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Seasonal Prevalence and Host-Related Patterns of *Neoechinorhynchus iraqensis* Infection in *Planiliza abu* and *Silurus triostegus* from Al-Gharraf River, Wasit Province, Iraq

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Abstract

The present study aimed to determine the seasonal prevalence and mean intensity of the acanthocephalan parasite *Neoechinorhynchus iraqensis* of two freshwater fish species Planiliza abu and Silurus triostegus from Al-Gharraf River, Wasit Province, Iraq. Fish were sampled monthly from January to December 2025 from three fixed stations along the river. In total, 720 fish, consisting of 360 P. abu and 360 S. triostegus were examined. The parasites were recovered from the intestine and relaxed in chilled distilled water, fixed in 70% ethanol, stained with Semichon's aceto-carmine, cleared, mounted and identified by reference to the standard morphological characters. Overall prevalence was 19.17% (138/720). Infection was significantly higher in P. abu (24.72%; 89/360) than in S. triostegus (13.61%; 49/360) (chi-square = 14.34, df = 1, $p < 0.001$). The prevalence was highest in summer, lowest in winter, and higher in spring. P. abu had the highest prevalence in June (53.33%) while S. triostegus had the highest prevalence in the month of June-July (30.00%). The monthly prevalence of P. abu and S. triostegus were strongly and positively correlated with water temperature ($r = 0.900$, $r = 0.883$, respectively and $p< 0.001$ for both). The results showed that host feeding ecology and the changes of thermal conditions during the seasons could be important factors in the transmission of N. iraqensis in the Al-Gharraf River system. The study gives new epidemiological data for Iraqi freshwater fish parasitology and gives a support to the integrated monitoring of fish parasites, fish ecology and environmental variables.

Keywords

Acanthocephala;
Al-Gharraf River;
Fish Parasites;
Neoechinorhynchus iraqensis;
Planiliza abu;
Silurus triostegus;
Seasonal Prevalence

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1. Introduction

Freshwater fish parasites are key elements in aquatic ecosystems, showing feeding patterns of their hosts, trophozoology, environmental quality, and seasonality of transmission. As long as the definitions are provided and applied in the same way, infection parameters like prevalence and mean intensity are frequently used in ecological parasitology to describe host-parasite relationships (Bush et al., 1997). Parasite communities may also be sensitive to environmental disturbance and the structure of the food-webs and are thus appropriate indicators in aquatic ecosystem assessment (Marcogliese, 2005; Sures et al., 2017).

Armed, retractable proboscis which penetrates the wall of the definitive host's intestine is the special feature of intestinal helminths, the Acanthocephalans. They typically demand an arthropod intermediate host and a vertebrate definitive host and transmission usually takes place when an intermediate host is ingested by a fish or other vertebrate definitive host (Kennedy, 2006; Taraschewski, 2000). Infective cystacanth development in intermediate hosts and feeding habits of definitive hosts are therefore key considerations in the understanding of seasonal and host related infection patterns.

This species was initially reported from the freshwater mullet Liza abu which in many taxonomic works is called Liza Planiliza in Iraq (Amin et al., 2001). In the subsequent Iraqi records, it was revealed that species of Neoechinorhynchus are distributed among freshwater and fish associated with the water of the Tigris-Euphrates and its tributaries, and marshes and drainage systems, as well as fishpond systems distributed widely in the Tigris-Euphrates basin (Mhaisen & Abdul-Ameer, 2024). More recently, molecular research has confirmed the presence of N. iraqensis in Iraqi freshwater fish, such as P. abu and S. triostegus (Ali & Abdullah, 2024; Gaber & Awad, 2025).

Ecology and feeding behaviors of the two host species studied are very different. Planiliza abu is a freshwater and brackish-water mullet found in Iraq and neighbouring areas of the Tigris-Euphrates system (Çiçek et al. 2023; Froese & Pauly, 2025a). The dietary studies from southern Iraq revealed that P. abu feed on diatoms, algae, detritus, higher plants, fish eggs and small zooplankton, which is representative of its non-specialized benthic and herbivorous-detritivorous feeding habit (Mohamed & Abood, 2019). S. triostegus is a big demersal catfish of the Mesopotamian basin, known to occur in rivers and marshes of Iraqi rivers (Froese & Pauly, 2025b; Jawad & Al-Janabi, 2016). These environmental differences could lead to exposure to infected intermediate hosts being different, and exposure is closely associated with feeding pathways for acanthocephalan transmission.

