Invertebrate survey of Mulloon Creek during wet period 2022-23

Third report on the biological and physical parameters of water quality in the Mulloon Creek catchment, New South Wales

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Mulloon Institute
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Acknowledgements
The authors of this report wish to acknowledge the help and input of Luke Peel and Peter Hazell throughout the study and the guidance for the appropriate sites to sample. Luke Peel previously contributed the map (Figure 1) of the Mulloon Creek and Reedy Creek sites. The Mulloon Rehydration Initiative is jointly funded through the Mulloon Institute, the NSW Government through its Environmental Trust and the Australian Government's National Landcare Program.

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Front cover pictures (clockwise):
Stone flies (Gripopterigidae) from Black Jackie
May fly (Leptophlebiadae)from Reedy Creek in January 2023
Beetle larvae (Ptilodactylidae) from Reedy Creek in April 2023
Black Jackie pond in October 2022

Introduction

Macroinvertebrates of freshwater can serve as an indicator of water quality as well as the changes that occur to water over a season (Rosenberg and Resh 1993). The types of macroinvertebrates are considered to vary in water as a result of differences in species in their ability to cope with the differing physical parameters of the water in which they dwell (Resh and Rosenberg 1984). The major physical changes in water are those that can affect the life cycle of the invertebrates, as well as their ability to survive over the short term (Johnson *et al.* 1993).

Although many studies have examined macroinvertebrates in Australia (*e.g.* (Thomson *et al.* 2012, Verkaik *et al.* 2014), few studies have considered the smaller catchments that may change as farming practices change, especially overtime and during droughts and extensive rain. The review by Lester and Boulton (2008) on placing timber into the waterways of agricultural streams was one of the rare papers to indicate ways of improving the water conditions following many years of agricultural activity. However, no studies in Australia have reported how water quality changes over time with restoration of agricultural landscapes, although a large literature exists in North America regarding the same question.

The study of macroinvertebrates can examine changes in either taxonomic groups or functional groups (usually based on feeding). These patterns can be compared to a number of stream conditions, such as the water temperature, dissolved oxygen, pH, conductivity and flow rate. Each of the environmental aspects affects some aspect of the macroinvertebrate life cycle (Johnson *et al.* 1993). For example, temperature determines the rate of growth of macroinvertebrates, but also influences the oxygen that can be dissolved in the water. As temperature increases, the capacity of water to hold oxygen decreases. Oxygen is necessary to meet the macroinvertebrate metabolic requirements. There is also a relation between temperature and metabolic rate, as the metabolic requirements of macroinvertebrates increase with temperature (Harrison *et al.* 2012), so not only does the requirement for oxygen increase as temperature increases, but the availability of oxygen typically decreases at the same time in water.

The dissolved salts in water can also affect the amount of oxygen that is present, and conductivity is an indication of dissolved salts in water (Randall *et al.* 2002). As conductivity increases, the amount of oxygen that is held in water decreases, but freshwater is typically defined as conductivity that is below the threshold for reducing oxygen concentration. However, salts and pH can affect the ability of invertebrates to regulate ions and water within their body, so that changes in these parameters can be reflected in the invertebrates that are present in various locations where these parameters change (Cooper 2004).

Finally, the rate at which water flows and the substrate that forms the anchor for invertebrates can cause a change in the composition of observed invertebrates. As vegetation increases on the substrate, the invertebrates have a greater ability to find locations to adhere, so that any change in water flow is limited in causing their movement down the stream (Lester and Boulton 2008), a situation that is exacerbated in droughts. However, rapid flows can reduce the vegetation, especially reeds, making fewer locations for insects to attach. All these physical components can become more important as drought extends the time between renewal of water movements between the series of ponds that exist during droughts, potentially benefitting some taxa, but decreasing populations of other taxa((Cooper 2022, Cooper and Wallenius 2017)).

In this report, I present information regarding the macroinvertebrate communities that are present at six sites along Mulloon and Reedy Creeks over 12 months (September 2022-August 2023) along with the physical conditions that were present in the separate sampling areas located at each site at that time. Mulloon Creek originates in the Tallagandra State Forest, flows in a northerly direction through agricultural land, and joins Reedy Creek before entering the Shoalhaven River system near Braidwood. I examined how the physical parameters may influence the composition of invertebrates at various locations as well as how stable the communities were over the period of measurement. This study can be compared to the previous studies when rain occurred in each month of the survey and the creek was flowing continuously and by an extended drought period when rain only fell in the last 2 months. The effects of extended rain periods measured over this year are important for understanding how future work and development along Mulloon Creek can influence the macroinvertebrate communities when rain input is not limiting.

Methods and sites

Study sites

Six sites along Mulloon Creek were chosen and are shown relative to the catchment in Figure 1 and are the same sites examined in the previous two studies (Cooper 2022, Cooper and Wallenius 2017).

Invertebrates were collected by sweep net, as water flow was high and conditions at most sites were deep water and high flow. At least 10 sweeps were done per location to standardize amount of water sampled, with three collections made per site. Although no effort was made to determine substrate type in detail, notes of the substrate were made to indicate potential variation among samples at each site (see Appendix 1 for site details). Invertebrates were collected at each site in October 2022 and January, April and July 2023. Physical measurements were made monthly from Septemberl 2022-August 2023, with access to some sites limited by the conditions of rainfall (especially Reedy Creek in October and November 2022, figure x)

Physicochemical analysis of water

Samples were made monthly at the various locations listed above for the parameters of water temperature, dissolved oxygen, conductivity, pH and water flow. Water temperature and dissolved oxygen measurements were made using battery operated electronic thermometer (˚C) (WP-90, TPS Australia) and oxygen electrode (% saturated) (WP-82, TPS Australia). Thermometer was calibrated against a digital thermometer traceable to NBS standards and oxygen electrode calibrated as directed in the instructions from TPS Australia. The conductivity was measured using a portable system (Activon) and measured to the nearest 0.001 mS cm^{-1}. pH was determined using pH paper (Merck) capable of measuring from pH 0-14. Water flow was measured throughout this year although access was occasionally limited as indicted previously. The maximum and minimum temperatures and monthly rainfall were obtained from weather stations at Mulloon Farm (courtesy of Tony Bernardi).

Macroinvertebrate analysis

Invertebrates were returned to the laboratory and ethanol added to preserve specimens until separation from the mixed substrate-specimen material could be made. Collections included large quantities of vegetation, as more vegetation was present in all sites. Invertebrates were removed under microscope and re-stored in individual vials containing 80% ethanol. The three samples from each site were individually used for counting and identification of macroinvertebrates as the numbers of insects were relatively high compared with previous years. At the end, the numbers for each individual sampling site were added together to summarise the collection at each site. Insects were classified to family, while other invertebrates were only described to order. Identifications were made using keys (Cairns *et al*. 2017, Gooderham and Tsyrlin 2002, Hawking 1986, Hawking and Smith 1997, http://keys.lucidcentral.org/keys/lwrrdc/public/Aquatics/main.htm)

Statistical analysis

Statistical analyses of macroinvertebrate and physical parameters have several alternative techniques to determine what is occurring within a stream (lotic) environment (Norris and Georges 2003). We used two techniques: principal component analysis and SIGNAL scores as done previously .

Principal component analysis on the physicochemical data (no data transformation) was used to determine how similar the various sites were based upon those parameters. Principal component analysis was used to determine how similar the various sites were with respect to composition and quantity of invertebrates. This analysis was done on the data using a natural log transformation (ln(x+1)), where x is the number of individuals collected in a family. Comparing the grouping of the sites was used to determine whether similarities existed between the physicochemical and biological information.

Signal scores were determined according to the procedures outlined by Chessman (2003). The current Signal analysis was compared with the 2006-08, 2016-17 and 2019-20 data to see how sites along Mulloon Creek may have changed in the intervening years.

Statistical analysis was done using Microsoft Excel and JMP 17 (SAS Corp).

Results

Physicochemical measurements

Water temperature at the various sites increased from September through to January, and then progressively decreased into winter (Figure 2). Temperatures varied among the sites, but were within 2-3˚C for any month.

Dissolved oxygen values were relatively constant throughout the year, being above 95% at all sites, except at Sandhills Creek when water was only slightly flowing and the dissoved O_2 had decreased to around or below 90%.

Conductivity was nearly the same at all sites along Mulloon Creek (~0.1 mS cm^{-1}) but Sandhills Creek was always higher ranging from 0.3-0.8 mS cm^{-1}. (Figure 4). Water input throughout the year maintained the lower salinity in Mulloon Creek, but clearly some salts were being washed down into the site on Sandhills Creek.

Very little difference was observed in pH across both the sites and seasons, as nearly all measurements indicated that the sites were circumneutral (pH 5-6.5) (Figure 5). The lower pH values observed may be associated with organic acids released into the water with leaf falls.

Water flow was high during all months along Mulloon Creek (>0.1 m s^{-1}). Water flow was significantly lower in Sandhills Creek compared with any site on Mulloon Creek ($F_{[5,48]}=3.65$, $p=0.007$).

Rain fell in most months (Figure 7). The rain in September was extreme and the creeks levels rose, leading to problems for collection (Figure 8). Seasonal maximum and minimum temperatures were similar to long-term average values (Bureau of Meteorology data from Braidwood).

To determine what physicochemical parameters may affect each site, a principal component analysis was performed on the data in Figures 2-6, with the graphical result presented in Figure 9 and eigenvectors in Table 1. The eigenvectors indicate that conductivity is the most important variable defining the sites, although dissolved oxygen and water flow rate are nearly as important (70% of variation in first 2 eigenvalues). Figure 9 shows that Sandhills Creek is separate from the Mulloon Creek sites as result of its high conductivity.

Macroinvertebrates

Overall, 16167 macroinvertebrates were identified, falling into 46 different taxa (see Appendix 2 for complete list). The largest number of invertebrates was collected in July (Figure 9), although the pattern differed somewhat among the sites (Figure 10). The number of taxa tended to increase as number of individuals increased, but that was dependent upon site (Figure 10). Peter’s Pond was the source of the highest number of individuals collected (July) and Sandhills Creek in April had nearly the same number collected. Triple Ponds (January), Peter’s Pond (April) and Sandhills Creek (April) were the most diverse in number of taxa collected. October had the fewest number of invertebrates collected at most sites. No clear seasonal pattern is indicated except for that collection period in October.

Even though the sites differed in the numbers of individuals collected, some taxa were commonly collected in all sites and accounted for at least 10% of the collected individuals at least once (Table 2). The most common taxon was the Leptophlebiidae in most seasons for all locations except Sandhills, where they were only collected in abundance in October. Chrionominae were also collected in abundance from most sites in most seasons except from Peter's Pond. Ceinidae (amphipods) were not collected in Black Jackie or Triple Ponds but were more numerous in the other locations. Various other taxa were collected in high numbers, but may be only found in certain sites.

