ECOLOGY EVALUATION OF BAP CONCERNS AND COMMENTS: INTERNAL REVIEW DRAFT JUNE 27, 2012
OFFERED FOR REVIEW TO DR. BRETT JULY 16, 2012

Concern or comment	Response to Concern	Expectation
Quality Assurance Project Plan: Spokane Regional Wastewater Phosphorus Bio-Availability Study, prepared by Spokane County Utilities Division, July 2009. Is there a final signed copy of the QAPP?		
Goal 1: Determine the fraction of total phosphorus in effluent from Spokane area WWTP pilot tertiary treatment processes that is biologically available. Did study accomplish this goal? If not, was there sufficient explanation? (QAPP, July 2009, p 4) Goal 2: Determine how advanced phosphorus removal technology affects the BAP of the effluent. Did study accomplish this goal? If not, was there sufficient explanation? (QAPP, July 2009, p 4)	The objective of this study was to use algal bioassays to determine the Bio-Available Phosphorus (BAP) of effluent treated by the pilot projects at the main WWTP discharges to the Spokane River. The percent BAP (%BAP) varied with different P removal levels. This study also tested whether more conventional, and easily carried out, measures of P composition could be used in place of BAP to quantify the eutrophication potential of effluents. Spokane: Trend shows decreasing BAP in effluent; variation in results increases at low concentrations of BAP due to analytical method. Coeur d'Alene: Trend shows decreasing BAP in effluent; variation in results due to process fluctuations. Post Falls: Variation for the effluent samples makes it challenging to distinguish what levels of P removal this plant was capable of versus what they actually achieved. Liberty Lake: Trend shows decreasing BAP in effluent; variation in results due to process fluctuations. Hayden Lake Area SWB: high variation associated with the P concentrations for most samples which was compounded by a small sample size. IEP: Removal performance is based on the result from only one influent sample.	The study shows consistent trends with respect to the relationship of BAP to TP. The data quality for all of the studies was qualified in by one or more of the following: • Analytical variation at low concentrations that made the accuracy and precision of low levels of BAP difficult to assess. • Variations due to process trends that created variability and/or what was classified as "outlier" data in the results. • Low sample counts, which made statistical analysis of the data quality difficult if not impossible (i.e., data based on one data point). Data quality must be rigorous if used for regulatory purposes (such as using datasets within a model and/or changing regulatory permit values). Standard Quality Assurance/Quality control procedures should be used to determine method detection limits, method quantification limits, matrix interferences, precision, accuracy, and reproducibility.

Goal 3: Determine if the bioavailability of phosphorus from Spokane area wastewater discharges varies seasonally. Did study accomplish this goal? If not, was there sufficient explanation? (QAPP, July 2009, p 4)	No locations were sampled in accordance with the QAPP schedule. No explanations were provided regarding deviance for QAPP schedule. Seasonal variability was discussed with the Spokane River WWTP and the Spokane River summer vs. winter conditions.	Scope and schedule are an integral part of the QAPP. Any deviations from the QAPP must be explained.
Effluent from WWTPs processed through the following pilot tertiary treatment processes will be used for evaluation in this study: City of Spokane: • Kruger Actiflo sand-ballasted sedimentation • Cambridge Water technology's CoMag ballasted sedimentation • Zenon membrane filtration • Corix conventional sedimentation • Blue Water continuous upflow filter • Corix multi-media granular filtration City of Coeur d'Alene • Blue Water continuous upflow filter • Zenon micro-filtration system • Zenon membrane bioreactor system Inland Empire Paper • Siemens Trident HS system tertiary treatment Were these systems tested? If not, was there sufficient explanation? (QAPP, July 2009, p 5)	It is not possible to determine if these systems were tested since there are multiple systems from single manufacturers with multiple names. City of Spokane • Kruger Actiflo sand-ballasted sedimentation (name not mentioned) • Blue Water continuous upflow filter (type not mentioned) Coeur d'Alene (nomenclature confusing) • Zenon membrane filter • Zenon membrane system	Terminology needs to be consistent and specific throughout the report: technology used, brand, model, model number, etc. Basically, the report should provide enough information that test procedures are "reproducible."

A parallel study at Northwestern University is being implemented to conduct detailed phosphorus speciation analysis of effluent samples from the same WWTPs. Sampling for both studies will be coordinated, when possible, to help avoid repeated analytical analysis and to allow the two studies to build off the associated results. Were the results of this study addressed in the report? Have the results of this study been made available in order to build off the associated results? (QAPP, July 2009, p 5)	Not addressed in the report.	If there is collaborative data, it should be provided. This is a major omission in the study that should be explained.
Project schedule start date: Sampling begins, July 2009 Project schedule end date: Final report, July 2010 Did project meet schedule milestones? If not, was sufficient explanation provided? (QAPP, July 2009, p 6)	There were numerous deviations from the schedule, which were not explained. See Table 2 at end of this report. City of Spokane: August 2009-April 2010 City of Coeur d'Alene: May 2010-August 2010 City of Post Falls: May 2010-August 2010 Liberty Lake SWD: April 2010-August 2010 HARSB: May 2010-August 2010 IEP: September 2009-June 2010 Spokane River: August 2009-March 2010	This is a seasonal study so deviation from schedule could impact the interpretation of the data. Any deviations from schedule must be noted, explained, and the potential impact on the results explained.
Did data collected meet the measurement quality objectives (Table "5")? If not, was there sufficient explanation? (QAPP, July 2009, p 8)	Of TP, TDP, and SRP, the only parameter that was measured was TP. Check standards/LCS, and matrix spikes were not done. Without these measurements, it is not possible to evaluate the precision or accuracy of the procedure for TP. BAP does not have a measurement quality objective, which is a failure of the QAPP to specify that this is necessary. See Table 1 at end of this document.	Future QAPPs should have rigorous measurement quality objectives and data quality objectives. Project researchers must develop procedures that demonstrate how those objectives will be achieved.

