

***Ergasilus luteusi* Al-Sahlany, Adday et Ali, 2024 (CYCLOPOIDA: ERGASILIDAE) parasite of two fishes and effect of length groups, gender and season in Al-Gharraf River, Southern Iraq**

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Abstract

The copepod *Ergasilus* Nordman, 1832 common ectoparasite of both freshwater and marine fishes; however the present work aim to study ecological aspect of new parasite *Ergasilus luteusi* Al-Sahlany, Adday et Ali, 2024 parasite of two freshwater cyprinid (*Planiliza abu* and *Carasobarbus lutues*) occurred in Al-Gharraf River, Thi Qar Province, Southern Iraq. The seasonal prevalence by *E. luteusi* with *C. luteus* differ little from that from *P. abu*; while the higher prevalence with the crustacean is found in the median group length of both fish species. The results are discussed according to the variation in environmental factor between seasons, physiological aspect between males and females and the correspondence between second antenna of the parasite and size of gill filaments.

Keyword: Ecology, Ectoparasite, Fish, Fresh water, gills, Thi Qar.

I. Introduction

The Gharraf River, the main branch of the Tigris River, is located in southern Iraq. It flows from the right side of the Tigris River at the Kut dam into the Euphrates Basin, passing through the provinces of Wasit and Dhi Qar, and ends in the Hammar Marsh northern Nasiriyah. The river passes through several cities, with the most important being Kut, Al-Muwafaqiyah, Al-Hayy, Al-Bashair, and many green villages with high human population in Wasit Province. It also flows through Dhi Qar Province, passing by the areas of Al-Fajr, Qalat Sukkar, Al-Rifai, Al-Nasr, and then to the Al-Bida'a area (Iraqi Ministry of Water Resources, 2006).

Iraq is considered one of the countries rich in water bodies, which include rivers, lakes, reservoirs, marshes, streams, and ponds that can be utilized for fish production (Coad, 2010). The presence or abundance of parasites can reflect the environmental condition, as ectoparasites with direct life cycles, such as ciliated parasites, prefer environments with high bacterial content; in contrast, endoparasites with complex life cycles prefer stable and unpolluted waters (Modu *et al.*, 2016). Host individuals are occupy different environments or may change their feeding habits depending on age, gender, or reproductive status, leading to variations in their exposure to parasitic infective stages (Nagy and Postawa, 2016).

Ergasilus luteusi very recently described as new species from Yellow barbel *Carassobarbus luteus* (Heckel) and Abu mullet *Planiliza abu* (Heckel) from Al-Gharraf River, Thi Qar Province (Al-Sahlany *et al.*, 2024). Some studies have been conducted in Iraq on the ecology of fish parasites, focusing on the impact of parasitic infestation on fishes and its relationship with the season. Some studies has been carried out on monogenean parasites in northern Iraq (Abdullah, 2007), as well as in central Iraq, of Baghdad (Al-Nasiri, 2000; Mansoor, 2009). Others crustacean parasites were conducted in central Iraq, Baghdad (Mansoor, 2009), northern Iraq, Erbil (Al-Marjan, 2016), and southern Iraq at Basrah (Khamees and Mhaisen, 2001; Eassa *et al.*, 2014).

Studies on acanthocephalan worms in southern Iraq, specifically in Dhi Qar Province, were conducted by Al-Kinnany and Al-Ubaydi (2017), while Al-Nasiri (2000) addressed several species of myxozoans and acanthocephalans in central Iraq, Baghdad Province.

Studies related to the relationship between parasitic infestation by monogenoids and fish length and sex were conducted in northern Iraq (Bashê and Abdullah, 2010b; Abdullah and Nasraddin, 2020) and in central Iraq (Al-Saadi, 2007). Research on parasitic crustaceans infesting the gills of abu mullet *P. abu* in southern Iraq, Basrah (Khamees and Mhaisen, 2002), and northern Iraq (Abdullah and Mhaisen, 2003) has also been conducted. Acanthocephalan studies were carried out in northern Iraq (Abdullah and Ali, 1999; Abdullah and Mhaisen, 2007a), central Iraq at Salah al-Din Province (Al-Ayash, 2011; Saleh, 2016), and Karbala Province (Taher *et al.*, 2009).