To see how the macroinvertebrate collections indicated the state of the various sites, two different methods for comparing the sites were undertaken. The first method was the calculation of SIGNAL scores, as that which was done previously in 2006-08, where sampling was done at the sites Below pump shed (compared with Black Jackie), William's Wallow (compared with Triple Pond), Peter's Pond and Palerang and subsequently in both 2015-16 and 2019-2020 at the same sites as here. The scores depend upon the quality of water grade of the macroinvertebrate family and the weighting based upon the number of individuals collected in that family (Chessman 2003). Comparison of the previous scores with the current analyses at the various sites is presented in Figure 11. The current scores are higher than the scores in 2019-20 during the drought, but comparable to those in 2015-16.

The principal component analysis (PCA) gives a slightly different picture of the sites (Figure 12) compared with the SIGNAL analysis. In the PCA, all sites have similar trajectories for their vectors, suggesting some similarity for all sites, but a clear difference is shown between the October collection and the collections in the other months. The weighting for the October collection is the much lower number of individuals collected compared with the other collection periods. The sites themselves are all clustered around the centre of the taxa distribution.

Comparison with figure 8 derived from the physicochemical characteristics of the sites are similar and not dependent upon physicochemical structure during this collection period.

Discussion

Differences were found among the study sites with respect to both physicochemical characteristics and biological macroinvertebrate assemblages using the principal component analyses. The patterns that were present between sites in the study of the previous two studies are not found during this collection period, as all sites on Mulloon Creek were similar and only Sandhills Creek were clearly different, but only as the flow of water decreased. Sandhills Creek was the most depauperate site previously, but when the water flow was high, the conductivity was reduced and the number and diversity of the collection was equal to that of the other Mulloon Creek sites. With the high and continuous water flow along Mulloon Creek, all sites were much more similar in both the physico-chemical patterns and also the taxa that were present than previously measured. The only exception was Sandhills Creek but that water originates from a different source and is clearly increasing in conductivity during the study period. Despite that change in conductivity, the differences in taxa were still slight even in the July collection. Presumably the increasing conductivity would lead to a change in taxa present in Sandhills Creek if further collections were taken later in the year. The slight change in conductivity in Reedy Creek resulted from the difference in water input from Sandhills Creek compared with the water travelling down Mulloon Creek.

The SIGNAL system gave a similar pattern of site structure with respect to water quality. In all collections, the SIGNAL was above three and many times above four. Using an AUSRIVAS analysis, good quality water would be anything near a SIGNAL score of 5 for this type of creek (Chessman 2003), suggesting that all sites were good and that little effect of high conductivity could be observed. Presumably, conductivity is not limiting the diversity, but other aspects of habitat, such as vegetation, may be influencing the taxa diversity. These anomalies demonstrate that some care must be taken when interpreting the results of the SIGNAL scores and that following the pattern of macroinvertebrates overtime may be the best method for understanding how the water quality varies (Chessman 2003).

Taxa appear to have an overwhelming similarity in all sites, as the first eigenvector accounts for nearly 77% of the variation (Figure 12, Table 2). Taxa such as the stoneflies (Plecoptera), caddisflies (Trichoptera) and Ptilodactylidae (Coleoptera) were indicators of a recovery of the waterway from the previous conditions during drought as none of those taxa were present during the drought (Cooper 2022). These three taxa were present collections at all sites, although they were never dominant. In contrast, Odonata were only slightly present in Peter’s Pond and lacking in numbers in most other collections. The high rate of flow in most other locations, such as Reedy Creek, removed the reeds that normally dominate in the waterway and are habitats for odonate larvae. If the habitat is modified to limit the present of perching sites, then odonates will be reduced in number. The water flow does bring in fresh and well-oxygenated water to the various sites that I measured, but the change of vegetation limited the number of odonates that were present. Peter’s Pond still had the vegetation present as the larger surface area of the pond reduced flow in the centre area, and that is where the odonates were most abundant in this year.

The presence of allochthonous wood and leaves can be used by a variety of macroinvertebrate functional groups, especially shredders and grazers, to enhance the biodiversity within streams (McKie and Cranston 2001). The leaves and wood result from vegetation addition into that stream section as a result of the surrounding and overhanging plants that will periodically add to the local carbon supply in both locations, although not all taxa appear to benefit. This allochthonous material would be a source of food and may explain the large numbers of Trichoptera (caddisflies) that were found in this year as most caddisflies are shredders and consume vegetation. Flowing water is important for most Trichoptera, but the addition of vegetation from both outside the stream and from displaced water vegetation, food may be extremely abundant (Gooderham and Tsyrlin 2002). The population of both the Leptoceridae and Calocidae/Helicophidae complex would benefit from the extra resources and high oxygen levels in the water. The high numbers of Ephemeroptera (Baetidae and Leptophlebiidae) probably also benefited from the same conditions as they also consume vegetation and use vegetation in the water as habitat (Gooderham and Tsyrlin 2002)

The overall result suggests that little change has occurred over the 25 years in macroinvertebrate diversity, but drought (20006-08 and 2019-20) is associated with a reduction in taxa, especially those dependent upon flowing water such as Trichoptera. As restoration continues, ways of including more overhanging natural vegetation should be considered in other sites to increase addition of food and habitat as the vegetation from plants falls into the water. Regions of gravel crossings (Triple Ponds and Palerang) may present difficult areas for restoration, although ponds built above Palerang Crossing were good habitat after the excavations done in 2019, but the hardest location for restoration will be Sandhills Creek as its flow is much more periodic (usually only during flood periods) compared with the rest of the Mulloon-Reedy system, a result that was even apparent when water flow was relatively high along Mulloon-Reedy Creek.

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Table 1. Eigenvectors derived from the principal component analysis of the physicochemical parameters that were measured during the course of this work. The large values of the first eigenvectors indicate that dissolved oxygen and conductivity were important for separating the sites, especially Sandy Hills and Reedy Creek, but pH was also an important variable. Water flow is important. The first two vectors account for 66.3% of the variation, and vector 3 accounts for an additional 18% of the variation. Although temperature did not appear to account for much of the variation in the univariate plots, it appears to play a role in characterising the sites as shown by the large values in the second and third eigenvectors.

	Eigenvector 1	Eigenvector 2	Eigenvector 3
Conductivity (mS/cm)	0.5617	0.1474	-0.2718
Temp (˚C)	0.2477	-0.7336	0.5479
Dissolved O_2 (%)	-0.5730	-0.2225	0.1200
pH	0.3215	0.4707	0.7101
H_2O flow (m/s)	-0.4375	0.4112	0.3262

Table 2. Taxa that made up more than 10% of the individuals collected at any one collection time for the various sites.

Site	Month	Taxa	Percent of total
Black Jackie	October, January, April, July	Chironominae	14.6, 18.9, 34.9, 46.5
	October, January, April, July	Leptophlebiidae	25.0, 11.4, 26.0, 10.9
	October	Oligochaeta	20.8
	April, July	Gripopterygidae	10.6, 30.3
	January	Ptilodacylidae	13.1
	January	Ecnomidae	19.4
Triple Ponds	April	Copepoda	11.6
	October, January, April, July	Chironominae	16.0, 21.7, 20.0, 37.7
	April, July	Leptophlebiidae	23.8, 16.5
	October	Oligochaete	13.3
	January	Economidae	26.2
	October	Leptoceridae	31.3
Peter's Pond	January, April, July	Ceinidae	35.0, 38.7, 31.3
	October	Ostracoda	26.7
	October	Simulidae	20.4
	October	Leptophlebiidae	27.3
	January, April, July	Leptoceridae	21.4, 25.2, 27.2
Palerang	January, April, July	Ceinidae	11.3, 21.7, 20.8
	January, April, July	Chironominae	15.9,13.8, 19.4
	October	Simulidae	64.1
	January,April, July,	Leptophlebiidae	31.0, 19.8,26.8
	April	Corixidae	18.5
Sandhills Creek	October	Ceinidae	13.6
	July	Copepoda	17.6
	July	Ostracoda	22.5
	October, January, April, July	Chironominae	26.1,50.0,49.7, 23.0
	October	Leptophlebiidae	19.6
	October	Oligochaeta	15.0
Reedy Creek	January, April	Ceinidae	22.1, 14.9
	October	Dytiscidae	41.7
	October, July	Copepoda	13.9, 14.2
	January, April, July	Chironominae	15.2, 14.9, 23.8
	October	Simulidae	16.7
	October	Gripopterygidae	13.9
	January	Baetidae	29.2
	January, April, July	Leptophlebiidae	14.9, 14.4, 20.6

Table 3. Eigenvalues and cumulative variation explained from the principal component analysis on the individual taxa relative to site. As shown by the cumulative percent column, one eigenvalue accounts for 77% of the variation indicating that the taxa are responding similarly in the various sites.

Number of eigenvalues	Eigenvalue	Cumulative Percent
1	3.085	77.12
2	0.522	90.17
3	0.230	95.93

Figure 1. Sites of sampling for physical attributes and macroinvertebrates along Mulloon Creek (courtesy of Luke Peel).

