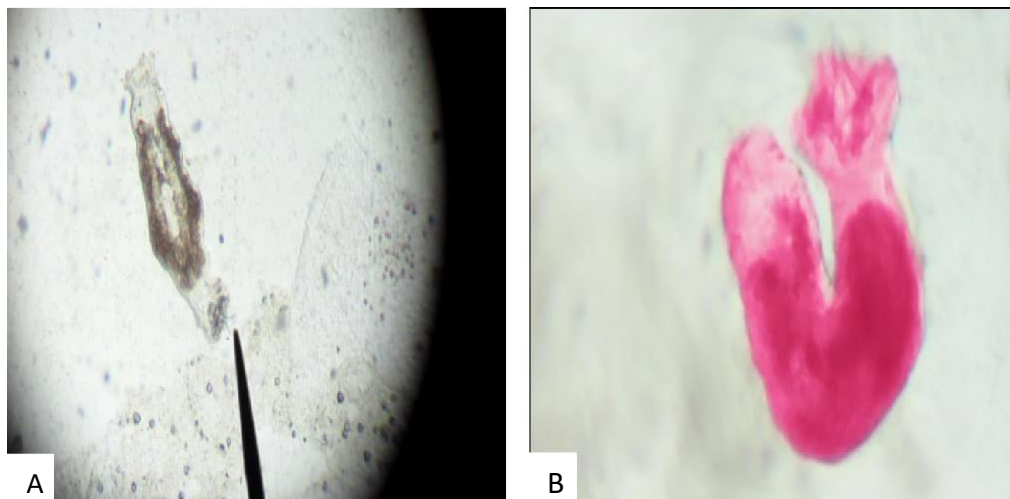
**Figure (5):** Gills of *Clarias gariepinus* infected with *Dactylogyus claridae* showing severe congestion (arrow).



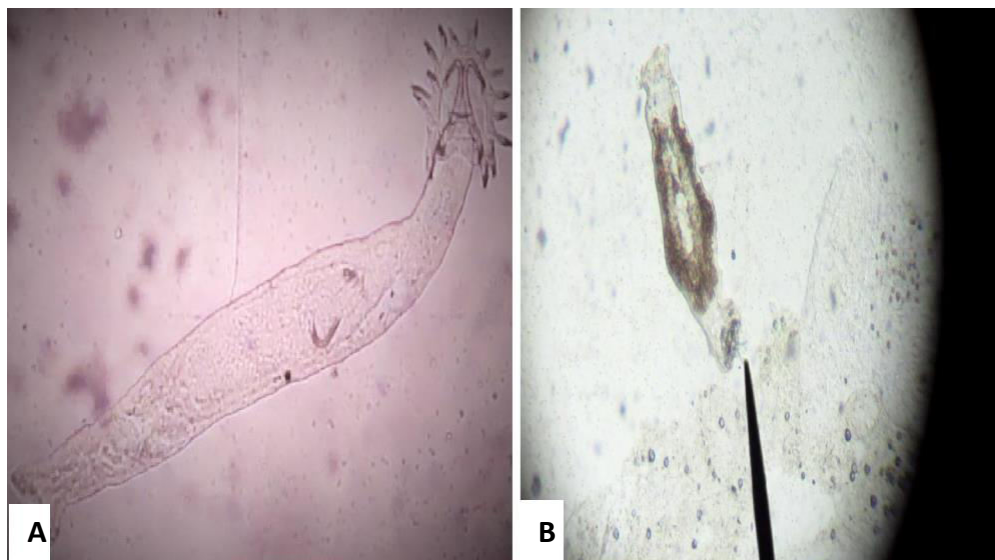
**Figure (6):** A- Wet mount of *Gyrodactylus cichilidae* (X10). B- Stained *Gyrodactylus cichilidae* by acetocarmine stain (X10).



**Figure (7):** A & B Wet mount of *Gyrodactylus claridi* (X10). C- Stained *Gyrodactylus claridae* by acetocarmine stain (X10).

