Center for Independent Experts (CIE) Independent Peer Review Report

Petition to list alewife (*Alosa pseudoharengus*) and blueback herring (*Alosa aestivalis*) ("River Herring") under the Endangered Species Act (ESA)

Review of Stock Structure and Extinction Risk Analysis Working Group Reports

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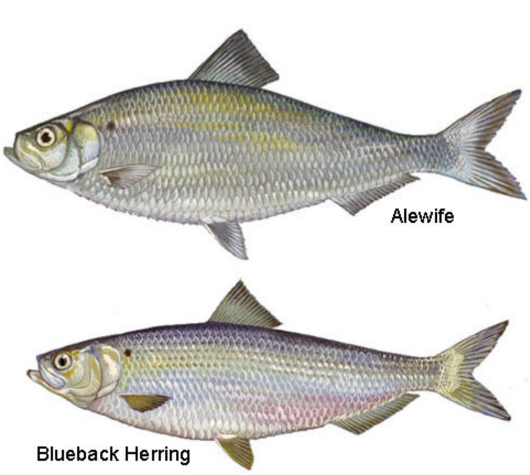

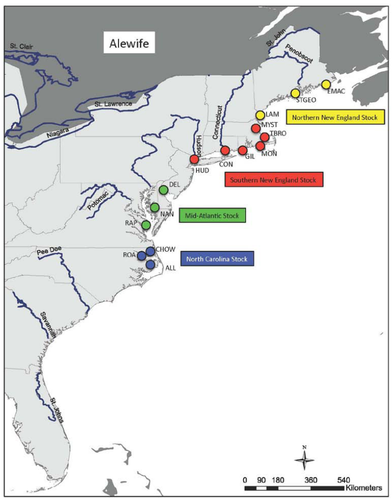

Executive Summary

River herring, a collective term for two species of anadromous fishes found along the Atlantic coast of North America, from the maritime provinces of Canada to the SE United States, Alewife (*Alosa pseudoharengus*) and blueback herring (*Alosa aestivalis*), represent among the oldest fisheries in the USA. They also comprise important components of both freshwater and marine ecosystems within which they serve as key predators and forage as prey. Despite their historical commercial importance, various factors relating to habitat disturbance and degradation, over-exploitation, bourgeoning by-catch mortality, and accelerating effects of climate change, both species have suffered marked population declines (>90% in some localities from the 1950 to 1970 average) and contraction in geographic range. In 2006 the National Marine Fisheries Service (NMFS) designated river herring as a *Species of Concern*, followed in August 2011 by a petition from the Natural Resources Defence Council (NRDC) to the NMFS to list both alewives and blueback herring, each as threatened throughout all or a significant portion of their range under the Endangered Species Act (ESA). If designation was not appropriate at the species level, alternatively there was a request that NMFS designate distinct population segments (DPS) of alewife and blueback herring as specified in the petition (Central New England, Long Island Sound, Chesapeake Bay and Carolina for alewives, and Central New England, Long Island Sound, and Chesapeake Bay for blueback herring). In November 2011, NMFS issued a positive 90 day finding indicating that the NRDC petitioned action may be warranted. In accordance with usual procedures relating to the ESA, it was necessary for the NMFS to review the best scientific evidence available to develop a listing determination.

NMFS agreed to use such information, together with that presented at three expert and consultative workshops covering areas of identified gaps in knowledge: (i) stock structure, (ii) extinction risk analysis (ERA) and (iii) climate change. The focus of the current CIE Expert Review is on (i) and (ii) only, though additional information on climate change taken from the literature and salient reports, and identified as such, will be referred to as appropriate. NMFS will use the information from these workshops to assess whether there are discrete and significant populations of alewives or blueback herring that might warrant separate protections under the joint US Fish and Wildlife Service and NMFS Distinct Population Segments (DPS) policy (61 FR 4722). Upon applying the DPS policy, the evidence gathered at the stock structure workshop will help NMFS to make an informed decision on whether the stock structure can adequately be protected as a single unit or, whether one or more DPS are necessary to best protect certain stock complexes of alewives or blueback herring that represent a discrete and significant unit to the taxon as a whole.

The remit of the current CIE Expert Review, conducted 20 August – 3 September 2012 as a desk review, was to provide a scientific peer review of Stock Structure and ERA reports prepared by the NMFS. The aim was in accordance with the terms of reference requesting objective opinions on whether the best scientific evidence available was contained within the reports and whether conclusions drawn from such data were supported sufficiently. It was anticipated initially that some estimates of extinction probability would be available for peer review, but such data were not forthcoming within the available time. Thus, the Reviewer's role for the ERA

component is focused on the validity of data to be used in the modelling and the rationale for selecting this information, together with the appropriateness of the methodology to be employed. Specifically, consideration will be given within the context of ToRs whether the decisions and recommendations proposed for the modelling approach and the input data are scientifically sound and appropriate for assessing extinction risk for river herring.

Evidence presented to assess stock structure derived from various sources, encompassing genetics (microsatellite diversity), life history, population dynamics, physiological, behavioural and morphological variation. Ecological sampling utilised past and Review-specific data, and in addition to standard survey and sampling approaches along the US Atlantic coast, including some information on selected Canadian populations, microchemistry data from otoliths were available for some populations. Among data sources, genetic evidence was the most coherent and robust available, enabling the testing of specific hypotheses relating to alternative models of stock structure – whether each species represented a continuous single stock complex throughout the entire range from the US to Canada, or whether evidence supported the existence of discrete groupings appropriate for consideration under DPS policy. While the genetic information was not exhaustive, and focused on spatial relationships among samples only, with no temporal (time-series of samples) component to explore stability of observed population structuring, significant genetic differentiation was detected, allowing the designation of populations into regional groupings. Considering the single or multiple stock complex hypotheses, genetic data supported strongly the latter, with a final proposition that alewife comprised five detectable stocks: Carolina (all alewife rivers south of, and including the Chowan River); Mid-Atlantic (all Virginia waters up to, and including New Jersey waters); Southern New England (all New York waters up to, and including Massachusetts waters) ; Northern New England (Lamprey up to and including the St. Croix River); Canada (all Canadian Rivers), and blueback herring comprising five stock complexes also: Southern (St. John River to Cape Fear River), Mid-Atlantic (Neuse River to Connecticut River); Southern New England (Gilbert-Stewart to Mystic River); Northern New England (Exeter River up to and including St. Croix River); Canada (all Canadian Rivers). While other non-genetic data were not appropriate for defining putative stock boundaries, they did support the case for discrete stocks of each river herring species. The putative boundaries proposed are essentially based on genetic data alone and represent geographically-defined units, each comprising a significant component of the biological diversity of the respective species, though such stock units should be viewed as a likely minimum number of complexes. It is very likely that biologically significant heterogeneity exists within each of the proposed categories. Such is the level of stock structuring that loss of individual components would represent a significant gap in the range of the respective species. Moreover, there is clear separation of alewife and blueback herring as discrete and demographically independent entities, each with their own suite of biological properties that impart differential responses to environmental threats, thereby requiring separate consideration under the ESA.

After considering a range of abundance survey data and life history information depicting the current knowledge of river herring distribution, biology and structure of putative stock complexes, it was proposed to undertake an ERA utilising the Multivariate Auto-Regressive State Space (MARSS) package. Probability of populations/stocks persisting into the future is to be quantified, and where possible, the effects of various environmental factors on such probabilities will be assessed. Only a brief preliminary analysis to establish efficacy of the approach was included within the ERA Working Group Report. The choice of input data, partitioned into previously defined stock complexes, was sound, and the MARSS approach is well suited to river herring through an ability to decompose probability estimates of extinction according to the various stock complex scenarios, and by accommodating the incompleteness of biological input data and the variable duration of time-series available.

Overall, the best available scientific and commercial data on stock structure and ERA for river herring were contained within the two submitted reports, utilising appropriate methodologies for the biological scales under study (between-species and intraspecific biological diversity). Additional genetic analysis on existing and future data are, however, recommended to better refine the spatial and/or temporal distribution and dynamics of putative DPS by the application of additional sampling, demographic analyses, a mixed stock analysis of the marine phase of river herring, and improved assessment of the impacts of stocking and hybridization between alewife and blueback herring. It is vitally important that all existing and forthcoming genetic data (and any associated trait data) be reassessed for the potential inclusion of misidentified specimens or the inclusion of hybrids. The apparent on-going confusion in identification of alewife and blueback herring must not be underestimated in terms of potential impact on DPS designation. As argued within the Report, at relatively low cost, such issues can be readily addressed by occasional use of high throughput independent verification of species status such as DNA barcoding.

Although the NMFS has determined that the NRDC pertains to anadromous populations of river herring only, the Reviewer recommends that the status of landlocked alewife be reassessed based on recent genetic evidence, not included in either Report, indicating multiple and independent origins of landlocked forms from anadromous counterparts, and the marked genetic and ecological divergence between these two forms over only 100-300 years. Landlocked and anadromous river herring should not be viewed as interchangeable forms of the same biological entity, and crossing the two forms for whatever purpose is strongly discouraged.

In general, the Stock Structure and ERA Working Reports provided a detailed and balanced overview of the key threats pertinent to ESA listing, and the best available data for determining 1) whether there is evidence of stock structure for alewife and blueback herring – the evidence supports strongly the existence of such stock structuring appropriate for DPS policy; and 2) the provision to NMFS of expert opinions on the extent (if any) of stock structure for alewife and blueback herring – the evidence (primarily genetic) supports the delineation of putative groupings that contain fish with genetically and biologically distinct attributes such that their loss would represent a significant gap in the range of the respective river herring species.

1. Background

1.1 Original time-line, context and scope of petition

Alewife (*Alosa pseudoharengus*) and blueback herring (*Alosa aestivalis*), two anadromous fishes collectively known as "river herring", represent among the oldest fisheries in the USA. They are found along the Atlantic coast of North America, from the maritime provinces of Canada to the SE United States (Mullen *et al*., 1986; Shultz *et al*., 2009). Despite their historical abundance (NRDC, 2011), populations of both species are estimated to be a fraction of their former size: Overall coastal landings of alewives and blueback herring have averaged approximately 1 million pounds over the last decade, a decline of more than 98% from the 1950 to 1970 average. A recent overview of Atlantic States Marine Fisheries Commission River Herring Stock Assessment, reported in the River Herring Climate Change Workshop Report (NMFS, 2012c) concluded that of the 52 stocks of alewife and blueback herring for which data were available, 23 were depleted relative to historic levels, one stock was increasing, and the status of 28 stocks could not be determined because the time-series of available data was too short. For example, while the status of Maine-based alewife populations appears to be better than that of more southerly stocks, the region has undergone marked contraction in freshwater range. It is worth noting, however, that the Stock Structure Working Group acknowledged that historical records, and even some recent records, for the two species taken separately likely do not distinguish alewives and blueback herring accurately because of similarities in their morphology. Nevertheless, there has been an undisputable marked decline in the abundance of river herring taken collectively. The fragmentation of habitat through historical damming reduced the potential habitat available to alewives by 95% by 1850 (Hall et al. 2011). Construction of dams and other impoundments, habitat degradation, disease, exploitation, the continued inclusion of river herring as by-catch and striped bass predation were all proposed as key factors associated with their decline. The original NRDC petition (NRDC, 2011) also highlighted the perceived insufficiency of existing regulatory authorities to manage river herring. In addition, there is increasing current concern about the impact of global warming, with potential impacts on recruitment, distribution, water quality and increased frequency of flooding.

In 2004 NMFS created the *Species of Concern* (see http://www.nmfs.noaa.gov/pr/species/concern), which encompass those species about which NMFS has some concerns regarding status and threats, but for which insufficient information is available to indicate a need to list the species under the Endangered Species Act (ESA). River herring were included under this category in 2006. In August 2011 NRDC petitioned the NMFS to list both alewives and blueback herring, each as threatened throughout all or a significant portion of their range under the Endangered Species Act (ESA). If designation was not appropriate at the species level, alternatively there was a request that NMFS designate DPS of alewife and blueback herring as specified in the petition (Central New England, Long Island Sound, Chesapeake Bay and Carolina for alewives, and Central New England, Long Island Sound, and Chesapeake Bay for blueback herring).

