Nutrient Assessment Protocols for Wadeable Streams in Nevada

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Algae at East Fork Carson River between Highway 88 and Muller Lane, July 2004

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Introduction

The purpose of this document is to provide some general guidance on possible steps for determining the nutrient impairment status of wadeable streams in Nevada. These protocols will be useful in addressing a two key issues:

> **303(d) List:** Of the some 100 waterbody reaches listed in 2004 303(d) List, nearly 50% are shown to have exceedances of total phosphorus standards. However, NDEP is not confident in the appropriateness of the phosphorus standards throughout the state. It is hoped that these protocols will be useful in determining whether or not these listed waters are truly impaired. Appropriate next steps (additional monitoring, water quality standards revisions, TMDL development, targeted 319 projects, etc.) can then be determined. Before a large amount of resources are devoted to developing TMDLs and control strategies, it is advisable to evaluate the eutrophication status of 303(d) Listed waters using these protocols.
>
> **Appropriate Nutrient Criteria:** As described in Nevada's Nutrient Criteria Strategy (NDEP, 2007), NDEP faces significant challenges in reviewing/revising existing nutrient criteria and establishing new criteria for new waters to be included in the regulations. Nutrient levels are believed to be a poor indicator of nutrient impairment. Rather parameters such as dissolved oxygen and algae density are much better measures of stream health as affected by nutrients. Assessments, such as described in this document, will increase the state's database on nutrients, algae levels and other factors. As a result, NDEP will be better equipped to move toward more appropriate nutrient criteria throughout Nevada.

This document is to be considered a living, changing report, which will be revised over time as NDEP obtains more data, tests these protocols, and gains more insight into Nevada's waters.

Background

Exceedances of total phosphorus standards are common in many of Nevada's streams. However in many cases, it is not known if the phosphorus levels are actually impacting the beneficial uses, e.g. aquatic life, recreation, etc. As discussed by TetraTech (2005), the use of nutrient concentrations alone are poor predictors of assessing eutrophication impacts. Also, Dodds et al. (2002) examined data from over 600 streams and found that nutrients concentrations accounted for less than half of the variance in the benthic algae biomass. They speculated that other factors, such as flow, light availability, channel conditions, grazing, were responsible for the remaining variability. In a detailed study of Colorado streams, Lewis, Jr. and McCutchan, Jr. (2005) found even less of a relationship between nutrient concentrations and benthic biomass, with DIN (dissolved inorganic nitrogen) accounting for only 15% of the variance. No statistically significant relationship was found between benthic biomass and other nitrogen and phosphorus species.

Given the problems of relying on nutrients concentrations to predict impairment, EPA Region IX RTAG (Regional Technical Advisory Group) has recommended the use of secondary indicators in determining impairment status (Tetra Tech 2006). Key indicators for streams include: 1) benthic algal biomass; 2) dissolved oxygen; and 3) pH (photosynthesis driven). It is believed that these three parameters are more direct indicators of stream use support/impairment status than N and P concentrations.

Of these three indicators, algae is the primary driver. While some algae is a necessary component of the ecosystem, excessive algae can impact the beneficial uses in a variety of ways. According to EPA (2000):

> *"Algae are either the direct or indirect cause of most problems related to excessive nutrient enrichment, e.g. algae are directly responsible for excessive, unsightly periphyton mats or surface plankton scums, and may cause high turbidity, and algae are indirectly responsible for diurnal changes in DO and pH"*

Secondary Indicator Thresholds

Algal Biomass: Table 1 presents a summary of various algal biomass limits that have been recommended by others for the protection of various uses. Suggested maximum chlorophyll-a densities range from 50 to 200 mg/m^2. Biggs (2000a) states that some of these values have been determined subjectively, and that more quantitative study is needed to define the public's perception of what might be "excess" algae for recreational and aesthetic uses. However, Biggs recommends biomass threshold levels for recreational/aesthetic uses that are similar to the other authors.

It appears that algae levels may impair other uses such as recreation before DO problems are experienced. According to EPA (2000), there are no apparent effects on DO, pH or benthic invertebrates at the 100 - 200 mg/m^2 (chl. A) level. For example, Truckee River data shows that biomass levels much higher than in Table 1 do not necessarily lead to DO problems. During June 2001, biomass levels at various transects between the TMWRF discharge and Derby Dam ranged from around 200 to over 15,000 mg/m^2, with the average for all transects about 2000 mg/m^2 (chl.-a), yet no DO problems (<5 mg/l) were measured.

Unfortunately, there appears to be limited information on appropriate biomass thresholds needed to protect aquatic life uses. Nordin (2001) presented 100 mg/m^2 and stated that this "*criterion is designed primarily to protect fish habitat and changes in communities of organisms such as invertebrates which are important themselves or which may be important fish-food organisms*." Biggs (2000a) recommended 50 mg/m^2 for protection of benthic invertebrate diversity based upon macroinvertebrate and corresponding biomass data for New Zealand streams. His studies showed that EPT invertebrates (mayflies, stoneflies, caddisflies) populations decreased sharply where periphyton biomass exceeds 13 mg/m^2 (chl-a), with the relative abundance of midges, worms and snails increasing greatly above this level.

