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Exploring the Rich Tapestry of Macroinvertebrates, Including Aquatic Oligochaetes, across Seasons in the Bao Rice (Deep Water Rice) Fields of Lakhimpur District, Assam, India

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Abstract: An extensive study of freshwater macroinvertebrates was conducted in two deep water rice (DWR) field, natively known as Bao or red rice fields in Lakhimpur District, Assam, India from June 2021 to May 2022. This study identified 43 species from 31 families and 15 orders, including Arthropods, Annelids, and Molluscs. Hemiptera was the most dominant order (33.99% RA), with 10 species across 7 families, followed by Tubificida (12.15% RA), with 5 species in 2 families. Odonata had 3 families and 4 species (11.85% RA), while Trombidiformes had the lowest dominance (1.15% RA). *Paraplea* sp. and *Baetis fuscatus* were notably abundant. The high Shannon index (3.2<H'<3.5) indicated significant species diversity, and the Evenness index (0.64<E<0.74) suggested balanced distribution. The Biomonitoring Working Party (BMWP) index (93-114) confirmed good water quality. Furthermore, six species of aquatic oligochaetes; *Aeolosoma headleyi, Branchiodilus* sp*., Dero spp., Aulophorus* sp*., Pristina brevista, and Limnodrilus hoffmeisteri* were recorded for the first time in the DWR field of Assam, North East India.

Keywords: Bao, Red rice, Macroinvertebrates Oligochaetes, Relative abundance, Shannon index, Evenness index

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Introduction

Freshwater macroinvertebrates, whether benthic or nektonic, are pivotal in nutrient cycling, decomposition, and material translocation within aquatic habitats (Wallace and Webster, 1996). In aquatic ecosystems, macroinvertebrates such as shredders, collectors, scrapers, piercers, and engulfers each occupy specific niches, offering vital services to the entire aquatic community (Watson-Ferguson *et al.,* 2006). Their high tolerance, abundance, and rapid colonization rate serve as indicators of ecosystem stability, often utilized in biomonitoring to assess anthropogenic impacts on aquatic ecosystems (Arimoro and Ikomi, 2008; Kripa *et al.*, 2013; Xu *et al.*, 2014). Rice fields host a variety of aquatic organisms, some of which complete their life cycles from larvae to adults in muddy aquatic environments (Bambaradeniya and Amerasinghe, 2004; Leitao *et al.*, 2007). Bao rice, also known as red rice or deep water rice (DWR), thrives in flood-prone areas of Assam, forming seasonal habitats for diverse aquatic organisms. Freshwater oligochaetes, capable of withstanding oxygen-deficient water, decomposing litter, and exhibiting benthophagous habits, serve as valuable indicators of environmental parameters and play significant roles in wetland trophic systems (Wissinger, 1999).

Due to minimal anthropogenic disturbance and stagnation of water which persist for a long duration, DWR fields support a large number of aquatic organisms throughout the year. Given the limited research on Bao rice fields and their aquatic organisms, particularly in the Northeastern region, this study aims to investigate the abundance of aquatic organisms and benthic oligochaetes during and after the harvesting period of Bao rice crops in Lakhimpur District, Assam, India.