In November 2011, NMFS issued a positive 90 day finding indicating that the NRDC petitioned action may be warranted. In accordance with usual procedures relating to the ESA, it was necessary for the NMFS to review the best scientific evidence available to develop a listing determination. The Atlantic States Marine Fisheries Commission (ASMFC) completed a stock assessment for river herring in May 2012, covering over 50 river specific stocks throughout the U.S. range of both species (ASMFC, 2012). NMFS agreed to use such information, together with that presented at three expert and consultative workshops covering areas of identified gaps in knowledge: (i) stock structure, (ii) extinction risk analysis and (iii) climate change. The focus of the current CIE Expert Review is on (i) and (ii) only, though additional information on climate change taken from the literature and salient reports, and identified as such, will be referred to where appropriate. In terms of stock structure the key issue was to ascertain under the ESA's DPS policy whether each species can adequately be protected as a single unit, or whether one or more distinct population segments are necessary to best protect certain stocks of alewives or bluebacks that represent a discrete and significant unit to the taxon as a whole. Information from diverse approaches was considered, including genetics (Atlantic coast-wide and finer scale between the state of Maine and Canada), life history (e.g., length at age, growth rates), abundance (e.g., run times, run counts, discards), evidence of straying and homing, tagging studies, stocking history, physiological variation and morphometrics. While the original NRDC petition specified species-specific stock structure hypotheses (number and identity of proposed DPS), experts at the stock structure workshop considered a range of stock structure hypotheses in relation to the lines of evidence presented. Utilising evidence presented in the stock structure workshop, an extinction risk analysis of alewife and blueback herring was assessed through a critical evaluation of the evidence to date and range of models that most effectively estimate extinction risk.

1.2 Stock structure, distinct population segments (DPS) and the Endangered Species Act

In relation to the current review, the concept of stock structure – its detection, spatial and/or temporal distribution and its role in the viability and persistence of natural fish resources in the face of environmental change, is core to both to the designation of so-called Distinct Population Segments (DPS), and the associated ERA. The perceived stock structure typically informs the appropriate spatial/temporal scale for the subsequent ERA, with critical decisions about whether to split or lump together various within-species groupings. The concept of stock structure is aimed at capturing the appropriate biological scale of diversity that most effectively encompasses the range of reproductively and biologically distinct assemblages within a given species (Carvalho & Hauser, 1994). Reproductive groupings, or in the context of river herring, spawning groups, especially those with significant natal homing, are likely to generate marked biological and genetic differentiation across their range – such that the resulting population diversity, also termed "biocomplexity", endows the species with enhanced flexibility and capacity to adapt to environmental change (Hilborn *et al*., 2003). In relation to river herring, taking each species separately, the challenge considered here is to what degree the best available scientific evidence (ideally from disparate methods) indicates the existence of stock structure and over what spatial or temporal scale should the boundaries of such distinct units be made? To conserve overall levels and patterns of genetic diversity within species as a resource for future adaptive change, the ESA encourages the designation of DPS (Waples, 1991; Kelly, 2010). The US Fish and Wildlife Service (USFWS) in 1996 developed the policy for determination of DPS for protection under the ESA (USFWS, 1996), focusing on three elements to designate a DPS: on the basis of their discreteness from other such units, by their perceived “significance” to the respective species they represent, and on the conservation status of a unit. Three significance criteria that are especially relevant in the current context are: (1) the persistence of a unit in an ecological setting that is unusual or unique for the taxon; (2) evidence that loss of the unit would result in a significant gap in the range of the taxon; (3) evidence that the unit differs markedly in its genetic characteristics relative to other populations of the same species. Discreteness in general can be estimated by using several approaches such as the use of genetics, life history data, demographic trends and morphological diversity, whereas significance equates to the degree to which a specific DPS encompasses a unique ecological setting or whose loss would result in a significant gap in the species' range (e.g. Grunwald *et al*., 2008 for sturgeon; Arden et al, 2011: for bullhead). A key challenge that faces those in recommending the listing of specific DPS is how to identify, for example in the current context for river herring, at what hierarchical level do any population groupings identified by specific data (or combined data sets) reach the appropriate boundary of discreteness and significance to the taxon? Compared with the other biological levels typically used by federal agencies in listing endangered or threatened species (species, sub-species), the notion of DPS are in reality more the construct of legal processes rather than biological (Kenny, 2010). As such, there is no common threshold or level of discreteness among identified DPS that is universally applicable across all species and scenarios, and it becomes crucial to examine critically the evidence that indicates an appropriate scale of boundaries and potential significance to the taxon. Such is the overall aim of the current review, to consider in the context of the ESA designation of DPS, whether and at what spatial scale, the existing scientific evidence identifies the case in support of DPS, or indeed, whether alewife and blueback herring can be listed as a single unit respectively. The findings are directly relevant to the subsequent ERA in determining the most appropriate spatial scale and framework to generate time-dependent extinction probabilities.

2. Description of Individual Reviewer's role in Review Activities

2.1 Nature of review

The original NRDC petition required that the NMFS should list the alewife and blueback herring as threatened species as a whole. The claim was that they represent unitary species likely to become endangered within the foreseeable future throughout all or significant portions of their ranges, including rivers in Maine, New Hampshire, Massachusetts, Connecticut, the Chesapeake Bay and its tributaries, and many coastal river systems in the Carolinas. If NMFS does not list the alewife and the blueback herring each as a threatened species as a whole, the agency proposed the specific listing of DPS. The present CIE Review was conducted as a desk peer review based on the submission of various documents described in Appendix 1 and was undertaken from 20 August – 3 September 2012. NMFS will use the information from the stock structure workshop to assess whether there are discrete and significant populations of alewives or blueback herring that might warrant separate protections under the ESA's DPS policy.

2.2 Terms of Reference

The remit was to provide a scientific peer review of Stock Structure (NMFS, 2012a) and Extinction Risk Analysis (NMFS, 2012b) reports prepared by the NMFS, National Oceanic and Atmospheric Administration, USA (and associated documents including copies of key workshop presentations, summaries of expert opinions, outline of workshop discussions) on river herring (alewife and blueback herring) in accordance with the following terms of reference:

1. Is the information regarding the life history and population dynamics of the species the best scientific information available? If not, please indicate what information is missing and if possible, provide sources.
2. Does the information on river herring genetics, physiological, behavioural, and/or morphological variation presented for the species' range represent the best scientific information available? If not, please indicate what information is missing and if possible, provide sources.
3. Based on the scientific information presented, are the conclusions regarding species, subspecies, or distinct population segment delineations supported by the information presented? If not, please indicate what scientific information is missing and if possible, provide sources.
4. Based on the scientific information presented in the extinction risk analysis report, does this analysis consider all of the best available data and are the conclusions appropriate and scientifically sound? If not, please indicate what information is missing and if possible, provide sources.
5. In general, are the scientific conclusions in the reports sound and interpreted appropriately from the information? If not, please indicate why not and if possible, provide sources of information on which to rely.
6. Where available, are opposing scientific studies or theories acknowledged and discussed? If not, please indicate why not and if possible, provide sources of information on which to rely.
7. In general, is the best scientific and commercial data available for the stock structure and extinction risk analysis of river herring presented in the reports? If not, please indicate or provide sources of information on which to rely.

In addition, the review process utilised additional literature as identified in References Cited, including background materials relating to the concept and application of the ESA to listing of species and the designation of DPS (Haig *et al.,* 2006; Palsbøll *et al*., 2006; Waples *et al*., 2007; Kelly, 2010). The original *NRDC Petition to List Alewife and Blueback Herring as Threatened Species and to Designate Critical Habitat* (NRDC, 2011) was also consulted.

While there is a plethora of data presented within the Stock Structure Report, the Extinction Risk Analysis Report is less complete than was anticipated, and will require some marginal reinterpretation of the wording of the respective ToRs. The original aim was that some estimates of extinction probability would be available for the peer review, but the data became available too late in the cycle. Thus, the Reviewer's role for the Extinction Risk component is focused on the validity of information that will be used in the modelling and the rationale for selecting such information, together with the appropriateness of the proposed methodology that will be employed in conducting the extinction risk analysis. Specifically, I will consider within the context of the ToRs whether the decisions and recommendations proposed for the modelling approach and the data to be used in the models was scientifically sound and appropriate for assessing extinction risk for river herring.

My specific expertise is in the application of genetics to stock structure analysis of aquatic species in the context of conservation and sustainability measures, including anadromous fishes, though I am familiar with approaches and use of evidence obtained from other sources including life histories, morphology, physiology, behaviour (migrations) and population demography. Additionally, I have knowledge of the methods employed and interpretation of extinction risk analysis within the context of conservation biology.

3. Summary of Findings for each ToR: weaknesses and strengths

3.1 Approach and focus of peer review

Each of the terms of reference will be considered in turn by including an initial brief statement of the current status based on submitted workshop reports and associated documents, followed by a critical evaluation of the evidence provided. Emphasis will be placed on the salient strengths and weaknesses of the data in terms of its completeness, statistical validity and ensuing interpretation. Opposing views will be commented upon, and while it was not the purpose of the stock structure and extinction risk workshops to generate a consensus of views, my aim here is to assess which of the alternative conclusions or recommendations are best supported by the documentary evidence. Notwithstanding, there are some parts of the evidence presented, identified further below, that is either available in a way that is difficult to assess because analyses of data only are presented rather than summary statistics of actual data (e.g. both of the genetic studies in the Stock Structure Report), and the work remains unpublished, or because aspects of the sampling or analysis were unclear (e.g. aspects of the information on morphology, physiology and marine migration). While river herring have been the target of fisheries for centuries, and the central role of these fishes recognised as key contributors to freshwater and marine ecosystems as forage and prey, I would regard both species as being "data-poor" in terms of the intensity and availability of data across respective geographic ranges. As such, and taken together with the inherent subjectivity of certain features of the ESA descriptors such as the meaning of "significant portion of its range" and the DPS concept, every effort will be made to generate balanced, objective and consistent opinions on the available evidence.

3.2 Key pertinent features of river herring stock structure

Before considering the various ToRs in relation to stock structure, there are several salient issues that are relevant to the review process.

3.2.1 Original NRDC petition listing

It was pointed out in the original NRDC petition that if alewife and blueback herring are not listed each as a threatened species as a whole, that the agency should designate specific DPS as follows: four DPS of alewife and three DPS of blueback herring - Central New England DPS of alewives, Long Island Sound DPS of alewives, Chesapeake Bay DPS of alewives, and Carolina DPS of alewives; Central New England DPS of blueback herring, Long Island Sound DPS of blueback herring, and Chesapeake Bay DPS of blueback herring. Such DPSs were proposed to encompass fish that originate from a river within the DPS and include the marine range of such fish. The original petition hypothesis will be considered alongside alternatives that were considered in the Stock Structure Working Group Report.

3.2.2 Severity of alewife and blueback herring population decline

In addition to the generally observed significant decline in the abundance and geographic range of blueback and alewife herring, the original NRDC petition documented some cases of particular concern (NRDC,2011), for example:

- The river herring fisheries of Chesapeake Bay and tributaries – which is regarded historically as the largest in the USA – is close to elimination, with landings in Virginia, Maryland, and from the Potomac River showing a decline of 99 percent or more from their mean catch levels between 1950-1970.
- The alewife count in two key Massachusetts river herring runs, in the Monument and Mattapoisett Rivers, declined almost 85 and 95 percent, respectively, between just 2000 and 2010.
- In South Carolina, alewife is considered extirpated.

