COMMENTARY

The utility of transcriptomics in fish conservation

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ABSTRACT

There is growing recognition of the need to understand the mechanisms underlying organismal resilience (i.e. tolerance, acclimatization) to environmental change to support the conservation management of sensitive and economically important species. Here, we discuss how functional genomics can be used in conservation biology to provide a cellular-level understanding of organismal responses to environmental conditions. In particular, the integration of transcriptomics with physiological and ecological research is increasingly playing an important role in identifying functional physiological thresholds predictive of compensatory responses and detrimental outcomes, transforming the way we can study issues in conservation biology. Notably, with technological advances in RNA sequencing, transcriptome-wide approaches can now be applied to species where no prior genomic sequence information is available to develop species-specific tools and investigate sublethal impacts that can contribute to population declines over generations and undermine prospects for long-term conservation success. Here, we examine the use of transcriptomics as a means of determining organismal responses to environmental stressors and use key study examples of conservation concern in fishes to highlight the added value of transcriptome-wide data to the identification of functional response pathways. Finally, we discuss the gaps between the core science and policy frameworks and how thresholds identified through transcriptomic evaluations provide evidence that can be more readily used by resource managers.

KEY WORDS: Conservation physiology, Endangered species, Molecular approaches

Introduction

The past two decades have seen an exciting increase in integrative, interdisciplinary research linking mechanistic responses at the cellular level with whole organism-, population-, community- and even ecosystem-level effects (Bay et al., 2017; Miner et al., 2012; Whitham et al., 2006). Genomic technologies have played key roles in these efforts (e.g. microarrays and more recently, high-throughput RNA and DNA sequencing approaches), contributing to significant advances in organismal physiology, evolutionary biology and ecology (Alvarez et al., 2015; De Wit et al., 2015; Evans, 2015; Miller and Maclean, 2008; Miller et al., 2014). The incorporation of

sequencing approaches into physiological and ecological research has largely been facilitated by technological advancements coupled with drastic cost reductions and improved bioinformatics resources. It is now feasible to employ these techniques for non-model species for which there are limited genomics resources (MacConaill, 2013). This has provided key insights into the evolution of adaptive phenotypes in natural populations as well as a mechanistic understanding of the physiological responses to various environmental stressors (e.g. Dayan et al., 2015; Narum and Campbell, 2015; Velotta et al., 2017).

Beyond fundamental advances, the application of genomics tools to evaluate genetic diversity, population size, phenotypic plasticity and adaptive potential to environmental and anthropogenic drivers of habitat change is increasingly contributing to conservation biology. Although many studies using genomic technologies in conservation biology have focused on population-level evolutionary processes (e.g. Allendorf et al., 2010; Garner et al., 2016; Kohn et al., 2006; McMahon et al., 2014; Shafer et al., 2015), interactions with changing environmental conditions involve numerous behavioral and physiological phenotypes (Kültz et al., 2013) that occur over shorter timescales and are heavily influenced by genomic variation between populations. Therefore, there are strong arguments for using both population genetics and transcriptomics in conservation management applications (Miller et al., 2014; Ouborg et al., 2010; Vandersteen Tymchuk et al., 2010). Together, these approaches can not only assess the capacity and constraints of natural populations to contend with changing environments, but also the potential of these populations to adapt to new challenges (Corlett, 2017). An example of this is provided by Velotta et al. (2017), who identified transcriptional mechanisms underlying adaptation to freshwater in alewife (*Alosa pseudoharengus*). Using saltwater- and freshwater-tolerant, landlocked populations of ancestrally anadromous alewife, the researchers identified parallel divergence in both expression and types of genes involved in osmoregulation, which suggested that not only quantitative but also qualitative changes in the gill transcriptome response to salinity might facilitate freshwater adaptation. Eliason et al. (2011) showed thermal adaptations in upstream spawning migration in Fraser River sockeye salmon (*Oncorhynchus nerka*) populations. Additionally, Jeffries et al. (2014b) identified differences in the transcriptome response across different populations and species of Fraser River Pacific salmon (*Oncorhynchus* spp.). Evidence of local adaptation to summer water temperatures has also been presented by Gradil et al. (2016), who demonstrated heritable differences in cardiac performance in distinct populations of Atlantic salmon (*Salmo salar*). These studies show how variable natal environments or stream conditions during migration can drive divergent selection among populations.

