

Testing a potential flow-specific invertebrate metric: LIFENZ

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Outcome 5. Macroinvertebrates and fish: stressor-specific metrics*

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Executive summary

Macroinvertebrate metrics that combine and summarise the sensitivity of taxa to stressors with taxa presence or abundance are commonly used to quantify human impacts on river ecosystems. A common criticism of such metrics is that they can be influenced by multiple stressors making determining the mechanistic cause of changes in metric scores difficult. However, despite the potential limitations, stressor-specific metrics can often distinguish the impacts of their stressor better than other metrics. In addition, when used with other metrics, they can help identify drivers of macroinvertebrate community change.

The Ministry for the Environment (MfE) recently commissioned a project to further develop and validate previously identified nutrient and sediment specific stressor metrics. Changes to natural river flow regimes that alter the magnitude, timing, intensity or duration of high and low flow events can also act as stressors on river ecosystems. This report contributes to the first project by testing a potential hydrologically-sensitive macroinvertebrate metric, LIFENZ. LIFENZ is modelled on the UK Lotic-invertebrate Index for Flow Evaluation (LIFE) and designed to respond to changes in flow regime.

Low flow conditions can stress riverine communities. Departure from a natural flow regime can also alter both the magnitude and impact of other stressors, such as stream-bed sedimentation or elevated nutrient concentrations. Using an invertebrate metric that is sensitive to hydrological conditions in conjunction with other stressor-specific metrics will assist in disentangling drivers of poor stream health. A flow sensitive invertebrate metric may also improve our understanding of eco-hydrological relationships, which are required for setting effective limits to water resource use.

How LIFENZ was tested

Within-site changes in LIFENZ were examined using data from 65 sites in the National River Water Quality Network (NRWQN). Each of the NRWQN sites had up to 29 years of annual invertebrate data and continuous flow records. Separate stepwise multiple regressions were used to identify the main environmental correlates (hydrology, dissolved nutrients, water temperature) of annual within-site changes in LIFENZ and the Quantitative Macroinvertebrate Community Index (QMCI) scores. Environmental conditions associated with between-site differences in LIFENZ and in environment–LIFENZ relationships were also investigated.

Redundancy of LIFENZ and QMCI

As would be expected for any community-based metric, LIFENZ and QMCI scores showed some redundancy (i.e., degree of correlation between the metrics) both between and within sites. However, the strength of correlations (maximum $r = 0.68$ among 65 within-site correlations and one between-site correlation) indicated that the metrics likely provide unique information about drivers of both spatial and temporal differences in invertebrate communities.

Is LIFENZ a hydrologically-sensitive metric?

Like many invertebrate metrics that respond to parameters other than the stressor they were designed to detect changes in, LIFENZ was influenced by predictors other than hydrological conditions (nutrient concentrations and water temperature) both within and between sites. However, hydrological indices generally had a larger influence on LIFENZ than physico-chemical conditions. In a series of stepwise site-specific regressions, a hydrological index was retained as a predictor for 82% of the sites at which within-site changes in LIFENZ scores were predictable, while

only 67% of QMCI models retained a hydrological index. Thus, while not entirely stressor-specific, LIFENZ scores seemed to respond most to changes in the environmental conditions it was developed to detect changes in.

Does LIFENZ respond to particular hydrological indices?

LIFENZ was influenced by one particular aspect of the hydrological regime between sites, but not within them. Between sites LIFENZ scores were more correlated with the average number of days accrual between high flows than the magnitude of floods or low flows. However, of the 28 sites for which within-site patterns in LIFENZ were predictable, there was no detectable pattern between the hydrological parameter, if any, that was retained in the model and site characteristics such as REC Source-of-Flow category.

Regardless of an inability to predict which hydrological indices influenced LIFENZ at a site, LIFENZ did respond to a range of different hydrological indices. This indicates that LIFENZ may be a useful indicator of a variety of different hydrological alterations. However, to ensure reasonable predictive power in LIFENZ-environment relationships for any given site, hydrological indices should be chosen that are considered to relate to the mechanisms most likely to influence macroinvertebrate communities at that site.

Suitability of the dataset

The NRWQN dataset used in the analyses is similar to most water quality monitoring networks in that stand-down periods for invertebrate sample collections after large flood events are in place. The purpose of this stand-down periods is to reduce the impact of flow variability on the collected samples and increase the chances of detecting changes in invertebrate communities caused by water quality or habitat degradation. However, almost all sites in the NRWQN dataset did have invertebrate samples collected after a variety of flow conditions, ranging from years with relatively stable antecedent flows to years in which large flow events occurred in the 30 days prior to sample collection. This indicates that the dataset is not likely to limit the conclusions that can be made about the strength of LIFENZ – hydrology relationships.

Application of LIFENZ and recommendations

Currently LIFENZ is most useful for investigating within-site changes in invertebrate communities. LIFENZ can be calculated from full count or coded abundance invertebrate data. These data can be historic or newly collected data. It is recommended that LIFENZ be calculated alongside other invertebrate metrics. The utility of LIFENZ should be evaluated in conjunction with other stressor-specific metrics. This is particularly important in rivers that have temporal periphyton and/or substrate composition data or have different characteristics from the NRWQN sites, e.g., are smaller headwater sites, or have data that span large hydrological alterations. Such investigations could 1) identify potential redundancy between LIFENZ and other stressor-specific metrics, 2) potentially explain more variation both within and between sites in LIFENZ through the use of periphyton data and 3) also test the generality of the relationships identified in this report in the relatively large NRWQN sites.

Sites that vary in hydrological regime will naturally differ in LIFENZ scores. Development of a ratio of observed LIFENZ scores relative to expected LIFENZ scores at that site if it was unimpacted by hydrological alteration could allow between site comparisons. Further investigation of methods to classify sites based on hydrological regime type and then generate expected LIFENZ scores under

natural hydrological conditions is recommended if between-site comparisons of the impact of hydrological alteration is useful.

Currently, LIFENZ could be used in specific site case studies and in conjunction with the nutrient and sediment specific metrics to:

1. Assess whether nutrient enrichment, sedimentation or flow alteration individually or in combination are the likely causes of degraded stream ecosystem health
2. Estimate the ecological effects of a significant disturbance events, such as a dam failure or extended drought
3. Measure success of flow restoration efforts and the impact of flow regulation or reductions

1 Introduction

1.1 Background

Benthic macroinvertebrates are used globally as indicators of human impacts to river ecosystems. Macroinvertebrate taxa often vary in their sensitivity to environmental impacts such as pollution or increased stream-bed sedimentation. Macroinvertebrate metrics that combine and summarise the sensitivity of taxa to impacts with their presence or abundance at a site can be used to quantify potential human impact.

A common criticism of invertebrate metrics is that they can be influenced by multiple factors that affect macroinvertebrate community composition (Boothroyd and Stark 2000). This makes determining the mechanistic cause of changes in metric scores difficult (Chessman and McEvoy 1998). To alleviate this problem, metrics designed to be stressor-specific may distinguish the impacts of their stressor better than other, non-specific, metrics (e.g., Monk et al. 2006, Kairo et al. 2012). Using stressor-specific metrics in combination, even when they are correlated, can help identify drivers behind changes in macroinvertebrate communities (Clews and Ormerod 2009).

1.1.1 Development and testing of stressor-specific macroinvertebrate metrics

The Macroinvertebrate Community Index (MCI) is widely used in New Zealand. The MCI was developed as a metric of water quality in stony streams (Stark 1985). However, the MCI is also sensitive to changes in other river conditions, such as floods and extended periods of low flow (Boothroyd and Stark 2000), particularly in pristine waterways (Death et al. 2009). The MCI was recently included as a compulsory monitoring tool for regional councils in the National Policy Statement for Freshwater Management (NPS-FM) (NZ Govt 2017). The Ministry for the Environment (MfE) also commissioned a project to investigate the sensitivity of MCI and other macroinvertebrate metrics to different human impacts (MfE contract no. 21630, Clapcott et al. 2017). One of the aims of that project was to establish proof-of-concept for nutrient-specific and sediment-specific invertebrates metrics.

MfE subsequently commissioned further development and validation of nutrient and sediment stressor-specific metrics (Wagenhoff et al. 2018). The current report concerns an additional component of that project; further testing of a potentially hydrologically sensitive macroinvertebrate metric, LIFENZ (Greenwood et al. 2016).

1.1.2 Alterations to river flow as a potential stressor

The timing and quantity of water flow within a river affects river geomorphology and habitat diversity and is often correlated with physico-chemical conditions such as water temperature or nutrient concentrations. This has led to river flow being described as a 'master variable' that affects the distribution and diversity of riverine organisms (Resh et al. 1988, Power et al. 1995).

Alterations to river flow that change natural patterns in magnitude, duration, timing and/or predictability of high and low flows have the potential to act as 'stressors' on riverine communities. Following Wagenhoff et al. (2018) In the current report, I define a 'stressor' as an attribute that, as a consequence of human activity, has exceeded its normal range of variation and affects macroinvertebrate taxa and communities. Stressful conditions relating to river flow regimes may include extended periods of low or no flow, a reduction in the frequency of bed-moving floods leading to excessive periphyton growth, or an increase in the frequency of bed-moving floods such that communities cannot recover between floods. Human activities such as impoundments or water

abstraction can alter river flow regimes, potentially leading to stressful conditions for riverine organisms.

Since water abstraction often allows increased land use intensity, rivers with altered flow regimes are likely to occur in areas where they may be exposed to other potential stressors such as contaminant run-off or increased deposition of fine sediment on the stream-bed (e.g., Matthaei et al. 2010). Such multiple-stressor conditions can make it difficult to disentangle the effects on riverine communities of individual stressors (e.g., changes to flow, nutrient enrichment, stream-bed sedimentation), complicating efforts to minimise impacts or to successfully restore river sections. An invertebrate metric specifically or predominantly influenced by changes to flow-driven hydraulic conditions could assist in detangling causes of poor river health at sites affected by multiple human impacts.

1.1.3 Development and initial testing of a potential hydrologically sensitive invertebrate metric for New Zealand

A potential hydrologically sensitive macroinvertebrate metric (LIFENZ) has been developed for New Zealand (Greenwood et al. 2016) by adapting the UK-specific Lotic-invertebrate Index for Flow Evaluation (LIFE; Extence et al. 1999). Both LIFE and LIFENZ are based on water velocity preferences of benthic macroinvertebrates. LIFE has been shown to respond to both natural and anthropogenic variations in flow in the UK (Extence et al. 1999, Clarke and Dunbar 2005) and been used to identify sites subjected to hydrological stress and to develop river management plans (Monk et al. 2008). Because LIFE is not directly applicable to regions with different fauna, LIFENZ was developed for use in New Zealand (Greenwood et al. 2016).

Development of LIFENZ

LIFENZ was developed following the methodology of Extence et al. (1999) and by assigning water velocity preference categories to aquatic invertebrate taxa using professional judgement. Metric scores are generated using a lookup table to assign a score to each taxon based on their velocity preference category and abundance within a collected macroinvertebrate sample. The abundance categories match those recommended in national sampling guidelines (Stark et al. 2001) and are based on samples collected from an area of 1 m². For full details regarding the development of LIFENZ see Greenwood et al. (2016).

Results from initial testing of LIFENZ

As part of the development of LIFENZ preliminary testing was conducted using 20 years of annual invertebrate data from 66 sites on the National River Water Quality Network (NRWQN). Results of the initial tests indicated that LIFENZ scores were higher in locations with greater water velocity (Greenwood et al. 2016). Hydrological indices relating to antecedent flow conditions, such as the length of time since a recent high flow also explained temporal variation in LIFENZ across the 66 sites.

Hydrological regime was also associated with differences in site-average LIFENZ scores. Sites with more stable flows had lower LIFENZ scores. A statistical method called path analysis indicated that this effect was partly a direct effect of flow on LIFENZ scores, but also linked to changes in periphyton coverage caused by hydrological conditions.

LIFENZ scores were correlated with scores for the most widely used macroinvertebrate metrics in New Zealand, MCI and QMCI, but unexplained variance indicated that the metrics are not entirely

redundant. MCI and QMCI were not affected by local water velocity, but did respond to many of the same environmental differences as LIFENZ within and between sites.

1.2 Project scope and aims

The overall goal of the current project was to extend the initial testing of the LIFENZ metric and assess its suitability as a hydrologically-sensitive invertebrate metric in comparison to existing macroinvertebrate metrics. The specific objectives were to:

1. Determine if LIFENZ scores vary predictably between sites that vary in environmental conditions such as flow regime, catchment size, climate, catchment land cover.
2. Determine whether LIFENZ is more sensitive to antecedent hydrological conditions than QMCI.
3. Investigate which flow metrics or aspects of the hydrological regime most influence differences in LIFENZ values both within and between sites.
4. Determine whether LIFENZ is more sensitive to hydrological conditions at sites with particular hydrological regimes, catchment sizes, climate or catchment land cover.

Greenwood et al. (2016) recommended that future testing of LIFENZ would be beneficial in stream types not found in the NRWQN, i.e., smaller headwater reaches. However, due to the lack of paired invertebrate and hydrological datasets with sufficient temporal replication, an updated NRWQN dataset was used here. This project builds on previous testing (Greenwood et al. 2016) by:

- Updating the end of the NRWQN dataset from 2011 to 2018, adding up to 7 years data to each site.
- Using samples from all seasons, not December to April as in the previous analysis.
- Running individual models for each site rather than a mixed effects model across all sites to better investigate factors correlated with LIFENZ scores within sites.
- Investigating whether site characteristics (e.g., upstream landcover, climate, river type) influence within-site relationships between LIFENZ and hydrological conditions.

2 Methods

2.1 Invertebrate data

Invertebrate data were available from 66 sites in New Zealand's National Rivers Water Quality Network (NRWQN) (Davies-Colley et al. 2011) (Figure 2-1). The network began in 1989 with the goal of monitoring long-term trends in water quality, biology and habitat (Smith et al. 1989). The river catchments in the network drain about half of New Zealand's total land area and are biased towards large rivers. Mean flows observed at gauging stations in the rivers vary from 0.8 to 567 m³s⁻¹ (Smith et al. 1989; see Greenwood et al. (2016) for more site information). Since 1989 samples of aquatic invertebrates have been collected annually, generally between late summer and early autumn, under baseflow conditions (flow < long-term median flow) (Scarsbrook et al. 2000). At each site, seven Surber samples (0.1 m² area, 250-µm mesh) are collected from cobble or gravel substrate (Smith et al. 1989). The samples from seven locations at each site are pooled into one sample for analysis. Samples are sorted in the laboratory using a full count method with sub-sampling of abundant (>100 individuals) taxonomic groups and identified to the lowest taxonomic resolution possible, most commonly genus level.

Data from 1990 to 2018 were used, which resulted in 1698 samples from 66 sites. Most samples (85% or 1428 samples) were collected in summer or early autumn (December to March; Figure 2-2). The 66 sites had annual invertebrate data for 17 to 29 years (Figure 2-3).

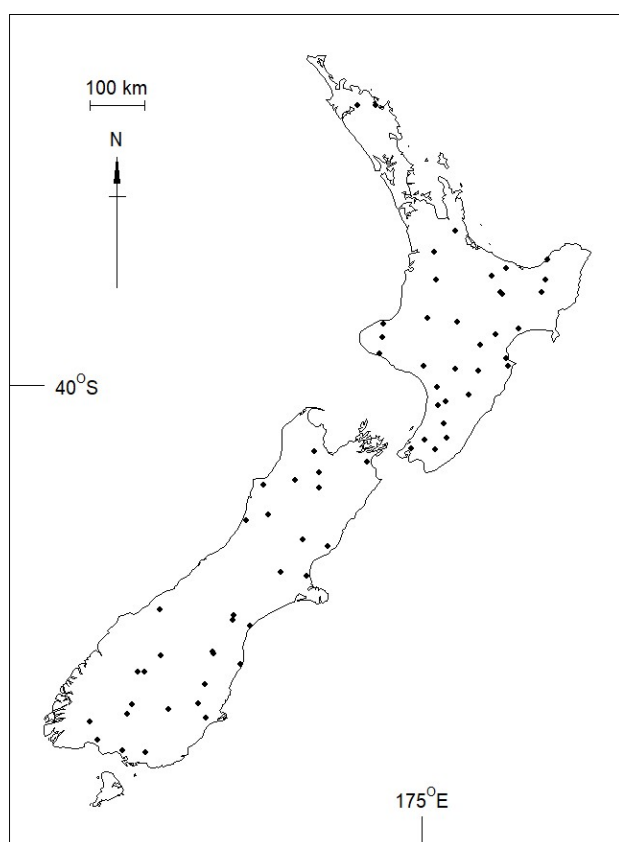


Figure 2-1: Location of the 66 NRWQN study sites. Each site had up to 29 years of annual invertebrate data that was used to test the responsiveness of the invertebrate metric LIFENZ to changes in river hydrology.

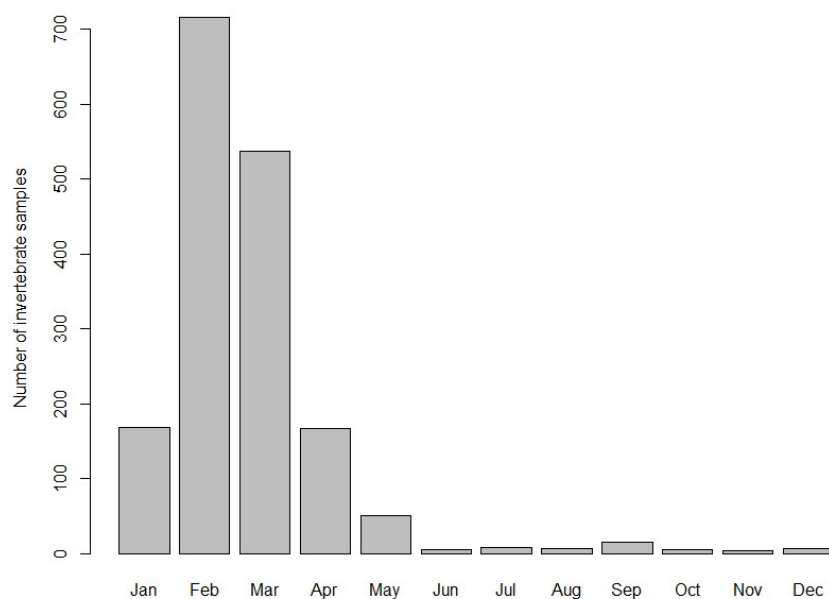


Figure 2-2: Number of invertebrate samples collected in each month. Annual collections of invertebrate samples at 66 national water quality monitoring sites between 1989 and 2018 resulted in a total of 1698 samples, most of which were collected during late summer and autumn.

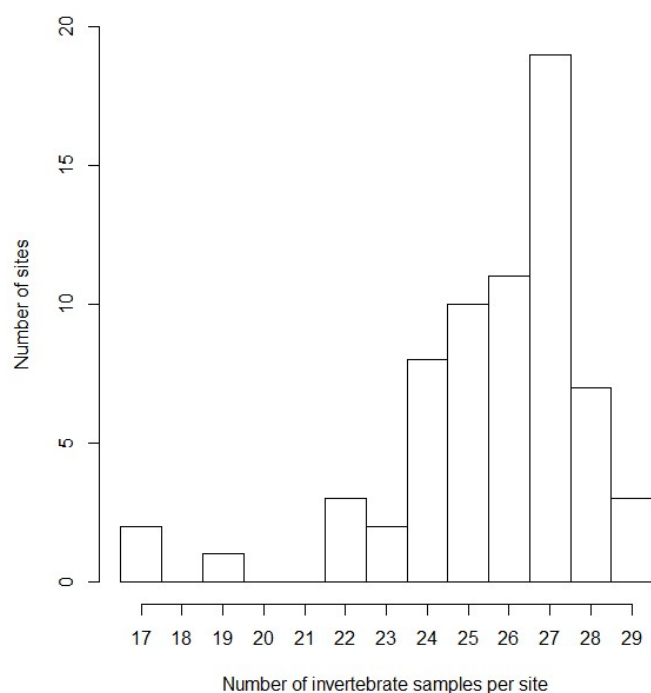


Figure 2-3: Number of national monitoring network sites with between 17 and 29 years of annual invertebrate data. Annual samples were collected between 1989 and 2018 with most sites having data for at least 22 of those years.

2.2 Hydrological data and indices

Most sites had nearby river gauging stations that provided continuous river flow information, although several have modelled flows derived from values observed at gauges several kilometres away. Mean daily flow data were used. Updated hydrological data were available for 65 sites, the remaining site was excluded from further analysis.

Numerous hydrological indices can be calculated from time-series flow data. Many of the indices are intercorrelated and a subset of uncorrelated indices is useful for concise descriptions of hydrological regimes (Clausen and Biggs 2000, Olden and Poff 2003). A limited suite of metrics that described antecedent flow conditions (prior to invertebrate sample collection dates) and site-average flow characteristics were calculated.

For each sampling date, the antecedent flow metrics were the mean, minimum and maximum flows in the previous 30 days (MeanFlow30, MinFlow30 and MaxFlow30), and the number of days since a flood N_m times the median flow, where $N_m = 1.5$ and 3 (DA1.5 and DA3). The number of days since a flow event correspond to days of accrual (DA) or the amount of time for periphyton and invertebrates to recover after a flow event, assuming a flow of that magnitude disturbs their communities. Three times the median flow was chosen as a flow threshold as flood events of this magnitude have previously been related to biological responses (Clausen and Biggs 1997). MeanFlow30, MinFlow30 and MaxFlow30 were standardised by the long-term mean flow to allow for comparisons between sites as well as within sites.

For each site, a range of annual hydrological metrics was calculated. The hydrological year was started on June 1 to ensure summer low flow calculations included flow conditions from a full summer period. These were the annual maximum and minimum flows over a 7-day rolling window (Annual Flood and ALF), the mean number of days between flows 1 and 3 times the median flow (Mean_AnnualDA1 and Mean_AnnualDA3), mean flows in February and July (MeanFeb and MeanJul), the number of times the flow hydrograph reversed (Reversals) and the mean duration of high flow events (PulseLengthHigh). High flow thresholds were defined as a flow magnitude that was exceeded <25% of the time.

