Fig. 34. Glutathione-S-transferase activity in *Lamellidens corrianus* bivalves collected from different sampling sites of Sirpur and Bilawali Tanks of Indore in different seasons.

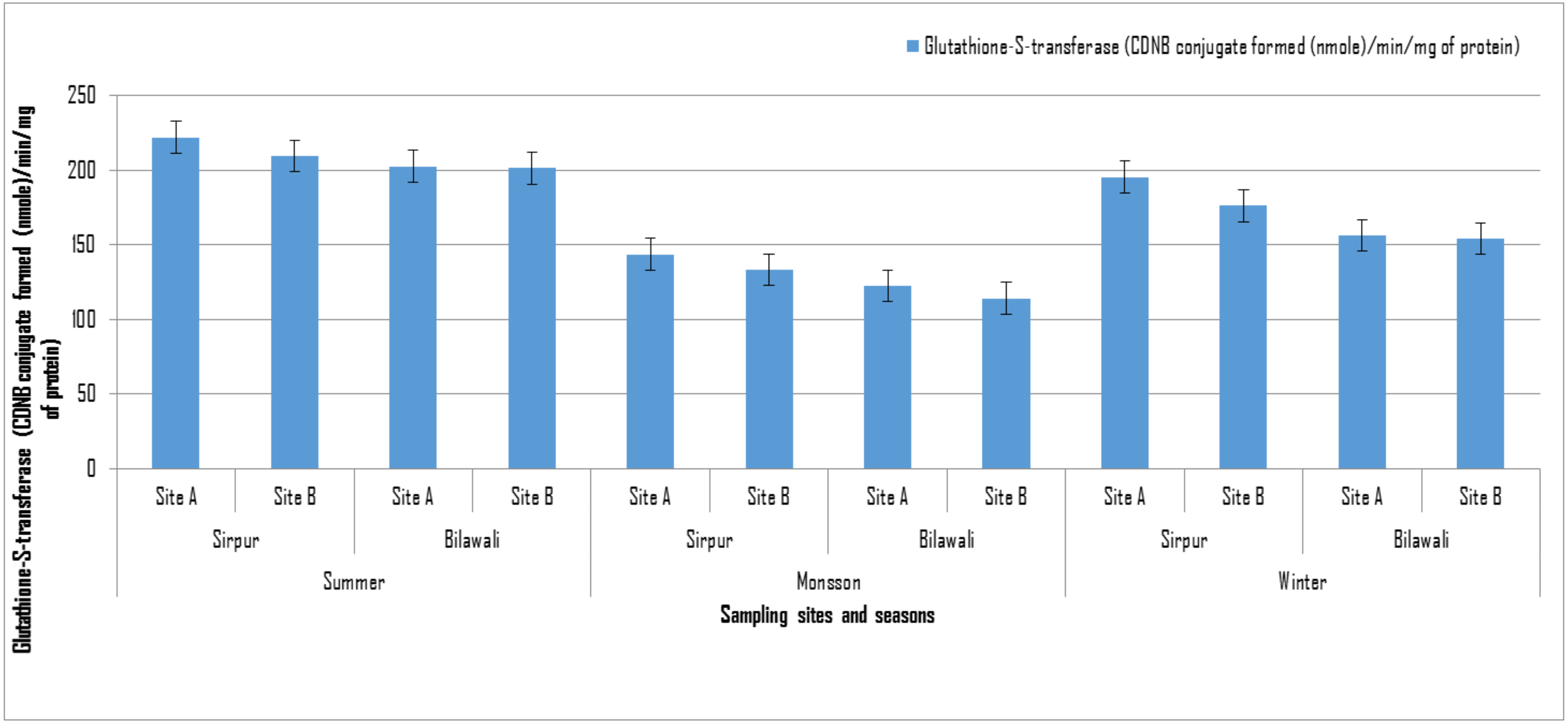
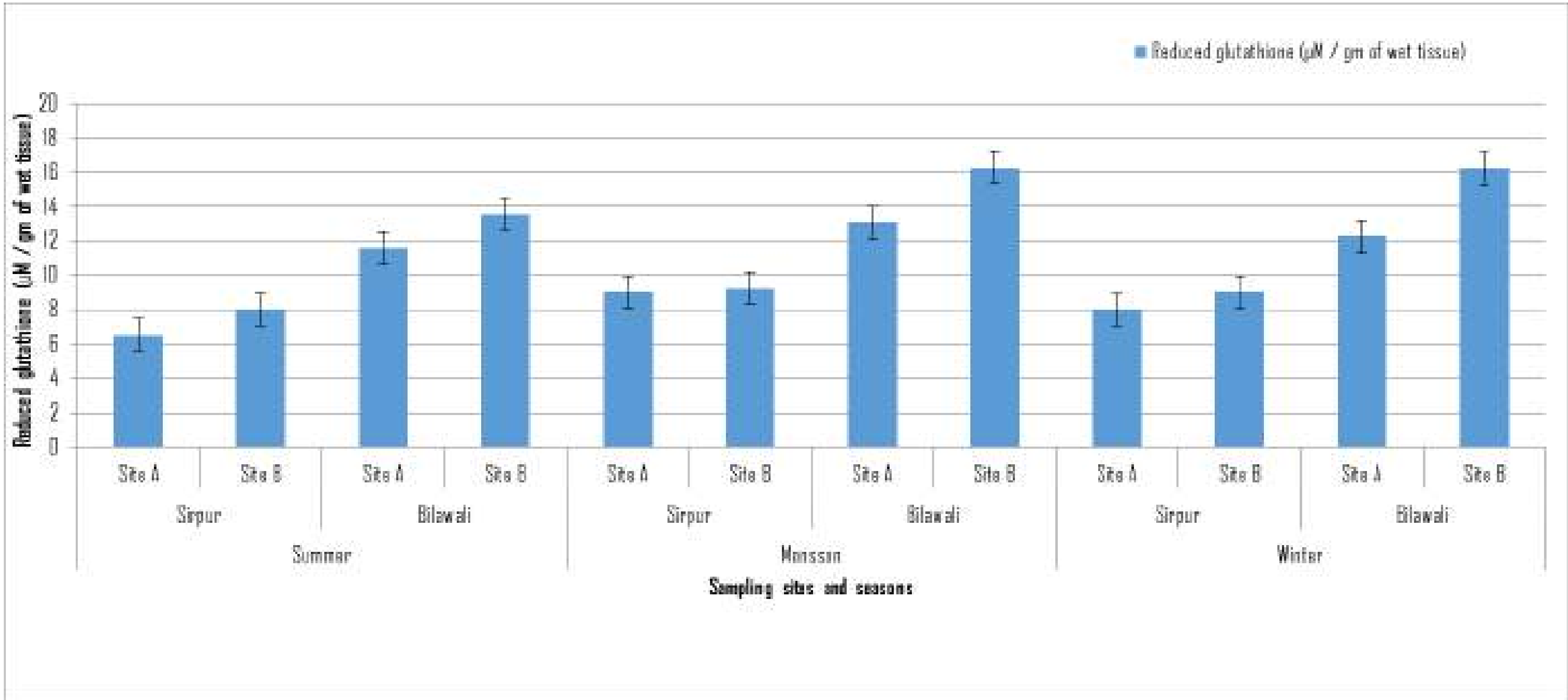


Fig. 35. Reduced glutathione activity in *Lamellidens corrianus* bivalves collected from different sampling sites of Sirpur and Bilawali Tanks of Indore in different seasons.



13.58±0.21 in summer; 13.09±0.13 and 16.28±0.11 in monsoon; 12.29±0.12 and 16.24±0.17 in winter seasons respectively.

3.4.1.4 Superoxide dismutase:

Superoxide dismutase with standard error per mg protein in collected *Lamellidens corrianus* bivalve samples of Sirpur and Bilawali Tank in summer, monsoon and winter seasons is shown in Table 7. Superoxide dismutase with standard deviation per mg protein in *Lamellidens corrianus* bivalve samples collected from Sirpur Lake site A and site B were 119.84±1.29 and 125.64±1.39 in summer; 149.84±1.92 and 169.24±2.09 in monsoon; 128.36±1.28 and 151.31±1.34 in winter seasons whereas, Bilawali Tank site A and site B were reported 135.47±2.58 and 143.57±1.97 in summer; 190.18±2.16 and 201.26±2.84 in monsoon; 149.75±1.19 and 160.75±1.17 in winter seasons respectively.

3.4.1.5 Catalase

Catalase with standard error per mg protein in collected *Lamellidens corrianus* bivalve samples of Sirpur and Bilawali Tank in summer, monsoon and winter seasons is shown in Table 7. Catalase with standard deviation per mg protein in *Lamellidens corrianus* bivalve samples collected from Sirpur Lake site A and site B were 92.35±1.14 and 93.27±1.18 in summer; 131.06±1.84 and 132.56±1.79 in monsoon;

Fig. 36. Superoxide dismutase activity in *Lamellidens corrianus* bivalves collected from different sampling sites of Sirpur and Bilawali Tanks of Indore in different seasons.

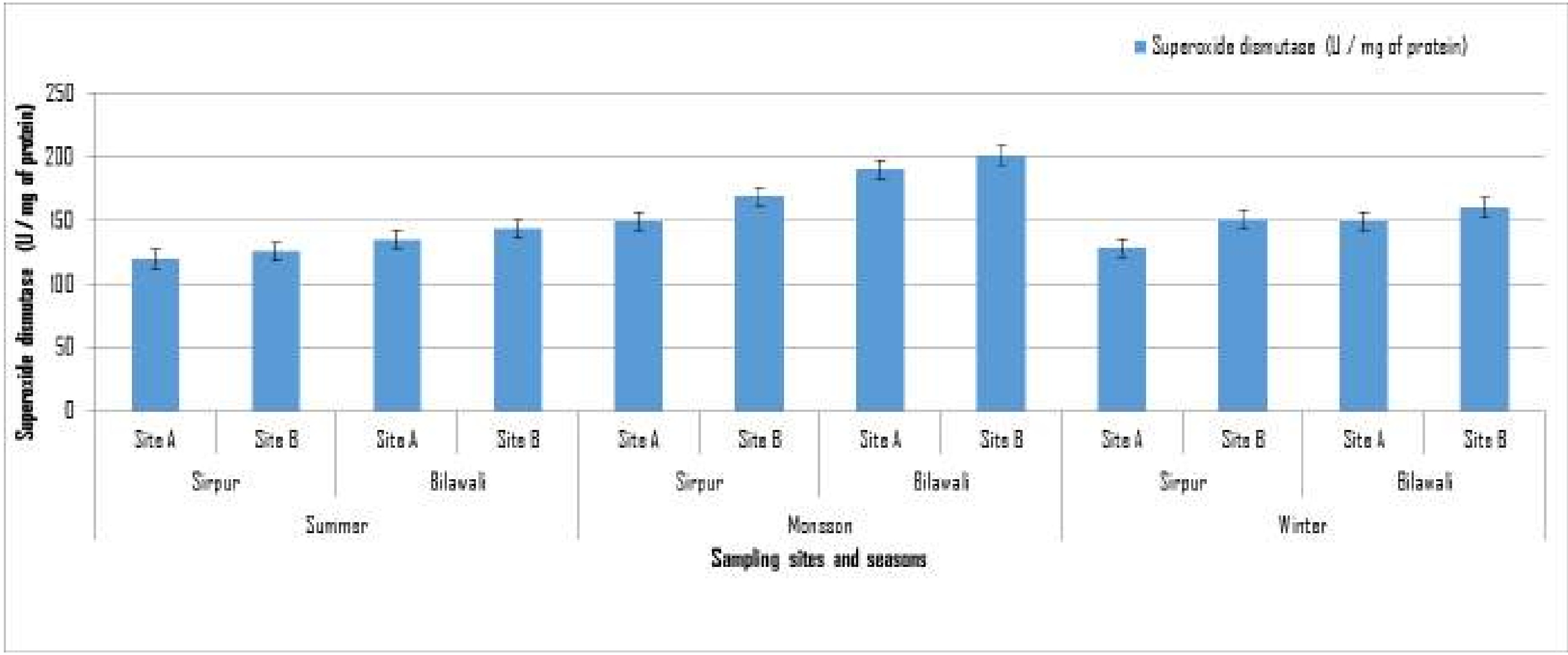
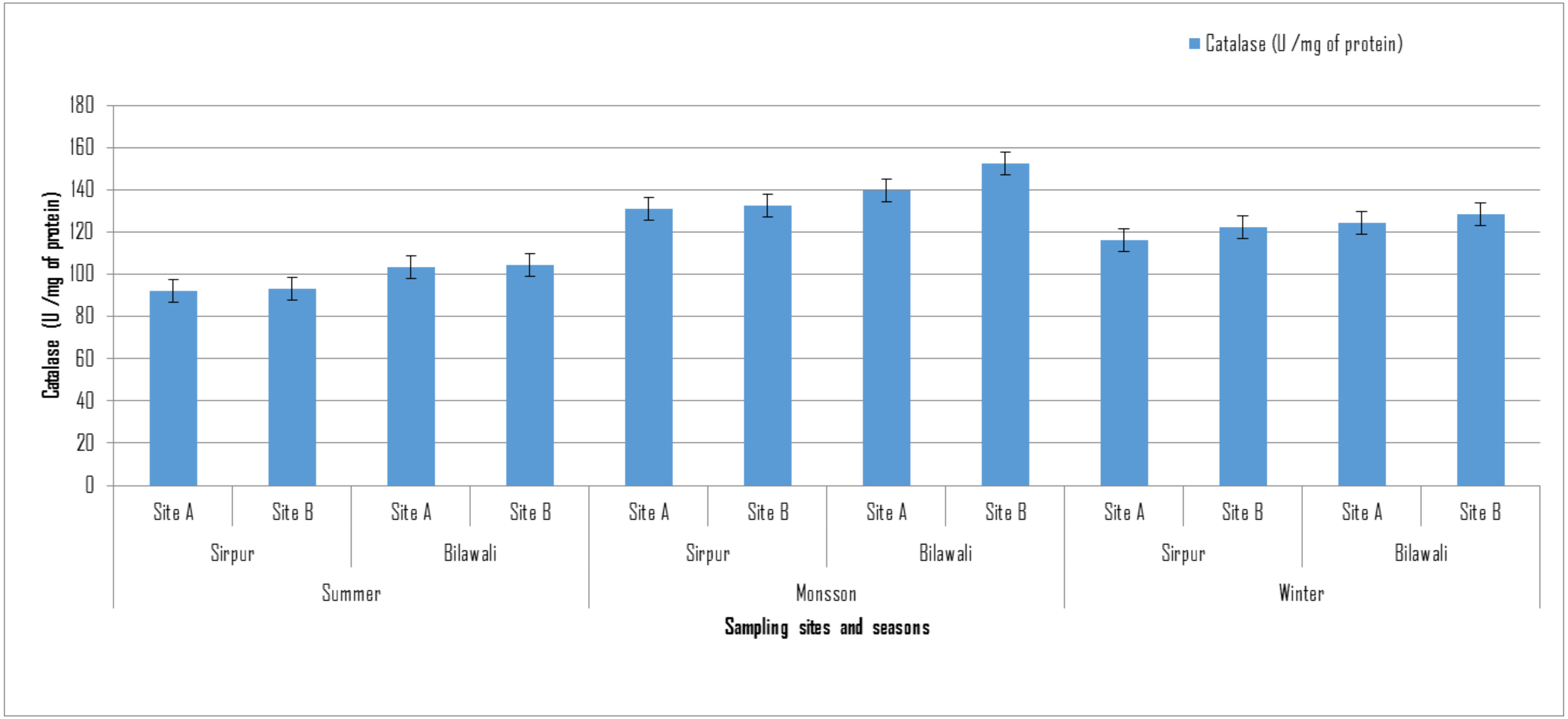


Fig. 37. Catalase activity in *Lamellidens corrianus* bivalves collected from different sampling sites of Sirpur and Bilawali Tanks of Indore in different seasons.

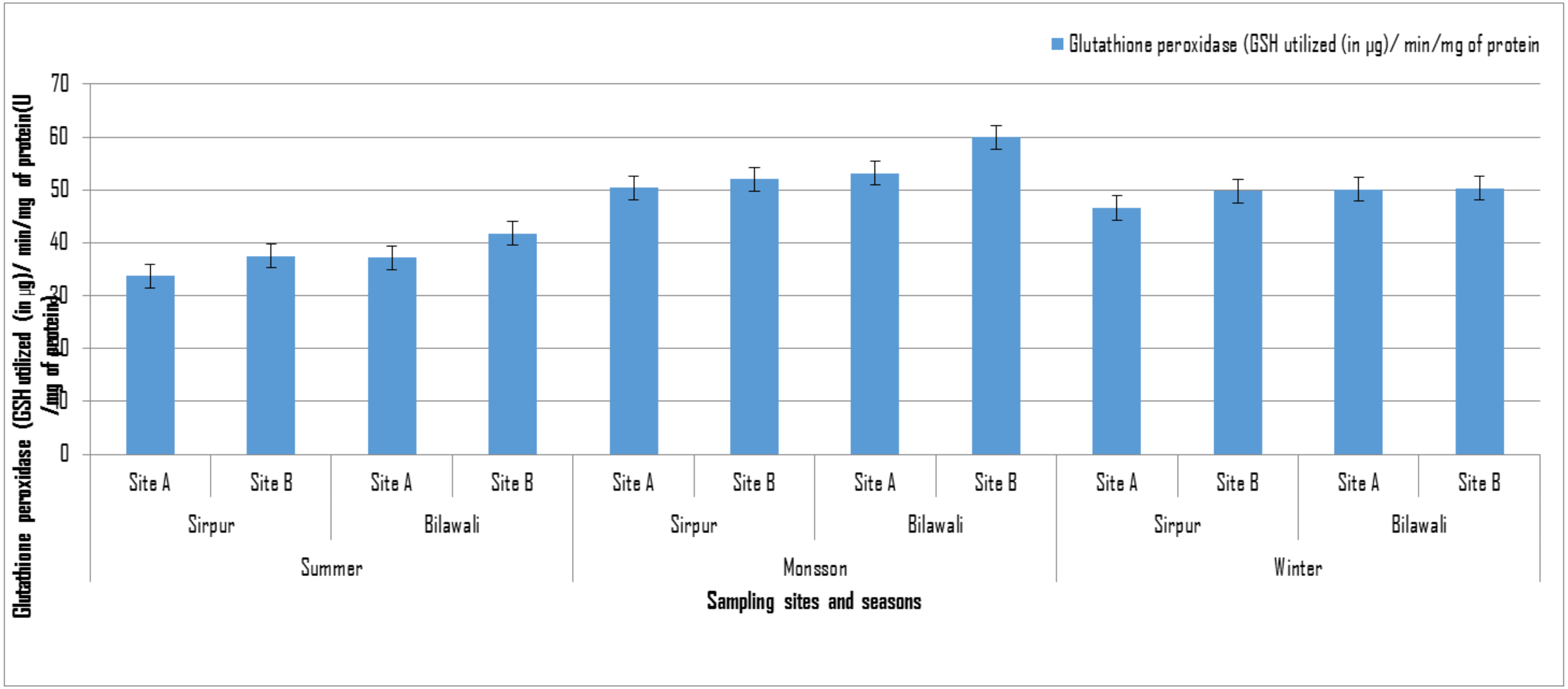


116.24±1.16 and 122.58±1.29 in winter seasons whereas, Bilawali Tank site A and site B were reported 103.52±2.13 and 104.26±1.96 in summer; 139.86±1.29 and 152.45±1.09 in monsoon; 124.51±1.05 and 128.36±1.76 in winter seasons respectively.

3.4.1.6 Glutathione peroxidase

Superoxide dismutase with standard error per mg protein in collected *Lamellidens corrianus* bivalve samples of Sirpur and Bilawali Tank in summer, monsoon and winter seasons is shown in Table 7. Superoxide dismutase with standard deviation per mg protein in *Lamellidens corrianus* bivalve samples collected from Sirpur Lake site A and site B were 33.75±0.26 and 37.45±0.23 in summer; 50.42±0.17 and 52.01±0.14 in monsoon; 46.59±0.11 and 49.78±0.11 in winter seasons whereas, Bilawali Tank site A and site B were reported 37.19±0.22 and 41.79±0.16 in summer; 53.18±0.11 and 60.01±0.14 in monsoon; 50.14±0.09 and 50.28±0.08 in winter seasons respectively.

Fig. 38. Glutathione peroxidase activity in *Lamellidens corrianus* bivalves collected from different sampling sites of Sirpur and Bilawali Tanks of Indore in different seasons.



3.4.2 *Lamellidens marginali*

Table 8 presents the mean with standard deviation of collected observations of oxidative biomarkers in *Lamellidens marginali* samples.

Table 8. Seasonal variations of oxidative stress in *Lamellidens marginali* collected from Sirpur and Bilawali Tanks of Indore.

Parameters	**Summer**				**Monson**				**Winter**			
	Sirpur		**Bilawali**		**Sirpur**		**Bilawali**		**Sirpur**		**Bilawali**	
	Site A	**Site B**	**Site A**	**Site B**	**Site A**	**Site B**	**Site A**	**Site B**	**Site A**	**Site B**	**Site A**	**Site B**
Lipid Peroxidation	216.59± 4.51	196.28± 4.44	191.26± 4.19	179.94± 4.63	143.24± 4.72	126.59± 3.87	121.05± 3.88	118.42± 3.09	189.64± 4.92	175.96± 3.51	167.26± 4.01	161.27± 3.12
Glutathione-S-transferase	249.56± 4.63	201.57± 4.06	213.25± 4.02	201.58± 4.17	163.28± 4.98	124.23± 3.67	134.26± 4.01	109.25± 3.11	199.24± 5.02	171.68± 3.45	172.36± 4.03	156.34± 3.09
Reduced glutathione	8.69±0.07	9.02±0.06	15.01±0.09	15.12±0.10	10.24±0.11	9.94±0.11	16.02±0.09	17.23±0.10	9.03±0.08	9.05±0.07	15.46±0.12	16.25±0.09
Superoxide dismutase	121.45± 1.86	123.51± 1.68	138.25± 1.28	149.56± 1.15	165.84± 1.72	178.42± 1.96	194.58± 2.06	194.53± 1.92	128.37± 1.11	149.68± 1.94	146.27± 2.31	155.24± 1.65
Catalase	91.2±1.23	99.27±1.22	98.57±1.14	108.69± 1.10	135.26± 1.39	141.26± 1.78	153.67± 0.09	158.69± 1.49	119.26± 1.09	128.67± 1.78	120.32± 1.98	131.27± 1.53
Glutathione peroxidase	33.01±0.13	38.46±0.12	40.41±0.11	45.62±0.08	46.27±0.12	48.36±0.13	49.15±0.14	54.27±0.14	37.36±0.05	42.26±0.09	44.12±0.10	48.56±0.10

3.4.2.1 Lipid peroxidation

Lipid peroxidation with standard error per mg protein in collected *Lamellidens marginali* bivalve samples of Sirpur and Bilawali Tank in summer, monsoon and winter seasons is shown in Table 8. Lipid peroxidation with standard deviation per mg protein in *Lamellidens marginali* bivalve samples collected from Sirpur Lake site A and site B were 216.59±4.51 and 196.28±4.44 in summer; 143.24±4.72 and 126.59±3.87 in monsoon; 189.64±4.92 and 175.96±3.51 in winter seasons whereas, Bilawali Tank site A and site B were reported 191.26±4.19 and 179.94±4.63 in summer; 121.05±3.88 and 118.42±3.09 in monsoon; 167.26±4.01 and 161.27±3.12 in winter seasons.

3.4.2.2 Glutathione-S-transferase

Glutathione-S-transferase with standard error per mg protein in collected *Lamellidens marginali* bivalve samples of Sirpur and Bilawali Tank in summer, monsoon and winter seasons is shown in Table 8. Glutathione-S-transferase with standard deviation per mg protein in *Lamellidens marginali* bivalve samples collected from Sirpur Lake site A and site B were 249.56±4.63 and 201.57±4.06 in summer;

Fig. 39. Lipid Peroxidation activity in *Lamellidens marginali* bivalves collected from different sampling sites of Sirpur and Bilawali Tanks of Indore in different seasons.