Integrative biology

In addition to improving knowledge of the fundamental biology of species, integrating transcriptomics with complementary

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Glossary

Conservation biology
Scientific discipline focused on the study of phenomena affecting the maintenance, loss and restoration of biological diversity, including development of approaches for effective protection and sustainable management of species, habitats and ecosystems.

Cytokine
Broad category of small proteins that play important roles in cell signaling, particularly as chemical messengers that control immune responses.

Driver
Natural or human-induced factor that directly or indirectly causes a change.

Functional genomics
The use of genomic data to describe gene functions and interactions.

Gene ontology
Framework used to describe gene function(s), and relationships between them.

Genotype
All of the heritable genetic information carried by an organism in its DNA; frequently refers to some relevant subset or component of the DNA passed to the organism by its parent.

High-throughput sequencing (HTS)
A category of approaches capable of processing multiple DNA and RNA sequences in parallel; sub-categories are often also referred to as 'next-generation sequencing', 'massively parallel sequencing' or 'second-generation sequencing'.

Mesothermal
Species that exhibit the capacity to tolerate and acclimate to a modest range of temperatures.

Microarray
A high-throughput transcriptome analysis platform involving a glass slide spotted with thousands of specific DNA sequences.

Phenotype
The set of observable characteristics (e.g. biochemistry, physiology, morphology, behavior) of an individual resulting from the interaction of its genotype with the environment.

Phenotypic plasticity
The capacity of an individual or a single genotype to produce more than one phenotype under different environmental conditions.

Quantitative polymerase chain reaction (qPCR)
The continuous collection of fluorescent signal obtained through polymerase chain reaction used to quantify gene transcript levels.

RNA sequencing (RNA-Seq)
High-throughput sequencing of RNA used to determine differential expression of transcripts between samples.

Smolt
Stage in the lifecycle of anadromous salmon characterized by hormonal and other physiological changes that prepare the fish for migration from fresh water to the sea.

Transcriptome
All messenger RNA (mRNA) molecules expressed within a cell or tissue sample.

physiological and ecological metrics can contribute to effective species management, population recovery and potentially aid captive rearing (e.g. aquaculture and hatcheries) by determining environmental requirements and critical thresholds, such as for optimal growth or stress responses. An environmental stressor, as defined by Somero et al. (2016) "…is any physical or chemical factor that can perturb the physiological and biochemical systems of organisms and, above some threshold value of intensity, lead to suboptimal values for these traits". Understanding the impact of sublethal stress and distinguishing compensatory responses (i.e. stress responses from which an organism can adjust its physiology and recover) from those that indicate irreversibly detrimental

effects, is particularly crucial within conservation contexts. The sensitivity of transcriptomics relative to coarser metrics of physiological performance, such as tolerance assays, makes it a valuable tool not only to identify specific functional physiological thresholds, but also to model responses that tease apart these differences. Carefully designed transcriptomic studies can reveal and utilize points of inflection in gene responses along a stress gradient (or duration of exposure) to delineate and differentiate between ranges of homeostasis and compensation, potential reversibility and recovery, as well as determine disease status and be predictive of mortality (Fig. 1). A common approach has been to rely on well-described biomarkers (e.g. heat shock protein levels to evaluate the magnitude of protein damage in response to a stressor) to assess specifically defined endpoints. Transcriptome-wide approaches, by contrast, identify pathway-specific responses, which can be used to develop both molecular and biochemical biomarker suites targeted at specific functional pathway assessments. Transcriptomics can thus be used in hypothesis-driven research as well as discovery research.

