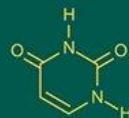


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Comparative analysis of GSI in relation to environmental factors in *Puntius sophore* and *Rasbora daniconius*

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Abstract

The research work was carried out to assess the Gonadosomatic Index (GSI) of two small indigenous freshwater fishes, *Puntius sophore* and *Rasbora daniconius* with respect to environmental factors. The examination aimed to examine the reproductive biology and spawning behaviour of these species. The results indicated significant seasonal fluctuations in GSI values in both fish species, reflecting variations in gonadal maturation and reproductive activity. The spawning season of *P. sophore* was identified as June–August with peak spawning in June–July (GSI peak in May: 8.46%), while *R. daniconius* spawned from April–July with peak in July (GSI: 2.21%). *Puntius sophore* showed significantly higher GSI and fecundity compared to *R. daniconius*, indicating greater reproductive investment per spawning event. Correlation analysis revealed significant positive GSI-fecundity relationships in both species, while *R. daniconius* uniquely showed significant negative GSI-body size correlations.

Keywords: Indigenous, fluctuation, fecundity, GSI, correlation

1. Introduction

Reproductive Biology is among the most important process of a fish population. Every fish has its own ecological habitat, hence, each one is characterized by a distinctive form of reproduction, possessing particular physiological, behavioural and morphological adaptations. Research into teleostean reproduction usually focuses on commercially valuable or indigenous species. Knowledge about their reproductive cycle and the factors that influence it are important aspects of fisheries biology. Reproductive factors such as Gonadosomatic index (GSI), fecundity, spawning period and egg diameter are widely used to evaluate comparative gonadal maturation and breeding efficiency in fishes. The reproductive process is significantly influenced by ambient environmental conditions including temperature, dissolved oxygen, rainfall pattern, and photoperiod and water quality. *Puntius sophore*, commonly known as Pool Barb, Spot-fin Swamp Barb, or Stigma Barb, is an important small indigenous fish species belonging to the family Cyprinidae under the order Cypriniformes. This species has a wide geographical distribution extending across India, Bangladesh, Myanmar, Nepal, Pakistan, China, Bhutan, and Afghanistan (Zen *et al.*, 2024). The species inhabits diverse freshwater habitats including rivers, streams, ponds, beels (floodplain wetlands), and inundated rice fields. *Rasbora daniconius*, commonly known as Slender Rasbora or Blackline Rasbora, is a small cyprinid fish belonging to the family Danionidae (previously included in Cyprinidae) under the order Cypriniformes. Several studies have described the reproductive biology of *Puntius sophore* and *Rasbora daniconius* individually; however, comparative information regarding the influence of environmental factors on GSI and reproductive activity remains limited. Hence, the present investigation was undertaken to compare the reproductive biology of these two fish species in relation to environmental parameters.

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2. Material and Method

The study was carried out to analyze the reproductive characteristics of *Puntius sophore* and *Rasbora daniconius* collected from Tighra Dam Gwalior, Madhya Pradesh. Fish specimens were sampled regularly with the help of cast nets and hand-operated fishing gears. After collection, the specimens were preserved carefully and transported to the laboratory for detailed examination.

In the laboratory, morphometric analysis including total body length and body weight were recorded using a measuring scale and digital balance. Each specimen was dissected carefully to remove the gonads for reproductive analysis. The weight of gonads was measured separately, and the Gonadosomatic Index (GSI) was calculated using the following formula:

$$\text{GSI (\%)} = (\text{Gonad Weight} / \text{Total Body Weight}) \times 100$$

Gonadosomatic Index (GSI): GSI is a widely used indicator for assessing the reproductive status and spawning periodicity of fish species. It represents the ratio of gonad weight to body weight and reflects the relative investment in reproduction. Monthly variations in GSI help identify breeding seasons and peak spawning periods, which is crucial for implementing closed seasons and protecting spawning stocks. Recent studies on small indigenous fishes have demonstrated that GSI peaks during pre-monsoon and monsoon periods in South Asian cyprinids (Singha *et al.*, 2024; Hossain *et al.*, 2024) [1].