There is thus little doubt, based on accumulating evidence, that there has been a contraction in respective species range, as well as marked trends in decline. It is worth emphasizing however that there is evidence indicating that the magnitude of decline in blueback populations appears to be markedly greater than in alewife herring – underlining the need that stock structure should be assessed and managed at the species level, with separate species-level stocks defined for the two species. For example, there has been a marked reduction in river herring abundance in the Oyster and Taylor river systems (New Hampshire), dominated historically by blueback, whereas some river herring populations where alewife dominate, although exhibiting inter-annual variation in abundance, have fluctuated around a mean value in recent years. Indeed, there is evidence indicating that there has been a shift in species composition in New Hampshire river herring towards dominance by alewife. It follows that it is likely that each species is exposed to divergent mortality rates arising from separate exploitation, natural mortality or predation in marine habitats (outside of spawning rivers). It is possible that such heterogeneity among species derives from species-specific variance in such features as seasonal movements, annual migratory or behavioural patterns – in principle, biological traits that might also be associated with stock structure, and issues of direct relevance to the designation of DPS. The wider ecosystem consequences of such species-level shifts in abundance do not appear to have been examined to date.

3.2.3 Stocking and hybridisation

It is worthwhile emphasizing prior to consideration of stock structure that the extent of biological and genetic divergence or discreteness will to a large degree depend upon the fidelity of respective spawning groups of each species returning for breeding back to natal sites. The expectation is that the higher the fidelity, the more opportunity there is for the evolution and persistence of discrete biological characters within each of the spawning groups. However, as with many salmonids for example (Harris & Milner, 2006), and species such as Atlantic herring (Ruzzante *et al.*, 2006), it is well acknowledged that fidelity is often not 100%, and that depending upon various factors such as age and experience of migrating individuals, habitat degradation or impoundments, as well as proximate factors such as food supply, a varying proportion of individuals may interbreed with other spawning assemblages ("straying"), which depending upon the success of offspring and subsequent recruitment, would be expected to reduce levels of discreteness. Evidence of such effects will be discussed below. It is worthwhile noting that anthropogenic factors such as habitat fragmentation and damming can impact directly on such processes, as well as the more indirect effects of reductions in population size through harvesting that might affect the rate and extent of divergence among groups (Hansen *et al*., 2002).

In addition to the direct implications of straying on stock structure, there is evidence discussed below indicating that there are variable levels of interbreeding or hybridisation between alewife and blueback herring. Such breakdown of species reproductive barriers can pose a significant threat to the integrity and adaptive potential of species (Perry *et al*., 2002), and is one feature of the river herring stock structure and associated extinction risk analysis that will require greater consideration than is perhaps given in either of the two Working Group Reports. Hybridisation between species might not only impact on long-term persistence of discrete species, depending upon largely the fitness of resultant hybrids, but as will be illustrated in the consideration of genetic determination of DPS, can also complicate the estimation of DPS putative boundaries.

3.3 ToR 1: Is the information regarding the life history and population dynamics of the species the best scientific information available? If not, please indicate what information is missing and if possible, provide sources.

Data pertinent to life history and population dynamics were based on diverse sources summarised in the Stock Structure (NMFS, 2012a) and Extinction Risk (NMFS, 2012b) Reports. Such sources encompassed historical data on catches, growth and survival of river herring, natal homing, run counts of specific rivers/regions, bottom trawl surveys, and spatial and temporal distribution of spawning condition. It was possible to establish local trends on the anadromous component of river herring, though based on available sources these were necessarily focused on only a portion of each species range. In general, all available data has been synthesized and presented, providing a coherent overview of the status of life history and population fluctuations. Information on natal homing is especially pertinent in relation to stock structure, and limited microchemistry data available for Poquestanuck Brook and Quinnipiac River (Gahagan *et al.,* 2012) found classification back to natal site at around 85% for alewives and blueback, though the study included only two sites with a single datum for each species. Such findings do, however, support the notion that individuals correctly classified experienced common environmental history over their lifetimes, consistent with a high degree of natal homing. The study did, however, emphasize that similarity in water chemistry among sites could largely homogenise any chemical signals, which together with difficulties of sampling “known” age-0 fish due to early life-history movements from nursery areas, render it challenging to use geochemical methods to detect natal origins among closely spaced river herring runs. Importantly, Gahagan *et al.* (2012) detected changes in Sr:Ca and Ba:Ca ratios across sectioned otoliths that indicated greater movement across salinity boundaries during the first year of life than was previously shown for anadromous alewives and blueback herring. Rulifson *et al.,* (2012) examined growth and survival of river herring using otolith microchemistry with variable natal assignments ranging from 0-64%. Inferences on straying suggested that older fish had higher straying rates than younger fish, and that such variance might derive from survival differences in other locations.

Overall, the evidence for significant natal homing is strong, and importantly coincides with other independent information such as marked genetic differentiation among populations of each species and population-level specific diversity in run dynamics. However, uncertainty in the origin and movements of sampled fish, together with insufficient magnitude or stability of geochemical signatures from water chemistry, especially among proximate rivers, indicates that otolith microchemistry should always be used in conjunction with independent approaches.

Considerable information based on trawl surveys was presented (NMFS, 2012a), including time-series, showing the spatial distribution of seasonal variance in river herring along the East Coast of the US (N Caroline to Maine). The Northeast Fisheries Science Centre (NEFSC) bottom trawl survey indices for alewife in the 1970s and 1980s show that the population was in a low state; however, more recent trends (2008 through to present) indicate some increases. In general, blueback herring are caught in lower abundances than alewife in the NEFSC bottom trawl survey, but such variance in species composition could arise from the timing of the survey in comparison to the timing of their seasonal migrations. Sufficient consideration of various drawbacks of using trawl data to assess population status were considered (NMFS, 2012a), including the effects of diel vertical migration of river herring, that the entire geographic range was not sampled (e.g. omitting the Southern coast of South Carolina), that trawl data did not detect the decline in the 1990s shown by several run counts, and that regular surveys began post 1975 after the period of high landings. Nevertheless, The NEFSC bottom trawl survey is the only coastwide index available for river herring, and has been consistently sampled for over 35 years, and is used for many species stock assessments. Findings identified significant herring by-catch as a mortality factor during marine migrations of river herring, encompassing a range of fisheries, most likely including squid, mackerel and Atlantic herring fisheries (McBride *et al.,* 2010). Data overall showed convincing differences in the distribution of age classes in marine waters, with offshore by-catch being dominated by adult river herring, whereas 0 and 1 alewives remained in near-shore coastal waters throughout the year. Thus, in estimating trends in population abundance and likely spawning biomass, it is imperative to incorporate spatial and age-specific mortality rates arising from by-catch, which is an ongoing factor for inclusion in subsequent risk analysis. However, in the case of river herring considerable caution should be exercised in such assessments because in addition to the globally common challenge of detecting representative levels of by-catch, especially in the face of Illegal, Unregulated or Unreported (IUU) fishing, it is especially difficult to derive accurate measures because such mortality occurs in multiple habitats.

Although trawling continues to represent a key method for estimating population status, its utility in characterisation of riverine spawning habitat for Alosines varies with the method employed (Harris & Hightower, 2012), and such variance in sampling efficiency must be incorporated into life history and population estimates. For example, the efficiency of oblique plankton tows and spawning pads for collecting eggs is known to differ, leading potentially to misleading estimates of spawning duration. For example, compared with spawning pads, oblique plankton tows had a higher chance of sampling river herring eggs, thereby identifying longer spawning periods. However, it was proposed that to obtain more detailed information on microhabitat use and reproduction, other approaches such as spawning pads, direct observation of spawning and examination of female histology would be more appropriate. For meaningful comparisons therefore it is important when assessing the dynamics and reproductive patterns of designated DPS to ensure that standardised and spatially-tailored approaches are employed.

While the spatial and temporal scale presented for river herring based on the Workshop Report (NMFS, 2012a) summarises the best available scientific evidence on life history and population dynamics, there are two aspects that should be incorporated. First, while it is recognised within the Report (NMFS, 2012a) that landlocked forms of alewives represent life histories distinct from the anadromous form, and thus representing levels of phenotypic plasticity within the species, the inference by the working group that such plasticity might endow alewives with the ability of landlocked and anadromous forms to quickly shift between such alternate life histories (p. 18, NMFS, 2012b) is likely to be erroneous and misleading. Recent work (Palkovacs *et al.,* 2008) using molecular genetic markers (sequence data from mitochondrial DNA control region and microsatellites) indicated that landlocked alewives have evolved multiple times independently from the anadromous forms no more than 5000 years ago and perhaps as recently as 300 years ago. Moreover, landlocked forms are genetically isolated, whereas anadromous forms exhibit some evidence of interbreeding. Importantly such isolation between landlocked and anadromous forms, even though separated relatively recently, has lead to significant divergence in foraging traits, with the landlocked forms exhibiting narrower gapes and smaller gill rakers than anadromous counterparts, suggesting that they are adapted to feeding on smaller prey items. It appears that such adaptation represents rapid evolution of foraging traits, possibly in response to changes in available resources associated with ecological isolation of prey communities. Subsequent work (Czesny *et al*., 2012), using highly sensitive “next generation sequencing”, supports evidence for such rapid evolution by examining the genetic divergence of alewives that recently invaded the North American Great Lakes. In addition to disrupting local food web structures, rapid population growth and range expansion has resulted in dramatic changes in growth rates, size at maturation and fecundity. By sequencing the transcriptomes (signatures of expressed genes) of individuals from Lake Michigan and the Atlantic Ocean, findings demonstrated that such phenotypic divergence is attributed to massive regulatory modifications rather than changes in coding genes. Such regulatory shifts offer an alternative route for adaptive shifts to new environments, nevertheless representing the separation of highly distinctive gene pools between anadromous and landlocked alewives.

Second, and as supported within the first set of genetic data presented in the Stock Structure Report (NMFS, 2012a), alewife and blueback can hybridize. While this has been recognised for some time, as discussed above, it is important to recognise that not only will the inadvertent inclusion of hybrids in samples for biological and genetic analysis prove misleading in terms of DPS integrity, importantly also, it can result in a significant threat to species integrity.

What are the implications of such findings on landlocked alewives (data not included in either NMFS Report) and alewife-blueback hybridization, and why are they important in the context of the current Expert Review? There are several key implications: 1. Depending upon the period of isolation, landlocked forms of alewives represent distinct and novel sources of genetic, life history and morphological diversity within the species, and as such should be recognised, each with a level of integrity (multiple independent origins from anadromous form) that might be expected to utilise habitat-specific resources more efficiently than anadromous counterparts. 2. The magnitude of phenotypic and genetic differentiation between landlocked and anadromous forms demonstrates that it is unlikely that shifts in the opposite direction (from landlocked to anadromous and *vice versa*) will occur quickly, or without significant mortality. They should therefore not be viewed as interchangeable forms of the same biological form. 3. The demonstration that alewives can adapt to environmental change relatively quickly (becoming landlocked, and invading new habitats) highlights the importance of maintaining high levels of genetic diversity within the species, thereby in principle securing high adaptive potential to altered environments, even if the rate of change is relatively rapid (decades to hundreds of years). 4. The observation by Czesny *et al*., (2012) that the invasion of new habitats by alewives can disrupt local food web structures poses an additional threat to existing freshwaters, especially within the context of altered migratory routes and enhanced opportunity for ecological and reproductive isolation. 5. Altered habitats or other environmental features that result in higher levels of hybridization between alewives and blueback herring represent a significant threat to the integrity and maintenance of each species, and will likely generate an increasing proportion of maladapted forms, with reduced potential for swift adaptive change.

Some additional pertinent information not contained within the Stock Structure Report (NMFS, 2012a) considered the spawning run of blueback herring (McBride *et al*., 2010), albeit restricted geographically to St John's River, Florida. The study found that spawning occurred across several days through the release of multiple batches (batch every 3-4d), and that spawning occurred far upstream (up to 400 km). While the stock was persistent, the smaller individual sizes evident today suggest that the population is still experiencing higher mortality than a few decades ago. Such observations are pertinent since it demonstrates the ongoing threats to contemporary populations, as well as the ecological diversity in spawning habit- a biological attribute that is likely to exhibit heterogeneity among designated DPS.