Were samples collected at 41 sampling sites over the sampling period, as proposed (Table 6)? (The sampling schedule will be finalized during the study.) If the preliminary sampling sites were not used, was there sufficient explanation? (QAPP, July 2009, p 9-10)	1) QAPP lists "Pilot A-F" and list of treatment processes. 2) No direct correlation in report due to nomenclature 3) Actual samples were taken from treatments in series. See Table 2 at end of this document regarding TOTAL planned events 32; limit of 45; actual 91 Proposed facilities 3; actual 6 No rationale provided for going outside the scope or schedule of the QAPP.	Scope and schedule are an integral part of the QAPP. Any deviations from the QAPP must be explained.
Effluent from at least seven treatment processes and two natural waters will be evaluated for phosphorus bioavailability. Was this objective accomplished? If not, was there sufficient explanation? (QAPP, July 2009, p 10)	6 plants, 14 effluent streams, 2 natural waters were evaluated.	No further action.
Samples will include at least 3 samples each from: • One municipal waste stream pilot from Coeur d'Alene • One municipal waste stream pilot from City of Spokane • One industrial source (Inland Empire Paper) Was this objective accomplished? If not, was there sufficient explanation? (QAPP, July 2009, p 10)	The report does not make a clear distinction between "samples" and "sampling events" • 5 sampling events from CdA • 8 sampling events from City of Spokane • 5 sampling events from IEP	No further action.

Five surface water samples may be tested up to four times. Was this objective accomplished? If not, was there sufficient explanation? (QAPP, July 2009, p 10)	2 surface water sample locations were tested. • One was sampled 5 times • One was sampled 1 time. QAPP does not specify procedures.	Future QAPPs should have rigorous measurement quality objectives and data quality objectives. Project researchers must develop procedures that demonstrate how those objectives will be achieved.
One sample site (Spokane Pilot A) will be tested monthly throughout the study period. Was this objective accomplished? If not, was there sufficient explanation? (QAPP, July 2009, p 10)	No. There was no discussion regarding deviation from the proposed monitoring schedule.	Scope and schedule are an integral part of the QAPP. Any deviations from the QAPP must be explained.
All samples, including WWTP effluent and surface water, will be analyzed for total phosphorus and total dissolved phosphorus. Was this objective accomplished? If not, was there sufficient explanation? (QAPP, July 2009, p 10)	No. Total dissolved phosphorus was not measured. There was no discussion regarding why TDP was not measured.	Scope and schedule are an integral part of the QAPP. Any deviations from the QAPP must be explained.
This analysis for total phosphorus will allow the determination of percent bioavailability in the TP sample. Was this objective accomplished? If not, was there sufficient explanation? (QAPP, July 2009, p 10)	Yes.	No further action.
Analysis of Total Dissolved Phosphorus will allow for speciation between the dissolved and particulate fraction. Was this objective accomplished? If not, was there sufficient explanation? (QAPP, July 2009, p 10)	No. Total dissolved phosphorus was not measured. There was no discussion regarding why TDP was not measured.	Scope and schedule are an integral part of the QAPP. Any deviations from the QAPP must be explained.

Other analysis, such as Soluble Reactive Phosphorus will depend on project funding and coordination with the parallel study at Northwestern University to avoid duplicate analytical procedures. Was there coordination and/or analysis for SRP? If not, was there sufficient explanation? "Soluble Reactive Phosphorus" refers to all forms of phosphorus present in a sample following filtration (usually through a .45 µm filter) that react to a specific analytical method. (QAPP, July 2009, p 10, 11)	No. There was no coordination and/or analysis for SRP nor was there sufficient explanation regarding whether or not this was not done.	Scope and schedule are an integral part of the QAPP. Any deviations from the QAPP must be explained.
Were the field procedures followed? If not, was there sufficient explanation? (QAPP, July 2009, p 11, paragraph 1)	Unclear. The number of samples collected at each site (replicates vs. samples) was not clearly described in the report.	Chain of custody procedures should be developed by the researcher so that there is documentation of the shipping procedures and adequate control of the samples. Data should be reported in a manner that clearly demonstrates the number of locations sampled, samples collected, and aliquots of samples analyzed.
The test procedure for determining phosphorus bio-availability assumes that "raw" (unfiltered and untreated samples of water) will be subjected to bioassay. Was this procedure followed? If not, was there sufficient explanation? (QAPP, July 2009, p 11, paragraph 4)	Unclear.	The report must contain a description indicating that the QAPP test procedure was followed and, if not followed, adequate explanation must be provided.

[The bio-assay] results will be coupled with the parallel Northwestern University study conducting detailed phosphorus speciation analysis. The combination of the two studies will allow an in-depth examination of phosphorus bio-availability, but the method lacks the advantage of direct biota growth measurements. Was this objective accomplished? If not, was there sufficient explanation? (QAPP, July 2009, p 11, paragraph 4)	Not addressed in the report.	This is a major omission in the study that should be explained.
Phosphorus bio-availability will be determined using the bioassay method described in Standard Method 8111. Was this method used? If not, was there sufficient explanation? (QAPP, July 2009, p 11-12)	Yes.	No further action.
Because of the precision of this method is lower than for standard wet chemistry approaches, four replicates of each sample will be incubated and the results averaged for the final calculations. (Four 50 ml aliquots of sample are incubated for 14 days). Was this method used? If not, was there sufficient explanation? (QAPP, July 2009, p 12)	Yes.	No further action.

