Response to ISRP Review Comments: Project ID 35029
Transfer IHN virus genetic strain typing technology to fish health managers

Comment 1: the tasks associated with the technology transfer do not include "blind" tests of virus that the regional labs would process in order to confirm the accuracy of their work. Since several regional labs would be trained in these needed techniques, provided the necessary equipment, and then expected to contribute to the monitoring and control of the IHN M-clade, we suggest that regular testing for confirmation of methods should be incorporated.

Response 1: This is an excellent suggestion, and it is easy to incorporate it fully into the proposal without any modification of the budget. We would add the following "blind" testing to our current plan. As each new lab becomes trained and equipped and ready to analyze their own virus samples, we would first send them three unknown virus isolates to sequence. The isolates would be selected by the staff person dedicated to this proposal, from the virus isolates typed recently in our own laboratory. Final sequence results from the trainee lab will be sent to us for alignment and confirmation of identical sequences. Where any discrepancies occur we will request the raw sequence profiles from the trainee and compare the un-edited data with our own profiles, to determine if the error occurred in sequence generation or in the editing and analysis steps. We will work with the individual researcher to identify and correct the problem, and then re-test with the same three isolates and one new isolate. Once exactly identical sequences are generated, we will send two blind test isolates every 3 months during the first year of activity for each trainee lab. If no errors occur we will thereafter test twice a year with two isolates. Where errors occur we will troubleshoot to identify the problem, and then continue with quarterly testing until the data generated by the trainee lab is consistently accurate. This will provide both us and the trainee laboratories with a high level of confidence in all data generated and added to the IHNV database.

Comment 2: The authors suggest that the overall goal of this work is to document the distribution of the M-clade and control its spread in the Columbia basin. The proposal is not very explicit, however, in how prevention or control of the spread would result. This should be more clearly explained in the proposal.

Response 2: The data generated in this proposal will be used to prevent or control the spread of M-clade IHNV through support of educated management decisions by the state and federal Fish and Wildlife agencies whose laboratories we will train in this proposal. Furthermore, we can initiate dialogue regarding recommendations for management strategies. We envision that the strain typing information generated will be used in making management decisions that impact M-clade virus control at individual hatchery facilities, and also on a broader management agency scale. Research in the focus sites (Objective 2) will also provide new information to managers. These sites were selected specifically to address the issue of management of M clade IHNV.

From an individual hatchery perspective, facility managers confronted with IHNV epizootics must consider various possible routes of contamination: virus in the water supply,

importation of infected fish, horizontal transmission from other fish stocks at the facility, vertical transmission from infected eggs, or contamination via equipment or personnel. The strain typing we will do in objectives 1 and 2 of this proposal, and later the typing done by other labs, will provide some idea of which of these routes is most probable in a specific case, thus providing direction for the most efficient use of available funds and personnel to reduce or eliminate the virus. For example, if virus is in the water supply, then efforts to eliminate carrier fish above the intake, or ozonation of the intake water should be undertaken. In contrast, if vertical transmission or contamination from within the hatchery are is found to be most likely, careful examination and improvement of the iodination procedures and hatchery hygiene would be more effective. Although these strategies will improve control of all IHNV strains detected, we would propose that if an M-clade virus is found this should be given greater effort and a higher priority for management changes and expenditure of money and personnel time, to maximize the probability of eliminating the virus. On an overall scale, this higher priority given to M-clade viruses should more effectively contain M clade viruses that appear in the basin outside the Hagerman Valley. Once virus strain typing data is provided to individual hatchery managers, we would propose that they consider more rapid and strict quarantine and/or destruction of fish stocks experiencing an M-clade epizootic or a high M-clade IHNV carrier rate. This suggestion will most likely be presented to hatchery managers by the fish health management agency personnel responsible for the facility, but the staff person funded by this proposal will be also be available for assistance as necessary.

On a larger scale, we anticipate that the staff of the fish health management agency laboratories trained in this proposal will become the most effective agents of M-clade virus control in the basin. They are responsible for management decisions regarding virus sampling, fish quarantine and/or destruction, and approval of fish transports within the basin. Each of these activities should be more strictly controlled when an M-clade virus is involved. In management decisions regarding fish transportations there are often many complex factors involved in the final decision, and fish health risks is only one of those factors. We would suggest that when an M-clade IHNV is detected, the importance of fish health risks should be elevated above many or all of the other factors. The most critical example would be proposed movements of fish stocks to sites in the middle and upper Columbia basin above the confluence with the Snake River. Since no M-clade IHNV has ever been detected in this large portion of the basin it is imperative that all fish to be moved there be thoroughly tested, and if M-clade virus is detected, the transport should be prevented.

These would be our recommendations, but we recognize that fish health workers and agency managers are likely to have more concrete and insightful ideas that will modify and improve their utility. We propose that this should be included as a topic of discussion at fish managers meetings, and these should be attended by the staff person supported by this proposal. The initial goal would be an informal set of recommendations aimed at control of M-clade IHNV in the basin. This will be written by our staff person, and continuously modified and improved in response to discussions with fish health managers. This informal document would be distributed to all trainee labs and any other interested parties, and eventually it may become more formal if the managers in the basin so desire. Since this M-clade control was part of the original proposal, no budget modifications would be necessary. The travel budget will be sufficient to allow the staff person to attend manager meetings on an annual basis.

Comment 3: We ask the proponents to consider a more active investigation of the M-clade distribution and control of its spread. The authors make a good case for the importance of this research and monitoring, but if the spread of M-clade is a threat to recovery, why not take an immediate active role in sampling and examination of the current distribution and then management of the virus? The budget could be adjusted appropriately.

Response 3: This is another excellent point indicating the need to more clearly define the intent of the original proposal. Although not explicitly stated as a separate objective, the elements of an active role in M-clade control were intended to be included in Objectives 1, 2 and 4 of the proposal. We are unlikely to undertake an active role in virus sampling, because this would be redundant with the already thorough sampling programs of the state and federal fish health agencies. These colleagues sample every spawning fish stock and every disease epidemic at all fish culture facilities in the basin, and they also sample wild fish populations whenever possible. We have always found them very willing to take additional virus samples when our research indicates it would be useful, and their collaboration is a major asset to this work. As they come to understand the high importance of M-clade IHNV, I have no doubt that we can agree on optimal sampling strategies to assure its accurate assessment. Thus, I see the staff person dedicated to this proposal spending the majority of their time assimilating all strain typing data and specifically focussing on the current distribution of M-clade IHNV in the basin, and on evaluating the success of control strategies undertaken during the course of the proposal. This will involve collating information from strain typing done in our own laboratory as case studies (objective 1) or more in-depth focus studies (objective 2) with data submitted from other laboratories once they are trained and active. By year 2 of the proposal we should have a reasonable idea of the M-clade virus distribution, which will be communicated to fish health managers at meetings as described above. Active management for M-clade control would be undertaken through development of a set of recommendations for management decisions involving M-clade virus, tapping the expertise of basin fish health managers as described in the response to comment 2. In this manner, management for control of M-clade virus will continue past the three year duration of this proposal, as will no doubt be necessary. Again, since this was part of the original intent of the proposal, no budget modification is necessary.