Similarly, studies on tapeworms were conducted in northern Iraq, Nineveh Province (Al-Niaeemi, 2011), and on the nematode larvae *Contraceacum* sp. in northern Iraq (Abdullah and Mhaisen, 2011c). Manhal (2016) examined the relationship between infestation by the eye parasite *Diplostomum* spp. and the length and weight groups of *C. luteus* in Basrah, southern Iraq. The newly described *Ergasilus* from two freshwater fishes in neglected area in regard to parasitology approach (Al-Gharraf River); led to encourage us to study the ecological aspects of this parasite, seasonal variations and the effect of gender, group length of the hosts.

II. MATERIALS AND METHODS

During 13 months (December 2022 till December 2023) of parasitological examination of 24 fishes species were caught from Al-Gharraf River 46°17'45''-46°11'64''E and 31°44'60''-31°58' 07''. Only two species of freshwater fishes represent with 436 specimens of Yellow barbel *Carassobarbus luteus* (Heckel) and 884 specimens Abu mullet *Planiliza abu* (Heckel) were infested with *Ergasilus luteusi*, which selected for designed the ecological fish parasite study. Coad (2010) and Froese and Pauly (2024) followed for identified the fishes and up-to-date the taxonomy respectively. The ecological terms (Percentage of Prevalence and mean of intensity) were calculated according to Bush *et al.* (1997).

III. RESULTS AND DISCUSSION

III.I. The relationship between parasitic infestation *Ergasilus luteusi* and the length of *P. abu*

The results showed that the number of 219 *Planiliza abu* infested by of *Ergasilus luteusi*, with a prevalence of 24.8%. The parasite infestation was studied throughout the seasons, and the examined fishes were classified according to their length groups during the study period as follows:

The prevalence infestation by *E. luteusi* between 4-2-5.3% at only the middle length groups during winter (Table 1). The highest prevalence (5.3) is recorded during winter at the 151-200 mm length group, with an infestation intensity of 3, while smaller and longer length groups were not infested (Table 1). The statistical analysis of variance by using LSD among means of length groups, indicated a significant differences only between the second and third groups in prevalence and mean of intensity.

Table 1: The prevalence and mean of intensity of different length groups of *Planiliza abu* by the *Ergasilus luteusi* during winter.

| Length group (mm) | Number of examined fish | Number of infested fish | Prevalence % | Number of parasites | Mean of intensity |
|-------------------|-------------------------|-------------------------|------------------|---------------------|-------------------|
| >100 | 1 | 0 | ^a *0 | 0 | ^a 0 |
| 100 -150 | 96 | 4 | ^b 4.2 | 7 | ^b 1.75 |
| 151 -200 | 19 | 1 | ^c 5.3 | 3 | ^c 3 |
| 201 -250 | 1 | 0 | 0 ^a | 0 | ^a 0 |

*Different letters indicate significant differences between the means of the different treatments.

In the spring season, **infestation** was observed in three length groups only. The highest prevalence and mean of intensity of infestation 58.3% and 2.7 was recorded at 151-200 mm length group respectively. No differences were detected in the infestation in other groups (Table 2). The least significant difference (LSD) analysis indicated a significant differences was found in longest length group only concerning the prevalence. While, there were no significant differences among all three length groups in mean of intensity.

Table 2: The prevalence and mean of intensity of different length groups of *Planiliza abu* by *Ergasilus luteusi* during spring.

| Length group (mm) | Number of examined fish | Number of infested fish | Prevalence % | Number of parasites | Mean of Intensity |
|-------------------|-------------------------|-------------------------|-------------------|---------------------|-------------------|
| >100 | 8 | 2 | ^a *25 | 4 | ^a 2 |
| 100 -150 | 124 | 34 | ^a 27.4 | 69 | ^a 2.02 |
| 151 -200 | 12 | 7 | ^b 58.3 | 19 | ^a 2.7 |

*Different letters indicate a significant differences between the means of the different treatments.