Five replicates each of seven standards (0, 10, 20, 35, 50, 75, and 100 µg P/L) are incubated simultaneously to establish a "standard curve." Was this method used? If not, was there sufficient explanation? (QAPP, July 2009, p 12)	No. Standard media with a known concentration series of KH_2PO_4 (0, 5, 10, 15, 20, 25, 30, 40 and 50 µg P·L-1) were incubated in triplicate to obtain a standard curve for algal growth yield.	Scope and schedule are an integral part of the QAPP. Any deviations from the QAPP must be explained.
Sample conditions were 24 ± 2° C under continuous fluorescent lighting of 4300 lm ± 10% for 14 days. Was this method used? If not, was there sufficient explanation? (QAPP, July 2009, p 12)	Yes.	No further action.
The test algae will be deprived of phosphorus prior to incubation. Was this method used? If not, was there sufficient explanation? (QAPP, July 2009, p 12)	Yes.	No further action.
The total phosphorus values provide a necessary baseline for calculating the percent bio-available phosphorus. Was this objective achieved? If not, was there sufficient explanation? (QAPP, July 2009, p 12)	The study shows consistent trends with respect to the relationship of BAP to TP. The data quality for all of the studies was qualified in by one or more of the following: • Analytical variation at low concentrations that made the accuracy and precision of low levels of BAP difficult to assess. • Variations due to process trends that created variability and/or what was classified as "outlier" data in the results. • Low sample counts, which made statistical analysis of the data quality difficult if not impossible (i.e., data based on one data point).	Data quality must be rigorous is used for regulatory purposes (such as using datasets within a model and/or changing regulatory permit values). Standard Quality Assurance/Quality control procedures should be used to determine method detection limits, method quantification limits, matrix interferences, precision, accuracy, and reproducibility.

The total phosphorus values will allow observation of the discharge phosphorus composition over the year. Was this objective achieved? If not, was there sufficient explanation? (QAPP, July 2009, p 12)	No locations were sampled in accordance with the QAPP schedule. No explanations were provided regarding deviance for QAPP schedule. Seasonal variability was discussed with the Spokane River WWTP and the Spokane River summer vs. winter conditions.	Scope and schedule are an integral part of the QAPP. Any deviations from the QAPP must be explained.
Determining the soluble reactive phosphorus will provide a base for comparing of the results of the somewhat tedious bio-available phosphorus test with the traditional analytical measure of biologically active phosphorus. Was this objective achieved? If not, was there sufficient explanation? (QAPP, July 2009, p 12)	No. Analysis of TRP allowed for speciation between the "reactive" and "non-reactive" fractions and provided a basis for comparison with the much more time intensive BAP assays.	Scope and schedule are an integral part of the QAPP. Any deviations from the QAPP must be explained.
Were laboratory measurements made in accordance with the parameters in Table 7? If not, was there sufficient explanation? (QAPP, July 2009, p 13)	1. Samples were not collected in accordance with the QAPP schedule 2. Total P and BAP exceeded the expected range of results 3. BAP reported as 1 or 0 and less than reporting limit	Samples that measured outside the laboratory measurement methods were not qualified.
Were field quality control procedures followed? Was a blind duplicate sample collected and analyzed for each sample run? If not, was there sufficient explanation? (QAPP, July 2009, p 13)	No.	Scope and schedule are an integral part of the QAPP. Any deviations from the QAPP must be explained. The researcher must prepare procedures to ensure that the QAPP requirements are met.

1. Were there check standards/laboratory control samples, method blanks, analytical duplicates, matrix spikes, and matrix spike duplicates? 2. Were 10% of the samples duplicates? 3. Was the average algal density of four aliquots used to determine sample precision and accuracy? 4. Were similar aliquot procedures used to develop the standard curve (with precision and accuracy?) 5. What was the result of the laboratory blank? 6. What was the result of the blind duplicate? If not, was there sufficient explanation? (QAPP, July 2009, p 13)	1. Only method blanks and analytical duplicates were used. 2. No 3. Yes 4. Triplicate not four times. 5. Not recorded 6. Not done	Scope and schedule are an integral part of the QAPP. Any deviations from the QAPP must be explained. The researcher must prepare procedures to ensure that the QAPP requirements are met. Final report should contain a discussion of each element of the QAPP and whether or not those elements were met. If not, there should be a discussion as to why there was a deviation from the QAPP and the implication that has on the final results.
Are the field and laboratory data on the excel spreadsheets available? If not, was there sufficient explanation? (QAPP, July 2009, p 14)	Not provided	Raw data (field and laboratory data, chain of custody forms, QA/QC charts) should be provided as an appendix to the report.
Were quarterly progress reports submitted to Ecology? Were deviations to schedule explained? (QAPP, July 2009, p 14)	No. No.	The researcher is responsible for conducting the project in accordance with the requirements of the QAPP.

Does the final report contain: • Project goals • Methods used • Results of the research (QAPP, July 2009, p 14)	Yes.	No further action with respect to this item.
Does the final report contain a section of data verification and validation, including: • Procedures used to collect and record data • Chain of custody for samples between sample collection and data reporting • Laboratory quality control procedures • Discussion of "holding times" between removal of aliquots and actual completion of analytical procedures If not, was there sufficient explanation? (QAPP, July 2009, p 14)	No.	The researcher is responsible for conducting the project in accordance with the requirements of the QAPP. The researcher must prepare procedures to ensure that the QAPP requirements are met.
Does the final report contain a section on data quality assessment in which: • Data is evaluated in terms of its relationship to the expected norms of variability? • Were deviations from the norms explained? • Were limitations on data due to the deviations interpreted or conclusions drawn? If not, was there sufficient explanation? (QAPP, July 2009, p 14)	Some statistical analysis was provided based on the analysis of replicate samples. Assessment of the precision, accuracy, and reproducibility was not made. Some conclusions were drawn regarding deviations from the norm. The limitations on data were not discussed, other than reference to variations caused by low levels and the analytical detection limit.	Final report should contain a discussion of each element of the QAPP and whether or not those elements were met. If not, there should be a discussion as to why there was a deviation from the QAPP and the implication that has on the final results.