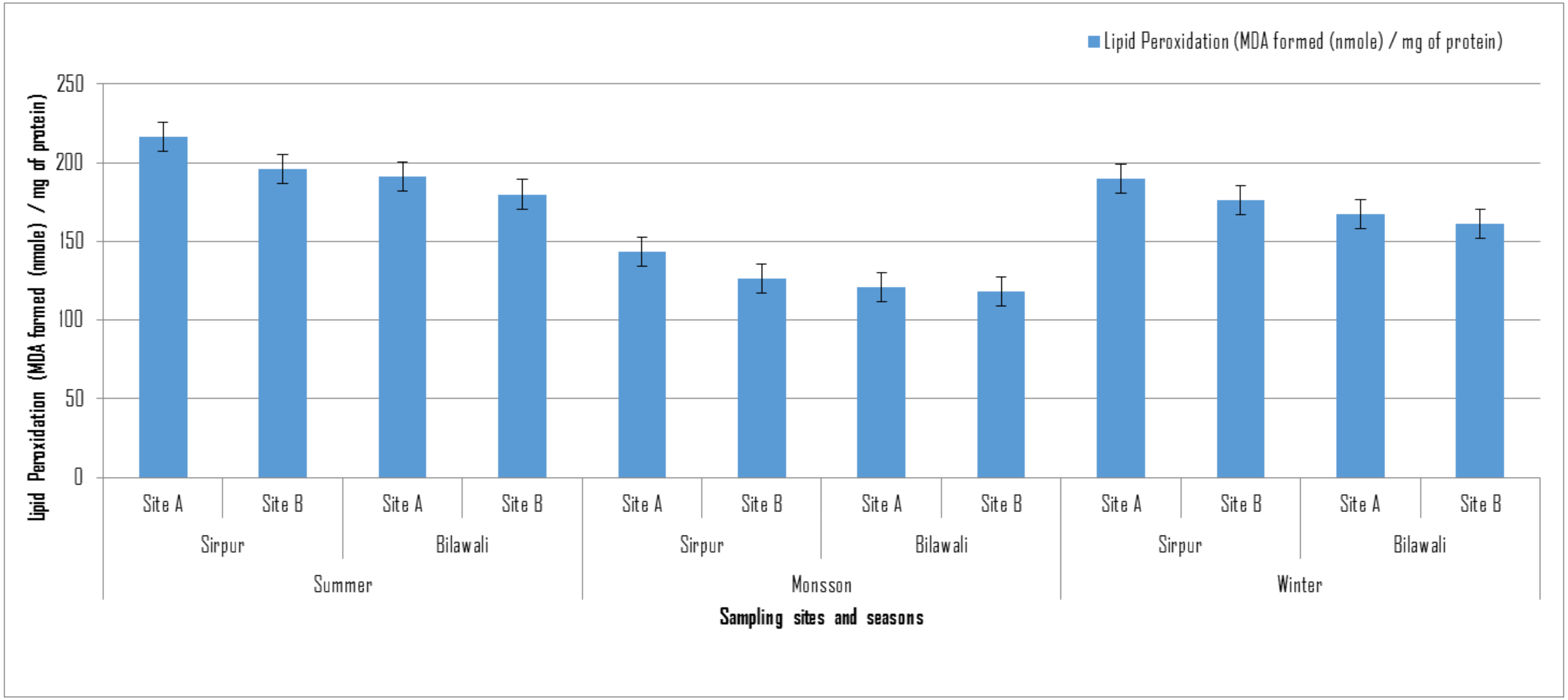
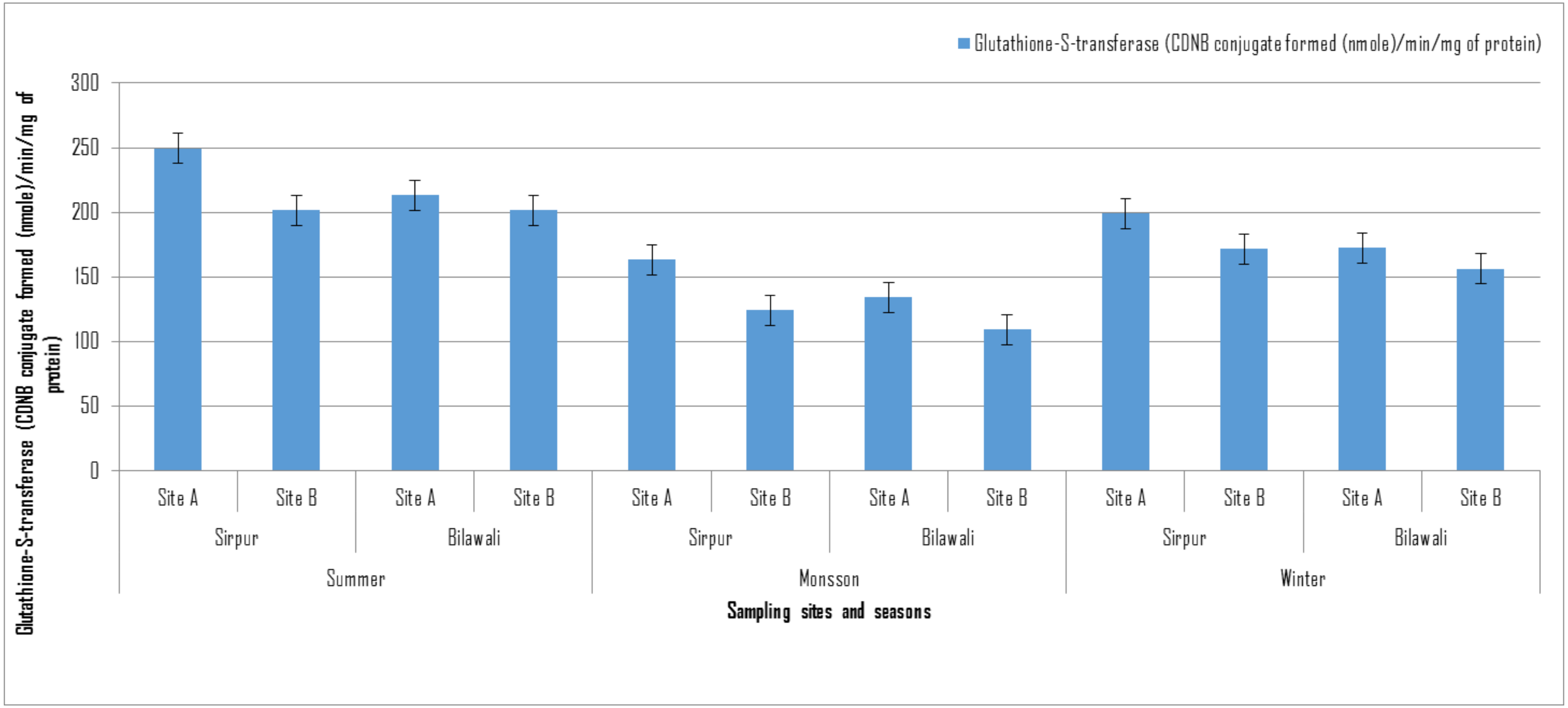


Fig. 40. Glutathione-S-transferase activity in *Lamellidens marginali* bivalves collected from different sampling sites of Sirpur and Bilawali Tanks of Indore in different seasons.



163.28±4.98 and 124.23±3.67 in monsoon; 199.24±5.02 and 171.68±3.45 in winter seasons whereas, Bilawali Tank site A and site B were reported 213.25±4.02 and 201.58±4.17 in summer; 134.26±4.01 and 109.25±3.11 in monsoon; 172.36±4.03 and 156.34±3.09 in winter seasons respectively.

3.4.2.3 Reduced glutathione

Reduced glutathione with standard error per mg protein in collected *Lamellidens marginali* bivalve samples of Sirpur and Bilawali Tank in summer, monsoon and winter seasons is shown in Table 8. Reduced glutathione with standard deviation per mg protein in *Lamellidens marginali* bivalve samples collected from Sirpur Lake site A and site B were 8.69±0.07 and 9.02±0.06 in summer; 10.24±0.11 and 9.94±0.11 in monsoon; 9.03±0.08 and 9.05±0.07 in winter seasons whereas, Bilawali Tank site A and site B were reported 15.01±0.09 and 15.12±0.10 in summer; 16.02±0.09 and 17.23±0.10 in monsoon; 15.46±0.12 and 16.25±0.09 in winter seasons respectively.

3.4.2.4 Superoxide dismutase:

Superoxide dismutase with standard error per mg protein in collected *Lamellidens marginali* bivalve samples of Sirpur and Bilawali Tank in summer, monsoon and winter seasons is shown in Table 8. Superoxide dismutase with standard deviation per mg protein in *Lamellidens*

Fig. 41. Reduced glutathione activity in *Lamellidens marginali* bivalves collected from different sampling sites of Sirpur and Bilawali Tanks of Indore in different seasons.

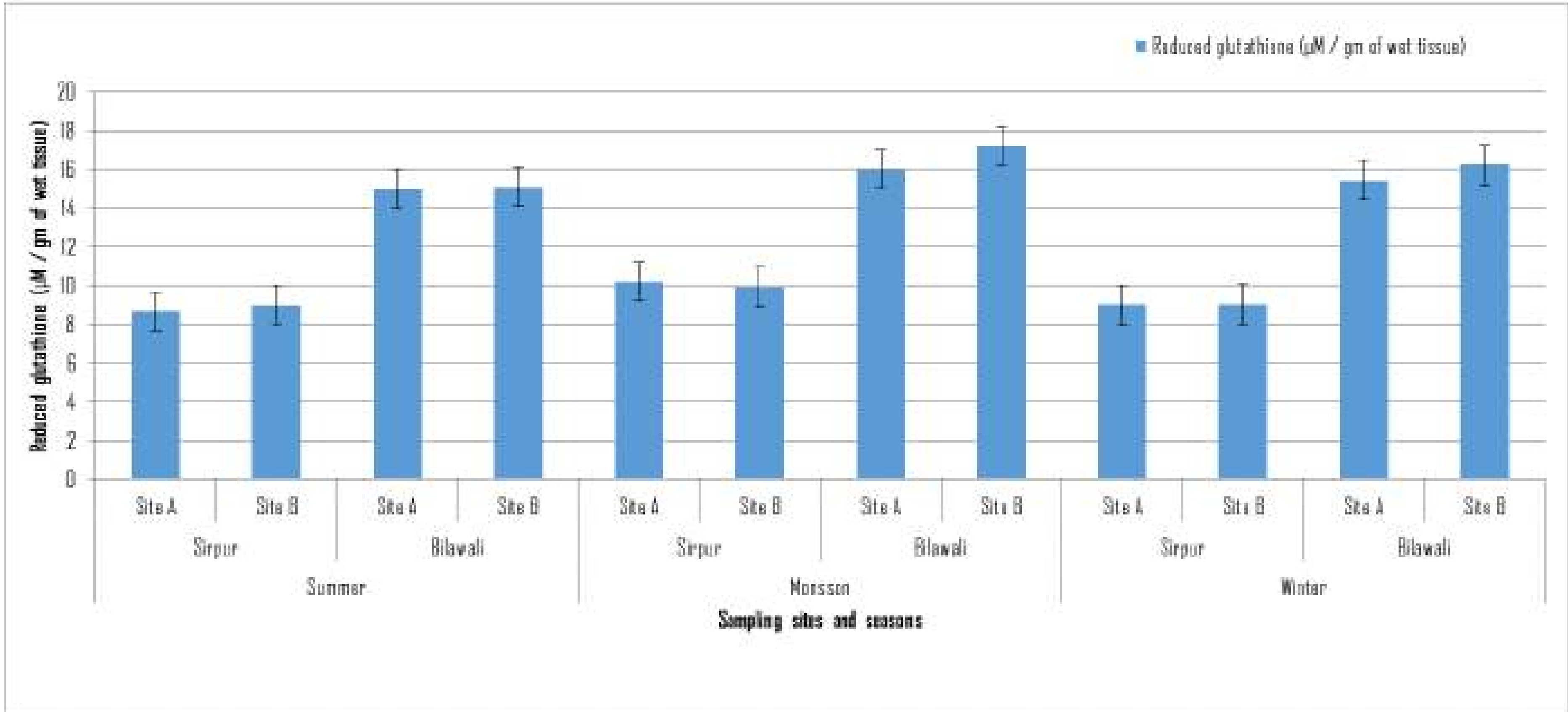
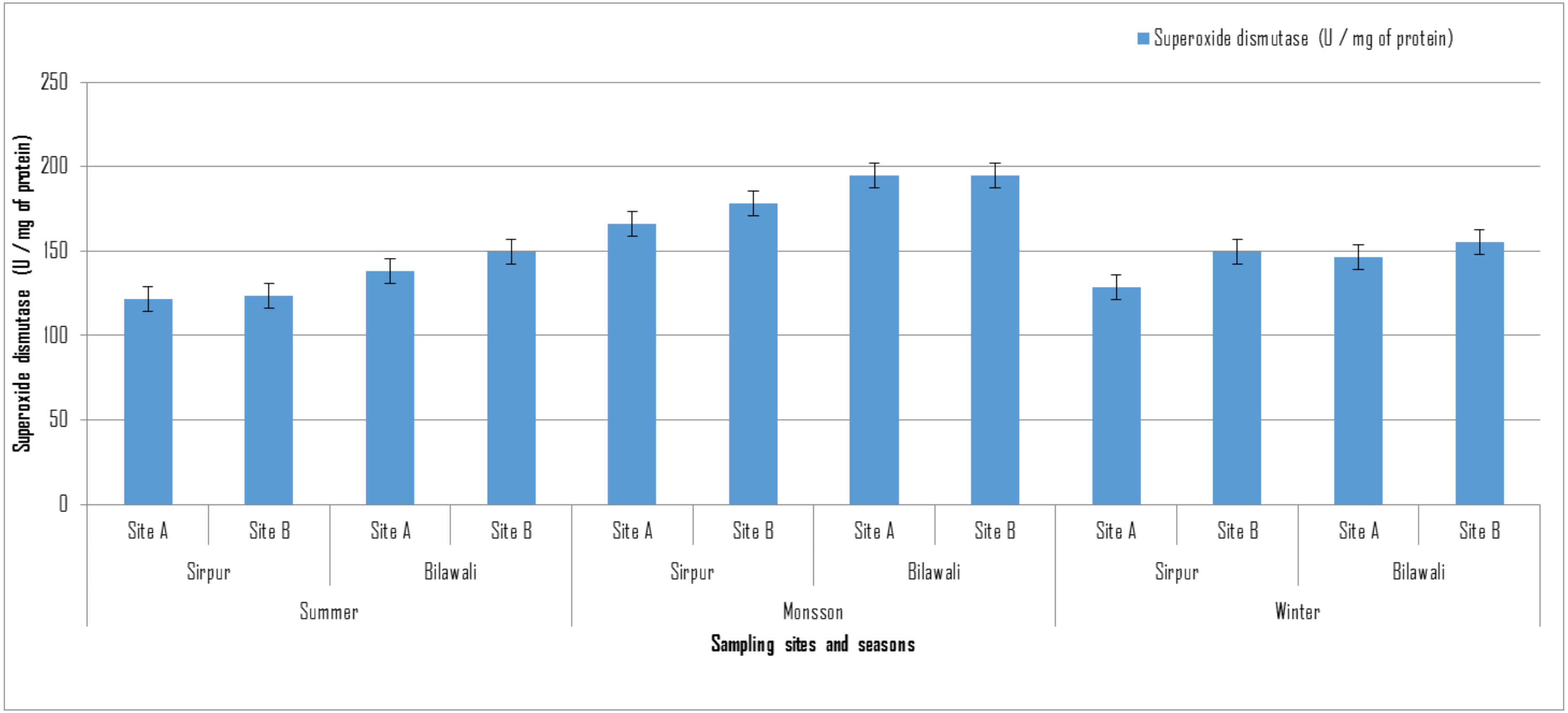


Fig. 42. Superoxide activity in *Lamellidens marginali* bivalves collected from different sampling sites of Sirpur and Bilawali Tanks of Indore in different seasons.



marginali bivalve samples collected from Sirpur Lake site A and site B were 121.45±1.86 and 123.51±1.68 in summer; 165.84±1.72 and 178.42±1.96 in monsoon; 128.37±1.11 and 149.68±1.94 in winter seasons whereas, Bilawali Tank site A and site B were reported 138.25±1.28 and 149.56±1.15 in summer; 194.58±2.06 and 194.53±1.92 in monsoon; 146.27±2.31 and 155.24±1.65 in winter seasons respectively.

3.4.2.5 Catalase

Catalase with standard error per mg protein in collected *Lamellidens marginali* bivalve samples of Sirpur and Bilawali Tank in summer, monsoon and winter seasons is shown in Table 8. Catalase with standard deviation per mg protein in *Lamellidens marginali* bivalve samples collected from Sirpur Lake site A and site B were 91.2±1.23 and 99.27±1.22 in summer; 135.26±1.39 and 141.26±1.78 in monsoon 119.26±1.09 and 128.67±1.78 in winter seasons whereas, Bilawali Tank site A and site B were reported 98.57±1.14 and 108.69±1.10 in summer; 153.67±0.09 and 158.69±1.49 in monsoon; 120.32±1.98 and 131.27±1.53 in winter seasons respectively.

3.4.2.6 Glutathione peroxidase

Superoxide dismutase with standard error per mg protein in collected *Lamellidens marginali* bivalve samples of Sirpur and Bilawali Tank in

Fig. 43. Catalase activity in *Lamellidens marginali* bivalves collected from different sampling sites of Sirpur and Bilawali Tanks of Indore in different seasons.

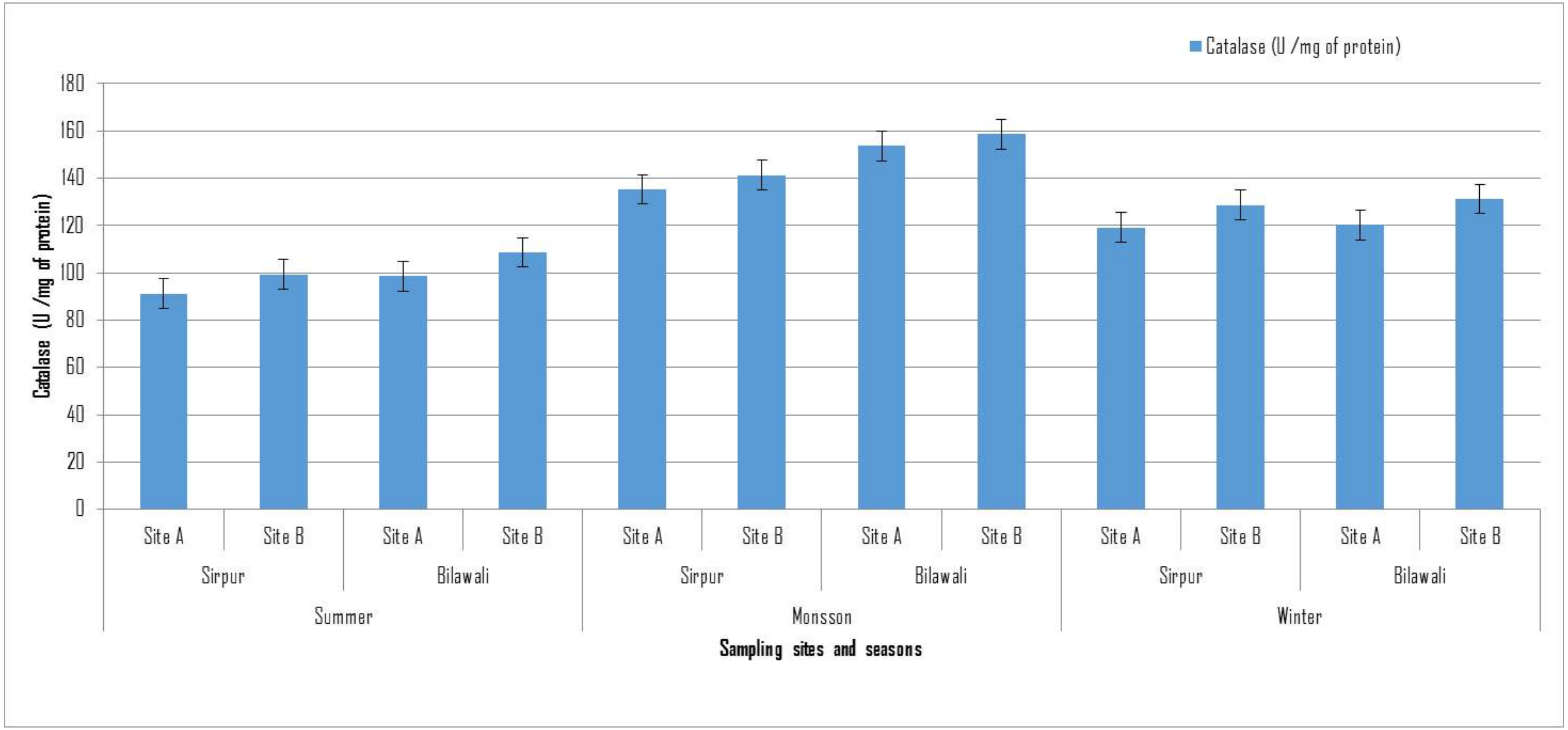
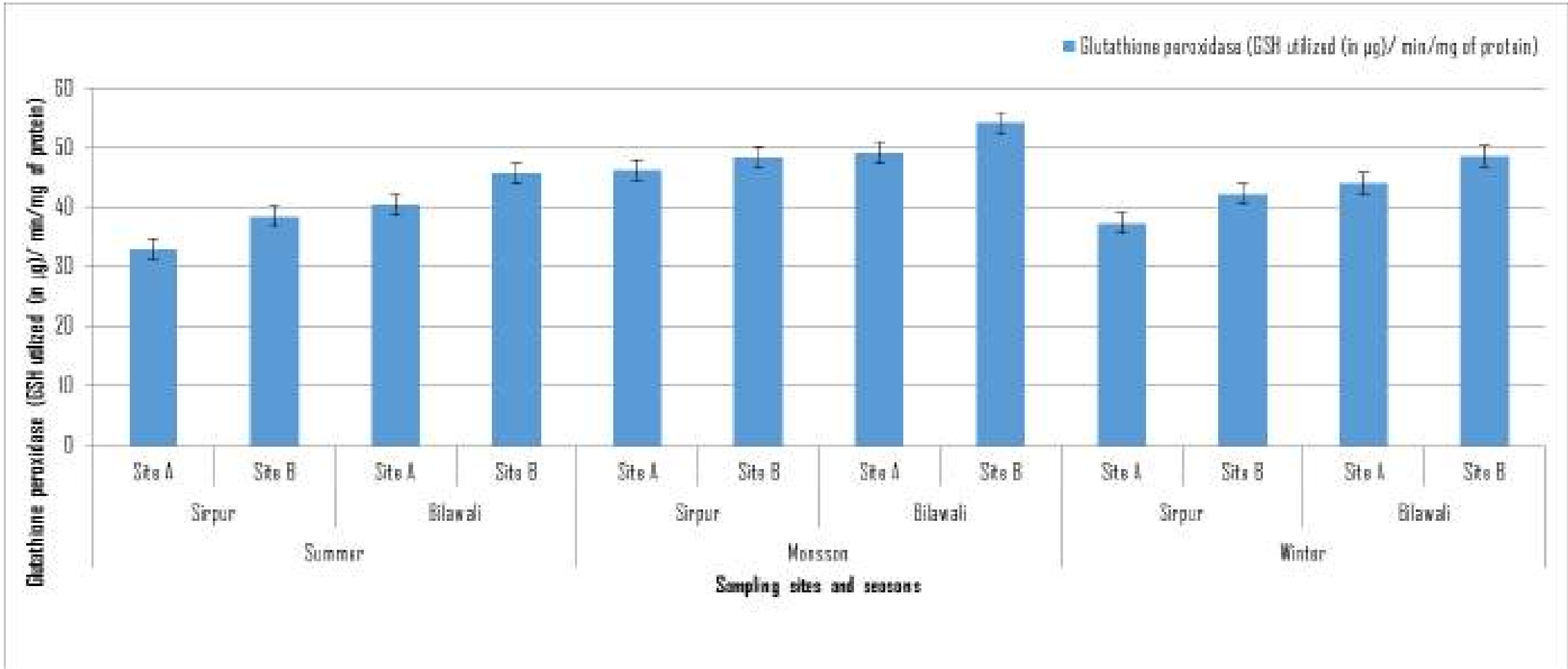


Fig. 44. Glutathione peroxidase activity in *Lamellidens marginali* bivalves collected from different sampling sites of Sirpur and Bilawali Tanks of Indore in different seasons.



summer, monsoon and winter seasons is shown in Table 8. Superoxide dismutase with standard deviation per mg protein in *Lamellidens marginali* bivalve samples collected from Sirpur Lake site A and site B were 33.01±0.13 and 38.46±0.12 in summer; 46.27±0.12 and 48.36±0.13 in monsoon; 37.36±0.05 and 42.26±0.09 in winter seasons whereas, Bilawali Tank site A and site B were reported 40.41±0.11 and 45.62±0.08 in summer; 49.15±0.14 and 54.27±0.14 in monsoon; 44.12±0.10 and 48.56±0.10 in winter seasons respectively.