Materials and Methods

Two important seasonally flood-affected Bao rice field (DWR) in Lakhimpur district, Assam, India viz. Magurmari (27º07'41.3'' N and 94º09'09.0''E) and Chutiakari (27º14'58.5'' N and 94º08'36.8'' E), were selected for study from June 2021 to May 2022. The sampling was conducted during the cultivation and post harvesting period. During post harvesting phases the sample were collected from the water bodies in small ponds and trenches within and around the bao rice field. The specimens were collected once in a month from 5 random sampling site of each field. A total 30 samples were collected in every seasons from both the sampling site by using D frame net and circular frame net with mesh size 500 µm. Standard Protocol as outlined by DiFranco (2006) for sampling of macroinvertebrates in freshwater wetlands was adopted in this study. Dip net was swept through the water column for a distance of one meter within approximately 30 sec to 1 min in an area where the bottom surface has not been disturbed earlier. Three such sweeps constituted a sample for each site. (Rosenberg and Resh, 1982; Subramanian and Sivaramakrishnan, 2007; Coccia *et al.*, 2016; Dalal and Gupta, 2016; Abhilash and Mahadevaswamy, 2022). For benthic organisms, a wooden frame cubes measuring 0.5m X 0.5 m X 0.5m open on the top and bottom side wrapped with nylon netting, were employed in different sites of bao rice field and after proper hand shaking of vegetation or uprooting them , the water sample along with oligochaetes and other components were collected into a container (Hegde and Sreepada, 2014). Macroinvertebrates were sorted, and smaller organisms were filtered using a 0.4 mm mesh net (Che Salmah, 1996) and 30 ml of samples were filtered out for microscopic observation. For mud-dwelling oligochaetes, the bottom layer was sieved and transferred to a container with water. Excess sediment was removed by rinsing, and the sample was placed in a water-filled petri dish for observation.

Identification of Macroinvertebrates:

Collected specimens were observed under photoplan microscope (MSW-807) and classified up to possible level of taxa on the basis of morphological characteristic. Identification keys and methodology adopted as per Imms (1957); Edmonson (1959); Richardson (2003); Bal *et al.* (2004); and Neiser and Chen (2005). Taxonomic characteristics like Habitat, Body pigmentation, Body contour, types and position of setae, shape of prostomium and anal part, position of mouth, formation of mucous tubes etc. were followed using standard literature (Harman *et al.,*1988; Naidu, 2005). Relative abundance (RA%) and dominant rate of seasonal and annually abundant species was assessed by using Engelmens dominance scale (Engelmann,1973). Statistical analysis (Shanon index, Evenness Magalef index and Berger–Parker) was done using PAST statistical software. Biomonitoring Working Party (BMWP) score obtained across all seasons, as described in studies by Mandaville (2002) and Barman *et al.* (2015).

Results and Discussion

In the present study, a total of 43 species (Table 1) of macroinvertebrates from the DWR field were identified, belonging to 31 different families (Fig. 1) and distributed across 15 orders (Fig. 2). Among these orders, Hemiptera was the most abundant, representing RA: 34% (Fig. 2) of the total organisms. Within the order Hemiptera, 10 species belonging to 7 families were observed, such as *Gerris* sp. (Garridae), *Hydrometra* sp. (Hydrometridae), *Backswimme*r sp. (Notonectidae), *Paraplea* sp. (Pleidae), *Ranatra* sp. (Nepidae), *Lethocerous indicus*, *Laccotrephes ruber, Diplonychus rusticus* and *Abedus herberti* (Belostomidae) and *Micronecta silva* (Micronectidae). Studies by Thongphak *et al.* (2016); Wakhid *et al.* (2020) also reported Hemipteran dominancy in the aquatic environments in Indonesia and Thailand, respectively. Furthermore, Das and Gupta (2010) reported the presence of 10 hemipteran species in rice fields in Cachar district, Assam, while Hazarika and Goswami (2010) working on the pond ecosystem of Guwahati, Assam, India cited 14 hemipteran species.

The study examined 6 *aquatic* Oligochaetes species: *Aeolosoma headleyi* (family Aeolosomatidae, order Aeolosomatida); *Aulophorus* sp., *Pristina brevista*, *Dero* sp., *Branchiodilus* sp. (family Naididae); and *Limnodrilus hoffmeisteri* (family Tubificidae), notably the first four species had no prior documentation from the northeastern region of India (Singh *et al.*, 2009), additionally there are no prior reports of investigations on these species, specifically in the context of the DWR field of Lakhimpur, Assam. Following Hemiptera, the order Tubificida, encompassing the families Naididae and Tubificidae of aquatic Oligochaetes was the second most dominant (RA: 12%), group. Order Odonata represented by four species across three families: *Somatochlora* sp. (family Cordulidae), *Sympetrum striolatum* (family Libellulidae), and *Ischnura* sp. and *Ceriagrion* sp. (family Coenagrionidae) accounted for RA: 11.8% of the total fauna. The order Coleoptera constitutes RA: 8.4%, with five species: *Cybister fimbriolatus*, *Cybister sugilatus*, *Hygrotus* sp., and *Laccophilus* sp. (family Dytiscidae), and the Water Scavenger Beetle (family Hydrophilidae). Similarly, order Orthoptera, although primarily terrestrial, includes the genus *Oxy* sp. (family Acrididae) and *Tetrix subulata* (family Tetrigidae), identified as aquatic and semi-aquatic species with a relative abundance of 8.1%. This observation supports findings by Merritt and Cummins (1996) regarding semi-aquatic and aquatic behavior in North American species. *Baetis fuscatus* (family Baetidae) a single representive species of the order Ephemeroptera accounts for 5.8% of the total fauna.