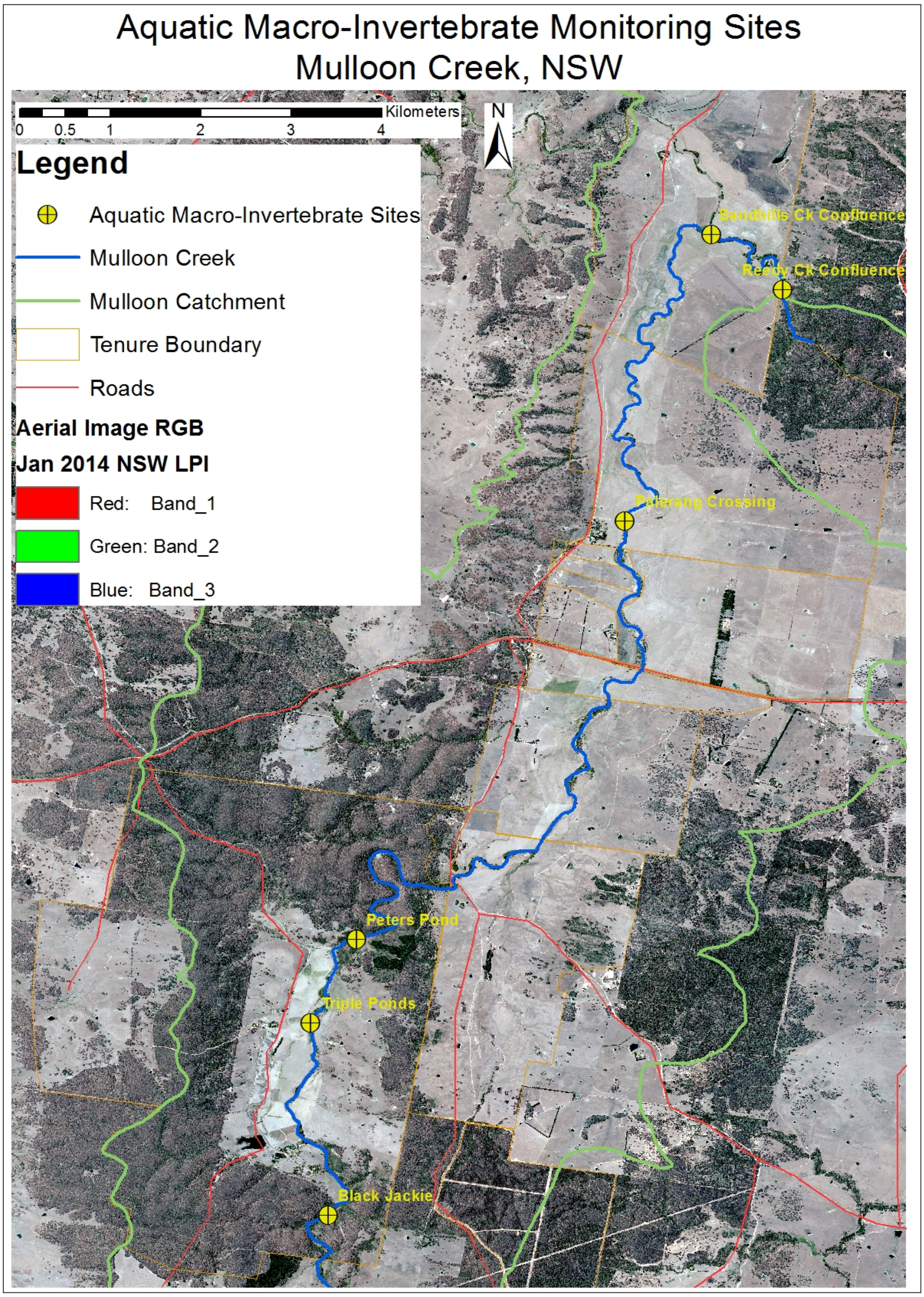


Figure 2. Water temperature at six sites along Mulloon Creek from Oct 2019-May 2020. Temperature is near 20 ˚C from December to February. Temperatures were only between 5-10˚C May to September then started increasing above 10˚C in October and November.

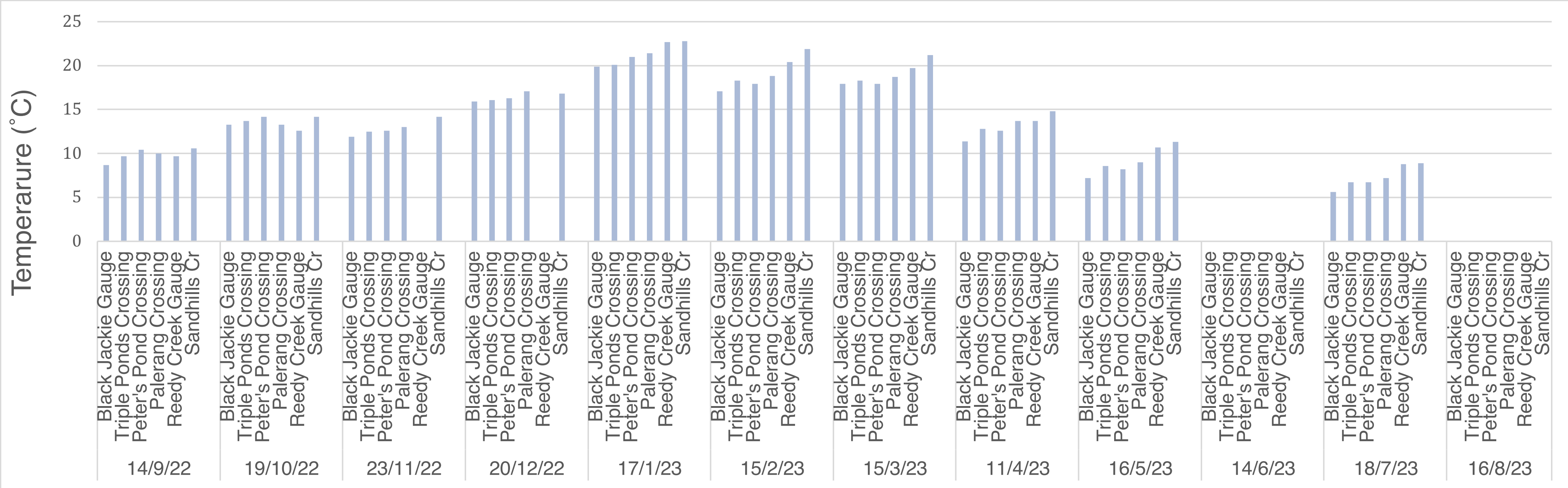


Figure 3. Dissolved oxygen (%saturation at measured temperature) for the six sites along Mulloon Creek from April 2019 –March 2020. Oxygen decreased as temperatures increased from April to January, but increased again after the rain. Dissolved oxygen is usually lowest in Sandhills, however oxygen in most ponds would have been maintained by both wind and vegetation within the ponds producing oxygen during the day.

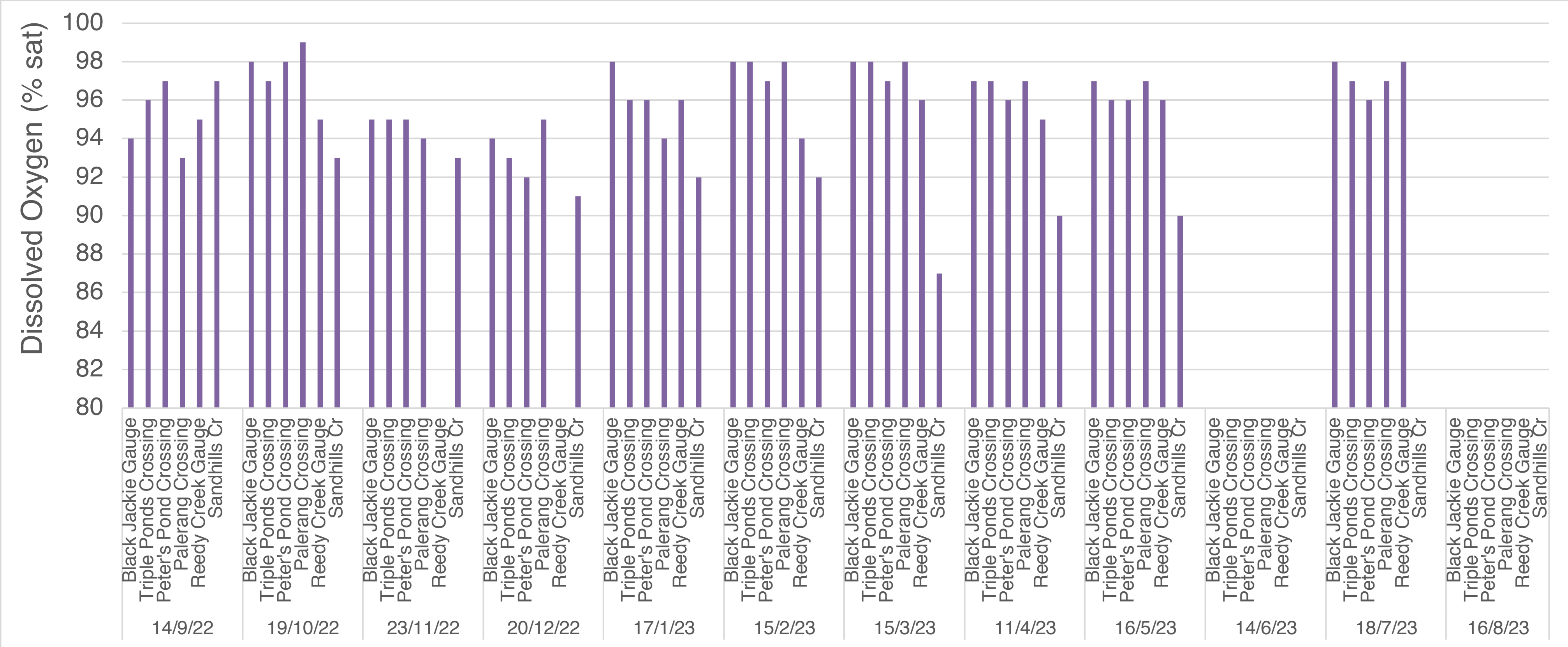


Figure 4. Changes in conductivity at 6 sites along Mulloon Creek from Apr 2019-March 2020. Reedy Creek and Sandhills are much higher in conductivity than the other sites, until rain occurred in February and March.