Was there an evaluation of the statistical error in the BAP estimates: • Uncertainty in the TP estimates for any particular sample • Error in the estimated intercepts and standard curves for the regression equations representing the relationship between the actual known phosphate concentrations and the algal cell density in the calibration curves • Statistical variability (the standard deviation of four replicate observations) of the results based on the four duplicate samples If not, was there sufficient explanation? (QAPP, July 2009, p 14)	Yes	No further action.
Were the standard deviations for triplicate measurements of the TP determined? Was the variation in the TP calibration curve represented by the outputs ($\pm$ 1 SD) for the statistical software (SPSS)? (QAPP, July 2009, p 14)	Yes	No further action
Was the bootstrapping technique used to account for variability in from all three sources to create a distribution of plausible independent estimates: • Random selection of TP value • Selection of likely standard curve • Selection of likely BAP value of original distributions • Repeat of process 1000 times (QAPP, July 2009, p 14)	No.	Scope and schedule are an integral part of the QAPP. Any deviations from the QAPP must be explained.

Unexpected complications occurred in evaluating some of the effluents and further work would be needed to resolve these issues (ECY 1/20/2011; cover letter)	Variations due to process operations occurred at several of the locations, which affected the schedule of the sampling and the analytical results. These were not fully addressed with respect to the actual conditions at the facilities at the time the samples were collected.	Scope and schedule are an integral part of the QAPP. Any deviations from the QAPP must be explained.
Request that comments on this study by other Spokane River stakeholders (dischargers, environmental groups, tribes, etc.) be made available for public review. (ECY 1/20/2011; p 1)	The comments made to this study were to be collected and added as an attachment to the Final Report.	Comments and response to comments should be an integral part of the final report.
All information available on the operation of the treatment process (effluent flow rates, chemical dosage rates, unusual operation conditions, etc.) of the facilities should be included in the report. (ECY 1/20/2011; p 1)	This information has not been provided.	Process conditions, and any deviations from normal operating conditions, must be fully explained within the report. Scope and schedule are an integral part of the QAPP. Any deviations from the QAPP must be explained.
Were additional split samples collected but not sent to UW for analysis? (ECY 1/20/2011; p 1)	No additional split samples were collected or sent to UW to analysis.	Scope and schedule are an integral part of the QAPP. Any deviations from the QAPP must be explained.
If split samples were collected and analyzed the dischargers should provide these results (including other parameters in addition to phosphorus) for inclusion into the report. (ECY 1/20/2011; p 1)	Researchers reported they did not have a mechanism to compel anyone else to provide data that was not collected and processed for their project.	Process conditions, and any deviations from normal operating conditions, must be fully explained within the report. Final report should contain a discussion of each element of the QAPP and whether or not those elements were met. If not, there should be a discussion as to why there was a deviation from the QAPP and the implication that has on the final results.
Please explain the significance of using KCl instead of K_2HPO_4. Is this a deviation from the standard methods? (ECY 1/20/2011; p 3, paragraph 2)	Reason provided is that this substitution (to create P-starved algae prior to the start of the experiment. Did not address the deviation from standard methods.	Scope and schedule are an integral part of the QAPP. Any deviations from the QAPP must be explained.
Please confirm that the samples were shipped to UW within established holding times. (ECY 1/20/2011; p 6, paragraph 1)	Confirmed in the response to comments.	Chain of custody procedures should be developed by the researcher so that there is documentation of the shipping procedures and adequate control of the samples.

It is unclear what the significance of the sample variability divided by the square root of the number of replicates processed is. Is this a standard way of showing low analytical uncertainty? (ECY 1/20/2011; p 10, paragraph 1)	No explanation was provided.	Provide further discussion in the QAPP (in the context of method and data quality objectives) regarding how data will be statistically evaluated for precision, accuracy, and reproducibility. Analytical uncertainty can be shown by evaluating the QA/QC samples (blanks, duplicates, spikes, matrix spikes, laboratory control samples) . . . which were not done. Procedures should be developed by the researcher so that there is documentation of that there is adequate control of the samples.
Identify which WWTP has the 17% variability (ECY 1/20/2011; p 10, paragraph 1)	Noted in response. Not included in the final report.	Comments and response to comments should be an integral part of the final report.
Is the high CV for BAP samples problematic or is this just a statistical outcome? Is seems that if the mean is low and the SD is also low, that's not a bad thing even if the CV is high. Should these instances be footnoted to the effect that these samples are not in fact problematic? (ECY 1/20/2011; p 11, paragraph 1)	Noted in response. Not included in the final report.	Final report should contain a discussion of each element of the QAPP and whether or not those elements were met. If not, there should be a discussion as to why there was a deviation from the QAPP and the implication that has on the final results. Comments and response to comments should be an integral part of the final report.
Please use the formal name of the City of Spokane WWTP (Riverside Park Water Reclamation Facility (RPWRF)) to distinguish it from other "Spokane WWTPs" throughout report, as per page 51. (ECY 1/20/2011; p 13)	Confirmed in response to comments and changed except for in the Executive Summary.	No further action.
Please add "with current (secondary) treatment methods" at the end of the first sentence discussing RPWRF. (ECY 1/20/2011; p 13, sentence 1)	Confirmed in response to comments.	No further action.
Do the pilot treatments come after the secondary clarifier? This is unclear as worded here.	Confirmed in response to comments.	No further action.