3.4.3 *Unio* sp.

Table 9 presents the mean with standard deviation of collected observations of oxidative biomarkers in *Unio* sp. samples.

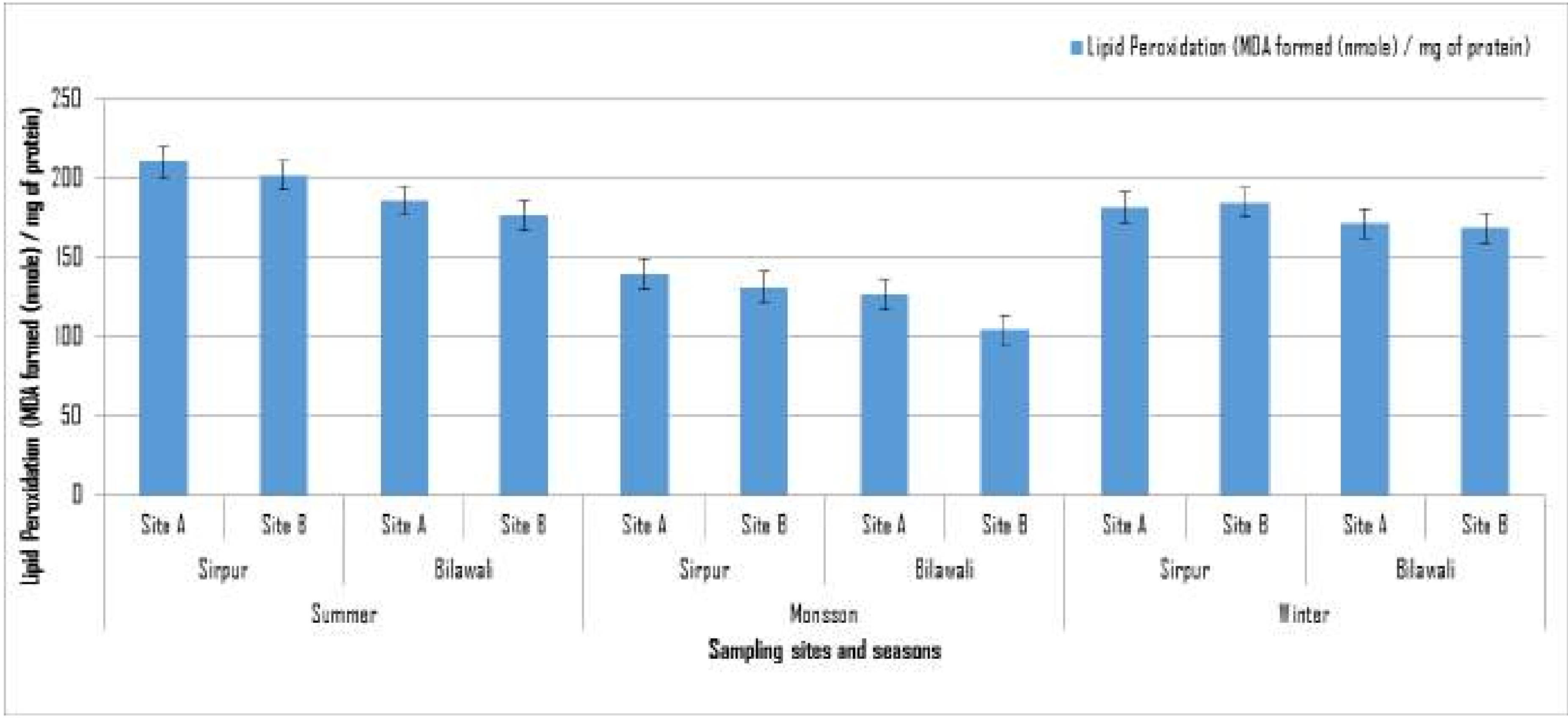
Table 9. Seasonal variations of oxidative stress in *Unio* sp. collected from Sirpur and Bilawali Tanks of Indore.

Parameters	**Summer**				**Monson**				**Winter**			
	Sirpur		**Bilawali**		**Sirpur**		**Bilawali**		**Sirpur**		**Bilawali**	
	Site A	**Site B**	**Site A**	**Site B**	**Site A**	**Site B**	**Site A**	**Site B**	**Site A**	**Site B**	**Site A**	**Site B**
Lipid Peroxidation	210.23±5.21	201.45±5.02	185.74±5.09	176.59±5.42	139.57±4.23	131.42±4.23	126.25±4.67	104.23±3.24	181.45±5.21	184.56±4.96	171.25±4.85	168.46±4.12
Glutathione-S-transferase	230.18±5.01	210.52±4.98	201.56±4.65	201.45±5.86	142.56±4.42	124.62±3.28	119.51±3.68	116.24±3.65	192.45±4.98	171.24±3.99	154.86±3.86	156.28±3.21
Reduced glutathione	5.86±0.09	6.76±0.11	9.23±0.09	11.23±0.08	7.01±0.10	7.59±0.12	11.2±0.11	14.26±0.12	6.95±0.11	6.95±0.12	11.03±0.09	13.06±0.1
Superoxide dismutase	120.45±2.12	125.64±2.03	130.45±1.98	140.25±1.48	160.58±1.56	180.46±1.14	186.54±1.69	191.42±1.94	125.34±2.01	145.36±3.02	141.29±2.38	151.24±2.16
Catalase	88.64±1.01	96.58±1.12	105.28±1.86	105.36±1.23	129.54±1.86	143.51±1.12	154.75±1.23	152.01±1.64	124.23±1.96	121.48±2.12	118.46±2.10	128.34±1.13
Glutathione peroxidase	32.56±0.16	34.27±0.11	36.15±0.14	40.81±0.11	40.16±0.07	46.02±0.10	47.2±0.13	51.2±0.11	39.56±0.12	40.58±0.13	41.03±0.11	44.52±0.12

3.4.3.1 Lipid peroxidation

Lipid peroxidation with standard error per mg protein in collected *Unio* sp. bivalve samples of Sirpur and Bilawali Tank in summer, monsoon and winter seasons is shown in Table 9. Lipid peroxidation with standard deviation per mg protein in *Unio* sp. bivalve samples collected from Sirpur Lake site A and site B were 210.23±5.21 and 201.45±5.02 in summer; 139.57±4.23 and 131.42±4.23 in monsoon; 181.45±5.21 and 184.56±4.96 in winter seasons whereas, Bilawali Tank site A and site B

Fig. 45. Lipid Peroxidation activity in *Unio* sp. bivalves collected from different sampling sites of Sirpur and Bilawali Tanks of Indore in different seasons.



were reported 185.74±5.09 and 176.59±5.42 in summer; 126.25±4.67 and 104.23±3.24 in monsoon; 171.25±4.85 and 168.46±4.12 in winter seasons.

3.4.3.2 Glutathione-S-transferase

Glutathione-S-transferase with standard error per mg protein in collected *Unio* sp. bivalve samples of Sirpur and Bilawali Tank in summer, monsoon and winter seasons is shown in Table 9. Glutathione-S-transferase with standard deviation per mg protein in *Unio* sp. bivalve samples collected from Sirpur Lake site A and site B were 230.18±5.01 and 210.52±4.98 in summer 142.56±4.42 and 124.62±3.28 in monsoon; 192.45±4.98 and 171.24±3.99 in winter seasons whereas, Bilawali Tank site A and site B were reported 201.56±4.65 and 201.45±5.86 in summer; 119.51±3.68 and 116.24±3.65 in monsoon; 154.86±3.86 and 156.28±3.21 in winter seasons respectively.

3.4.3.3 Reduced glutathione

Reduced glutathione with standard error per mg protein in collected *Unio* sp. bivalve samples of Sirpur and Bilawali Tank in summer, monsoon and winter seasons is shown in Table 9. Reduced glutathione with standard deviation per mg protein in *Unio* sp. bivalve samples collected from Sirpur Lake site A and site B were 5.86±0.09 and 6.76±0.11 in summer; 7.01±0.10 and 7.59±0.12 in

Fig. 46. Glutathione-S-transferase activity in *Unio* sp. bivalves collected from different sampling sites of Sirpur and Bilawali Tanks of Indore in different seasons.

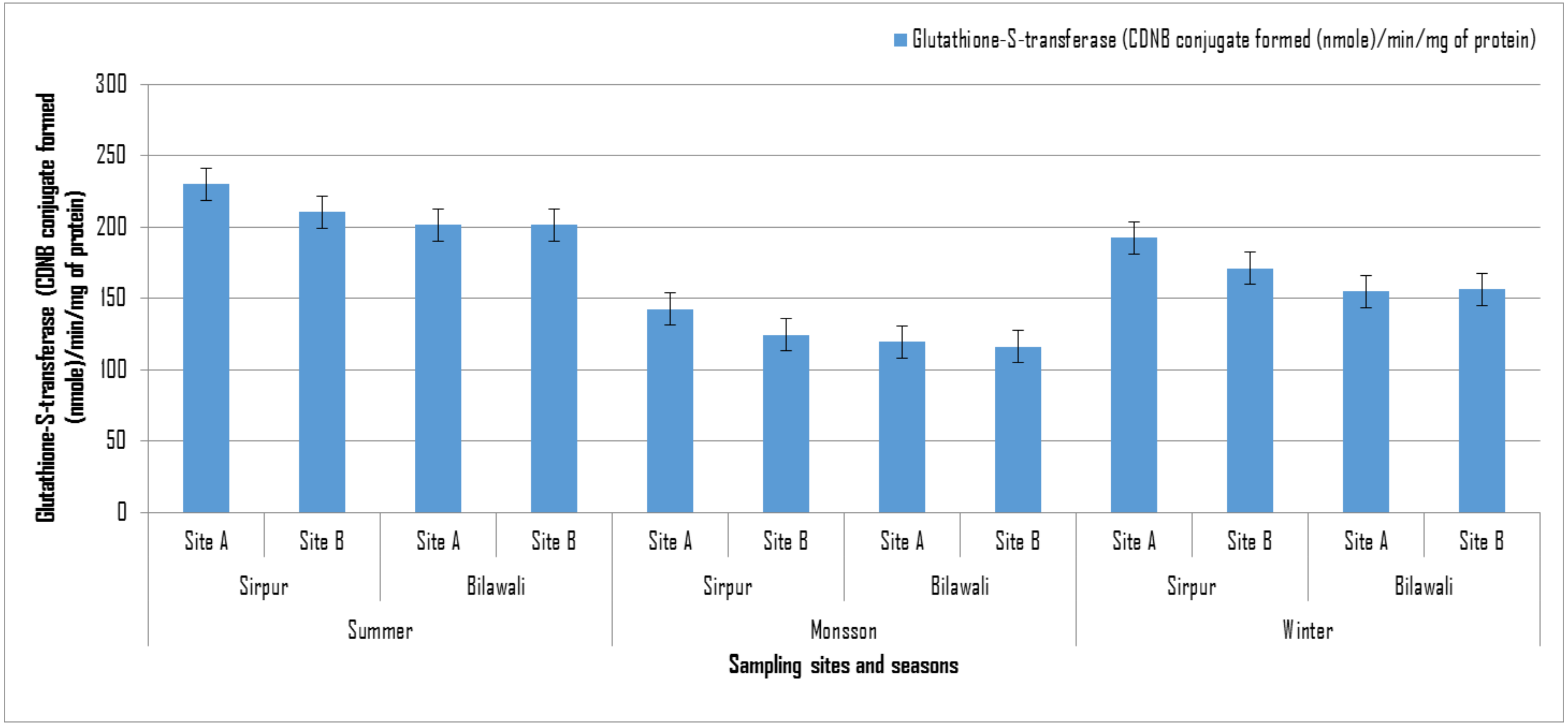
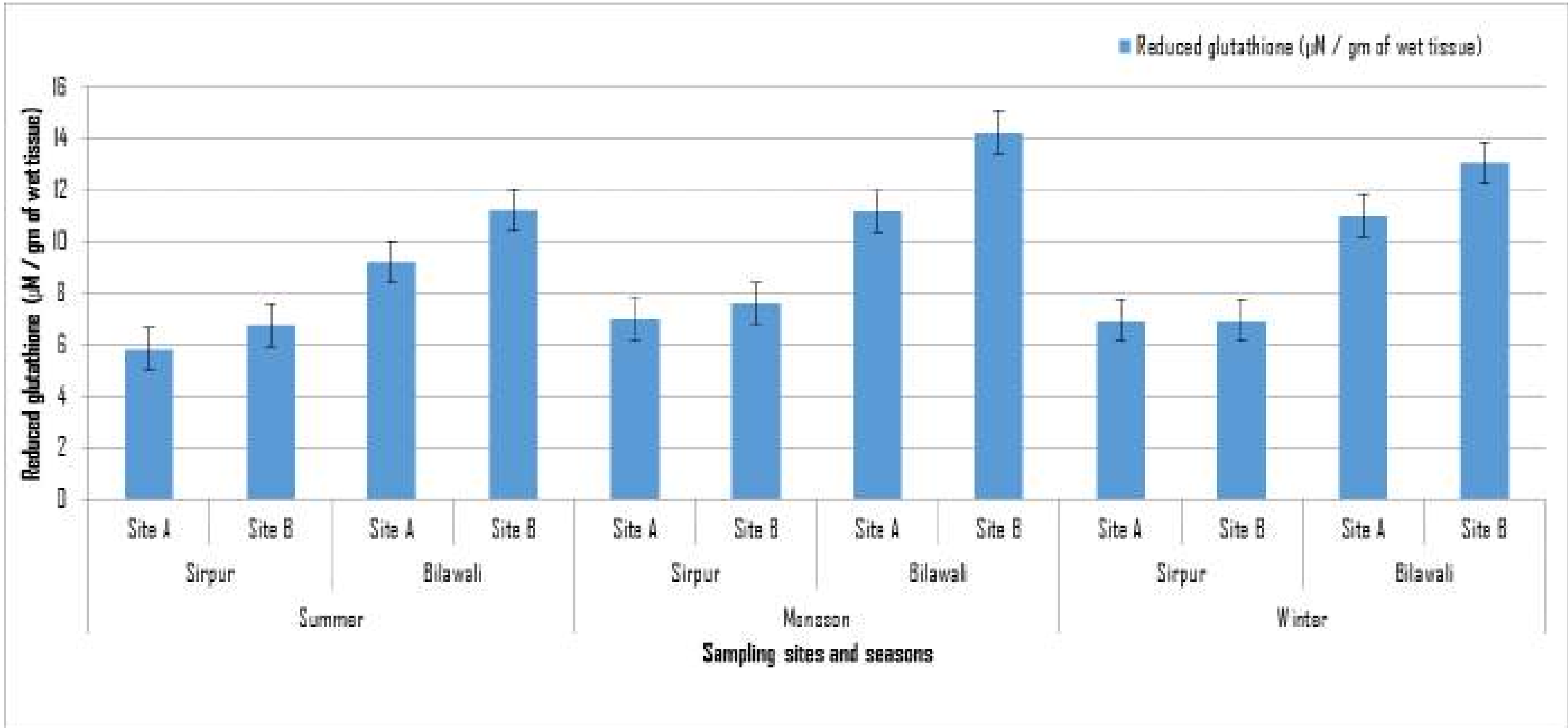


Fig. 47. Reduced glutathione activity in *Unio* sp. bivalves collected from different sampling sites of Sirpur and Bilawali Tanks of Indore in different seasons.



monsoon; 6.95±0.11 and 6.95±0.12 in winter seasons whereas, Bilawali Tank site A and site B were reported 9.23±0.09 and 11.23±0.08 in summer; 11.2±0.11and 14.26±0.12 in monsoon; 11.03±0.09 and 13.06±0.1 in winter seasons respectively.

3.4.3.4 Superoxide dismutase:

Superoxide dismutase with standard error per mg protein in collected *Unio* sp. bivalve samples of Sirpur and Bilawali Tank in summer, monsoon and winter seasons is shown in Table 9. Superoxide dismutase with standard deviation per mg protein in *Unio* sp. bivalve samples collected from Sirpur Lake site A and site B were 120.45±2.12 and 125.64±2.03 in summer; 160.58±1.56 and 180.46±1.14 in monsoon; 125.34±2.01 and 145.36±3.02 in winter seasons whereas, Bilawali Tank site A and site B were reported 130.45±1.98 and 140.25±1.48 in summer; 186.54±1.69 and 191.42±1.94 in monsoon; 141.29±2.38 and 151.24±2.16 in winter seasons respectively.

3.4.3.5 Catalase

Catalase with standard error per mg protein in collected *Unio* sp. bivalve samples of Sirpur and Bilawali Tank in summer, monsoon and winter seasons is shown in Table 9. Catalase with standard deviation per mg protein in *Unio* sp. bivalve samples collected from Sirpur Lake site A

Fig. 48. Superoxide activity in *Unio* sp. bivalves collected from different sampling sites of Sirpur and Bilawali Tanks of Indore in different seasons.

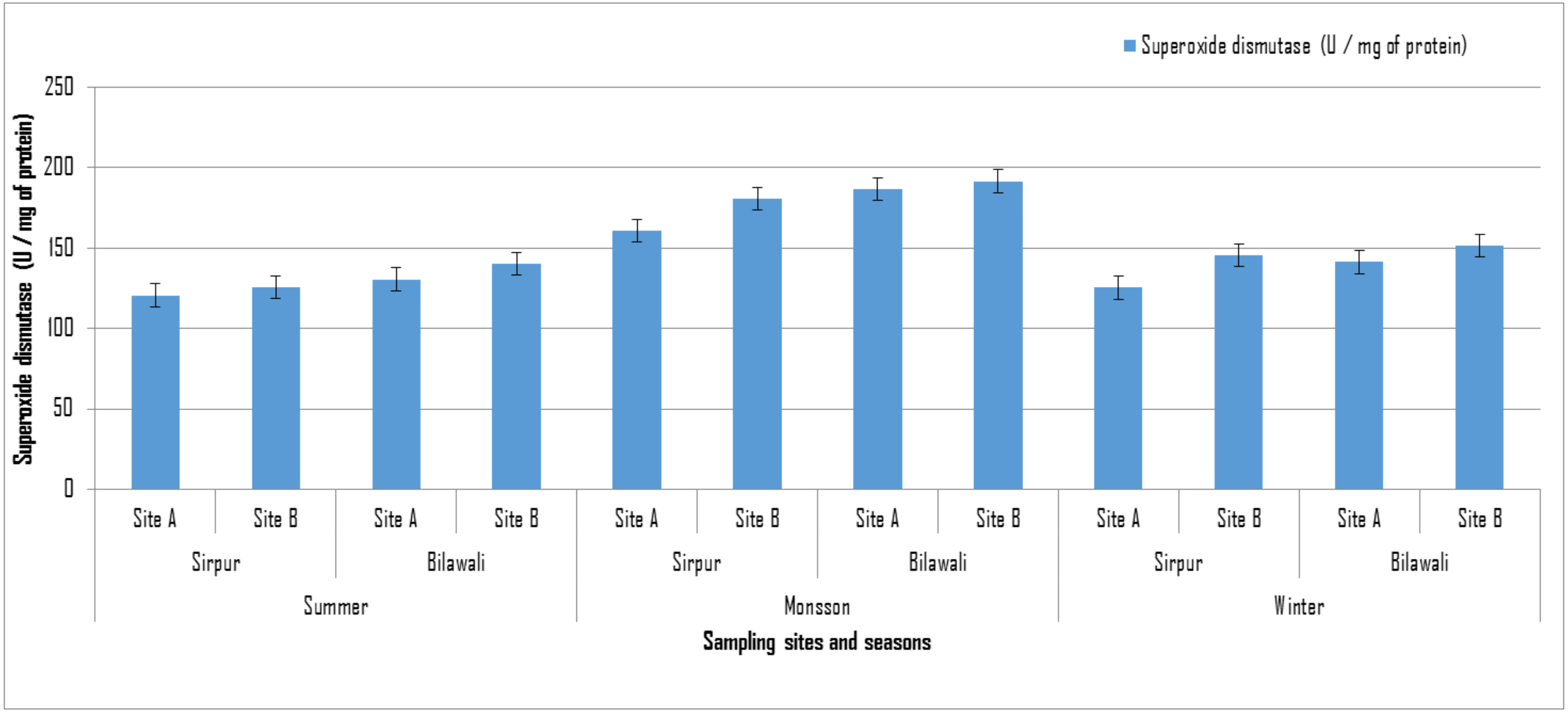
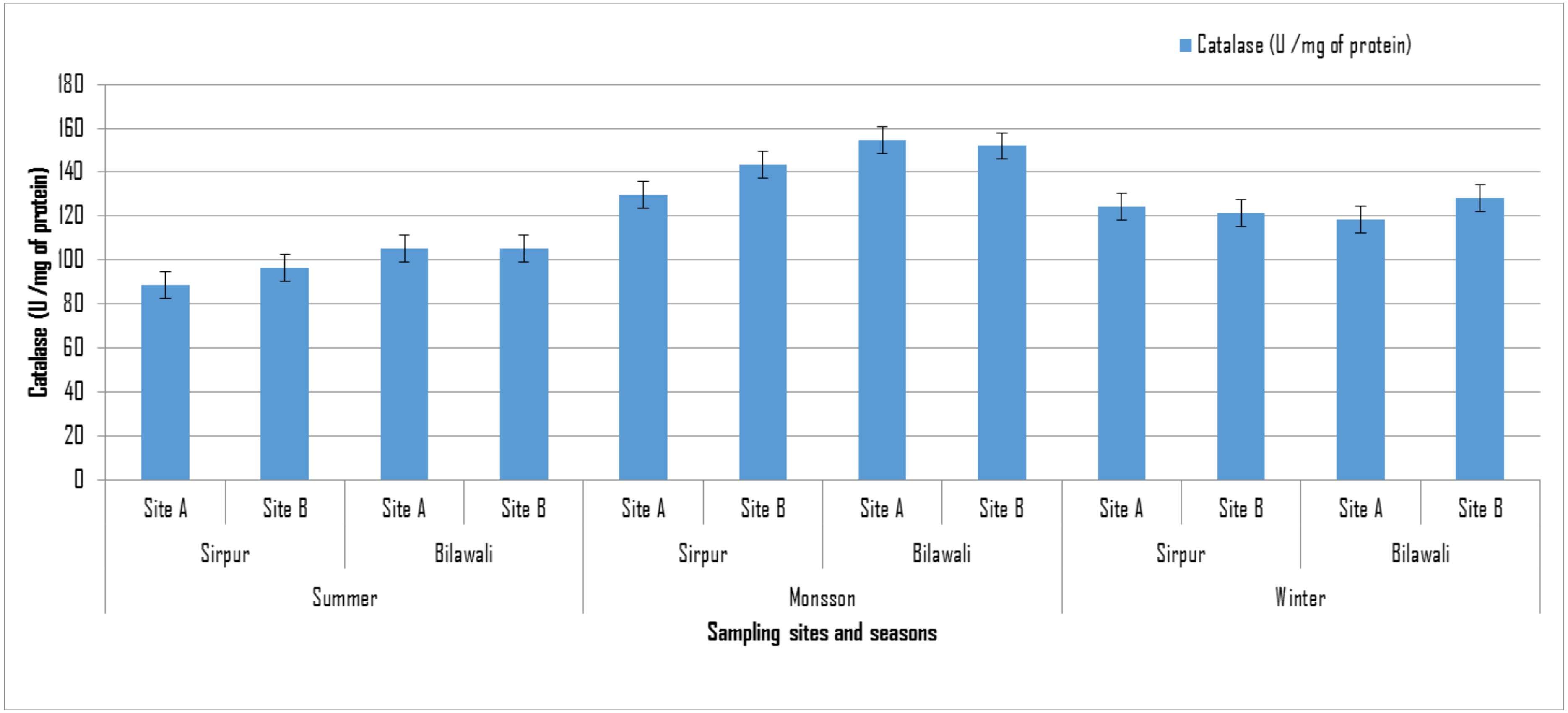


Fig. 49. Catalase activity in *Unio* sp. bivalves collected from different sampling sites of Sirpur and Bilawali Tanks of Indore in different seasons.