Another important factor which may affect host-parasite dynamics in aquatic environments is temperature. Climate change can impact parasite development, intermediate host abundance, feeding habits of fish and the distribution of host and parasite (Marcogliese, 2016). In warmer river systems, like those of southern and central Iraq, therefore, seasonal differences in temperature may be a factor in explaining the temporal differences in parasite prevalence or intensity. The local information on seasonal dynamics of N. iraqensis in Al-Gharraf River is still scant, especially in the comparison between two different trophic strategy hosts.

The objectives of the present study were to estimate the monthly and seasonal prevalence and mean intensity of N. iraqensis in P. abu and S. triostegus collected from Al-Gharraf River in 2025; compare the infection prevalence and mean intensity of N. iraqensis in P. abu and S. triostegus; investigate the relationship between water temperature and monthly prevalence of N. iraqensis, record the main laboratory operations for recovery and morphological identification of the parasite, and interpret the results in relation with the literature obtained from Iraqi and international sources related to the ecology and environmental parasitology of Acanthocephalans.

2. Materials and Methods

2.1 Study Area and Sampling Design

Sampling in the field was done every month from January to December 2025 in Al-Gharraf River, Wasit Province, Iraq. Three fixed sampling stations were selected, representing different riverine microhabitats: the first station was near the head regulator of Kut Barrage, the second was near the middle of Al-Muwaffaqiyah sector and the third was near Al-Hai district. The approximate coordinates used to describe these sectors were 32°30′N, 45°48′E for Station 1; 32°21′N, 45°56′E for Station 2; and 32°11′N, 46°00′E for Station 3. If rigorous sampling coordinates are required by the journal to which the manuscript is being submitted, the coordinates in this listing should be verified against the original field GPS record.

In the period of the study, a total of 720 specimens of fish were examined, of which 360 were P. abu and 360 S. triostegus. Thirty individuals of each host species were taken from all sampling stations for each month. The monthly sample was distributed between the three stations as evenly as possible, depending upon the availability of fish. Cast nets, gill nets and electrofishing were used to collect fish, as appropriate and legal. A calibrated field thermometer was used to take water temperature readings each month at the time of sampling.

2.2 Fish Transport and Ethical Handling of Fish

The live fishes were taken to the aerated polyethylene tanks filled with river water and then transported to the Ichthyoparasitology Laboratory, Department of Biology, College of Science, University of Wasit. They were transferred in about 2 hours after their capture. Those fish which could not be examined immediately were stored at 4°C for a maximum of 12 hours to reduce post-mortem tissue damage and the fallout of parasites.
Total length and weight of each fish to the nearest 0.1 cm and 0.1 g respectively was recorded prior to dissection. All procedures involved in handling and euthanasia were carried out in a humane manner and complied with with the appropriate institutional committee.

2.3 Fish Health and Disease Assessment

The fish were all laid on their dorsal side in a stainless-steel dissecting tray. A median incision made from anal end to the pectoral end; the abdominal wall was reflected to expose the abdominal cavity. Anterior esophagus to rectum digestive tract was taken and transferred to Petri dish with physiological saline solution (0.9% NaCl). The intestine was separated into three parts (foregut, midgut and hindgut) and opened lengthways with fine scissors and blunt probes without harming any worms attached to the intestine.

The intestinal mucosa was studied in a stereomicroscope. Acanthocephalans were removed carefully by fine needles which were used to spread the surrounding mucosa, not by pulling directly on the body. The purpose of this procedure was to avoid damage to the proboscis and hooks, which are required for morphological identification (Kennedy, 2006; Taraschewski, 2000).

2.4 Relaxation, Fixation, Staining and Mounting

The recovered worms were transferred to distilled water at 4°C for 14-18 hours to evert their proboscis and to relax their bodies. The relaxed specimens were preserved in 70% ethanol. Whole-mount worms were fixed in worms, rehydrated in descending ethanol, rinsed with distilled water, stained with Semichon's aceto-carmine for 20-45 minutes, differentiated in acid alcohol (only when necessary), dehydrated in ascending ethanol, cleared in xylene and mounted in Canada balsam. Slips were dried and examined under the microscope.