(ECY 1/20/2011; p 10, sentence 2)		
Identify that the colored boxes represent where samples were taken. Please add similar, consistent diagrams for other facilities (particularly where samples are taken). (ECY 1/20/2011; p 14, figure 3)	Confirmed in response to comments.	No further action.
This section does not clearly answer the question posed as to whether TP can be used as a conservative measure of %BAP in this pilot study. (ECY 1/20/2011; p 18, paragraph 3)	Confirmed in response to comments.	This should be addressed in the final report, with a statistical analysis as to the accuracy, precision, and reproducibility.
What are the units in this section? Are these numbers ratios? (ECY 1/20/2011; p 19, paragraph 2)	Confirmed in response to comments.	No further action.
Why is BAP/TRP relationship presented as a ratio in this figure and not in a regression such as in Figure 5? (ECY 1/20/2011; p 20, figure 6)	Confirmed in response to comments.	Comments and response to comments should be an integral part of the final report.
What is meant by a “sustainability perspective?” Depending on the expertise of the reviewing staff, sustainability perspective has been interpreted differently. One reviewer suggests checking with Prof. Dave Stensel to provide extra clarity and perspective to the statement. Alternately, section 0.3 of the USEPA Nutrient Control Design Manual, August 2010 could be consulted. (ECY 1/20/2011; p 20, paragraph 2, sentence 3)	Confirmed in response to comments.	Comments and response to comments should be an integral part of the final report.

Please refer to appropriate figure (Figure 5?) for the statement in the first sentence. It's unclear where this statement comes from since there is no statement that TP overestimates BAP elsewhere in the results section. Are the authors saying that TP, which is used in permitting, is assumed to be 100% bioavailable in wastewater treatment permits and that this is an overestimation? That would be a correct statement but BAP is a fraction of TP so TP is always going to be an "overestimate" of BAP. (ECY 1/20/2011; p 21, paragraph 2)	Confirmed in response to comments.	Comments and response to comments should be an integral part of the final report.
Figure 5 shows there's some relationship between TP and BAP but this section puts those findings aside and moves on to TP and BAP ratios without explaining why TP and BAP relationships can't be used. (ECY 1/20/2011; p 21, paragraph 2)	Confirmed in response to comments.	Comments and response to comments should be an integral part of the final report.
Define "protracted" as it relates to the reference cited. (ECY 1/20/2011; p 22, paragraph 1, last sentence)	Confirmed in response to comments.	Comments and response to comments should be an integral part of the final report.
It would be easier on the reader if you present the layout of the WWTP pilot treatment and where samples were collected first as you did for the City of Spokane samples. This section starts right off with results with no context or explanation of the treatment technology. (ECY 1/20/2011; p 24)	Confirmed in response to comments.	No further action.
Carry [above] suggestion thorough for remaining sections. (ECY 1/20/2011; p 24)	Confirmed in response to comments.	No further action.

Why were some samples composited and others were were grabs? Could spikes be missed or muted by either approach? (ECY 1/20/2011; p 26, paragraph 1)	Noted in response to comments. Spikes were not collected.	Comments and response to comments should be an integral part of the final report. Final report should contain a discussion of each element of the QAPP and whether or not those elements were met. If not, there should be a discussion as to why there was a deviation from the QAPP and the implication that has on the final results.
It is unclear how the BAP outliers are caused by mean BAP values approaching the analytical limits for the bioassay by looking at the values in Table 4c. In short, this last sentence doesn't make sense without further explanation. Is the quantitation limit several times the detection limit for other BAP tests as it is for most wet chemistry tests? (ECY 1/20/2011; p 29, paragraph 2)	Noted in response to comments. Quantitation limit not adequately discussed. Method data objectives from the QAPP were not met nor discussed.	Comments and response to comments should be an integral part of the final report. Final report should contain a discussion of each element of the QAPP and whether or not those elements were met. If not, there should be a discussion as to why there was a deviation from the QAPP and the implication that has on the final results.
Missing legend symbol for %BAP (ECY 1/20/2011; p 30, figure 13)	Confirmed in response to comments.	No further action.
Please verify whether first sentence is correct ("Prior to any treatment . . . "). Figure 7 shows that there is at least primary treatment prior to the treatment plant influent. Did you mean before the tertiary treatment for P removal? (ECY 1/20/2011; p 30, paragraph 3)	Confirmed in response to comments.	No further action.
Please highlight difference in pilot influent samples at Post Falls compared to City of Spokane and Coeur d'Alene samples. Post Falls influent is true, raw influent and not post treatment into a pilot facility. This should be mentioned in the opening paragraphs for the Post Falls chapter. (ECY 1/20/2011; p 32)	Confirmed in response to comments.	No further action.
Typo, strike word "that" following "one set of effluent samples (LLE) …" (ECY 1/20/2011; p 37, sentence 2)	Confirmed in response to comments.	No further action.

Replace word “located” with “taken [?]” (ECY 1/20/2011; p 37, paragraph 1, last sentence)	Confirmed in response to comments.	No further action.
Clarify whether there is any treatment prior to influent sample or, if like Post falls, the influent sample is raw sewage and the effluent samples are following existing treatment, not pilot (small scale) treatment technology. This point needs to be made very clear for facilities where raw effluent is tested because we are essentially looking at “scaled up” existing technology BAP removal performance at these two facilities (notwithstanding the outliers and low sample size). (ECY 1/20/2011; p 37, figure 18)	Confirmed in response to comments.	No further action.
Please describe what is meant by “quality of P in effluent.” Is this describing the composition of P species? (ECY 1/20/2011; p 40, paragraph 3)	Noted in response to comments.	Comments and response to comments should be an integral part of the final report.
Until more information becomes available from HARSB, it doesn’t seem useful to include any further report of this facility beyond the first paragraph. Suggest deleting rest of chapter after introduction on this page. (ECY 1/20/2011; p 41)	Did not follow suggested comment.	Comments and response to comments should be an integral part of the final report.
Suggest preceding the term “classic algal growth bioassay” with “as determined in this study using the …” to clarify that this study is in fact uses the classic growth bioassay. (ECY 1/20/2011; p 45)	Confirmed in response to comments, on page 51.	Comments and response to comments should be an integral part of the final report.
Clarify the type of particles being described; algae, sediment, other? Always precede term “particles” with “algae” to avoid confusion in this section please. (ECY 1/20/2011; p 45, sentence 2)	Did not follow suggested comment.	Comments and response to comments should be an integral part of the final report.