At summer season, the prevalence of infestation was recorded in three length groups, with very similar prevalence (21.2% to 22.2%) and an infestation intensity between 2 and 4.6 (Table 3). The analysis of variance table showed no significant differences between any of the length groups in the prevalence of infestation, and a significant difference was observed in first length group regarding to mean of intensity.

Table 3: The prevalence and mean of intensity of different length groups of *Planiliza abu* by *Ergasilus luteusi* during the summer.

| Length group (mm) | Number of examined fish | Number of infested fish | Prevalence % | Number of parasites | Mean of intensity |
|-------------------|-------------------------|-------------------------|-------------------|---------------------|-------------------|
| >100 | 19 | 02 | ^a 121. | 91 | ^{b*} 64. |
| 150 -100 | 025 | 53 | ^a 21.2 | 155 | 2.9 ^a |
| 200 -151 | 9 | 2 | ^a 22.2 | 4 | ^a 2 |

*Different letters indicate significant differences between the means of the different treatments

At autumn season, the prevalence of infestation was recorded in three length groups, with very similar infestation prevalence (34.1% to 36.2%) and an infestation intensity between 1.7 and 3.32 (Table 4). However there are a significant differences between the three length groups in relation to both the prevalence and mean intensity of infestation.

Table 4: The prevalence and mean of intensity of different length groups of *Planiliza abu* by the *Ergasilus luteusi* during autumn.

| Length group (mm) | Number of examined fish | Number of infested fish | Prevalence % | Number of parasites | Mean of intensity |
|-------------------|-------------------------|-------------------------|--------------------|---------------------|-------------------|
| >100 | 69 | 25 | ^{c*} 36.2 | 84 | ^c 33. |
| 150 -100 | 203 | 71 | ^b 134. | 121 | 1.70 ^b |
| 200 -151 | 1 | 0 | ^a 0 | 0 | ^a 0 |

*Different letters indicate significant differences between the means of the different treatments

The current study demonstrated the relationship between fish length and prevalence of infestation across the seasons. It was found that fish of intermediate lengths were more susceptible to infestation compared to smaller fish. In other words, smaller fish were relatively less susceptible to infestation than other length groups, and the prevalence increased with the length increasing (Oniye *et al.*, 2004; Akinsanya and Otubanjo, 2006).

The middle length of the host is typically for most parasitic crustaceans (Timi and Lanfranchi, 2005). This may be due to the fact that middle lengths possess gill filaments that are suitable for the shape of the second antennae, which crustaceans used for an attachment. In smaller fish, the gill filaments may be too small for adult crustaceans to attachment, while as fish grow to larger sizes, the filaments may become too suitable for the parasites to establish in safe mode (Timi and Lanfranchi, 2005).

Regarding the variation in prevalence with the seasons, the decrease in infestation during winter was largely influenced by temperature, which had the greatest effect on infestation prevalence in this season. The drop in temperatures during winter leads to reduced activity of microorganisms and parasites due to a decrease in metabolic and enzymatic processes when water temperatures decline (Weiner, 2000). On the other hand, the efficiency of the host's immune system it's most important defense mechanism against parasitic infestation greatly depends on temperature in ectothermic organisms, including the fishes (Magnadóttir, 2006). However, there are some exceptions; for instance, the immune system in three-spined sticklebacks (*Gasterosteus aculeatus* L.) is more active at relatively low temperatures compared to higher temperatures (Dittmar *et al.*, 2014; Franke *et al.*, 2017).