Finally, Welch and Jacoby (2004) conclude that 150 mg/m^2 may serve as an appropriate threshold:

> *"While the 150* mg/m^2 *level cannot be supported as an absolute threshold above which adverse effects on water quality and benthic habitat readily occur, it nonetheless is a level below which aesthetic quality will probably not be appreciably degraded…or other adverse effects…such as taste and odours in water supplies and fish flesh, impediment of water movement, clogging of water intakes, restriction of intra-gravel water flow and DO replenishment or…adverse effects on DO/pH in the water column, or degradation of benthic habitat."*

In a recent effort, Suplee and others (2008) conducted surveys to evaluate the public's perceptions of stream bottom algae levels in Montana's rivers and streams. Respondents looked at eight photographs of Montana rivers and streams, each representing a different level of stream bottom algae (ranging from 40 to 1280 mg/m^2 chl-a). Figures 1 and 2 show two of the photographs used depicting 150 and 400 mg/m^2

chl-a. For each photograph, respondents decided if the level of algae shown would be "desirable" or "undesirable" for their major form of recreation on rivers and streams. Surveys showed that as benthic algal chlorophyll *a* (Chl *a*) levels increased, desirability for recreation decreased. For the public majority, mean benthic Chl *a* levels ≥ 200 mg/m2 were determined to be undesirable for recreation, whereas mean levels ≤ 150 mg Chl *a* /m2 were found to be desirable.

Table 1. Algal Biomass Limits (modified from Welch and Jacoby, 2004)

Maximum Chlorophyll a – mg chl/m^2		% Cover		Use	Source
Diatoms	**Filamentous Algae**	**Diatoms**	**Filamentous Algae**		
100 – summer mean 150 - maximum				Unknown	Montana Water Quality Standards, Clarks Fork River
150 - 200				Aesthetics	Suplee et al. (2009)
100 - 200			20%	Based upon perceived impairment	Welch et al. (1988, 1989)
100-200				Nuisance	Dodds et al., 1997
200				Eutrophy	Dodds et al., 1998
150				Nuisance	Watson and Gestring, 1996
n/a	120	60% of diatoms > 0.3 cm thick	30% of filamentous green algae > 2 cm long	Contact recreation & aesthetics	Biggs, 2000a
200	120		30% of filamentous green algae > 2 cm long	Protection of trout habitat	
50	50			Protection of benthic biodiversity	
100				Invertebrates	Nordin, 1985
100			40%	Nuisance	Quinn, 1991
150				Nuisance	Sosiak, 2002

Due to its importance as an impairment indicator, rapid algae monitoring will be considered as an initial step in the proposed assessment protocols. However, it is important to recognize that algal biomass levels can be highly variable with time and space requiring monitoring of numerous site over extended time periods. As a result, algae monitoring can be quite expensive depending on the rigor of the approach. A variety of methodologies for characterizing algal biomass have been developed by others – ranging from the qualitative (the least rigorous consisting of photographs and visual estimates of biomass) to the quantitative (the most rigorous consisting of algal sampling and laboratory analyses).

Dissolved Oxygen: Algal activity leads to fluctuations in stream dissolved oxygen as photosynthesis and respiration occur. Typically, DO levels will be highest in the afternoon during peak photosynthesis and lowest near sunrise just prior to the restart of photosynthesis.

Figure 1. Algae Levels at 150 mg/m^2 chl-a (Suplee et al., 2009)

Figure 2. Algae Levels at 400 mg/m^2 chl-a (Suplee et al., 2009)

The Nevada Administrative Code (NAC) includes dissolved oxygen limits for many of Nevada's streams. For Class Waters, minimum DO limits vary from 5.0 mg/l for waters with trout, to 6.0 mg/l for waters without trout. Lower quality waters (Class D) have DO limits of 3.0 mg/l. Most other main stem streams have DO limits set at 5.0 mg/l regardless of fish species present. The NAC values are to be considered as instantaneous minimums.

EPA's Gold Book (EPA, 1986) provides somewhat different DO thresholds (Table 2). The high values for coldwater early life stages (9.5 and 8.0) may not be realistic for Nevada's waters. For example: for a stream at Elevation 5,000 feet and temperature of 21° C (typical summer temperature criteria in NAC), its DO saturation level is 7.4 mg/l. Without any algal photosynthesis activity, this water would not be able to meet the Gold Book Criteria during the peak afternoon temperatures.

Table 2. EPA Dissolved Oxygen Water Quality Criteria

Parameter	Description	Coldwater Criteria		Warmwater Criteria	
		Early Life Stages[1,2]	Other Life Stages	Early Life Stages[2]	Other Life Stages
30-day Mean	Average DO over a 30-day period	---	6.5	---	5.5
7-day Mean	Average DO over a 7-day period	9.5 (6.5)	---	6.0	---
7-day Mean Minimum	Average daily minimum DO for 7 days	---	5.0	---	4.0
1-day Minimum	Minimum DO for 1 day	8.0 (5.0)	4.0	5.0	3.0

[1]These are water column concentrations recommended to achieve the required intergravel DO concentrations (in parentheses) to support salmonid redds.
[2]Includes all embryonic and larval stages and all juvenile forms to 30 days following hatching.