Transcriptomics and environmental change
The growing field of conservation physiology has spearheaded the movement of incorporating physiological tools into assessments of species responses to anthropogenic stressors and environmental change (Wikelski and Cooke, 2006). In particular, now that functional genomics are being increasingly applied in non-model species, transcriptomics approaches are well poised to enhance the field of conservation physiology. Recent studies have begun to realize some of these possibilities, such as how functional transcriptome-wide data can be used to improve or refine adaptive management plans geared towards species conservation (Connon et al., 2012; Jeffries et al., 2014a; Komoroske et al., 2015; Mykles et al., 2016; Narum and Campbell, 2015; Wellband and Heath, 2017). However, despite rapid growth in the field of conservation physiology, there are relatively few tangible examples where these approaches have been incorporated into species management efforts (Cooke et al., 2012, 2013). To shed light on the state of this field and motivate future advances, here we broadly discuss how transcriptomics tools (i.e. high-throughput qPCR, microarray and RNA-sequencing studies) can be leveraged to address needs in applied fisheries biology and conservation. We highlight key recent advances and forthcoming applications, as well as challenges in functional knowledge-translation towards facilitating management efforts. We primarily draw on examples and issues from applied fisheries biology to demonstrate these concepts because these taxa have been extensively studied in fundamental science, but there are also clear research needs towards the conservation management of natural populations and the cultivation of sustainable aquaculture. RNA-sequencing technology can be used to conduct functional genomic assessments on species for which there is no prior molecular information available and can be used to investigate sequence variation between individuals or populations to identify specific genotypes involved in a phenotypic response (e.g. Manousaki et al., 2013; Zhang et al., 2015). Species of conservation concern are often not amenable to established approaches owing to sensitivity to captivity, limited availability as a result of their conservation listing, or when information on fundamental life history parameters precludes physiological assessments (Ekblom and Galindo, 2011; Jeffries et al., 2016; Luo et al., 2014). Because transcriptomic technologies and approaches avoid some of these challenges, they are increasingly emerging for species of conservation concern. Our focus here is thus
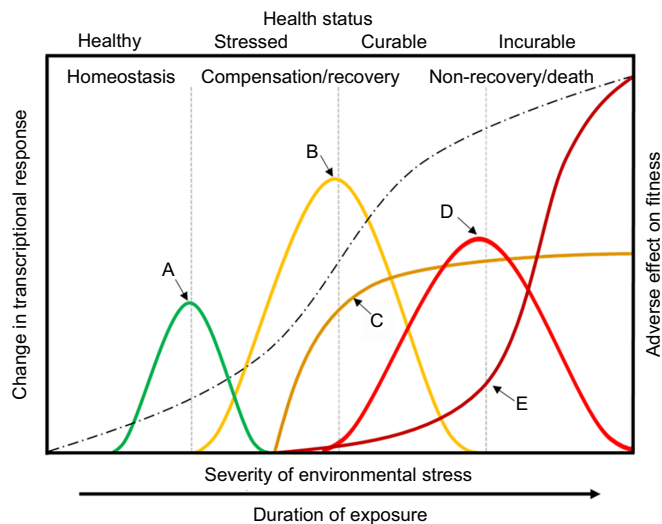
Fig. 1. Conceptual transcriptional response profiles across a multitude of functional pathways with increasing severity of environmental stress or duration of exposure. Points of inflection are likely indicative of specific adverse effects and functional thresholds; homeostasis and compensation (A), potential reversibility and recovery (B), or predictive of disease and subsequent mortality (C,D,E). The dashed black line represents a hypothetical, sigmoidal relationship between adverse effect on fitness (right-hand y-axis) and severity of environmental stress. Adapted and updated from Depledge et al. (1993), Moore (2002) and Connon (2003).

on the use of transcriptomics in conservation physiology as a systems-wide approach to understand mechanisms of tolerance and adaptation to environmental change. We emphasize that transcriptomics is by no means a substitute for whole-organism physiology or ecological measures, but rather a complementary tool that can deepen our understanding of the mechanisms underlying physiological responses manifesting as sensitivity, tolerance, phenotypic plasticity and adaptation.