2.5 Morphological Identification

A compound light microscope with an ocular micrometer was used to examine the parasites. The proboscis structure, the number and arrangement of hooks, the body shape, the presence or absence of lemnisci, reproductive organs and other diagnostic features were used to make a morphological identification. The main criteria used for identification were the original description and taxonomic accounts of N. iraqensis and related species (Amin, 2002; Amin et al., 2001). Taxonomic confirmation was made by referring to the updated morphological and molecular account of Amin et al. (2026).

This study was not a taxonomic redescription, but rather an ecological and epidemiological survey. As such no new morphometric range is recommended. If possible, a representative mounted specimen(s) should be housed in an institutional parasitological collection to assist in future verification.

2.6 Epidemiological and Statistical Analysis. The parasitological terms were taken from Bush et al. (1997). Prevalence was calculated as percentage, percentage of infected fish/total number of fish examined x 100. Intensity of mean was determined by counting the number of parasites recovered from the infected fish. The overall mean intensity was computed as a weighted mean using the infected counts of each month and the monthly mean values of the intensities.

The chi-square test (Yates' correction) was used to test for differences in prevalence between host species. Pearson correlation was used to evaluate water temperature – prevalence relationship on a monthly basis. Descriptive summary of seasonal trends was used since the main purpose was to describe the annual temporal patterns. A p value less than 0.05 was considered statistically significant. SPSS version 26.0 was used for analysis.

3. Results

3.1 Overall Infection Pattern

A total of 720 fish were examined, with 19.17% (138 cases) infected with N. iraqensis. The percentage of infected fish was significantly higher in P. abu (24.72%) than in S. triostegus (13.61%) when all fish (360) were examined. The difference between the two host species was statistically significant (chi-square = 14.34, df = 1, p < 0.001). The overall weighted mean intensity was also higher in P. abu (4.6 worms/fish) than in S. triostegus (2.8 worms/fish).

Table 1. Monthly distribution of water temperature and *N. iraqensis* infection parameters in *P. abu* and *S. triostegus* during 2025.

Month	**Water temp. (°C)**	**P. abu infected**	**P. abu prevalence (%)**	**P. abu mean intensity**	**S. triostegus infected**	**S. triostegus prevalence (%)**	**S. triostegus mean intensity**
January	12.5	1	3.33	1.0	0	0.00	0.0
February	14.2	2	6.67	1.0	1	3.33	1.0
March	18.5	4	13.33	1.5	2	6.67	1.0
April	22.1	9	30.00	3.2	5	16.67	2.0
May	27.4	13	43.33	5.1	8	26.67	3.1
June	31.8	16	53.33	7.2	9	30.00	4.2
July	33.2	15	50.00	6.5	9	30.00	3.8
August	32.5	11	36.67	4.8	6	20.00	2.5
September	29.1	7	23.33	3.0	4	13.33	1.8
October	24.3	5	16.67	2.1	2	6.67	1.5
November	19.0	4	13.33	1.2	2	6.67	1.0
December	14.8	2	6.67	1.0	1	3.33	1.0
Total/mean	23.3	89	24.72	4.6	49	13.61	2.8

Note. Prevalence was computed with the sample size of 30 fish per host species/month. The overall prevalence was adjusted to 24.72% for P. abu and 13.61% for S. triostegus. The overall mean intensity for the total/mean row is a weighted average of the infected counts and the monthly mean intensities.

3.2 Seasonal Infection Pattern

There was a definite unimodal pattern in the seasonal analysis. The infection was significant in spring, highest in summer and decreased in autumn and winter. Prevalence was highest in summer (46.67%) and lowest in winter (5.56%) in P. abu. The same trend was noted in S. triostegus with prevalence being 26.67% in summer and 2.22% in winter. The seasonal mean intensity was similar, with the highest in summer for both host species.

Table 2. Seasonal stratification of *N. iraqensis* prevalence and mean intensity in both host species.