Site-specific hydrological metrics were calculated from both the antecedent and mean annual hydrological metrics. Mean values of the annual antecedent metrics included; MaxFlow30 and MinFlow30, DA1.5 and DA3. Mean values of annual metrics included; MeanFeb, MeanJul, reversals, Mean_AnnualDA3, DA1.5, DA3 and PulseLengthHigh. BFI (mean annual 7day minimum flow/mean flow) was also calculated for each site. The metrics SpecificFlood and SpecificMALF were calculated as the mean of Annual Flood and MALF at a site, respectively, divided by the site catchment area. Variables were standardised by mean flow or catchment area where required to allow for between site comparisons. Standardising by area allows high flow catchments to be compared with low flowing catchments, whereas standardising by mean flow removes this influence, retaining differences in flow variability not flow magnitude between sites. See Table 2-1 for further descriptions of mean site hydrological indices. Many of these indices are used in standard flow regime-hydrological analyses such as the Range of Variability (RVA) approach (Richter et al. 1997).

Table 2-1: Hydrological indices extracted for between sites analyses. Not all indices were included in final analyses. Principal components analysis (PCA) was used to reduce the number of terms (see Section 2.4.1).

Parameter	Description	Indicator of	Range
Mean_AnnualDA3	Mean mean-annual number of days between flow events > 3 times the median flow.	Annual average days accrual	16-364
Mean_AnnualDA1	Mean number of days between flow events > median flow	Annual average days accrual	11 - 96
Mean_DA1.5	Mean number of days since a flow events > 1.5 times the median flow.	Antecedent days accrual	23 - 787
Mean_DA3	Mean annual number of days since a flow events > 3 times the median flow.	Antecedent days accrual	37-2680
Mean_MeanFlow30	Mean-annual mean flow in the 30 days prior to invertebrate sample collection standardised by long-term mean flow	Antecedent mean flow (standardised by flow)	0.4-1.2
Mean_MaxFlow30	Mean-annual maximum flow in the 30 days prior to invertebrate sample collection standardised by long-term mean flow	Antecedent maximum flow (standardised by flow)	1.7 - 993
SpecificFlood	Max annual 7- day high flows / catchment area. Log10 transformed	High flow magnitude (standardised by catchment area)	0.03 – 0.8
Mean_MeanFeb	Mean flow in February	River size during summer	0.37 – 488 m ³ s ⁻¹
Mean_MeanJul	Mean flow in July	River size during winter	1.7 – 491 m ³ s ⁻¹
BFI	Mean annual 7-day minimum flow / mean flow	Low flow magnitude (standardised by flow)	0.09 – 0.7
SpecifcMALF	Mean annual 7-day minimum flow / catchment area.	Low flow magnitude (standardised by catchment area)	0.0007 – 0.04
Mean_PulseLengthHigh	Mean number of days in each flow event > 75 th flow percentile	Duration of high flows	2.3 – 18.0
Mean_Reversals	Number of times the hydrograph switches between rising and falling or and vice versa	Flow variability	90-199

2.3 Other environmental information

Monthly measurements of dissolved nutrient concentrations, spot water temperature, conductivity and visual assessments of periphyton cover were undertaken at the sampling sites (for full sampling details see Smith and Maasdam 1994). From these data the nearest prior monthly measurements of water temperature, conductivity, dissolved reactive phosphorus (DRP) and nitrate-nitrite-N (NO_x-N) prior to each annual aquatic invertebrate sample collection were extracted. As nitrate generally comprises the majority of NO_x-N, nitrate-nitrite-N is referred to as nitrate in this report. Maximum DRP and nitrate concentrations were also extracted for each year at each site. Spot water temperature values will vary according to the time of day the measurements are taken. However, sites are generally visited in the same order and thus at the same time of day.

Site-average periphyton coverage data were included in the analysis in Greenwood et al. (2016). Periphyton data were not included because of several limitations with the temporal resolution of the data. Periphyton data consist of in situ, visual assessments of cover. Visual assessments cannot be made when water levels are too deep or fast to stand safely in the water, or when the water is too turbid to see the periphyton. These limitations led to temporal gaps in periphyton data for many sites, and an almost complete lack of data at some sites. Periphyton cover data were determined to be too incomplete to be included as an explanatory parameter in the within-site analysis and the between-sites analyses.

River network segment numbers (NZReaches) were used to extract parameters representing the environmental characteristics of the NRWQN sites from existing databases (REC version 1 and Freshwater Ecosystems New Zealand, Snelder and Biggs 2002, Leathwick et al. 2011). Environmental variables describing catchment morphology, landcover type, slope, and temperature have previously been shown to influence macroinvertebrate communities (Leathwick et al. 2011, Clapcott et al. 2012). Variables were extracted that related to upstream land cover, local climate and topography of the catchment and sampling reach (Table 2-2). REC Climate and Source-of-flow categories were also assigned to each site.

Table 2-2: Description and summary statistics of parameters describing catchment and segment characteristics. REC Climate and Source-of-flow categories were extracted from the River Environment Classification (Snelder and Biggs 2002); pastoral land cover was calculated from Land Cover Data Base Version 3, and catchment and segment predictors were extracted from the Freshwater Ecosystems New Zealand database (Leathwick et al. 2011).

Group	Predictors	Description	Median (range)
Land cover	usPastoral	Proportion upstream catchment in pastoral land cover	28% (0-91)
Climate	usRain50	Catchment rain days > 50 mm / month	0.19 (0.02-1.1)
Climate	segAveTwarm	Segment January mean air temperature	16.7 °C (13.8 – 19.6)
Topography	Sqrt(SegSlope)	Square root segment slope	0.002 ° (0-0.03)
Topography	Log10(Cat_area)	Log upstream catchment area	1172 Km ² (11-16548)
REC categories	REC Climate	Climate categories from REC	-
REC categories	REC Source-of-flow	Source-of-flow categories from REC	-

2.4 Statistical analyses

2.4.1 Between sites

Pearson correlation was used to investigate the degree of correlation, and hence redundancy between site average LIFENZ and QMCI scores.

Selection of hydrological parameters

Following Olden and Poff (2003), principal components analysis (PCA) was used to create a subset of independent hydrological indices between sites. All hydrological indices in Table 2-1 were included in the PCA. The goal of the analysis was to identify a subset of indices that describe the major source of hydrological variation between sites while minimising redundancy (i.e., strong correlations between selected indices). The significance of axes was assessed using the broken stick method. Because principal components axes are by definition orthogonal, indices that aligned with separate axes were selected to ensure that chosen indices are relatively independent. Three indices for use in further between-sites analyses were selected. See Section 3.1.1 for selected indices.

Variation in LIFENZ between sites

LIFENZ scores generally vary between sites that differ in local water velocity. However, water velocity data are not widely available or easily generalisable between sites. REC Climate and Source-of-flow

categories can differentiate hydrological regime types (Snelder and Booker 2013). If LIFENZ is influenced by overall river hydrological regime then these categories may be able to assist in identifying ranges of expected LIFENZ scores between sites. Separate one-way analysis of variance (ANOVAs) were used to test whether mean site QMCI and LIFENZ scores differed between REC Climate and Source-of-flow categories, with post-hoc Tukey tests identifying any significant pairwise differences between categories. Because only one site occurred in the glacial mountain Source-of-flow category, this site was combined with the mountain category.

Categorising sites can be a useful and efficient way to investigate whether biota-environment relationships are similar at sites with similar physical characteristics. However, environmental conditions are often correlated and identifying a limited number of categories that represent differences in all environmental parameters that influence biota is difficult. Due to the limited number of sites (65), the hierarchical nature of the REC categories (i.e., combined Climate – Source-of-flow categories) was not able to be used as there were too few sites in any one combined category for analysis. Instead multiple regression was used to investigate the relationships between multiple continuous environmental predictors with LIFENZ and QMCI site scores. To select environmental predictors for inclusion in the models, four categories of environmental conditions likely to affect macroinvertebrate metrics were considered. These were land cover, climate, hydrology and site and reach topographic conditions. Correlations between candidate environmental parameters in each category were used to reduce the number of parameters included in the final models.

For the land-cover category, the proportion of upstream catchment that was pastoral land cover (usPastoral) was used as the sole parameter. Median maximum annual DRP and nitrate concentrations were positively correlated ($R = 0.7$) and also correlated with upstream pastoral land cover individually (DRP: $R = 0.6$, nitrate: $R = 0.7$), indicating the pastoral land cover was a reasonable proxy for dissolved nutrient concentrations.

The number of heavy rainfall days (>50 mm) per month (usRain50) and summer air temperature (segAveTWarm) were used to indicate climatic differences between sites. Local river slope (segSlope) and upstream catchment area (cat_area) were used as site and catchment topography parameters. See Table 2-2 for further details of these parameters.

Three hydrological parameters were also included based on the results of the PCA on hydrological indices described above.

Variance inflation factors (VIF) were calculated for the LIFENZ and QMCI models to assess collinearity of predictors. $VIF < 5$ for each parameter was deemed acceptable.

The fit of each regression model was assessed using leave-one-out cross-validation. This process involves using the predictor variables to generate a series of models omitting one datapoint each time. Each model is used to predict the value of the response variable (LIFENZ or QMCI) for the omitted datapoint. Observed values were plotted against predicted values from these models and Nash Sutcliffe Efficiency (NSE) values were generated to assess predictive power. NSE is commonly used to assess predictive power in hydrological models (Nash and Sutcliffe 1970) and ranges from $-\infty$ to 1. A value of 1 indicates perfect model fit, 0 indicates model predictions are as accurate as the mean of the observed data and negative values indicate that the mean is a better predictor than the model.

Parameters were log10 or square-root transformed where required to approximate normal distributions.

2.4.2 Within sites

Antecedent conditions relative to full year

Invertebrate samples cannot be collected during floods because of risks to the person collecting the sample. Many sampling protocols for monitoring designed to detect human impacts on water quality or in-stream habitat also recommend samples are not collected immediately following a high flow event. This reduces the influence of flow conditions on the invertebrate community and allows for better comparison between sampling events. In line with this, invertebrate samples for the NRWQN are generally collected in summer and generally after a stand-down period after high flows. This means that flow conditions prior to invertebrate sample collection (antecedent flow) may not encompass the full range of flow conditions experienced by a site. While this is ideal for investigating long-term trends in water quality or habitat conditions, it is a potential limitation for testing the responsiveness of the LIFENZ index to antecedent flow conditions.

To investigate whether flow conditions prior to invertebrate sample collection were a good representation of flow conditions experienced by the invertebrate community throughout the year, the ratio of maximum annual flow (calculated across a 7-day rolling window) and the maximum flow in the 30 days prior to invertebrate samples was calculated annually for each site (Max flow ratio). A ratio of 1 or greater indicates that a flow equal or greater to the maximum annual 7-day flow event occurred in the 30 days prior to sample collection. A ratio of 0.5 indicates the maximum flow in the 30 days prior to invertebrate sample collection was half the size of the maximum annual flow.

Correlation between QMCI and LIFENZ scores within sites

Pearson correlations were used to investigate the degree of correlation, and hence potential redundancy, between QMCI and LIFENZ at each site.

Predicting variation in QMCI and LIFENZ scores within sites

Environmental parameters that vary temporally within a site and that are likely to influence invertebrate community composition were assessed for inclusion in multiple regressions predicting LIFENZ and QMCI scores over time within each site.

Parameters considered for inclusion in the models were the nearest prior monthly nutrient (DRP and nitrate) concentrations and spot water temperature, various antecedent flow metrics (maximum, minimum, mean flows in the 30 days prior to invertebrate collection and days accrual since a flow event > three times the median flow) and several annual mean flow statistics (e.g., mean annual low flow (MALF) within the year of invertebrate sample collection). To reduce the terms included in the analysis, Pearson correlation matrices were generated for the candidate predictor variables for each site and results were used to guide subsequent variable selection for the models. The same parameters were included in models for each site.

Two stepwise multiple regressions were conducted for each site, one each predicting QMCI and LIFENZ scores, using function 'step' in R. Terms were removed using 'backwards elimination', where the full model with all predictors is the starting point and terms are removed individually until there is not improvement in model fit statistics (in this case, AIC).

An assumption of linear regression is that residuals are independently and normally distributed. With repeat measurements from the same site over time there is potential for residuals of a model to be correlated over time (autocorrelation). To account for this the following procedure was performed for each site:

- the final stepwise model was run both with and without a residual autocorrelation structure that accounts for covariation in the residuals. This was done using the 'correlation' option in the GLS function in R with AR-1 autocorrelation as the autocorrelation structure;
- likelihood ratio tests ($\alpha = 0.05$) were used to test whether addition of the autocorrelation structure improved model fit. If it did, the structure was included in the final model, otherwise the model was run without the autocorrelation structure.

The fit of each final model was assessed using leave-one-out cross-validation, as described in Section 2.4.1. In summary, once a model was selected using the stepwise procedure, the variables included in the selected model were used to generate a series of models omitting one datapoint each time, each of which was used to predict the value of the omitted datapoint.

In view of issues raised in the literature about the validity of stepwise linear regression (Whittingham et al. 2006, Mundry and Nunn 2009), for selected models, a procedure that identifies the best subsets of models given a selection of predictor variables and ranks them based on goodness of fit statistics was also run. The best subsets procedure was performed using 'bestglm' in R with models ranked by Bayesian information criterion (BIC). The final models identified in the stepwise procedure were compared with those identified by the best subsets procedure to ensure that no good alternative models were ignored.

The within sites multiple regression analyses were run on 63 sites as two sites did not have events > 3 times the median flow and were removed from the analysis.

Do site characteristics influence LIFENZ model performance?

Separate one-way ANOVAs were used to test whether upstream pastoral land cover, catchment area and conductivity differed significantly between sites where within-site patterns in LIFENZ scores were (NSE of final model >0) and were not (NSE of final model <0) explained by temporal changes in water temperature, dissolved nutrients and flow conditions. The same analyses were applied to QMCI scores.

3 Results

3.1 Between sites

A high correlation between mean site LIFENZ and QMCI scores would indicate strong similarities between the metrics and suggest that LIFENZ provides little additional information to that provided by QMCI alone. However, while mean LIFENZ and QMCI site scores were positively correlated (Figure 3-1), the strength of the correlation was not overly high ($r = 0.62$). This indicates that LIFENZ provides unique information about the factors causing differences in invertebrate communities between sites, depending on the variables that each index responds to.

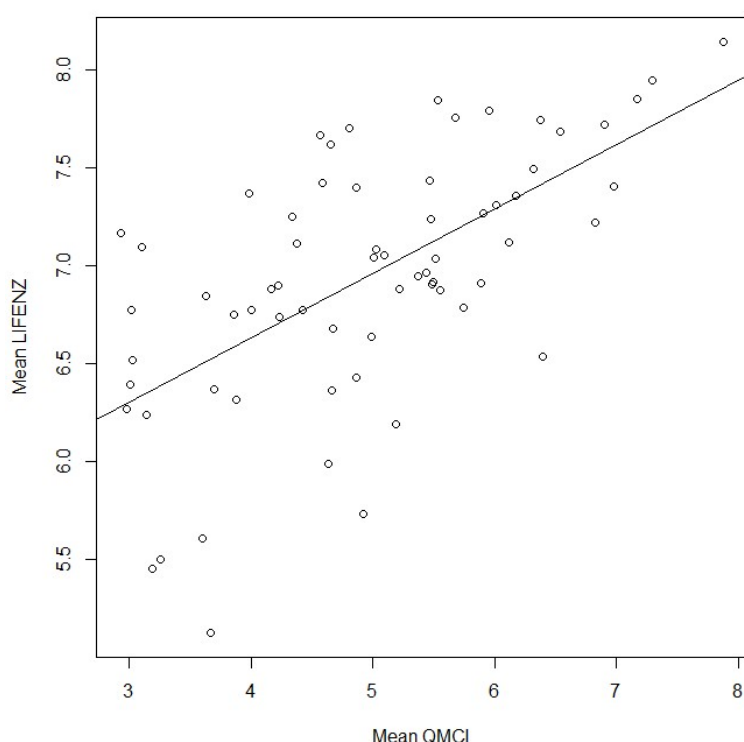


Figure 3-1: Correlation between mean LIFENZ and mean QMCI site scores. Solid line is linear regression ($n = 65$, $r = 0.62$).

3.1.1 Selection of Hydrological parameters

Thirteen hydrological indices, based on mean antecedent and mean annual flow conditions, were included in the PCA. The broken stick method identified that the first two principal components axes (PC1 and PC2) should be retained resulting in a 2-dimensional plot (Figure 3-2). The total variance explained by the PCA was 70%, with PC1 explaining 45% and PC2 25% of the variation in hydrological indices between sites. SpecificFlood, Mean_AnnualDA3 and SpecificMALF (see Table 2-1 for definitions) were selected as the indices to include in the between-site regression analyses. These three indices were aligned with different axes in the PCA and had some of the stronger loadings on the axes (Figure 3-2 and Table 3-1). However, each of these indices was correlated with other hydrological indices.

Table 3-1: PCA axis loading correlations of the three hydrological indices selected for between-site regression analyses. A rank correlation strength of 1 indicates that the hydrological index had the highest correlation with that axis. Thirteen indices were included in the PCA. See Table 2-1 for parameter details.

	PC1		PC2	
	Correlation	Rank corr. Strength	Correlation	Rank corr. Strength
Mean_AnnualDA3	-0.36	1	-0.21	8
SpecificMALF	-0.19	11	0.30	4
Specific Flood	0.12	13	0.34	2

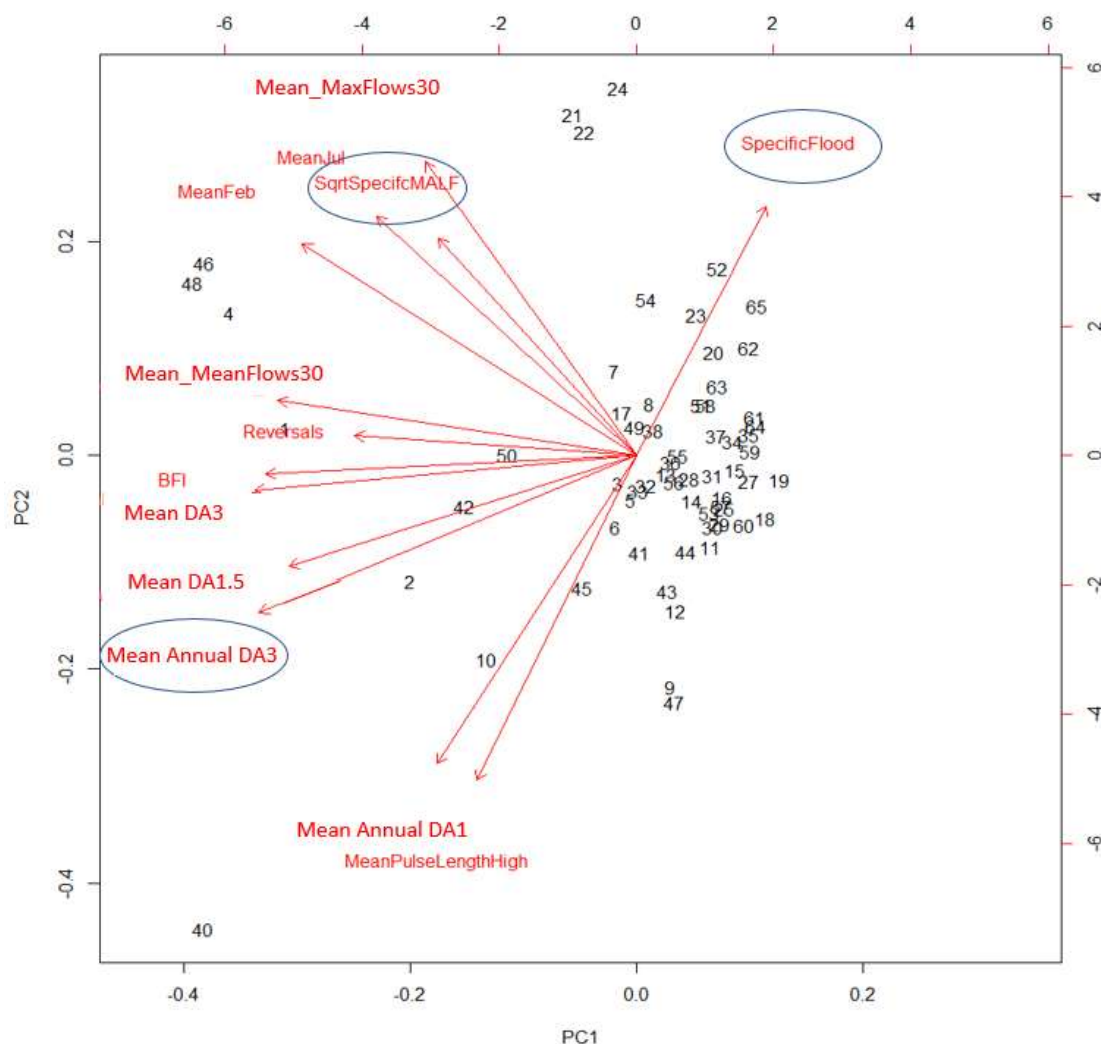


Figure 3-2: Ordination from the principal components analysis (PCA) of the 65 sites based on hydrological indices. Numbers indicate locations of sites. Arrows indicate the loading of each index on the two PCA axes. Hydrological indices with arrows that point in similar directions show similar patterns between the sites. Circled indices were selected for between-site regression analyses.