Understanding the mechanistic responses to environmental conditions

Translating genomics technology for use in conservation biology remains a challenging effort, and the basic approach is presented in
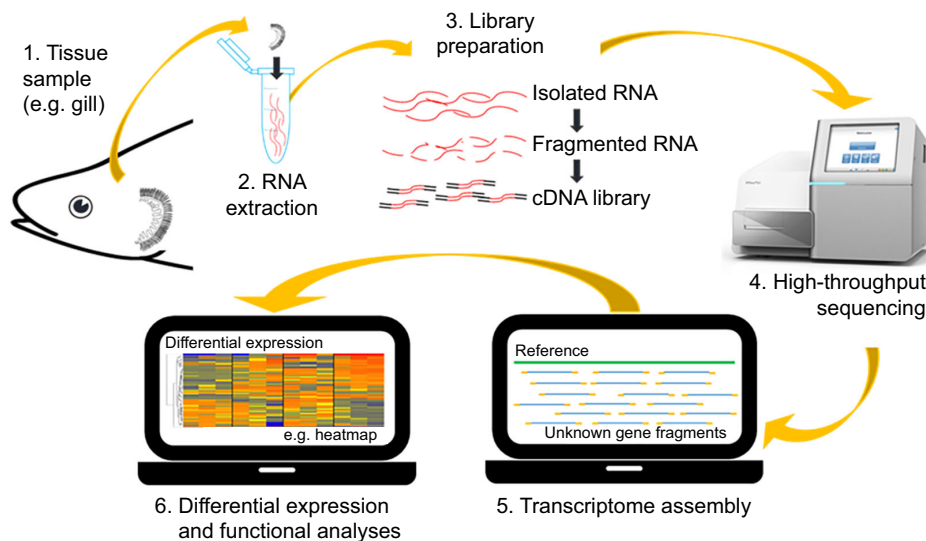
Fig. 2. The utility of transcriptomics is clear in fundamental research seeking to uncover molecular and biochemical mechanisms underlying a phenotypic trait or functional ability under different environmental conditions. However, from an applied perspective, the knowledge gained from transcriptomics approaches that cannot be attained through traditional assessments is not always initially apparent. While toxicological and medical research, for example, focus primarily on development and disease in model species [e.g. zebrafish (*Danio rerio*), fathead minnows (*Pimephales promelas*), Japanese medaka (*Oryzias latipes*)], physiological ecology research attempts to use species relevant to a particular system to evaluate and associate responses from individuals to the level of populations, communities and ecosystems.

It is important to underscore that a fundamental understanding of how organisms function and respond to their environments is a linchpin of effective applied biology. While traditional physiological and ecological metrics such as tolerance, growth, reproductive output and survival provide crucial information on the capacity of populations to persist under different environmental conditions, they do not provide insight into the mechanism of how the organisms respond the way they do. These mechanisms become evident when transcriptome-wide assessments are utilized to understand specific ecological questions. For example, in a comparative transcriptomics study evaluating thermal tolerance differences between two ecologically divergent populations of redband trout (*Oncorhynchus mykiss gairdneri*), Narum and Campbell (2015) identified adaptive patterns of gene expression underlying thermal adaptation to warmer aquatic climates. Adaptive gene expression responses were associated not only with thermal stress, but also with oxygen limitation at elevated temperatures. Furthermore, comparative transcriptomics has been used to link transcriptome plasticity with thermal tolerance and differential invasion success in round goby (*Neogobius melanostomus*) and tubenose goby (*Proterorhinus semilunaris*), two non-native species in the Great Lakes of North America (Wellband and Heath, 2017). Transcriptomic assessments, as previously discussed, are most powerful when they are integrated with other more conventional metrics to link across biological levels – that is, associating molecular mechanisms to crucial physiological and ecological endpoints. This provides insight into processes that can form a predictive framework to assess potential effects under new, untested