Is the “expected size distribution graph the typical pattern observed for other WWTPs in this study? In other words, this is an expected distribution for what? Wastewater effluent, streams, lakes, etc.? (ECY 1/20/2011; p 45, figure 26)	Confirmed in response to comments.	Comments and response to comments should be an integral part of the final report.
This paragraph needs a heading to reflect the conjecturing into low BAP from IEP being presented. Suggest “Potential causes of Low BAP” as the heading or something similar. (ECY 1/20/2011; p 46, paragraph 1)	Confirmed in response to comments.	No further action.
Add “pilot” between “advance” and “tertiary.” (ECY 1/20/2011; p 46, paragraph 2, sentence 2)	Confirmed in response to comments.	No further action.
Ecology agrees that IEP’s installation of a pilot plant is a “proactive commitment: but why is this term missing for the other treatment plants that have also installed tertiary pilot systems in advance of the TMDL? (ECY 1/20/2011; p 46, paragraph 2)	Noted in response to comments.	Comments and response to comments should be an integral part of the final report.
It would be helpful to have a treatment diagram for IEPs treatment system as the report has for the other systems. (ECY 1/20/2011; p 46, paragraph 2)	Confirmed in response to comments.	No further action.
What are the potential shortcomings of only having one influent sample? One sample doesn’t seem to be enough to characterize the quality. (ECY 1/20/2011; p 47, paragraph 2)	Noted in response to comments.	Comments and response to comments should be an integral part of the final report.
Last sentence is awkwardly worded. Please revise to something like “Our initial results suggest this effluent may be a poor substrate for . . .” (ECY 1/20/2011; p 47, paragraph 3)	Confirmed in response to comments.	No further action.

Same comment as the one regarding the influent sample. It really needs to be highlighted that there is only one influent sample to consider; more so than just saying "if one merely considers the result for the one influent sample. . ." The report makes much of the fact that there are a few samples for the other facilities but make little of the same situation for the influent at IEP. (ECY 1/20/2011; p 48, sentence 1)	Confirmed in response to comments.	No further action.
Typo, replace "like" with "likely." (ECY 1/20/2011; p 49, paragraph 3)	Confirmed in response to comments.	No further action.
Same comment as for page 46, last paragraph; this section needs a heading to clearly show authors speculation, discussion and conclusions as to what the likely causes of low BAP are in IEP effluent. (ECY 1/20/2011; p 49, paragraph 3)	Confirmed in response to comments.	No further action.
Please provide intro sentence as to why samples were collected from the river and lake; what was the objective for this part of the study (take from the QAPP?) In general, the report should have a consistent organization I all chapters, i.e., intro, sampling, results, conclusions. (ECY 1/20/2011; p 51)	Confirmed in response to comments.	No further action.
The correct term for the City of Spokane WWTP is introduced here but needs to be introduced at the beginning of the report and use the same term throughout the rest of the report. (ECY 1/20/2011; p 51, paragraph 1)	Confirmed in response to comments.	No further action.
Please provide exact locations of where Spokane River samples were taken. From which bridge, outfall, etc. (ECY 1/20/2011; p 51, paragraph 1)	Confirmed in response to comments.	No further action.

From where did the “upstream” concerns come from? What were the concerns (DO, algae, other)? How is upstream defined? Why was stateline chosen and not some other upstream location from Lake Spokane and the RPWRF (there are three other discharges between stateline and RPWRF)? Stateline was not a location from the QAPP. This needs to be clearly defined as to what the concern was, why this location was chosen and why it was sampled. (ECY 1/20/2011; p 51, last sentence)	Confirmed in response to comments.	Comments and response to comments should be an integral part of the final report.
Could there be another explanation for the high BAP in winter other than cessation of alum from the RPWRF? What about lake turnover or other seasonal factors that affect nutrient cycling? This should at least be acknowledged and discussed. (ECY 1/20/2011; p 52, paragraph 1)	Confirmed in response to comments.	No further action.
Regarding the statement “the algae bioassays indicated that most of the phosphorus was unavailable to algae,” an alternative explanation is that the most readily bioavailable phosphorus was already used by algae and macrophytes in the river. With the possible exception of the pools behind upstream dams, the water in the Spokane River is shallow enough that the entire water column is euphotic. Trying to determine what percentage of phosphorus *still in the water column* is bioavailable is uncertain under the best of conditions. In Lake Spokane, taking composite samples from the euphotic zone, thin interflow zone, and the hypolimnion give SRP/TP ratios of 16%, 82% and 86% respectively. This is not due to actual	Noted in response to comments.	Comments and response to comments should be an integral part of the final report.

differences in the bio-availability of the phosphorus, rather the fact that a portion of the available phosphorus has already been taken up by algae or macrophytes. (ECY 1/20/2011; p 52, paragraph 2)		
There should be a discussion about the fact that at the state line, the river is a losing reach to groundwater and you also have Post Falls dam upstream, which can act as a sink for algae and phosphorus before it hits state line. These factors should be considered in the evaluation of this one sample. The report should also mention that Ecology has a long data record for this and numerous other sites throughout the river, which provide a much better characterization of water quality than this one sample. (ECY 1/20/2011; p 53, paragraph 3)	Noted in response to comments.	Comments and response to comments should be an integral part of the final report.
Please define “raw sample.” Is this unfiltered river water? (ECY 1/20/2011; p 54, paragraph 1)	Noted in response to comments.	Comments and response to comments should be an integral part of the final report.
Typo, “Executive” Summary. This should be at the beginning of the report. (ECY 1/20/2011; p 56)	Confirmed in response to comments.	No further action.
Replace “very hard” with “impossible.” (ECY 1/20/2011; p 57, paragraph 1, last sentence)	Confirmed in response to comments.	No further action.