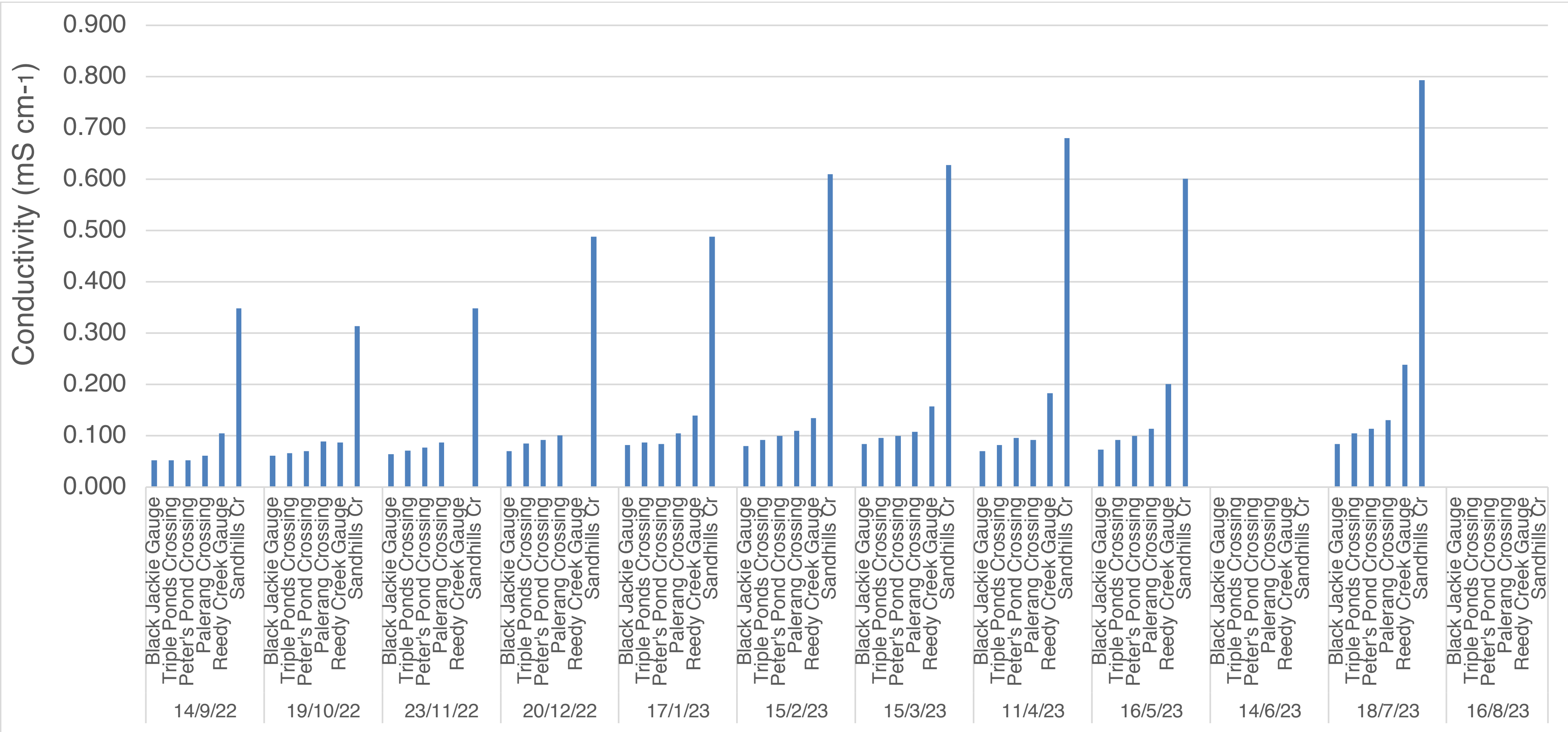


Figure 5. pH was circumneutral in all sites ranging from 5-7 throughout the sampling period.

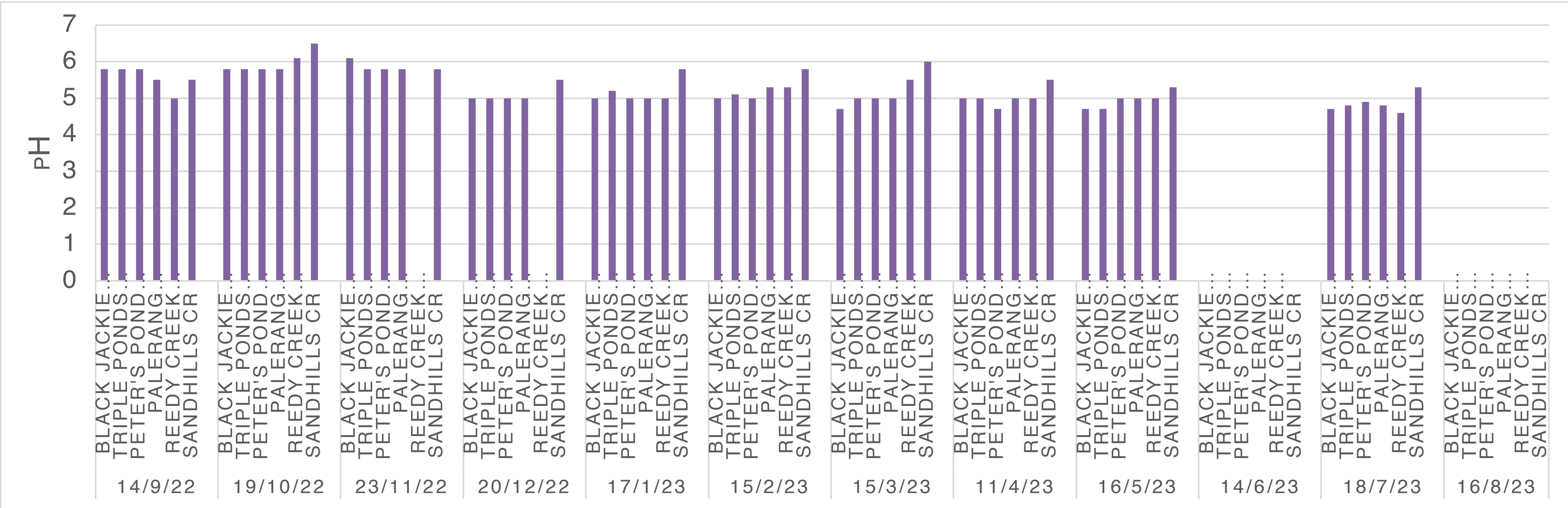


Figure 6. Rate of water flow (ms^{-1}) measured at the various sites during the study period. No flow occurred from April-January during the study.

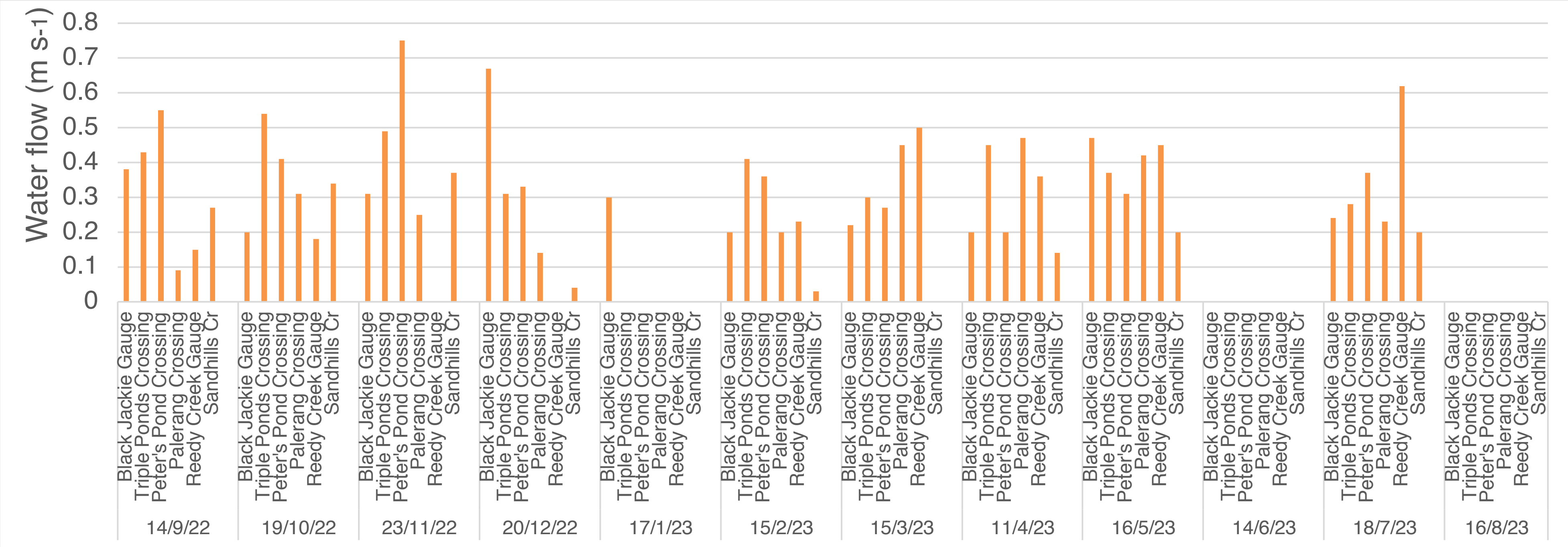


Figure 7. Maximum and minimum temperatures and rainfall taken from Mulloon Creek weather information courtesy of Tony Bernardi.

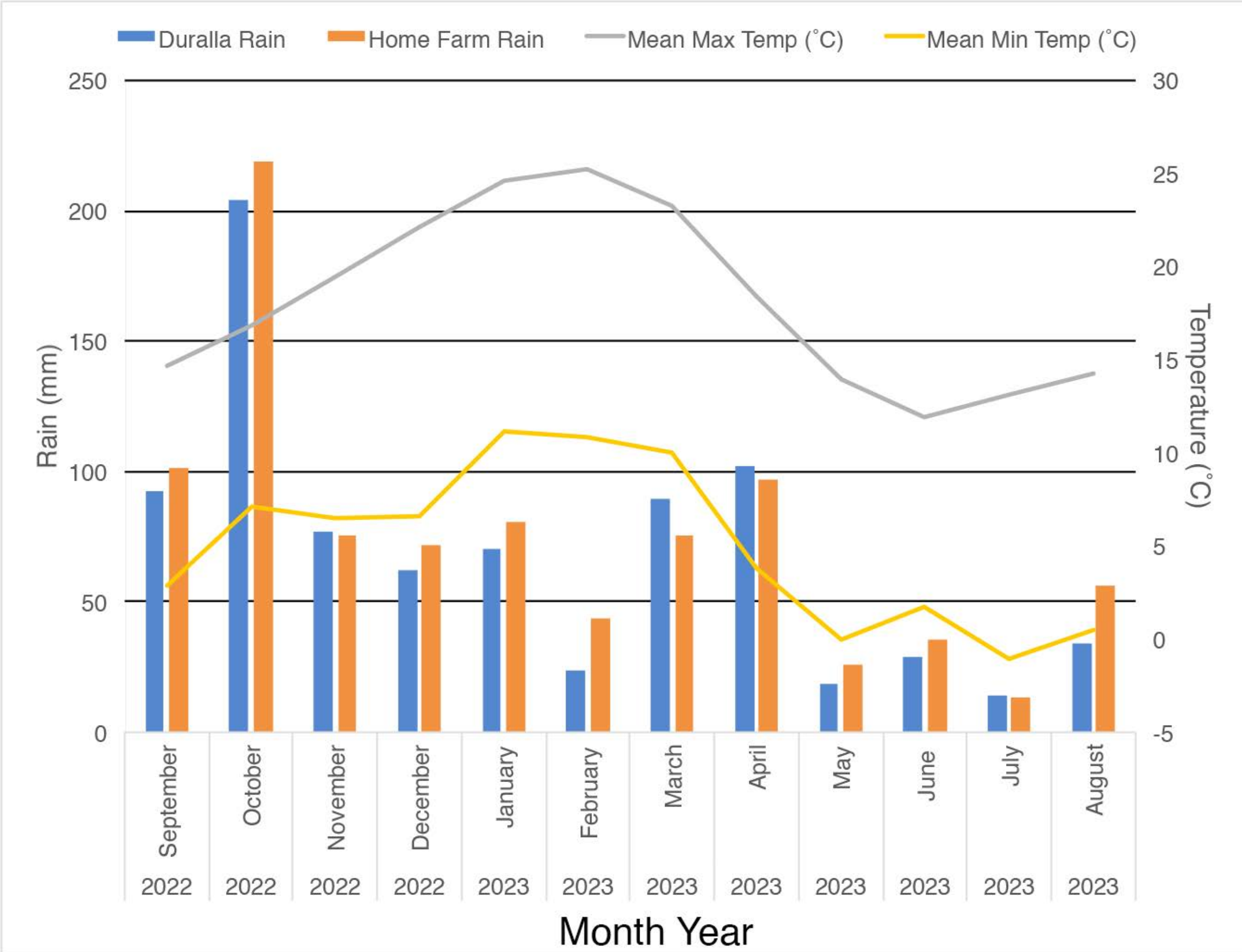


Figure 8. Picture of all the vehicles that had to come to my rescue when I was bogged in the paddock near Reedy Creek Gauge as a result of the rain during that period of September 2022.