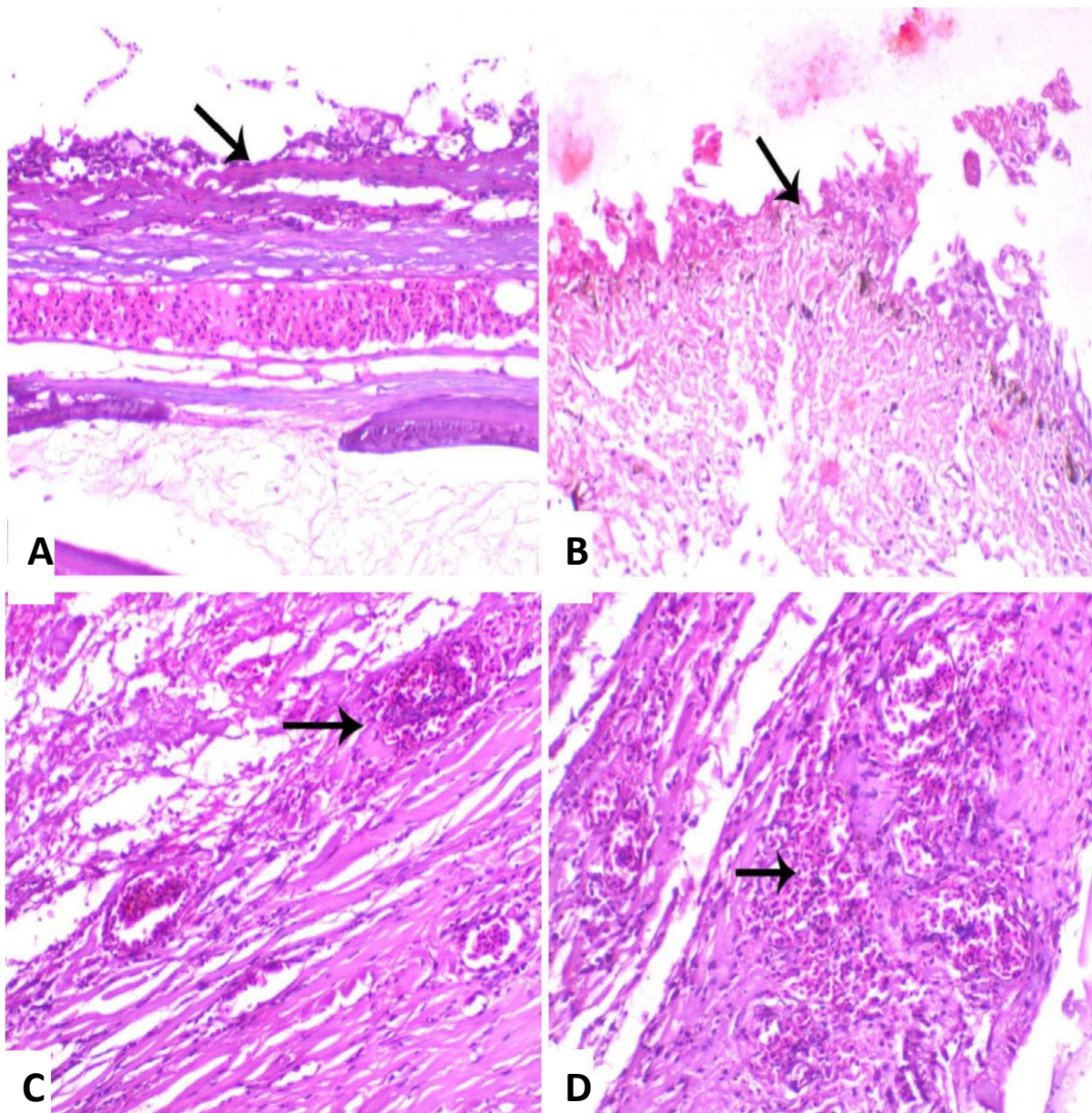


**Figure (8):** A- Wet mount of *Dactylogyrus claridi* (X10). B- Stained *Dactylogyrus claridae* by acetocarmine stain (X10).