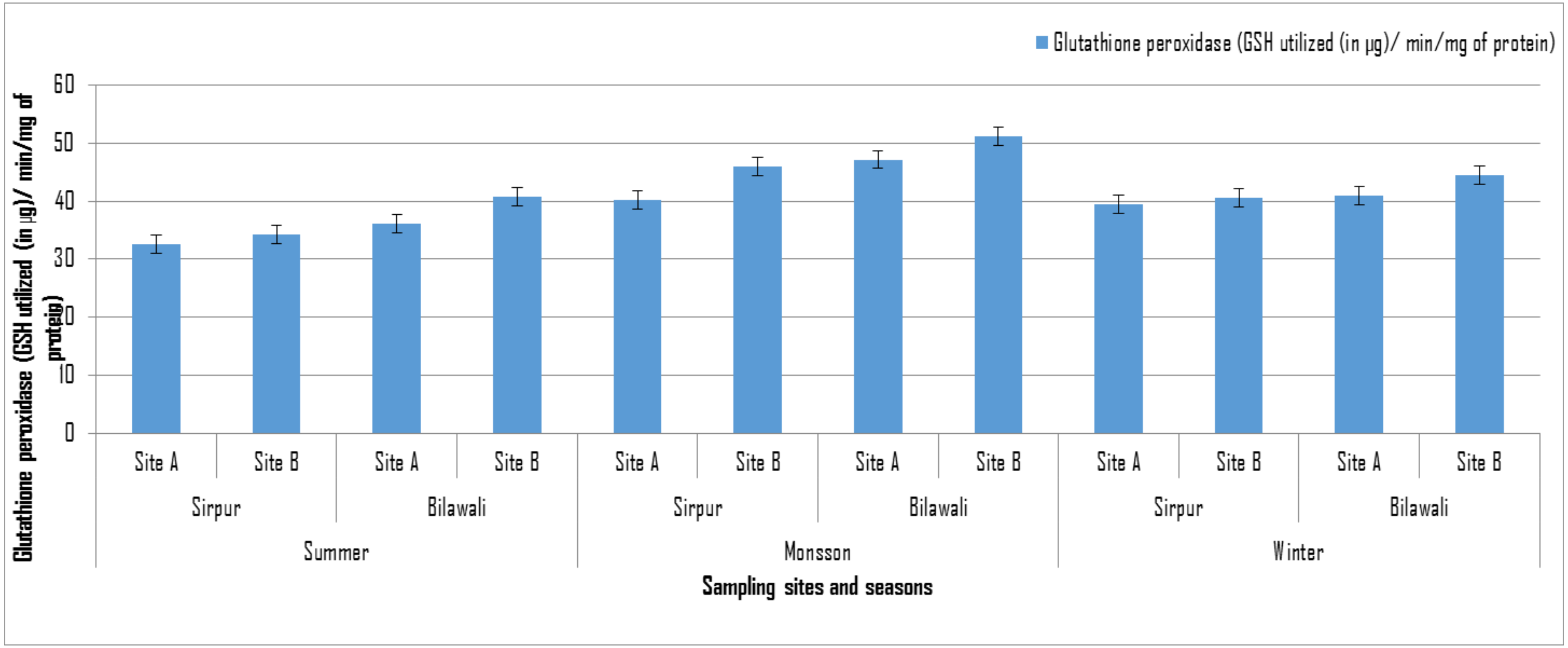


and site B were 88.64±1.01 and 96.58±1.12 in summer; 129.54±1.86 and 143.51±1.12 in monsoon 124.23±1.96 and 121.48±2.12 in winter seasons whereas, Bilawali Tank site A and site B were reported 105.28±1.86 and 105.36±1.23 in summer; 154.75±1.23 and 152.01±1.64 in monsoon; 118.46±2.10 and 128.34±1.13 in winter seasons respectively.

3.4.3.6 Glutathione peroxidase

Superoxide dismutase with standard error per mg protein in collected *Unio* sp. bivalve samples of Sirpur and Bilawali Tank in summer, monsoon and winter seasons is shown in Table 9. Superoxide dismutase with standard deviation per mg protein in *Unio* sp. bivalve samples collected from Sirpur Lake site A and site B were 32.56±0.16 and 34.27±0.11 in summer; 40.16±0.07 and 46.02±0.10 in monsoon; 39.56±0.12 and 40.58±0.13 in winter seasons whereas, Bilawali Tank site A and site B were reported 36.15±0.14 and 40.81±0.11 in summer; 47.2±0.13 and 51.2±0.11 in monsoon; 41.03±0.11 and 44.52±0.12 in winter seasons respectively.

Fig. 50. Glutathione peroxidase activity in *Unio* sp. bivalves collected from different sampling sites of Sirpur and Bilawali Tanks of Indore in different seasons.



3.5 Biochemical studies in bivalves:

Biochemical studies viz. Protein, Ascorbic acid ,RNA and DNA were assessed in *Lamellidens corrianus, Lamellidens marginali* and *Unio* sp bivalves samples collected from two different sites of Sirpur and Bilawali Tanks, Indore in summer, monsoon and winter seasons and presented in table 10-12.

3.5.1 *Lamellidens corrianus*

Table 10 presents the mean with standard deviation of collected observations of biochemical studies in *Lamellidens corrianus* samples.

Table 10. Seasonal variations of biochemical parameters in *Lamellidens corrianus* collected from Sirpur and Bilawali Tanks of Indore.

Body part	Summer				Monson				Winter			
	Sirpur		Bilawali		Sirpur		Bilawali		Sirpur		Bilawali	
	Site A	Site B	Site A	Site B	Site A	Site B	Site A	Site B	Site A	Site B	Site A	Site B
PROTEIN												
Gills	49.59±1.25	52.01±1.34	52.42±1.23	53.21±1.47	60.25±1.58	61.56±1.37	63.24±1.34	63.15±1.28	59.68±1.19	61.29±1.39	61.02±1.49	61.05±1.39
Digestive gland	47.56±1.12	51.2±1.42	50.27±1.42	51.12±1.49	61.10±1.29	61.03±1.29	62.13±1.31	64.29±1.22	57.62±1.18	59.68±1.38	59.61±1.38	60.31±1.39
Whole soft body	48.51±1.14	49.35±1.29	49.23±1.29	50.01±1.43	58.67±1.27	60.01±1.37	60.03±1.27	61.20±1.31	53.24±1.11	56.24±1.34	57.23±136	59.64±1.32
ASCORBIC ACID												
Gills	0.712±0.001	0.786±0.003	0.805±0.001	0.814±0.001	1.005±0.005	1.132±0.009	1.142±0.011	1.112±0.009	0.986±0.009	1.002±0.009	1.009±0.008	1.008±0.009
Digestive gland	0.856±0.002	0.886±0.003	0.926±0.002	0.953±0.002	1.103±0.005	1.296±0.010	1.298±0.012	1.236±0.010	1.023±0.009	1.124±0.010	1.143±0.010	1.112±0.008
Whole soft body	0.702±0.001	0.736±0.002	0.792±0.001	0.756±0.001	1.009±0.004	1.124±0.010	1.136±0.009	1.152±0.012	0.958±0.008	0.995±0.009	0.995±0.009	1.020±0.008
DNA												
Gills	1.12±0.23	1.18±0.22	1.23±0.21	1.29±0.21	1.46±0.23	1.36±0.21	1.76±0.22	1.86±0.23	1.62±0.12	1.54±0.21	1.56±0.22	1.59±0.23
Digestive gland	1.19±0.20	1.25±0.19	1.31±0.22	1.35±0.23	1.79±0.23	1.85±0.22	1.89±0.23	1.97±0.22	1.63±0.17	1.59±0.22	1.62±0.23	1.68±0.22
Whole soft body	1.23±0.21	1.32±0.14	1.41±0.23	1.42±0.21	1.94±0.25	1.99±0.23	1.95±0.23	1.96±0.21	1.79±0.21	1.86±0.23	1.95±0.22	1.94±0.23

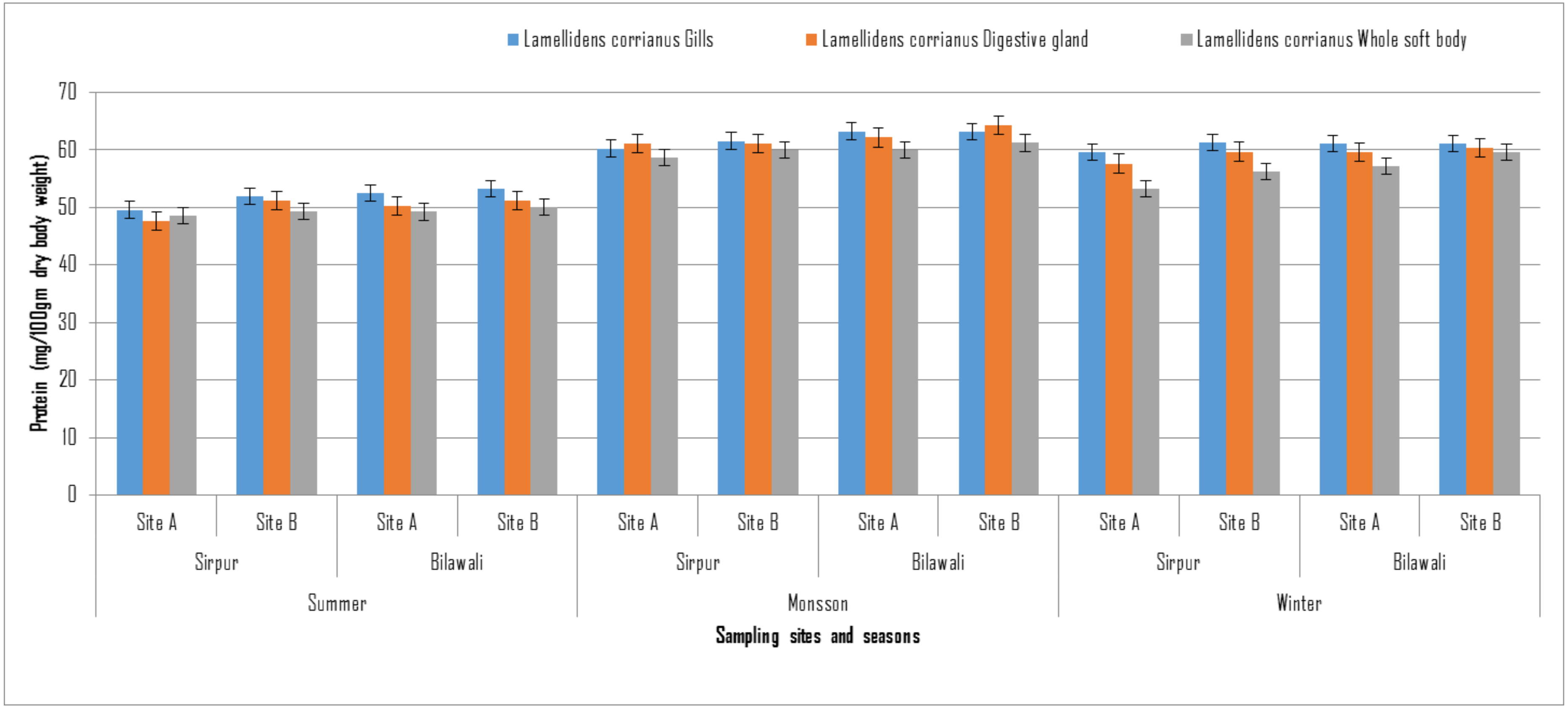
RNA												
	Summer				**Monsoon**				**Winter**			
	Sirpur		**Bilawali**		**Sirpur**		**Bilawali**		**Sirpur**		**Bilawali**	
Body part	**Site A**	**Site B**	**Site A**	**Site B**	**Site A**	**Site B**	**Site A**	**Site B**	**Site A**	**Site B**	**Site A**	**Site B**
Gills	5.01±0.34	5.98±0.41	6.07±0.33	6.12±0.30	7.01±0.29	8.36±0.33	8.49±0.41	8.50±0.39	7.50±0.38	8.96±0.42	9.08± 0.37	9.10±0.43
Digestive gland	6.39± 0.41	6.89± 0.33	7.02± 0.37	7.19±0.32	8.94±0.37	9.63±0.27	9.82±0.43	9.98±0.42	9.57±0.42	9.61±0.39	9.96± 0.39	9.69±0.39
Whole soft body	4.42± 0.29	4.93± 0.27	5.96± 0.36	6.02±0.44	6.18±0.28	6.89±0.27	8.34±0.41	8.36±0.39	6.62±0.34	7.38±0.43	8.91± 0.41	8.95±0.47

3.5.1.1 Protein

Protein with standard error per mg dry weight of gills in collected *Lamellidens corrianus* bivalve samples of Sirpur and Bilawali Tank in summer, monsoon and winter seasons is shown in Table 10. Protein with standard deviation per mg dry weight of gills in *Lamellidens corrianus* bivalve samples collected from Sirpur Lake site A and site B were 49.59±1.25 and 52.01±1.34 in summer; 60.25±1.58 and 61.56±1.37 in monsoon; 59.68±1.19 and 61.29±1.39 in winter seasons whereas, Bilawali Tank site A and site B were reported 52.42±1.23 and 53.21±1.47 in summer; 63.24±1.34 and 63.15±1.28 in monsoon; 61.02±1.49 and 61.05±1.39 in winter seasons respectively. Protein with standard deviation per mg dry weight of digestive gland in *Lamellidens*.

corrianus bivalve samples collected from Sirpur Lake site A and site B were 47.56±1.12 and 51.2±1.42 in summer; 61.10±1.29 and 61.03±1.29 in monsoon; 57.62±1.18 and 59.68±1.38 in winter seasons whereas, Bilawali Tank site A and site B were reported 50.27±1.42 and 51.12±1.49 in summer; 62.13±1.31 and 64.29±1.22 in monsoon; 59.61±1.38 and 60.31±1.39 in winter seasons respectively. Protein with standard deviation per mg dry weight of whole body soft tissue in *Lamellidens corrianus* bivalve samples collected from Sirpur Lake site A and site B were 48.51±1.14 and 49.35±1.29 in summer; 58.67±1.27 and 60.01±1.37 in monsoon; 53.24±1.11 and 56.24±1.34 in winter seasons whereas, Bilawali Tank site A and site B were reported 49.23±1.29 and 50.01±1.43 in summer; 60.03±1.27 and 61.20±1.31 in monsoon; 57.23±136 and 59.64±1.32 in winter seasons respectively.

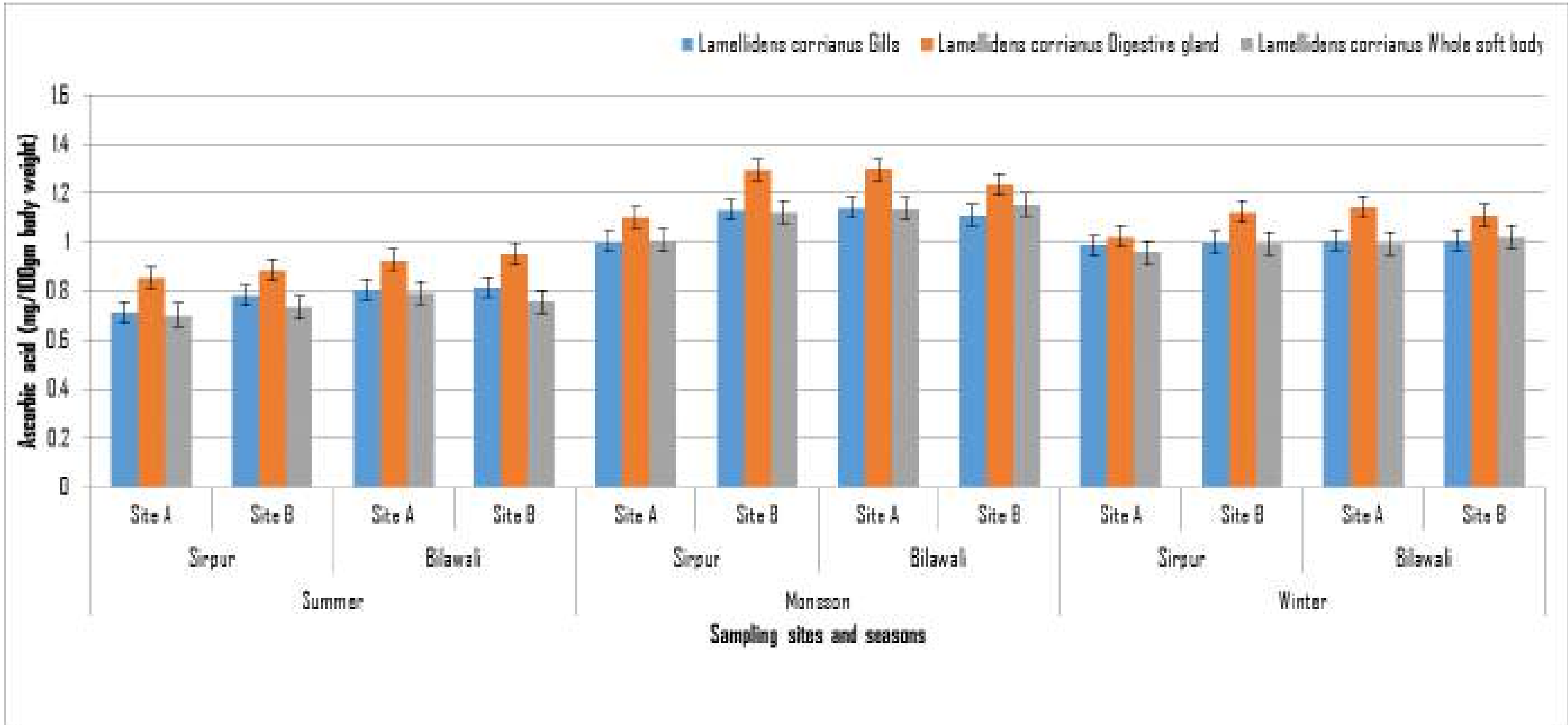
Fig. 51. Total protein in gills, digestive gland and whole soft body of *Lamellidens corrianus* bivalves collected from different sampling sites of Sirpur and Bilawali Tanks of Indore in different seasons.



3.5.1.2 Ascorbic acid

Ascorbic acid with standard error per mg dry weight of gills in collected *Lamellidens corrianus* bivalve samples of Sirpur and Bilawali Tank in summer, monsoon and winter seasons is shown in Table 9. Ascorbic acid with standard deviation per mg dry weight of gills in *Lamellidens corrianus* bivalve samples collected from Sirpur Lake site A and site B were 0.712±0.001 and 0.786±0.003 in summer; 1.005±0.005 and 1.132±0.009 in monsoon; 0.986±0.009 and 1.002±0.009 in winter seasons whereas, Bilawali Tank site A and site B were reported 0.805±0.001 and 0.814±0.001 in summer; 1.142±0.011 and 1.112±0.009 in monsoon; 1.009±0.008 and 1.008±0.009 in winter seasons respectively. Ascorbic acid with standard deviation per mg dry weight of digestive gland in *Lamellidens corrianus* bivalve samples collected from Sirpur Lake site A and site B were 0.856±0.002 and 0.886±0.003 in summer; 1.103±0.005 and 1.296±0.010 in monsoon; 1.023±0.009 and 1.124±0.010 in winter seasons whereas, Bilawali Tank site A and site B were reported 0.926±0.002 and 0.953±0.002 in summer; 1.298±0.012 and 1.236±0.010 in monsoon; 1.143±0.010 and 1.112±0.008 in winter seasons respectively. Ascorbic acid with standard deviation per mg dry weight of whole body soft tissue in *Lamellidens corrianus* bivalve samples collected from Sirpur Lake site A and site B were 0.702±0.001 and 0.736±0.002 in summer; 1.009±0.004 and 1.124±0.010 in monsoon; 0.958±0.008 and 0.995±0.009 in winter seasons whereas, Bilawali Tank site A and site B were reported 0.792±0.001 and 0.756±0.001 in summer; 1.136±0.009 and 1.152±0.012 in monsoon; 0.995±0.009 and 1.020±0.008 in winter seasons respectively.

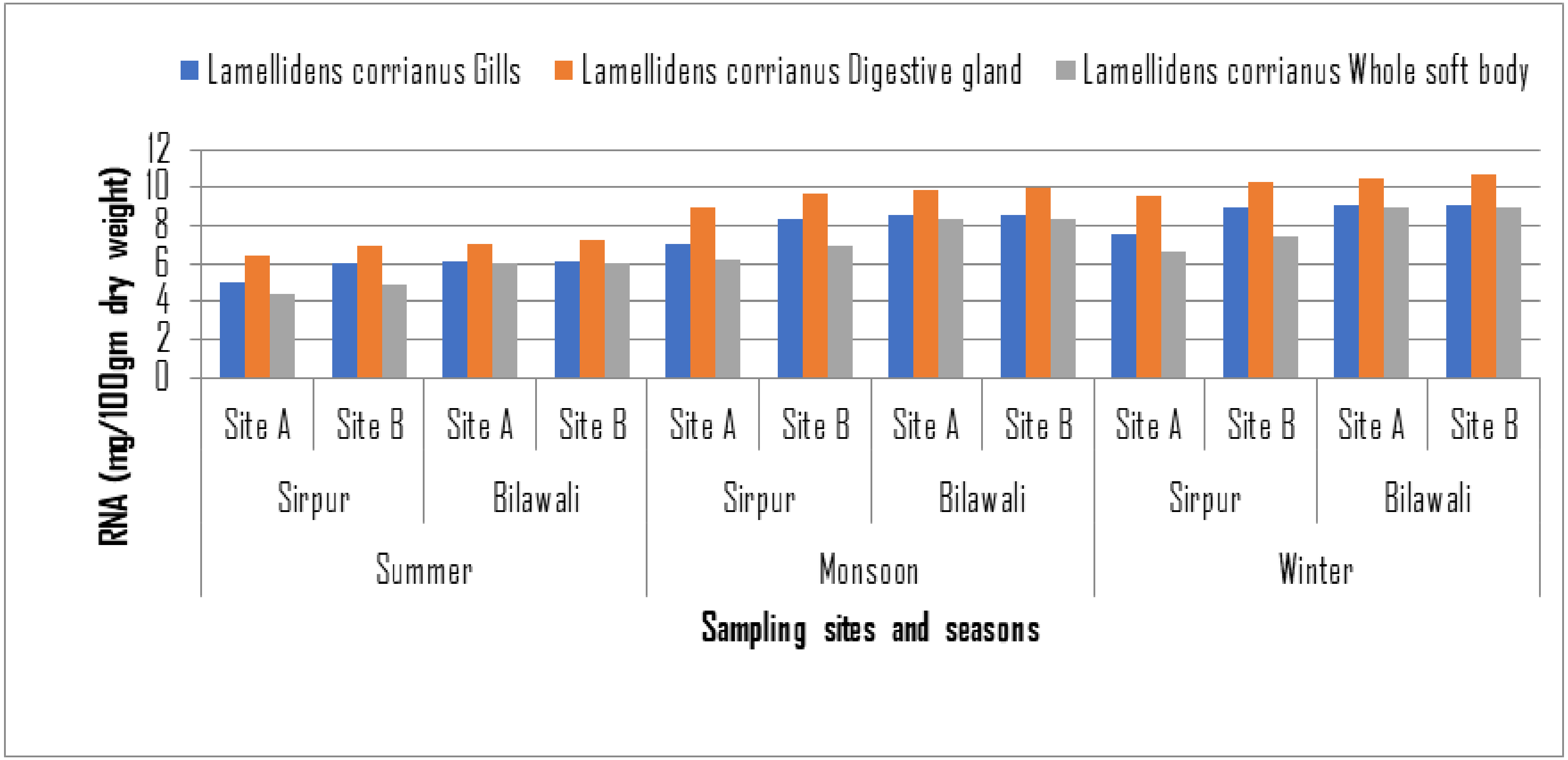
Fig. 52. Ascorbic acid in gills, digestive gland and whole soft body of *Lamellidens corrianus* bivalves collected from different sampling sites of Sirpur and Bilawali Tanks of Indore in different seasons.