Figure 9. Principal component analysis of the physicochemical measurements shows how the individual parameters determine differences among the sites. Most sites are clustered near the middle of the distribution. The eigenvectors indicate that conductivity is the major difference and results in the isolation of Sandhills Creek compared with other sites, but an important secondary effect is the rate of water flow. (BJ=Black Jackie Gauge, TP= Triple Ponds Crossing, PP= Peter's Pond Crossing, PC= Palerang Crossing, SHC= Sandhills Creek Gauge, RC= Reedy Creek Gauge)

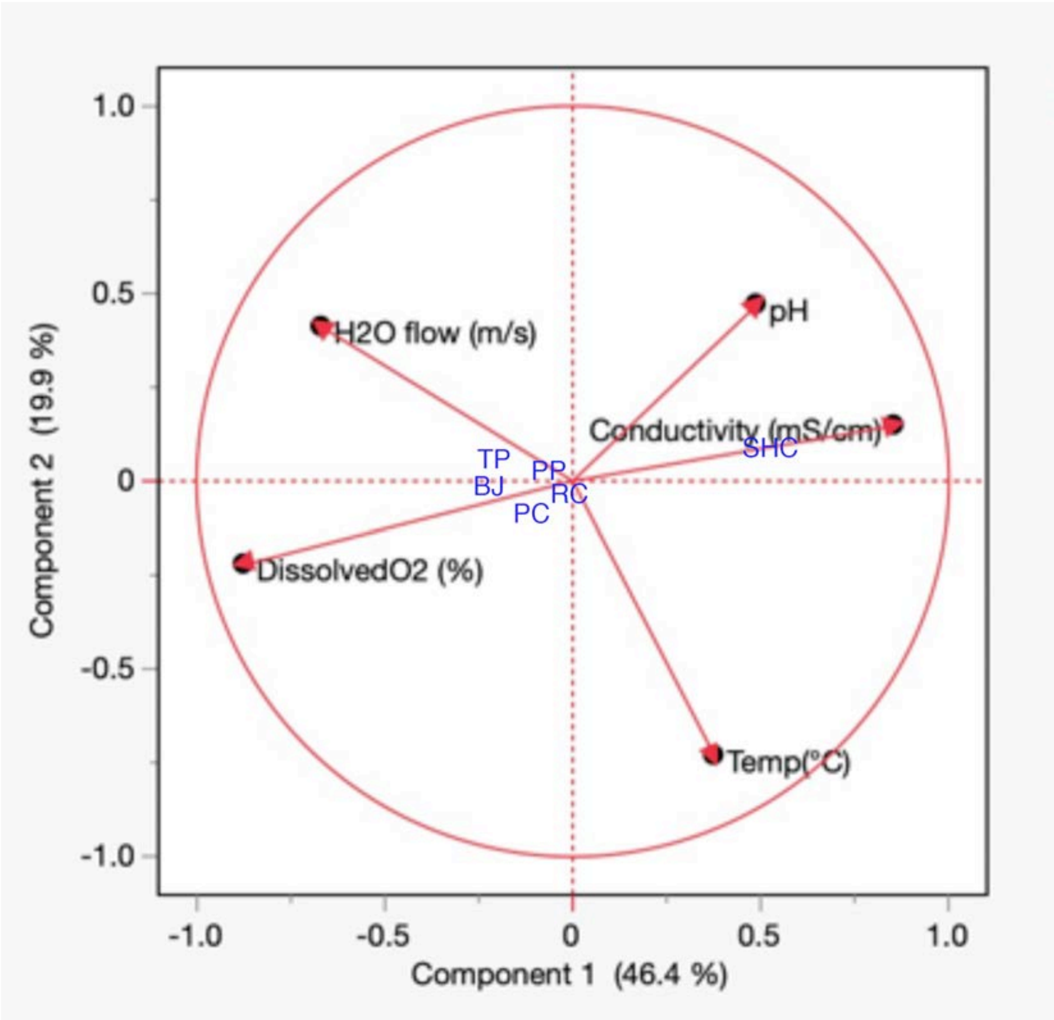


Figure 9. Distribution of total number and taxa of macroinvertebrates collected during the study. January had both the greatest number of invertebrates collected and the most number of taxa represented.

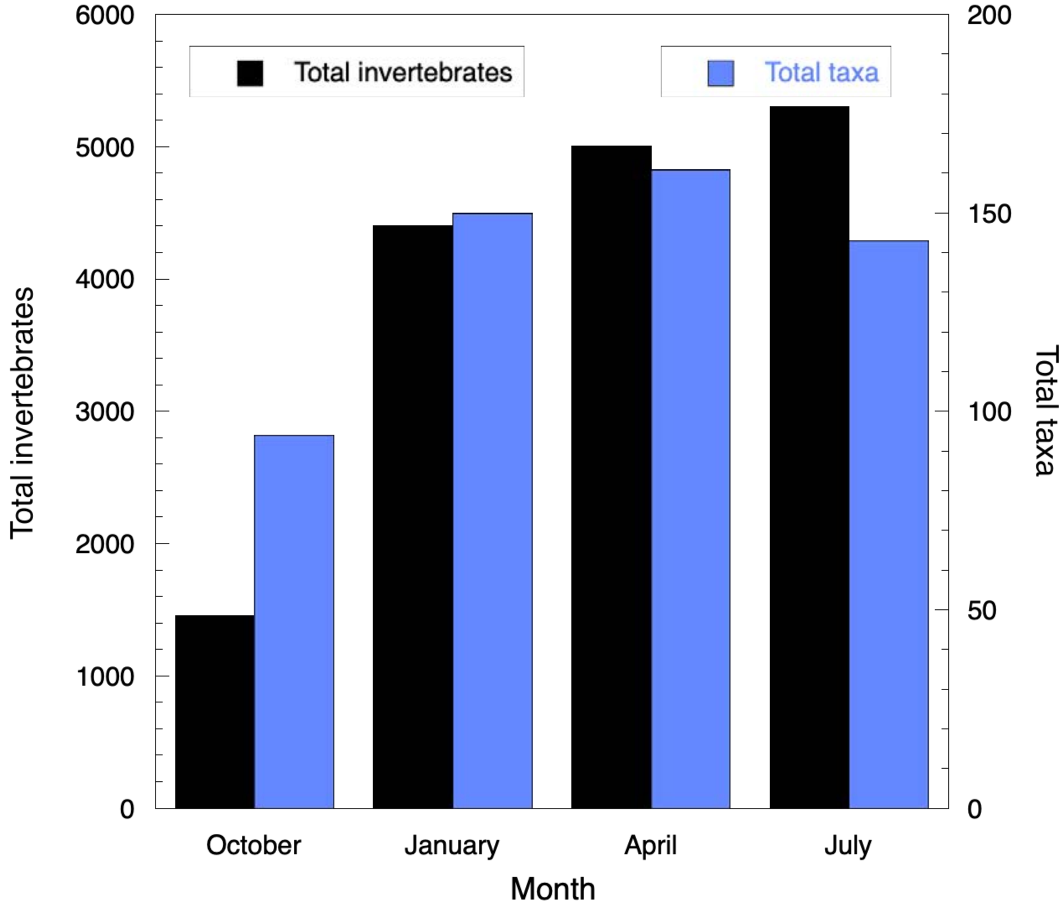


Figure 10. Monthly distribution of numbers of invertebrates and taxa collected at each site. Peter's Pond yielded the greatest number of individuals of all sites in April with Sandhills Creek having the next most individuals. Peter's Pond had the highest number of taxa at 33 in April, with greater than 30 collected in Triple Ponds in January and Sandhills in April. BJG= Black Jackie Gauge, TPC= Triple Ponds Crossing, PP= Peter's Pond, PC= Palerang Crossing, RCG=Reedy Creek Gauge, SHG=Sandhills Creek Gauge.

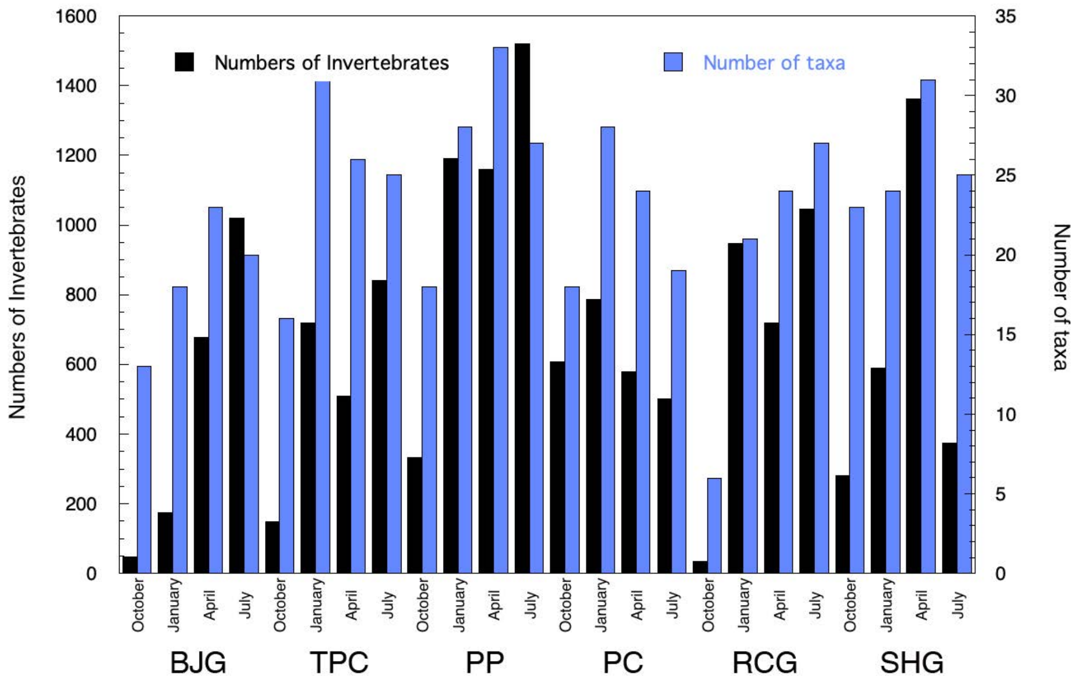


Figure 11. SIGNAL scores for 2006-08,2015-16, 2019-20 and 2022-23. Scores for 2015-16 are higher than the drought conditions in 2019-20, but 2022-23 is more similar to d 2015-16. BJG= Black Jackie Gauge, TPC= Triple Ponds Crossing, PPC= Peter's Pond, PC= Palerang Crossing, RCG=Reedy Creek Gauge, SHC=Sandhills Creek Gauge.