Enumeration of the aquatic bodies reveal five semi-aquatic and aquatic spider species, comprising RA: 4.5% of the total population. These species include the Thin-legged wolf spider and Spotted fishing spider from the family Lycosidae; *Dolomedes* sp., the White Flank spider from the family *Pisauridae* and *Argyroneta aquatica* from the family Cybaeidae within the order Araneae. The unique aquatic habitat of *Argyroneta aquatica* is corroborated by Kang *et al.* (2014), who noted that this species is the only spider that spends its entire life submerged in water. In contrast, species from the families Pisauridae and Lycosidae are semi-aquatic and are frequently observed near ponds, wetlands, and rivers, as documented by Carico (1973) and Perevozkin *et al.* (2004). Subsequent to the order Araneae, the order Diptera was represented by *Chironomous* sp. (family Chironomidae) and a Limoniid Crane Fly larva (family Limoniidae), collectively accounting for RA: 3.1%. The water mite *Hydracarina* sp. (family Hydrachnidae), as noted by Pesic *et al.* (2013) representing the order Trombidiformes, constituted RA: 1.1% of the total population.

Table 1: Species wise RA (%) of Macro invertebrates in Bao rice (DWR) field of Lakhimpur District

S. No.	Taxa.	Monsoon		Early Post monsoon		Late Post monsoon		Pre-Monsoon	
1	Thin legged wolf spider	0.63	SBR	1.84	RCD	0.22	SBR	0.98	SBR
2	Spotted fishing spider.	0.33	SBR	0.91	SBR	0.17	SBR	0.48	SBR
3	*Dolomedes* sp.	0.42	SBR	0.77	SBR	0.25	SBR	0.82	SBR
4	White flank water spider	1.03	SBR	2.85	RCD	0.06	SBR	0.99	SBR
5	*Argyroneta aquatica*	0.63	SBR	3.33	SBD	0.41	SBR	2.01	RCD
6	Water Mite (Hydracarina)	1.54	RCD	0.81	SBR	0.80	SBR	1.44	RCD
7	*Macrobrachium lammerii*	1.57	RCD	0.86	SBR	0	---	0	---
8	*Liotelphusa laevis*	0.25	SBR	0.22	SBR	0.15	SBR	0	---
9	*Lobothelphusa woodmasoni*	1.24	RCD	0	---	0.92	SBR	0.41	SBR
10	*Oxya* sp.	1.84	RCD	6.93	SBD	1.52	RCD	1.30	RCD
11	*Tetrix subulata*	10.55	DMN	7.90	SBD	0.85	SBR	2.41	RCD
12	*Somatochlora* sp	4.12	SBD	1.31	RCD	0.63	SBR	2.44	RCD
13	*Sympetrum striolatum*	1.55	RCD	1.03	SBR	1.68	RCD	3.68	SBD
14	*Ischnura* sp	10.38	DMN	1.90	RCD	0.90	SBR	9.17	SBD
15	*Ceriagrion* sp.	2.29	RCD	1.10	RCD	2.75	RCD	1.82	RCD
16	*Baetis fuscatus*	11.00	DMN	3.12	RCD	0	---	9.14	SBD
17	*Gerris* sp.	2.89	RCD	1.61	RCD	2.99	RCD	5.56	SBD
18	*Hydrometra* sp	2.47	RCD	1.40	RCD	10.49	DMN	4.25	SBD
19	Backswimmer	3.72	SBD	0.86	SBR	2.26	RCD	9.19	SBD
20	*Paraplea* sp.	2.27	RCD	1.04	SBR	10.21	DMN	9.42	SBD
21	*Ranatra* sp.	1.36	RCD	3.67	SBD	10.10	DMN	6.13	SBD
22	*Lethocerus indicus*	0.60	SBR	1.78	RCD	1.92	RCD	3.14	RCD
23	*Laccotrephes ruber*	2.27	RCD	0.30	SBR	4.50	SBD	1.20	RCD
24	*Diplonychus rusticus*	2.10	RCD	1.27	RCD	4.80	SBD	0.37	SBR
25	*Abedus herberti*	1.96	RCD	0.92	SBR	4.32	SBD	0.30	SBR
26	*Micronecta silva*	1.94	RCD	1.08	RCD	2.12	RCD	4.88	SBD
27	*Cybister fimbriolatus*	0.27	SBR	2.68	RCD	2.48	RCD	0.93	SBR