3.5.1.3 RNA

RNA with standard error per mg dry weight of gills in collected *Lamellidens corrianus* bivalve samples of Sirpur and Bilawali Tank in summer, monsoon and winter seasons is shown in Table 9. RNA with standard deviation per mg dry weight of gills in *Lamellidens corrianus.* bivalve samples collected from Sirpur Lake site A and site B were 5.01± 0.34 and 5.98± 0.41 in summer; 7.01± 0.29 and 8.36± 0.33 in monsoon; 7.50± 0.38 and 8.96± 0.42 in winter seasons whereas, Bilawali Tank site A and site B were reported 6.07± 0.33 and 6.12± 0.30 in summer; 8.49± 0.41 and 8.50± 0.39 in monsoon; 9.08± 0.37 and 9.10± 0.43 in winter seasons respectively. RNA with standard deviation per mg dry weight of digestive gland in *Lamellidens corrianus* bivalve samples collected from Sirpur Lake site A and site B were 6.39± 0.41 and 6.89± 0.33 in summer; 8.94± 0.37 and 9.63± 0.27 in monsoon; 9.57± 0.42 and 9.61± 0.39 in winter seasons whereas, Bilawali Tank site A and site B were reported 7.02± 0.37 and 7.19± 0.32 in summer; 9.82± 0.43 and 9.98± 0.42 in monsoon; 9.96± 0.39 and 9.69± 0.39 in winter seasons respectively. RNA with standard deviation per mg dry weight of whole body soft tissue in *Lamellidens corrianus* bivalve samples collected from Sirpur Lake site A and site B were 4.42± 0.29 and 4.93± 0.27 in summer; 6.18± 0.28 and 6.89± 0.27 in monsoon; 6.62± 0.34 and 7.38± 0.43 in winter seasons whereas, Bilawali Tank site A and site B were reported 5.96± 0.36 and 6.02± 0.44 in summer; 8.34± 0.41 and 8.36± 0.39 in monsoon; 8.91± 0.41 and 8.95± 0.47 in winter seasons respectively

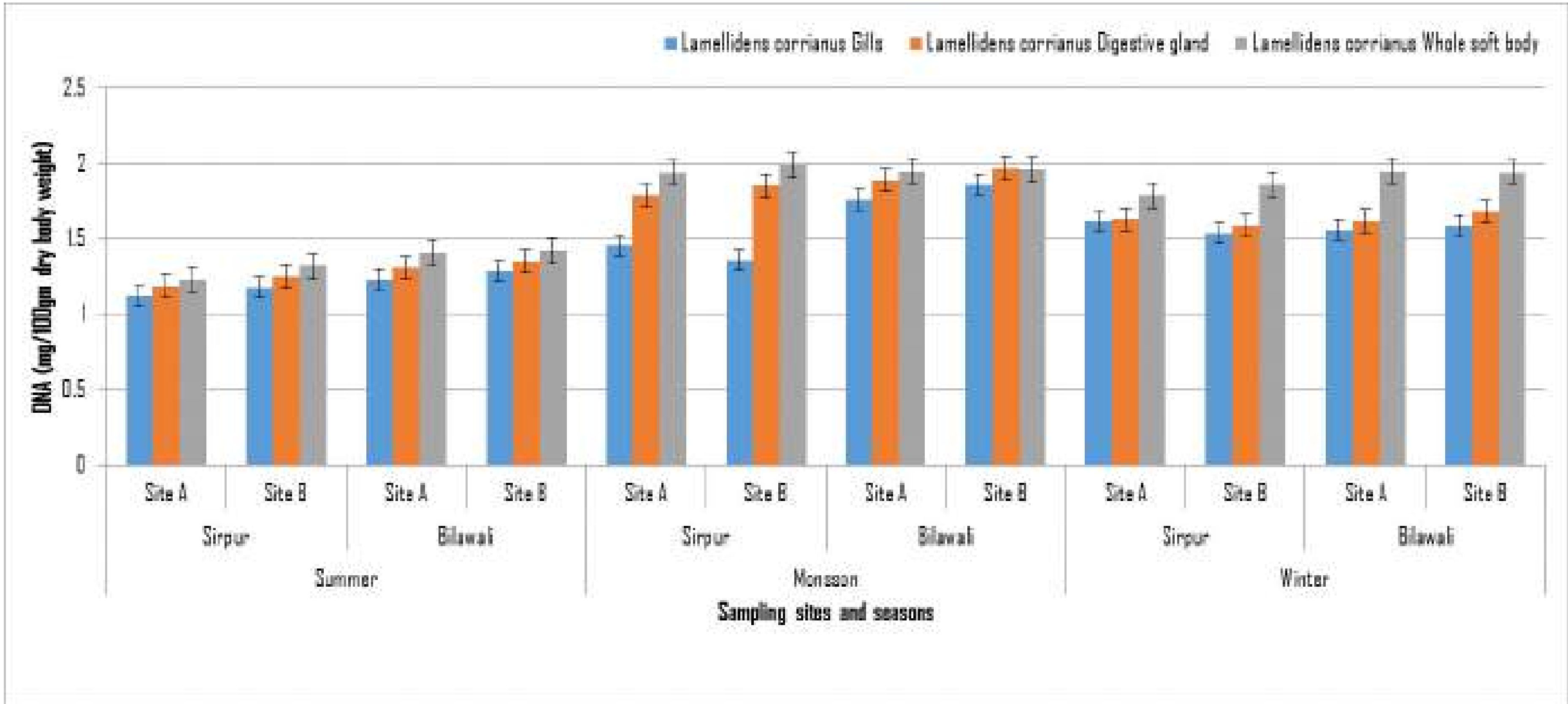
Fig. 53. RNA in gills, digestive gland and whole soft body of *Lamellidens corrianus* bivalves collected from different sampling sites of Sirpur and Bilawali Tanks of Indore in different seasons



3.5.1.4 DNA

DNA with standard error per mg dry weight of gills in collected *Lamellidens corrianus* bivalve samples of Sirpur and Bilawali Tank in summer, monsoon and winter seasons is shown in Table 9. DNA with standard deviation per mg dry weight of gills in *Lamellidens corrianus bivalve* samples collected from Sirpur Lake site A and site B were 1.12±0.23 and 1.18±0.22 in summer; 1.46±0.23 and 1.36±0.21 in monsoon; 1.62±0.12 and 1.54±0.21 in winter seasons whereas, Bilawali Tank site A and site B were reported 1.23±0.21 and 1.29±0.21 in summer; 1.76±0.22 and 1.86±0.23 in monsoon; 1.56±0.22 and 1.59±0.23 in winter seasons respectively. DNA with standard deviation per mg dry weight of digestive gland in *Lamellidens corrianus* bivalve samples collected from Sirpur Lake site A and site B were 1.19±0.20 and 1.25±0.19 in summer; 1.79±0.23 and 1.85±0.22 in monsoon; 1.63±0.17 and 1.59±0.22 in winter seasons whereas, Bilawali Tank site A and site B were reported 1.31±0.22 and 1.35±0.23 in summer; 1.89±0.23 and 1.97±0.22 in monsoon; 1.62±0.23 and 1.68±0.22 in winter seasons respectively. DNA with standard deviation per mg dry weight of whole body soft tissue in *Lamellidens corrianus* bivalve samples collected from Sirpur Lake site A and site B were 1.23±0.21 and 1.32±0.14 in summer; 1.94±0.25 and 1.99±0.23 in monsoon; 1.79±0.21 and 1.86±0.23 in winter seasons whereas, Bilawali Tank site A and site B were reported 1.41±0.23 and 1.42±0.21 in summer; 1.95±0.23 and 1.96±0.21 in monsoon; 1.95±0.22 and 1.94±0.23 in winter seasons respectively.

Fig. 54. DNA in gills, digestive gland and whole soft body of *Lamellidens corrianus* bivalves collected from different sampling sites of Sirpur and Bilawali Tanks of Indore in different seasons.



3.5.2 *Lamellidens marginali*

Table 11 presents the mean with standard deviation of collected observations of biochemical studies in *Lamellidens marginali* samples.

Table 11. Seasonal variations of biochemical parameters in *Lamellidens marginali* collected from Sirpur and Bilawali Tanks of Indore.

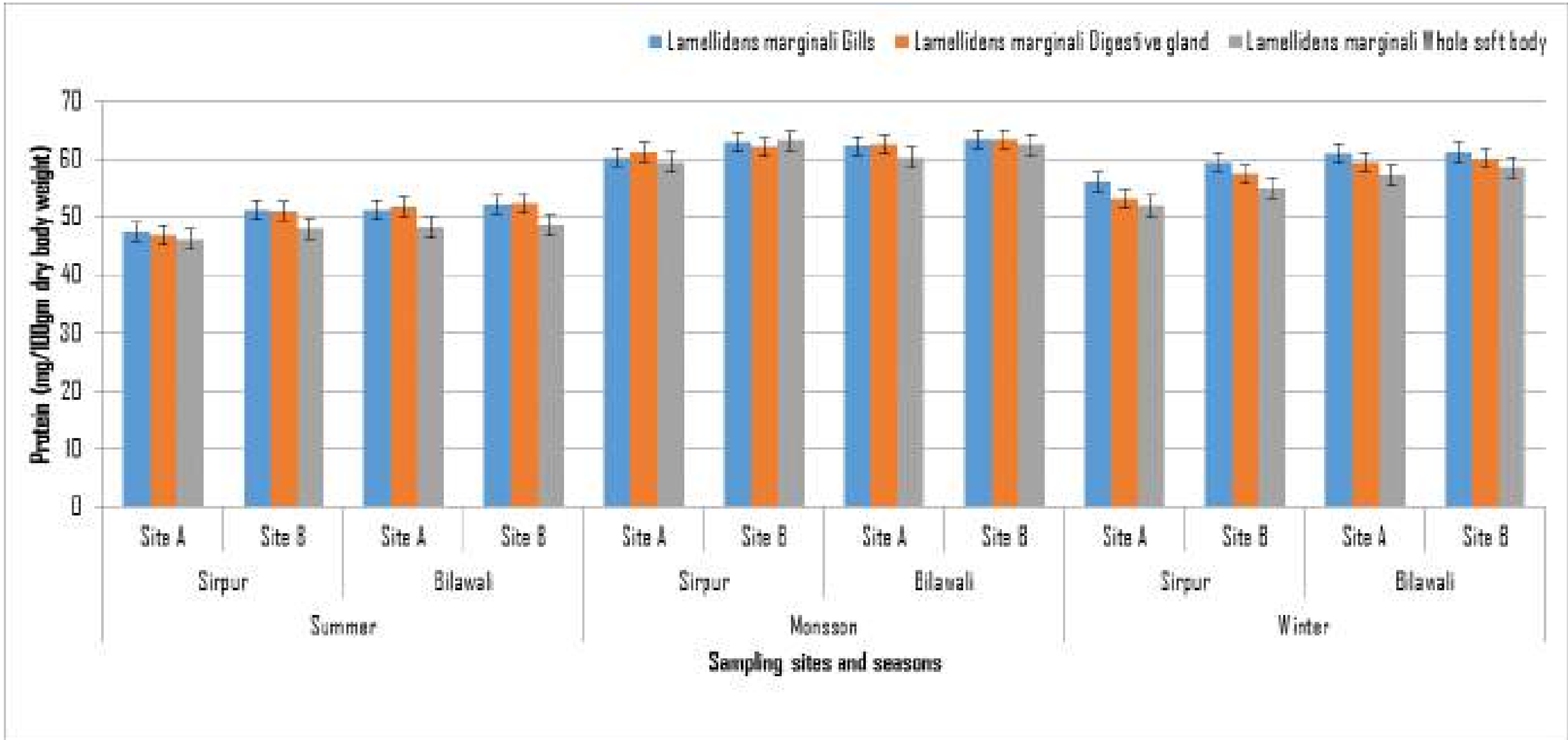
Body part	Summer				Monson				Winter			
	Sirpur		Bilawali		Sirpur		Bilawali		Sirpur		Bilawali	
	Site A	Site B	Site A	Site B	Site A	Site B	Site A	Site B	Site A	Site B	Site A	Site B
PROTEIN												
Gills	47.52±1.23	51.24±1.41	51.24±1.29	52.16±1.47	60.29±1.34	62.98±1.41	62.46±1.41	63.59±1.36	56.16±1.15	59.63±1.29	61.02±1.36	61.25±1.41
Digestive gland	46.89±1.15	51.11±1.39	51.82±1.30	52.36±1.49	61.24±1.45	62.14±1.28	62.53±1.39	63.49±1.32	53.24±1.14	57.42±1.27	59.64±1.41	60.12±1.36
Whole soft body	46.25±1.10	48.10±1.15	48.24±1.41	48.67±1.23	59.58±1.39	63.25±1.32	60.28±1.27	62.59±1.37	52.03±1.14	55.12±1.21	57.26±1.29	58.59±1.30
ASCORBIC ACID												
Gills	0.776±0.001	0.812±0.003	0.812±0.002	0.824±0.002	1.100±0.006	1.129±0.009	1.149±0.012	1.186±0.009	0.986±0.008	0.997±0.009	0.997±0.008	1.061±0.008
Digestive gland	0.882±0.002	0.945±0.004	0.964±0.003	0.942±0.003	1.235±0.006	1.325±0.011.	1.324±0.019	1.321±0.010	1.023±0.009	1.129±0.010	1.124±0.010	1.123±0.009
Whole soft body	0.721±0.001	0.795±0.002	0.752±0.001	0.806±0.002	1.112±0.004	1.142±0.009	1.145±0.012	1.154±0.010	0.984±0.008	0.998±0.009	1.023±0.010	1.006±0.010
DNA												
Gills	1.12±0.21	1.24±0.26	1.22±0.19	1.31±0.23	1.63±0.21	1.79±0.20	1.88±0.21	1.84±0.22	1.48±0.20	1.62±0.21	1.62±0.19	1.68±0.20
Digestive gland	1.22±0.28	1.25±0.23	1.31±0.18	1.33±0.19	1.68±0.21	1.91±0.19	1.76±0.19	1.96±0.21	1.39±0.23	1.54±0.21	1.64±0.19	1.71±0.1
Whole soft body	1.29±0.22	1.35±0.22	1.42±0.23	1.42±0.19	1.94±0.19	1.98±0.20	1.98±0.19	1.98±0.19	1.89±0.21	1.92±0.22	1.95±0.20	1.93±0.29

RNA												
	Summer				**Monsoon**				**Winter**			
	Sirpur		**Bilawali**		**Sirpur**		**Bilawali**		**Sirpur**		**Bilawali**	
Body part	**Site A**	**Site B**	**Site A**	**Site B**	**Site A**	**Site B**	**Site A**	**Site B**	**Site A**	**Site B**	**Site A**	**Site B**
Gills	5.07± 0.28	6.01±0.38	6.13± 0.34	6.24± 0.29	7.09± 0.37	8.40± 0.39	8.58± 0.44	8.66± 0.33	7.59± 0.33	9.00± 0.41	9.17± 0.42	9.28± 0.43
Digestive gland	6.22± 0.33	7.03±0.24	7.20± 0.41	7.31± 0.27	8.70± 0.39	9.49± 0.31	9.81± 0.45	9.59± 0.37	9.31± 0.37	9.43± 0.46	9.92± 0.62	9.63± 0.53
Whole soft body	4.59± 0.21	5.71±0.39	6.04± 0.38	6.22± 0.35	6.42± 0.27	7.98± 0.33	8.45± 0.39	8.63± 0.41	6.87± 0.38	8.55± 0.46	9.03± 0.39	9.25± 0.35

3.5.2.1 Protein

Protein with standard error per mg dry weight of gills in collected *Lamellidens marginali* bivalve samples of Sirpur and Bilawali Tank in summer, monsoon and winter seasons is shown in Table 11. Protein with standard deviation per mg dry weight of gills in *Lamellidens marginali* bivalve samples collected from Sirpur Lake site A and site B were 49.59±1.25 and 52.01±1.34 in summer; 60.25±1.58 and 61.56±1.37 in monsoon; 59.68±1.19 and 61.29±1.39 in winter seasons whereas, Bilawali Tank site A and site B were reported 52.42±1.23 and 53.21±1.47 in summer; 63.24±1.34 and 63.15±1.28 in monsoon; 61.02±1.49 and 61.05±1.39 in winter seasons respectively. Protein with standard deviation per mg dry weight of digestive gland in *Lamellidens marginali* bivalve samples collected from Sirpur Lake site A and site B were 47.56±1.12 and 51.2±1.42 in summer; 61.10±1.29 and 61.03±1.29 in monsoon; 57.62±1.18 and 59.68±1.38 in winter seasons whereas, Bilawali Tank site A and site B were reported 50.27±1.42 and 51.12±1.49 in summer; 62.13±1.31 and 64.29±1.22 in monsoon; 59.61±1.38 and 60.31±1.39 in winter seasons respectively. Protein with standard deviation per mg dry weight of whole body soft tissue in *Lamellidens marginali* bivalve samples collected from Sirpur Lake site A and site B were 48.51±1.14 and 49.35±1.29 in summer; 58.67±1.27 and 60.01±1.37 in monsoon; 53.24±1.11 and 56.24±1.34 in winter seasons whereas, Bilawali Tank site A and site B were reported 49.23±1.29 and 50.01±1.43 in summer; 60.03±1.27 and 61.20±1.31 in monsoon; 57.23±136 and 59.64±1.32 in winter seasons respectively.

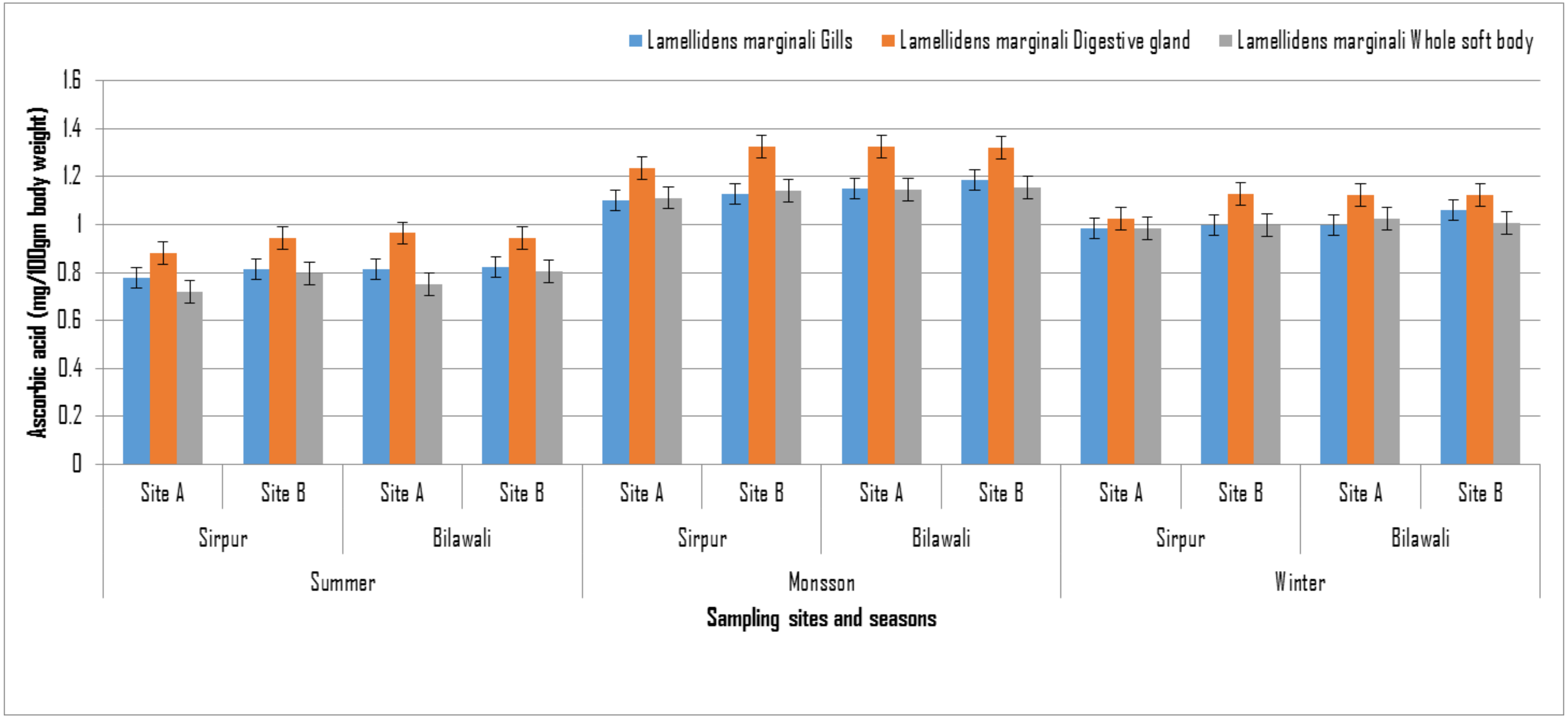
Fig. 55. Total protein in gills, digestive gland and whole soft body of *Lamellidens marginali* bivalves collected from different sampling sites of Sirpur and Bilawali Tanks of Indore in different seasons.



3.5.1.2 Ascorbic acid

Ascorbic acid with standard error per mg dry weight of gills in collected *Lamellidens marginali* bivalve samples of Sirpur and Bilawali Tank in summer, monsoon and winter seasons is shown in Table 11. Ascorbic acid with standard deviation per mg dry weight of gills in *Lamellidens marginali* bivalve samples collected from Sirpur Lake site A and site B were 0.712±0.001 and 0.786±0.003 in summer; 1.005±0.005 and 1.132±0.009 in monsoon; 0.986±0.009 and 1.002±0.009 in winter seasons whereas, Bilawali Tank site A and site B were reported 0.805±0.001 and 0.814±0.001 in summer; 1.142±0.011 and 1.112±0.009 in monsoon; 1.009±0.008 and 1.008±0.009 in winter seasons respectively. Ascorbic acid with standard deviation per mg dry weight of digestive gland in *Lamellidens marginali* bivalve samples collected from Sirpur Lake site A and site B were 0.856±0.002 and 0.886±0.003 in summer; 1.103±0.005 and 1.296±0.010 in monsoon; 1.023±0.009 and 1.124±0.010 in winter seasons whereas, Bilawali Tank site A and site B

Fig. 56. Ascorbic acid in gills, digestive gland and whole soft body of *Lamellidens marginali* bivalves collected from different sampling sites of Sirpur and Bilawali Tanks of Indore in different seasons.