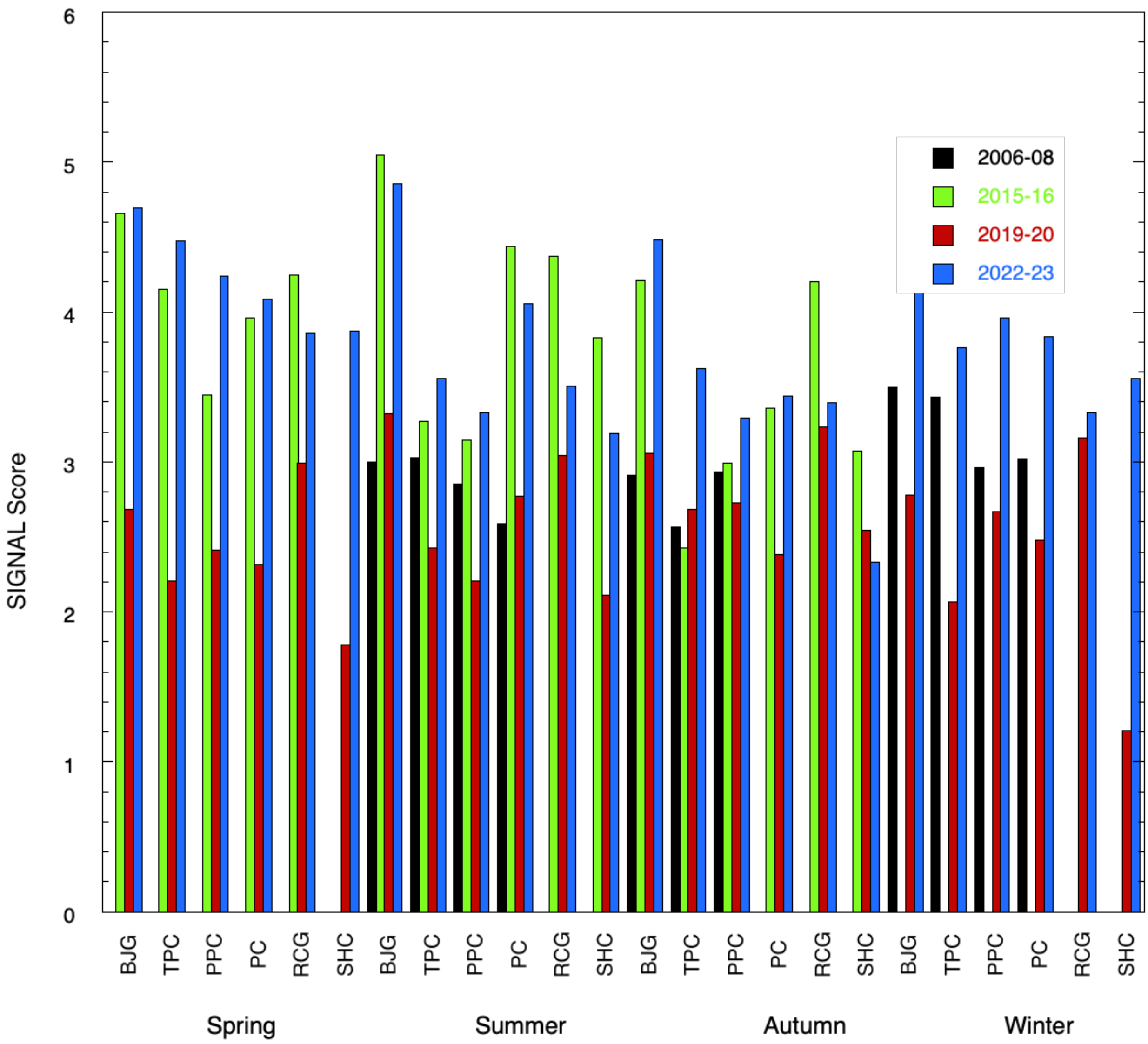
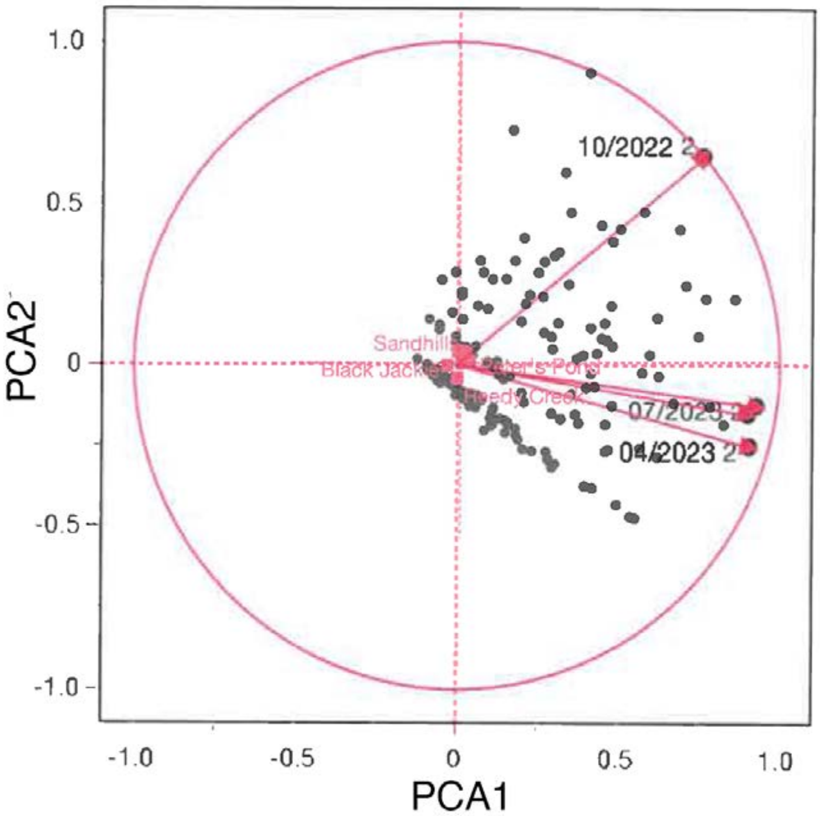


Figure 12. Site comparison across the seasons based upon the macroinvertebrate collection. All sites were similar and grouped together near the centre of the distribution. The big difference in the principal component analysis for this series of collections was the October 2022 collection, as the biggest rainfall had occurred during that month and the water movement and depth may have limited the number and diversity of macroinvertebrates collected.



Appendix 1.
Description of sites

Black Jackie- Cool, shady location. Most of the collection time fast flowing cool water was present (Figure 14a and b)

Triple Pond – Deep fast flowing substrate below crossing. Pebbles were present at crossing, but rare elsewhere. Deep pools above crossing were sampled during the study period (Figure 15).

Peter's Pond – Three collection locations 1) above the pond near rain gauge below rocks, 2) in lily pads and reeds near spillway, 3) at exit of water near willows (Figure 16)

Palerang – Deeper water at crossing and deep pools above crossing. (Figure 17a and b)

Sandhills Creek – Flowing water nearly throughout the sampling period with vegetation from overhanging trees. Gauge not working most of year as the flow caused the gauge to be dislodged (Figure 18).

Reedy Creek – Downstream from gauge where rocks in water present. No reeds downstream of rocks as, reeds had been dislodged by flow before sampling had begun (Figure 19).

Figure 14a and b. Black Jackie sampling site in October (a) 2022 looking upstream and March (b) 2023 looking upstream that was sampled.

a)

b)

Figure 15. Triple Pond crossing, showing water movement of fluid in September 2022.

Figure 16. Peter's Pond just below collecting site shown.

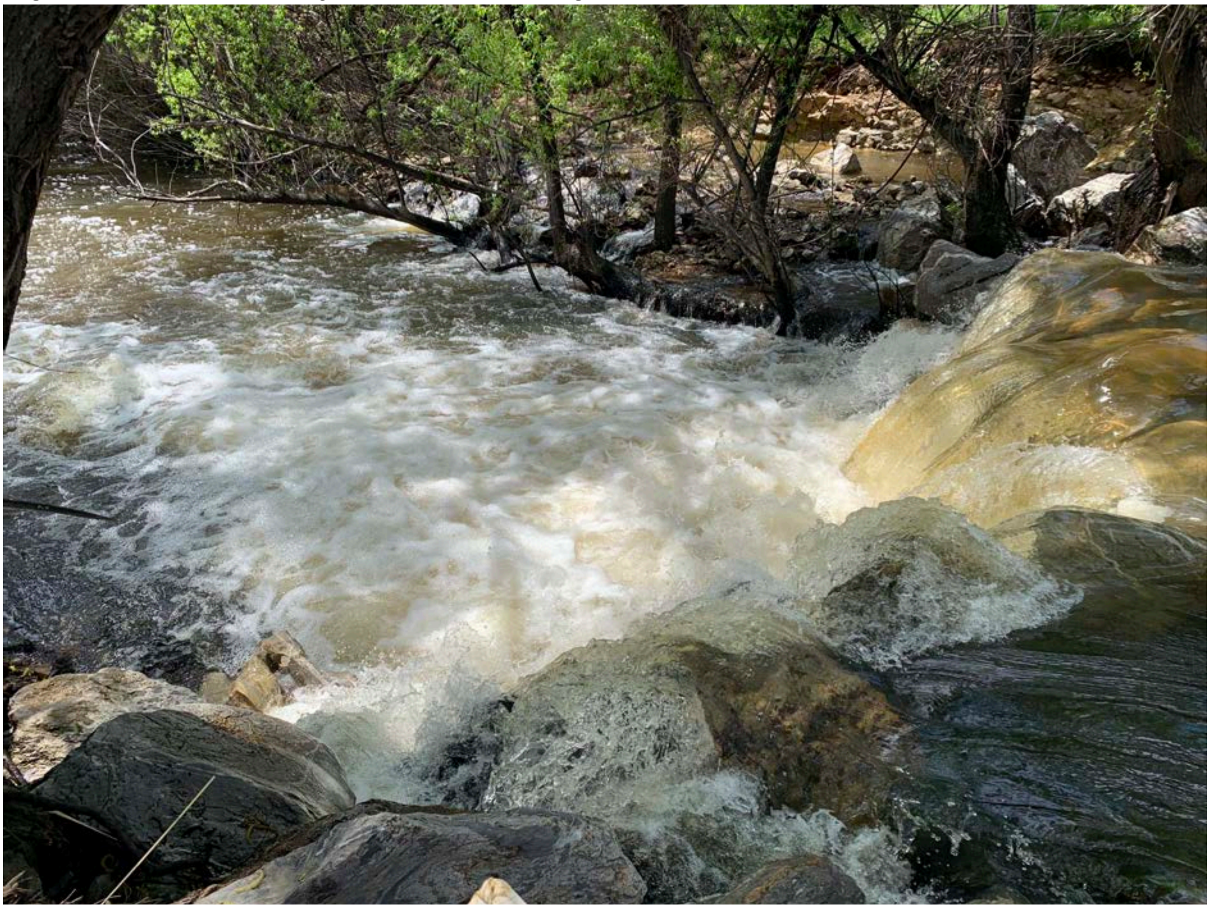


Figure 17. Palerang crossing before (a) downstram and (b) upstream, just above crossing in September 2022..

a)

b)

Figure 18. Sandhills Creek gauge showing pool that extended to Mulloon Creek (September 2022)

Figure 19. Reedy Creek looking downstream below rocks in March 2023.