3. Result and discussion

The Gonadosomatic Index (GSI) is the most widely used and reliable indicator of the reproductive state and spawning readiness of fish (Lagler, 1956; De Vlaming *et al.*, 1982; West, 1990) [3, 4, 6]. Higher GSI values indicate active gonadal development and approaching spawning, while lower values signify quiescent, immature, or spent reproductive phases. The GSI is extensively used to determine the spawning season, duration of the reproductive period, and the reproductive effort of fish populations (Murua *et al.*, 2003) [5].

In the present study, the GSI was calculated for all specimens on a monthly basis to determine the temporal pattern of gonadal development and to identify the peak spawning period of both species in Tighra Dam, Gwalior. One-way analysis of variance (ANOVA) was used to test for significant monthly variation in GSI values.

3.1 *Puntius sophore* (Female)

The monthly variation in GSI of *P. sophore* females is presented in Table 1 and illustrated in Figure 1. The annual mean GSI was $3.05 \pm 0.59\%$ (SD = 6.212, range: 0.13 to

40.92%). The extremely wide range and high standard deviation reflect the fact that the sampled population included fish at all stages of gonadal development from immature fish with minimal gonad weight to fully ripe fish with heavy, yolk-laden ovaries.

The GSI showed a distinct seasonal pattern that clearly delineated the reproductive cycle

- **Winter Phase (November–February):** The GSI values were at their lowest during this period, with November ($0.31 \pm 0.08\%$) and February ($0.31 \pm 0.08\%$) recording the minimum mean values. January showed a slightly higher value ($1.27 \pm 0.69\%$) but was based on only 2 specimens and should be interpreted with caution. During this period, the ovaries were predominantly in the immature or spent condition (recovering from the previous spawning season), with minimal gonadal activity. This phase represents the gonadal quiescent or resting period of the annual reproductive cycle.
- **Pre-Monsoon Development Phase (March–May):** A gradual increase in GSI was observed from March ($0.50 \pm 0.15\%$) through April ($1.68 \pm 0.82\%$), reaching markedly elevated levels in May ($8.46 \pm 5.03\%$). The high standard error in May reflects the co-occurrence of fish at different stages of maturation within the same month some individuals had already reached advanced vitellogenesis while others were still in the early maturing phase. The month of May recorded the highest mean GSI in the entire study, suggesting that the initial cohort of females attained peak gonadal maturity just before the onset of the southwest monsoon.
- **Monsoon Spawning Phase (June–August):** GSI remained elevated during the monsoon months June ($3.90 \pm 2.49\%$), July ($4.16 \pm 2.15\%$), and August ($7.03 \pm 1.81\%$). The sustained high GSI values during this three-month period confirm prolonged reproductive activity coinciding with the monsoon season. The high GSI in August (7.03%) is noteworthy and suggests that some individuals may undertake a second or extended spawning bout during the late monsoon period. This is consistent with the batch-spawning strategy identified through ova diameter studies (Section 4.10).
- **Post-Monsoon Decline Phase (September–October):** A decline in GSI was observed from September ($2.95 \pm 1.16\%$) through October ($0.88 \pm 0.29\%$), indicating the cessation of spawning activity and the progressive transition of ovaries to the spent condition. The large sample size in September (N = 25) provides a robust estimate, and the elevated standard error indicates the simultaneous presence of some fish still in reproductive condition alongside predominantly spent individuals.

Table 1: Monthly variation in Gonadosomatic Index (GSI, %) of *Puntius sophore* (Female) from Tighra Dam, Gwalior