The mean intensity of infestation increases from spring to summer and autumn, while it decreases in winter, indicating that a new generation of crustaceans is released in spring (Retief *et al.*, 2007; Morales-Serna *et al.*, 2024). Several researchers have noted that the prevalence of infestation in some species of *Ergasilus* is seasonally higher in spring, summer, and autumn compared to winter (Aladetohun *et al.*, 2013; Öztürk, 2013). Some male crustacean males, such as *Ergasilus luteusi*, have free living, while only the females parasitizing the fishes (Vasconcelos and Tavares-Dias, 2016). This associated with findings from other studies that showed an increase in zooplankton abundance in spring compared with other seasons (Abbas *et al.*, 2014). The maximum density of the zooplankton community occurs in spring and summer, while lower densities are observed in winter and autumn. Spring is considered the most favorable period for the growth and reproduction of zooplankton species (Ajeel *et al.*, 2015). The larval stage (copepodites) of the *Ergasilus* is found abundantly in the water, and due to its motile behavior, it is conclude that fish become infested with adult *Ergasilus* when contact with final larval stage (Aleksiev *et al.*, 2021). The key factors contributing to the spread of parasitic **infestation** contain host-related factors and environmental factors e.g. the species and number of parasites, the host's behavior, age, sex, nutrition, and both biotic and abiotic factors (Alasadiy, 2018; Bdair, 2018).

III.II Relationship between parasitic infestation and fish gender and the seasons for *P. abu*

The highest prevalence was observed in *P. abu*, males reaching 57.62% during autumn, compared to 38.27% in females during spring. The lowest infestation prevalence for males was 15.38% during winter, while it was 2.88% for females during winter. In the summer the prevalence in both genders are similar. Statistical analysis showed that there were no significant differences in the prevalence of infestation between the genders in the summer and autumn, while significant differences were found in prevalence of infestation in the winter and spring. There were also significant differences in the prevalence of infestation between the different seasons in both males and females, and there was a similar gradual increase in prevalence of infestation between the genders, starting in winter and ending in fall (Table 5).

Table 5: Number of fish, gender, and its relationship with infestation by *Ergasilus luteusi* on *Planiliza abu* in relation to seasons.

| Seasons | Number of examined fish | No. Males | No. Females | No. infested fish | No. infested males | % Prevalence for males | No. infested females | % Prevalence for females |
|---------|-------------------------|-----------|-------------|-------------------|--------------------|------------------------|----------------------|--------------------------|
| Winter | 117 | 13 | 104 | 5 | 2 | 15.38 ^{aB} | 3 | 2.88 ^{aA*} |
| Spring | 144 | 63 | 81 | 43 | 12 | 19.04 ^{bB} | 31 | 38.27 ^{bA} |
| Summer | 350 | 103 | 247 | 75 | 23 | 22.33 ^{cC} | 52 | 21.05 ^{cC} |
| Autumn | 273 | 59 | 214 | 96 | 34 | 57.62 ^{dD} | 62 | 28.97 ^{dD} |

*Different lowercase letters indicate a significant differences between the means of the different treatments within each column, while uppercase letters indicate a significant differences between the means of the treatments within each row.

Generally the idea that females utilized more energy in reproduction by producing greater gametes than the males; consequently, the females become more susceptible to parasitic **infestation** due to the greater stress, as previously suggested in various host-parasite systems (Deerenberg *et al.*, 1997; Nordling *et al.*, 1998; Sanz *et al.*, 2001); This idea clearly reflect on the prevalence in female higher than



that of male during spring. As well as the imbalance in the numbers of each gender during the season might negatively effects on the total prevalence in each season (only 13 males vs 104 females in winter; 103 vs 247 in summer and 59 vs 214 in autumn), This idea clearly reflect the higher prevalence than that of female in winter.

The host's gender is one of the important factors in the interactions between the host and the parasite (Lizama *et al.*, 2005). No variation in the prevalence concerning the host's gender may be probably due to the similarities behavior and size between males and females (Diniz *et al.*, 2022)

The prevalence increase from winter, spring, summer and autumn when reach to maximum value 57.62 in males and 28.97 in female. This increase in prevalence is likely related to the zooplankton blooming in the autumn, which means that males of the genus *Ergasilus* are available in greater numbers, mating, and the presence of more mature females.