Appendix 2. Identified aquatic invertebrates collected during the study for each site for each month collection was undertaken. Predators: Hemiptera, Dytiscidae (Coleoptera), Tanypodinae (Diptera), most Odonata, Ecnomidae (Trichoptera)

Black Jackie	Acarina	Acarina	0	1	0	1
Black Jackie	Arachnid	Araneae	0	0	2	0
Black Jackie	Coleoptera	Ptilodactylidae	0	23	49	8
Black Jackie	Megaloptera	Corydalidae	0	0	4	0
Black Jackie	Collembola	sp.	0	0	1	0
Black Jackie	Crustacea	Copepoda	0	3	6	30
Black Jackie	Crustacea	Ostracoda	1	1	3	6
Black Jackie	Diptera	Ceratopogonidae	1	1	2	0
Black Jackie	Diptera	s-f Chironominae	7	33	236	475
Black Jackie	Diptera	s-f Orthocladiinae	0	0	0	1
Black Jackie	Diptera	s-f Tanypodinae	1	6	30	20
Black Jackie	Diptera	Simulidae	0	1	22	3
Black Jackie	Diptera	Tipulidae	3	1	0	1
Black Jackie	Plecoptera	Gripopterygidae	3	0	72	309
Black Jackie	Plecoptera	Nournotmidae	0	0	0	5
Black Jackie	Ephemeroptera	Baetidae	4	35	23	24
Black Jackie	Ephemeroptera	Caenidae	1	2	9	3
Black Jackie	Ephemeroptera	Leptophlebiidae	12	20	176	111
Black Jackie	Hemiptera	Veliidae	0	0	2	1
Black Jackie	Nematomorpha		0	0	2	0
Black Jackie	Odonata	Aeshnidae	0	1	0	0
Black Jackie	Odonata	Gomphidae	0	0	4	0
Black Jackie	Oligochaeta	Oligochaeta	10	2	2	4
Black Jackie	Trichoptera	Ecnomidae	1	34	22	9
Black Jackie	Trichoptera	Hydroptilidae	0	6	3	5
Black Jackie	Trichoptera	Leptoceridae	4	5	4	5
Black Jackie	Porifera		0	0	3	0
Triple Ponds	Acarina	Acarina	0	1	0	0
Triple Ponds	Arachnid	Araneae	0	3	1	0
Triple Ponds	Amphipoda	Ceinidae	0	4	4	19
Triple Ponds	Bivalvia	Sphaeriidae	1	1	3	8
Triple Ponds	Coleoptera	Dytiscidae	0	13	4	6
Triple Ponds	Coleoptera	Gyrinidae	1	0	0	0
Triple Ponds	Coleoptera	Hydrophilidae	0	1	0	0
Triple Ponds	Coleoptera	Ptilodactylidae	0	0	1	1
Triple Ponds	Megaloptera	Corydalidae	0	0	3	0
Triple Ponds	Collembola	sp.	0	6	0	0
Triple Ponds	Crustacea	Cladocera	0	2	46	19

Triple Ponds	Crustacea	Copepoda	0	53	59	30
Triple Ponds	Crustacea	Ostracoda	11	28	1	26
Triple Ponds	Decapoda	Atyidae	0	5	1	1
Triple Ponds	Diptera	Ceratopogonidae	0	4	0	0
Triple Ponds	Diptera	Dolichopodidae	0	2	0	0
Triple Ponds	Diptera	s-f Chironominae	24	156	102	317
Triple Ponds	Diptera	s-f Orthocladiinae	0	0	6	6
Triple Ponds	Diptera	s-f Tanypodinae	8	33	25	52
Triple Ponds	Diptera	Simulidae	5	12	17	2
Triple Ponds	Diptera	Tipulidae	1	1	0	0
Triple Ponds	Plecoptera	Gripopterygidae	7	2	2	50
Triple Ponds	Ephemeroptera	Baetidae	3	42	49	56
Triple Ponds	Ephemeroptera	Caenidae	1	3	0	5
Triple Ponds	Ephemeroptera	Leptophlebiidae	9	59	121	139
Triple Ponds	Gastropoda	Planorbidae	0	1	0	4
Triple Ponds	Hemiptera	Corixidae	0	6	1	12
Triple Ponds	Hemiptera	Notonectidae	1	0	0	0
Triple Ponds	Hemiptera	Veliidae	0	2	0	0
Triple Ponds	Hirudinea	Glossiphoniidae	0	2	0	1
Triple Ponds	Hydra		0	0	5	9
Triple Ponds	Odonata	Lestidae	0	0	1	0
Triple Ponds	Oligochaeta	Oligochaeta	20	3	11	8
Triple Ponds	Trichoptera	Calamoceratidae	0	0	0	1
Triple Ponds	Trichoptera	Calosidae	10	10	1	9
Triple Ponds	Trichoptera	Ecnomidae	1	188	33	32
Triple Ponds	Trichoptera	Hydropsychidae	0	1	1	0
Triple Ponds	Trichoptera	Hydroptilidae	0	20	6	0
Triple Ponds	Trichoptera	Leptoceridae	47	55	5	29
Peter's Pond	Araneae		0	1	1	0
Peter's Pond	Amphipoda	Ceinidae	9	417	449	476
Peter's Pond	Bivalvia	Sphaeriidae	0	0	4	2
Peter's Pond	Coleoptera	Psephenidae	1	0	1	0
Peter's Pond	Coleoptera	Dytiscidae	1	0	3	1
Peter's Pond	Collembola	sp.	0	1	0	0
Peter's Pond	Crustacea	Cladocera	1	16	38	0
Peter's Pond	Crustacea	Copepoda	2	29	77	47
Peter's Pond	Crustacea	Ostracoda	89	8	46	61
Peter's Pond	Decapoda	Atyidae	0	2	1	1
Peter's Pond	Diptera	s-f Chironominae	20	97	37	98
Peter's Pond	Diptera	s-f Orthocladiinae	0	0	0	0
Peter's Pond	Diptera	s-f Tanypodinae	1	27	11	82
Peter's Pond	Diptera	Simulidae	68	1	0	1

Peter's Pond	Diptera	Stratiomyidae	0	2	5	1
Peter's Pond	Diptera	Tipulidae	0	0	2	1
Peter's Pond	Plecoptera	Gripopterygidae	14	16	0	25
Peter's Pond	Plecoptera	Notonourmiridae	0	0	0	0
Peter's Pond	Ephemeroptera	Baetidae	11	86	21	9
Peter's Pond	Ephemeroptera	Caenidae	0	1	1	0
Peter's Pond	Ephemeroptera	Leptophlebiidae	91	70	29	132
Peter's Pond	Gastropoda	Planorbidae	3	87	11	33
Peter's Pond	Hemiptera	Belostomadidae	0	0	1	0
Peter's Pond	Hemiptera	Notonectidae	0	3	8	0
Peter's Pond	Hemiptera	Veliidae	0	6	1	0
Peter's Pond	Hirudinea	Glossiphoniidae	0	3	2	3
Peter's Pond	Hydra		0	3	4	19
Peter's Pond	Odonata	Aeshnidae	0	0	7	2
Peter's Pond	Odonata	Gomphidae	0	2	1	13
Peter's Pond	Odonata	Coenagrionidae	0	39	52	4
Peter's Pond	Odonata	Lestidae	0	1	4	1
Peter's Pond	Oligochaeta	Oligochaeta	1	3	4	12
Peter's Pond	Trichoptera	Calamoceratidae	0	0	3	6
Peter's Pond	Trichoptera	Calosidae	2	6	22	68
Peter's Pond	Trichoptera	Ecnomidae	8	0	20	7
Peter's Pond	Trichoptera	Hydropsylidae	0	0	1	0
Peter's Pond	Trichoptera	Hydroptilidae	1	5	0	0
Peter's Pond	Trichoptera	Leptoceridae	10	255	292	414
Peter's Pond	Turbellaria	Dugesiidae	0	3	1	1
Palerang	Arachnid	Acarina				
Palerang	Arachnid	Araneae	0	1	1	0
Palerang	Amphipoda	Ceinidae	2	89	126	104
Palerang	Coleoptera	Dytiscidae	0	0	3	0
Palerang	Coleoptera	Elmidae	0	1	0	0
Palerang	Coleoptera	Hydrophilidae	0	1	0	0
Palerang	Coleoptera	Ptilodactylidae	1	10	3	1
Palerang	Crustacea	Cladocera	3	2	1	1
Palerang	Crustacea	Copepoda	4	9	3	35
Palerang	Crustacea	Ostracoda	15	3	10	4
Palerang	Decapoda	Atyidae	0	1	0	0
Palerang	Diptera	Ceratopogonidae	0	2	1	0
Palerang	Diptera	Dolichopodidae	0	1	0	0
Palerang	Diptera	s-f Chironominae	46	125	80	97
Palerang	Diptera	s-f Orthocladiinae	1	5	3	2
Palerang	Diptera	s-f Tanypodinae	8	71	14	28
Palerang	Diptera	Simulidae	390	1	9	4