Fig. 2. Schematic representation of a transcriptomic evaluation approach. Transcriptomic approaches such as RNA-Seq involve the sampling of specific tissues (e.g. fish gill) of interest (1), extraction of RNA from the tissue (2), construction of cDNA libraries (3) and use of high-throughput sequencing technology that produce short fragmented reads (4). These fragments are then aligned and assembled based on homology to the reference genome of a closely related species or through a *de novo* assembly of a reference transcriptome (5). Differential expression of transcripts between samples of interest can then be computed and functionally analyzed towards describing gene interactions and potential consequences (6).

conditions. For example, using a transcriptomics approach, researchers uncovered a molecular signature in adult sockeye salmon (*Oncorhynchus nerka*) that failed to survive upriver spawning migration that was consistent with a response to a pathogen infection, explaining a potential cause of premature mortality occurring in an economically and ecologically important species (Miller et al., 2011).

Several studies conducted on multiple salmonid species have not only supported, but heightened, the importance of such mechanistic studies. Prior transcriptome-wide assessments were used in the determination of differing metabolic shifts in adult Fraser River sockeye salmon (*O. nerka*) during their spawning migration, as they return through estuarine environments towards their freshwater spawning sites (Miller et al., 2009). While early starvation in saltwater resulted in elevated protein turnover associated with arrival in the estuarine environment and a shift from anaerobic glycolysis to oxidative phosphorylation in muscle tissue, genes responding within freshwater environments were associated predominantly with thermal stress and immune system function, corresponding with elevated migration mortality (Miller et al., 2009). Immune system activation has also been shown to be common in transcriptomic studies that evaluate the effects of temperature on wild salmonids, as chronic exposure to high water temperatures leads to increased incidence of disease (Jeffries et al., 2012; Miller et al., 2014). Significant associations between metabolic gene transcription and thermal regimes, as well as cytokine gene transcription and pathogen abundance, have been demonstrated in rainbow trout (*O. mykiss*) metabolic and immune challenge studies (Wellband and Heath, 2013). In fact, studies that focused on disease have been highly successful in using transcriptome data to predict salmonid migration success (Connon et al., 2012; Jeffries et al., 2014a; Miller et al., 2011).

Understanding when exceeding sublethal thresholds leads to fitness consequences

One of the major challenges of understanding the impact of changing environmental conditions on fishes is determining the point at which sublethal responses to stressors begin to adversely impact the organism (Somero et al., 2016). Exposure to an environmental stressor or stressors can be considered detrimental when there are impacts on ecological fitness (Schulte, 2014), most simply reproduction and survival. Although lethal impacts are conceptually easier to demonstrate, and articulate to resource managers, sublethal impacts can lead to altered reproductive output or susceptibility to disease and contribute to population declines over generations. Characterizing such sublethal impacts of environmental change is an equally important consideration from a management perspective. Therefore, an understanding of when sublethal thresholds that lead to adverse impacts on a species are exceeded is crucial to understanding habitat requirements of a species of conservation concern and facilitating their management in a changing environment. For example, Komoroske et al. (2014, 2015) evaluated physiological and transcriptomic responses to thermal stress at multiple levels of biological organization on the delta smelt (*Hypomesus transpacificus*), a mesothermal endangered fish species. These studies demonstrated that sublethal critical thresholds occur 4–6°C below upper acute tolerance limits for this species. This work also provided evidence of limited thermal plasticity due to an inability to effectively make physiological adjustments, resulting in elevated risks associated with chronic thermal stress. Similarly, Jeffries et al. (2016) used transcriptomics to evaluate and contrast thermal stress responses of delta smelt with

another native species of conservation concern, the longfin smelt (*Spirinchus thaleichthys*). The niche of the larval stage for these two species overlap spatially *in situ*; however, the acute thermal tolerance for larval longfin smelt is ∼3°C lower than that of larval delta smelt. Combining transcriptomics with whole-organism respirometry and tolerance assays, this study determined mechanistic differences in the thermal sensitivity between these two species. By evaluating responses at multiple levels of biological organization, these studies provide insight into physiological mechanisms contributing to the variation in thermal tolerances observed across species. Additionally, the findings suggest that both species might already be living close to their thermal limits and could be experiencing sublethal impacts, given that water temperatures in their habitat range frequently occur beyond critical sublethal stress thresholds. The conservation utility of these data was further demonstrated when sublethal and lethal thermal thresholds for delta smelt were integrated with climate change model projections to forecast future habitat suitability (Brown et al., 2016). This work revealed that a large proportion of the delta smelt's existing habitat range is forecast to be thermally suboptimal or unsuitable within several decades, and likely to result in strong habitat compression for this species.