Armstrong (NMFS, 2012a) presented evidence that there was significant river-specific heterogeneity in alewife length-at-age in four rivers: the Nemasket, Town Brook, Monument, and the Mystic, which are listed from north to south respectively. Nemasket had returning alewives that exhibited consistently greater length-at-age than rivers to the north, and such patterns varied with latitude for the other three rivers examined. Although it is not possible to determine whether such life history variance is genetically-based from the current data, the trends observed do indicate fine-scale structuring indicative of significant biological differentiation, possibly linked to co-varying latitudinal factors such as temperature or productivity of systems. Notwithstanding the underlying causes, the observations coincide with the subsequent observations provided by genetic data that river herring exhibit marked demographic independence, sometimes across relatively fine spatial scales, allowing the opportunity for largely independent divergence of significance overall to the respective taxon.

3.4 TOR 2: Does the information on river herring genetics, physiological, behavioral, and/or morphological variation presented for the species' range represent the best scientific information available? If not, please indicate what information is missing and if possible, provide sources.

3.4.1 Genetics

The application of genetics to the analysis of stock structure in river herring was based on two primary data sets, both of which applied DNA markers known as microsatellites (NMFS, 2012). There are no other such expansive genetic data sets known to this Reviewer that are available from the published literature.

Microsatellites represent the vanguard of contemporary genetic markers used to explore the population structure of fish (Hauser & Carvalho, 2008) and many other species. These are short repetitive nuclear DNA sequences that are usually distributed randomly throughout the genome of taxa, and in general are applied based on the assumption of evolutionary "neutrality". The latter means that alternative forms (alleles) at respective microsatellite loci do not normally influence the fitness (survival or reproductive success) of carriers, and that their distribution in time and space are normally a consequence of demographic processes such as population size and gene flow (the successful interbreeding between migrants and individuals in the recipient population), as well as mutation rates (the generation of new variants or alleles as a microsatellite locus. Such markers are therefore deemed to be effective markers that identify varying degrees of interbreeding among groups, with the predicted relationship that the higher the restriction to gene flow, the greater the level of genetic differentiation, and *vice versa*.

Two key aspects of microsatellite diversity are pertinent to the current analysis of stock structure in river herring: first, the use of several loci, as in the described studies, can reveal information on the *temporal and spatial distribution of genetic diversity* – for example whether most genetic diversity is contained within (indicating large scale random mating) or among (indicating restricted interbreeding and gene flow among units) populations - and on the *amount or levels of genetic diversity* in the samples analysed. Both aspects are relevant here – the former can act as a proxy to determine the extent and distribution of breeding assemblages or units, and the latter provides an estimate of genetic diversity, a proxy often used by conservation biologists to indicate whether levels of diversity are typical or aberrant compared to expectations. While microsatellites are not typically employed to assess directly the level of adaptively significant diversity within or among populations, they can reveal properties of wild populations that indicate the relative magnitude of potential for local adaptation (Hansen *et al*., 2002). The general expectation is that where there is restricted gene flow among population samples (high between population genetic differentiation), there is increased opportunity for local adaptation (compared with high levels of gene exchange). Thus, based on the available genetic evidence provided in the current Review, it is possible to draw inferences on levels of reproductive isolation among units and the associated potential for adaptive (and thereby unique) variation that might be associated with such assemblages. While deciphering the spatial scale of population sub-division, or in the current context, the occurrence of spatial putative boundaries in DPS, presumably grouping together those samples most genetically similar (and by inference, biologically similar as well), always carries some level of subjectivity, it is possible to combine several independent lines of evidence, thereby strengthening the spatial or temporal framework of DPS proposed.

To recall, the key objective here is to apply the microsatellite data to a range of river herring samples collected in time and space, and to ask whether it is likely that they represent overall a random mix of individuals, without any need to designate DPS (since the loss of any specific population(s) is not likely to be of significance to that taxon), or indeed, if evidence supports the existence of DPS (and thereby becomes significant to the taxon as a whole), what is the most likely pattern of DPS (stock structure) supported by the genetic (and other) evidence? It is possible to decompose a critical review of such genetic data into clear sub-questions: (i) based on the sampling design employed *are the data representative?*; (ii) *are the data robust?*, that is, is there sufficient information content (levels of diversity assayed by the markers) , and have all appropriate actions been taken to secure quality assurance?; (iii) are the spatial patterns of genetic diversity revealed by the microsatellite markers *statistically valid* (appropriate tests), and *which of the various stock structure hypotheses* do they most likely support? These evaluation criteria will be applied to each of the two studies described (including the additional supplementary information on alewives provided after the workshop (08/13/12; NMFS, 2012a) by Palkovacs *et al*. on the alewife dataset to examine the uniqueness of the (tentatively) designated Connecticut River alewife stock.

The first study presented by Gephard (NMFS, 2012a) used 15 microsatellites from Maine to Florida on alewife (778 individuals – not "samples" as referred to in the Report) from 15 spawning runs in different rivers and blueback (1201 individuals) from 20 different rivers . So-called "Bayesian" analysis ("A decision-making analysis that …"*permits the calculation of the probability that one outcome is superior based on the observed data and prior beliefs…subjectivity of beliefs is not a liability, but rather explicitly allows different options to be formally expressed and evaluated"*), indicated five putative alewife stocks (using two independent Bayesian tests-STRUCTURE and BAPS): 1) Northern New England; 2) Southern New England; 3) Connecticut River; 4) Mid-Atlantic, and; 5) North Carolina, and for blueback, no single optimum solution was reached using both Bayesian tests: for blueback, STRUCTURE was unable to identify an optimal classification of groups, whereas one of the tests (BAPS), suggested four genetically identifiable stock complexes: 1) Northern New England; 2) Southern New England; 3) Mid Atlantic; 4) and Southern.

In terms of the evaluation criteria described above: (i) based on the sampling design employed *are the data representative?* There was a reasonable geographic coverage along the USA coast, though as recognised, collection of specimens on their upstream spawning run may pool samples from what are truly distinct spawning populations within the major river drainages sampled; thereby underestimating the extent of stock diversity within rivers. No samples were collected from marine forms of either species, thereby leaving uncertainty in the linkages between mixed marine aggregations and the relative contributions of specific spawning groups. The use of 15 microsatellite loci, the vast majority of which provided independent sources of information (that is, were not linked on the same chromosome), is well within the acceptable number typically used in population surveys of genetic diversity, and is therefore deemed to sample genetic diversity across the genome in a representative manner. Sample sizes were of an appropriate order of magnitude, with numbers varying between 24-138/sample (alewife, 37-69; blueback, 24-138), though most samples contained more than 40 individuals/site (alewife, 13/15 = 87%; blueback, 13/15 = 87%). While the sample sizes are similar to many microsatellite studies on fishes, I would suggest that they are at the lower boundary, and because individual genetic diversity summary statistics (e.g. heterozygosity, allelic richness and diversity) were not presented, it was not possible to state unequivocally that the sample sizes were entirely representative. For example, it is known that depending upon mutation rates and population size, the number of alleles likely to be detected in any sample of individuals is a function of the sample size, with generally more alleles being detected in larger sample sizes (Ruzzante, 1998), though there is a threshold above which the relationship reaches an asymptote, and only relatively rare (and contributing little information) alleles will be found at higher sampling intensity. Many anadromous and other fishes have numbers of alleles that sometimes reach >100 across samples, and in such situations, dependent upon relative allele frequencies in those samples, to obtain a representative estimate of genetic diversity and associated trends in stock structure, it is necessary to have large sample sizes, typically between 60-80 individuals/site. Thus, it is possible here that the level of genetic diversity was underestimated, which would typically have the consequence of underestimating the number of discrete assemblages or DPS. It is therefore fair to say that the current stock hypotheses proposed represent *the minimum number of identifiable assemblages, or DPS*, for listing.

In relation to criterion (ii) *are the data robust?* That is, is there sufficient information content (levels of diversity assayed by the markers), and have all appropriate actions been taken to secure quality assurance? Among the appropriate tests employed in assessing the quality of data are so-called Hardy-Weinberg (H-W) tests that examine the distribution of diversity at each microsatellite locus according to expectations, and tests of linkage disequilibrium, that show whether each locus provides an independent source of information, rather than being linked and thereby co-varying with each other. The latter effect would reduce the level of sampling across the genome of target samples. There were some significant deviations from H-W (4 loci), and it appears that no further action was taken: typically, unless some viable cause can be identified (using such software as MICRO-CHECKER, van Osterhout *et al.,* 2004), loci are sometimes discarded and not used in subsequent analysis. Here, there appears to be no such consideration, and in the absence of additional detail, data from these four loci should be interpreted with some caution. On the positive side, it appears that there was some agreement across loci in the spatial scales of differentiation that was detected. Only one case of potential linkage disequilibrium was detected, and is unlikely to have any major impact of the findings. While there is some uncertainty in the quality control of four microsatellite loci in alewife samples, overall the data generated would appear robust.

In relation to criterion (iii) are the spatial patterns of genetic diversity revealed by the microsatellite markers *statistically valid* (appropriate tests), and *which of the various stock structure hypotheses* do they most likely support? In addition to widely accepted measures of genetic differentiation among samples (F_{st} tests that quantify the level of between vs. within genetic diversity of pairwise/multiple comparisons), insightful Bayesian tests (STRUCTURE and BAPS) were applied to identify the most likely number of genetic clusters or units given the distribution of genetic diversity observed. It is not unusual for different statistical tests of genetic structuring to generate conflicting outputs, and the fact that it was not possible to arrive at coincident patterns in blueback herring from both BAPS and STRUCTURE is of little concern. In such circumstances, additional independent evidence or patterns are examined: in this case, the use of a comparable data set from alewife, together with the likelihood of BAPS classification based on other biological or geographic information, indicated that the stock complexes for blueback were well supported. Both the five and four stock complexes proposed for alewife and blueback are likely, though subsequent information provided by Palkovacs *et al*. on the alewife stock complexes, proposed a reduction in the number from five to four (excluding Canadian populations- so five in total for the purposes of the Review) by grouping the Hudson and Connecticut Rivers with the Southern Atlantic stock. The shift in designation does illustrate the tentative nature of such data based only on spatial surveys of genetic diversity, as well as the impact of additional complexity that might only become apparent through closer scrutiny of the data. Here, for example hybrids (alewife – blueback) and misidentified samples (arising from morphological similarity) were found and subsequently removed. Palkovacs *et al.* found that upon removal of these individuals from the Connecticut River alewife dataset, the Connecticut and Hudson Rivers belong to the Southern New England stock. Such findings do cause concern since the inclusion of hybrid individuals and misidentified individuals can clearly have a marked impact on the subsequent designation of DPS for listing. However, and despite various shortcomings detailed below, the evidence for the existence of distinct river herring assemblages is strong and convincing, and in conjunction with complementary information on other biological levels of differentiation, there are solid grounds for asserting that the various groupings represent significant components to each taxon (blueback and alewife), such that the loss of any specific assemblages would represent a gap in the species range as well as a loss of ecologically and genetically distinct units. Additional caveats to the study, discussed within the workshop (NMFS, 2012a) include a more detailed analysis of population structure within the major stocks identified to explore the extent of significant sub-structuring within each. Additionally, while it is acceptable to use neutral markers in the current context, they do not directly reveal directly the spatial or temporal levels of adaptively significant diversity. Indeed, recent studies show an increasing pattern of "hidden" adaptively divergent groups of fish, not previously detected by neutral markers (Hauser & Carvalho, 2008). New approaches are now becoming available to assess adaptive variation in the wild (Allendorf *et al., 2010*). The implication is that the current proposed boundaries should be regarded as the minimum likely number of units that might qualify for DPS designation, and that additional and likely more fine-scale stock heterogeneity that is relevant to the long-term dynamics and persistence of threatened river herring species likely exists. The levels of hybridisation between alewife and blueback are not well documented, and may also influence the results of the species-specific analyses.