Season	P. abu examined	P. abu infected	P. abu prevalence (%)	P. abu mean intensity	S. triostegus examined	S. triostegus infected	S. triostegus prevalence (%)	S. triostegus mean intensity
Winter (Dec-Feb)	90	5	5.56	1.00	90	2	2.22	1.00
Spring (Mar-May)	90	26	28.89	3.89	90	15	16.67	2.45
Summer (Jun-Aug)	90	42	46.67	6.32	90	24	26.67	3.62
Autumn (Sep-Nov)	90	16	17.78	2.27	90	8	8.89	1.52

Note. Winter was defined consistently as December-February. Therefore, winter prevalence was corrected to 5.56% for *P. abu* and 2.22% for *S. triostegus*.

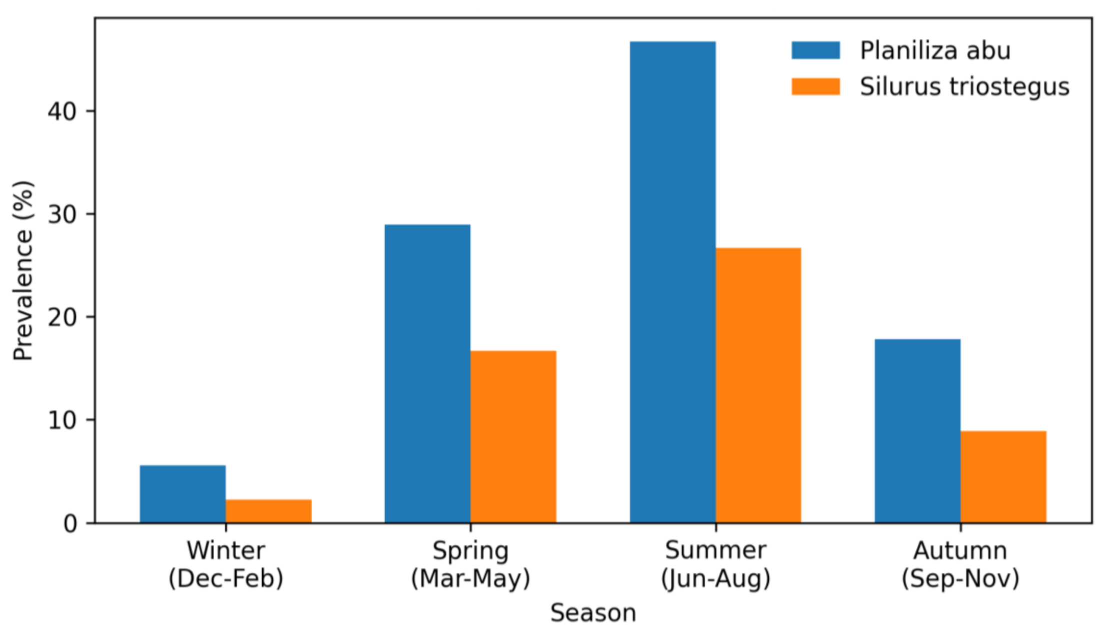

Figure 1. Seasonal prevalence of *N. iraqensis* in *P. abu* and *S. triostegus* from Al-Gharraf River.

3.3 Relationship with Water Temperature

The temperatures of the water ranged from 12.5°C in January to 33.2°C in July. The monthly prevalence values of both host species were highly and positively correlated with water temperature. The correlation was slightly stronger in P. abu ($r = 0.900$, $p < 0.001$) than in S. triostegus ($r = 0.883$, $p < 0.001$). The highest mean worm intensity was observed in June (7.2 worms/ infected fish) corresponding to early summer warm temperature.