Palerang	Diptera	Tipulidae	0	1	1	1
Palerang	Plecoptera	Gripopterygidae	55	4	0	37
Palerang	Ephemeroptera	Baetidae	4	70	7	13
Palerang	Ephemeroptera	Caenidae	0	3	6	1
Palerang	Ephemeroptera	Leptophlebiidae	47	244	115	134
Palerang	Gastropoda	Planorbidae	2	1	2	0
Palerang	Hemiptera	Corixidae	0	0	107	0
Palerang	Hemiptera	Veliidae	3	0	0	0
Palerang	Hydra		0	0	0	3
Palerang	Nematomorpha		0	2	0	0
Palerang	Odonata	Aeshnidae	0	0	1	0
Palerang	Odonata	Gomphidae	0	0	2	0
Palerang	Odonata	Coenagrionidae	0	1	0	0
Palerang	Oligochaeta	Oligochaeta	16	20	40	23
Palerang	Trichoptera	Calosidae	1	5	1	0
Palerang	Trichoptera	Ecnomidae	9	67	41	6
Palerang	Trichoptera	Hydroptilidae	1	32	0	2
Palerang	Trichoptera	Leptoceridae	0	14	3	4
Reedy Creek	Amphipoda	Ceinidae	0	209	107	101
Reedy Creek	Bivalvia	Sphaeriidae	0	0	34	12
Reedy Creek	Coleoptera	Dytiscidae	15	48	0	0
Reedy Creek	Collembola	sp.	0	0	0	1
Reedy Creek	Crustacea	Cladocera	0	0	89	16
Reedy Creek	Crustacea	Copepoda	5	1	36	148
Reedy Creek	Crustacea	Ostracoda	0	0	3	5
Reedy Creek	Diptera	Ceratopogonidae	0	0	1	0
Reedy Creek	Diptera	s-f Chironominae	2	144	107	249
Reedy Creek	Diptera	s-f Orthocladiinae	0	4	2	2
Reedy Creek	Diptera	s-f Tanypodinae	0	43	47	80
Reedy Creek	Diptera	Simulidae	6	0	2	2
Reedy Creek	Plecoptera	Gripopterygidae	5	0	0	19
Reedy Creek	Plecoptera	Notonemuridae	3	0	1	11
Reedy Creek	Ephemeroptera	Baetidae	0	276	52	59
Reedy Creek	Ephemeroptera	Caenidae	0	9	6	2
Reedy Creek	Ephemeroptera	Leptophlebiidae	0	141	103	215
Reedy Creek	Gastropoda	Ancylidae	0	0	0	1
Reedy Creek	Gastropoda	Planorbidae	0	0	4	4
Reedy Creek	Hemiptera	Corixidae	0	14	12	35
Reedy Creek	Hirudinea	Glossiphoniidae	0	1	0	0
Reedy Creek	Hydra		0	1	1	8
Reedy Creek	Lepidoptera	Pyralidae	0	1	1	0
Reedy Creek	Odonata	Aeshnidae	0	0	0	1

Reedy Creek	Odonata	Coenagrionidae	0	3	1	0
Reedy Creek	Odonata	Lestidae	0	4	1	0
Reedy Creek	Oligochaeta	Oligochaeta	0	1	29	17
Reedy Creek	Trichoptera	Calamoceratidae	0	1	0	0
Reedy Creek	Trichoptera	Calosidae	0	5	14	9
Reedy Creek	Trichoptera	Ecnomidae	0	18	30	13
Reedy Creek	Trichoptera	Hydropsychidae	0	0	0	1
Reedy Creek	Trichoptera	Hydroptilidae	0	11	0	4
Reedy Creek	Trichoptera	Leptoceridae	0	11	35	30
Reedy Creek	Trichoptera	Odontoceridae	0	0	0	1
Sandhills	Acarina	Acarina	1	0	0	1
Sandhills	Amphipoda	Ceinidae	38	26	3	3
Sandhills	Bivalvia	Sphaeriidae	0	2	0	1
Sandhills	Coleoptera	Dytiscidae	9	28	26	11
Sandhills	Coleoptera	Scirtidae	0	0	0	1
Sandhills	Collembola	sp.	0	1	6	3
Sandhills	Crustacea	Cladocera	2	46	149	1
Sandhills	Crustacea	Copepoda	8	55	98	66
Sandhills	Crustacea	Ostracoda	1	26	80	84
Sandhills	Diptera	s-f Chironominae	73	294	676	86
Sandhills	Diptera	s-f Orthocladiinae	0	0	2	0
Sandhills	Diptera	s-f Tanypodinae	6	28	22	32
Sandhills	Diptera	Psychodidae	0	0	0	0
Sandhills	Diptera	Stratiomyidae	0	0	3	0
Sandhills	Diptera	Tipulidae	1	0	0	0
Sandhills	Plecoptera	Gripopterygidae	4	0	0	1
Sandhills	Plecoptera	Notonourmiridae	0	0	1	0
Sandhills	Ephemeroptera	Baetidae	1	1	3	0
Sandhills	Ephemeroptera	Caenidae	6	8	5	2
Sandhills	Ephemeroptera	Leptophlebiidae	55	2	105	7
Sandhills	Gastropoda	Planorbidae	1	4	18	5
Sandhills	Hemiptera	Corixidae	4	10	17	7
Sandhills	Hemiptera	Notonectidae	1	4	10	2
Sandhills	Hemiptera	Veliidae	0	6	22	0
Sandhills	Hirudinea	Glossiphoniidae	0	0	1	1
Sandhills	Hydra		0	1	6	0
Sandhills	Nematomorpha		0	4	0	1
Sandhills	Odonata	Aeshnidae	0	0	2	0
Sandhills	Odonata	Coenagrionidae	1	1	4	1
Sandhills	Odonata	Lestidae	0	0	4	0
Sandhills	Oligochaeta	Oligochaeta	42	7	32	27
Sandhills	Trichoptera	Calamoceratidae	2	0	2	0

Sandhills	Trichoptera	Calosidae	3	0	1	1
Sandhills	Trichoptera	Coenosuchidae	7	2	1	0
Sandhills	Trichoptera	Ecnomidae	7	1	37	17
Sandhills	Trichoptera	Hydroptilidae	0	26	21	12
Sandhills	Trichoptera	Leptoceridae	7	5	2	1
Sandhills	Turbellaria	Dugesiidae	0	0	2	0

Appendix 3. Differences in SIGNAL score and principal component analysis of macroinvertebrates.

In this report, we show differences among the various sites on the basis of two mathematical ways of determining macroinvertebrate numbers and taxa, the SIGNAL score and a principal component analysis.

The SIGNAL (**S**tream **I**nvertebrate **G**rade **N**umber – **A**verage **L**evel) score is derived from a simplified system to give an indication of water quality (Chessman 2003). The score is based on a table that gives each taxon a grade from 1-10 that is derived from the perception of how representative that taxon is for indicating high quality water. The higher the grade the less tolerant that taxon is considered to be to pollution, the lower the grade the more pollution tolerant. The grade for the taxon is then multiplied by a weighting associated with the numbers of that taxon collected, but the weighting only varies from 1-5 as shown in Table 4.1.

Table A3.1. Relationship between number of any single taxon collected and weighting for that number in SIGNAL calculation. This table shows that collecting more than 20 from any taxon is not considered in the calculation of a SIGNAL score.

Number of taxon collected	Weighting
1-2	1
3-5	2
6-10	3
11-20	4
>20	5

As the SIGNAL score is determined by the calculation in equation 1, a few high grade macroinvertebrates can have a much higher influence on the overall score than many lower grade macroinvertebrates, especially as no more than 21 are considered in the actual calculation.

$$\text{SIGNAL score} = \frac{\sum Grade \times weighting}{\sum taxa\ weight} \qquad \text{(Equation 1)}$$

The SIGNAL score is easy to determine using an Excel spread sheet and also minimises the time for sorting macroinvertebrates as only 21 individuals from a taxon are considered in the calculation, therefore counting more than that is unnecessary (for grades used in this report, see Table A3.2).

In contrast, a principal component analysis does not consider any taxon different from another, but only considers how many taxa and how many individuals for each taxa were collected. The comparison is therefore not weighted towards any single taxa, but makes the comparison as though nothing is known about which taxa may represent a certain freshwater condition. Because of this lack of precondition expectation, the analysis will be more robust towards comparing biodiversity at different locations and a few specimens will not unduly bias the analysis. The data is also natural log transformed to reduce the influence of highly common species on the analysis as mentioned in the results. A principal component analysis is not as easily calculated as a SIGNAL score, but requires specialised computer/statistical programs, but for making site-by-site comparisons as required for ecological studies presents a more complete picture of what is happening in each location.

Table A3.2. Grades of taxa used in this report taken from Chessman (2003). No grades are given for Copepoda, Cladocera or Ostracoda as grades were omitted in Chessman (2003).

Taxon	**Family**	**Signal Grade**
Acarina	Acarina	6
Amphipoda	Ceinidae	2
Bivalvia	Sphaeriidae	5
Coleoptera	Dytiscidae	2
Coleoptera	Gyrinidae	4
Coleoptera	Haliplidae	2
Coleoptera	Hydrophilidae	2
Coleoptera	Psephenidae	6
Coleoptera	Scirtidae	6
Collembola	sp.	1
Crustacea	Cladocera	
Crustacea	Copepoda	
Crustacea	Ostracoda	
Decapoda	Atyidae	3
Decapoda	Parastacidae	4
Diptera	Ceratopogonidae	4
Diptera	Culicidae	1
Diptera	s-f Chironominae	3
Diptera	s-f Orthocladiinae	4
Diptera	s-f Tanypodinae	4
Diptera	Simulidae	5
Diptera	Stratiomyidae	2
Ephemeroptera	Baetidae	5
Ephemeroptera	Caenidae	4
Ephemeroptera	Leptophlebiidae	8
Gastropoda	Lymnaeidae	1
Gastropoda	Physidae	1
Gastropoda	Planorbidae	2
Hemiptera	Corixidae	2
Hemiptera	Naucoridae	2
Hemiptera	Notonectidae	1
Hemiptera	Veliidae	3
Hirudinea	Glossiphoniidae	1
Megaloptera	Corydalidae	7
Odonata	Aeshnidae	4
Odonata	Coenagrionidae	2
Odonata	Gomphidae	5
Odonata	Hemicorduliidae	5
Odonata	Lestidae	1
Odonata	Libellulidae	4
Odonata	Synlestidae	7
Odonata	Telephlebiidae	9
Oligochaeta	Oligochaeta	3
Plecoptera	Gripopterygidae	8

Trichoptera	Calamoceratidae	7
Trichoptera	Calosidae	9
Trichoptera	Conosucidae	7
Trichoptera	Ecnomidae	4
Trichoptera	Glossosomatidae	9
Trichoptera	Helicophidae	10
Trichoptera	Hydribiosiidae	8
Trichoptera	Hydropsychidae	6
Trichoptera	Hydroptilidae	4
Trichoptera	Leptoceridae	6
Trichoptera	Philoreithridae	8
Turbellaria	Dugesiidae	2
Turbellaria	Temnocephalidae	5

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