As with thermal stress, transcriptomic evaluations have been highly successful in determining mechanisms underlying tolerance thresholds and adaptations to osmotic stress (Brennan et al., 2015; Evans and Somero, 2008; Lockwood and Somero, 2011; Somero, 2010; Whitehead et al., 2011, 2012), providing a framework for the assessment of osmotic habitat requirements for species of conservation concern. Komoroske et al. (2016) coupled transcriptome-wide assessments with organismal measures of condition and tolerance in delta smelt to evaluate mechanistic relationships contributing to habitat range limitations correlated with salinity gradients. The observed altered gene expression associated with osmoregulation and growth, along with whole-organism physiological responses (Komoroske et al., 2014), suggested that, although delta smelt can tolerate and effectively osmoregulate at salinities substantially greater than their current limited natural distribution, higher salinities would likely impose sublethal energetic costs on physiological performance. Thus, by linking mechanisms across biological levels and relating findings to environmental conditions *in situ*, the authors used transcriptomic tools to shed light on the dynamics of compensatory sublethal responses.

Biomarker development for rapid assessment of physiological condition

Transcriptomic approaches, such as those described above, have led to the development of mechanistic tools that can be used within an assessment framework towards management of species of conservation concern. An excellent example of using functional genomics approaches as a component of conservation physiology comes from Pacific salmon research (Cooke et al., 2012). Following a pivotal study that used transcriptomics screening approaches to identify a disease-related immune signature predictive of migration mortality in sockeye salmon (Miller et al., 2011), increased efforts have been made to develop biomarkers of diseased fish for managing Pacific salmon migration in British Columbia. Using current molecular tools available for Pacific salmon species, biomarkers of stress, immune responses and common salmon pathogens, have been developed for rapid screening of wild-caught salmon in the Fraser River system (Miller et al., 2014). This screening approach was used to identify a viral pathogen that

contributes to migration mortality in seemingly healthy wild sockeye salmon smolts (Jeffries et al., 2014b). Transcriptomics has also been successfully used to assess the effects of pathogen infection in Chinook salmon in a field setting (Bass et al., 2017) and sockeye salmon in a laboratory setting (Teffer et al., 2017). Transcriptome screening is an extremely valuable predictive approach for monitoring wild fish health as, often, fish that become symptomatic of a disease die off and are not collected in population surveys in the river or on the spawning grounds. Therefore, detecting evidence of infection early can help explain some of the causes of premature mortality that occur in natural populations. To our knowledge, except within the field of ecotoxicology (in particular associated with endocrine disruption), the described conservation physiology studies on salmonid species are some of the best examples of successfully using transcriptomics in association with evaluating detrimental impacts in natural populations, and therefore these salmonid studies can serve as a model system for other species of conservation concern.

Transferring our understanding of mammalian functional systems to species of conservation concern