Month	N	Mean±SE	SD	Min	Max	Median	Season	Stage
January	2	1.27±0.69	0.97	0.58	1.95	1.27	Winter	SPENT
February	6	0.31±0.08	0.20	0.13	0.59	0.25	Winter	IMMATURE
March	7	0.50±0.15	0.39	0.13	1.17	0.36	Pre-Mon.	MATURING
April	8	1.68±0.82	2.31	0.13	6.09	0.49	Pre-Mon.	MATURING
May	8	8.46±5.03	14.22	0.21	40.92	1.44	Pre-Mon.	MATURING
June	10	3.90±2.49	7.88	0.13	24.29	0.45	Monsoon	SPAWNING
July	10	4.16±2.15	6.79	0.13	18.42	1.22	Monsoon	SPAWNING
August	11	7.03±1.81	6.00	0.30	18.84	6.67	Monsoon	MATURING
September	25	2.95±1.16	5.82	0.14	25.17	0.63	Monsoon	SPENT
October	16	0.88±0.29	1.17	0.13	4.22	0.33	Post-Mon.	SPENT
November	6	0.31±0.08	0.20	0.17	0.63	0.25	Post-Mon.	SPENT
December	3	1.66±0.77	1.34	0.30	2.97	1.71	Winter	MATURING
Annual	112	3.05±0.59	6.21	0.13	40.92	0.71	--	--

ANOVA: F = 1.676, df = 11, 100, p = 0.090 (Not Significant); Values expressed as Mean ± Standard Error

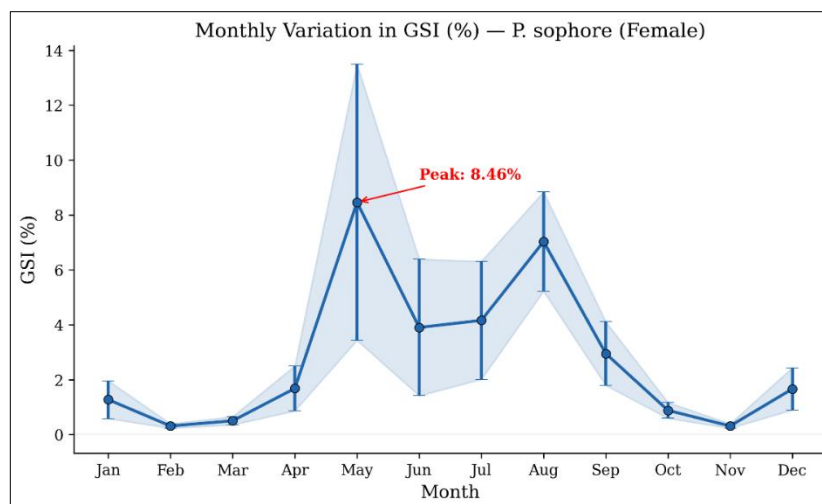


Fig 1: Line graph showing monthly variation in mean GSI (%) of *P. sophore* (Female) from Tighra Dam, Gwalior with vertical bars indicating $\pm$ Standard Error

One-way ANOVA revealed that the monthly variation in female GSI was not statistically significant at the 5% level ($F = 1.676$, $df = 11, 100$, $p = 0.090$), although it approached significance. The non-significant result despite the obvious seasonal trend in mean values is likely attributable to the extremely high within-month variability (overall $CV = 203.7\%$). Within any given month, the population comprised fish at multiple stages of gonadal maturity some immature, some maturing, some ripe, and some already spent resulting in high intra-monthly variance that reduces the power of the ANOVA to detect between-month differences. This phenomenon is commonly reported in multi-spawning tropical fish species and does not negate the biological significance of the observed seasonal GSI pattern (West, 1990; Murua *et al.*, 2003) [5, 6].

The peak GSI period observed in the present study (May–August) is broadly consistent with published reports on *P. sophore* from the Indian subcontinent. Rafiquzzaman *et al.* (2018) reported the highest GSI values during April (15.43 ± 2.20) from Gazipur and June (15.60 ± 1.74) from Jessore, Bangladesh, indicating regional variation in peak spawning time. Choudhury *et al.* (2015) documented GSI peaks between March and July from Tripura, India. (Kant *et al.* (2016) [7] reported a single highest peak in July (14.06 ± 1.59) from Jammu waters. Hossain *et al.* (2006) [8] observed peak GSI during June–July from the Padma River, Bangladesh. The GSI values reported in the present study (maximum mean: 8.46% in May) are somewhat lower than the values reported from Bangladeshi populations (15.43–15.60%), which may be attributed to differences in body

size, nutritional status, environmental conditions, and the analytical approach (the present study includes all maturity stages, while some published studies selectively analysed mature females only).