Finally, in relation to the first genetic study on USA river herring (but also applicable to the second genetic study described below), there are several additional analyses of existing or additional genetic data that would yield higher confidence in evaluating the designation of stock structure and associated DPS. (1) It is well recognised that greater confidence can be ascribed to spatial patterns of genetic diversity by including a temporal component to the analyses (Waples, 1998): there is unavoidable error in estimating any population parameters in the wild simply because of sampling effects: *statistical* significance does not always equate to *biological* significance. The resampling of the same populations over time, and then comparing the distribution of genetic diversity consecutively, allows for tests of coincidence in the existence of putative stock or population boundaries: consistency in patterns increases the probability that any such patterns are indicative of *biological* and not simply *statistical* processes, and thus increasing the confidence in ascribing any such system of units for conservation and management. In addition to such temporal genetic analyses, other data sources such as morphological, behavioural or life history divergence among units if they coincide with genetic data, can also increase confidence that any such spatial framework is biologically realistic. In addition to comparing samples collected across a few years, the availability of archived historical fish scales or otoliths from which DNA may be retrieved (Iwamoto *et al*., 2012), can provide an additional temporal dimension of comparison over longer time scales, and can reveal the impact of anthropogenic disturbance. (2) Well established methods are available for the detection of interspecies hybrids in microsatellite and other genetic data (e.g. Randi, 2007), and it is recommended that such tests are applied to collated genetic information and all future such studies. (3) Relatively straight forward tests on genetic data are also available to examine evidence for demographic effects (Cristescu *et al*., 2010; Tallmon *et al*., 2010), indicating whether populations that have declined show signatures of population bottlenecks as determined by patterns of genetic diversity across time. Indications that genetic diversity has been lost from populations not only increase the potential uniqueness of semi-isolated gene pools, but can also influence the ability to adapt to new challenges. Such information can therefore be insightful in the extent to which units are grouped or separated into viable assemblages. (4) As emphasized above, increased recognition of the discreteness and potential significance of landlocked alewives should be incorporated into stock structure and DPS considerations. (5) While there remains variable impacts of stocking on genetic structure in river herring, preliminary studies indicate marked geographic variance, with some rivers or drainages being heavily impacted (as illustrated by the second genetic study described below). Particular caution should be exercised both in the sampling of fish for stock structure analysis to minimise the chance of inadvertently including unrepresentative and aberrant individuals likely to generate unrepresentative spatial patterns of diversity, as well as the use of such practices in management, with careful choice and documentation of stocked individuals.

The second genetic study, presented by Willis (NMFS, 2012a) examined the diversity of alewives in Maine and maritime Canada, as well as exploring the regional effects of stocking on river herring in Maine. In the former, alewife and blueback herring (881 individuals) were examined from 15 sites throughout mid-coast Maine at 10 microsatellite loci. The latter part of the study examined 2000 alewife individuals from Maine and Atlantic Canada using 14 microsatellite loci. It was not possible to ascertain the range of sample sizes employed from either the information contained within the Stock Structure Working Group Report (NMFS, 2012a), or the presentation slides. However, the subsequent evaluation is based on the assumptions of relatively large (~ 40 individuals/sample) and equal sample sizes across samples, and within the magnitude of the previous study presented by Gephard (NMFS, 2012a). The genetic analysis of alewives in Maine and Atlantic Canada revealed a so-called Isolation by Distance (IBD) pattern, that is, a significant correlation between geographic separation and genetic differentiation, such that more proximate populations exhibited higher levels of genetic similarity, indicative of higher levels of interbreeding. Based on the documented life history and migratory habits of alewives, such patterns lend support to the notion that natal homing underpins the generation of genetically distinguishable populations. Various tests, including a multivariate test of population grouping based on F_{st} estimates identified four putative groupings: (1) Cape Breton, Nova Scotia, Gulf of St Lawrence; (2) East shore of Nova Scotia; (3) Bay of Fundy and (4) Maine. The same Bayesian approach employed in the first genetic study (STRUCTURE) suggested hybridization between alewife and blueback herring, as well as marked impacts of stocking on stock structure in Maine. It appears that alewife populations in Maine have indeed been subjected to extensive within and out of basin stocking for enhancement, recolonisation of locally extinct populations, and stock introduction. Although there has been considerable stocking and translocation within Maine, documentary evidence is scant. Stocking was proposed to be much less common in Canada, as supported by the geographic patterns of microsatellite variation.

In relation to the three evaluation criteria discussed more fully in the first genetic study: (i) based on the sampling design employed *are the data representative?* Only 10 microsatellite loci were employed in the population survey, while acceptable in deciphering regional diversity, is perhaps on the lower limit of number of loci to be employed. The use of 14 loci in the exploration of stocking impacts is certainly sufficient to detect such effects, especially since markedly divergent and regionally aberrant populations, typical of translocated or out of basin stocked fish, will be more readily detected. It is possible that the use of additional loci in the population survey, while not altering the substantive finding of marked and significant genetic differentiation among alewife populations, that additional more fine-scale divergence might be disclosed. No additional comments can be made on the adequacy of sample sizes. With the second criterion (ii) *are the data robust?* No details have been provided on tests of equilibrium (H-W, or linkage disequilibrium), though it has to be anticipated based on the extensive experience of Experts who conducted the study that such issues will have been addressed. Finally, in relation to criterion (iii) are the spatial patterns of genetic diversity revealed by the microsatellite markers *statistically valid* (appropriate tests), and *which of the various stock structure hypotheses* do they most likely support? In keeping with findings from the first genetic study, there is strong and convincing evidence from entirely independent population samples and analyses that river herring (specifically alewife in the second study) exist as a mosaic of genetically discrete assemblages likely to represent significant components of the species range. The independent disclosure of hybridization between alewife and blueback herring highlight again the urgency in assessing the extent and dynamics of such potential threats to each of the species as a whole, and in particular how recent and ongoing environmental and habitat change might increase the incidence of such interbreeding between two distinct *Species Of Concern* (NMFS, 2006), each with their own characteristic biological and distributional features that underpin persistence and adaptive potential in the face of environmental change.

Collectively, genetic data indicate a strong case for the designation of DPS independently within each of the alewife and blueback herring species, and the putative groupings proposed likely represent the minimum levels of separation into DPS, with likely biologically significant, more fine-scale sub-structuring within each. Additional to the evidence presented here, and as discussed above, there is strong evidence supporting the case for landlocked alewives to receive separate designation.

In relation to historical shifts in the extent and patterns of stock structure in river herring, it is likely that river or wider-scale (e.g. drainage) – specific units existed in the past, it is expected that loss of habitat, together with stocking in particular regions, has resulted in some homogenization of genetic structuring, contributing to the generally observed ~100km scale of structuring determined by genetic data. The combined inclusion of demographic tests to explore population bottlenecks, as well as utilisation of archived DNA from past populations might assist in reconstructing the spatial and temporal scale of such past events, providing an insightful dimension for interpreting contemporary patterns.

3.4.2 Physiological, behavioral, and/or morphological variation

Various independent lines of evidence were explored in order to assess the extent of stock structure in river herring. Geometric morphometrics and analysis of otolith shape was examined in Maine populations of alewife and one population in Massachusetts. Convincing evidence of stock discreteness was revealed by relatively high correct classification of individuals back to source populations – for example, otolith shape classified all populations correctly from between 70-90% of the time, and morphometrics classified eastern from western Maine, and eastern Maine from Massachusetts only 58% of the time. There is evidently significant variance in both approaches, most likely being attributed to environmental variation and the fact that fish are known to be among the most highly phenotypically plastic of all vertebrates. Thus, while morphological traits can be employed in conjunction with other markers, especially if a framework of stock distribution if already available (e.g. from genetic studies), these additional tools can be targeted at testing specific hypotheses. It appears that while otolith shape offers potential, morphometrics is less discriminatory.

Data presented by Sullivan summarised findings relating to river herring returns to six rivers along New Hampshire's16 km coastline. Although there was some consistency in spawning run estimates among rivers, evidence was also presented for independent return response between the alewife dominated rivers (Cocheco, Lamprey, and Exeter), notably the Cocheco and Lamprey Rivers, supporting the notion of a level of demographic independence across relatively small spatial scales.

One key feature of river herring is their anadromous habit, which means that for a considerable period of their life history they occupy and utilise marine habitats and resources. There is, however, relatively scant information available on the dynamics and abundance of marine populations, some of which has been reviewed above in 3.3. Data provided by Rulifson (not present at the workshop) summarised results from a tagging study completed from 1985-1986 in the Bay of Fundy. Although there was a low tag return rate (overall 0.39%), it appears that some river herring migrate considerable distances in their marine phase: for example, alewives and blueback herring tagged in the Bay of Fundy were likely to be of different origins, with some alewives from as far away as New England, while blueback herring recaptures were likely not regional fish, but those of U.S. origin from the mid-Atlantic region. There is evidently an urgent need to more closely monitor the dynamics and seasonal movements of mixed marine aggregations ("mixed" in *both* the sense of alewife and blueback species and the diversity of species-specific populations originating from distinct rivers/drainages). The application of mixed stock analysis (Bekkevold *et al*., 2007) using genetic markers would, for example, likely generate an effective framework for exploring behavioural and migratory diversity among stock complexes. Its utility will, however, depend upon the completeness and level of population-level discreteness in the reference data base. When such tools are in place, it is then possible in principle to assess not only the relative contributions of particular stocks to marine feeding aggregations, but importantly also, it will provide the opportunity to adjust harvesting in accordance with the strength (estimated abundance) of particular source populations. Moreover, a spatially defined framework that incorporates seasonal variation in distribution and abundance will also serve to identify those regions (and specific stock contributions) that are especially vulnerable to the effects of by-catch and other mortality factors: information that can feed directly into an ERA.

Collectively, the data presented on behaviour, physiology and migration support the notion of varying degrees of demographic independence among putative regional populations/stocks, but that considerable uncertainty remains in the distribution and dynamics of such units in the marine environment. The implications of such uncertainty amplify the challenge of coupling the marine and freshwater phases of the lifecycle of river herring: both being key contributors to spawning success and species/DPS persistence. While the current petition appears to focus on the freshwater phases, representing the necessary foundation for listing of any designated DPS, there is an urgent need to more closely scrutinise the dynamics of river herring populations in the sea in view of their core contribution to subsequent spawning and recruitment.

3.5 ToR 3: Based on the scientific information presented, are the conclusions regarding species, subspecies, or distinct population segment delineations supported by the information presented? If not, please indicate what scientific information is missing and if possible, provide sources.

Overall, the evidence provided supports the key elements of the framework for listing of alewife and blueback as comprising a range of independent population/regional units appropriate for designation under DPS policy. Evidence is clear that each of the alewife and blueback species, despite varying and perhaps largely undefined levels of hybridization should be recognised separately within the context of the ESA listing. Each species differs in aspects of its biology (e.g. timing and duration of spawning, migration) and distribution, and evidence within the Working Group Report (NMFS, 2012,a) supports the notion that each species is experiencing distinct levels of exploitation, natural mortality and by-catch in line with species-level diversity in seasonal movements, annual migratory and behavioural patterns. Thus, evidence supports strongly the need to designate stock structure at the species level, with separate stocks defined for alewife and blueback herring.