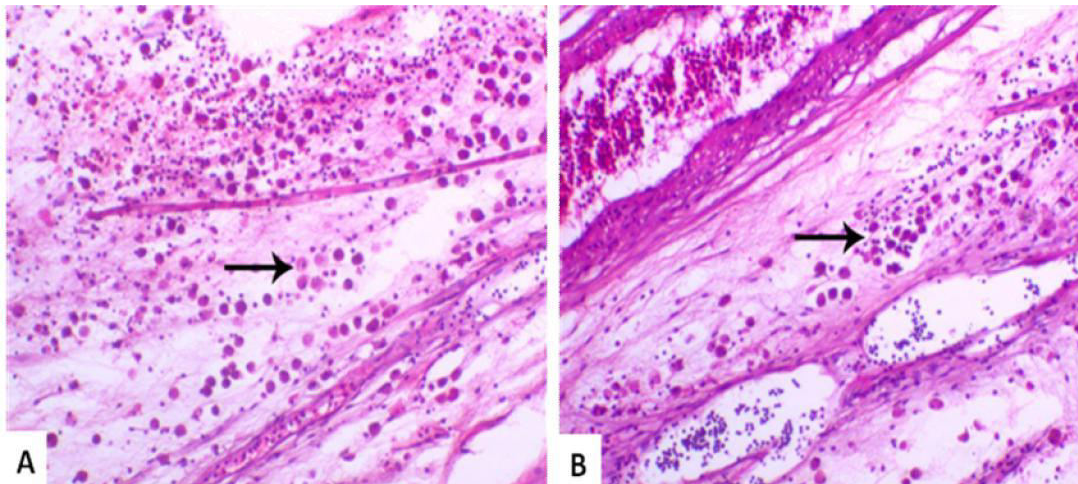


**Figure (9):** A- Wet mount from the skin of *Clarias gariepinus* showing *Gyrodactylus claridi* (X10). B- Wet mount from gills of *Clarias gariepinus* infected with *Dactylogyrus claridi* (X10).

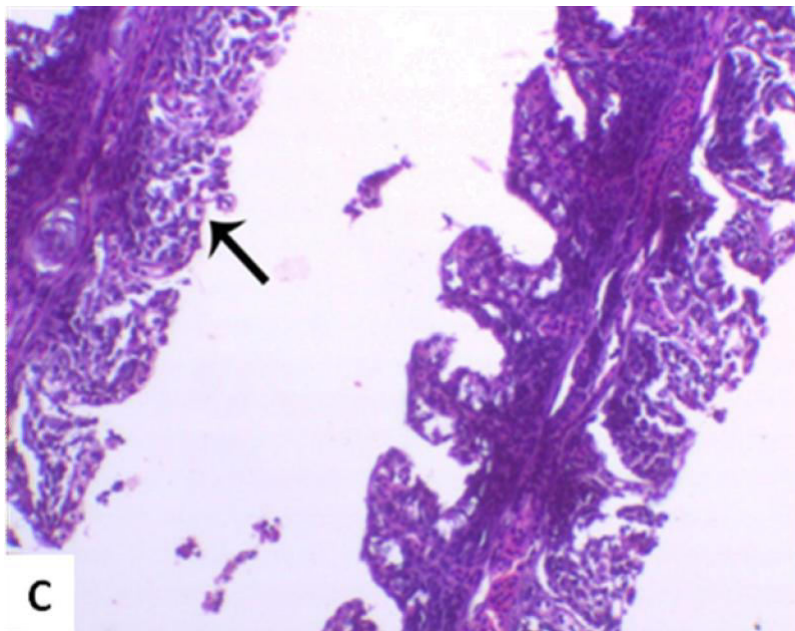




**Figure (10):** Skin of affected African catfish (*Clarias gariepinus*). **A)** showing erosion of the stratified skin layer (arrow) with marked congestion of the subcutaneous blood vessels. **B)** showing ulceration and desquamation of the stratified skin layer (arrow). **C)** showing subcutaneous congestion of the blood vessels (arrow). **D)** showing severe vascular congestion and hemorrhage (arrow) in the subcutaneous tissue of the skin. H & E stain. A X200; B X200; C X200; D X200.



**Figure (11): A-B...** Skin of affected *C. gariepinus*. **A)** showing marked infiltration of eosinophilic granular inflammatory cells infiltration (arrow) associated with oedema. **B)** showing vascular congestion, lymphatic dilatation and peri-lymphatic eosinophilic granular cell infiltration (arrow).



**C)** Gill lamellae of affected *C. gariepinus* showing severe degree of fusion of gill lamellae as a result of marked leucocytic infiltration (arrow). H & E stain. A X200; B X200; C X200; D X200.