The monsoon-associated spawning pattern observed in *P. sophore* is characteristic of most Indian freshwater cyprinids and has been attributed to a combination of environmental triggers including rising water levels, increased turbidity, changes in water temperature, photoperiod alterations, and the onset of rainfall (Jhingran, 1991; Lowe-McConnell, 1987; Welcomme, 1979) [9, 10, 11]. These environmental cues are believed to stimulate the hypothalamic-pituitary-gonadal (HPG) axis, leading to increased gonadotropin secretion and subsequent gonadal maturation and spawning (Zohar, 1989; Pankhurst and Munday, 2011). The pre-monsoon GSI peak in May suggests that some individuals may complete their gonadal maturation in response to rising temperatures and early rainfall events before the full onset of the southwest monsoon in June.

3.2 *Puntius sophore* (Male)

The GSI of *P. sophore* males (Table 2) was considerably lower than that of females, as is characteristic of teleost fishes where the female gonad (ovary) constitutes a much larger proportion of body weight than the male gonad (testis) due to the metabolic investment in yolk production during oogenesis (Wootton, 1990). The annual mean GSI for males was $0.38 \pm 0.02\%$ ($SD = 0.158$, range: 0.13 to 0.82%), approximately one-eighth of the female annual mean (3.05%).

Table 2: Monthly variation in GSI (%) of *Puntius sophore* (Male) from Tighra Dam, Gwalior

Month	N	Mean $\pm$ SE	SD	Min	Max	Season	Stage
February	4	0.17 $\pm$ 0.02	0.04	0.13	0.22	Winter	Developing
March	5	0.20 $\pm$ 0.04	0.09	0.13	0.35	Pre-Mon.	Resting
April	5	0.38 $\pm$ 0.05	0.10	0.24	0.50	Pre-Mon.	Maturing
May	6	0.45 $\pm$ 0.04	0.09	0.34	0.59	Pre-Mon.	Mature
June	6	0.50 $\pm$ 0.05	0.13	0.33	0.72	Monsoon	Mature
July	7	0.57 $\pm$ 0.08	0.20	0.33	0.82	Monsoon	Mature
September	8	0.38 $\pm$ 0.02	0.05	0.31	0.46	Monsoon	Spent
October	11	0.39 $\pm$ 0.01	0.03	0.34	0.44	Post-Mon.	Spent
November	4	0.16 $\pm$ 0.02	0.04	0.13	0.22	Post-Mon.	Spent
December	2	0.37 $\pm$ 0.01	0.01	0.36	0.38	Winter	Spent
Annual	58	0.38 $\pm$ 0.02	0.16	0.13	0.82	--	--

ANOVA: $F = 8.993$, $df = 9, 48$, $p < 0.001$ (**); No specimens collected in January and August