The composition of the bioassay samples for non-phosphorus constituents may be quite different depending on the ratio of media to effluent in the test sample, which would have been determined by . . . the initial phosphorus concentration. Thus, the difference in algae growth between the diluted "influent and intermediate process effluent samples" and the undiluted pilot plant effluent samples may be due at least in part to effluent toxicity or some other limiting factor as opposed to differences in the bioavailability of the phosphorus in the samples . . . While inconclusive, the Spokane River results are consistent with non-P limitation . . . river P comes from disparate sources including existing treatment plants, which produce 56-82% BAP . . . Thus the low BAP estimates for the river samples are unexpected . . . The high TRP in the river samples could be partially explained by an *in situ* limitation due to low water temperature and light availability . . . because samples are provided with ample light and warm temperatures during the assay, this does not explain the low BAP in the river samples. Previous studies have shown that upper Spokane River is N-limited. (EPA 2/25/2011; p 1, paragraph 1)	No response	Comments and response to comments should be an integral part of the final report.

The report does not acknowledge the possibility of non-P limitation for effluents containing low concentrations of phosphorus. (EPA 2/25/2011; p 2, paragraph 2)	In response to comment does not believe this to be the case but the only way to settle this is to conduct follow-up experiments. (UW, 2/28/2011 paragraph 4)	Aluminum and Zinc can be limiting factors and have been found in local effluents in concentrations that could be toxic. The Spokane River also contains relatively high concentrations of metals (cadmium, lead, and zinc.) (EPA 3/10/11; paragraph 4) Phase II proposal addresses this concern.
It is not clear on Page 6 whether the threshold P concentration above which samples were diluted is 100 or 50 µg/L, nor is it clear whether the dilution threshold is based on TP or TRP. The report should be edited to clarify which samples were diluted. (EPA 2/25/2011; p 2, paragraph 2)	No response.	Comments and response to comments should be an integral part of the final report.
[The data suggests that] low carbon content for the advanced treatment effluents, which can influence algae growth in bottle tests, and effluent micronutrient concentrations are unknown. The report should acknowledge the possibility on non-P nutrient limitation in undiluted, low-P samples from both advanced wastewater treatment effluent and from the river. (EPA 2/25/2011; p 2 paragraph 3)	Algae in closed bottle tests might be carbon (CO_2) limited, but for this experiment the bottles were open and continuously shaken. (UW, 2/28/2011 paragraph 5). Noted and acknowledged by EPA. (EPA 3/10/2011, paragraph 3).	While low N and C concentrations and toxicity can influence algae growth in bottle tests of undiluted samples, the effluents will be diluted by the receiving water, and natural processes can compensate for deficiencies if N and C in lakes and reservoirs . . . these factors, which may have influenced assay results, will not be present in the environment. Therefore, the possibility of limitation by nutrients other than P or the presence of toxicity in undiluted effluents from advance treatment facilities must be ruled out or controlled for before the results of this study could be used to inform regulatory decisions. (EPA 2/25/2011; p 3 paragraph 3)
Another option [for testing non-P limitation] would be to adapt the procedure described in EPA's whole effluent toxicity (WET) test for green algae [without adding P]. (EPA 2/25/2011; p 2 paragraph 4)	Agrees with comment, would need to first determine how many samples are sufficient. Would it be enough to run these experiments only once for each effluent type tested in the initial experiment? (UW, 2/28/2011 paragraph 2)	Determining the number of samples is a balancing act between cost and minimizing uncertainty. (EPA 3/10/11; last paragraph)
The report should acknowledge the possibility of toxicity for all low-P effluents requiring little or no dilution prior to the assay, not just those from IEP. (EPA 2/25/2011; p 3 paragraph 2)	No response.	Comments and response to comments should be an integral part of the final report.

[regarding the toxicity of effluents to algae] Another option would be to test the effluents for toxicity using EPA Method 1003.0. (EPA 2/25/2011; p 3 paragraph 2)	Agrees with comment, would need to first determine how many samples are sufficient. Would it be enough to run these experiments only once for each effluent type tested in the initial experiment? (UW, 2/28/2011 paragraph 2)	Determining the number of samples is a balancing act between cost and minimizing uncertainty. (EPA 3/10/11; last paragraph)
The use of cultured algal species provides little insight into how complex natural assemblages adapted to nutrient supply conditions of their native habitat would respond to N and P availability . . . the effluents being tested are ultimately discharged into Lake Spokane . . . P that is not initially bioavailable can become bioavailable over time under certain conditions. (EPA 2/25/2011; p 3 paragraph 5)		The "whole lake experiment" of installing treatment and watching water quality improve will be the ultimate test of the BAP study (and the model, TMDL, and permits.) (EPA 3/10/11; paragraph 2)
The fact that N and P chemistry constantly changes in the environment is the reason EPA recommends nutrient water quality criteria and monitoring be based on total P and total N. (EPA 2/25/2011; p 4 paragraph 1)	If this is the official position of the EPA, then it is simply wrong. (UW, 2/28/2011 paragraph 6)	EPA's nutrient criteria have not changed over the last 10 years. EPA's position has not changed on this since nutrient criteria were recently promulgated for Florida using total P and total N. (EPA 3/10/11; last paragraph)
The report should acknowledge the limitations on the ability of a small-scale, short term bioassay using a cultured algal species to accurately predict the impact of the effluents upon natural waters. (EPA 2/25/2011; p 4 paragraph 1)	No response.	Comments and response to comments should be an integral part of the final report.
IEP Test Data, 2011 Daily sample collected for a minimum of 14 days beginning on April 26, 2011. (Additional IEP Data Test Plan, 4/26/2011)	Data not received	Data should be submitted as requested.
IEP will operate the Trident system each day for a time sufficient to collect a minimum of 8 samples approximately one hour apart. (Additional IEP Data Test Plan, 4/26/2011)	Data not received	Data should be submitted as requested.