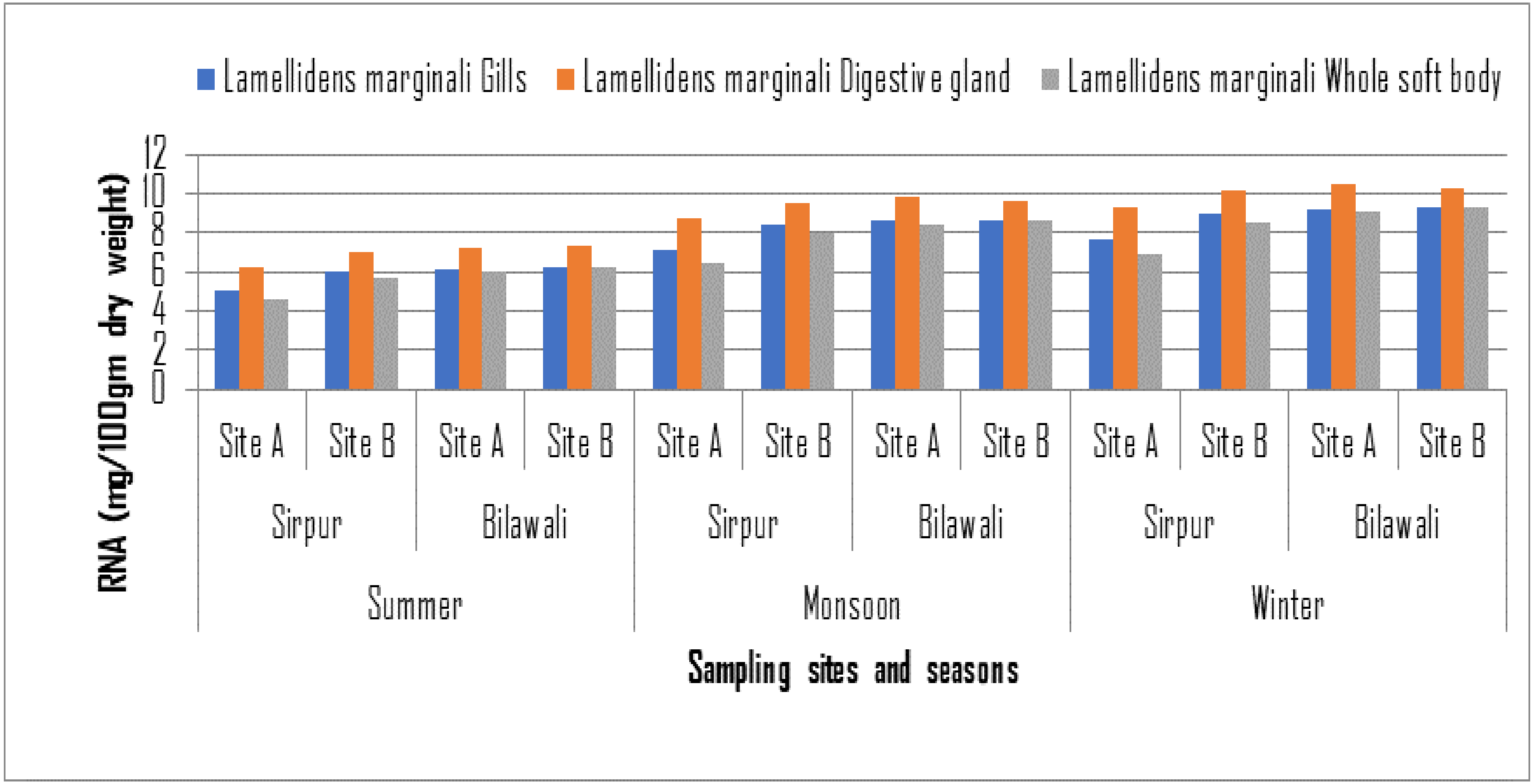
were reported 0.926±0.002 and 0.953±0.002 in summer; 1.298±0.012 and 1.236±0.010 in monsoon; 1.143±0.010 and 1.112±0.008 in winter seasons respectively. Ascorbic acid with standard deviation per mg dry weight of whole body soft tissue in *Lamellidens marginali* bivalve samples collected from Sirpur Lake site A and site B were 0.702±0.001 and 0.736±0.002 in summer; 1.009±0.004 and 1.124±0.010 in monsoon; 0.958±0.008 and 0.995±0.009 in winter seasons whereas, Bilawali Tank site A and site B were reported 0.792±0.001 and 0.756±0.001 in summer; 1.136±0.009 and 1.152±0.012 in monsoon; 0.995±0.009 and 1.020±0.008 in winter seasons respectively.

3.5.1.3 RNA

RNA with standard error per mg dry weight of gills in collected *Lamellidens marginali* bivalve samples of Sirpur and Bilawali Tank in summer, monsoon and winter seasons is shown in Table 11. RNA with standard deviation per mg dry weight of gills in *Lamellidens marginali.* bivalve samples collected from Sirpur Lake site A and site B were 5.07± 0.28 and 6.01± 0.38 in summer; 7.09± 0.37 and 8.40± 0.39 in monsoon; 7.59± 0.33 and 9.00± 0.41 in winter seasons whereas, Bilawali Tank site A and site B were reported 6.13± 0.34 and 6.12± 0.30 in summer; 8.58± 0.44 and 8.66± 0.33 in monsoon; 9.17± 0.42 and 9.28± 0.43 in winter seasons respectively. RNA with standard deviation per mg dry weight of digestive gland in *Lamellidens marginali* bivalve samples collected from

Sirpur Lake site A and site B were 6.22± 0.33 and 7.03± 0.24 in summer; 8.70± 0.39 and 9.49± 0.31 in monsoon; 9.31± 0.37 and 9.43± 0.46 in winter seasons whereas, Bilawali Tank site A and site B were reported 7.20± 0.41 and 7.31± 0.27 in summer; 9.81± 0.45 and 9.59± 0.37 in monsoon; 9.92± 0.62 and 9.63± 0.53 in winter seasons respectively. RNA with standard deviation per mg dry weight of whole body soft tissue in *Lamellidens marginali* bivalve samples collected from Sirpur Lake site A and site B were 4.59± 0.21 and 5.71± 0.39 in summer; 6.42± 0.27 and 7.98± 0.33 in monsoon; 6.87± 0.38and 8.55± 0.46 in winter seasons whereas, Bilawali Tank site A and site B were reported 6.04± 0.38 and 6.22± 0.35 in summer; 8.45± 0.39 and 8.63± 0.41 in monsoon; 9.03± 0.39 and 9.25± 0.35 in winter seasons respectively

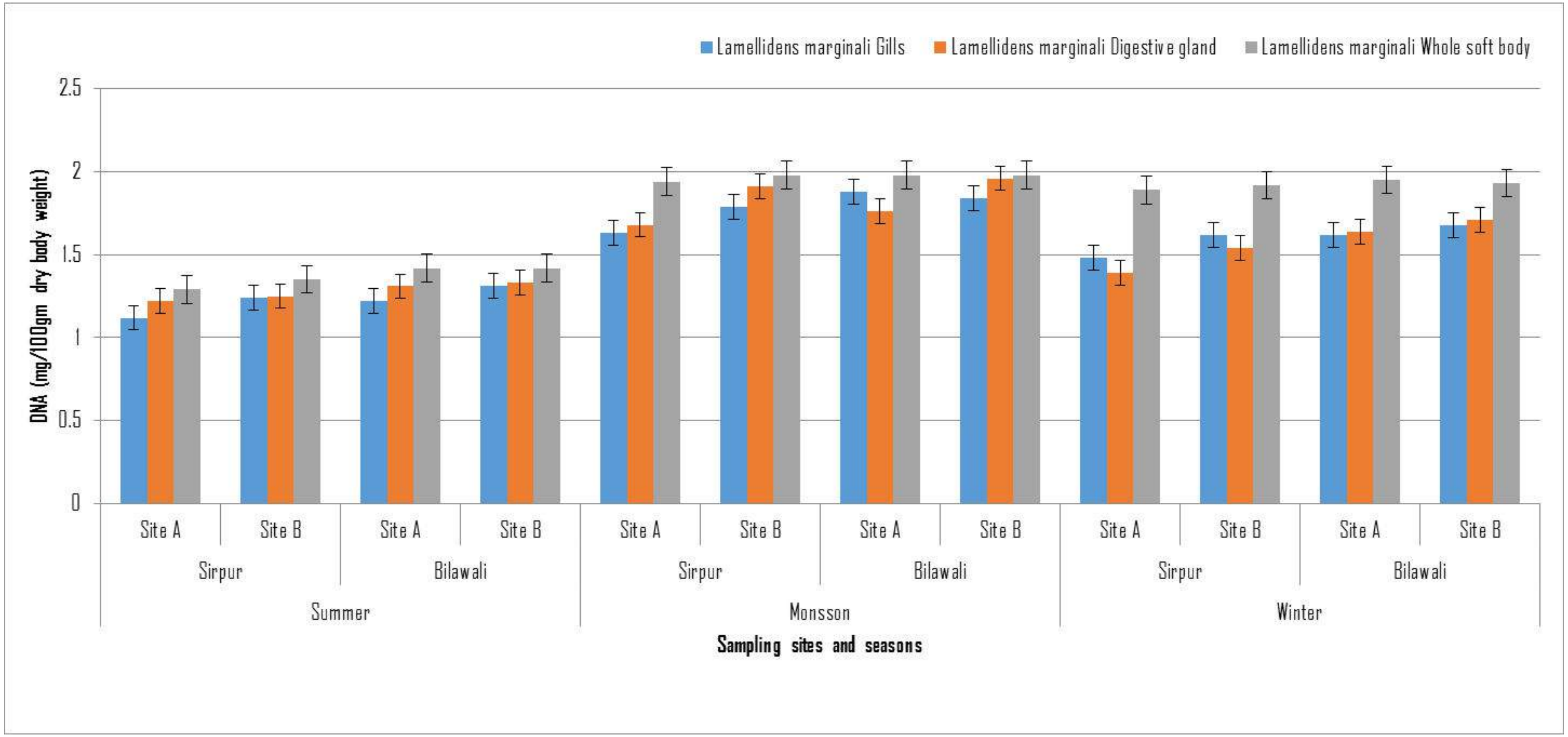
Fig. 57. RNA in gills, digestive gland and whole soft body of *Lamellidens marginali* bivalves collected from different sampling sites of Sirpur and Bilawali Tanks of Indore in different seasons



3.5.1.4 DNA

DNA with standard error per mg dry weight of gills in collected *Lamellidens marginali* bivalve samples of Sirpur and Bilawali Tank in summer, monsoon and winter seasons is shown in Table 11. DNA with standard deviation per mg dry weight of gills in *Lamellidens marginali* bivalve samples collected from Sirpur Lake site A and site B were 1.12±0.23 and 1.18±0.22 in summer; 1.46±0.23 and 1.36±0.21 in monsoon; 1.62±0.12 and 1.54±0.21 in winter seasons whereas, Bilawali Tank site A and site B were reported 1.23±0.21 and 1.29±0.21 in summer; 1.76±0.22 and 1.86±0.23 in monsoon; 1.56±0.22 and 1.59±0.23 in winter seasons respectively. DNA with standard deviation per mg dry weight of digestive gland in *Lamellidens marginali* bivalve

Fig. 58. DNA in gills, digestive gland and whole soft body of *Lamellidens marginali* bivalves collected from different sampling sites of Sirpur and Bilawali Tanks of Indore in different seasons.



samples collected from Sirpur Lake site A and site B were 1.19±0.20 and 1.25±0.19 in summer; 1.79±0.23 and 1.85±0.22 in monsoon; 1.63±0.17 and 1.59±0.22 in winter seasons whereas, Bilawali Tank site A and site B were reported 1.31±0.22 and 1.35±0.23 in summer; 1.89±0.23 and 1.97±0.22 in monsoon; 1.62±0.23 and 1.68±0.22 in winter seasons respectively. DNA with standard deviation per mg dry weight of whole body soft tissue in *Lamellidens marginali* bivalve samples collected from Sirpur Lake site A and site B were 1.23±0.21 and 1.32±0.14 in summer; 1.94±0.25 and 1.99±0.23 in monsoon; 1.79±0.21 and 1.86±0.23 in winter seasons whereas, Bilawali Tank site A and site B were reported 1.41±0.23 and 1.42±0.21 in summer; 1.95±0.23 and 1.96±0.21 in monsoon; 1.95±0.22 and 1.94±0.23 in winter seasons respectively.

3.5.3 *Unio* sp.

Table 12 presents the mean with standard deviation of collected observations of biochemical studies in *Unio* sp. samples.

Table 12. Seasonal variations of biochemical parameters in *Unio* sp. collected from Sirpur and Bilawali Tanks of Indore.

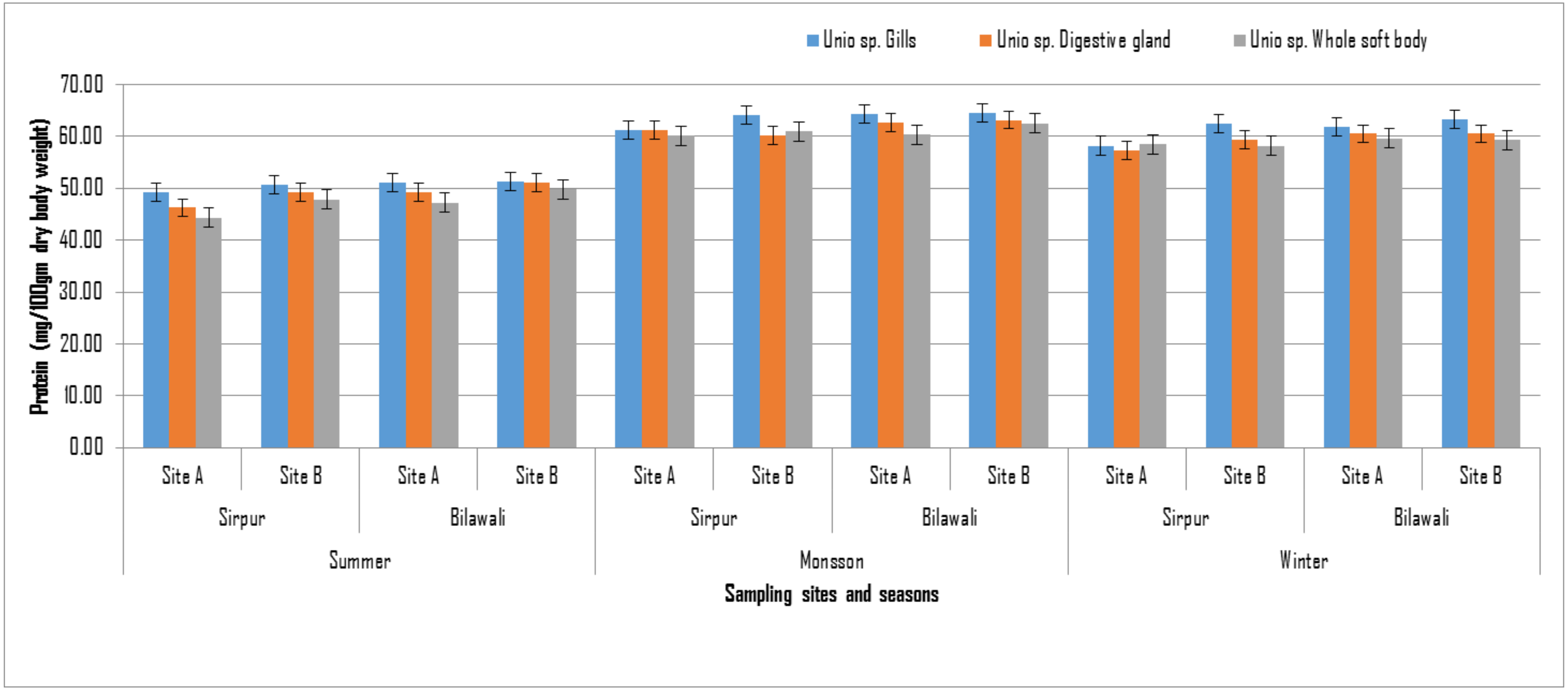
Body part	Summer				Monson				Winter			
	Sirpur		Bilawali		Sirpur		Bilawali		Sirpur		Bilawali	
	Site A	Site B	Site A	Site B	Site A	Site B	Site A	Site B	Site A	Site B	Site A	Site B
PROTEIN												
Gills	49.21±1.12	50.67±1.62	51.02±1.42	51.28±1.21	61.24±1.60	64.16±1.29	64.23±1.19	64.57±1.29	58.21±1.12	62.38±1.39	61.85±1.29	63.24±1.27
Digestive gland	46.24±1.28	49.16±1.52	49.26±1.39	51.09±1.12	61.25±1.18	60.25±1.32	62.58±1.32	63.16±1.32	57.31±1.14	59.42±1.37	60.49±1.21	60.49±1.24
Whole soft body	44.32±1.13	47.86±1.28	47.21±1.38	49.75±1.13	60.16±1.38	60.95±1.27	60.28±1.19	62.54±1.33	58.42±1.15	58.12±1.34	59.64±1.23	59.25±1.11
ASCORBIC ACID												
Gills	0.774±0.001	0.812±0.003	0.812±0.002	0.818±0.001	1.110±0.006	1.119±0.009	1.136±0.012	1.169±0.011	0.916±0.008	1.012±0.010	1.041±0.010	1.004±0.008
Digestive gland	0.920±0.003	0.912±0.003	0.965±0.002	0.924±0.002	1.254±0.006	1.321±0.011	1.321±0.014	1.325±0.015	0.998±0.007	1.109±0.010	1.116±0.009	1.128±0.008
Whole soft body	0.719±0.002	0.795±0.002	0.789±0.002	0.795±0.002	1.110±0.006	1.294±0.011	1.142±0.015	1.176±0.015	0.937±0.006	0.994±0.009	0.994±0.008	1.012±0.006
DNA												
Gills	1.16±0.19	1.17±0.19	1.21±0.21	1.26±0.16	1.43±0.19	1.65±0.20	1.82±0.22	1.78±0.17	1.42±0.21	1.46±0.19	1.65±0.21	1.69±0.24
Digestive gland	1.14±0.23	1.20±0.17	1.24±0.22	1.29±0.21	1.62±0.28	1.79±0.21	1.86±0.21	1.95±0.19	1.41±0.18	1.47±0.24	1.58±0.21	1.68±0.24
Whole soft body	1.25±0.21	1.31±0.22	1.39±0.23	1.39±0.17	1.98±0.28	1.99±0.20	1.84±0.21	1.97±0.19	1.95±0.19	1.95±0.25	1.89±0.20	1.93±0.29

RNA												
	Summer				**Monsoon**				**Winter**			
	Sirpur		**Bilawali**		**Sirpur**		**Bilawali**		**Sirpur**		**Bilawali**	
Body part	**Site A**	**Site B**	**Site A**	**Site B**	**Site A**	**Site B**	**Site A**	**Site B**	**Site A**	**Site B**	**Site A**	**Site B**
Gills	5.02± 0.33	5.43±0.34	5.61± 0.34	5.98± 0.36	7.02± 0.39	7.59± 0.37	7.85± 0.39	8.30± 0.41	7.51± 0.34	8.13± 0.43	8.39± 0.40	8.89± 0.39
Digestive gland	6.13± 0.39	6.03±0.29	6.14± 0.29	6.25± 0.34	8.58± 0.41	8.42± 0.34	8.59± 0.41	8.68± 0.43	9.18± 0.39	9.03± 0.39	9.18± 0.39	9.30± 0.37
Whole soft body	4.87± 0.27	5.37±0.34	5.47± 0.24	5.84± 0.32	6.81± 0.39	7.50± 0.32	7.65± 0.39	8.11± 0.37	7.29± 0.46	8.04± 0.38	8.18± 0.46	8.69± 0.39

3.5.3.1 Protein

Protein with standard error per mg dry weight of gills in collected *Unio* sp. bivalve samples of Sirpur and Bilawali Tank in summer, monsoon and winter seasons is shown in Table 12. Protein with standard deviation per mg dry weight of gills in *Unio* sp. bivalve samples collected from Sirpur Lake site A and site B were 49.21±1.12 and 50.67±1.62 in summer; 61.24±1.60 and 64.16±1.29 in monsoon; 58.21±1.12 and 62.38±1.39 in winter seasons whereas, Bilawali Tank site A and site B were reported 51.02±1.42 and 51.28±1.21 in summer; 64.23±1.19 and 64.57±1.29 in monsoon; 61.85±1.29 and 63.24±1.27 in winter seasons respectively. Protein with standard deviation per mg dry weight of digestive gland in *Unio* sp. bivalve samples collected from Sirpur Lake site A and site B were 46.24±1.28 and 49.16±1.52 in summer; 61.25±1.18 and 60.25±1.32 in monsoon; 57.31±1.14 and 59.42±1.37 in winter seasons whereas, Bilawali Tank site A and site B were reported 49.26±1.39 and 51.09±1.12 in summer; 62.58±1.32 and 63.16±1.32 in monsoon; 60.49±1.21 and 60.49±1.24 in winter seasons respectively. Protein with standard deviation per mg dry weight of whole body soft tissue in *Unio* sp. bivalve samples collected from Sirpur Lake site A and site B were 44.32±1.13 and 47.86±1.28 in summer; 60.16±1.38 and 60.95±1.27 in monsoon; 58.42±1.15 and 58.12±1.34 in winter seasons whereas, Bilawali Tank site A and site B were reported 47.21±1.38 and 49.75±1.13 in summer; 60.28±1.19 and 62.54±1.33 in monsoon; 59.64±1.23 and 59.25±1.11 in winter seasons respectively.

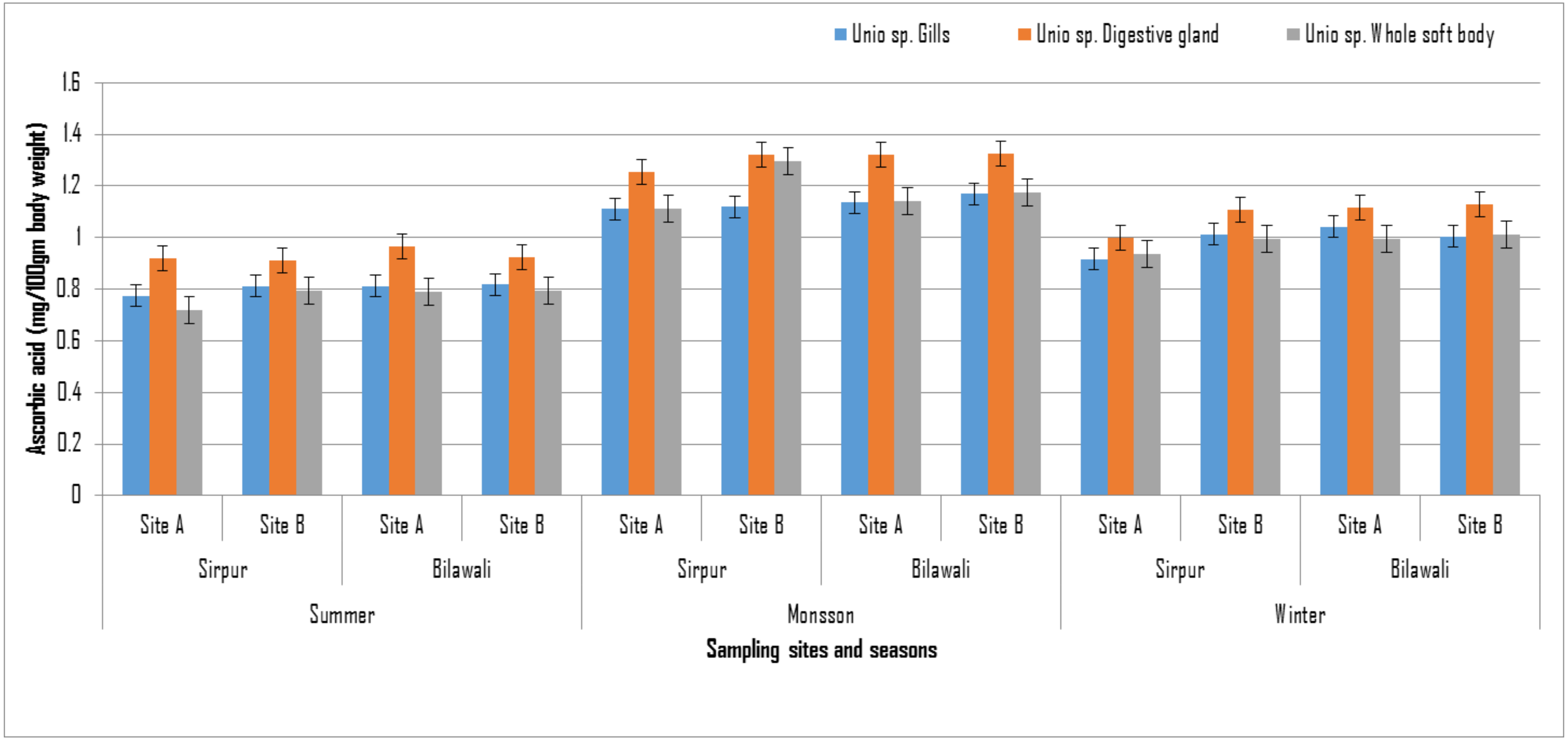
Fig. 59. Total protein in gills, digestive gland and whole soft body of *Unio* sp. bivalves collected from different sampling sites of Sirpur and Bilawali Tanks of Indore in different seasons.