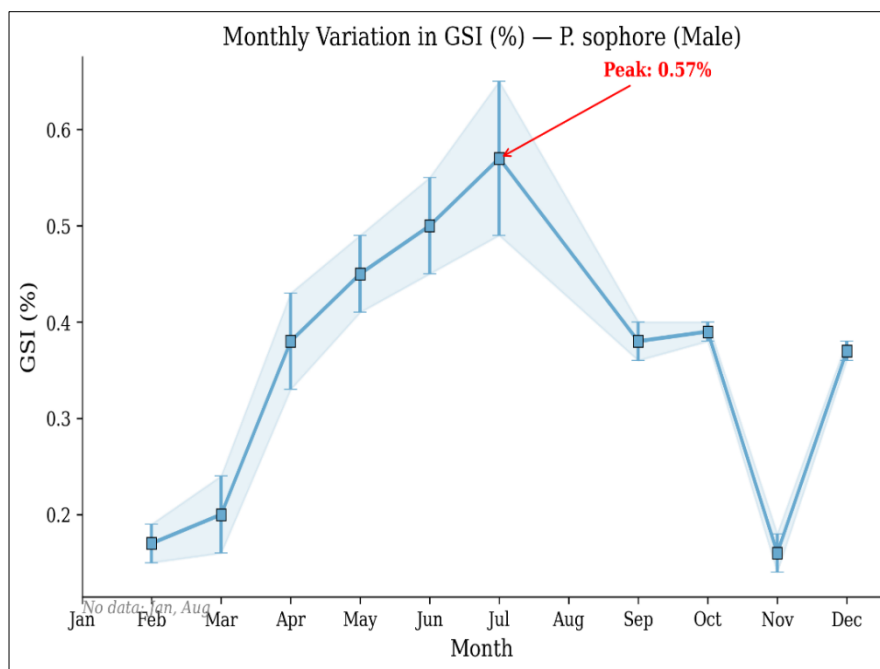


Fig 2: Line graph showing monthly variation in mean GSI (%) of *P. sophore* (Male) from Tighra Dam, Gwalior

The highest male GSI was recorded in July ($0.57 \pm 0.08\%$), followed by June ($0.50 \pm 0.05\%$) and May ($0.45 \pm 0.04\%$), closely corresponding to the female GSI pattern and confirming synchronous gonadal maturation in both sexes during the pre-monsoon and monsoon months. The lowest values were recorded during November ($0.16 \pm 0.02\%$) and February ($0.17 \pm 0.02\%$), corresponding to the resting/recovery phase. No male specimens were collected in January and August, which precluded GSI estimation for these months. One-way ANOVA revealed highly significant monthly variation in male GSI ($F = 8.993$, $df = 9, 48$, $p < 0.001$). This contrasts with the non-significant result obtained for females and can be explained by the substantially lower within-month variability in male GSI ($CV = 41.7\%$ for males vs. 203.7% for females). The more uniform pattern of spermatogenesis compared to the complex, multi-stage process of oogenesis results in lower

intra-monthly variance in male GSI, thereby increasing the statistical power of the ANOVA to detect seasonal differences (Wootton, 1990). Post-hoc analysis (Tukey's HSD) revealed that the GSI values in May, June, and July were significantly higher than those in November, February, and March ($p < 0.05$), confirming the pre-monsoon to monsoon period as the peak male reproductive phase.

3.3 *Rasbora daniconius* (Female)

The GSI of *R. daniconius* females (Table 3) showed an annual mean of $1.02 \pm 0.15\%$ ($SD = 1.318$, range: 0.11 to 7.00%), which was markedly lower than that of *P. sophore* females ($3.05 \pm 0.59\%$). The peak GSI was recorded in July ($2.21 \pm 0.65\%$), followed by October ($2.02 \pm 0.79\%$) and June ($1.36 \pm 0.43\%$). The lowest GSI values were observed during winter months February ($0.21 \pm 0.05\%$) and January ($0.32 \pm 0.10\%$).

Table 3: Monthly variation in GSI (%) of *Rasbora daniconius* (Female) from Tighra Dam, Gwalior

Month	N	Mean±SE	SD	Min	Max	Season	Stage
January	5	0.32±0.10	0.23	0.11	0.65	Winter	Spent
February	5	0.21±0.05	0.11	0.11	0.40	Winter	Immature
March	6	0.47±0.15	0.37	0.16	1.10	Pre-Mon.	Matured
April	6	0.96±0.21	0.51	0.32	1.63	Pre-Mon.	Spawning
May	7	0.53±0.12	0.31	0.17	1.00	Pre-Mon.	Spawning
June	7	1.36±0.43	1.13	0.30	3.42	Monsoon	Spawning
July	8	2.21±0.65	1.84	0.36	5.88	Monsoon	Matured
August	7	0.73±0.20	0.52	0.15	1.50	Monsoon	Spent
September	1	0.80±0.00	--	0.80	0.80	Monsoon	Spent
October	10	2.02±0.79	2.49	0.15	7.00	Post-Mon.	Spent
November	5	0.39±0.13	0.30	0.11	0.76	Post-Mon.	Immature
December	7	0.75±0.02	0.06	0.68	0.83	Winter	Spent
Annual	74	1.02±0.15	1.32	0.11	7.00	--	--

ANOVA: $F = 2.315$, $df = 10, 63$, $p = 0.022$ (*); September: only 1 specimen interpret with caution