#### 4. Discussion

Nowadays, more attention have being paid to improve fish aquaculture in Egypt, in a trial to solve the shortage of animal protein. The present work studied the seasonal incidence of monogenean trematodal diseases among naturally infested *Oreochromis niloticus* and *C. gariepinus* in Kafr El-Sheikh governorate.

Parasitological examination of 400 *O. niloticus* and 400 *C. gariepinus* revealed the presence of monogenean trematodes in 70 and 136 positive infested cases (17.5% & 34%), respectively. The

highest seasonal incidence was recorded in summer (39% and 63% in *O. niloticus* and *C. gariepinus*, respectively). This result totally agree with that reported by **Osman (2005)** and **Noor El Deen (2007)** where the highest prevalence was in summer. These variations in results might be attributed to the physical (Depth, water current and temperature) and chemical (Oxygen, salinities) factors of the environment and to fish species as well (**Paperna (1996)**; **Endraws (2001)**; **El- Tantawy (2001)** and **Diab et al. (2006)**). A very interesting fact could support the results of the current study in which high infestation of

monogeneans occurred in summer, that the high water temperature increases the rate of transmission of *Gyrodactylus* due to increased parasite and/or host activity (Bakke *et al.*, 1991). Despite, stress (high water temperature) was accompanied with decreases of circulating lymphocytes, increase of macrophage cells, and enhanced red blood cell degradation resulting in increases the susceptibility of fish to diseases (Peters and Schwarzen, 1985).

The gross clinical appearance of infested *Oreochromis niloticus* with monogenean *Gyrodactylus sp.* and *Dactylogyus sp.*, demonstrated dark or pale body coloration, detachment of scales in some areas of the body, excessive mucous secretion on the external body surface, ascites, scattered hemorrhagic patches and ulcerative areas in different parts of the skin as well as severe congestion on the isthmus region and over the two gill covers. However, the infested *C. gariepinus* showed emaciation, dark or pale body coloration, excessive mucous secretion on the external body surface, erosion of the skin with some small wounds, hemorrhagic ulcers especially on the ventral abdomen, and severe congestion of the gills. These results were similar to that recorded by (Kuperman and Matey 2000, El-Tantawy 2001, Noor El-Deen *et al.*, 2015 and Gado *et al.*, 2017).

Excessive mucous secretion might be released to relieve the irritating inflammatory reaction caused by continuous irritation of monogenean trematodes (Marzouk, 2002; Khalil, 2010; Noor El-Deen *et al.*, 2015 and Gado *et al.*, 2017).

Concerning the scattered hemorrhagic patches with small wounds or ulcers on the body surface together with darkening of skin of some fishes infected with *Gyrodactylus*; this might be related to that *Gyrodactylus* (skin fluke); is provided with a pair of too long and strong anchors in the opisthaptor and 7 pairs of small strong hooklets used for fixation firmly on the external body surface of its host to resist the external water currents as well as continuous regularly locomotion and relocation from side to side and around the fin margin and frequently cross over the body surface to another fin; the caudal, pectoral and pelvic ones. These results nearly similar to that recorded by Sterud *et al.* (1998); Osman (2005) and Gado *et al.* (2017).

Infested fishes appeared exhausted and/or asphyxiated; this might be attributed to low oxygen intake resulting from destructed gill epithelium, which caused by feeding activity, attachment, fixation and locomotion of monogenea causing massive destruction of the respiratory epithelial cells which may be similar to that reported by Eissa *et al.* (2010); Noor El-Deen *et al.* (2015) and Gado *et al.* (2017).

Investigated congested gills may be attributed to destruction of the efferent vessels by monogenea;

where the blood pressure is low causing extensive hemorrhage and clotting of blood leading to rapid occlusion of the vessel, ischemia and necrosis in some areas; which may progress into pale gills giving the Marbling appearance (Eissa, 2006; Noor El-Deen *et al.*, 2015 and Gado *et al.*, 2017).

Microscopic smears taken from skin, gills, eye and mouth of examined *O. niloticus* and *C. gariepinus* revealed the presence of both *Dactylogyus sp.* and *Gyrodactylus sp.* over skin and gills.

The adult worms isolated from the skin and gills (monogenean trematodes; *Gyrodactylus sp.* and *Dactylogyus sp.*), of infested *Oreochromis niloticus*; were morphologically and parasitologically described and were nearly similar to the descriptions given by Yamaguti (1963); El-Asely *et al.*, (2015) and Noor El-Deen *et al.*, (2015).