There appears, however, from the literature and evidence provided by the Working Group Reports (NMFS, 2012 a,b) to be ongoing confusion in distinguishing these species in the field and within samples. For example, the genetic studies presented in NMFS (2012a) initially included incorrectly assigned species, resulting not surprisingly in the need to revise proposed designation of stock structure. It is perhaps worth noting that relatively little effort is needed, either in situations where taxonomic ambiguity between alewife and blueback ensues, or even on a regular basis in assessing genetic diversity and/or stock structure in either species, to DNA sequence samples at the DNA barcoding gene (*COI, cytochrome oxidase I;* Costa and Carvalho, 2007). Surveying the global DNA barcoding data base, *The Barcode of Life Data*

System (BOLD: http://www.barcodinglife.org/), reference sequences for each species have already been deposited from several regions across the species' range, allowing relatively quick and simple verification of species status since each species exhibits a species-specific profile with relatively little within-species variance. Moreover, as data indicate, each species exhibits independent responses to environmental variation, emphasizing the need to properly designate individuals to the correct taxon. Taking each species as a whole, it is clear to see from evidence provided, as well as from diverse historical records, that river herring have in general suffered significant population decline, which is largely ongoing, with continued threats from habitat disturbance, pollution, exploitation and by-catch. The geographic range of both species has contracted: for example, alewife appears to exhibit contraction at the southern edge of its range of distribution, with populations locally extinct from South Carolina, and now possibly from southern North Carolina. Notably, the potential impact of climate change poses a particular additional effect that might act synergistically with other mortality factors. Elevated water temperatures will increase the incidence of hypoxic zones in spawning and nursery areas such as Chesapeake Bay and the Delaware River. Shifts in weather patterns will also likely increase water flow rates and associated contaminant loadings, with ensuing threats to the suitability of particular habitats. Any related shifts in the community or ecosystem structure of oceanic, estuarine and riverine environments may further affect migratory cues, thereby directly threatening the persistence of locally adapted and discrete assemblages through interference with natal homing or increased straying. Since the anadromous habit of river herring necessitates segregation into river-specific populations, there is limited scope for range shifts, especially in the short-term. Finally, it is evident from the burgeoning nature of river herring decline and reductions in distribution range that there is currently no appropriate regulatory framework in place.

In terms of stock structure and the designation of DPS- it is the genetic data that are most well placed to formulate hypotheses and test each against available data. All expert opinions expressed in the NMFS (2012a) Report suggested evidence of regional stock structure (~ 100 km scale) for both alewife and blueback populations. However, views on the exact boundaries differed among Experts, though a general statement of hypotheses was provided, as detailed below, which was to be taken forward for an ERA. As indicated in detail throughout the Review, marked spatial and/or temporal genetic differentiation among populations/regions is a powerful tool for identifying biologically significant assemblages within an obvious geographic framework. While a range of alternative hypotheses with varying levels of grouping samples were discussed within the Stock Structure Report (NMFS, 2012,a), the strongest supported are summarised below:

Alewives- Updated on 08/13/12

• **Hypothesis 1:**

- One continuous stock complex throughout the entire range from US to Canada

• **Hypothesis 2**: Five stock complexes

- Carolina (all alewife rivers south of, and including the Chowan River)
- Mid-Atlantic (all Virginia waters up to, and including New Jersey waters)
- Southern New England (all New York waters up to, and including Massachusetts waters)

- Northern New England (Lamprey up to and including the St. Croix River)
- Canada (all Canadian Rivers)

Blueback herring

• **Hypothesis 1:**

- One continuous stock complex throughout the entire range from US to Canada

• **Hypothesis 2**: Five stock complexes

- Southern (St. John River to Cape Fear River)
- Mid-Atlantic (Neuse River to Connecticut River)
- Southern New England (Gilbert-Stewart to Mystic River)
- Northern New England (Exeter River up to and including St. Croix River)
- Canada (all Canadian Rivers)

Data presented on genetics, life history, physiological, behavioural and morphological diversity provides no support for Hypothesis 1 in either species. There is compelling evidence that each species independently exists as a complex of discrete population units, each with specific genetic characteristics and most likely associated biological properties. While there was overall no apparent coincidence in the range of populations examined for each of the data sources, collectively they support strongly varying levels of demographic independence, reproductive diversity, distributional variance and population-specific variation in migratory and behavioural diversity among putative stocks. Hypothesis 2 for each of the species is most strongly supported, proposing the existence of at least five stock complexes (including Canadian populations) based primarily on genetic data. It is important to note, however, that there are significant geographic gaps in the sampling survey, and that the grouping of Canadian population into a single separate stock is based on political and geographic expediency, rather than biological reality. It is highly likely that the Canadian river herring populations support the categorisation of additional distinct units. Indeed, evidence presented did suggest that three stock complexes exist within Canadian waters for alewife: Inner Bay of Fundy (U.S./Canada border up to but not including the Tusket River); East Coast of Nova Scotia (Tusket River up to Cape Breton Island), and the Gulf of St. Lawrence (Cape Breton Island to the Miramichi River, possibly further into the Gulf of St. Lawrence) (Bentzen and Willis, unpublished data).

Overall, and as indicated in the collective consideration of genetic and other evidence, such a complex of stocks will likely represent the *minimum* number of units for consideration under DPS policy. While I would not favour designation of each and every river as a separate DPS or stock, it is likely that marked genetic and biological heterogeneity of significance to the respective taxon as a whole exists within each of the stock complexes proposed. Such designation would be a conservative approach to recognising and conserving population diversity, though it is important for the purposes of any translocations or enhancement programmes (which are not necessarily to be encouraged), or in situations where proximate or local populations increasingly converge due to habitat disturbance, to recognise the high probability that additional "hidden", though biologically significant diversity exists within defined categories.

Additionally, as argued above (section 3.3), a complete consideration of alewife DPS should recognise the unique evolutionary and biological discreteness of landlocked forms. Based on evidence discussed in 3.3, indicating independent origins or landlocked forms from anadromous counterparts, and strong local adaptation, it is not appropriate to consider these two forms as interchangeable variants. Additionally, intentions to cross such forms for the purposes of enhancement or any other reason are strongly discouraged.

Experts in the Working Group Report (NMFS, 2012a) identified several significant gaps in their consideration of stock structure in alewife and blueback herring, namely:

- Inconsistencies and uncertainties in the proper identification of alewives and blueback herring in river herring datasets;
- Genetic structure of mixed stocks at sea;
- Information on movements and migrations at sea;
- Longer and finer scale genetic data for returning spawners;
- Otolith microchemistry range wide and at a finer scale;
- Straying rate data;
- Information on hybridization and conditions that contribute to hybridization (e.g. climate change, dams);
- Information on whether the abundance of Atlantic herring differentially affect bluebacks and/or alewives;
- Understanding if fishways inadvertently select for certain phenotypes or certain species;
- Understanding the hatchery effects of stocking on genetic diversity.

Reference is made to many of the above caveats within the current Expert Review, and although such matters represent appropriate priorities for additional work, collectively they do not undermine the core conclusions presented here, but rather represent priorities for ongoing determination of status. Evidence supports the two following conclusions:

(1) The clear separation of alewife and blueback herring as discrete and independent species, each with their own suite of biological properties that impart differential responses to environmental threats, and thereby requiring separate consideration under the ESA, and
(2) The existence of discrete stock complexes within each of the two river herring species, suitable for designation under DPS policy.

3. 6 ToR 4: Based on the scientific information presented in the extinction risk analysis report, does this analysis consider all of the best available data and are the conclusions appropriate and scientifically sound? If not, please indicate what information is missing and if possible, provide sources.

As explained at the outset, it was not possible to review the outcome of the planned ERA within the current review. Thus, my role for this Extinction Risk component of the Review is focused on the validity of information that will be used in the modelling and the rationale for selecting this information, together with the appropriateness of methodology to be employed in conducting the extinction risk analysis. Specifically, I will consider within the context of the ToRs whether the decisions and recommendations proposed for the modelling approach and the data to be used in the models were scientifically sound and appropriate for assessing extinction risk for river herring.

It is important to emphasize initially that while extinction ERA have been conducted for many taxa in recent years, and such outputs have been utilised by conservation biologists and management bodies, there continues to exist significant disparity in the most appropriate methods to employ. The exercise of an ERA typically can be broken down into the selection and gathering of empirical data for inclusion, and the choice of various methods, often including varying forms of a Population Viability Analysis (PVA; e.g. Gong *et al*., 2012). The resulting analyses usually allows for inclusion of uncertainty in terms of data values, as well as the ability to model a range of scenarios over varying periods of time. Crucial input decisions concerning biological parameters of target species include a consideration of the length of time series available, the spatial resolution of data, and the extent of sampling effort. Careful consideration must also be given to the underpinning model assumptions and an ability to assess the validity of outputs through a so-called "sensitivity" analysis (e.g. Bretagnolle *et al*., 2004). Each of such factors must be taken into account prior to drawing conclusions from any ERA. Indeed, it has been suggested (Wesley & Damon-Randall, 2008) in the context of the ESA, that where data are limiting accurate quantitative forecasts of extinction risk, that a standardised and qualitative five-factored decision analysis is employed to evaluate extinction risk, based on the criteria specified in Section 4(a)(1) of the ESA:

(a) The present or threatened destruction, modification, or curtailment of habitat or range;
(b) Overutilization for commercial, recreational, scientific, or educational purposes;
(c) Disease or predation;
(d) The inadequacy of existing regulatory mechanisms; or
(e) Other natural or manmade factors affecting its continued existence.

Considerations can be made at the level of specific population subunits, with a subjective score. While such an approach may be inadequate to use in the absence of quantitative estimates, it does have the advantage of incorporating the key elements that are relevant to risk assessment within the ESA designations, as well as providing an additional, and largely independent bench mark with which to judge formal quantitative assessment, especially in data-poor species.

In the current context, it is perhaps realistic to state that river herring are essentially "data-poor" species, with many unknowns in terms of population parameters, especially the extension of specific measures across the species range. Experts have been asked to undertake an ERA for each of the two species according to the two specific hypotheses described in 3.4 above. Since it is possible that one or more stock complexes may be combined into a single DPS or multiple DPS in the ESA listing determination, it was proposed that any trajectories generated for hypothesis 2 of either species should allow for the possibility of combining results from stock complexes subsequently.

Participants at the ERA workshop reviewed information on the river herring petition and ESA consideration process, an overview of the stock structure discussions, data on the ASMFC stock assessment, available data for potential inclusion and the range of most appropriate models to utilise, as well as various ERA methodologies for other species that have been employed in the past. A modification of a PVA was initially considered, namely a diffusion approximation method of a PVA for river herring. PVAs are used typically to predict the chance of a population persisting into the future or the probability of it dropping below some pre-defined threshold (quasi-extinction). There exists many variants of such methods, ranging from highly complex (i.e., Atlantic salmon in Legault, 2005) to more simple extrapolating trends (i.e., diffusion approximation as with Dennis *et al*., 1991). Consideration was given in the ERA Report (NMFS, 2012b) on ways to potentially improve estimates of population growth (μ) by incorporating life history and year class effects. A time-series of data are available for inclusion from 10 years. However, as if often the case in data-poor species, careful consideration has been exercised in the subsequent selection of input data taking into account the fact that information on river herring is geographically patchy (e.g. majority of run count data were available only for the New England region), the appropriateness of bottom trawl survey data to ascertain relative abundance, and the recognition that the choice of choice of years for diffusion approximation PVA can strongly affect model conclusions.

Additional consideration was given to the use of the Multivariate Auto-Regressive State Space (MARSS) package in R. The approach provides a method for fitting linear MARSS models to multivariate time-series data, and is considered to better compensate for both process and observation error. In terms of data input it was proposed to utilise NEFSC spring and fall bottom trawl survey indices in coastwide applications, and that for the stock complex applications (hypothesis 2 of respective species), that run counts where available, and regional fishery-independent surveys could yield reliable input data. The use of bottom trawl survey data to assess abundance, while not ideal for a species such as river herring, is in the present context credible if diel vertical distribution of the species is taken into account, and also the relatively long time-series available, from 1975, and with the potential ability to extend back into the 1960s. The longer time-series might capture the decline in relative abundance in the 1960s and early 1970s. Overall, and despite limited data, confidence can be given to the estimation of probabilities that populations might fall below a critical threshold. Applying the model to various river herring populations, it became evident that with longer time-series of data, improved accuracy of forward forecasting was possible. Moreover, if significant associations could be detected between specific environmental factors and recruitment variance, it would be possible to analyze such effects for time trends that can potentially be projected forward to account for such effects as climate change estimations.