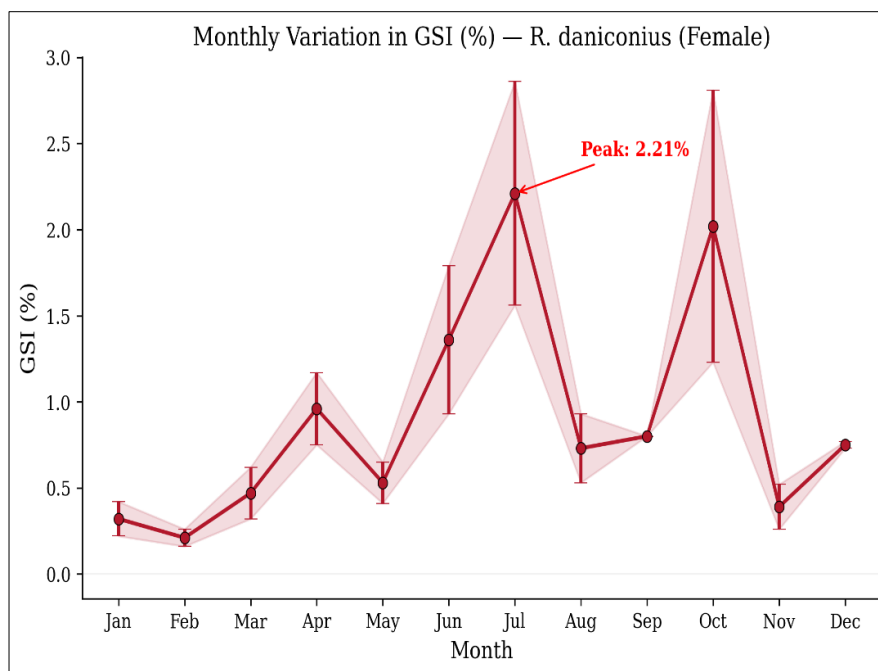


Fig 3: Line graph showing monthly variation in mean GSI (%) of *R. daniconius* (Female) from Tighra Dam, Gwalior

One-way ANOVA revealed significant monthly variation in GSI ($F = 2.315$, $df = 10, 63$, $p = 0.022$). The secondary GSI peak in October ($2.02 \pm 0.79\%$) is noteworthy and may suggest a bimodal spawning pattern or a late-season reproductive cohort in *R. daniconius*. However, this October peak should be interpreted cautiously due to the high standard error and the presence of one individual with GSI = 7.00% (an outlier) that substantially elevated the mean. The lower GSI in August (0.73%) and the shift to spent condition suggest that the primary spawning event concludes by August, and the October elevation may represent residual reproductive activity or delayed maturation in a subset of the population. Comparative

Analysis between the two species using the independent t-test revealed a highly significant difference in female GSI (*P. sophore*: $3.05 \pm 0.59\%$ vs. *R. daniconius*: $1.02 \pm 0.15\%$; $t = 3.350$, $p = 0.001$, Cohen's $d = 0.45$). The medium effect size ($d = 0.45$) indicates a biologically meaningful difference between the species. *P. sophore* invests proportionally more energy in gonadal development relative to body weight compared to *R. daniconius*. This may reflect the contrasting reproductive strategies: *P. sophore* as a more prolific spawner producing larger numbers of smaller eggs, while *R. daniconius* may adopt a strategy of producing fewer but relatively larger eggs, investing more energy per offspring (Smith and Fretwell, 1974; Wootton, 1990).

3.4 Comparative GSI analysis

Table 4: Comparative monthly variation in GSI (%) of *P. sophore* and *R. daniconius* from Tighra Dam, Gwalior