3.1.2 REC Climate and Source-of-flow categories

LIFENZ scores may differ naturally between sites that vary in climatic conditions, particularly sites that have different rainfall patterns, which will influence the natural hydrological regime. Most of the NRWQN sites used in this study are in the Cool-Wet REC Climate category (41 of 65 sites), making a robust test of whether LIFENZ varies between REC Climate classes difficult. Neither QMCI ($F_{4,61} = 1.4$, $P = 0.3$) nor LIFENZ ($F_{4,61} = 0.4$, $P = 0.8$) differed significantly between the REC Climate categories.

Mean LIFENZ scores did vary significantly between REC Source-of-flow categories ($F_{3,62} = 15.0$, $P < 0.001$), with values generally higher in categories where higher flow variability is likely to occur, i.e., hill and mountain rivers, compared to lowland or lake-fed rivers (Figure 3-3). Overall, LIFENZ scores were highest in mountain and hill rivers, lower in lowland rivers and lower still in lake-fed rivers (Figure 3-3). QMCI scores also varied significantly between REC Source-of-flow categories ($F_{3,62} = 8.4$, $P < 0.001$), but there were fewer significant pairwise differences between categories. Like LIFENZ, QMCI values were higher in hill and mountain rivers than lowland or lake-fed rivers, but values in lowland sites were not significantly different from sites in lake-fed rivers (Figure 3-4).

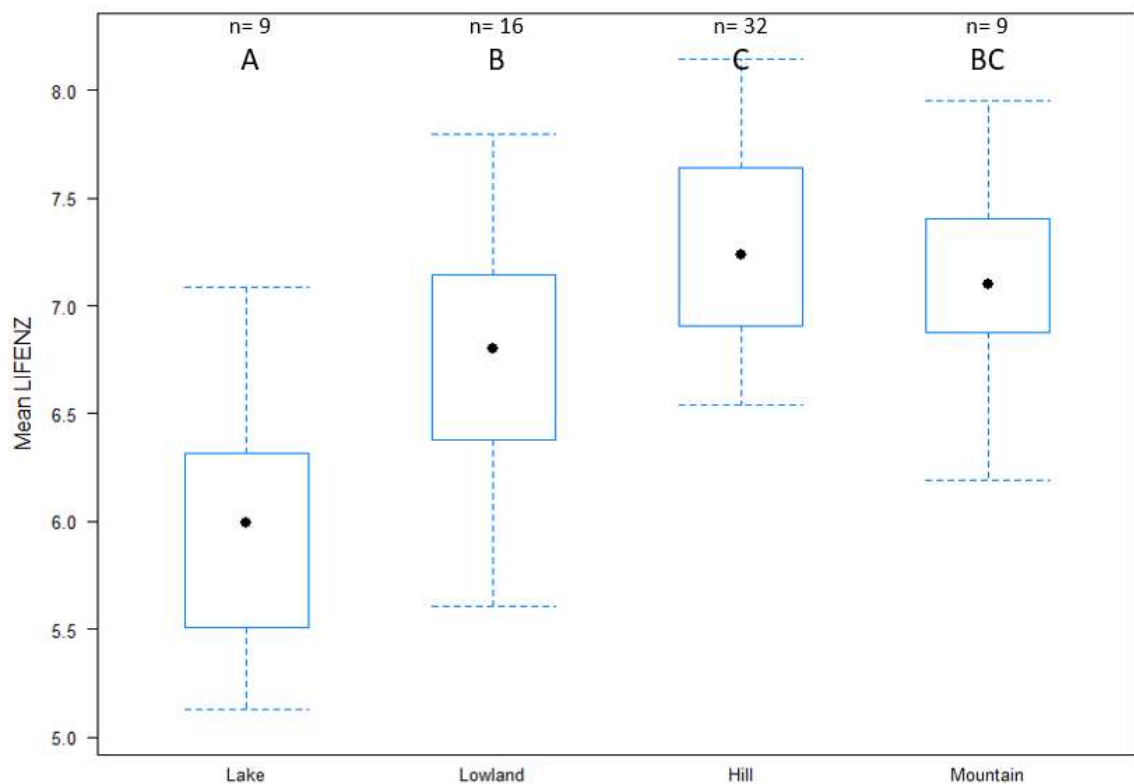


Figure 3-3: Box and whisker plots of mean site LIFENZ scores by REC Source-of-flow categories. Dots indicate medians, boxes end at the 25th and 75th percentile and whiskers at the 5th and 95th percentiles. Letters denote significant pairwise differences between categories; categories with the same letter are not significantly different. Sample sizes in each category are indicated above the boxes. The glacial mountain category was combined with the mountain category.

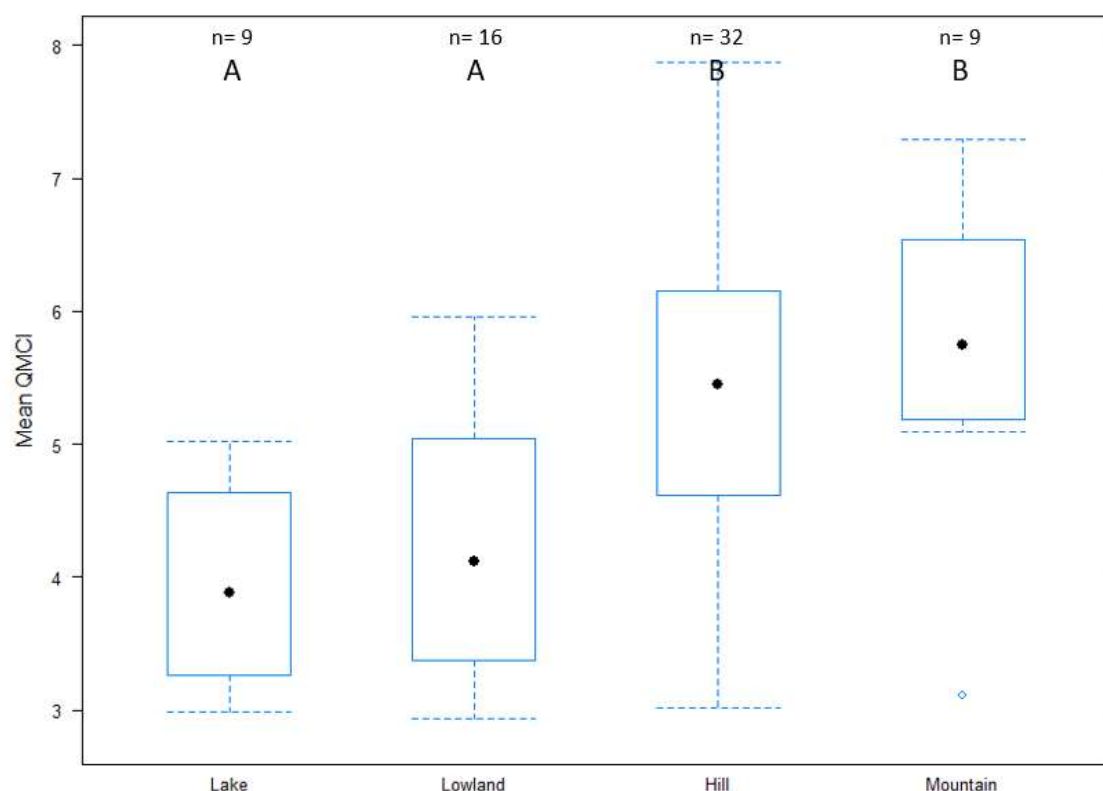


Figure 3-4: Box and whisker plots of average site QMCI values by REC Source-of-flow categories. Dots indicate medians, boxes end at the 25th and 75th percentile and whiskers at the 5th and 95th percentiles. Letters denote significant pairwise differences between categories; categories with the same letter are not significantly different. Sample sizes in each category are indicated above the boxes. The glacial mountain category was combined with the mountain category.

3.1.3 Environmental gradients

The three hydrological indices selected for regression analysis (see Section 3.1.1) were added to the other predictor variables (see Table 3-2 for predictor details). VIF indicated low collinearity between predictors (VIF <5 for all parameters).

The LIFENZ model performed better than the QMCI model in the cross-validation assessments, however both models had positive NSE values, indicating that the models could explain some between-site differences in both macroinvertebrate metrics (NSE for LIFENZ = 0.38, QMCI = 0.17). Predictions of QMCI showed slightly more bias than LIFENZ, with greater underpredictions for higher observed values (Figure 3-5).

Upstream pastoral land cover, segment slope, upstream catchment area and mean days of accrual between flow events were significant predictors of between-site differences in both QMCI and LIFENZ (Table 3-2). Both QMCI and LIFENZ scores increased at sites with decreasing upstream pastoral land cover, decreasing days of accrual, decreasing catchment area, and increasing river gradient (Table 3-2). QMCI values were also higher at sites where the summer air temperature was lower. While both QMCI and LIFENZ were influenced by similar predictors, the significant hydrological predictor (mean days accrual) had a more significant effect on LIFENZ ($p < 0.001$) than QMCI ($p = 0.01$). Sites with longer average accrual times had lower average LIFENZ and QMCI values.

Table 3-2: Descriptions of predictors and results from separate multiple regressions between the predictors and LIFENZ and QMCI. Non-significant terms (alpha = 0.1) are indicated by –. Wald test (F) statistics, significance (p values) and the direction of the relationship (in brackets in the F column) are shown for significant predictors.

Group	Predictors	Description	LIFENZ		QMCI	
			F	p	F	p
Land use	usPastoral	Proportion upstream catchment in pastoral land cover	6.3 (-)	0.02	4.7 (-)	0.03
Climate	Log10(usRain50)	Catchment rain days > 50 mm / month	-	-	-	-
Climate	segAveTwarm	Segment summer air temperature	-	-	5.9 (-)	0.02
Topography	Sqrt(SegSlope)	Square root segment slope	12.1 (+)	<0.001	10.1 (+)	0.002
Topography	Log10(Cat_area)	Log upstream catchment area	18.3 (-)	<0.001	11.2 (-)	0.001
Hydrology	Log10(meanDA)	Log mean annual mean number of days between flows > 3 times long-term median flow; mean days accrual	23.4 (-)	<0.001	7.0 (-)	0.01
Hydrology	Sqrt(SpecificMALF)	Square root of mean annual 7-day low (m3 s ⁻¹) flow divided by catchment area (km ²)	-	-	-	-
Hydrology	Log10(SpecificFlood)	Log of mean annual 7-day maximum flow (m3 s ⁻¹) divided by catchment area (km ²)	-	-	-	-

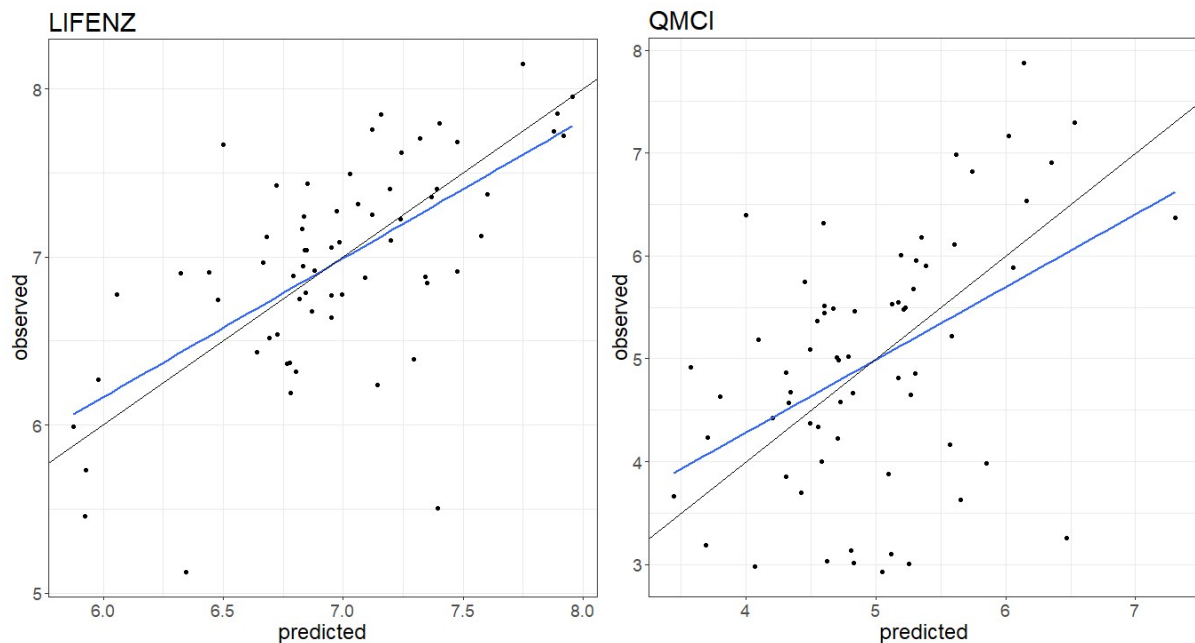


Figure 3-5: Observed against hold-one-out cross validation predicted values for LIFENZ and QMCI (n = 65). Black line represents 1:1, blue line is linear regression.

3.2 Within sites

3.2.1 Antecedent conditions relative to full year

The median annual maximum flow ratio for all sites was less than 1, indicating that for all sites maximum flows in the 30 days prior to sample collection are generally lower than the maximum flow a site experiences during a year (Figure 3-6). However, almost all sites experienced several years where the maximum flow ratio was >1 , indicating that antecedent maximum flows did exceed maximum annual flows (calculated over a 7-day rolling window; Figure 3-6). For several sites (e.g., WN5, TK4, TK6) median flow ratios were relatively close to 1, indicating that antecedent maximum flows were close to annual maximum flows in almost half of the years sampled. For other sites, antecedent maximum flows were much less than maximum annual flows in most years (median flow max ratio $\ll 1$ e.g., WA 7, TK5, HV2), although even at these sites antecedent maximum flows were greater than annual maximum flows (flow max ratio >1) in at least several years (Figure 3-6). As invertebrate samples were largely collected in summer, the sites with maximum flow ratios $\ll 1$ in most years may have more floods in winter than in the summer-autumn sampling period. Sites with higher maximum flow ratios in most years may have floods distributed relatively evenly through the year.

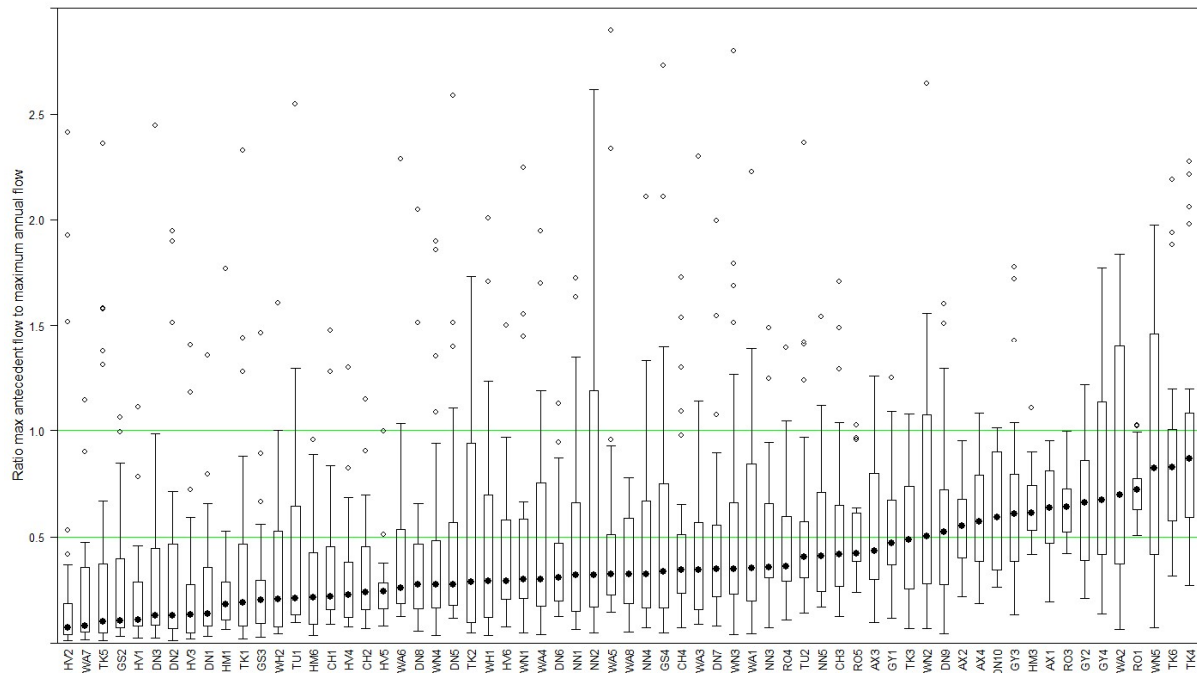


Figure 3-6: Box and whisker plots of the annual ratio of maximum flow in the 30 days prior to collection of an invertebrate sample (antecedent flow) to maximum annual flow (calculated across a 7 day rolling window) in the year prior to invertebrate sample collection. A ratio of <1 indicates that the maximum flow in the month prior to invertebrate sample collection was lower than the annual maximum flow for that year. Green lines indicate ratios of 0.5 and 1. n ranges between 17 and 29 for each site, median = 26.

A similar analysis and pattern was observed for antecedent nitrate concentrations (the most recent prior nitrate concentration) and the maximum nitrate concentration observed at the site within the sampling year. At most sites the most recent nitrate concentrations prior to an invertebrate sample collection were less than 50% of maximum nitrate concentrations observed over the year (Figure 3-7). However, at 28 sites (43%) the maximum annual nitrate concentration occurred within the 30 days prior to the collection of the invertebrate sample.

Thus, while antecedent maximum flow and nitrate concentrations were generally lower than maximum values experienced throughout the rest of the year, at most sites the temporal dataset included at least one year where antecedent flow and nitrate conditions were equal to or greater than maximum annual values. A dataset where invertebrate sample collections were distributed evenly over the year, and which corresponded more closely to the range of flows at each site could increase the degree to which LIFENZ discriminates the effects of antecedent flows. However, almost all sites included a range of maximum flow ratios, including at least one year where high flows occurred just prior to invertebrate sample collection.

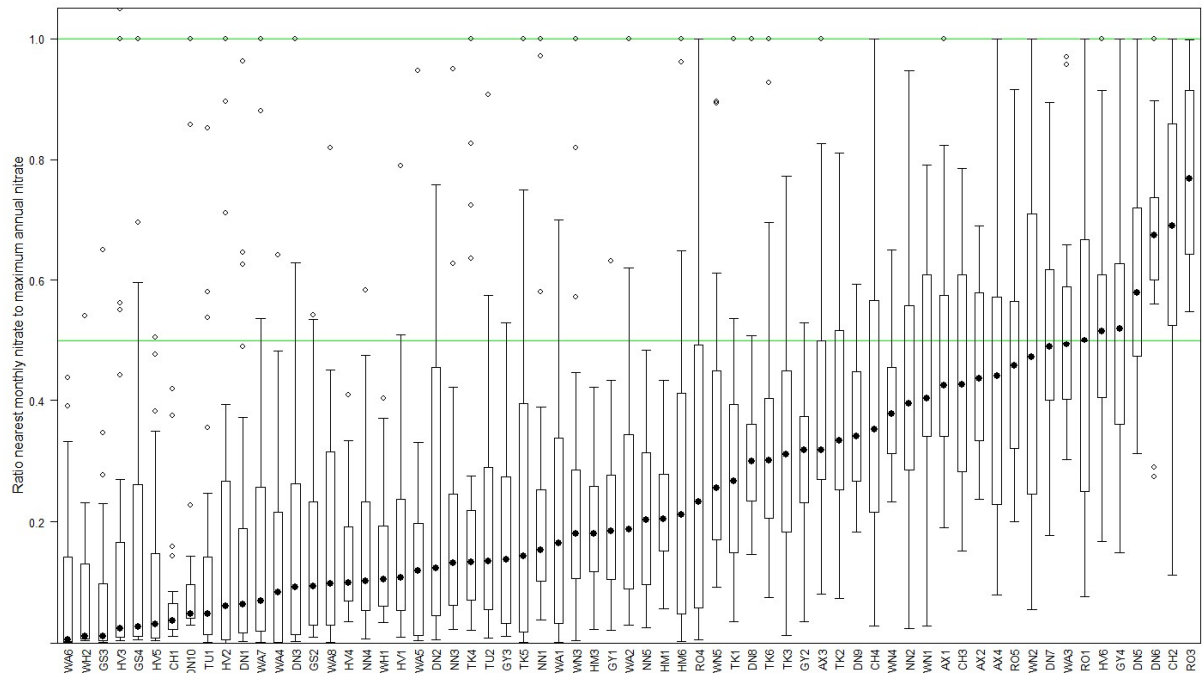


Figure 3-7: Box and whisker plots of the annual ratio of the nearest nitrate concentration available prior to invertebrate sampling to annual maximum nitrate concentrations. A ratio of <1 indicates that nitrate concentrations just prior to invertebrate sample collection were lower than the annual maximum concentration for that year. Green lines indicate ratios of 0.5 and 1. N ranges between 17 and 29 for each site, median =26.

3.2.2 Within-site correlations between QMCI and LIFENZ scores

At most sites QMCI and LIFENZ scores were positively correlated (Figure 3-8 and Figure 3-9), although four sites had slightly negative correlation coefficients (Figure 3-9). More than 40% of the sites had correlation coefficients <0.3, and the maximum correlation between QMCI and LIFENZ scores at a site was 0.68. These relatively low within-site correlations indicate that the two metrics can provide unique information regarding temporal changes in invertebrate community composition.