As discussed earlier, available information suggests that algal biomass may need to be rather high (much higher than values in Table 1) before DO problems (<5 mg/l) are observed during a given day. Recent investigations in the lower South Fork Humboldt River showed night DO levels dropped to about 3 mg/l (Figure 3) as a result of the high algae growth (Figure 4).

Due to the diel fluctuations, the early morning DO is the most important value to measure as part of a monitoring scheme. Fortunately, DO monitoring has gotten much easier in recent years. The simplest approach for determining low DO levels for given site is to take readings near sunrise using a held-held DO monitor. A more intensive approach is the deployment of continuous DO monitoring sondes.

Figure 3. South Fork Humboldt River ~3 Miles Upstream of Humboldt River - DO, Temperature and pH - August 11-12, 2008

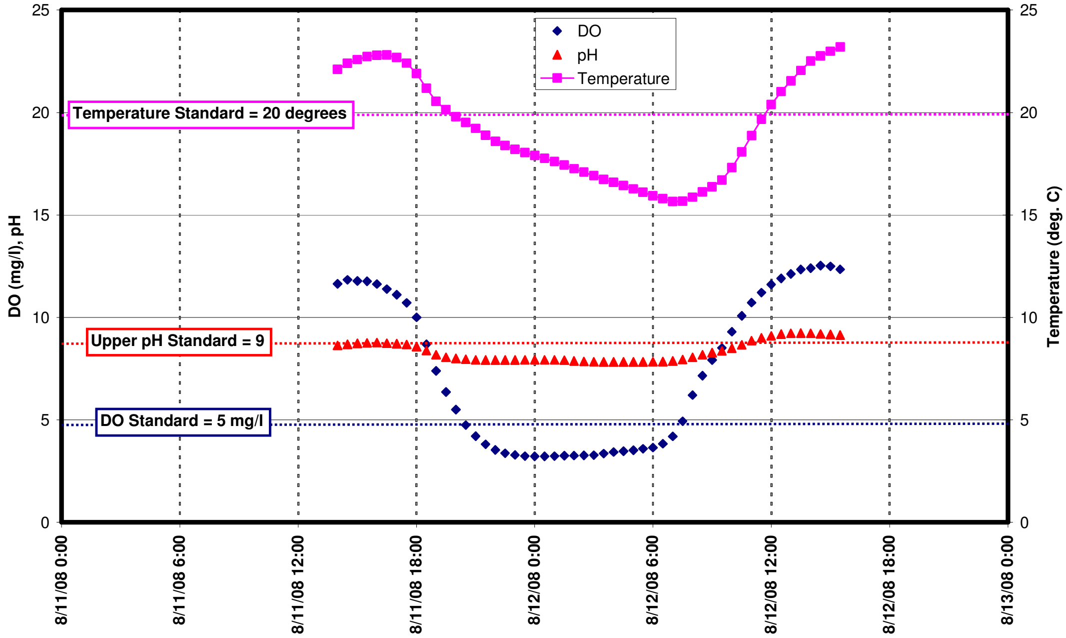

Figure 4. High Algal Biomass in Lower South Fork Humboldt River

pH: Algal photosynthesis and respiration alter water pH with the uptake or release of carbon dioxide. As a result, pH levels can vary considerably over one day depending upon the buffering capacity (alkalinity) of the stream. Like DO, algal activity can lead to maximum pH levels during the afternoon with minimum pH levels prior to sunrise. However, streams with lower buffering capacity (lower alkalinity) can show greater diurnal swings in pH (EPA, 2000).

Most of the streams identified in the NAC includes pH limits of 6.5 – 9.0 for the protection of aquatic life based upon EPA guidance. These criteria are considered to represent instantaneous thresholds for pH measurements taken at any time during the data.

Due to the diel fluctuations, the early morning pH is the most important value to measure as part of a monitoring scheme. The simplest approach for determining low/high pH levels for given site is to take readings near sunrise and in the mid-afternoon using a hand-held pH monitor. A more intensive approach is the deployment of continuous pH monitoring sondes.

Proposed Nutrient Assessment Protocols for Wadeable Streams

This section presents a multi-tiered approach (modeled after the New Mexico approach (2005)) for assessing nutrient impairment status (Figure 5). In general, the assessment tiers are as follows. For new waters (waters not yet identified in the NAC), the first step could be to compare median nutrient levels to indicator thresholds that have been developed by NDEP (April 2009). If indicators are exceeded, a Level I investigation may be an appropriate next step. For 303(d) listed waters, the Level I assessment may be the appropriate starting point for assessment. A Level I assessment is primarily qualitative in nature allowing for rapid assessments of numerous sites. If the Level I assessment indicates a possible nutrient problem, a Level II assessment may be initiated which involves more quantitative measurements. If the Level II assessment is inconconclusive, more detailed biological assessments (protocols yet to be developed) may be needed.

It must be kept in mind that these steps (and their level of effort) are not carved in stone and any can be bypassed based upon BPJ. These levels are provided as general guidelines describing possible ways to assess nutrient impairment status. As always, in determining impairment status, professional judgment will be necessary on a case-by-case basis. NDEP must have flexibility in how these assessments can be undertaken, as dictated by the stream in question. It is fully expected that as experience is gained with these protocols, modifications will be made based upon lessons learned. Clearly stream access is a significant factor affecting the utility of this approach. Public access may be limited along some streams, reducing the number of sites that can be readily visited. The ability to draw conclusions about a water's status could be restricted if insufficient sites are assessed.