Month	<i>P.s</i> ♀ (N)	<i>P.s</i> ♀ GSI	<i>P.s</i> ♂ (N)	<i>P.s</i> ♂ GSI	<i>R.d</i> ♀ (N)	<i>R.d</i> ♀ GSI
Jan	2	1.27 ± 0.69	0	--	5	0.32 ± 0.10
Feb	6	0.31 ± 0.08	4	0.17 ± 0.02	5	0.21 ± 0.05
Mar	7	0.50 ± 0.15	5	0.20 ± 0.04	6	0.47 ± 0.15
Apr	8	1.68 ± 0.82	5	0.38 ± 0.05	6	0.96 ± 0.21
May	8	8.46 ± 5.03	6	0.45 ± 0.04	7	0.53 ± 0.12
Jun	10	3.90 ± 2.49	6	0.50 ± 0.05	7	1.36 ± 0.43
Jul	10	4.16 ± 2.15	7	0.57 ± 0.08	8	2.21 ± 0.65
Aug	11	7.03 ± 1.81	0	--	7	0.73 ± 0.20
Sep	25	2.95 ± 1.16	8	0.38 ± 0.02	1	0.80 ± 0.00
Oct	16	0.88 ± 0.29	11	0.39 ± 0.01	10	2.02 ± 0.79
Nov	6	0.31 ± 0.08	4	0.16 ± 0.02	5	0.39 ± 0.13
Dec	3	1.66 ± 0.77	2	0.37 ± 0.01	7	0.75 ± 0.02
Annual	112	3.05 ± 0.59	58	0.38 ± 0.02	74	1.02 ± 0.15

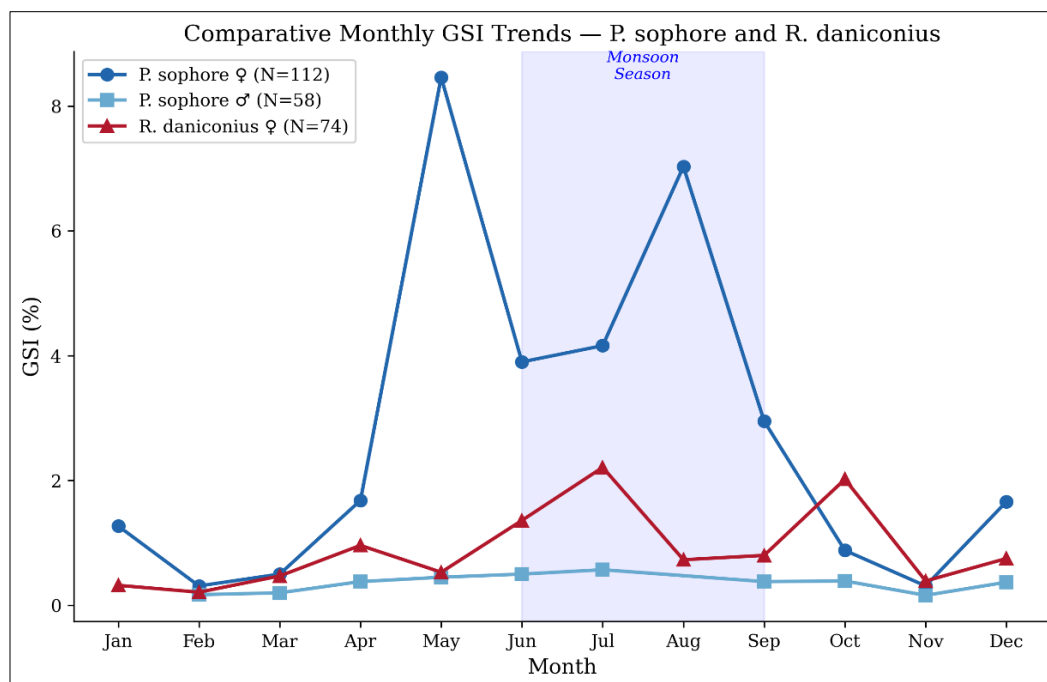


Fig 4: Comparative line graph showing monthly GSI trends of *P. sophore* (♀ and ♂) and *R. daniconius* (♀) from Tighra Dam, Gwalior

The comparative GSI analysis reveals that while both species exhibit monsoon-associated spawning, their peak GSI months differ slightly. *P. sophore* females showed the highest GSI in May (8.46%), while *R. daniconius* peaked in July (2.21%). This temporal offset in peak reproductive activity may represent an adaptive mechanism to reduce inter-specific competition for spawning habitat, food resources for larvae, and nursery grounds a phenomenon commonly observed in sympatric fish communities in tropical freshwaters (Lowe-McConnell, 1987; Welcomme, 1979) [10, 11].

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