28	*Cybister sugilatus*	0.86	SBR	4.50	SBD	2.63	RCD	2.07	RCD
29	*Hygrotus* sp.	1.09	RCD	3.18	SBD	2.88	RCD	0.25	SBR
30	*Laccophilus* sp.	0.65	SBR	1.45	RCD	2.64	RCD	0.16	SBR
31	Water Scavanger Beetle	0.85	SBR	2.13	RCD	2.79	RCD	0	SBR
32	*Chironomous* sp.	1.29	RCD	0.37	SBR	2.64	RCD	4.23	SBD
33	Limoniid crane fly larva	0.88	SBR	0.32	SBR	2.66	RCD	0	----
34	*Planorbis* sp.	1.46	RCD	10.02	DMN	0.85	SBR	0	----
35	*Brotia costula*	1.32	RCD	3.07	RCD	0.79	SBR	0	----
36	*Margaritifera* sp.	1.61	RCD	3.74	SBD	0.87	SBR	0.37	SBR
37	*Pila globossa*	1.33	RCD	2.73	RCD	0.79	SBR	0.30	SBR
38	*Aeolosoma headleyi*	5.56	SBD	2.74	RCD	0	---	1.03	SBR
39	*Branchiodilus* sp.	3.59	SBD	4.79	SBD	2.12	RCD	2.95	RCD
40	*Dero* sp.	2.24	RCD	1.66	RCD	2.51	RCD	1.41	RCD
41	*Aulophorus* sp.	3.59	SBD	3.40	SBD	2.20	RCD	1.72	RCD
42	*Pristina brevista*	2.53	RCD	1.39	RCD	1.99	RCD	0.96	SBR
43	*Limnodrilus hoffmeisteri*	0	---	3.23	SBD	4.36	SBD	2.13	RCD

*Relative abundance: <1= Subrecedent (SBR); 1.1-3.1 = Recedent (RCD); 3.2-10 = Subdominant (SBD); 10.1-31.6=Dominant (DMN); and >31.7%=Eudominant