The Level I/II approach was initially developed to screen and prioritize 303(d) listed waters for potential next steps such as TMDL development and 319(h) funded nonpoint source projects. In subsequent discussions with EPA, it has been agreed that this basic approach can also be used to list/delist waters based upon response variables (DO, algae biomass, etc.) regardless of whether or not phosphorus levels are exceeding water quality standards. At this time, it is unknown what level of information is needed to justify delisting/listing. Given the high variability of algae biomass, etc., it is expected that a significant number of sites may need to be investigated for a given stream reach.

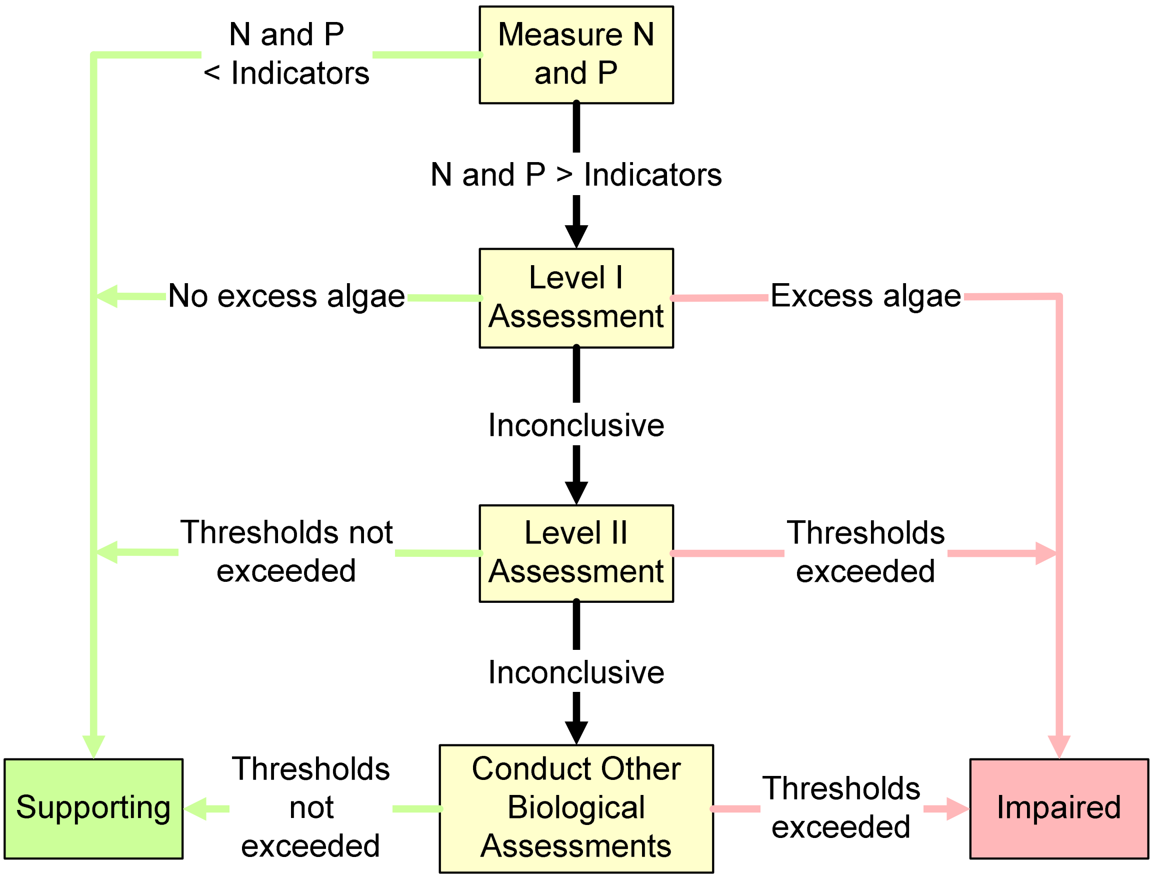

Figure 5. Proposed Multi-Tiered Nutrient Assessment Approach

As discussed early, the flow chart of activities does not have to be rigidly followed and any of the assessment tiers in Figure 5 can be initiated for any water of interest, depending upon the needs of the investigator. In other words, a waterbody does not have to be exceeding its nutrient criteria or indicators for one to pursue any of these assessment tiers. As described in Nevada's Nutrient Criteria Strategy (NDEP, 2007), significant data are needed for a range of waters (from pristine to poor quality) throughout the state. Through the collection of data and analysis, NDEP will be able to test and refine its nutrient criteria strategy and these assessment protocols.