However, monogenean trematodes; (*Gyrodactylus sp.* and *Dactylogyus sp.*), isolated from infested *O. niloticus* and *C. gariepinus*; were morphologically and parasitologically described and were nearly similar to the descriptions given by Yamaguti (1963); Abo Esa *et al.* (2008); Abd El-Maged (2009); Abd El-Latif *et al.*, (2009); Abou Zaid (2011); Hamouda (2014) and Gado *et al.* (2017).

The effect of different monogenetic trematodes on the infested *O. niloticus* and *C. gariepinus* on different hematological parameters are recorded. Although Azevdo *et al.* (2006) stated that the total number of erythrocytes, leucocytes haven't relation with the ectoparasites infections, the results in the current study revealed significant decrease in Hb amount, non-significant decrease in WBCs count and non-significant increase in RBCs count; the results which disagree with that recorded by Murad and Mostafa (1988); Tavares-Dias *et al.*, (2002) and Ibtisam (2004) where they reported lower erythrocytic, hemoglobin levels and a higher leucocytic count especially in catfish. The recorded results of differential leucocytic count was partially agree with Murad & Mostafa (1998) and El-Seify *et al.* (2003).

Recognizing the effect of different ectoparasites either Monogeneans (*Dactylogyus* & *Gyrodactylus*) on blood serum components of *O. niloticus* and *Clarias gariepinus*; lower level of total serum protein was recorded in all infected cases; albumin amount was decreased in catfish only and globulins was increased in all infected cases.

The blood serum Aspartate aminotransferase (AST), Alanine aminotransferase (ALT) Alkaline phosphatase (ALP) enzymes activities and Creatinine values were elevated in the infested *O. niloticus* but, decreased in all cases of infested *C. gariepinus* with external parasites than the non infected fishes; this indicate that the external parasites stimulated the

activities of ALT, AST and ALP liver enzymes as well as Creatinine. This agree with **Younis, (1999)** and **(Adhams, 2002)** which recorded that aspartate aminotransferase (AST) and alanine aminotransferase (ALT) showed significant increase in *O. niloticus* infected with monogenetic trematodes. This may be due to hepatic cells injury or increased synthesis of the enzymes by the liver (**Yang and Chen, 2003**). Also these findings may be attributed to the inflammatory reactions and intoxications produced by the parasite in the affected fish.

Results of phagocytic activity and index are summarized in table (6). Both phagocyte activity and Phagocyte index was increased in both *Oreochromis niloticus* and *Clarias gariepinus*. This increase in the PA and PI values were also reported by **Stosik (2002)**, **Kollner et al. (2004)**, **Tavares-Dias et al., (2007)** and **Rashed (2013)**. Also, **Tavares-Dias et al., (2002)** mentioned that Ichthyophthiriasis in Nile tilapia showed an increase in phagocytic activity. **Coles (1986)** stated that the increasing phagocytic activity was attributed to the increasing lymphocytic numbers.

Histopathological alterations of the skin and gills of the infested *O. niloticus* and *C. gariepinus* was recorded and the results are similar to that recorded by **Aly et al. (1998)**. The skin and gill damage might be induced by feeding activity, attachment, fixation and locomotion of monogeneans causing massive destruction of the respiratory epithelial cells and/or cutaneous cells. These results agreed with **Abd El-Hady (1998)**; **Noor El-Deen (2007)** **Roberts (2012)** and **Gado et al., (2017)**.

### Conclusion

From the current study, it can be concluded that *Dactylogyrus sp.* have the highest rate of the infestation (17.5% and 34%) in *O. niloticus* and *C. gariepinus*, respectively. While, *Gyrodactylus sp.* infection was found to be a mixed infection in all cases with *Dactylogyrus sp.*; showing a prevalence rate (0.25% and 2.25%) in *O. niloticus* and *C. gariepinus*, respectively. The highest seasonal incidence was recorded in summer (39% and 63% in *O. niloticus* and *C. gariepinus*, respectively. Total serum proteins, liver enzymes and creatinine were increased due to the inflammatory reaction induced by monogenean infestation. Phagocytic activity and phagocytic index was increased due to increased lymphocytic number.

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