III.III-The relationship between *Ergasilus luteusi* infestation and the length of *C. luteus* during the seasons

The results showed that the number of *C. luteus* infested with *Ergasilus luteusi* reached 436 (7.34%) of all examined fish. The parasitic infestation according to the seasons are categorized into length groups, *C. luteus* were free from infestation by *Ergasilus luteusi*, during winter, despite examined 71 fish specimens belonging to four length groups.

At spring, prevalence was observed within three length groups, with the highest prevalence recorded in the largest group (201-250 mm) at 18.2%, with an intensity of infestation of 1.5. The prevalence and mean of intensity in the other two length groups are similar. No infestations were recorded in the smallest length group (<100 mm, see Table 6). The statistical analysis indicated a significant differences (0.05) in the prevalence and intensity at three length groups.

Table 6: The prevalence and mean of intensity of different length groups of *Carasobarbus luteus* by *Ergasilus luteusi* during spring.

| Length group (mm) | No. examined fish | No. infested Fish | Prevalence % | Number of parasites | Mean of intensity |
|-------------------|-------------------|-------------------|-------------------|---------------------|-------------------|
| >100 | 0 | 0 | ^a 0 | 0 | ^a 0 |
| 150 -100 | 66 | 1 | ^b 1.5 | 3 | ^b 3 |
| 200 -151 | 44 | 3 | ^c 6.8 | 7 | ^c 2.3 |
| 250 -201 | 22 | 4 | ^d 18.2 | 6 | ^d 1.5 |

*Different letters indicate a significant differences between the means of the different treatments.

During the summer, there was a clear variation in prevalence of infestation among different length groups, with the highest prevalence of 40% recorded in the largest length group (201-250 mm). In contrast, mean of intensity have similar value in different length group (Table 7). Statistical analysis indicated a significant differences (0.05) in the prevalence among the length groups, while no significant differences were observed in the mean of intensity among the length groups during the summer.

Table 7: Changes in prevalence of different length groups of *C. luteus* by *Ergasilus luteusi* during summer.

| Length group (mm) | No. examined fish | No. infested fish | Prevalence % | Number of parasites | Mean of intensity |
|-------------------|-------------------|-------------------|------------------|---------------------|-------------------|
| >100 | 1 | 0 | ^a 0 | 0 | ^a 0 |
| 150 -100 | 80 | 2 | ^b 2.5 | 4 | ^a 2 |
| 200 -151 | 44 | 11 | ^c 25 | 32 | ^a 2.9 |
| 250 -201 | 5 | 2 | ^d 40 | 3 | ^a 1.5 |

*Different letters indicate significant differences between the means of the different treatments.

During the autumn, prevalence varied significantly among the length groups of *C. luteus* infested *E. luteusi*. The highest prevalence is recorded (35%) in the length group of 151-200 mm. The other length group showed prevalence between 1.6% and 12.5%, with mean of similar value in the mean of intensity. No infestations were recorded in the smallest length group (Table 8). The statistical analysis indicated significant differences (0.05) in both prevalence and mean of intensity among different length groups.

Table 8: Changes in the infestation of different length groups of *Carasobarbus luteus* by *Ergasilus luteusi* during autumn.

| Length group (mm) | No examined fish | No infested fish | Prevalence % | Number of parasites | Mean of intensity |
|-------------------|------------------|------------------|-------------------|---------------------|-------------------|
| >100 | 2 | 0 | ^a 0 | 0 | ^a 0 |
| 150 -100 | 64 | 1 | ^b 1.6 | 3 | ^c 3 |
| 200 -151 | 20 | 7 | ^d 35 | 10 | ^b 1.4 |
| 250 -201 | 8 | 1 | ^c 12.5 | 4 | ^d 4 |