Level I Assessment

The Level I assessment relies primarily upon qualitative estimates of algal biomass as an indicator for possible next assessment steps. In addition to monitoring site location data, other information collected as part of the Level I assessment includes:

- ❑ Flow conditions (quantity, turbidity, etc.)
- ❑ Flow width and depth
- ❑ Canopy cover conditions
- ❑ Substrate type
- ❑ Indicate dominant species (rooted aquatic plants, attached algae)
- ❑ Percent cover of stream bottom with algae

Given the spatial and temporal variability of algal biomass, it may be necessary to evaluate numerous locations two or more times during the growing season. As water conditions can be highly variable, it may be necessary to visit the assessment sites during two or more years. Further Level I guidance and field sheets are provided in Appendices A and B. The number of sites and frequency of visits are somewhat determined by the goals of the investigation and can be based upon best professional judgment. If the goal is to attempt to delist a water, it is likely that a significant number of sites will need to be evaluated. It is uncertain how much information will be needed to actually delist a water based upon Level I assessment information.

The literature provides no clear cut levels (% cover) at which impairment may be assumed to occur. As shown in Table 1, some researchers have identified algae cover levels of 20 to 40% as affecting recreation and aquatic life uses. In New Mexico's approach, the >50% cover is used to determine whether or not a Level II assessment is needed. However, the New Mexico protocols do not discussion reasons for selecting the >50% cover threshold. Nevertheless, the >50% cover threshold appears to be an appropriate value to use at this time. According to data from Biggs (2000) for the Waipara River in New Zealand, 50% cover conditions were comparable to algal biomass levels of about 200 mg/m^2 chl-a, a value at the upper end of suggested chlorophyll-a thresholds in Table 1.

As with the %cover threshold, the literature does not provide clear guidance of microalgae and macrophyte[1] levels that could indicate nutrient problems. The New Mexico protocols incorporate a rating of microalgae based upon thickness, with a thickness of >1 mm used to determine whether or not a Level II assessment is appropriate. They also note if stream reaches have macrophyte cover less than or greater than 50%. However, this information is not used by New Mexico as a flag for potential nutrient enrichment since "…bottom sediments act as the primary nutrient source for rooted macrophytes…" Nevertheless excessive macrophytes may need to be examined closer, as nutrient rich sediments may be the result of high erosion or other upstream activities. Madsen and Cedergreen (2002) pointed out that while macrophytes may obtain a majority of their nutrient from the stream substrate, many macrophytes can also utilize nutrients in the water column. However, higher nutrient availability in the sediment may lead the plants to utilize this source.

[1] Macroalgae are those benthic algae having a thallus (structure) visible to the naked eye (such as cladophora), while microalgae have a thallus that cannot be distinguished without a microscope (such as diatoms) (Allan and Castillo, 2007). Hauer and Lamberti (2007), considers macrophytes to consist of those "large plants" which have true roots. Typically, macrophytes can be either emergent (attached to the bottom via roots but extend above the water surface), submerged (attached to the bottom with most of their mass below the water surface) or free-floating (not attached to the bottom but with roots hanging in the water) plants.

For purposes of multi-tiered assessment, it is suggested that the following thresholds be used in evaluating the Level I information:

Indicate "No Excess" Algae:

- <25% cover by microalgae (> 1mm thick) and filamentous algae
- <25% cover by macrophytes

Indicate Inconclusive Findings:

- 25 - 75% cover by microalgae (> 1mm thick) and filamentous algae
- 25 - 75% cover by macrophytes

Indicate "Excess" Algae:

- >75% cover by microalgae (> 1mm thick) and filamentous algae
- >75% cover by macrophytes

It must be noted that these thresholds are not absolutes, and that staff can always choose to undertake Level II assessments based upon best professional judgment and other factors. These thresholds will need to be reviewed as more data are collected.

It is likely that situations will be encountered where only a portion of a long reach is experiencing elevated algae (and perhaps low dissolved oxygen levels), with the average algal cover for the long reach falling below the 25% threshold. It may be appropriate to only seek to delist the portion with low algae, and to undertake additional investigations for the problem area.

Level II Assessment

Under the Level II assessment, more quantitative measurements of algal biomass along with water quality sampling and measurements of daily minimum/maximum DO and pH levels can be taken for comparison to the water quality standards or indicators. Professional judgment will be needed to determine how much data are needed to make a nutrient impairment status determination. Some waters may require more data than others to reach a conclusion. For example, it may be appropriate to collect limited early morning (near sunrise) DO data for the stream reach in question, or one may opt to deploy a water quality monitoring sonde to collect more detailed information.

As part of the Level II assessment, it may be necessary to determine whether or not the DO and pH problems are due to algal activity and not other causes (low flow, acid mine drainage, etc.). In any assessment, it must be recognized that nutrient impairment problems can be greatly exacerbated by low flow and poor riparian shading conditions. It must also be considered that algae may impact beneficial uses (aquatic life, recreation) even though no DO and pH problems are identified.

Recommendations for Collecting and Evaluating Algal Biomass Data: Table 1 presents a range of chlorophyll-a thresholds that have been recommended by others for the protection of various uses. All of these sources recommend chl-a levels less than or equal to 200 mg/m^2, with a range of 100 – 200 mg/m^2 common. As with most criteria, one size does not fit all. Nevada streams have great diversity across the varying landscape, with streams beginning in the mountains as high gradient waters but flowing out into a valley at a much lower gradient. It is unrealistic to expect that one algal biomass threshold is appropriate for all these different streams and reach. However at this time, no gradient of thresholds are suggested in the literature. As suggested in Table 1, 150 to 200 mg/m^2 (chl-a) could initially be considered as a threshold above which streams would be considered impaired. The appropriateness of this threshold should be tested over time. It may be found that lower chl-a values may be appropriate for high quality streams in areas such as the Tahoe Basin, upper Jarbidge basin, Ruby Mountains, etc.