Subsequent specific consideration was given to the nature and amount of data available within the context of the proposed stock complexes, all of which is appropriate to assess extinction risk. Indeed, despite the relative scarcity of data for river herring across broad geographic regions, the range of information available could be decomposed into relevant stock and regional scales. While numerous and evidently disparate data sources exist for input to models at the stock complex level for each species, it should be recognised that issues of quality assurance of data, variance in sampling procedures and effort will likely impart additional variance to extinction probability estimates. In such situations it will be important to undertake appropriate sensitivity studies, as well as perhaps a partial subjective scoring analysis of the type utilised by Wesley & Damon-Randall (2008) for additional bench-marking.

Based on available data, a range of potential models for inclusion were discussed, including depletion-based stock reduction analysis (DBSRA), age-structured projection models, CPUE, MARSS models, and diffusion approximation. Pertinent applications of each were discussed from other studies (NMFS, 2012b), and a realistic assessment given on the caveats or advantage of alternatives. For alewife, the MARSS package was the model proposed to best accommodate the key biological attributes of the species and the available data. An advantage of the MARSS approach is that it can be run with missing data within observational time series, thereby removing the need for matching time-frames across all data sets. Critically, and perhaps most importantly here, MARSS additionally permits the inclusion of more than one hidden state, which could represent one stock complex or a series of individual stocks. A preliminary run was undertaken with the MARSS model using spring and fall trawl survey data. Population growth rate and initial biomass were assessed, and the forward projections generated of the target population to estimate the probability of extinction over 100 years, and although there were issues relating to quality of outputs, it was concluded that the MARSS approach was best suited for ERA in river herring. A standardised approach for both species based on the 4-year running sum for all run counts and young-of-year (YOY) surveys is proposed.

Based on the available scientific evidence describing input data, and range of ERA models considered, the decision to employ a MARSS approach is well supported. Of the various options, it is an approach that has inherent flexibility for encompassing respective stock complex scenarios, as well as accommodating limitations in the data, either through missing values or with restricted time-series. Input data, although presenting potential caveats, have been recognised sufficiently by the ERA Working Group, and should be reviewed when interpreting model outputs. Additionally an appropriate sensitivity analysis (various options are available; e.g. Reed et al., 2002) to assess the relative contribution of various drivers that most affect population growth or quasi-extinction probability should be undertaken.

3.7 ToR 5: In general, are the scientific conclusions in the reports sound and interpreted appropriately from the information? If not, please indicate why not and if possible, provide sources of information on which to rely.

The key conclusions arising from both Working Group Reports can be summarised thus:

(1) The clear separation of alewife and blueback herring as discrete and independent species, each with their own suite of biological properties that impart differential responses to environmental threats, and thereby requiring separate consideration under the ESA, and
(2) The existence of discrete stock complexes within each of the two river herring species, suitable for designation under DPS policy.
(3) By utilising a range of abundance survey data and life history information decomposed according to various stock complex scenarios for alewife and blueback herring, it is proposed to undertake an ERA utilising the Multivariate Auto-Regressive State Space package. Probability of populations/stocks

persisting into the future will be quantified, and where possible, the effects of various environmental factors on such probabilities will be assessed.

In general, the conclusions are sound in that they have been suitably considered in relation to available data, the approaches incorporate advanced and informative methodologies and critical consideration of most caveats in design or interpretation have been provided. Either through an occasional lack of detail (e.g. sample sizes for some genetic studies), or because of the nature of submitted evidence (e.g. slide presentations), it was not always possible to assess the quality of all data sets. Nevertheless, evidence which derives from independent sources in support of the case for separate species delineation of DPS is strong. Additional genetic analysis on existing and future data would perhaps better refine the spatial and/or temporal distribution of putative DPS by incorporating a temporal analysis of genetic diversity to better assess the stability of putative stocks and associated biological significance of such structuring, additional demographic analyses (e.g. assessment of population bottlenecks), enhancing the geographic range of sampling for all data sources, to examine within-stock complex heterogeneity in genetic and biological traits, to reassess the status of landlocked alewife, to explore the composition and seasonal movements of the marine phase of river herring, and to more accurately estimate the impacts of stocking and hybridization on population integrity. It is vitally important that all existing and forthcoming genetic data (and any associated trait data) be reassessed for the potential inclusion of misidentified specimens or the inclusion of hybrids. The update provided by Palkovacs *et al*. on alewife stock complexes demonstrates with force the effect that such misclassification can have on stock designation. The ensuing outputs from the projected ERA should, however, be interpreted with some caution until a broader geographic range of populations can be assessed, together with more detailed (sampling effort) input variables.

3.8 ToR 6: Where available, are opposing scientific studies or theories acknowledged and discussed? If not, please indicate why not and if possible, provide sources of information on which to rely.

In general, contributors to both Working Group Reports (NMFS, 2012a, and b) are to be commended for considering a range of alternative scenarios in terms of stock structure, and the validity of data and appropriateness of different methodologies for inclusion in the ERA. Not only were the strength and weaknesses of alternative stock complex scenarios considered, but a useful summary of independent Expert opinions were provided following the Stock Structure Working Group Report. While there was disparity in the placement of putative boundaries to group specific populations, there was unanimity in the existence and need to recognize discrete population units of each species suitable for consideration under the ESA DPS policy. It does of course have to be recognized that the proposed stock boundaries, while an appropriate framework based on available data, must be seen as labile, with additional biologically significant heterogeneity likely existing within stock categories. Additional emphasis on the status of landlocked alewife (Palkovacs *et al.,* 2008; Czesny *et al*., 2012) should be given. A variety of modelling approaches for ERA were reviewed (NMFS, 2012b), and an appropriate trade-off was achieved in balancing the most insightful assessment with the nature and volume of available data, for what can be regarded as an essentially "data poor" species. There does, however, remain a need to exercise caution in ERA outputs when compared across the various putative stock categories, both because of unequal sampling effort and volume of data, but also because of a lack of standardisation in approaches or quality control inherent in independently-conducted surveys.

3.9 ToR 7: In general, is the best scientific and commercial data available for the stock structure and extinction risk analysis of river herring presented in the reports? If not, please indicate or provide sources of information on which to rely.

As stated and discussed more fully above in related ToRs, in general, the best available scientific and commercial data for stock structure and ERA of river herring have been presented. Without repeating the detail of points made, some additional analyses at the level of distinct stocks for alewife and blueback herring would potentially strengthen the overall case for listing, as well as better refining the spatial resolution of stock structuring that is significant to the respective species.

4. Conclusions and Recommendations in accordance with ToRs

- River herring have experienced in recent decades a significant decline in population abundance and overall contraction in geographic range. Indications are that threats arising from various sources such as habitat alteration, pollution, directed exploitation and by-catch, as well as likely escalating effects of climate change, continue to impact on alewife and blueback herring distribution and persistence.
- There is clear separation of alewife and blueback herring as discrete and demographically independent entities, each with their own suite of biological properties that impart differential responses to environmental threats, thereby requiring separate consideration under the ESA.
- There is compelling evidence from genetic studies and population/regional-diversity in life history, morphology, migratory patterns and behaviour that support the existence of discrete stock complexes within each of the two river herring species, suitable for consideration under DPS policy: respective populations or groupings thereof, differ markedly in their genetic and other biological properties relative to other such units. While the putative boundaries proposed, essentially based on the genetic (microsatellite diversity) data, represent discrete assemblages, each comprising a significant component of the biological diversity of respective species, stock units should be viewed as a likely *minimum* number of complexes. It is very likely, based on our knowledge of other anadromous species with varying levels of natal homing, that biologically significant heterogeneity exists within each of the proposed categories. Such is the level of stock structuring that loss of individual components would represent a *significant gap* in the range of the respective species.
- After considering a range of abundance survey data and life history information depicting the current knowledge of river herring distribution, biology and structure of putative stock complexes, it was proposed to undertake an ERA utilising the Multivariate Auto-Regressive State Space (MARSS) package. Probability of populations/stocks persisting into the future will be quantified, and where possible, the effects of various environmental factors on such probabilities will be assessed. Only a brief preliminary

analysis was included within the ERA Working Group Report establishing the feasibility of the approach. The choice of input data was sound, and the MARSS approach is well suited to river herring through an ability to decompose probability estimates of extinction according to the various stock complex scenarios, and by accommodating the incompleteness of biological input data and the variable duration of time-series available.

- Overall, the best available scientific and commercial data on stock structure and ERA for river herring were contained within the two submitted Reports, utilising appropriate methodologies for the biological scales under study (between species and intraspecific biological diversity). Additional genetic analysis on existing and future data are recommended to better refine the spatial and/or temporal distribution and dynamics of putative DPS by (a) incorporating a temporal analysis of genetic diversity to better assess the stability of putative stocks and associated biological significance of discrete units; (b) additional demographic analyses (e.g. assessment of population bottlenecks) to examine the impact of past population declines, and by extension, the impact of projected future threats; (3) enhancing the geographic range of sampling for all data sources; (4) examining within-stock complex heterogeneity in genetic and biological traits; (5) undertaking such studies as mixed stock analyses of river herring in marine aggregations to explore the composition and seasonal movements of the marine phase, and (6) accurately estimating more fully the impacts of stocking and hybridization on population integrity. It is vitally important that all existing and forthcoming genetic data (and any associated trait data) be reassessed for the potential inclusion of misidentified specimens or the inclusion of hybrids.
- The apparent on-going confusion in identification of alewife and blueback herring must not be underestimated in terms of potential impact on DPS designation under ESA policy. As argued within the Report, at relatively low cost, such issues can be readily addressed by occasional use of high throughput independent verification of species status using such methods as DNA barcoding.
- Although the NMFS has determined that the NRDC pertains to anadromous populations of river herring only, the Reviewer recommends that the status of landlocked alewife be reassessed based on recent genetic evidence indicating multiple and independent origins of landlocked forms from anadromous counterparts, and the marked genetic and ecological divergence between these two forms over only 100-300 years. Landlocked and anadromous river herring should not be viewed as interchangeable forms of the same biological entity, and crossing the two forms for whatever purpose is strongly discouraged.
- In general, the Stock Structure and ERA Working Reports provided a detailed and balanced overview of the key threats pertinent to ESA listing, and the best available data for determining 1) whether there is evidence of stock structure for alewife and blueback herring; and 2) the provision to NMFS of expert opinions on the extent (if any) of stock structure for alewife and blueback herring.

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6. Appendix 1: Bibliography of materials provided for review

1. NMFS. 2012. River Herring Stock Structure Working Group Report. Report to the National Marine Fisheries Service, Northeast Regional Office. August 13, 2012, 60pp
2. NMFS, 2012. River Herring Extinction Risk Analysis Working Group Report. Report to the National Marine Fisheries Service, Northeast Regional Office. August 13, 2012. 40 pp.
3. Copies of presentations from the Stock Structure and Extinction Risk Analysis Workshops.
4. Appendix A: Expert Opinions from the River Herring Stock Structure Workshop.
5. Appendix B: Updated genetic data from Palkovacs *et al*. on alewife stock complexes
6. 2012 River Herring Workshop Groups: Goals, Agenda & Notes for Stock Structure Meeting, June 20-22, 2012, NMFS Northeast Regional Office, Gloucester, MA
7. Supplementary Materials to the Extinction Risk Analysis Working Group Report for CIE Peer Review

7. Appendix 2: A copy of the CIE Statement of Work

Attachment A: Statement of Work for Dr. Gary Carvalho

External Independent Peer Review by the Center for Independent Experts

River Herring (Alewife and Blueback Herring)
Stock Structure and Extinction Risk Analysis

Scope of Work and CIE Process: The National Marine Fisheries Service's (NMFS) Office of Science and Technology coordinates and manages a contract providing external expertise through the Center for Independent Experts (CIE) to conduct independent peer reviews of NMFS scientific projects. The Statement of Work (SoW) described herein was established by the NMFS Project Contact and Contracting Officer's Technical Representative (COTR), and reviewed by CIE for compliance with their policy for providing independent expertise that can provide impartial and independent peer review without conflicts of interest. CIE reviewers are selected by the CIE Steering Committee and CIE Coordination Team to conduct the independent peer review of NMFS science in compliance the predetermined Terms of Reference (ToRs) of the peer review. Each CIE reviewer is contracted to deliver an independent peer review report to be approved by the CIE Steering Committee and the report is to be formatted with content requirements as specified in **Annex 1**. This SoW describes the work tasks and deliverables of the CIE reviewer for conducting an independent peer review of the following NMFS project. Further information on the CIE process can be obtained from www.ciereviews.org.