Comprehensive reviews are available on best practices and challenges of using transcriptomics in comparative biology and ecological genomics (e.g. Alvarez et al., 2015; Mykles et al., 2016), many of which are broadly applicable to non-model species and particularly relevant for using transcriptomics in conservation physiology and translating findings to inform effective species management and conservation. One important challenge is the need to improve the transferability and understanding of mammalian (human models) functional response pathways to those of fishes as well as other vertebrate and non-vertebrate species. As discussed earlier, because RNA-sequencing technology can be used to conduct *de novo* transcriptome-wide assessments on species with no prior molecular information available, these approaches can now be applied to fishes of immediate conservation concern (Jeffries et al., 2016). Analytically, the responding transcripts are often grouped together based on functional pathways to assess the 'functional response' in the transcriptome profiles. This approach requires annotating and assigning functions to the mRNA sequences, and using analysis approaches that involve predicted gene functions [i.e. gene ontologies (GOs); Primmer et al., 2013]. However, it is important to recognize that although the GOs are intended to be species-neutral, much of the information regarding gene functions are in practice derived from mammals. This can especially be an issue when studying tissues that do not have direct homologs in mammals, or organs that have multiple analogous functions, such as the fish gill, where high sequence homology and functionality has been identified between gill and branchial cell remodeling (Lam et al., 2014), or renal epithelia (Evans et al., 2005). Furthermore, there are often species-specific transcripts for which no functional annotation is assigned (Primmer et al., 2013), despite having been discovered as significant in the response to a particular stressor. Caution has been proposed regarding annotation of transcripts with highly evolutionarily divergent species (Alvarez et al., 2015) – instead, researchers should create databases of unannotated transcripts to contribute to identifying the ecological function of these transcripts (Pavey et al., 2012). Likewise, researchers using transcriptomics in conservation physiology studies should recognize the role of these transcripts in the response to environmental stressors, rather than focusing the interpretation of the expression data on transcripts with annotation or transcripts with well-defined functions. A collaborative approach

to address this challenge is to form large consortia that work towards identifying the ecological functions of transcripts specific to a species or group of species. One such organization, serving as an example of the direction that genomics research on species of conservation concern should go, is the Functional Analysis of All Salmonid Genomes (FAASG) initiative (Macqueen et al., 2016). This is a particularly exciting initiative as these efforts will strive to provide valuable functional annotation for the genomes of a family of species with enormous economic, cultural and ecological importance globally.

Much of our understanding of gene functions is sourced from developmental biology and toxicology studies. There is the potential for genes to have different or multiple functions in evolutionarily divergent species (Primmer et al., 2013), and many genes function across numerous functional pathways (crosstalk), resulting in differential gene expression that can confound data interpretation. These factors currently limit our understanding of the true functional responses determined via transcriptomics studies in species of conservation importance. However, while current functional annotation tools rely on gene-function overlap that might fail to correctly identify altered pathways, there are efforts under way to establish analytical processes by which functional association networks can improve pathway annotations and function (Ogris et al., 2017). As these resources continue to improve, researchers can also advance our understanding of gene function within specific pathways by designing robust studies that integrate functional genomics with phenotypic metrics, such as physiological threshold evaluations.

Mining existing transcriptomic datasets to evaluate and further interpret mechanistic responses to stress

A recent increase in publications on multiple stressor transcriptomic response, along with strict requirements for archiving transcriptomic data in publically accessible repositories (e.g. Gene Expression Omnibus, https://www.ncbi.nlm.nih.gov/geo/; Sequence Read Archive, https://trace.ncbi.nlm.nih.gov/Traces/sra/sra.cgi?), provides a wealth of knowledge to predict risk. Archived data provide the means for conducting meta-analyses, providing a scaffold on which to evaluate and further interpret mechanistic thresholds of stress. Efforts should be made to mine existing transcriptomics datasets or conduct meta-analyses to identify transcriptomic responses (gene groups or functional pathways) characteristic of particular stressors that can be used to develop biomarker suites for responses across species. Wang et al. (2016) provide a powerful example of how a meta-analysis approach can be used towards developing data-driven bioinformatics applications for determining adverse outcomes. Utilizing human connectivity mapping (Cmap) – an *in silico* approach developed for biomedical research – the researchers evaluated over 3500 zebrafish (*D. rerio*) and fathead minnow (*P. promelas*) transcriptome profiles; driven by mechanistic physiological responses, through which the investigators identified stressor-specific signatures, this methodology aided the development of biomarkers focused on stressors of concern. Another example was recently described by Miller et al. (2017), who utilized transcriptomics metadata analysis for the development of biomarkers capable of distinguishing an active viral disease state from bacterial and parasitic disease states in fishes. While metadata approaches such as these are confounded by issues such as data heterogeneity resulting from differences in laboratory practices, experimental design and high variability in expression among individuals, across different tissue types, sex, age and physiological status (Simmons et al., 2015), the vastness of the datasets serves to

strengthen the identification of response commonalities across multiple species and can also be targeted at baseline response information. This approach, however, is not necessarily amenable to evaluating response singularities associated with sensitive species of conservation concern, as the basis of this sensitivity is likely related to physiological characteristics that might themselves have resulted in the species' endangered status. The power of transcriptomics in predicting responses to environmental stressors ultimately relies on the strength of analytical systems and models that are biologically meaningful and reflective of underlying processes (Kish et al., 2016) specific to the populations being studied.