Ideally to apply the 150 to 200 mg/m^2 (chl-a) thresholds, algal biomass data should be collected using the same protocols used to develop these thresholds. Unfortunately, this information is not known. Nevertheless, there are a number of available protocols that could potentially be utilized. Currently under NDEP's bioassessment program, streams are being sampled following EMAP type protocols[2], but it is believed that this may not be the appropriate approach when comparing results to the Table 1 thresholds. For many streams, the study reach can become rather long encompassing multiple habitats (riffles, runs, pools). Barbour et al. (1999) provides a discussion of this approach, but recommends that single habitats (rather than multiple) be sampled when the intent is to quantify algal biomass. According to EPA (1999), the algae sampling should focus on the portion of the stream where algae is most likely to occur and to conflict with beneficial uses. This would be expected to occur in those riffle/run areas with cobble, gravel, etc. for attached algae to grown. EPA went on to recommend collecting 10 to 20 samples over a reach of at least 100 meters.

Others have also recommended protocols focusing on single habitat sections. Biggs (2000) has recommended a chlorophyll-a threshold of 120 mg/m^2 for the average within a reach that is a relatively homogenous section of stream channel. Use of this threshold would seem to suggest that sampling needs to be targeted to a riffle or run area.

Even if biomass sampling is restricted to a smaller reach than EMAP protocols would call for, there can still be significant variability within the subject reach. Typically, higher biomass occurs in areas with larger, more stable substrate material (Biggs, 2000a). Tett et al. (1978) and Biggs and Shand (1987) found that isolated boulders/bedrock can contain up to 15 times more biomass than that found on nearby unconsolidated sands and silts. This variability can make it challenging if the goal of the sampling is to quantify average algae levels for a given transect, reach or other division. One approach taken by the Desert Research Institute for Truckee River sampling (Memmott et al., 2008) was to collect 3 samples from each unit within a transect - sampling low, medium and high algal coverage areas yielding a composite sample for the unit.

The literature indicates there is no shortage of possible protocols for Nevada to implement as part of it nutrient assessment protocols. Given the variety of protocols available, guidance on an appropriate sampling method was sought from EPA, Region IX. In support of this need, EPA contracted with TetraTech (2008) to test an approach on Dry Creek (Reno area) and make recommendations. The project entailed the pilot testing of two hydrid sampling protocols that incorporated components of protocols used by the State of Montana (MDEQ, 2007), Stevenson and Bahl, 1999) and EPA (2003). Key components of the protocols included:

1. 100 meter reaches were selected and 11 equally spaced transects with established (Figure 5).
2. At each transect, the substrate type (cobble, sand, etc.) was characterized at 10 equally spaced points using a viewing bucket. This information was used to estimate the %rock, % sand, etc. within the study reach.
3. At each transect, one attached algae sample was collected from each substrate type. For each of the subreaches, one attached algae sample was collected from each substrate type. Sample locations were chosen so as be representative of the algae levels for the transect/subreach. Samples were composited based upon substrate type and sample location. For Dry Creek which has substrate types of rock and sand, four different composite samples were collected: a) transect samples – rock; b) transect samples – sand; c) subreach samples – rock; d) subreach samples – sand.

[2] In general, a reach equal to 40 times the channel width is selected for sampling. The selected reach is divided into 11 equally space transects with one algae sample collected from each transect, which are composited into 1 sample for laboratory analyses.

4. Sampling occurred 5 times during July and August, 2008.
5. Using the laboratory results along with the percentages for substrate types, average reach area-weighted (by substrate type) chlorophyll-a levels (mg/m^2) were calculated for the transect samples and the subreach samples.

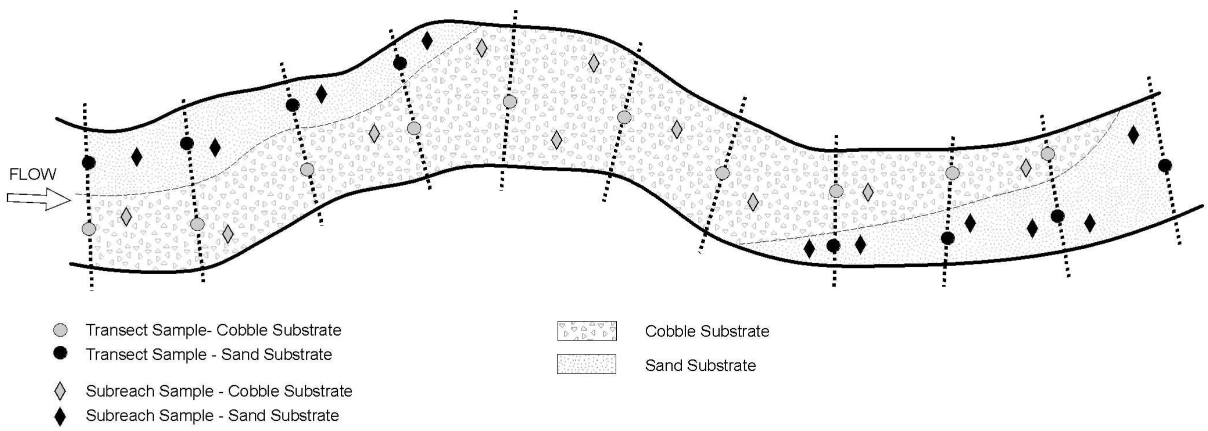

Figure 6. Algae Sampling Approaches Tested by TetraTech (2008)