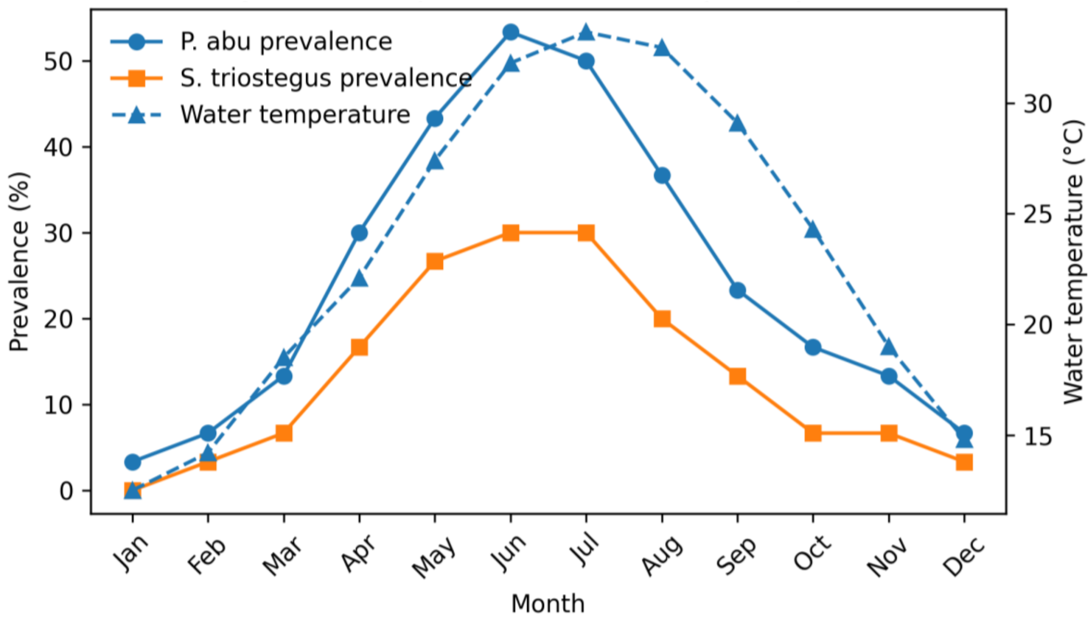

Figure 2. Monthly water emperature and prevalence of N. *iraqensis* in the two host species during 2025

4. Discussion

The present study revealed that the prevalence as well as the mean intensity of infection of N. iraqensis was significantly higher in P. abu than in S. triostegus. Trophic exposure, habitat use, and host feeding ecology are most likely responsible for this difference. The mode of transmission is usually trophozoological (Kennedy, 2006; Taraschewski, 2000), by ingestion of infected intermediate hosts or prey items containing infective larval stages by the definitive host. Thus, species of fish that have an intensive feeding on benthic material, detritus and small invertebrates are likely to be exposed to infective stages more than the fish with a more predatory feeding pattern.

The high infection rate of P. abu could be attributed to its wide benthic and herbivorous-detritivorous feeding habit. A dietary study from the southern waters of Iraq has revealed that *P. abu* feeds on diatoms, algae and detritus as well as fish eggs and small zooplankton, thus suggesting frequent association with the benthic substrates and small aquatic organisms (Mohamed & Abood, 2019). This feeding activity could lead to the ingestion of infected intermediate hosts or the contact with benthic related parasite stages. S. triostegus is a large demersal catfish species, with distinct ecological and feeding characteristics (Froese & Pauly, 2025b; Jawad & Al-Janabi, 2016). The reduced prevalence in the present study may be due to a decrease in direct contact with infected small crustaceans. However, this cannot be discounted, as predatory fish can pick up some parasites by ingesting infected prey. There is need to further investigate this possibility by stomach content analysis and examination of possible intermediate or paratenic host.

The percentage prevalence exhibited in this study showed a low level in winter, followed by rising levels in spring, a definite peak in summer, and a decrease in autumn. This pattern is consistent with the overall trend that the transmission of aquatic parasites is influenced by changes in water temperature, feeding activity of the host and availability of intermediate hosts (Marcogliese, 2016). The higher water temperatures during warm months can improve fish metabolism and feeding behavior, which can lead to higher chances of consuming infected intermediate hosts. Meanwhile, the temperature can affect development of parasites, and the numbers of intermediate hosts, but this effect can be variable depending on the type of parasite, host species and aquatic ecosystem.

Both water temperature and prevalence of *Neoechinorhynchus iraqensis* showed strong positive correlations with the month in both host species suggesting that water temperature was very important in influencing the trapping rate of *Neoechinorhynchus iraqensis* in Al-Gharraf River during 2025. The correlation between temperature and infection, however, was not entirely the same for both host species, suggesting that this relationship cannot be fully accounted for by temperature. It appears to be affected by the host behavior, habitat preference, feeding strategy and trophic exposure. More environmental parameters should be measured in future investigations, including dissolved oxygen, salinity, pH, turbidity, organic matter and seasonality of intermediate host abundance. The consideration of these factors would cover a more comprehensive picture of the ecological drivers that influence the transmission of acanthocephalans in freshwater systems of Iraq.