Project Description: NOAA's National Marine Fisheries Service (NMFS) was petitioned to list alewife (*Alosa pseudoharengus*) and blueback herring (*Alosa aestivalis*), collectively referred to as river herring, under the Endangered Species Act (ESA) on August 5, 2011. NMFS reviewed the petition and published a positive 90-day finding determining that the information in the petition, coupled with information otherwise available to the agency, indicated that the petitioned action may be warranted. As a result of the positive finding, the agency is required to review the status of the species to determine if listing under the ESA is warranted. River herring are commercially important US-Canada transboundary species that have an expansive coast-wide range; therefore, determinations from this process have the potential to be highly controversial.

Approximately three years ago, the Atlantic States Marine Fisheries Commission (ASMFC) technical committee began working on a river herring stock assessment. The ASMFC is scheduled to complete the assessment in May 2012. NMFS is collaborating with ASMFC on this effort and intends to use the information in the stock assessment as a primary source of information in making the 12-month listing determination. Because the stock assessment does not contain all elements needed to make a listing determination under the ESA, NMFS has identified the missing required elements and intends to hold specific workshops focused on addressing these information gaps. Two of the workshops organized for this purpose will address River Herring Stock Structure and Extinction Risk Analysis, and reports from each workshop will be compiled this summer.

The extinction risk and stock structure meetings will bring together appropriate scientists to discuss the available information and perform the necessary analyses. The invited participants for these meetings will not come to a consensus; rather, they will provide their individual expert opinions related to stock structure and various methods to determine extinction risk of these two species. NMFS will take this information as compiled in the reports and determine which extinction risk method and stock structure analysis will best inform the listing determination. These reports will not contain any listing advice or reach any ESA listing conclusions – such synthesis and analysis is solely within the agency's purview. NMFS will use these reports along with the ASMFC river herring stock assessment to develop an ESA listing determination and is required to publish its finding in the *Federal Register* on or before August 5, 2012 (within 12 months of receiving the petition).

Given the significant public interest in river herring, it will be critical for NMFS to obtain a transparent and independent review of the associated meeting reports. The information and analysis in these reports will likely contain essential factual elements upon which the agency may base its ESA listing determination. Accordingly, it is critical that these reports contain the best available information on the stock structure and extinction risk of the species, and that all scientific findings be both reasonable and supported by valid information contained in the documents. Therefore, we seek a CIE review of the scientific information in the workshop reports on river herring based on the Terms of Reference (ToRs) to be developed. The CIE reviewers will help to ensure an independent, scientific review of information for a management process that is very public and is likely to be highly controversial no matter what NMFS' listing decision is. The Terms of Reference (ToRs) of the peer review are attached in **Annex 2**.

Requirements for CIE Reviewers: Three CIE reviewers shall conduct an impartial and independent peer review in accordance with the SoW and ToRs herein. CIE reviewers shall have combined working knowledge and recent experience in one or all of the following: 1) fisheries population dynamics, expertise in stock assessment and life history of anadromous species; and/or 2) expertise in extinction risk analysis and population modeling; and/or 3) expertise in stock structure and genetics analysis. It is desirable that the extinction risk analysis expertise be familiar with applications in fisheries, particularly anadromous species. Each CIE reviewer's duties shall not exceed a maximum of 10 days to complete all work tasks of the peer review described herein.

Location of Peer Review: Each CIE reviewer shall conduct an independent peer review as a desk review, therefore no travel is required.

Statement of Tasks: Each CIE reviewers shall complete the following tasks in accordance with the SoW and Schedule of Milestones and Deliverables herein.

<u>Prior to the Peer Review</u>: Upon completion of the CIE reviewer selection by the CIE Steering Committee, the CIE shall provide the CIE reviewer information (full name, title, affiliation, country, address, email) to the COTR, who forwards this information to the NMFS Project Contact no later than the date specified in the Schedule of Milestones and Deliverables. The CIE is responsible for providing the SoW and

ToRs to the CIE reviewers. The NMFS Project Contact is responsible for providing the CIE reviewers with the background documents, reports, and other pertinent information. Any changes to the SoW or ToRs must be made through the COTR prior to the commencement of the peer review.

Pre-review Background Documents: Two weeks before the peer review, the NMFS Project Contact will send (by electronic mail or make available at an FTP site) to the CIE reviewers the necessary background information and reports for the peer review. In the case where the documents need to be mailed, the NMFS Project Contact will consult with the CIE Lead Coordinator on where to send documents. CIE reviewers are responsible only for the pre-review documents that are delivered to the reviewer in accordance to the SoW scheduled deadlines specified herein. The CIE reviewers shall read all documents in preparation for the peer review.

Desk Review: Each CIE reviewer shall conduct the independent peer review in accordance with the SoW and ToRs, and shall not serve in any other role unless specified herein. **Modifications to the SoW and ToRs can not be made during the peer review, and any SoW or ToRs modifications prior to the peer review shall be approved by the COTR and CIE Lead Coordinator.** The CIE Lead Coordinator can contact the Project Contact to confirm any peer review arrangements.

Contract Deliverables - Independent CIE Peer Review Reports: Each CIE reviewer shall complete an independent peer review report in accordance with the SoW. Each CIE reviewer shall complete the independent peer review according to required format and content as described in Annex 1. Each CIE reviewer shall complete the independent peer review addressing each ToR as described in Annex 2.

Specific Tasks for CIE Reviewers: The following chronological list of tasks shall be completed by each CIE reviewer in a timely manner as specified in the **Schedule of Milestones and Deliverables**.

1) Conduct necessary pre-review preparations, including the review of background material and reports provided by the NMFS Project Contact in advance of the peer review.
2) Conduct an independent peer review in accordance with the ToRs (**Annex 2**).
3) No later than 4 September 2012, each CIE reviewer shall submit an independent peer review report addressed to the "Center for Independent Experts," and sent to Mr. Manoj Shivlani, CIE Lead Coordinator, via email to shivlanim@bellsouth.net, and CIE Regional Coordinator, via email to Dr. David Sampson david.sampson@oregonstate.edu. Each CIE report shall be written using the format and content requirements specified in Annex 1, and address each ToR in **Annex 2**.

Schedule of Milestones and Deliverables: CIE shall complete the tasks and deliverables described in this SoW in accordance with the following schedule.

9 August 2012	CIE sends reviewer contact information to the COTR, who then sends this to the NMFS Project Contact.

13 August 2012	NMFS Project Contact sends the stock assessment report and background documents to the CIE reviewers. Background documents may be sent to the CIE reviewers one week earlier.
20 August – 2 September 2012	Each reviewer conducts an independent peer review as a desk review.
4 September 2012	CIE reviewers submit draft CIE independent peer review reports to the CIE Lead Coordinator and CIE Regional Coordinator.
18 September 2012	CIE submits the CIE independent peer review reports to the COTR.
25 September 2012	The COTR distributes the final CIE reports to the NMFS Project Contact and regional Center Director.

Modifications to the Statement of Work: This 'Time and Materials' task order may require an update or modification due to possible changes to the terms of reference or schedule of milestones resulting from the fishery management decision process of the NOAA Leadership, Fishery Management Council, and Council's SSC advisory committee. A request to modify this SoW must be approved by the Contracting Officer at least 15 working days prior to making any permanent changes. The Contracting Officer will notify the COTR within 10 working days after receipt of all required information of the decision on changes. The COTR can approve changes to the milestone dates, list of pre-review documents, and ToRs within the SoW as long as the role and ability of the CIE reviewers to complete the deliverable in accordance with the SoW is not adversely impacted. The SoW and ToRs shall not be changed once the peer review has begun.

Acceptance of Deliverables: Upon review and acceptance of the CIE independent peer review reports by the CIE Lead Coordinator, Regional Coordinator, and Steering Committee, these reports shall be sent to the COTR for final approval as contract deliverables based on compliance with the SoW and ToRs. As specified in the Schedule of Milestones and Deliverables, the CIE shall send via e-mail the contract deliverables (CIE independent peer review reports) to the COTR (William Michaels, via William.Michaels@noaa.gov).

Modifications to the Statement of Work: This 'Time and Materials' task order may require an update or modification due to possible changes to the terms of reference or schedule of milestones resulting from the fishery management decision process of the NOAA Leadership, Fishery Management Council, and Council's SSC advisory committee. A request to modify this SoW must be approved by the Contracting Officer at least 15 working days prior to making any permanent changes. The Contracting Officer will notify the COTR within 10 working days after receipt of all required information of the decision on changes. The COTR can approve changes to the milestone dates, list of pre-review documents, and ToRs within the SoW as long as the role and ability of the CIE reviewers to complete the deliverable in accordance with the SoW is not adversely impacted. The SoW and ToRs shall not be changed once the peer review has begun.

Acceptance of Deliverables: Upon review and acceptance of the CIE independent peer review reports by the CIE Lead Coordinator, Regional Coordinator, and Steering Committee, these reports shall be sent to the COTR for final approval as contract deliverables based on compliance with the SoW and ToRs. As specified in the Schedule of Milestones and Deliverables, the CIE shall send via e-mail the contract deliverables (CIE independent peer review reports) to the COTR (William Michaels, via William.Michaels@noaa.gov).

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Key Personnel:

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Annex 1: Format and Contents of CIE Independent Peer Review Report

1. The CIE independent report shall be prefaced with an Executive Summary providing a concise summary of the findings and recommendations, and specify whether the science reviewed is the best scientific information available.

2. The main body of the reviewer report shall consist of a Background, Description of the Individual Reviewer's Role in the Review Activities, Summary of Findings for each ToR in which the weaknesses and strengths are described, and Conclusions and Recommendations in accordance with the ToRs.

3. The reviewer report shall include the following appendices:

Appendix 1: Bibliography of materials provided for review
Appendix 2: A copy of the CIE Statement of Work

Annex 2: Terms of Reference for the Peer Review

River Herring (Alewife and Blueback Herring)
Stock Structure and Extinction Risk Analysis

Provide a scientific peer review of Stock Structure and Extinction Risk Analysis reports on river herring (alewife and blueback herring) in accordance to the following terms of reference:

1. Is the information regarding the life history and population dynamics of the species the best scientific information available? If not, please indicate what information is missing and if possible, provide sources.
2. Does the information on river herring genetics, physiological, behavioral, and/or morphological variation presented for the species' range represent the best scientific information available? If not, please indicate what information is missing and if possible, provide sources.
3. Based on the scientific information presented, are the conclusions regarding species, subspecies, or distinct population segment delineations supported by the information presented? If not, please indicate what scientific information is missing and if possible, provide sources.
4. Based on the scientific information presented in the extinction risk analysis report, does this analysis consider all of the best available data and are the conclusions appropriate and scientifically sound? If not, please indicate what information is missing and if possible, provide sources.
5. In general, are the scientific conclusions in the reports sound and interpreted appropriately from the information? If not, please indicate why not and if possible, provide sources of information on which to rely.
6. Where available, are opposing scientific studies or theories acknowledged and discussed? If not, please indicate why not and if possible, provide sources of information on which to rely.
7. In general, is the best scientific and commercial data available for the stock structure and extinction risk analysis of river herring presented in the reports? If not, please indicate or provide sources of information on which to rely.