3.5.1.2 Ascorbic acid

Ascorbic acid with standard error per mg dry weight of gills in collected *Unio* sp. bivalve samples of Sirpur and Bilawali Tank in summer, monsoon and winter seasons is shown in Table 12. Ascorbic acid with standard deviation per mg dry weight of gills in *Unio* sp. bivalve samples collected from Sirpur Lake site A and site B were 0.774±0.001 and 0.812±0.003 in summer; 1.110±0.006 and 1.119±0.009 in monsoon; 0.916±0.008 and 1.012±0.010 in winter seasons whereas, Bilawali Tank site A and site B were reported 0.812±0.002 and 0.818±0.001 in summer; 1.136±0.012 and 1.169±0.011 in monsoon; 1.041±0.010 and 1.004±0.008 in winter seasons respectively. Ascorbic acid with standard deviation per mg dry weight of digestive gland in *Unio* sp. bivalve samples collected from Sirpur Lake site A and site B were 0.920±0.003 and 0.912±0.003 in summer; 1.254±0.006 and 1.321±0.011 in monsoon; 0.998±0.007 and 1.109±0.010 in winter seasons whereas, Bilawali Tank site A and site B were reported 0.965±0.002 and 0.924±0.002 in summer; 1.321±0.014 and 1.325±0.015 in monsoon; 1.116±0.009 and 1.128±0.008 in winter seasons respectively. Ascorbic acid with standard deviation per mg dry weight of whole body soft tissue in *Unio* sp. bivalve samples collected from Sirpur Lake site A and site B were 0.719±0.002 and 0.795±0.002 in summer; 1.110±0.006 and 1.294±0.011 in monsoon; 0.937±0.006 and 0.994±0.009 in winter seasons whereas, Bilawali Tank site A and site B were reported 0.789±0.002 and 0.795±0.002 in summer; 1.142±0.015 and 1.176±0.015 in monsoon; 0.994±0.008 and 1.012±0.006 in winter seasons respectively.

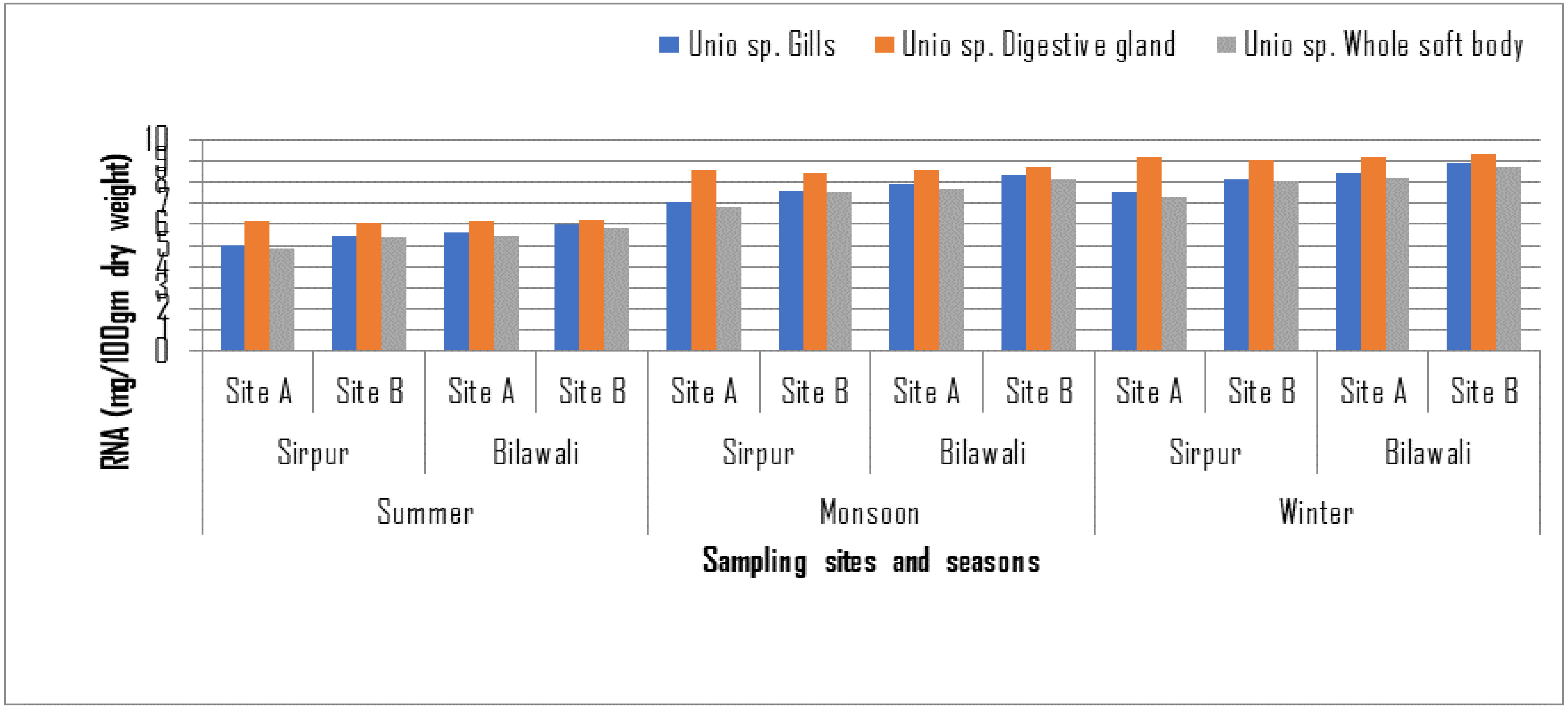
Fig. 60. Ascorbic acid in gills, digestive gland and whole soft body of *Unio* sp. bivalves collected from different sampling sites of Sirpur and Bilawali Tanks of Indore in different seasons.



3.5.1.3 RNA

RNA with standard error per mg dry weight of gills in collected *Unio* sp. bivalve samples of Sirpur and Bilawali Tank in summer, monsoon and winter seasons is shown in Table 12. RNA with standard deviation per mg dry weight of gills in *Unio* sp. bivalve samples collected from Sirpur Lake site A and site B were 5.02± 0.33 and 5.43± 0.34 in summer; 7.02± 0.39 and 7.59± 0.37 in monsoon; 7.51± 0.34 and 8.13± 0.43 in winter seasons whereas, Bilawali Tank site A and site B were reported 5.61± 0.34 and 5.98± 0.36 in summer; 7.85± 0.39 and 8.30± 0.41 in monsoon; 8.39± 0.40 and 8.89± 0.39 in winter seasons respectively. RNA with standard deviation per mg dry weight of digestive gland in *Unio* sp. bivalve samples collected from Sirpur Lake site A and site B were 6.13± 0.39 and 6.03± 0.29 in summer; 8.58± 0.41 and 8.42± 0.34 in monsoon; 9.18± 0.39 and 19.03± 0.39 in winter seasons whereas, Bilawali Tank site A and site B were reported 6.14± 0.29 and 6.25± 0.34 in summer; 8.59± 0.41 and 8.68± 0.43 in monsoon; 9.18± 0.39 and 9.30± 0.37 in winter seasons respectively. RNA with standard deviation per mg dry weight of whole body soft tissue in *Unio* sp. bivalve samples collected from Sirpur Lake site A and site B were 4.87± 0.27 and 5.37± 0.34 in summer; 6.81± 0.39 and 7.50± 0.32 in monsoon; 7.29± 0.46 and 8.04± 0.38 in winter seasons whereas, Bilawali Tank site A and site B were reported 5.47± 0.24 and 5.84± 0.32 in summer; 7.65± 0.39 and 8.11± 0.37 in monsoon; 8.18± 0.46 and 8.69± 0.39 in winter seasons respectively.

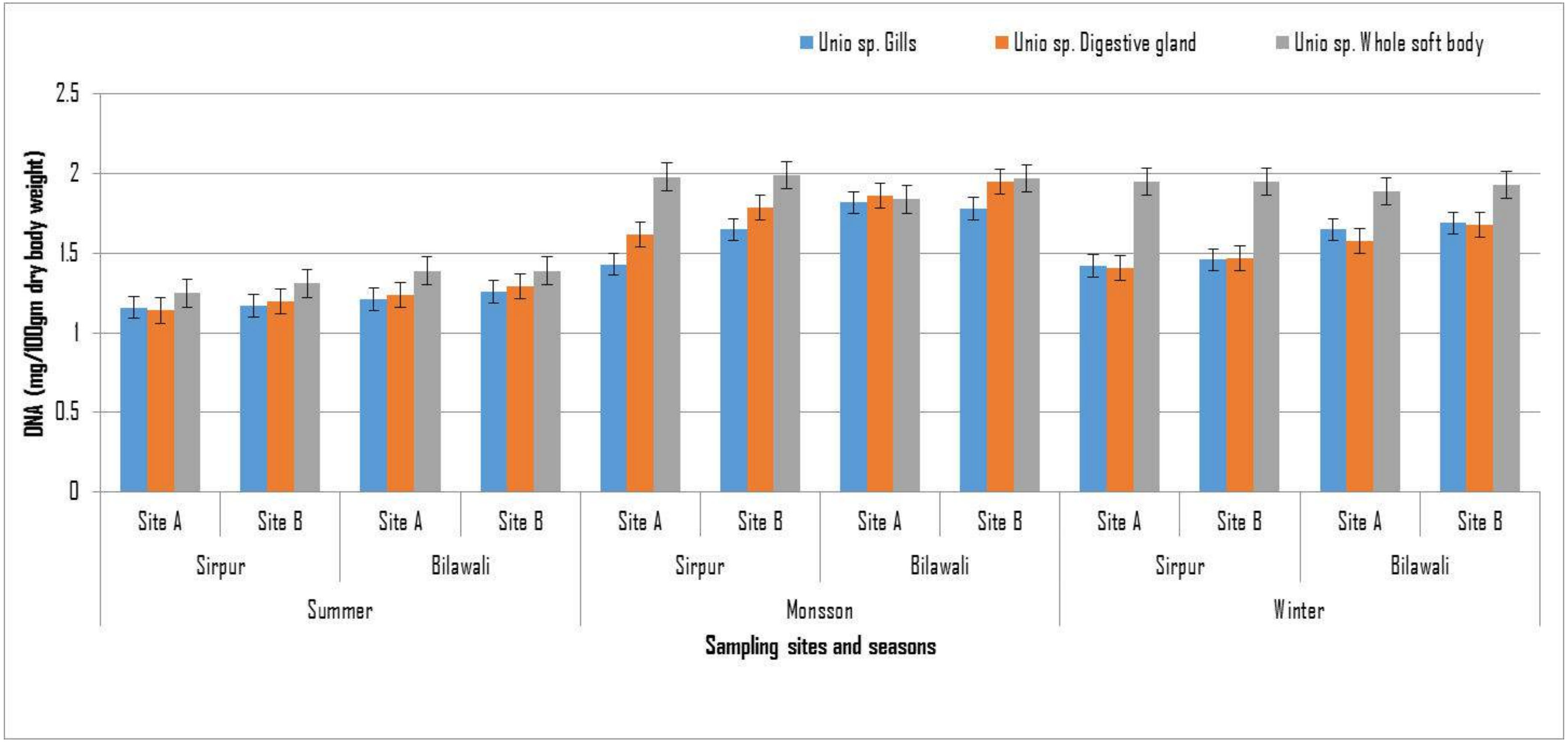
Fig. 61. RNA in gills, digestive gland and whole soft body of *Unio* sp. bivalves collected from different sampling sites of Sirpur and Bilawali Tanks of Indore in different seasons.



3.5.1.3.4 DNA

DNA with standard error per mg dry weight of gills in collected *Unio* sp. bivalve samples of Sirpur and Bilawali Tank in summer, monsoon and winter seasons is shown in Table 12.DNA with standard deviation per mg dry weight of gills in *Unio* sp. bivalve samples collected from Sirpur Lake site A and site B were 1.16±0.19 and 1.17±0.19 in summer; 1.43±0.19 and 1.65±0.20 in monsoon; 1.42±0.21 and 1.46±0.19 in winter seasons whereas, Bilawali Tank site A and site B were reported 1.21±0.21 and 1.26±0.16 in summer; 1.82±0.22 and 1.78±0.17 in monsoon; 1.65±0.21 and 1.69±0.24 in winter seasons respectively. DNA with standard deviation per mg dry weight of digestive gland in *Unio* sp. bivalve samples collected from Sirpur Lake site A and site B were 1.14±0.23 and 1.20±0.17 in summer; 1.62±0.28 and 1.79±0.21 in monsoon; 1.41±0.18 and 1.47±0.24 in winter seasons whereas, Bilawali Tank site A and site B were reported 1.24±0.22 and 1.29±0.21 in summer; 1.86±0.21 and 1.95±0.19 in monsoon; 1.58±0.21 and 1.68±0.24 in winter seasons respectively. DNA with standard deviation per mg dry weight of whole body soft tissue in *Unio* sp. bivalve samples collected from Sirpur Lake site A and site B were 1.25±0.21 and 1.31±0.22 in summer; 1.98±0.28 and 1.99±0.20 in monsoon; 1.95±0.19 and 1.95±0.25 in winter seasons whereas, Bilawali Tank site A and site B were reported 1.39±0.23 and 1.39±0.17 in summer; 1.84±0.21 and 1.97±0.19 in monsoon; 1.89±0.20 and 1.93±0.29 in winter seasons respectively.

Fig. 62. DNA in gills, digestive gland and whole soft body of *Unio* sp. bivalves collected from different sampling sites of Sirpur and Bilawali Tanks of Indore in different seasons.



DISCUSSION

DISCUSSION

Physico-chemical analysis of surface water and heavy metal determination in water and soil sediments of Sirpur and Bilawali Tanks of Indore was studied. Dry weight whole body; metal concentration per unit body weight (µg/g); metal Body burden (µg/individual); BWAF (Bio-water accumulation factor); BSAF (Bio-sediment accumulation factor); protein content, ascorbic acid content and DNA content in gills, digestive gland and whole soft body in of three species of bivalves viz. *Lamellidens corrianus, Lamellidens marginali* and *Unio* sp. collected from Sirpur Lake and Bilawali Tank, Indore were studied to determine effect of pollution on such species in this study and discuss with previous studies.

Physico-chemical studies of surface water samples

Quality water is needed to fulfill the need of human settlement. This is required for domestic purpose, drinking, fishing, irrigation and industries (Pruss, 1998; Thornburn *et al.,* 2003). India has 15 different ecological regions where 1.42 million population lives. About 37.7 million populations of this country are affected by waterborne diseases annually, in which approximately 1.5 million children suffer with diarrhea and allied disease every year. This is results nearly 600 million dollar burden to Indian economy. Over exploitation and pollution are main causes of deterioration of water quality (Pandey *et al.,* 2003). Polluted water quality may affect the aquatic life, animals, plants and humans (Chang *et al.,* 1998; Kara and Comlekci, 2004). A regular monitoring of water quality is needed to maintain the quality of water. Physico-chemical parameters, heavy metal assessment and bioaccumulation studies will needed regular assessment of water quality. Water qualities, heavy metals assessment of water, sediment and bioaccumulation studies in bivalves of Sirpur and Bilawali Tank of Indore Madhya Pradesh in different seasons were studied. Results have shown the higher values of Total alkalinity (mg/l) in Sirpur Lake site A during summer season followed by Sirpur site B, Bilawali site A, Bilawali site B in summer; Sirpur site A, site B, Bilawali site A site B in monsoon and winter seasons. Higher values of Dissolved oxygen (mg/l) in Bilawali Tank site A during winter season and reported gradually decreased values in Sirpur site B, site A, Bilawali site B in winter; Bilawali site B, site A Sirpur site B, site A in monsoon and summer seasons. Highest total hardness (mg/l) was recorded in Sirpur site A and then Sirpur site B, Bilawali site A, site B in summer season. Gradually decreased values were observed in Sirpur site A, site B, Bilawali site A, site B in winter season; Sirpur site A, site B, Bilawali site A, site B in monsoon season. Highest Salinity (mg/l) was recorded in Sirpur site A and then Sirpur site B, Bilawali site A, site B in summer season. Gradually decreased values were observed in Sirpur site A, site B, Bilawali site A, site B in winter season; Sirpur site A, site B, Bilawali site A, site B in monsoon season. Highest Chloride (mg/l) was recorded in Sirpur site A and then Sirpur site B, Bilawali site A, site B in summer season. Gradually decreased values were observed in Sirpur site A, site B, Bilawali site A, site B in winter season; Sirpur site A, site B, Bilawali site A, site B in monsoon season. Temperature was recorded within normal range everywhere. Highest pH (unit) was recorded in Sirpur site A in every season in comparison to other sampling sites. This result clearly indicated that Site A (Sirpur lake) was more polluted than Bilawali Tank sites and Sirpur site B. Obtained results are statistically proven with $P<0.05$The higher pH, total hardness ,total alkalinity, chlorides, salinity, and lowest dissolved oxygen levels were found in Site A surface water (Sirpur lake) may be due to huge agricultural runoff, domestic waste, textile, and other manufacturing residues. Catchment areas of Sirpur Lake Site A are surrounded by agricultural field, which is polluted by different fertilizers and pesticides. Earlier report told the high pH values were recorded, may be due to heavy input of textile, printing and other effluent with municipal waste (Kalff and Knoechel, 1998). Richardson (1988) worked on pollution of water and reported heavy contamination in water, which alter the physico-chemical properties of water. Dehadri (1990) also reported the direct discharge of industrial effluents deplete different characters of water viz. dissolved oxygen, pH, increase the CO_2 level in the water. Kolhe *et al.*, (2014) also told a story of pH modification after fusion of household and byproducts in water. In this analysis, higher pH was observed during the summer season than in the winter and monsoon seasons. This can be due to increased photosynthetic absorption of inorganic carbon dissolved by planktons (Subba rao and Govind, 1967; Goldman, 1972; Farrell *et al.,* 1979). Sharma and Jain (2000) recorded low pH in winter as well as higher in summer season.Mishra and Tripathi (2001); Singh *et al.,* (2002) also supported the results of like reports. The high pH recorded for Venkatesharaju (2010) in winter is due to a decreased rate of decomposition.

Heavy Metal Concentrations in sediment and surface water samples

Assessments of heavy metals in surface water samples and sediment of two different sites of Sirpur and Bilawali Tanks of Indore were performed and Median concentrations recorded of Zn, Cu, Pb Cd and Mn in surface water of Sirpur Site A and Site B were higher than site A and B of Bilawali Tank in summer and gradually lower in similar manner in winter and monsoon. The main reason behind the pollution of Site A (Sirpur lake), is catchment areas of surrounded by agricultural field, which is polluted by fertilizers and pesticides, these could acts as source of heavy metals Cu, Mn, Pb, Zn and As (Verkleji, 1993; de Meeus *et al.*, 2002). Discharges of untreated household waste and water into water bodies are a major cause of heavy metal contamination (Hadjmohammad, 1988; Rashed, 2001; Ravera, 2004; Aksoy *et al.*, 2005). Abaychi and DouAbul (1985) Stated that treated and untreated urban, industrial waste, agricultural run-off leads to the emissions from heavy metals Cu, Pb, Cd and Zn. Lokhande et al., (2011) stated that the major industries contributing to the contamination of Mn, Pb and Cu in the aquatic environment are colours, paints, textile industries. Jaishree and Khan (2014) claimed that textile dyeing and printing wastewater effluents include colours, bleaching agents, salts, acids, and heavy metals such as Cr, Cu, Pb, and Zn. The results show that the concentrations of metals in sediments varied widely and show variations in values of Mn, Cu, Pb, Cd and Zn between different reservoirs. The results showed a concentration pattern close to that of its abundance in water . Sirpur Lake Site A carries enormous amounts of domestic waste, garment, printing, industrial dyeing and other effluent together with organic matter, run-off from agricultural waste and traffic run-off, which is driven by higher amounts of Mn, Cu, Pb ,Mn and Zn heavy metals in sediment samples collected from Site B. Halcrow et al., (1973) reported earlier that concentrations of heavy metals in sediments increased. Presley et al. (1980) reported that the elemental sediment concentrations depend not only on anthropogenic sources, but also on the quality of organic matter, textural characteristics, mineralogical composition and sediment deposition environment. Harland et al., (2000) researched and documented the reliance on organic matter and particle size of metal concentrations in the sediments. Accumulation of heavy metals in sediments may affect concentrations of heavy metals in aquatic organisms living in these sediments (Pourang, 1996; Wildi *et al.,* 2004; Kim and Kim, 2006). Overall results showed that highest concentration of heavy metals Mn, Cu, Pb, and Zn in sediments samples of Sirpur Lake Site A than Sirpur Lake Site B, Bilawali Tank Site A and Bilawali Tank Site. However, tracing Hg in both water and soil sediments was a cumbersome process with high degree of error.

Bioaccumulation Study

Concentrations of heavy metals viz. Mn, Cu, Pb, Cd and Zn metal body burden, BWAF and BSAF values were determined in dry weight of whole soft body tissues of three different freshwater bivalve species i.e. *Lamellidens corrianus*, *Lamellidens marginalis* and *Unio.sp*. collected from Sirpur Lake Site A, Site B; Bilawali Tank Site A, Site B of Indore Madhya Pradesh during monsoon, summer and winter seasons. Results indicate that mean highest values of concentrations of heavy metals in body, metal body burden and BWAF and BSAF in dry soft body tissues of all sampled bivalve species collected from Sirpur Lake Site A than Sirpur Lake Site B, Bilawali tank Site A and Bilawali tank Site B. Reported higher metal concentrations and body burden in whole dry soft body tissues of all sampled bivalve species collected from Sirpur Lake Site A might be due to exposure of higher pollutant than other sampling sites. Shinde (2013) reported in his study that metal concentrations in the dry soft body tissues of mollusc were related to metal levels present in water bodies and soil. Deshmukh (2013) also supported this report. It includes the BWAF / BSAF values used to identify the most suitable sentinel species to track heavy metal contamination in water. In this study BWAF values refer to the concentration of a specific metal in bivalve tissues per concentration of that metal in water. Biosediment Accumulation Factor is specified as the metal concentration ratio of the bivalve tissue to that in the sediment (Usero *et al.,* 2005; Szefer *et al.,* 1999). In moonsoon, the concentrations of heavy metals, metal body load, BWAF and BSAF throughout all bivalve species were low compared to summer and winter seasons, this could be due to the rise in water levels in water bodies (Patil *et al.*, 2004; Deshmukh, 2013).