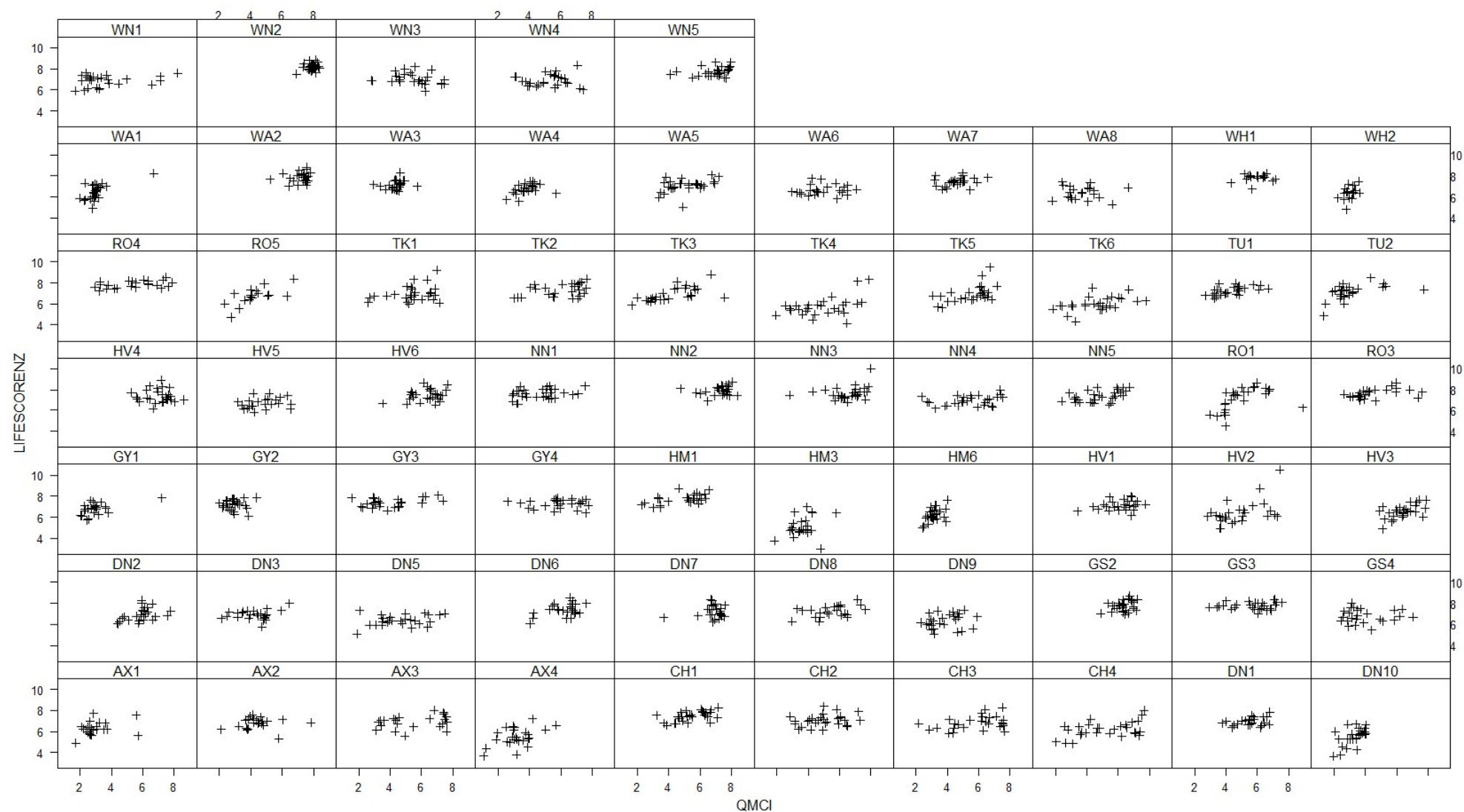


Figure 3-8: Plots of annual QMCI and LIFENZ at 65 sites. n ranges between 17 and 29 for each site.

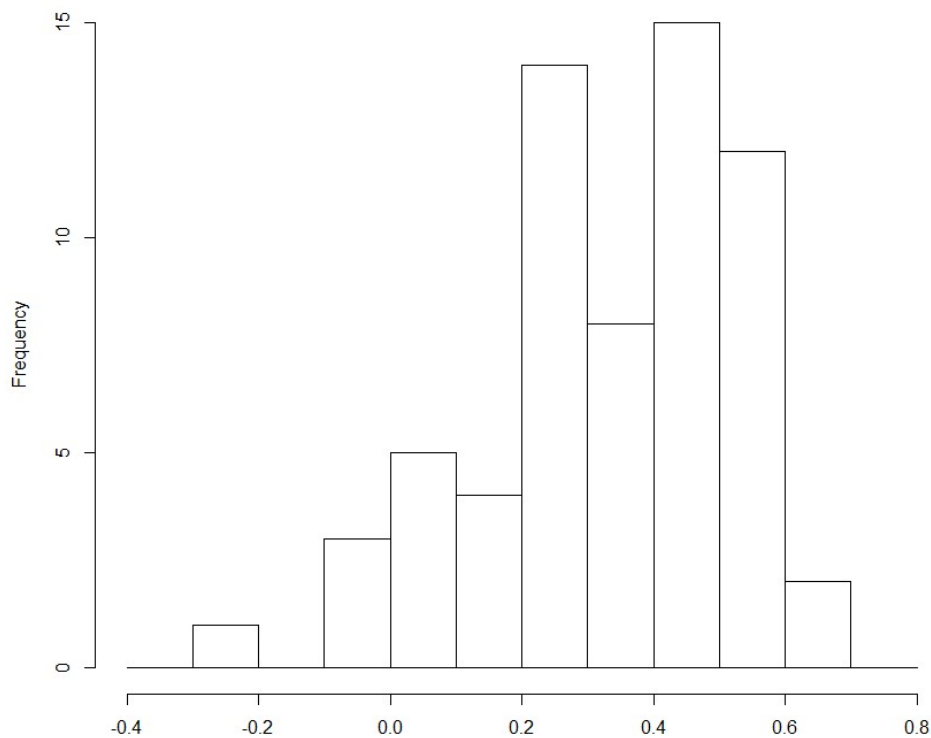


Figure 3-9: Frequency histogram of Pearson correlation coefficients of relationship between QMCI and LIFENZ for each site. For 65 sites. n = between 17 and 29 for each site.

3.2.3 Predicting within-site patterns in QMCI and LIFENZ scores

Correlations between predictors

Based on the availability of predictor variables, knowledge of mechanisms that influence within-site patterns in invertebrate communities, and correlations between potential predictors, a subset of predictors was identified to include in the stepwise multiple regressions. The same predictors were included in regressions across all sites and for regressions with both QMCI and LIFENZ as the response. The predictors included in all models were:

- Log10 nearest prior monthly nitrate-nitrite N concentration
- Log10 nearest prior monthly DRP concentration
- Nearest prior monthly spot water temperature
- Log10 maximum flow in the 30 days preceding invertebrate sample collection
- Log10 days accrual since a flow > three times the median flow
- Annual min annual 7-day low flow for year in which invertebrate sample was collected

While I attempted to select predictor variables that were unlikely to be highly correlated, it was inevitable that some correlations would occur across the 65 sites. When interpreting the results of the regression analyses the following common correlations between predictors were identified:

1. Nitrate and DRP concentrations were positively correlated ($r > 0.4$) at 36% of the sites.
2. Dissolved nutrient concentrations were correlated with hydrological indices and/or water temperature at several sites. Changes in river flow and seasonal patterns in biogeochemical patterns can also cause nutrient concentrations to vary widely over shorter time periods. Dissolved inorganic nitrogen concentrations in many New Zealand rivers often show a seasonal decline in late summer when uptake by periphyton is highest. This pattern of lower nitrate concentrations when water temperature was higher was observed in just over a third (35%) of sites. Nitrate concentrations are also commonly higher when river flows are higher. In nine of the NRWQN sites nitrate concentrations were positively correlated ($r > 0.4$) with the maximum flow in the previous 30 days.
3. Mean days of accrual and maximum flow in the 30 days prior to sample collection were significantly and negatively correlated at almost all sites (average $r = 0.58$, 90% of sites $r > 0.4$). The individual terms were retained in models for different sites, but for most sites their influence should be considered together, i.e., a longer accrual period also means a lower maximum flow prior to invertebrate sample collection.

Graphs of correlations between predictors are in Appendix B for sites where within-site patterns in LIFENZ scores were predicted ($NSE > 0$). VIF values, indicating collinearity of parameters retained in final models, are reported in Appendix A. No sites had $VIF > 0.5$ for any of the retained predictors.

Model selection

The stepwise procedure identified models identical or close to those ranked as the best models in the bestglm procedure. For sites where the model predictive power (as indicated by NSE) was relatively low, some differences in models identified by the bestglm and stepwise procedure occurred but in general both methods resulted in similar final models. Results reported here are from the backwards stepwise selection using AIC.

Best models for predicting LIFENZ

Addition of the temporal autocorrelation structure did not improve the fit in any of the models and the autocorrelation structure was therefore excluded from final models.

At 28 sites (44%) LIFENZ models had some predictive power ($NSE > 0$) using the predictor parameters included in the models (Table 3-3). Models predicting QMCI had some predictive power ($NSE > 0$) at 34 sites (54%; Table 3-3).

At 13 sites only the LIFENZ model had predictive power, while at 19 sites only the QMCI model had predictive power. At 15 sites both QMCI and LIFENZ models had predictive power. See Appendix A for summary statistics and parameters retained for sites where NSE for LIFENZ models was > 1 .

The R^2 of the relationship between observed values and those predicted by hold-one-out cross validation is a more conservative measure of model predictive power than NSE. Using the criteria of a positive R^2 value, there was some predictive power for both LIFENZ and QMCI in 86% of sites. The two metrics both had positive R^2 at the same 46 sites (71 %) and each had positive R^2 at eight (12 %) different sites.

Table 3-3: Summary statistics for stepwise regressions predicting within-site patterns of LIFENZ and QMCI across 65 sites. A positive Nash Sutcliffe Efficiency (NSE) was used to indicate that a regression model for a particular site and invertebrate metric had predictive power. NSE ranges from $-\infty$ to 1. A value of 1 indicates perfect model fit, 0 indicates model predictions are as accurate as the mean of the observed data and negative values indicate that the mean is a better predictor than the model. The number of sites with $R^2 > 0$ for the relationship between observed and hold-one-out cross validation predicted values is a more conservative method to identify models with some predictive power.

	LIFENZ	QMCI
Number of sites with positive NSE	28 (44%)	34 (54%)
Number of sites with positive R^2	54 (86 %)	54 (86%)
Positive NSE median (range)	0.11 (0.01-0.48)	0.11 (0.02-0.40)
All sites R^2 median (range)	0.07 (0-0.49)	0.10 (0- 0.41)
% positive NSE sites retaining hydrology predictors	82 % (23)	67% (74)
% positive NSE sites retaining water temperature	50 % (14)	35% (12)
% positive NSE sites retaining nutrient predictors	64 % (18)	59% (20)
% positive NSE sites retaining only hydrology predictors	18% (5)	29 % (10)
No sites (percentage) where DA retained	10 (36%)	12 (36%)
No sites (percentage) where Max flow retained	6 (21%)	13 (38%)
No sites (percentage) where Min flows retained	14 (50 %)	8 (24%)

3.2.4 Do site characteristics influence LIFENZ performance?

For sites where models of within-site patterns in LIFENZ and QMCI scores had some predictive power (positive NSE values), I investigated whether REC Source-of-flow categories (lake, lowland, hill, or mountain) influenced the amount of variance explained. For LIFENZ, models for sites in lake-fed rivers explained more within-site variation in LIFENZ than other Source-of-flow categories ($F_{3, 24} = 4.4$, $p = 0.01$, Figure 3-10). This difference between Source-of-flow categories was not observed for QMCI ($F_{3, 30} = 0.9$, $p = 0.47$, Figure 3-10).

QMCI models for sites with high upstream pastoral land cover explained more temporal variation than for sites with low upstream pastoral land cover ($F_{1, 32} = 4.6$, $p = 0.04$, Figure 3-11). This did not occur for LIFENZ ($F_{1, 26} = 0.3$, $p = 0.6$, Figure 3-11).

Sites where within-site patterns in LIFENZ or QMCI scores were able to be explained by changes in water temperature, dissolved nutrients and antecedent flow conditions did not vary significantly in upstream pastoral land cover, catchment area or conductivity from sites where within-site patterns in LIFENZ and QMCI were less predictable.

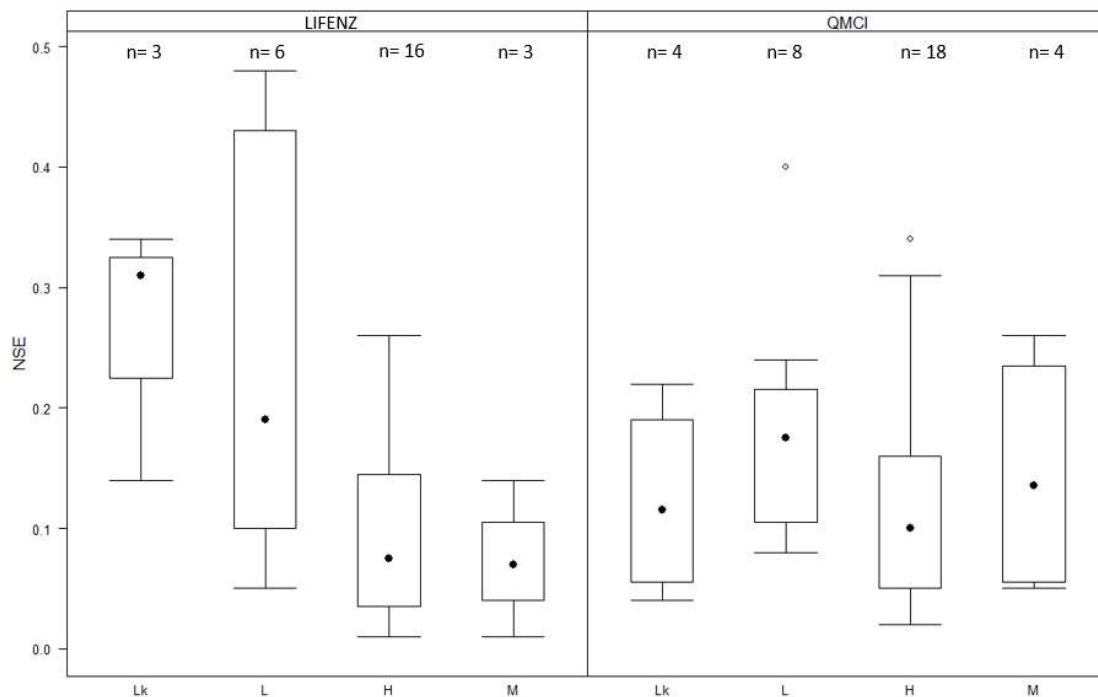


Figure 3-10: Differences between REC Source-of-flow categories in predictive power (of models that had some predictive power) for within-site patterns in both LIFENZ and QMCI scores. Predictive power is defined as a Nash Sutcliffe Efficiency (NSE) value >0. NSE was calculated during hold-one-out cross validation.

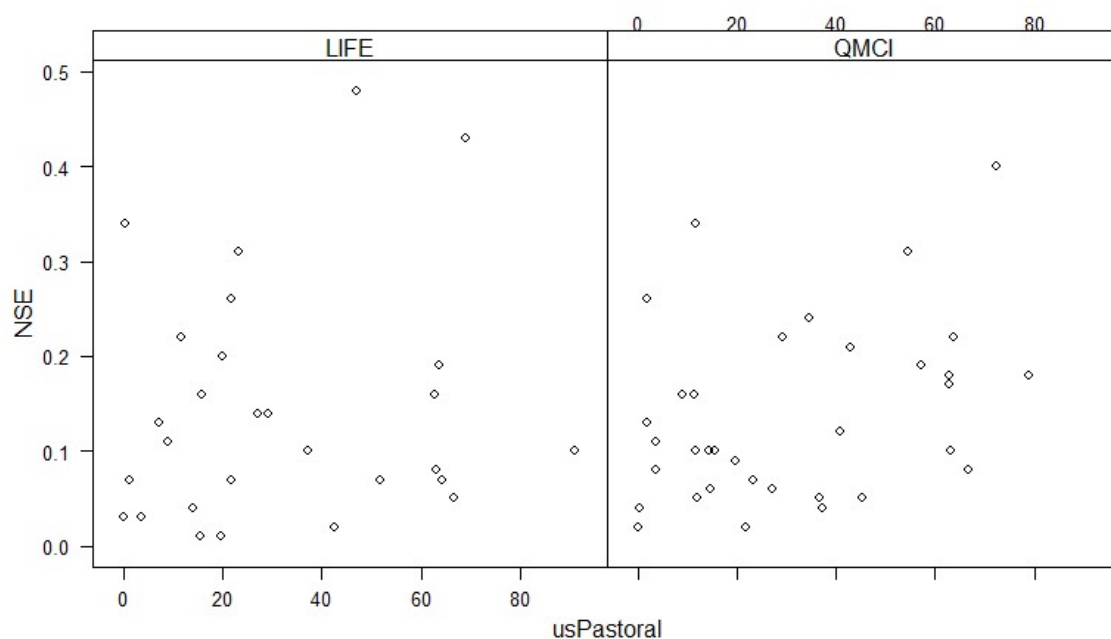


Figure 3-11: Relationship between upstream pastoral land cover and predictive power (of models that had some predictive power) for within-site patterns in both LIFENZ and QMCI scores. Predictive power is defined as a Nash Sutcliffe Efficiency (NSE) value >0. NSE was calculated during hold-one-out cross validation.

4 Discussion

Flow regulation and water abstraction are major factors influencing river systems globally (Stanford et al. 1996, Nilsson et al. 2005). Alterations to natural flow regimes can stress riverine communities and alter the magnitude and impact of other stressors such as stream-bed sedimentation or elevated nutrient concentrations (Matthaei et al. 2010). Using a macroinvertebrate metric that is sensitive to hydrological conditions, in conjunction with other stressor-specific metrics (e.g., Wagenhoff et al. 2018), will assist in disentangling drivers of poor stream health (e.g., Clews and Ormerod 2009). A flow sensitive macroinvertebrate metric may also improve our understanding of eco-hydrological relationships, which are required for setting effective river flow limits (Arthington et al. 2006, Poff et al. 2010).

For LIFENZ to be a useful addition to the suite of macroinvertebrate metrics used in New Zealand, it needs to add information not provided by the metrics in current use, respond primarily to changes in hydrology, and be relatively insensitive to other stressors.

4.1 Redundancy of LIFENZ and QMCI

As expected with community-based metrics, LIFENZ and QMCI showed some redundancy (degree of correlation) both on average between sites, and through time within sites. However, the relatively moderate strength of correlation between LIFENZ and QMCI (maximum $r = 0.68$ among 65 within-site correlations, $r = 0.62$ for the between site correlation) indicated that the metrics may provide unique information about drivers of both spatial and temporal differences in invertebrate communities. The number of sites at which potential drivers of temporal change in invertebrate communities could be identified increased from 34 sites when QMCI was used alone to 47 sites if LIFENZ was also calculated. Even when a more conservative model fit statistic was used (R^2), which resulted in a total of 56 sites with “predictive power”, the use of LIFENZ in addition to QMCI made interpretation of drivers of invertebrate community change possible at an additional 8 sites. Moreover, at sites where both metrics were able to be predicted, differences in the models for QMCI and LIFENZ should help identify stressors leading to changes in invertebrate communities (e.g., Clews and Ormerod 2009).

4.2 Is LIFENZ a hydrologically-sensitive metric?

Attributing changes in LIFENZ purely to changes in hydrological conditions within an empirical study is difficult due to the co-variation between hydrological conditions and other explanatory variables such as landcover, nutrients and temperature. Predictors other than hydrological conditions influenced LIFENZ scores both within and between sites. A high proportion of the site-specific stepwise LIFENZ regressions (82%) retained parameters relating to within-site changes in water temperature or nutrient concentrations. Between-site differences in LIFENZ scores were also associated with upstream pastoral land cover. However, LIFENZ scores did respond more strongly to hydrological indices than the other environmental parameters. In the site-specific regressions, a hydrological index was retained as a predictor for many (82%) of the sites at which temporal change in LIFENZ was predictable, while only 67% of QMCI models retained a hydrological index. A similar pattern was observed for site average LIFENZ scores, where the most significant predictors were the average number of days accrual between high flows, and parameters likely to influence local water velocity (e.g., river slope). Many invertebrate metrics appear to respond to parameters other than the stressor they were designed to detect changes in (Chessman and McEvoy 1998, Boothroyd and Stark 2000). However, like Wagenhoff et al. (2018)’s metrics, LIFENZ was most correlated with changes in the environmental conditions it was developed to detect changes in.

4.3 Which hydrological indices does LIFENZ respond to?

LIFENZ was correlated with one hydrological index between sites, but with several different indices within sites. Between sites LIFENZ scores were more correlated with the average number of days of accrual between high flows than the magnitude of floods or low flows. However, when the stepwise models for the 28 sites for which within-site patterns in LIFENZ were predictable from environmental variables were investigated there was no detectable pattern between the hydrological parameter, if any, that was retained in the model and site characteristics such as REC Source-of-Flow category.

The fact that LIFENZ did not respond to a common hydrological index across all sites is not surprising, because firstly, hydrological indices are correlated with each other and other environmental predictors and secondly, hydrological conditions interact with the geomorphological setting to create the hydraulic patterns that influence invertebrate communities. The hypothesis that the hydrological indices that influence LIFENZ would be predictable based on the site characteristics (indicated here by REC Source-of-flow categories) was tested. The component of the hydrological regime that influences invertebrate communities and hence LIFENZ at any site will vary depending on the mechanisms through which hydrology is affecting invertebrate communities at that site. Such mechanisms include high or low flows that remove or kill invertebrates (Death 1991, Arscott et al. 2010), change in food (periphyton) availability (Death and Zimmermann 2005) and/or alteration of local habitat conditions, e.g., water velocity (Greenwood et al. 2016). For any given site the magnitude of the flow that mobilises sediment, periphyton and/or invertebrates will vary depending on the geomorphology of the reach and the size and mobility of substrate. The fact that REC Source-of-flow categories could not explain which hydrological indices influenced LIFENZ may indicate that the categories are not good indicators of the parameters that influence LIFENZ-hydrology relationships, or that the hydrological indices chosen did not relate strongly to the processes that affect invertebrate communities. Antecedent flow conditions were also relatively well correlated with each other (and other environmental variables) at many of the sites, particularly days accrual since a flow event and antecedent maximum flows, making disentangling causal mechanisms of the changes in LIFENZ score difficult.