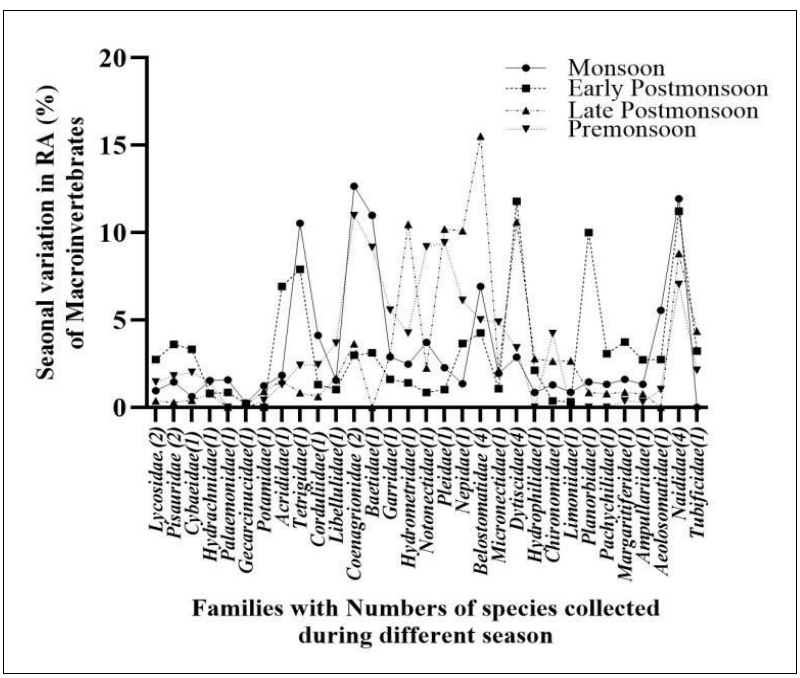

Fig. 1: Family wise abundance and dominance of macroinvertebrates during different seasons.

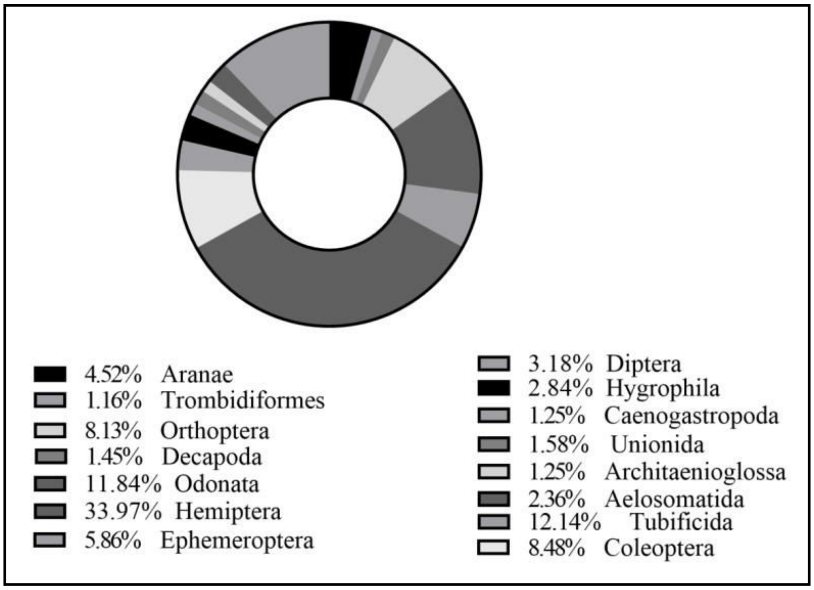

Fig. 2: Order wise Relative abundance (%) of aquatic Macroinvertebrates.

Decapoda, comprising 1.5% of the total population, includes three species: *Macrobrachium lammerii* (family Palaemonidae), *Liotelphusa laevis* (family Gecarcinidae), and *Lobothelphusa woodmasoni* (family Potamidae). Among Mollusca, Hygrophila is moderately represented with a 2.4% relative abundance, featuring *Planorbis* sp. (family Planorbidae). The order Caenogastropoda, represented by *Brotia costula* (family Pachychilidae), Architaenioglossa, represented by *Pila globossa* (family Ampullariidae), and the Unionida, represented by *Margaritifera* sp. (family Margaritiferidae), each accounted for less than 1.5% of the total population.