The results of the TetraTech study showed that the subreach sampling method and the transect sampling methods yielded similar results, with no one method showing consistently higher or lower results than the other method. The final recommendations by TetraTech involved the collection of the algal samples within each of the ten subreaches, at locations representative of algae conditions in the subreach.

Based upon the current work by TetraTech (2008) and other approaches, initial protocols have been derived for the purposes of estimating average attached algae density for reaches of interest (see Appendix C). Key factors in the protocols include:

1. Locate 100 meter section of stream (regardless of stream width). Depending upon the goals of the investigation, it may be appropriate to limit the study reach to a "high potential" area such as a riffle.

2. Divide the reach into 10 equal length subreaches.

3. For each subreach, visually estimate the percentage of substrate type (rock, fines/sand)

4. As presented, the goal of this protocol is to estimate "average algal density" for a study reach. One could expect that the more homogenous the algae levels for a reach, the fewer number of samples needed to quantify average chlorophyll-a levels. Conversely, the more sporadic the algae, the more samples that are likely needed to improve the accuracy of an algal coverage estimate. Because no two streams are alike, it is recommended that these protocols not be overly prescriptive and that the field personnel be allowed to make best professional judgments on the number of samples appropriate to estimate the reach-wide average algae density. Two options are provided for initial use:

    a. Alternative 1 - Within each subreach (and for each substrate type), collect one or more "representative" samples. To limit bias, the number of samples collected should be consistent from subreach to subreach. Only samples collected from the same substrate type should be composited.

b. Alternative 2 - Within each subreach (and for each substrate type), collect one sample from high algal cover area, one sample from medium cover area, and one sample from low cover area. Only samples collected from the same substrate type should be composited.

5. From the substrate type data and the algae laboratory analyses, the average (area weighted) chlorophyll-a density and AFDW (ash free dry weight) density are calculated for the 100 meter study reach.

As discussed earlier, it is unrealistic to expect that one algal biomass threshold is appropriate for all the diverse streams in Nevada. However at this time, no gradient of thresholds are suggested in the literature. It is suggested that a reach average algal threshold of 200 mg/m^2 (chl-a) be initially used for identifying impaired waters. The appropriateness of this threshold will need to be tested over time. For the higher quality waters in the Tahoe Basin, upper Jarbidge basin, Ruby Mountains, etc., it may be valuable to collect algal data at minimally-impacted streams and use these data to develop more restrictive attached algal thresholds.

Some streams have been found to be dominated by macrophytes rather than attached algae. At this time, no macrophyte thresholds have been found in the available literature, and none are recommended for the Level II assessment.

DO, pH, temperature monitoring: Under the Level III assessment, it may be appropriate to collect hourly DO, pH, and temperature data using datalogging water quality sondes. Examples of sonde installations are provided by current Truckee River monitoring efforts and by recent Carson River efforts (Latham, 2006). This type of monitoring can be labor intensive requiring frequent site visits to ensure proper operation of sondes. A less rigorous approach would involve instantaneous readings taken near sunrise and mid-afternoon to estimate daily minimum and maximum DO levels.

Summer DO water quality standards range from 5 to 6 mg/l as allowable daily minimums. Again, it is recommended that these protocols not be overly prescriptive in how DO impairments are determined from the DO data. In general, waters have been considered impaired if standards are exceeded for more than 10% of the samples collected. While the 10% can be used as a guideline, it is important to consider factors other than nutrients (flow, temperature, etc.) that can have detrimental effect on DO levels. Ultimately, the determination of nutrient impairment may need to be based upon a certain level of best professional judgment.

Some initial investigations by NDEP and a review of available information from others suggest that rather high algae levels are generally needed before DO problems are experienced in a stream. Algal levels typically need to be quite a bit higher than the 200 mg/m^2 (chl-a) threshold before DO levels consistently fall below 5 mg/l.

Water quality sampling for nutrient species: Frequent (possibly weekly) sampling for a suite of nutrient species may be needed to support a Level II investigation, if detailed water quality modeling is anticipated. Like biomass, DO, and pH, nutrient levels can significantly vary from day to day and from year to year. However if the sole purpose of the Level II assessment is to collect only algal and DO data to determine the nutrient impairment status, it may not be necessary to collect detailed nutrient data. Lesser efforts may be appropriate and should be decided on a case-by-case basis.