Regardless of an inability to predict which hydrological indices influenced LIFENZ at a site, LIFENZ did respond to a range of different hydrological indices. This indicates that LIFENZ may be a useful indicator of a variety of different hydrological alterations.

4.4 Site characteristics influence LIFENZ-environment relationships

While REC Source-of-flow categories did not show consistent patterns in hydrology-LIFENZ relationships, overall within-site patterns in LIFENZ were better explained in lake-fed rivers than other REC Source-of-flow categories. At sites with more steady flow regimes (e.g., lake-fed) the flow indices that were selected (days accrual, recent high flows, annual low flows) are more likely to represent the recent hydrological conditions compared with sites with more flashy flow regimes. This is partly because we used mean daily flow time-series. It is possible that hydrological indices that reflect within-day flow variability are required to better distinguish flow conditions at sites with more flashy flow regimes. Flow information with greater temporal resolution may better represent the processes that affect invertebrate communities, e.g., periphyton accrual rates or sediment mobilisation. In lake-fed rivers the flow hydrograph is moderated, and large flows occur rarely, but likely impact strongly on algal and invertebrate communities when they do occur. Likewise, under such relatively stable flow conditions, differences in water temperature and dissolved nutrient concentrations are more likely to influence periphyton biomass (Biggs 1990), which is linked with

changes in LIFENZ scores (Greenwood et al. 2016). At sites with more variable hydrological regimes large flow events occur frequently. Short accrual times between floods reduce the correlation between size of a recent event and impact on periphyton or invertebrates as communities may still be recovering from a prior event. Likewise, due to the over-riding influence of variable flow conditions, water temperature and dissolved nutrients are less likely to be correlated with periphyton growth and LIFENZ. LIFENZ is responsive to a range of different hydrological indices. However, to ensure reasonable predictive power in LIFENZ-environment relationships for any given site, hydrological indices should be chosen that are considered to relate to the mechanisms most likely to influence macroinvertebrate communities at that site. This may be possible using a method similar to Hoyle et al. (2017), who suggested that the frequency of sediment mobility at a site is likely to be a better predictor of periphyton abundance than hydrological indices similar to those used in this report.

River geomorphology may also influence LIFENZ - hydrology relationships based on site specific relationships between changes in river flow and local water velocity. The availability of local refugia, such as stable tributaries or deep pools will also influence how invertebrate communities recover from hydrological events (Resh et al. 1988). LIFENZ was designed based on taxa water velocity preferences (Greenwood et al. 2016) and for any given location flow - water velocity relationships are affected by site characteristics such as channel morphology and substrate type. For example, in general, low river flows are predicted to have impact stream invertebrate communities (Arscott et al. 2010). However, at sites where the flow is confined into a narrower bed as flow decreases it is possible that a flow reduction may increase local water velocity, at least temporarily, and lead to an otherwise unexpected increase in LIFENZ score (Gary Rushworth, pers. obs. for the UK-based LIFE). Such LIFENZ-hydrology relationships would indicate that the flow regime change is not acting as a stressor to the macroinvertebrate community in these sites. This circumstance is unlikely to occur in many rivers, and prolonged or greater decreases in flow would eventually act as a stressor on the invertebrate community and should lead to reductions in LIFENZ scores.

4.5 Does LIFENZ vary predictably between river types?

Because LIFENZ scores vary with local water velocity (Greenwood et al. 2016), sites that differ in hydraulic conditions, but have similar other environmental conditions, will naturally have different LIFENZ scores. Development of a ratio of observed LIFENZ scores relative to those expected if a site was unimpacted by hydrological alteration could allow between site comparisons (e.g., RIVPACS; Clarke et al. 2003). Expected values could be generated following existing methodology where locations are classified into groups and then expected scores for each group are generated by either identifying current unimpacted reference sites for each group (e.g., RIVPACS; Clarke et al. 2003) or by predicting expected metric values for unimpacted conditions based on current gradients of impacts (e.g., Clapcott et al. 2013).

REC Source-of-flow categories have some ability to distinguish sites based on their hydrological regime (Snelder and Booker 2013) and could be useful categories for generating expected LIFENZ scores. More pairwise differences in Source-of-flow categories were identified for LIFENZ than QMCI, and as would be expected, had higher values in categories where water velocities are likely to be higher (i.e., hill and mountain > lowland > lake-fed). Between-site differences in LIFENZ were also largely explained by hydrological indices (mean days accrual) and parameters likely affecting local water velocity (river slope) but were also affected by the network position of the site (catchment area) and upstream pastoral land cover. Any development of expected LIFENZ values should include

consideration of pastoral landcover and network position as well as differences in hydrological regime.

4.6 Suitability of the dataset

The procedures applied in collection of the NRWQN dataset used in the analyses are similar to most water quality monitoring networks in that stand-down periods for invertebrate sample collections after large flood events are in place (Scarsbrook et al. 2000). The function of this stand-down is to reduce the impact of flow variability on the collected samples and increase the chances of detecting changes in invertebrate communities caused by long-term water quality or habitat degradation. However, almost all sites in the NRWQN dataset did have invertebrate samples collected after a variety of flow conditions, ranging from years with relatively stable antecedent flows to years in which large flow events occurred in the 30 days prior to sample collection. This indicates that the dataset is not likely to limit the conclusions that can be made about LIFENZ – hydrology relationships. The dataset used in this study is probably typical of the datasets that LIFENZ is likely to be calculated from in the future, as it would be cost-prohibitive to design or modify current sampling programmes to capture both ideal water quality and hydrological alteration monitoring conditions.

Periphyton abundance and/or coverage often influences invertebrate communities (e.g., Liess et al. 2012), and has been shown to affect LIFENZ scores (Greenwood et al. 2016). However, dissolved nutrient concentrations are rarely directly correlated with periphyton cover or abundance at a site due to seasonal patterns in nutrient uptake rate and are also not often identified as affecting invertebrate metrics directly (Greenwood et al. 2016, Wagenhoff et al. 2018). Availability of periphyton data would have likely increased explanatory power for the invert metrics. A dataset that included temporal periphyton data, and that included samples collected over a full range of flow conditions, particularly after extreme hydrological conditions would likely allow better identification of the mechanisms causing within-site variation in LIFENZ.

4.7 Application of LIFENZ and recommendations

LIFENZ can be calculated using full count or coded abundance invertebrate data and for historic or newly collected datasets. Currently LIFENZ is most useful for investigating within-site changes in invertebrate communities. The calculation and evaluation of LIFENZ in conjunction with other stressor-specific metrics, particularly in rivers that have temporal periphyton and/or substrate size data, have different characteristics from the NRWQN sites (e.g., are smaller headwater sites), or have data that span large hydrological alterations is recommended. Such investigations could: a) identify sources of the potential redundancy between LIFENZ and other stressor-specific metrics; b) explain more variation both within and between sites in LIFENZ through the use of periphyton data; and c) test the generality of the relationships identified here in relatively large river sites.

Further exploration of the use of existing methods to classify sites based on hydrological regime type and then generate expected LIFENZ scores under unimpacted conditions is warranted if a between-site metric indicating the ecological impact of hydrological alteration is deemed to be useful.

Currently, and similar to the recommendations in Wagenhoff et al. (2018), LIFENZ could be used in conjunction with the nutrient and sediment specific metrics within specific sites to:

1. Assess whether nutrient enrichment, sedimentation or flow alteration individually or in combination are the likely causes of degraded stream ecosystem health

2. Estimate the ecological effects of a significant disturbance events, such as a dam failure or extended drought
3. Measure success of flow restoration efforts and the impact of flow regulation or reductions

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Appendix A Summary statistics for LIFENZ within site regressions

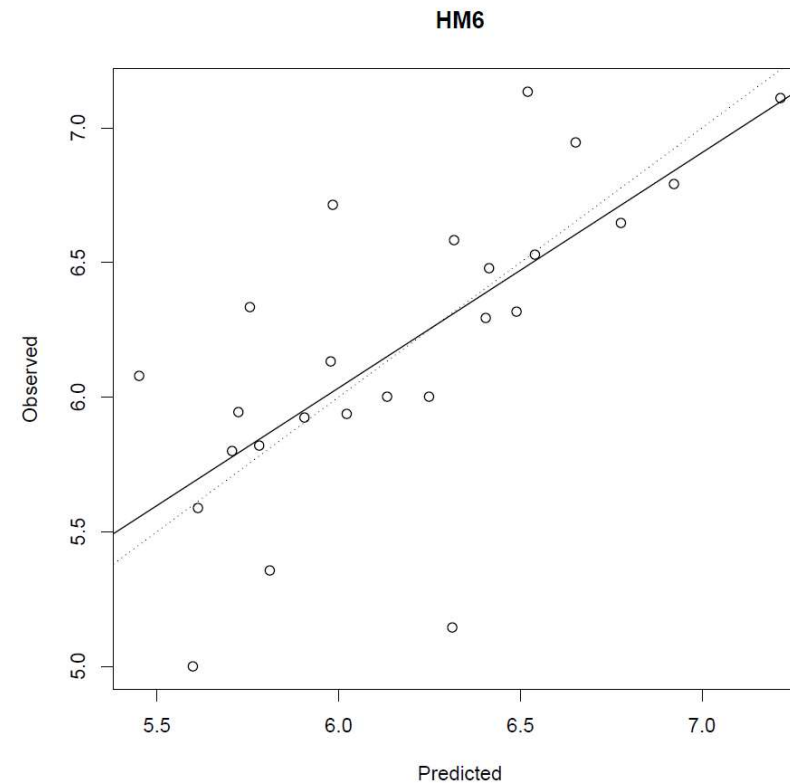
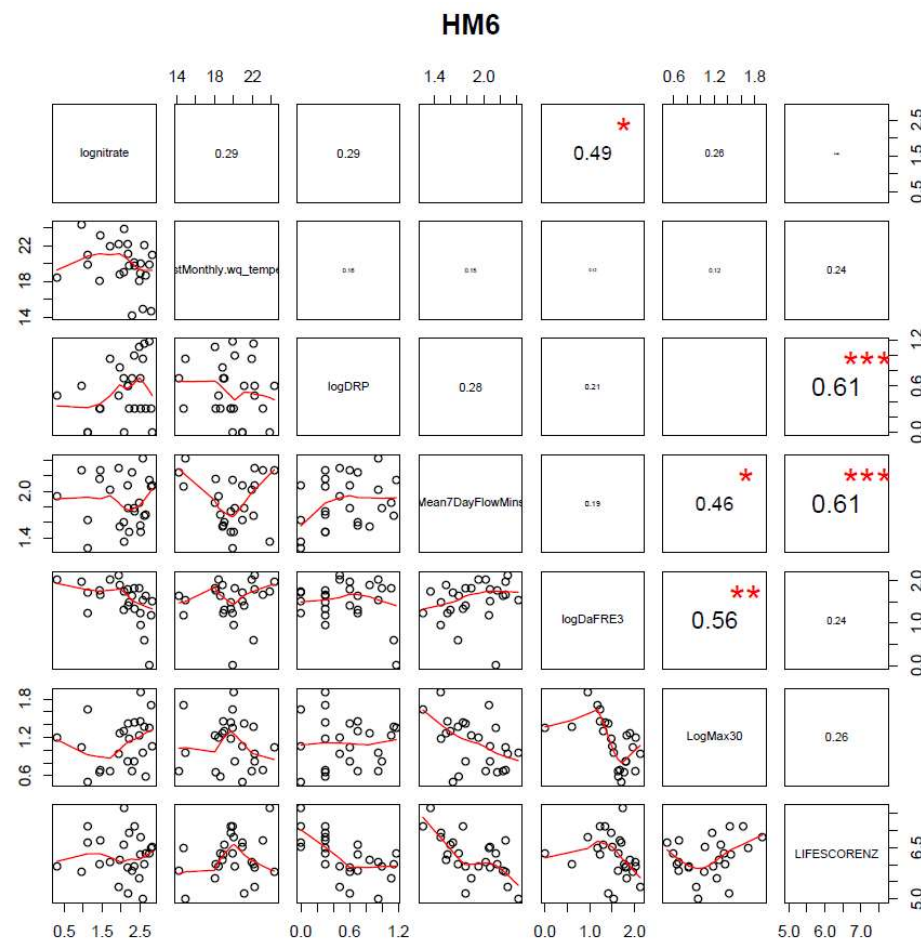
Details of sites where regressions of within-site patterns in LIFENZ had some explanatory power (positive NSE) ordered by predictive power. Nash Sutcliffe Efficiency (NSE) indicates predictive power from hold-one-out cross validation and ranges from $-\infty$ to 1. A value of 1 indicates perfect model fit, 0 indicates model predictions are as accurate as the mean of the observed data and negative values indicate that the mean is a better predictor than the model. The number of years data included in the model (n), maximum VIF for parameters and the terms retained after stepwise elimination are reported. VIF is variance inflation factor. Values above 5 for any parameter indicate collinearity. DRP = nearest monthly DRP concentration prior to invertebrate sample, Nitrate = nearest monthly $\text{NO}_x\text{-N}$ concentration prior to invertebrate sample, MaxFlows = maximum flows in the 30 days prior to the invertebrate sample, MinFlows = minimum annual 7-day low flow for year of invertebrate sample, DA = number of days since a flow greater than three times the median flow, Temp = nearest monthly water temperature spot measurement prior to invertebrate sample.

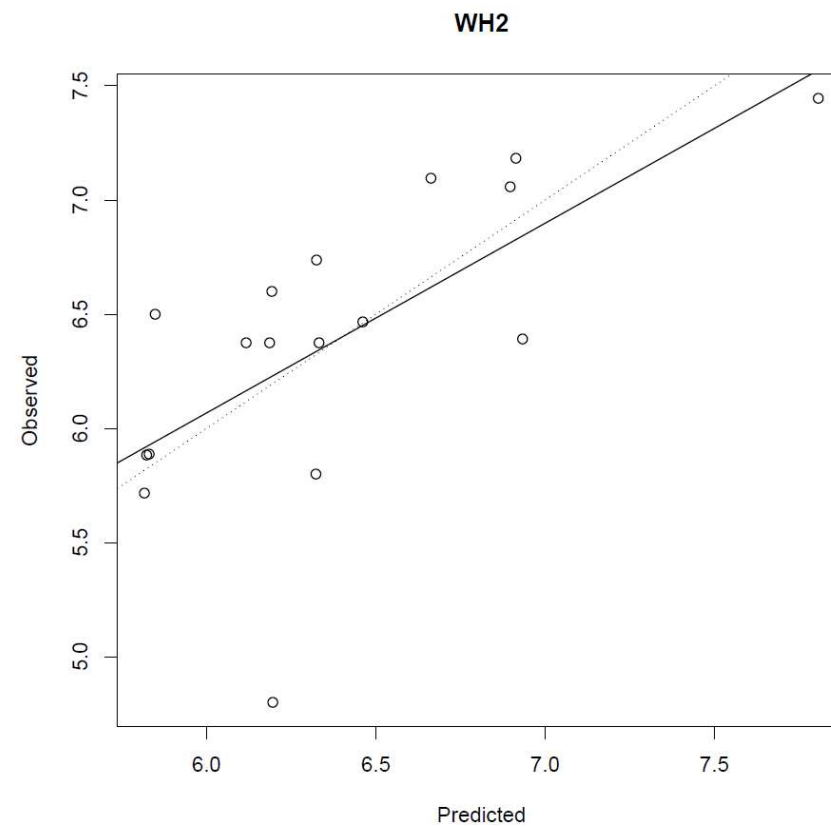
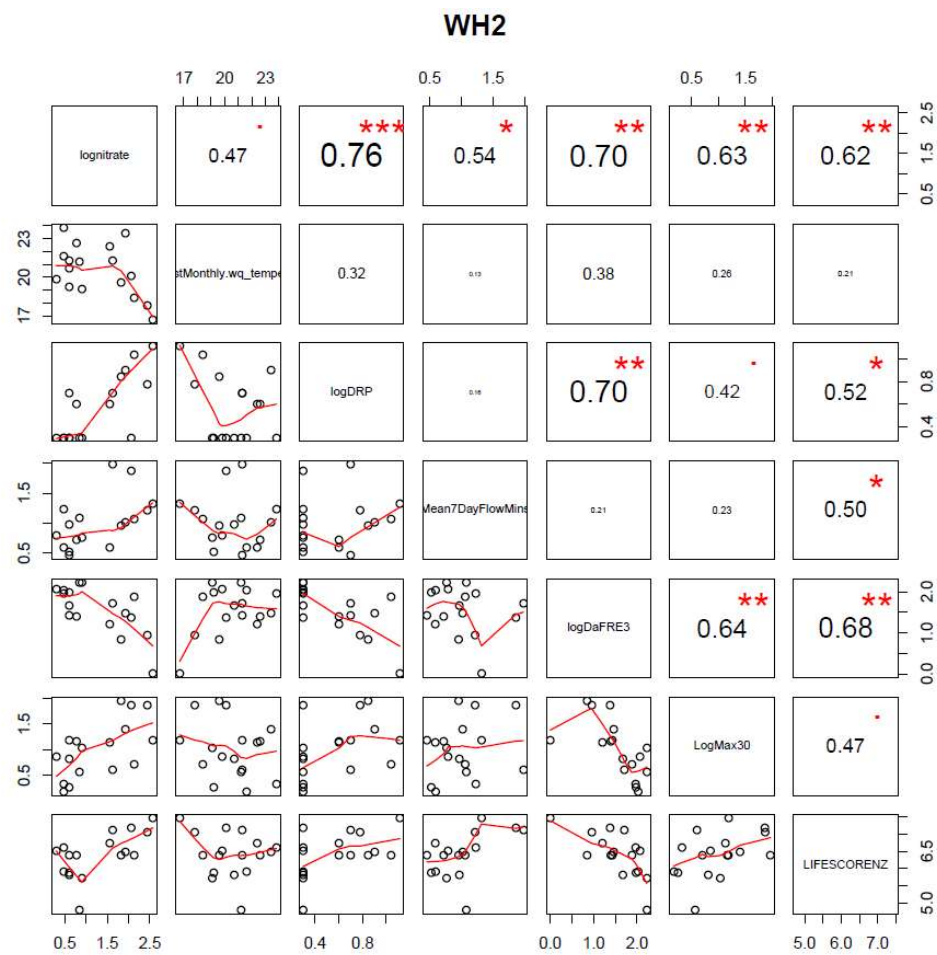
Site	Site name	n	NSE	Max VIF	Terms retained
HM6	Ohinemuri at Karangahake	25	0.48	1.3	DRP, MinFlows, DA
WH2	Waitangi at Wakelins	17	0.43	1.1	MinFlows, DA
DN10	Monowai at Below Control	25	0.34	2.9	Nitrate, MinFlows, Temp
TK4	Waitaki at Kurow	24	0.31	1.1	Nitrate, DRP, Temp
WA2	Manganui at SH3	26	0.26	1.1	DRP, MinFlows, MaxFlows, DA, Temp
WN1	Hutt at Boulcott	26	0.22	2.5	DRP, MinFlows, MaxFlows, DA
GS4	Motu at Houputo	24	0.20	1.8	Nitrate, DRP, Temp
DN3	Taieri at Outram	23	0.19	1.5	MaxFlows, Temp
DN5	Mataura at Seaward Downs	26	0.16	1.0	Nitrate, MaxFlows
RO5	Rangitaiki at Te Teko	19	0.16	4.2	Nitrate, DRP, MinFlows, DA, Temp
AX4	Clutha at Clutha @ Mill	21	0.14	1.0	MaxFlows, Temp
CH4	Waimakariri at Old Highway	28	0.14	1.1	DA, Temp
GY1	Buller at Te Kuha	22	0.13	1.1	Nitrate, DRP, DA
NN5	Buller at Longford	29	0.11	NA	MinFlows
WA3	Waingongoro at SH45	24	0.1	NA	DA
TU1	Whanganui at Te Maire	25	0.1	1.2	Nitrate, MinFlows, Temp
GS3	Motu at Waitangirua	27	0.08	2.2	Nitrate, DRP, Temp
HV4	Ngaruroro at Kuripapango	26	0.07	1.0	MinFlows, Temp