The relative abundance and dominance rank of macroinvertebrates in the DWR field were calculated using Engelman's Scale and are detailed in Table 1. Despite the consistent number of taxa observed across seasons, their relative abundance varied. During the monsoon and early post-monsoon seasons, 42 species were recorded, while the late post-monsoon and pre-monsoon seasons had 40 and 38 species, respectively (Table 2). The minimal human and animal interference, coupled with rapid rice stem growth during the monsoon in the bao rice field, resulted in higher species populations compared to other crop fields. Dominant species in the monsoon included *Baetis fuscatus*, *Tetrix subulata*, and *Ischnura* sp., alongside 5 subdominant, 12 subrecedent, and 22 recedent species (Table 1). Previous reports by Abhilash and Mahadevaswamy (2022) also noted the dominance of *B. fuscatus* and *Ischnura* sp. during the monsoon in Mysore, India. In the early post-monsoon, while the species count remained unchanged, only *Planorbis* sp. showed dominance, accompanied by 10 subdominant, 12 subrecedent, and various recedent species (Table 1). During the late post-monsoon, sporadic water accumulation in rice fields and ponds resulted in 40 taxa, with *Hydrometra* sp., *Paraplea* sp. and Ranatra sp. as dominant species, 4 subdominant, 15 subrecedent, and 18 recedent species (Table 1). In the early pre-monsoon, ploughing activities led to fewer specimens, but post-first rains, the faunal community increased, with 10 subdominant, 15 subrecedent, and 12 recedent species, although no dominant taxa were recorded (Table 1).

The derived seasonal Shannon-Wiener index values (Table 3) for the DWR field were 3.2, 3.27, 3.33, and 3.5 during the monsoon, early post-monsoon, late post-monsoon, and pre-monsoon

Table 2: Species wise Seasonal dominance of different macro invertebrates

No. of Species	Species	Monsoon	Early Post-Monsoon	Late Post-Monsoon	Pre-Monsoon	Total
1	Thin legged wolf spider	63	151	21	88	323
2	Spotted fishing spider	33	74	16	43	166
3	*Dolomedes* sp.	42	63	24	74	203
4	White flank water spider	103	234	5	89	431
5	*Argyroneta aquatica*	63	273	40	181	557
6	Water Mite. (Hydracarina)	154	66	79	130	429
7	*Macrobrachium lammerii.*	157	70	0	0	227
8	*Liotelphusa laevis*	25	18	14	0	57
9	*Lobothelphusa woodmasoni*	124	0	91	37	252
10	*Oxya* sp.	184	569	151	117	1021
11	*Tetrix subulata*	1055	649	84	217	2005
12	*Somatochlora* sp.	412	107	62	220	801
13	*Sympetrum striolatum*	155	84	167	332	738
14	*Ischnura* sp.	1038	156	89	827	2110
15	*Ceriagrion* sp.	229	90	273	164	756
16	*Baetis fuscatus*	1100	256	0	825	2181
17	*Gerris* sp.	289	132	297	502	1220
18	*Hydrometra* sp.	247	115	1042	383	1787
19	Backswimmer	372	70	224	829	1495
20	*Paraplea* sp.	227	85	1015	850	2177
21	*Ranatra* sp.	136	301	1004	553	1994
22	*Lethocerus indicus*	60	146	190	283	679
23	*Laccotrephes ruber*	227	24	447	108	806
24	*Diplonychus rusticus*	210	104	477	33	824
25	*Abedus herberti*	196	75	429	27	727
26	*Micronecta silva*	194	88	210	440	932
27	*Cybister fimbriolatus*	27	220	246	84	577
28	*Cybister sugilatus*	86	369	261	187	903
29	*Hygrotus* sp.	109	261	286	23	679
30	*Laccophilus* sp.	65	119	262	14	460
31	Water Scavanger Beetle	85	175	277	0	537
32	*Chironomous* sp.	129	30	262	382	803
33	Limoniid crane fly larva	88	26	264	0	378
34	*Planorbis* sp.	146	823	84	0	1053
35	*Brotia costula*	132	252	78	0	462
36	*Margaritifera* sp.	161	307	86	33	587
37	*Pila globossa*	133	224	78	27	462
38	*Aeolosoma headleyi*	556	225	00	93	863
39	*Branchiodilus* sp.	359	393	210	266	1228
40	Dero spp.	224	136	249	127	736
41	*Aulophorus* sp.	359	279	218	155	1011
42	*Pristina brevista*	253	114	197	87	651
43	*Limnodrilus hoffmeisteri*	000	265	433	192	901
		10007	8218	9942	9022	37189