The occurrence of *Neoechinorhynchus iraqensis* in Planiliza abu is similar to the original description from the Iraqi freshwater mullet (Amin et al., 2001). Since then, the species has been documented in various aquatics in Iraq and recent checklists include several fish hosts containing species of Neoechinorhynchus in the Tigris, Euphrates, Shatt Al-Arab, marshes, tributaries, drainage systems, ponds and floating cages (Mhaisen & Abdul-Ameer, 2024). The presence of acanthocephalan in the freshwater mugilid fish Liza abu was also mentioned in historical records, confirming the association between intestinal helminths and mullets in Iraqi waters, which was previously reported by (Mhaisen et al. 1988).

The prevalence values obtained in the present study also have been similar to some recent reports in Iraq. (Al-Musaedi and Alsaady , 2022) reported *Neoechinorhynchus iraqensis* from Planiliza abu and Silurus triostegus, and they found that Planiliza abu was more infected with the parasite than Silurus triostegus. Thi-Qar Province molecular studies of *Neoechinorhynchus iraqensis* in Planiliza abu, Aspius vorax and Silurus triostegus were based on 28S rDNA, and confirmed the presence of this parasite in the three genera (Gaber & Awad, 2025). Similarly, molecular identification was carried out from Darbandikhan Lake and confirmed the presence of *Neoechinorhynchus iraqensis* in Planiliza abu (Ali & Abdullah, 2024). These results collectively indicate that the distribution of *Neoechinorhynchus iraqensis* is widespread within the freshwater habitats of Iraq and that the prevalence of the parasite could be influenced by host species, locality, season and environment.

Careful recovery, relaxation, fixation and mounting of parasites is important in the accurate identification of acanthocephalans. In some species, such as those of the genus Neoechinorhynchus, proboscis form and the disposition of the hooks are significant characters helpful for the identification and damage to the proboscis or hook rows may affect this. The relaxation, fixation, staining, clearing and mounting have been in accord with the normal morphological technique employed in the taxonomy of acanthocephalans.

The identification in this study is nevertheless considered to be "morphology based" since no DNA sequence was obtained for the present specimens. It is therefore not intended that molecular diagnosis was carried out in this study, but rather that updated molecular references were included to aid in the taxonomic interpretation and comparison. Future studies/works should include photomicrograph, scanning electron microscopy (when available) and molecular confirmation (using 18S, 28S or mitochondrial markers) to strengthen the morphological diagnosis, particularly in studies designed to confirm species identity or to assess genetic variation (Amin et al., 2026). It must be recognised that there are some restrictions.

The study primarily emphasized monthly prevalence and mean intensity and its measurements were not quantitative of intermediate hosts. However, host sex, age class, size class, stomach content and various water quality parameters were not available for analysis and could affect parasite transmission.

In addition, there was no morphological confirmation of molecular sequencing in the present set of data. Further studies are needed to be conducted in conjunction with intermediate-host surveys, stomach-content analysis and histopathology, as well as the use of photomicrography and molecular tests. This integrated approach would yield more information about the transmission pathways, host specificity, seasonal patterns and environmental triggers of infection with N. iraqensis in Iraqi freshwater systems.

5. Conclusion

In this study it was shown that N. iraqensis is present in both P. abu and S. triostegus in Al-Gharraf River, Wasit Province, Iraq and the mean intensity and prevalence of N. iraqensis in P. abu were significantly higher than in S. triostegus. There was a clear seasonality in infection, with an increase in infection during spring, a peak in summer and a decrease in autumn and winter.

In both host species, water temperature was a significant correlate of monthly prevalence, indicating that the seasonal thermal conditions play a role in the transmission cycle. The benthic and detritivorous feeding habits of P.

abu might be responsible for the greater infection burden, by causing greater exposure to the infected intermediate hosts. The results of this study give the new local information for fish parasite ecology in Al-Gharraf River and point out the need to combine the host ecology, seasonality monitoring and environmental assessment in future studies of fish parasites in Iraq.

Conflict of Interest

The authors declare that they have no conflict of interest.

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