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Appendix A
Level I Assessment Guidance

Appendix A - Level I Assessment Guidance

Site Selection

Since algae biomass can have great spatial variability, it may be important to visit as many sites as reasonably possible (and as time/resources allows) within the waterbody of interest. As always, land ownership and physical constraints may greatly restrict the choice of accessible sites. The number of sites that need to be investigated can be based upon best professional judgment and is somewhat driven by the goals of the investigation. If limited resources are available but it is desirable to get an initial sense of conditions, it may be appropriate to only monitor those sites with the highest potential for algae growth (areas with limited shading, adequate substrate for algae growth, such as riffles). It is possible that if these "high potential" sites have limited algae, other "lower potential" areas of the stream will also have limited algae.

If the goal is investigate the possibility of delisting/listing a given water, it would be appropriate to visit a significant number of sites in order to get a comprehensive evaluation of overall algae levels. Sites should not be limited to the highest potential areas, but should look at a variety of physical conditions (pools, runs, riffles).

These protocols do not call for a specific reach length that needs to be investigated. This should be based upon conditions in the field and the best professional judgment of the crew. For sites on the larger rivers, it may be appropriate to record algal cover within riffle, run, or pool areas on separate field sheets. In this instance, the reach length would be dictated by the length of the riffle, run or pool. For smaller streams, a reach can be selected which includes riffles, runs and pools.

Monitoring Timing/Frequency

The temporal variability of algae biomass suggests that the selected sites need to be visited several times in an attempt to capture the peak biomass period. It is likely that peak algae biomass levels will occur during periods of higher temperatures, lower flows, clear water and maximum sunlight – typically July through September. Therefore, these conditions should be targeted in the timing of the surveys. Areas with high algal cover (such as the EF Carson River between Highway 88 and Muller Lane) have been found to still have significant (50-75 % cover) algal levels even into November. Also, there can be variability from year to year so sites may need to be visited over multiple years.

As discussed above, Biggs (2000a) has shown that the time since the last scouring flow event can have a significant affect on the algal biomass on a given day. This needs to be kept in mind when planning the surveys. If it is known that high flows have recently occurred, it may not be appropriate to visit the site.

Some streams of interest may be dry at the time of the field visit. It is possible to look for signs of dried algae on the substrate and record the findings.

Gather Background Information

Prior to going out into the field, it is highly recommended that various background information be gathered. If sufficient information already exists that describes the eutrophication status of a given waterbody (or a portion), then actions other than a visual survey may be appropriate.

Background information may be valuable in selecting sites and the timing of the surveys. For example, NDEP and EPA have been performing physical habitat characterizations throughout Nevada. These surveys may provide some insight into algal conditions in the subject waters. Other sources of information may include BLM allotment assessments, fisheries assessments, etc. Personal communications with BLM, USFWS, NDOW, USFS field staff may also yield valuable background information.

Field Data Collection

For each reach under investigation, the crew should fill out the assessment form in Appendix B. Also, it is highly recommended that photographic documentation accompany any visual survey effort. This would include general photographs of the site along with photographs of the streambottom. However, it may be appropriate to establish photo points/transects that can be revisited during the algal growing season. For a transect, a series of photographs could be taken while walking across the stream and facing upstream/downstream depending upon the lighting and other conditions. As always, provide documentation of any photographs taken.

Appendix B
Level I Assessment Field Form

Level I Nutrient Assessment Form

Nevada Division of Environmental Protection

Site Number					
Stream Name					
Reach Number		**Reach Length**			
%Riffle		**%Run**		**% Pool**	
Description					
Latitude/Northing		**Longitude/Easting**			
Datum		**Elevation (ft)**			
Survey by		**Date**		**Time**	
Weather Conditions	**Current**				
	Recent flushing flows?				

Stream Features	**Average width (ft)**		**Average depth (ft)**	
	Flow conditions (quantity, turbid?)		**Canopy Cover**	❑ Mostly open ❑ Partly open ❑ Mostly shaded
Riparian Vegetation	**Indicate the dominant type**	❑ Trees ❑ Willows	❑ Shrubs ❑ Grasses	
	Dominant species			
Substrate	**Indicate the dominant type(s)**	❑ Silt/Sand - <2mm (up to ladybug size) ❑ Gravel – 2 to 64 mm (ladybug to tennis ball) ❑ Cobble – 64 to 256 mm (tennis ball to basketball)	❑ Boulder - >256 mm (larger than basketball) ❑ Bedrock	
	Percent of stream bottom with gravel or larger	❑ < 25% ❑ 25 to 50%	❑ 50 to 75% ❑ >75%	
Aquatic Vegetation	**Indicate the dominant type(s)**	❑ Macrophytes – Non-algae	❑ Attached algae	
	Percent of stream covered by macrophytes	❑ < 25% ❑ 25 to 50%	❑ 50 to 75% ❑ >75%	
	Algae — **Percent of stream covered by microalgae > 1 mm thick AND filamentous algae**	❑ < 25% ❑ 25 to 50%	❑ 50 to 75% ❑ >75%	
	• **Percent microalgae > 1 mm**			
	• **Percent filamentous algae**			
Comments				

Level I Nutrient Assessment
Photo Point Monitoring Sites – Description and Location
Nevada Division of Environmental Protection

Site Number		**Reach Number**	
Stream Name			
Location Description			

Map

Appendix C
Level II Protocols

Yet to be completed