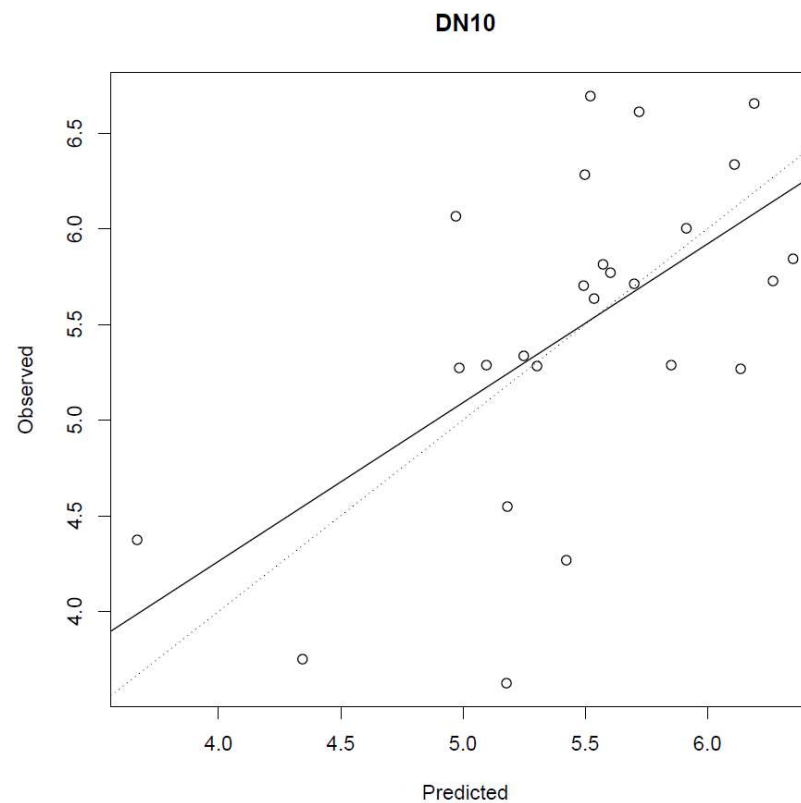
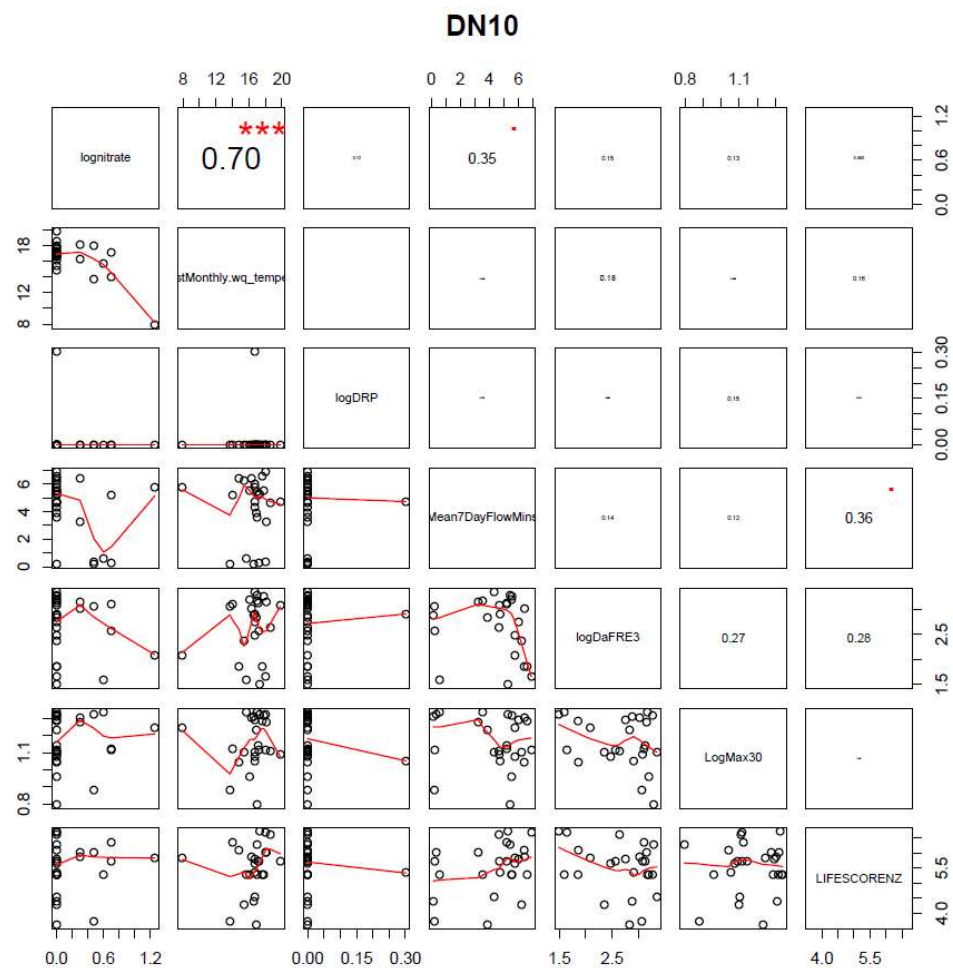
Site	Site name	n	NSE	Max VIF	Terms retained
NN4	Wairau at Tuamarina	27	0.07	1.8	Nitrate, DRP, DA
WA6	Rangitikei at Kakariki	24	0.07	1.1	DRP, MinFlows, DA
TK1	Opihi at Waipopo	27	0.07	NA	DRP
WN3	Ruamahanga at Waihenga	27	0.05	NA	DRP
HV5	Mohaka at Raupunga	25	0.04	NA	MinFlows
RO4	Whirinaki at Galatea	25	0.03	1.8	Nitrate, DRP, MinFlows, Temp
WN2	Hutt at Kaitoke	27	0.03	1.1	DRP, MinFlows, Temp
HM1	Waipa at Otewa	24	0.02	NA	MaxFlows
CH3	Waimakariri at Gorge	28	0.01	1.7	MinFlows, Temp
NN1	Motueka at Woodstock	29	0.01	1.0	MinFlows, DRP

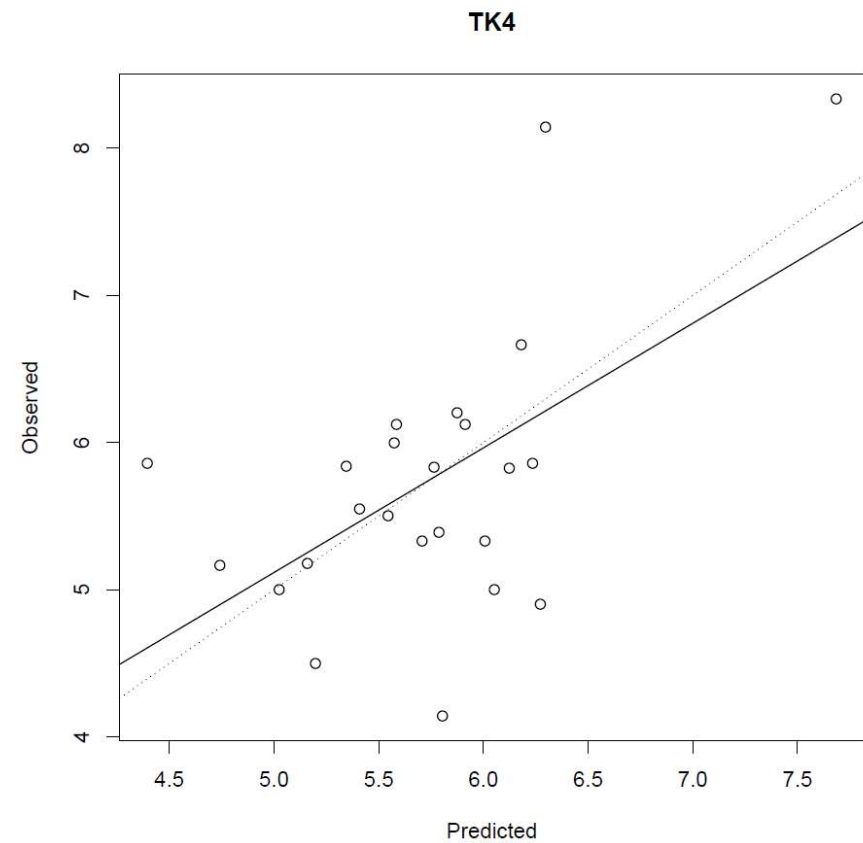
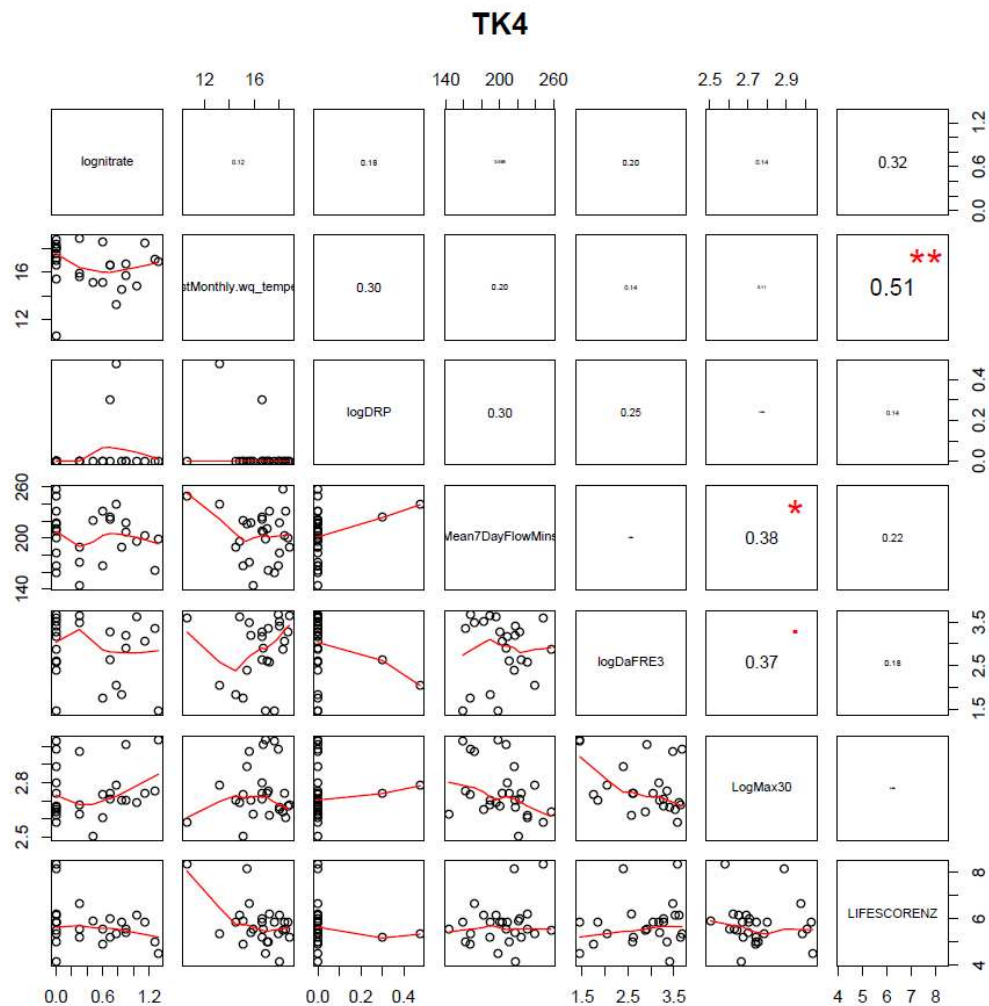
Appendix B Predictor correlations and cross-validation plots for sites with predictive power for within-site variation in LIFENZ

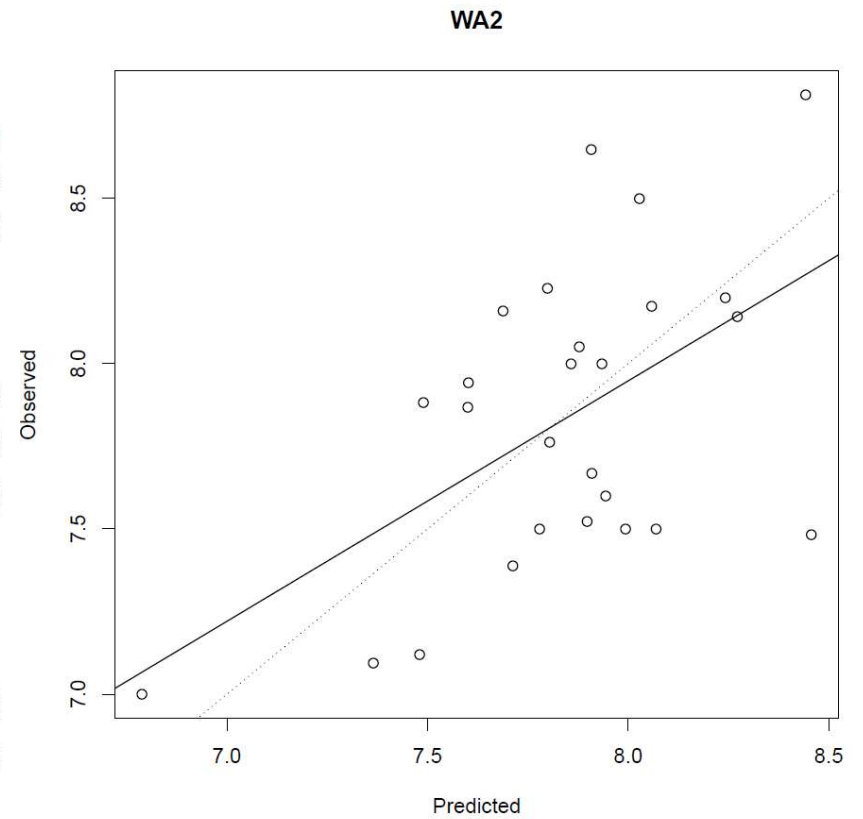
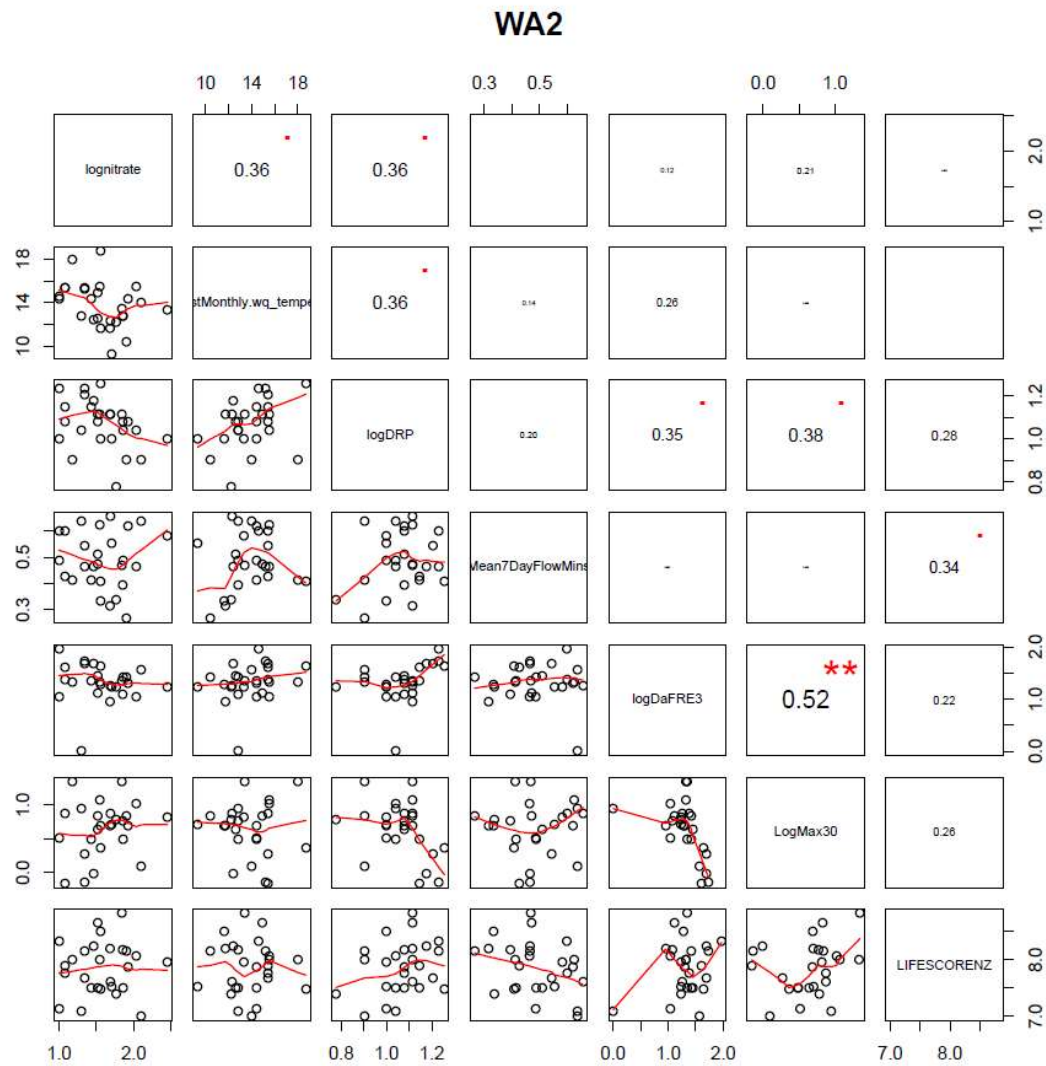
For each site with a regression model that can predict some within-site variation in LIFENZ (NSE > 0.1) the following plots are shown: 1) pairwise correlation plots of predictors included in the regression and 2) the hold-one-out cross validation plots showing predicted and observed LIFENZ scores. Sites are ordered from higher predictive power to lower. See Appendix A for regression summary statistics including NSE values.

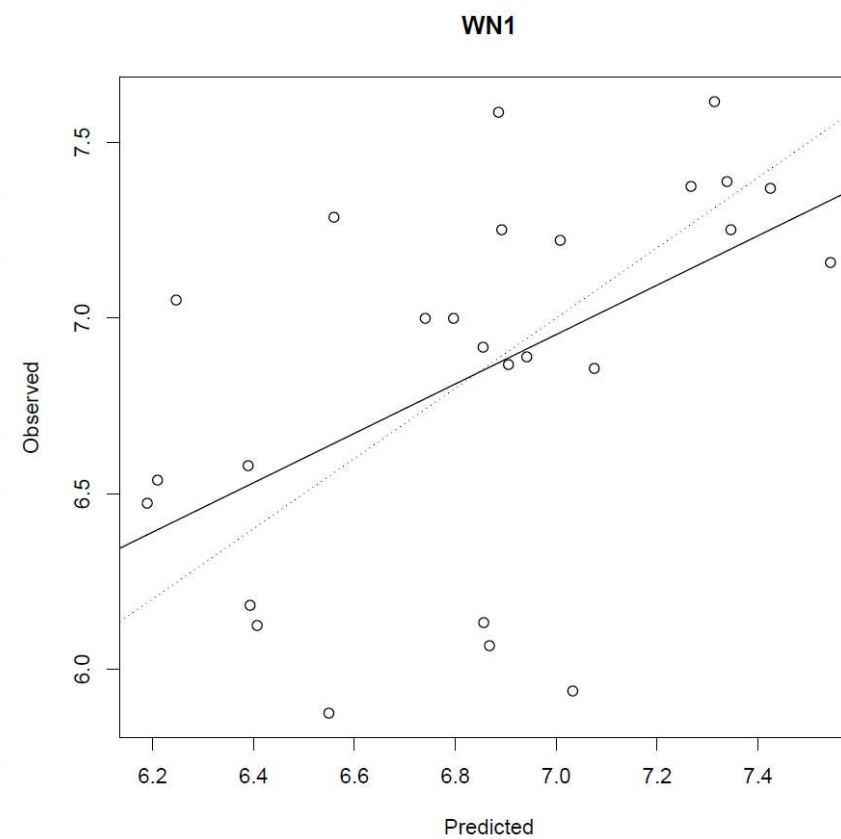
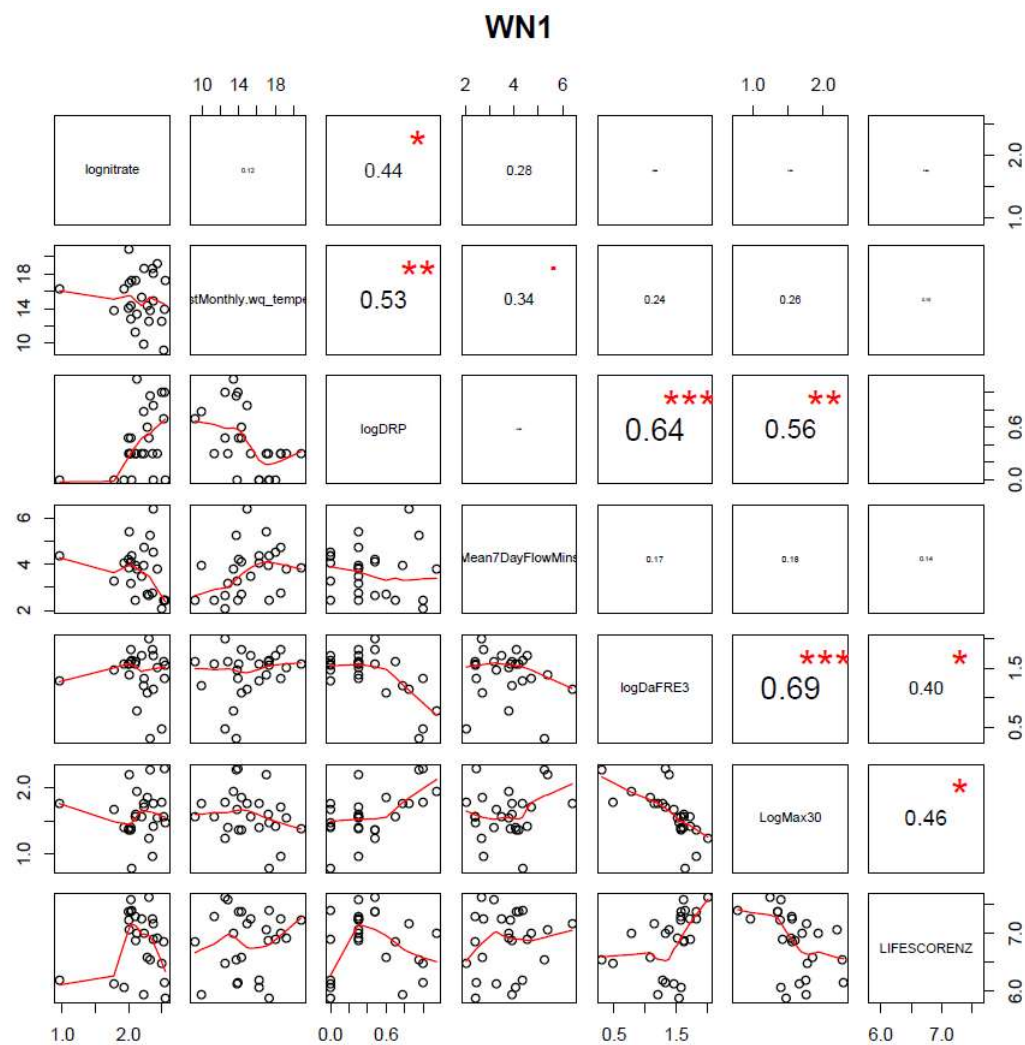