Communicating functional data to resource managers

Ultimately, transcriptomics can only truly contribute to conservation if it is effectively integrated into management decisions and policies. As is the case in a variety of disciplines, many scientific publications state that transcriptomic data 'will be informative towards conservation efforts'; however, the majority of this knowledge never reaches the regulatory agencies and managers who need it in an adequate and useable form. Enhanced efforts are needed to communicate research findings to stakeholders and regulators. Mismatches between academic and management language (i.e. jargon), requirements and decision frameworks often hamper translation of information. Cooperative efforts are under way to establish more effective interactions between resource managers and research groups in order to determine specific regulatory requirements and how the vast data sets produced using transcriptomics approaches can be translated into a useable numerical endpoint, as is commonly needed in species management. Additionally, it is common for managers to be unfamiliar with interpreting complex molecular data, and researchers are not often aware of the structure and process of biological regulatory policy development. As a result, despite the rapidly growing number of studies on species of concern, utilization of transcriptomic knowledge in conservation currently remains relatively rare.

To overcome these barriers, physiologists must build partnerships with regulatory agencies, working directly with conservation biologists and resource managers to translate transcriptomics into management actions and policies. Functional responses and thresholds identified through transcriptomic evaluations provide evidence that can be used in risk assessment. For example, using differential expression to identify mechanistic response thresholds beyond which physiological functions such as osmoregulation are impaired (Komoroske et al., 2016). This approach can be used directly to identify a suitable habitat where similar or more severe stress responses would be elicited, and thus would require diversion of resources towards enhanced management or adjustments to restoration planning. Changes to immune system function, hormone synthesis, detoxification, neurodevelopment and a multitude of other developmental pathways, can thus be used towards determining specific management-relevant thresholds. These thresholds are particularly powerful when they are associated with responses that are manifested at higher levels of biological organization that are more familiar to managers. For example, linking molecular sublethal thresholds with well-accepted measures of whole organismal fitness (e.g. growth, disease risk, or thermal tolerance traits) that are often used to set environmental regulatory criteria can be one way to find common ground and integrate results into management frameworks. Well characterized molecular thresholds can serve as sensitive markers that can be used to predict whether organisms have the plasticity to evoke compensatory mechanisms that could lead to their recovery, or if they are more likely to suffer detrimental consequences (i.e. non-recovery; Fig. 1). Transcriptomic responses on their own can also provide a conservative benchmark for managing the habitat requirements of species of conservation concern in aquatic systems.

Concluding remarks

Transcriptomic assessments are highly successful in providing a mechanistic framework to understand organismal capacities and limitations for tolerance to changing environments. Mechanistic responses can point to sublethal thresholds beyond which there is a negative impact on individual and population fitness. As the number of transcriptome profiles across multiple species are archived in public repositories continues to increase and meta-analytical tools are further developed, our capacity to elucidate specific functional response pathways will also improve. This will further our understanding of the mechanisms that regulate an organism's ability to contend with environmental change. Enhanced efforts are needed to communicate research findings to stakeholders and regulators. Conservation physiologists must build partnerships with regulatory agencies, working directly with conservation biologists and resource managers to translate transcriptomics into management actions and policies.

Competing interests

The authors declare no competing or financial interests.

Author contributions

Conceptualization: R.E.C., K.M.J., L.M.K., A.E.T., N.A.F. Writing - original draft: R.E.C., K.M.J., L.M.K. Writing - review & editing: A.E.T., N.A.F.

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