*Different letters indicate significant differences between the means of the different treatments

The current study showed a relationship between the length of *C. luteus* and prevalence of infested fish during the seasons. The nature of the infestation is influenced by length, and it was found that larger fish are more susceptible to infestation compared to smaller fish. Specifically, larger fish had a higher prevalence than smaller-sized fish. Smaller fish were less susceptible to infestation than other length groups. Several studies have provided that the number of parasites per host increases with the length of the fish (Isaac *et al.*, 2000; Guidelli *et al.*, 2003; Poulin and Morand, 2004). The size of the host is an important factor that determines the richness and abundance of parasites because the larger fish providing the surface area, the greater space available to acquire new parasites (Poulin *et al.*, 2011). The positive relationship between the infestation and host length may be attributed to the factor of the fish's age (Tekin-Özan *et al.*, 2008).

The prevalence in summer, spring, and autumn was higher than in winter, during which the infestation completely disappeared. The prevalence was higher in the dry season than in the wet season, and the responsible factor for this is over-nutrition, which often increases the parasitism, as the accompanying increase in productivity enhances the abundance of zooplanktons, including microcrustaceans, along with *Ergasilus* (Laffery and Kuris, 1999). Furthermore, the general effects of increased temperature may impacts on the parasite majurity (embryonic development and faster hatching), as well as effects on the host behavior (nutrition shifting and reduced the host resistance) and transmission e.g. early reproduction in spring, more new generations throughout the seasons, and the likelihood of transmission throughout the year (Marcogliese, 2001). This is attributed not only to the direct effects of temperature on the metabolic processes of the parasites, but also to the developed the immune activity of the hosts (Morley and Lewis, 2014). The effect of low

temperatures on parasite growth in ectothermic hosts is due to reduced metabolic activity (Wharton, 1999).

6-The relationship between parasitic infestation and fish gender throughout the seasons in *C. luteus* infested with *Ergasilus luteusi*

The results of the statistical analysis indicated a significant differences among the seasons and also between the prevalence between males and females. The results indicate the prevalence was low in all seasons except in the fish males during summer. There are no significant differences were found between female and male of *C. luteus* during spring and autumn; and only significant differences were found in prevalence of *E. luteusi* of the males during summer.

Table 9: Shows the number of fish, gender ratio, and its relationship with the infestation of the crustacean *Ergasilus luteusi* on *Carasobarbus luteus* according to the seasons.

| Seasons | NO examined Fish | No. Males | No. Females | No. Infested Fish | No. Infested males | % Prevalence for males | No. Infested females | % Prevalence Females |
|---------|------------------|-----------|-------------|-------------------|--------------------|------------------------|----------------------|----------------------|
| Winter | 78 | 37 | 41 | 0 | 0 | 0 ^{aA} | 0 | 0 ^{aA} |
| Spring | 134 | 35 | 99 | 8 | 2 | 5.7 ^{aA} | 6 | 6 ^{aA} |
| Summer | 130 | 23 | 107 | 15 | 4 | 47.82 ^{bB} | 11 | 10.28 ^{aA*} |
| Autumn | 94 | 29 | 65 | 9 | 1 | 3.45 ^{aA} | 8 | 12.3A ^a |

*Different lowercase letters indicate significant differences between the means of the different treatments within each column, while uppercase letters indicate significant differences between the means of the treatments within each row

The reason for increase the prevalence in summer might to idea that Ergasildae individuals begin to breeding during the spring and gradually increase until they reach their peak during the summer, then back to decrease in the autumn during end the life span, these strategy clearly reflect the increase of prevalence (Morales-Serna *et al.*, 2024) or related to dry season compared with wet season, when due to the copepod are directly linked to water temperature, and the high temperature supplied serious egg producing (de Santos *et al.*, 2017).

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CONFLICT OF INTEREST

To the knowledge of the investigator, there are no conflicts of interest related to the publishing of the whole study.

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