Oxidative Stress

Antioxidant enzyme activity: viz. SOD, CAT, GPx and GST; levels of GSH and LPO of three different freshwater bivalve species were assessed in the dry weight of the digestive gland tissues i.e. *Lamellidens corrianus*, *Lamellidens marginalis* and *Unio. sp.* collected from Sirpur Lake Site A, Site B; Bilawali Tank Site A, Site B of Indore Madhya Pradesh during monsoon, summer and winter seasons. Digestive gland is the main site of metal accumulation in the body and as it contains greater level of metallothenin (Pipe *et al.,* 1999; Canesi *et al.,* 2008; Waykar and Shinde, 2013; Deshmulh, 2013). Even digestive glands often also show higher level of antioxidant enzymes n body (Irato *et al.,* 2003). Different toxicant is generating reactive oxygen species and induces the LPO formation. This may increase the activity of GST, decrease the level of GSH and alter different antioxidant enzyme viz. SOD, CAT and GPx activities in different mollusk (Vasseur and Leguille, 2003; Box, *et al.,* 2007; Osman *et al.,* 2007; Shinde, 2013; Deshmukh, 2013). Such studies have indicated the oxidative stress indicator and act as a powerful and cost-effective tool to get information of environment and effect of contaminants on living biological resources. Highest level of LPO and Glutathione-S-transferase activity and lowest superoxide dismutase, glutathione peroxidase and catalase activity and low levels of decreased glutathione (GSH) in the bivalve species ' digestive glands, *Lamellidens corrianus*, *Lamellidens marginalis* and *Unio.sp* collected from Sirpur lake Site A than others. On the other side, the lowest level of lipid peroxidation and Glutathione-S-transferase activity and the highest activity in the digestive glands of superoxide dismutase, glutathione peroxidase and catalase and reduced glutathione (GSH) of three bivalve species collected from Bilawali tank Site B than others. Lipid peroxidation is a key aspect of cell injury and seems to be largely results by free radical reactions in biological membranes, being rich in polyunsaturated fatty acids (Chesseman, 1982). Metals are known inducers of Reactive oxygen species which covalently bind to macromolecules and inflict peroxidative degeneration of the endoplasmic reticulum lipid membrane; which is rich in polyunsaturated fatty acids. This leads to formation of lipid peroxide, which in turn gives products like, MDA that causes damage to membrane. The substantial increase in lipid oxidation (MDA) may signify the vulnerability of lipid molecules to reactive oxygen species and the degree to which these molecules suffer oxidation harm (Jamil, 2001). Increased lipid peroxide (LPO) is one of the most important contributors to the loss of cell function in oxidative stress conditions (Hermes-Lima *et al.*, 1995). The glutathione-s-transferase (GST) enzyme involved in the detoxification of environmental pollutants, as well as endogenous toxic compounds such as lipid peroxidation products (Ketterer and Chistodoulides, 1994; Awasthi *et al.*, 1995). In the current investigation the highest activity of glutathione-S-transferase (GST) was observed in digestive glands of three bivalve species collected from Site A (Sirpur lake) than other three sites might be due to bivalve Species have been exposed to greater levels of toxins than three other sites. Evidence collected in the present study showed the highest concentrations of heavy metals Zn, Cu, Pb, Mn and Cd in surface water; soil sediments and three bivalve species inhabiting at this reservoir than other three studied reservoirs. Metals are known inducers of reactive oxygen species (ROS). Higher GST activity at Site A (Sirpur lake) in bivalve digestive glands, the ability of the digestive glands to metabolize xenobiotics could be related; eliminate waste products (Gamble *et al.,* 1995) and it also suggest the defensive action against reactive oxygen radicals. Increase of GST enzyme activity also indicating activation of detoxification complexes in the digestive glands could be a good indicator of the pollutant exposure. Increased GST activity can therefore be due to higher detoxification of hydroperoxides. Many researchers revealed that the increase in activities of GST after exposure to toxicants. Sheehan *et al.,* (1995), reported an increase in GST activity in mussel, *Mytilus edulis* exposed to pollutants. Canesi *et al.,* (1999) reported an increase in GST activity in gill and digestive glands of *M. edulis* after exposure to Cu and Hg. Schuliga *et al.,* (2002) and Ventura-Lima *et al.,* (2007) have shown the modulation of GST activity on arsenic exposure. Compare to Site A (Sirpur lake) the bivalve species inhabiting at Site B (Sirpur lake)showed lesser level of LPO and activity of glutathione-S-transferase (GST) in digestive glands of three bivalve species, this might be related to bioaccumulated metal concentration in bivalves. Results indicate that bivalve inhabiting at Site B (Sirpur lake) are exposed to low levels of pollutants than as in Site A (Sirpur lake). In current study gathered data revealed the low concentration of heavy metals Zn, Cu, Pb, Mn and Cd in surface water, soil sediments and three bivalve species at Site B (Sirpur lake) than Site A (Sirpur lake). Therefore, this study clearly indicated that Site B (Sirpur lake) was less polluted than Site A (Sirpur lake). Reduced glutathione (GSH) is the major non-protein thiol and is the important endogenous antioxidant which plays a crucial role in the defense against oxidative damage (Gohary *et al.,* 1999; Sies, 1999). GSH could be the first line of defense against metal contamination by metal chelation or participation in the GPx or GST detoxification process (Sies, 1999). It has been proved that reduced glutahione is One of the most effective ROS scavengers produced as cell metabolism by-products or during oxidative stress (O'Brien *et al.,* 2001; Tsukamoto *et al.,* 2002; Han *et al.,* 2008). GSH have antioxidant properties, and Its protective function toward oxidative stress induced toxicity is well known in aquatic organisms (Hasspielar *et al.,* 1994; Gohary *et al.,* 1999). Aquatic organisms have evolved a suite of enzymatic and non-enzymatic defenses to cope with the production of reactive oxygen species (ROS). The antioxidant defense enzyme system includes several enzymes such as Catalase (CAT), Superoxide Dismutase (SOD), and Glutathione peroxidase (GPx). Many among these antioxidants interact in a concerted manner to eliminate reactive oxygen species and prevent damage to cellular components. These enzymes activities can be affected by reactive oxygen species (ROS) and therefore they may represent indicators of oxidative stress (Valavanidis *et al.,* 2006 ;Pavlovic *et al.,* 2004). Various antioxidant enzyme activities are mostly likely used as indicators of oxidative stress (Cargnelutti *et al.,* 2006; Bocchetti *et al.,* 2008 ;Banni *et al.,* 2008; Zhou *et al.,* 2008). The lowest activity of CAT , SOD and GPx activity was associated with enhanced LPO formation. A deficiency in these cellular defense enzymes might decline the capacity of aquatic organism to neutralize the production of ROS. Numerous researchers showed that the toxicants initiate the LPO formation, increases the activity of GST, decrease GSH level and alter antioxidant enzyme (SOD, CAT and GPx) activities in mollusk (Vasseur and Leguille, 2003; Osman *et al.*, 2007; Box, *et al.*, 2007; Shinde, 2013; Deshmukh, 2013).

Biochemical Study

In the present investigation, biochemical constituents like protein, ascorbic acid, RNA and DNA contents were determined from the soft body tissues like gills, digestive glands and whole soft body tissues of three bivalve species, *Lamellidens corrianus*, *Lamellidens marginalis* and *Unio.sp* inhabiting the two reservoirs of Indore district during three seasons and gathered results were compiled in table nos. 10 to 12 and fig. nos. 51 to 62.

The results revealed lowest protein, ascorbic acid and DNA in soft body tissues of three groups of bivalves collected from Site A (Sirpur lake) than Site B (Sirpur lake), Site A (Bilawali tank) and Site B (Bilawali tank), though high protein, ascorbic acid and DNA content of three bivalve species sampled from Site B (Bilawali Tank) were observed in soft body tissues reservoir than other three studied reservoirs. The findings of the ANOVA test showed that the variation between the mean values of biochemical components between reservoirs, seasons and bivalve species ($P<0.05$) was significantly different.

In the present study obtained results showed the low level of protein content, ascorbic acid content RNA and DNA content in soft body tissues of three bivalve species located from Site A (Sirpur lake) than other three observed sites of both reservoirs, could be because of the bivalves present at Site A (Sirpur lake) were subject to higher pollutant load than three other sites of both reservoirs recorded. The findings of the present study showed highest concentrations of Zn, Cu, Mn, Pb and Cd heavy metals in surface water, sediments and three bivalve species sampled from Site A (Sirpur lake) than other three analyzed sites of reservoirs. The gained heavy metals were enough to interrupt the natural equilibrium of oxidation / reduction in the cells by generating reactive oxygen species which lead to oxidative stress (Abdullah *et al.*, 2004). In the appearance of reactive oxygen species (ROS), proteins can be weakened by oxidative attack, resulting in peptide chain fragmentation, site-specific variations of amino acids, disrupted electrical load, accumulation of cross-linked reaction products and increased proteolysis sensitivity (Grune, 2000; Requena *et al.*, 2003). The low protein content found in different tissues suggests that environmental pressure decreases translations levels or increases protein catabolism to meet the high energy demands of toxicants (Vincent *et al.*, 1995; Waykar and Lomte, 2001a). The lesser protein composition could be the result of cell destruction / necrosis and resulting degradation of protein synthetic machinery (Umminger, 1977; Bradbury *et al.*, 1987). Protein and amino acid catabolism contribute greatly to total energy output. It is known that structural proteins are used as energy source under stressful conditions

(Claybrook, 1983). Pottinger *et al.,* (2002) disclosed that protein synthesis, represents disruption of normal metabolic processes, may be suppressed at high pollution stress. This work provides information regarding the heavy metal level in surface water and soil sediment and in three bivalve species and biomarker responses in relation to heavy metal accumulation at two reservoirs of Indore district. This research focused on seasonal fluctuations in heavy metal concentrations of surface water and soil sediments, bivalve organisms, and their effects on biomarker responses and biochemical components.

CONCLUSION AND RECOMMENDATIONS

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Reports of heavy metal accumulation in bivalves and their presence in water as well as in sediment are helpful in designing a system of heavy metals removal from water bodies and compliance. Bivalves are proven their usefulness as bio-indicator organisms and provide ideally, an estimate of trace elements availabilities in biomass of water bodies. Heavy metal body burden in bivalves have been used to identify and sketch the areas with exceedingly higher levels of trace metals and organic pollutants. This is due bivalves can be used as bio-monitors for aquatic environment. Bivalves can also providing the details of toxicity and alarm if anyone eat bivalves as food. The baseline data on metal concentrations in Sirpur Lake and Bilawali Tank may contribute in evaluating future impact of anthropogenic activities and progress. Biochemical studies are providing baseline data for assessment of bio-monitoring studies as reference. This study will deal the importance of biomarker research and incorporating the Indian freshwater monitoring program. Results of this study demonstrate the mean concentrations of Zn, Cu, Cd , Pb and Mn in surface water and soil sediment of Sirpur Site A and Site B were higher than site A and B of Bilawali Lake in summer and gradually lower in similar manner in winter and monsoon. Reported values were also higher than the recommendation of WHO limits for drinking water standard. Therefore, central water treatment plants urgently need to treat water and extract heavy metals before using this water for crop and drinking purposes. The haphazard anthropogenic activity near water bodies need to be controlled. Effective environmental monitoring exercise should also be encouraged to check the flow of heavy metals and other pollutant.

SUMMARY

SUMMARY

Human populations are increasing rapidly in the world and to fulfill the requirement of their needs, anthropological activities are progressively increasing.

Water is an elixir of life, so urbanization near water bodies is establishing. Settlement of population needs leads to technology development, industrialization, farming, and many more activities. These all have been adversely affecting environment and ecosystem.

Heavy metals viz. As, Zn, Cu, Pb, Cd, Mn, Hg, Ni and Cr are some trace metals present in polluted water. Higher concentration of these heavy metals makes harmfully affect to population of water bodies and even they are non-biodegradable and are persistent in nature.

Heavy metals may be transferred through accumulation in lower trophic organisms to higher trophic organism through food chain and leads to bad effect on ecological balance of environment and diversity of aquatic organisms.

The body burdens of metals in most bivalves have been used to identify and map areas with exceedingly high levels of trace metals and organic pollutants hence they can be used as biomonitors for aquatic environment.

Potential of bivalve to accumulate metals from water/sediments into its tissues can be estimated using Bio-water Accumulation Factor (BWAF) and Bio-sediment Accumulation Factor (BSAF). By comparing BWAF/BSAF values,

one can compare the potential of different bivalve species to accumulate metals from water and sediments in to their body tissues.

Bioaccumulation only detects the levels of heavy metals, in aquatic organisms, but a true evaluation of the damage inflicted by heavy metals should come from comprehensive biomarker studies.

India has 15 different ecological regions where 1.42 million population lives. About 37.7 million populations of this country are affected by waterborne diseases annually, in which approximately 1.5 million children suffer with diarrhea and allied disease every year. This is results nearly 600 million dollar burden to Indian economy. Over exploitation and pollution are main causes of deterioration of water quality. Polluted water quality may affect the aquatic life, animals, plants and humans. A regular monitoring of water quality is needed to maintain the quality of water.

In the present study different native species of freshwater bivalves, *Unio* sp., *Lamellidens corrianus*, *Lamellidens marginalis* were selected to establish a local environmental monitoring network using bivalves as bioindicator species to evaluate the trends of bioaccumulation of Zn, Cu, Pb and Mn in freshwater ecosystem.

Sirpur Lake and Bilawali tank of Indore, Madhya Pradesh was taken for this study. Surface water, sediments soil and three species of bivalve viz. *Unio sp*., *Lamellidens corrianus* and *Lamellidens marginalis* were sampled from two different sites of each water bodies.

Surface water, sediments soil and three species of bivalve viz. *Unio sp*., *Lamellidens corrianus* and *Lamellidens marginalis* were sampled.

Physico-chemical parameters viz. total alkalinity, dissolved oxygen, total hardness, salinity and chloride; heavy metals viz. Zn, Cu, Pb and Mn were determined in laboratory as per standard method of APHA 1998.

Metal bioaccumulation in bivalve species, Biowater Accumulation Factor (BWAF) and Biosediment Accumulation Factor (BSAF) was calculated. The BWAF factor is defined as the ratio between the concentration of metal in the organism and that in the water. The BSAF is defined as the ratio between the concentration of metal in the organism and that in the sediments.

Digestive glands of five animals of each species was collected, separated, surface blotted with tissues paper and thoroughly washed with phosphate buffer (50mM; pH 7.4). LPO, GSH, SOD, CAT, GPx and GST was performed for such samples.

Whole soft body tissues were removed and dried at 70^0 to 80^0C in the oven till the constant weight of dry tissues was obtained. Protein, ascorbic acid, DNA and RNA content was estimated.

Results have shown the higher values of Total alkalinity (mg/l) in Sirpur Lake site A during summer season followed by Sirpur site B, Bilawali site A, Bilawali site B in summer; Sirpur site A, site B, Bilawali site A site B in monsoon and winter seasons.

Higher values of Dissolved oxygen (mg/l) in Bilawali Lake site A during winter season and reported gradually decreased values in Sirpur site B, site A, Bilawali site B in winter; Bilawali site B, site A Sirpur site B, site A in monsoon and summer seasons.

Highest total hardness (mg/l) was recorded in Sirpur site A and then Sirpur site B, Bilawali site A, site B in summer season. Gradually decreased values were observed in Sirpur site A, site B, Bilawali site A, site B in winter season; Sirpur site A, site B, Bilawali site A, site B in monsoon season.

Highest Salinity (mg/l) was recorded in Sirpur site A and then Sirpur site B, Bilawali site A, site B in summer season. Gradually decreased values were observed in Sirpur site A, site B, Bilawali site A, site B in winter season; Sirpur site A, site B, Bilawali site A, site B in monsoon season.

Highest Chloride (mg/l) was recorded in Sirpur site A and then Sirpur site B, Bilawali site A, site B in summer season. Gradually decreased values were observed in Sirpur site A, site B, Bilawali site A, site B in winter season; Sirpur site A, site B, Bilawali site A, site B in monsoon season.

Temperature was recorded within normal range everywhere. Highest pH (unit) was recorded in Sirpur site A in every season in comparison to other sampling sites. This result clearly indicated that Site A (Sirpur lake) was more polluted than Bilawali Lake sites and Sirpur site B. Obtained results are statistically proven with $P<0.05$.

The higher values of pH, total alkalinity, total hardness, chlorides, salinity and lowest dissolved oxygen were observed in surface water of Site A (Sirpur lake) may be due to huge agricultural runoff, domestic waste, textile and other industrial effluents. Catchment areas of Sirpur Lake Site A are surrounded by agricultural field, which is polluted by different fertilizers and pesticides. There are two sugar and one cotton mill are located in the basin area of this Lake. Kalwan, Satana, Deola and some others are neighboring towns of this Lake and discharges untreated domestic waste to this Lake. Maximum pH in winter is due to decreased decomposition rate.

Assessments of heavy metals in surface water samples and sediment of two different sites of Sirpur and Bilawali Lakes of Indore were performed and reported the mean concentrations of Zn, Cu, Pb and Mn in surface water of Sirpur Site A and Site B were higher than site A and B of Bilawali Lake in summer and gradually lower in similar manner in winter and monsoon. The main reason behind the pollution of Site A (Sirpur lake), is rivulets of Site A (Sirpur lake) river namely Tamdi, Punand, Aram, Mosam, Masa Nadi, Baindki and markhandi etc. The catchment areas of these rivulets are surrounded by agricultural field, which is polluted by fertilizers and pesticides, these acts as source of heavy metals Cu, Cd, Pb, Zn and As. Overall results showed that highest concentration of heavy metals Mn, Cu, Pb, and Zn in sediments samples of Sirpur Lake Site A than Sirpur Lake Site B, Bilawali Tank Site A and Bilawali Tank Site B.

Concentrations of heavy metals viz. Mn, Cu, Pb, Cd and Zn metal body burden, BWAF and BSAF values were determined in dry weight of whole soft body tissues of three different freshwater bivalve species i.e. *Lamellidens corrianus*, *Lamellidens marginalis* and *Unio.*species collected from Sirpur Lake Site A, Site B; Bilawali Tank Site A, Site B of Indore Madhya Pradesh during monsoon, summer and winter seasons. Results indicate that mean highest values of concentrations of heavy metals in body, metal body burden and BWAF and BSAF in dry soft body tissues of all sampled bivalve species collected from Sirpur Lake Site A than Sirpur Lake Site B, Bilawali tank Site A and Bilawali tank Site B. Reported higher metal concentrations and body burden in whole dry soft body tissues of all sampled bivalve species collected from Sirpur Lake Site A might be due to exposure of higher pollutant than other sampling sites.

Digestive gland is the main site of metal accumulation in the body and as it contains higher level of metallothenin. Different toxicant is generating reactive oxygen species and induces the LPO formation. This may increases the activity of GST, decrease the level of GSH and alter different antioxidant enzyme viz. SOD, CAT and GPx activities in different mollusk. Such studies have indicated the oxidative stress indicator and act as a powerful and cost-effective tool to get information of environment and effect of contaminants on living biological resources.

Highest level of LPO and Glutathione-S-transferase activity and lowest activity of superoxide dismutase, catalase and glutathione peroxidase and low level of reduced glutathione (GSH) in the digestive glands of bivalve species, *Lamellidens corrianus*, *Lamellidens marginalis* and *Unio.sp* collected from Sirpur lake Site A than others. On other hand lowest level of lipid peroxidation and glutathione-S-transferase activity and highest activity of superoxide dismutase, catalase and glutathione peroxidase and level of reduced glutathione (GSH) in the digestive glands of three bivalve species collected from Bilawali tank Site B than others.

Lipid peroxidation is a main aspect in cellular injury and results largely from free radical reactions in biological membranes, which are rich in polyunsaturated fatty acids (Chapman, 1997). Metals are known inducers of reactive oxygen species which binds covalently to the macromolecules and induce peroxidative degeneration of the lipid membrane of endoplasmic reticulum, which is rich in polyunsaturated fatty acids. This leads to formation of lipid peroxide, which in turn gives products like, MDA that causes damage to membrane. The significant increase in lipid oxidation (MDA) may indicate the susceptibility of lipid molecules to reactive oxygen species and the extent of oxidative damage imposed on these molecules.

Increased lipid peroxide (LPO) is one of the most important contributors to the loss of cell function in oxidative stress conditions (Hermes-Lima *et al.*, 1995). The glutathione-s-transferase (GST) enzyme involved in the detoxification of environmental pollutants, as well as endogenous toxic compounds such as lipid peroxidation products

In the present investigation the highest activity of glutathione-S-transferase (GST) was observed in digestive glands of three bivalve species collected from Site A (Sirpur lake) than other three reservoirs might be due to bivalve species were exposed to higher level of pollutants than other three reservoirs. In the present study obtained data revealed highest concentrations of heavy metals Zn, Cu, Pb and Mn in surface water, soil sediments and three bivalve species inhabiting at this reservoir than other three studied reservoirs. Metals are known inducers of reactive oxygen species (ROS).

Higher GST activity at Site A (Sirpur lake) in the digestive glands of the freshwater bivalve might be related to the capacity of the digestive glands to metabolize xenobiotics, eliminate waste products and it also suggests the protective action against reactive oxygen radicals. Increase of GST enzyme activity indicating activation of detoxification mechanism in the digestive glands could be a good indicator of pollutant exposure. Increase of GST activity can therefore be due to increased detoxification of hydroperoxides.

Lowest level of LPO and activity of Glutathione-S-transferase (GST) in digestive glands of three bivalve species, this might be related to bioaccumulated metal concentration in bivalves. Results indicate that bivalve inhabiting at Site B (Sirpur lake) was exposed to low level of pollutants than Site A (Sirpur lake). In the present study obtained data revealed the low concentration of the heavy metals Zn, Cu, Pb,Cd and Mn in surface water, soil sediments and three bivalve species at Site B (Sirpur lake)than Site A (Sirpur lake).

This study clearly indicated that Site B (Sirpur lake)was less polluted than Site A (Sirpur lake). Reduced glutathione (GSH) is the major non-protein thiol and is the important endogenous antioxidant which plays a central role in the defense against oxidative

In the present investigation the biochemical constituents like protein, ascorbic acid, DNA and RNA contents were determined from soft body tissues like mantle, gills, digestive glands and whole soft body tissues of three bivalve species, *Lamellidens corrianus*, *Lamellidens marginalis* and *Unio.sp* in habiting the two reservoirs of Indore district during three seasons. The results showed, lowest protein, ascorbic acid and DNA a contents in soft body tissues of three bivalves species sampled from Site A (Sirpur lake) than Site B (Sirpur lake), Site A (Bilawali tank) and Site B (Bilawali tank), while highest protein, ascorbic acid, DNA and RNA contents were observed in soft body tissues of three bivalve species sampled from Site B (Bilawali Tank) reservoir than other three studied reservoirs. The results of ANOVA test indicated that the difference between the mean values of biochemical constituents were varied significantly between reservoirs, seasons and bivalve species ($P<0.05$). In the present study obtained results showed the low level of protein, ascorbic acid, DNA and RNA contents in soft body tissues of three bivalve species collected from Site A (Sirpur lake) than other three studied reservoirs, might be due to bivalves inhabiting at Site A (Sirpur lake) were exposed to higher load of pollutants than other three studied reservoirs.

Seasonal variation of heavy metal concentrations in surface water and soil sediment and bivalve species and their effect on biomarkers responses and biochemical components are focused in this study and reported information regarding heavy metal level in surface water and soil sediment and in three bivalve species and biomarker responses in relation to heavy metal accumulation at reservoirs of Indore district.

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Appendices