Composite samples will be collected over approximately an 8 hour period (once/hour) with each sample being at least .25 liter, in accordance with the test plan requirements. (Additional IEP Data Test Plan, 4/26/2011)	Data not received	Data should be submitted as requested.
The 3rd party lab will provide all appropriate Chain of Custody documentation, in accordance with the test plan requirements. (Additional IEP Data Test Plan, 4/26/2011)	Data not received	Data should be submitted as requested.
Samples will be analyzed by a lab accredited for o-phosphate and TP using method SM4500-PE/PF in accordance with the reporting limits specified in the test plan. (Additional IEP Data Test Plan, 4/26/2011)	Data not received	Data should be submitted as requested.
At least 4 replicate samples shall be submitted to the lab over the test period . . . The replicate samples shall be collected from the composite samples approximately once every three to four days, in accordance with the procedures in the test plan. (Additional IEP Data Test Plan, 4/26/2011)	Data not received	Data should be submitted as requested.
The system will be operated for 14 days, ending approximately Tuesday, May 10 (assuming no operational difficulties, mill outages, or conditions that would result in unrepresentative samples. (Additional IEP Data Test Plan, 4/26/2011)	IEP had some troubles with the sampling event and getting some weird results (SRP higher than TP). They'll keep pursuing o-phosporus but over the first cycle.	Data should be submitted as requested
Daily samples were collected for a minimum of two weeks. (Email ECY to IEP, 4/21/2011)	Data not received	Data should be submitted as requested.
Samples will be composited with a minimum of 8 subsamples each. (Email ECY to IEP, 4/21/2011)	Data not received	Data should be submitted as requested.
Include a minimum of four split samples, distributed evenly through time. (Email ECY to IEP, 4/21/2011)	Data not received	Data should be submitted as requested.

Provide documentation of appropriate sample collection/handling procedures (e.g., holding times, sample preservation, filtering, bottles, etc.) (Email ECY to IEP, 4/21/2011)	Data not received	Data should be submitted as requested.
Samples will be analyzed by a lab accredited for o-phosphate and TP using method SM4500-PE/PF. Reporting limit for o-phosphate < 0.003 mg/L. Reporting limit for TP < 0.10 mg/L (Email ECY to IEP, 4/21/2011)	Data not received	Data should be submitted as requested.
The upper confidence limit for the mean o-phosphorus fraction will be calculated based on a one-sided t-distribution and a 95% confidence level (t0.05(1)[n-1]) using individual sample o-phosphate fractions. (Email ECY to IEP, 4/21/2011)	Data not received	Data should be submitted as requested.
Statistical calculations will substitute one-half the reporting limit for samples below the reporting limit. (Email ECY to IEP, 4/21/2011)	Data not received	Data should be submitted as requested.
The TMDL model will be run changing only the IEP effluent limit (70 ppb seasonal average TP) and the o-phosphorus fraction using the upper confidence limit calculated above. (Email ECY to IEP, 4/21/2011)	Data not received	Data should be submitted as requested.
Equivalency will be determined based on the same criteria used for the seasonal limits evaluation (posted on the alternate seasonal limit web page). (Email ECY to IEP, 4/21/2011)	Data not received	Data should be submitted as requested.

Table 1: Comparison of QAPP Requirements vs. Actual measurements

Parameter	Check stds/LCS	Duplicate samples RPD	Matrix Spikes Recovery	Matrix Spikes Duplicates (RPD)	Lowest concentration
TP	Not done	Not calculated	Not done	Not calculated	➢ MQO
TDP	Not measured	Not measured	Not measured	Not measured	Not measured
SRP	Not measured	Not measured	Not measured	Not measured	Not measured

Table 2 QAPP Proposed Sample schedule (blue = proposed) vs. Actual samples (X = sample)

Site	**8/9**	**9/9**	**10/9**	**11/9**	**12/9**	**1/10**	**2/10**	**3/10**	**4/10**	**5/10**	**6/10**	**7/10**	**8/10**	**Proposed/ Actual Events**	**Actual Samples**
SR 9 mile	X	XX		X	X									4/5	5
3 Springs														4/0	0
SR State line								X						0/1	1
Spo Kruger														9/0	0
Spo "Influent"	X	XX	X	X	X			X	X					0/8	8
Spo Co Mag	X	XX	X	X	X			X	X					3/8	8
Spo Zenon membrane filtration	XX	XX	XX	XX	XX			XX	XX					4/7	14
Spo Corix conv sedimentation	XX	XX XX	XX	XX	XX			XX	XX					3/7	16
Spo Blue Water cont upflow filter	XX	XX XX	XX	XX	XX			XX	XX					0/7	16
Spo Corix MM Granular filt.	XX	XX XX	XX	XX	XX			XX	XX					0/7	16
CdA Influent										X	X X	X	X	0/5	5
CdA Blue Water cont upflow filter										X	X X	X	X	0/5	5
CdA Zenon micro filt														4/0	0
CdA Zenon memb filt										X	X X	X	X	0/5	5
CdA Zenon Memb bio rxtr										XX	XX XX	XX	XX	0/5	10
IEP Influent											X			0/1	1
IEP Trident HS		X X	X	X	X			X	X		X			5/8	8
Post Falls										XX	XX XX	XX	XX	0/5	10
LLSWD									XX	XX	XX XX	XX	XX	0/6	12
HARSB										XXX	XX	XX	XX	0/5	9

TOTAL	11	22	11	12	12	0	0	12	13	12	22	11	11	32/91	145

Table 3 Comparison of QAPP Proposed Laboratory Methods vs. Actual

Analyte	Samples 12/monthly	Expected Range of Results/Actual	Reporting limit	Above reporting limit	Sample Preparation Method	Analytical Method
Total P	No	0-100 ppb/7-8444 ppb	2 ppb	yes	yes	yes
TDP	Not analyzed	n/a	n/a	n/a	n/a	n/a
SRP	Not analyzed	n/a	n/a	n/a	n/a	n/a
BAP	No	0-50 ppb/0-5075 ppb	2 ppb	usually		yes