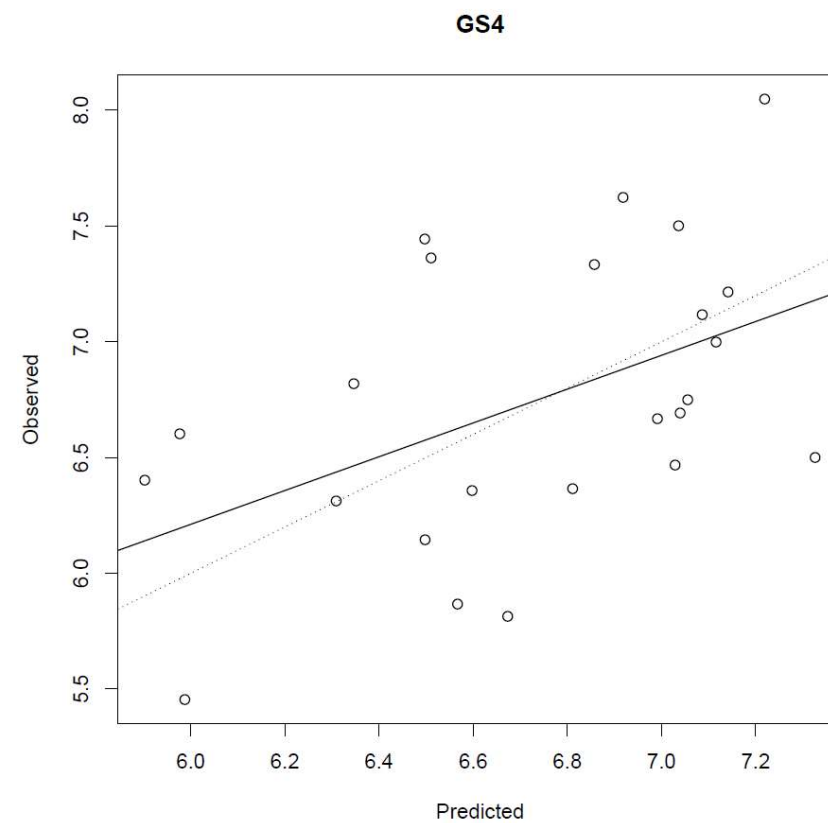
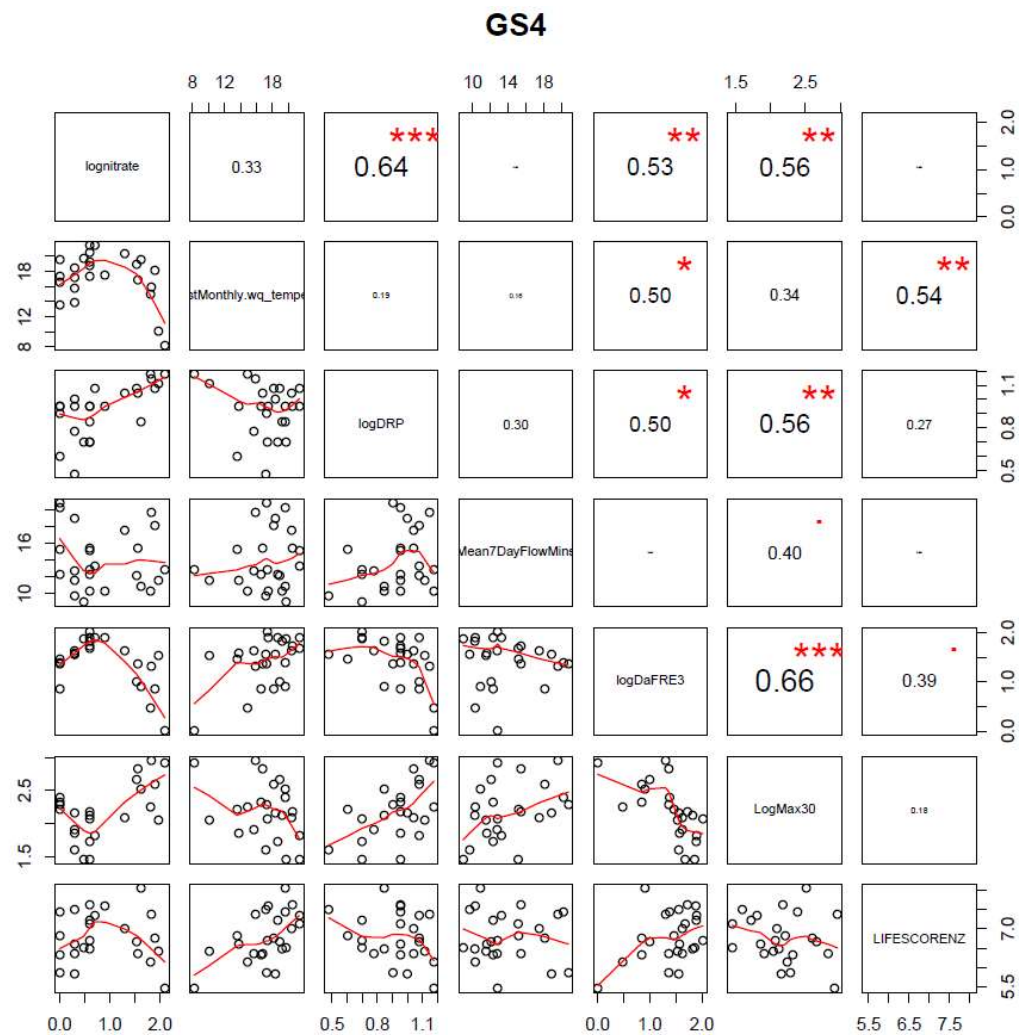


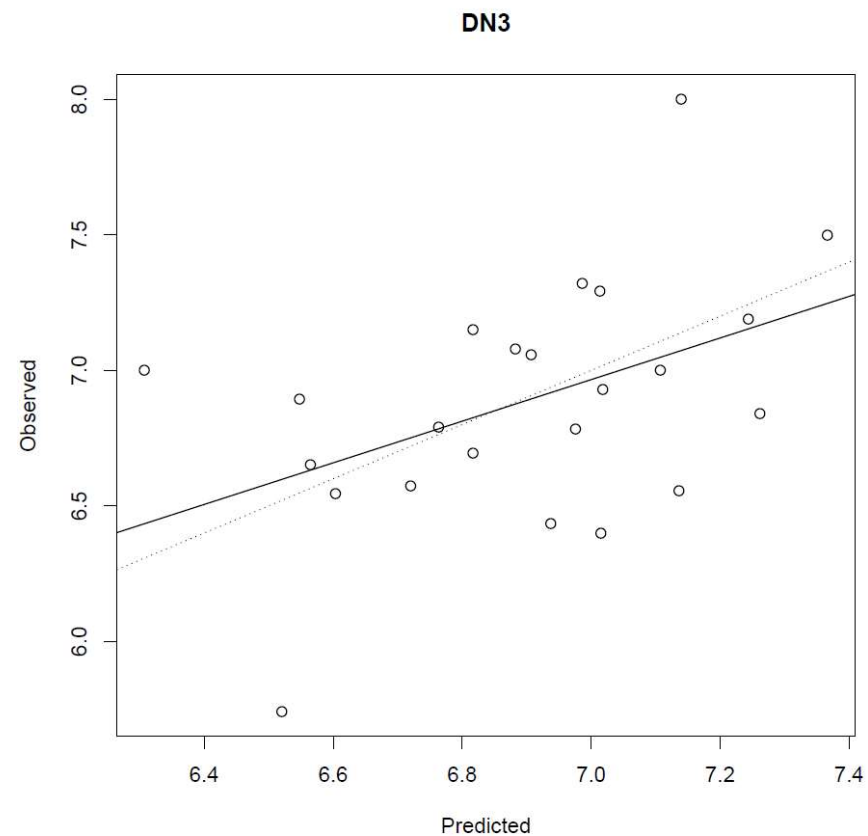
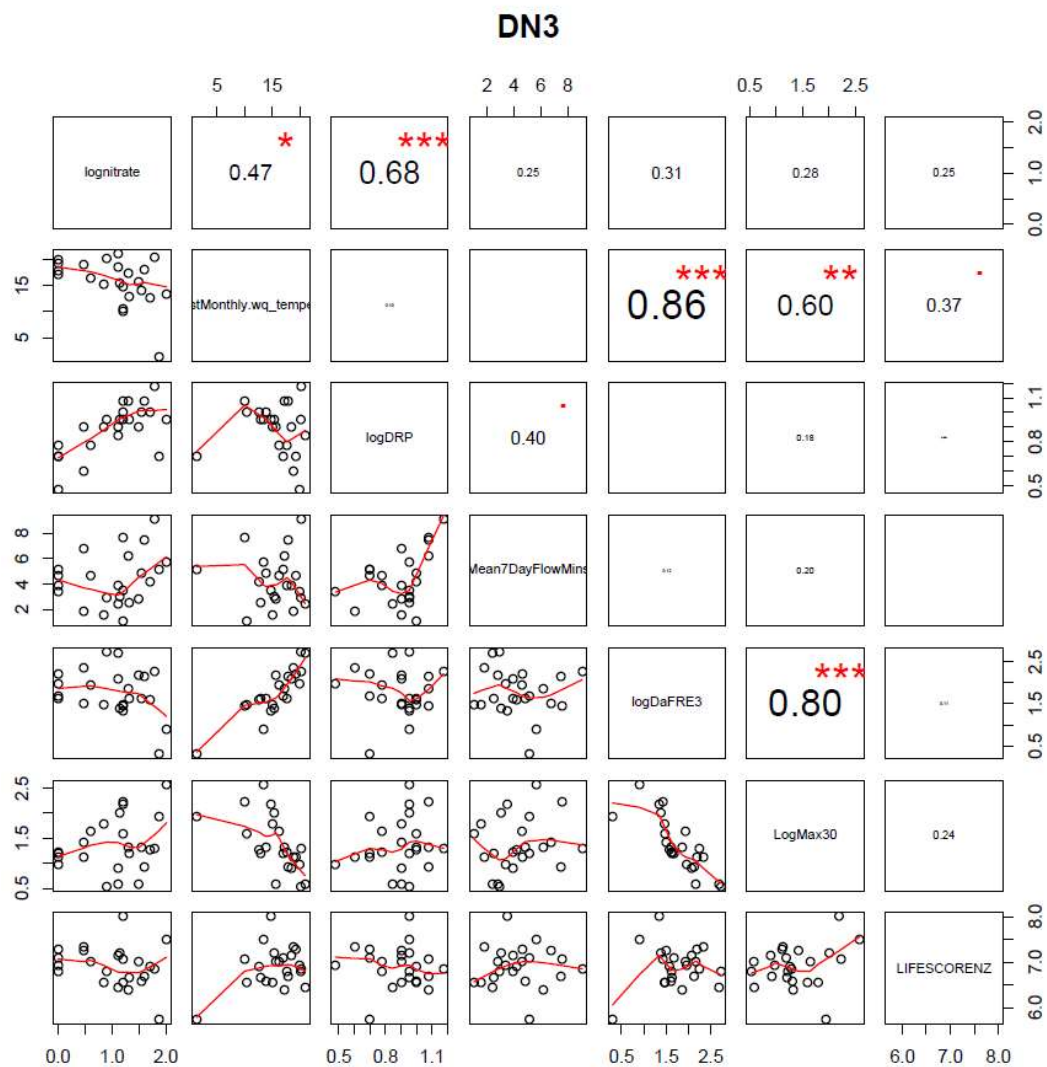


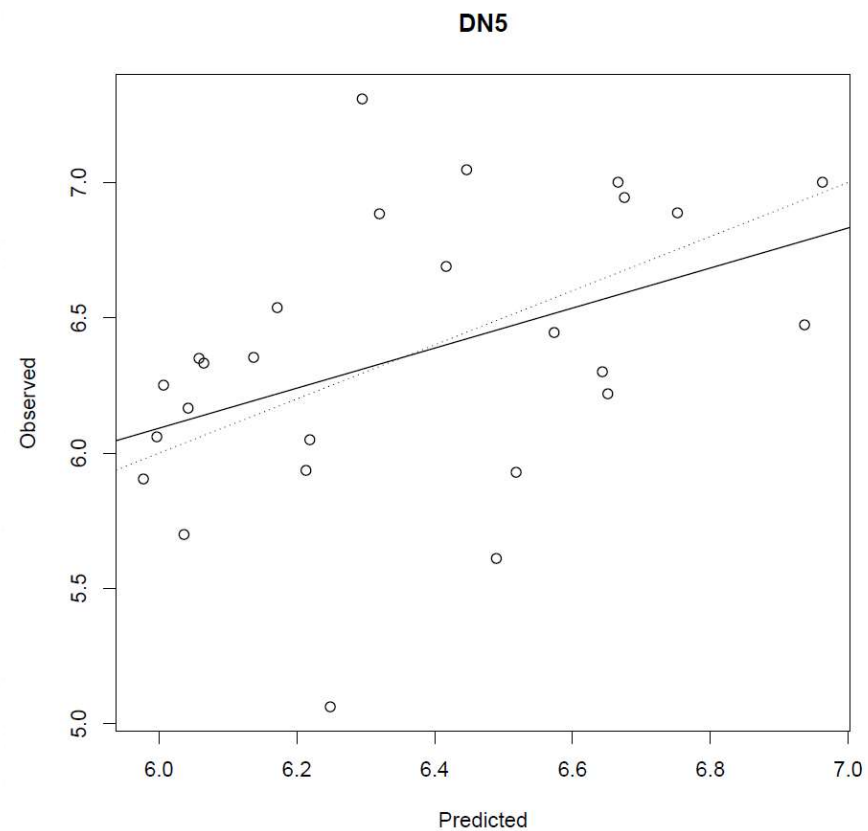
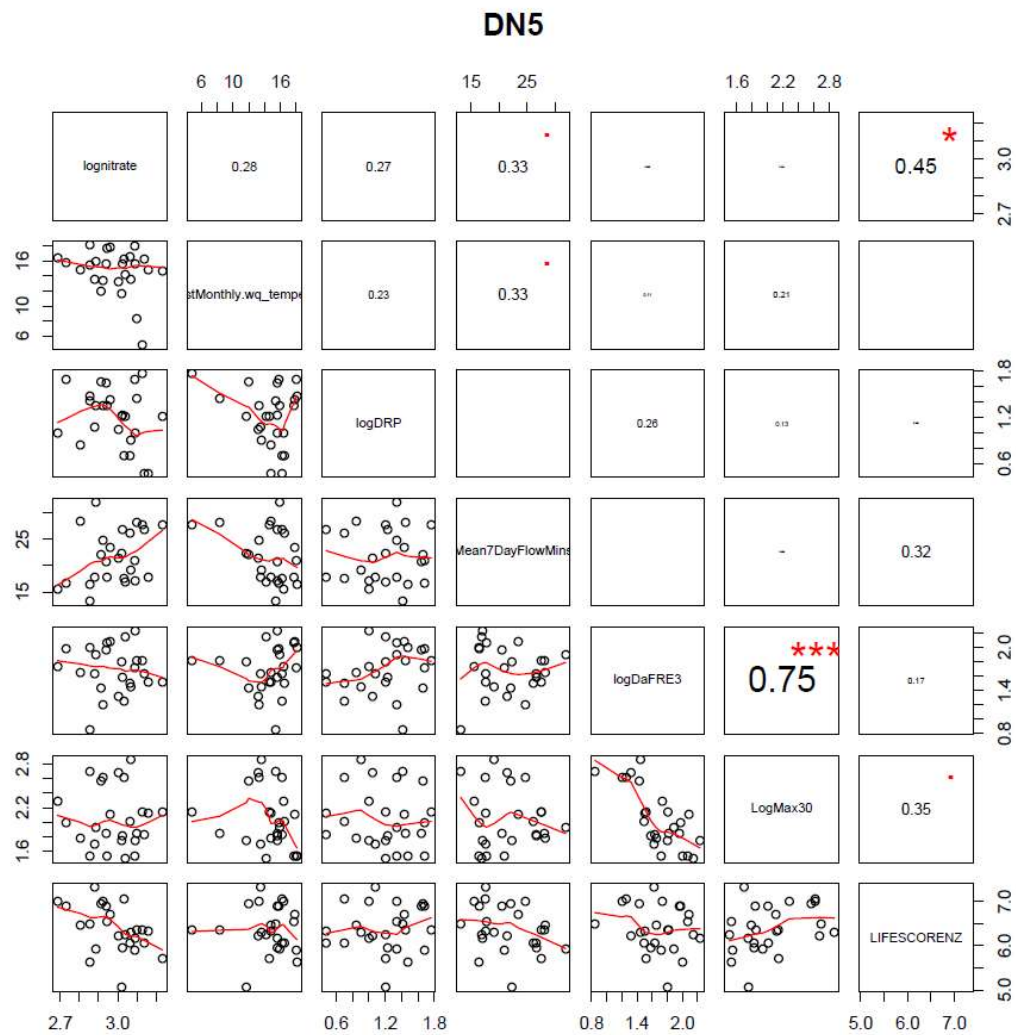


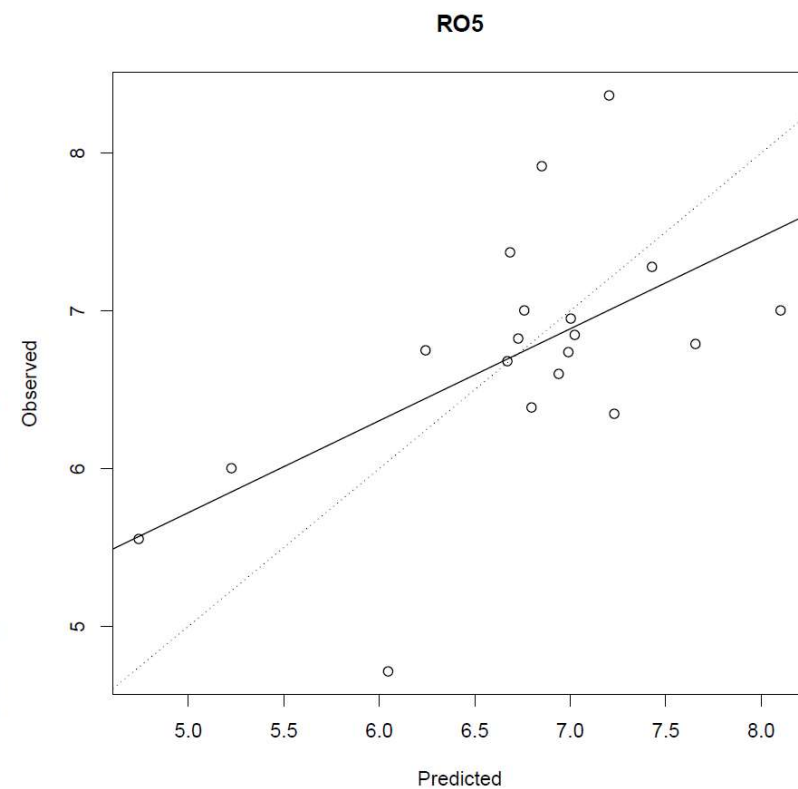
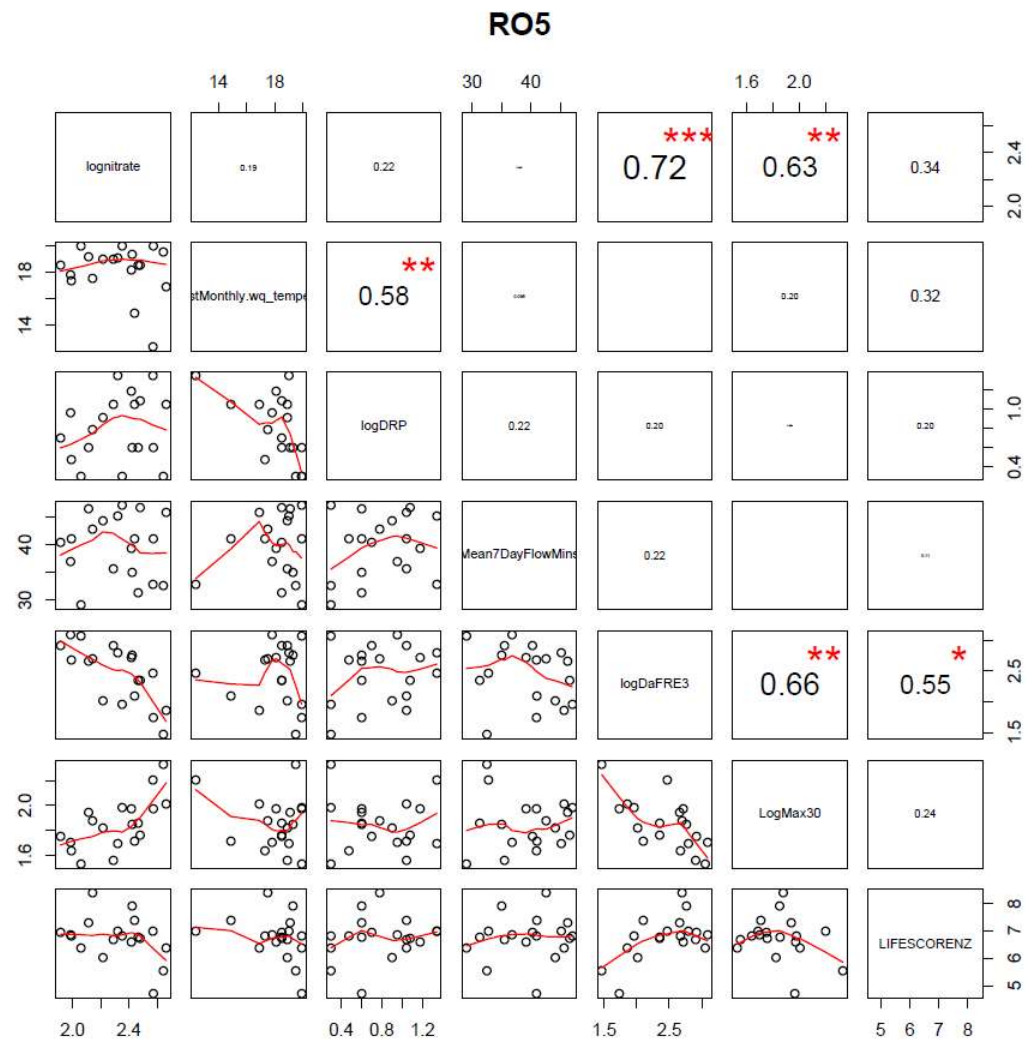