seasons, respectively. These values indicate a healthy ecosystem with high species diversity, as higher Shannon index values correspond to more diverse and robust ecosystems (Metcalfe, 1989). The evenness score ranged from 0.64 to 0.74, with the highest in the early post-monsoon season, suggesting a semi-balanced habitat (Pielou, 1975). The Berger-Parker index values, ranging from 0.09 to 0.1, indicate moderate pollution levels, supporting the semi-balanced habitat without dominant species (Padmanabha, 2011). Margalef's diversity index ranged from 8.12 to 9.38 (Table 3) reflecting high species richness and clean conditions year-round. Govindasamy *et al.* (2016)

Table 3: Seasonal variation of relative abundance in the diversity indices scores of macro invertebrates species in DWR field

Indices	Monsoon	Early Post-monsoon	Late Post-monsoon	Pre-monsoon
Taxa_S	42	42	40	37
Shannon_H	3.331	3.437	3.278	3.17
Evenness_e^H/S	0.6658	0.74	0.6629	0.6436
Margalef	9.35	9.38	8.95	8.13
Berger-Parker index	0.1099	0.09979	0.09978	0.08996
BMWP	114	109	104	93
ASPT	4.95	4.73	4.95	4.65

reported lower Margalef index values in South Indian rice fields. The BMWP index ranged from 93 to 114, indicating acceptable to good water quality, with the highest score of 119 during the monsoon due to pollution-sensitive species like *Baetis fuscatus*, *Ischnura* sp*.,* and *Tetrix subulata* (Kazanci, 2010). The ASPT scores ranged from 4.6 to 4.9 (Table 3), further indicating good water quality. The presence of pollution-resistant families, such as Hydrometridae (RA 10.4%), Pleidae (RA 10.2%), Belostomidae (RA 15.5%), and Dytiscidae (RA 10.6%), during the late post-monsoon (Table 1, Fig. 1) suggests habitat alteration due to crop harvesting (Foltz and Dodson, 2009; Edegbene *et al.*,2022). The prevalence of Baetidae in the pre-monsoon (RA 9.2%) and early post-monsoon (RA 3.2%) indicates a healthy, stable habitat.

Freshwater Oligochaetes, vital for fish and small aquatic animals (Wissinger, 1999), and major components of submerged rice field invertebrates (Simpson *et al.,* 1993), were collected throughout the seasons. Addressing the unexplored status of aquatic oligochaetes in Assam and the Northeast India (Fernando, 1993; Singh *et al.,* 2009), this study identified six aquatic Oligochaete species from the DWR fields in Lakhimpur district, Assam, viz. *Aeolosoma headleyi* (Aulosomatidae), *Branchiodilus* sp., *Dero* sp., *Aulophorus* sp., *Pristina brevista* (Naididae), and *Limnodrilus hoffmeisteri* (Tubificidae). Four of these species are reported for the first time from Assam, North East India, highlighting the importance of DWR fields for biodiversity and as a genetic reservoir for aquatic organisms in a flood-prone region.

Conclusion

The current study revealed that the Red rice field (DWR) is an anthropogenically undisturbed wetland, serving as a vital habitat for numerous aquatic macroinvertebrates. 43 macroinvertebrate species from the DWR field were identified, distributed across 31 families and 15 orders. Seasonal variations in community structure and species diversity were minimal, with a notable even distribution of species in the early post-monsoon season, indicating it as a favorable period for the emergence of various taxa. The presence of pollution-sensitive species like *Baetis fuscatus* and high species richness, as indicated by Margalef's diversity index, reflect a healthy ecosystem. The discovery of six aquatic Oligochaete species, four newly documented in Assam, underscores the DWR field's importance in supporting biodiversity and acting as a genetic reservoir for aquatic organisms in flood-prone regions. Given the lack of comprehensive studies on aquatic macroinvertebrates and Oligochaete diversity in the BAO rice fields, continuous research is essential to establish baseline data for future investigations.

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