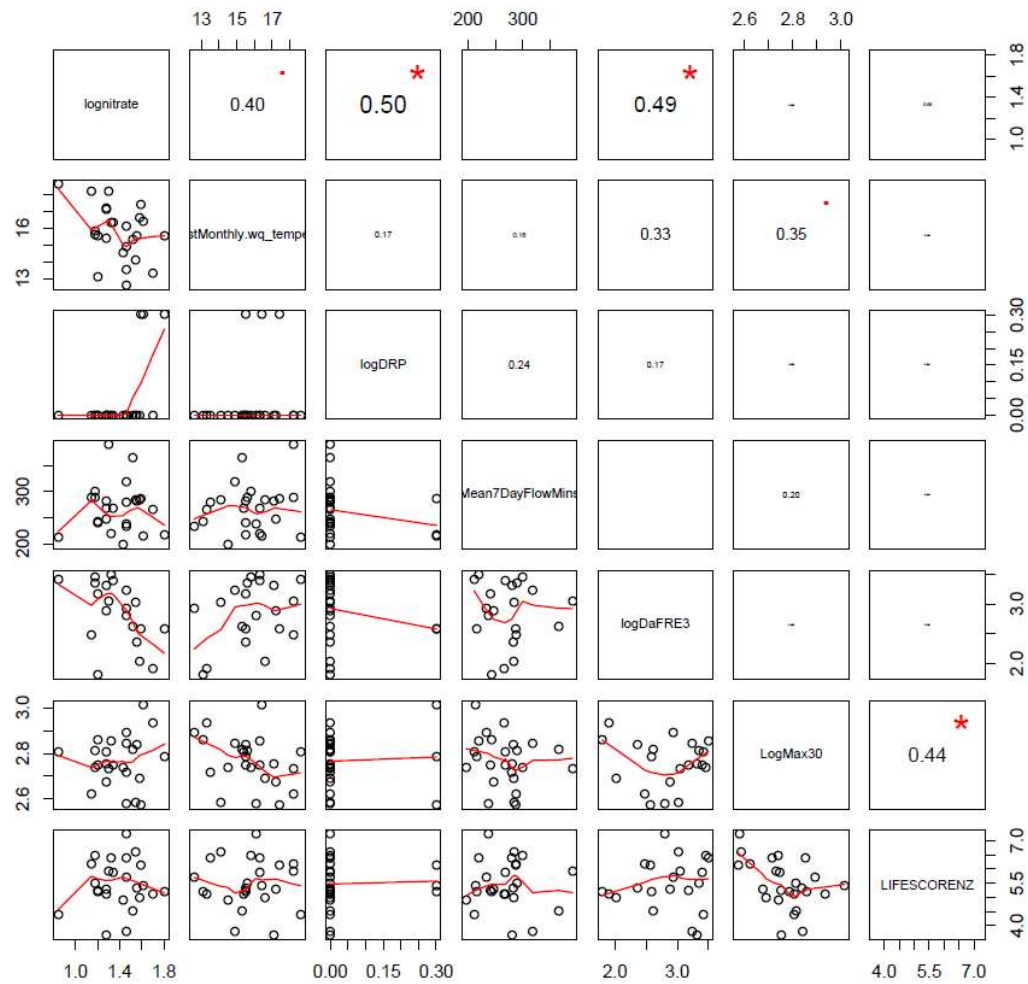




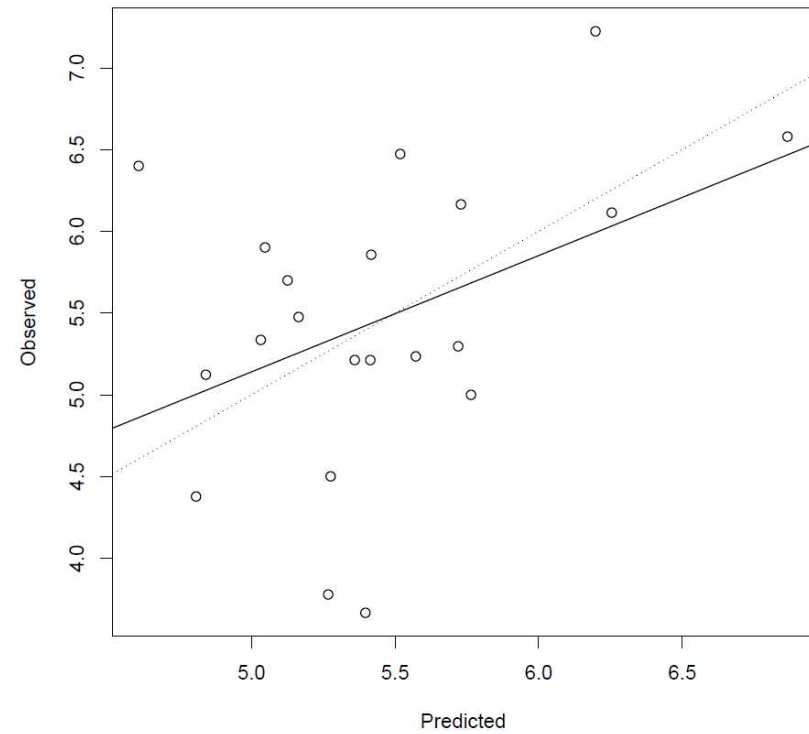


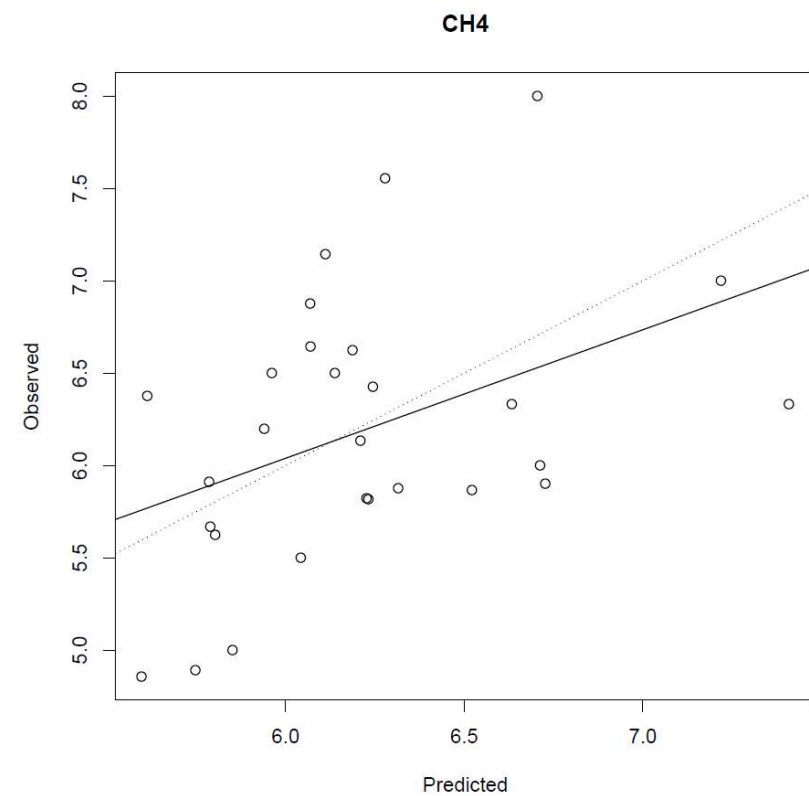
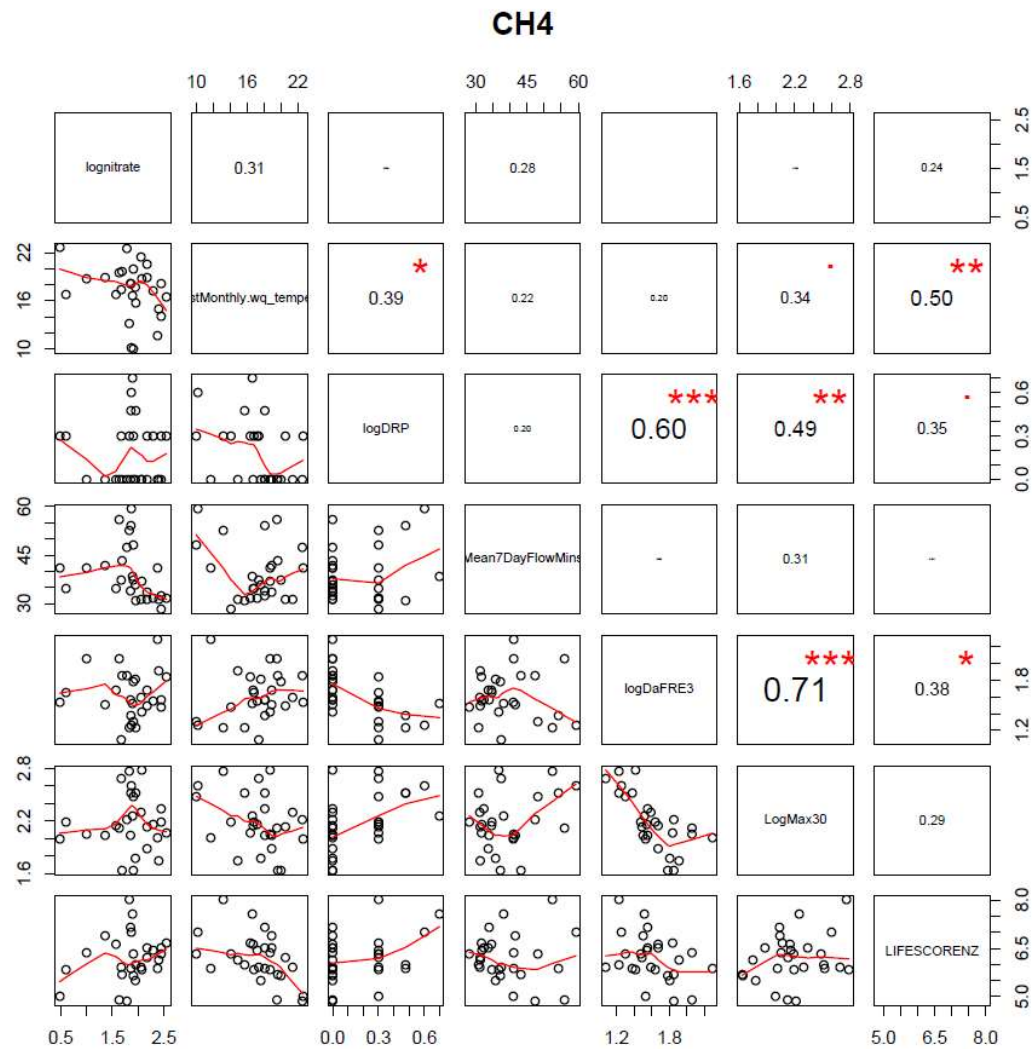


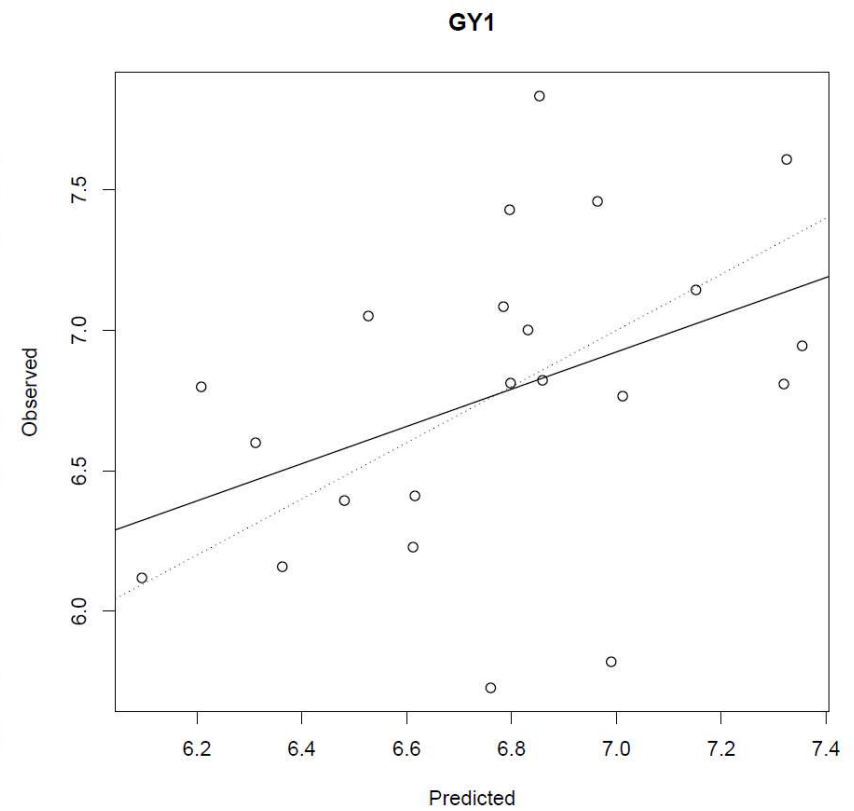
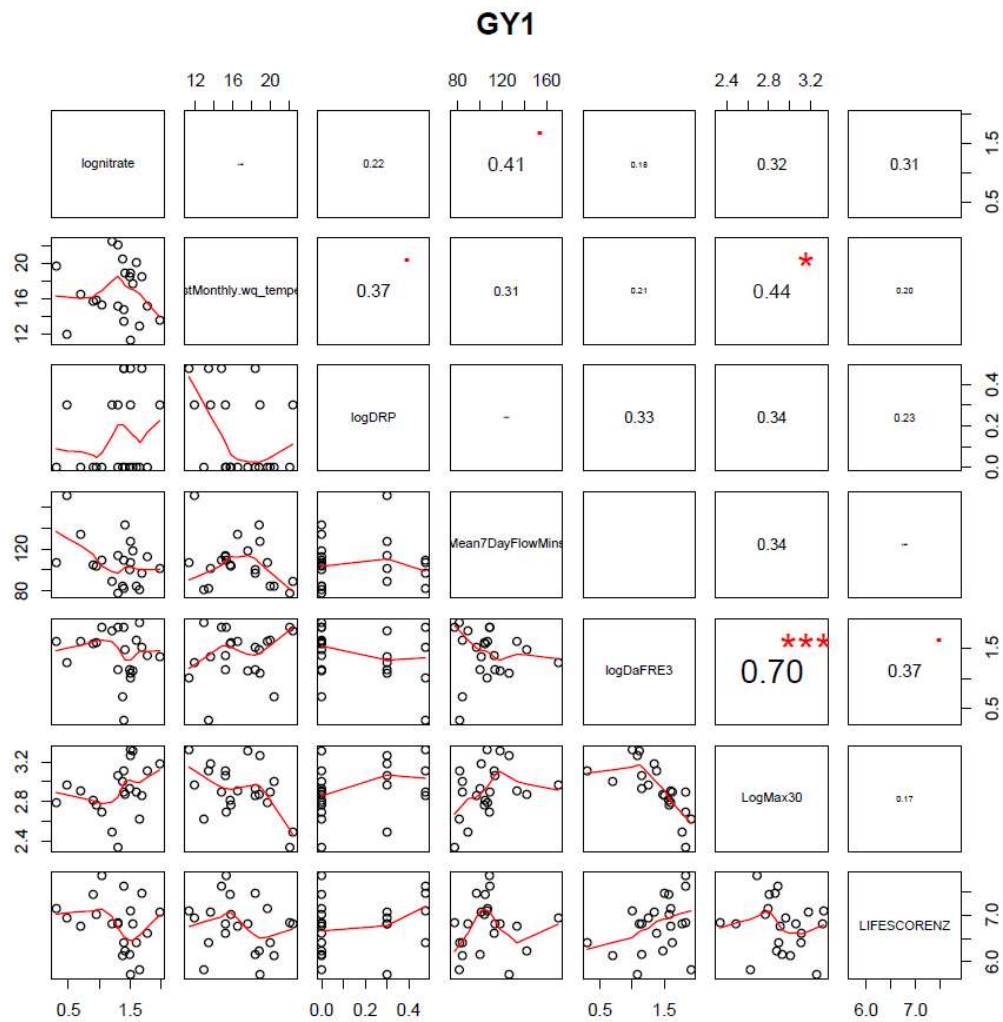
AX4

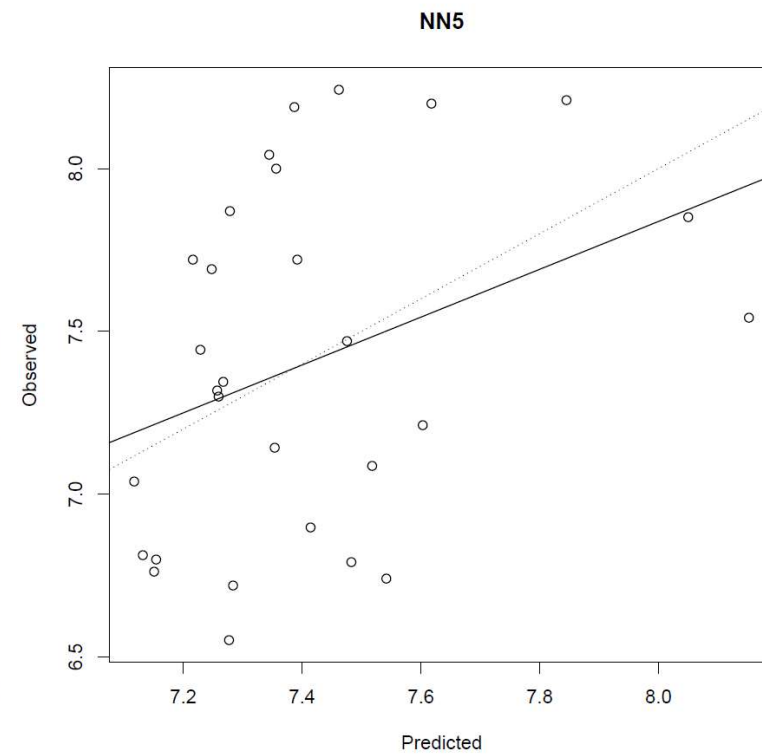
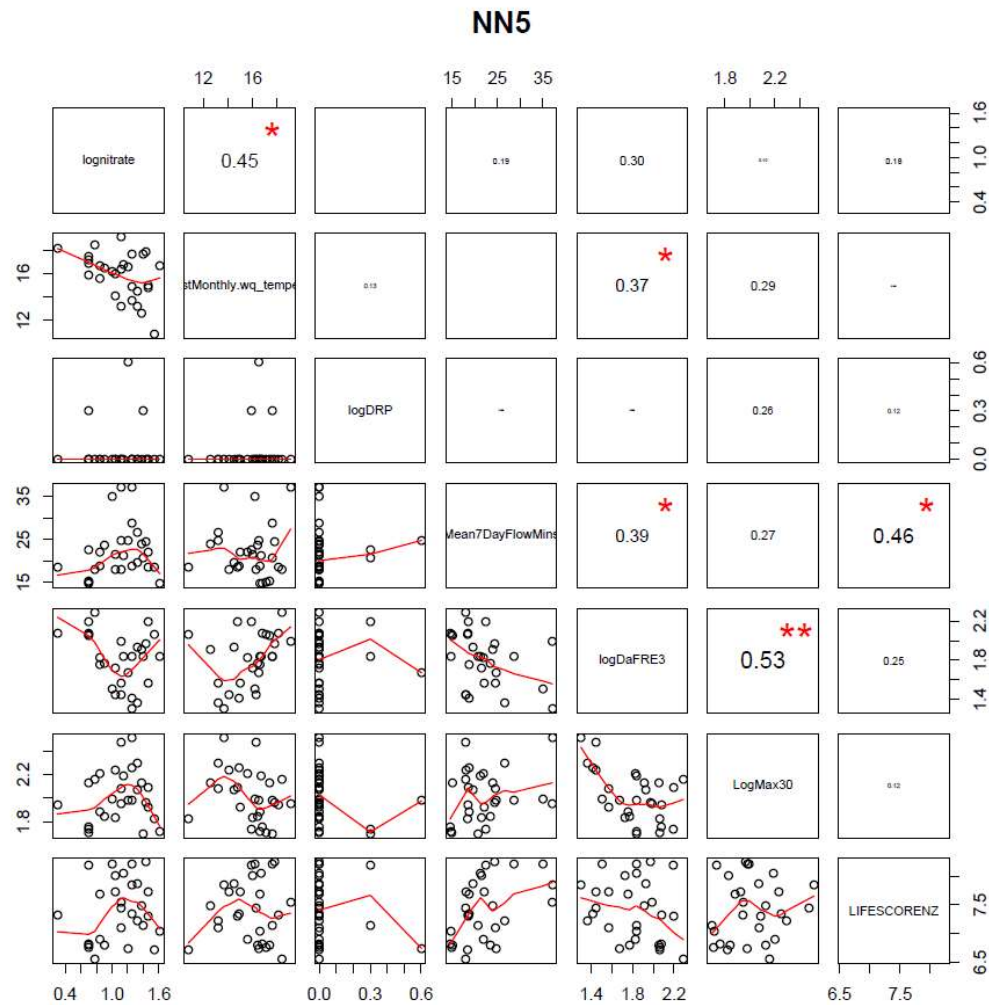


AX4









Appendix C Temporal patterns in flow, nitrate, QMCI and LIFENZ

This appendix includes plots for each site showing temporal patterns in mean daily flow, nitrate concentrations, QMCI and LIFENZ over the period for which mean daily flows were available.

