THE ECOLOGY OF LAKES AND RIVERS IN THE SOUTHERN BOREAL SHIELD: WATER QUALITY, COMMUNITY STRUCTURE, AND CUMULATIVE EFFECTS

by

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ABSTRACT

Cumulative effects are the collective ecological effects of multiple human activities. Cumulative effects assessment (CEA) is concerned with quantifying effects of natural environmental factors and human activities. CEA has not lived up to its promise as a precautionary instrument for sustainability, in part because our knowledge of stressors and their effects is elementary; monitoring systems (needed to characterize ecological condition and how it changes over time) are insufficient; and numerical methods for associating stressors and effects, and for forecasting development outcomes, are lacking.

This thesis reviews the environmental appraisal literature to synthesize CEA's theoretical underpinnings, articulate its impediments, and establish that ecological monitoring and modelling activities are critical to success. Three research chapters overcome several scientific barriers to effective CEA. Data from spatial and temporal surveys of lake and stream water chemistry and benthic community structure are used to evaluate candidate monitoring indicators, identify minimally impacted reference waterbodies, characterize baseline water quality and biological condition, and quantify cumulative effects of land use and natural environmental variation (spatial survey: 107 lakes and 112 streams sampled in 2012 or 2013; temporal survey: 19 lakes sampled between 1993 and 2016).

The research was conducted in Canada's Muskoka River Watershed, a 5660 km^2 area of Precambrian Shield that drains to Lake Huron. This area's combination of extensive remaining natural areas and pervasive human influence makes it ideal for studying cumulative effects. It is also characterized by many lakes and their connecting stream and river channels, which integrate effects of stressors in their catchments and constitute logical focal points for CEA. Moreover, the local planning authority (District

Municipality of Muskoka) is striving to implement CEA and establish a cumulative effects monitoring program centered on water as its foremost resource; therefore, practical applications of the research have, been identified.

Universal numerical methods, which are transferrable to other study areas, are used. Random forest models (an extension of the algorithm used to produce classification or regression trees) are shown to model the singular and collective effects of land-use and natural factors on water chemistry and benthic community structure, and to quantify the sensitivities, and identify the important drivers of various chemical and biological indicators of aquatic ecosystem condition. Partial dependencies from the random forests (i.e., the mean predicted values of a given indicator that occurred across the observed range of a selected predictor) are paired with TITAN (Threshold Indicator Taxa Analysis) to investigate biological and chemical "onset-of-effect" thresholds along gradients of human development. Declining calcium concentrations and amphipod abundances are demonstrated in lakes, and generalized linear models forecast an average 57% decrease in the abundances of these animals to occur by the time lake-water calcium concentrations reach expected minima.

As its key findings, the thesis highlights sensitive indicators that should be included in a cumulative effects monitoring program, and are to be preferred when forecasting outcomes of changed land-use or environmental attributes. Empirical break-points, where effects of stressor exposures become detectable, are also identified. These thresholds can be used to distinguish reference and impacted conditions, so that normal indicator ranges and associated assessment criteria (important CEA precursors) can be objectively derived. In addition, the potential severity of cumulative effects is exemplified by marked declines in the abundances of lake dwelling amphipods, which could propagate through food webs to substantially alter soft-water Boreal ecosystems.

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Part 1: INTRODUCTION

Background

As the branch of biology that deals with interactions of organisms with one another, and with their physical environment, Ecology's fundamental goal is to explain and predict the occurrences and abundances of taxa (Townsend 1989, Belovsky et al. 2004, Temperton and Hobbs 2004). Its sub-discipline, Community Ecology, is concerned with explaining the distributions, abundances, and interactions of taxa (Liebold et al. 2004) that co-occur as assemblages at different places or times.

Community assembly refers to the processes by which communities arise. Current paradigms emphasize dispersal, niche limitation (taxa not capable of tolerating the physical conditions present at a given location being excluded from that location), biotic interactions (e.g., predation, competition), evolution, and neutral (random) processes of extirpation and colonization as most important (Belyea and Lancaster 1999, Hubbell 2001, Fussmann et al. 2007)[1].

Stressor exposures and natural disturbances are universal determinants of the taxonomic structure and functions of ecosystems (Hutchinson 1957). The term *ecological effect* describes a change to the structure or function of the ecosystem — an alteration of habitat, biota, or (and) their interactions. Such changes are typically measured against the normal range of variability associated with a specified spatial or temporal baseline (Kilgour et al. 1998). Effects can accumulate via repeated insults of a single stressor, or via the interaction of multiple stressors.

[1] These processes are arranged in a visually intuitive way in the Ecological Filters metaphor of community assembly, in which community assembly is viewed as a series of filters or sieves through which the regional pool of taxa must pass to determine community composition at any place or time (Mueller-Dombois and Ellenberg 1974, Van der Walk 1981, Drake 1990, Poff 1997, and Patrick and Swan 2011).

Stressor exposures, the type and intensity of disturbances, and the magnitude of the resulting ecological responses generally depend on one's reference point[2] (Belyea and Lancaster 1999, Weiher et al. 2011), and can vary spatially and temporally (Sousa 1980, 1984; Resh et al. 1988; Miller et al. 2011). Such complexity makes it difficult to understand and predict ecological effects (Lamothe et al. 2018). Indeed, our present ecological theories, concepts, and paradigms lack predictive power as a rule (Peters 1991, Keddy 1992a and 1992b), which is problematic because, "as ecology matures and as the world's environmental problems continue to multiply, the need for general predictive models also grows" (Shapiro 1993). Quantitative and predictive models of community assembly would lend themselves to scientific testing, and would have many deployments against the World's problems — application in environmental appraisal being an obvious example.

The worldwide process of environmental impact assessment was borne out of the 1969 US National Environmental Policy Act (Benson 2003, Pope et al. 2013), as a way to identify, predict, evaluate and mitigate the effects of development projects (Glasson et al. 2013). Scrutiny of the process has demonstrated that environmental impact assessment remains a young and unproven field (Bérubé 2007) in which cumulative effects, the combined effects of multiple human activities (Scherer 2011), have not been adequately considered (Damman et al. 1995, Duinker and Greig 2006, Gunn and Noble 2011).

Cumulative effects assessment (CEA) is both an applied science (Cashmore 2004), and a sub-discipline of environmental impact assessment (Morrison-Saunders et

[2] Scale is critically important because our ability to understand local and regional community assembly depends on the spatial scale at which communities are defined and the scale at which assembly processes operate (Weiher et al. 2011).

al. 2014). It was proposed as a way to evaluate the collective environmental effects of human actions (Dubé 2003, Seitz et al. 2011); however, its theoretical underpinnings have not been fully articulated, so it remains unclear precisely what CEA is *supposed to* achieve (Cashmore 2004, Judd et al. 2015). For this reason, agreement between its theory and practice has not been assessed (Lawrence 2000, Pope et al. 2013), and many signals of the deepening unsustainability of human enterprise — the remarkable transformation of Earth's surface, anthropogenic increases of atmospheric CO_2 concentrations, hijacking of the World's nitrogen cycle, excessive use and alteration of fresh water, and extinctions of numerous species (Vitousek et al. 1997, Foley et al. 2005, Steffen et al. 2007) —suggest it has been ineffectual as a precautionary sustainability strategy.

Because of different legislative and regulatory contexts, there is regional variation in the way CEA is practiced; however, it usually proceeds via the following six steps: (1) valued ecosystem components are identified to focus the appraisal on a small number of important ecosystem features (Canter and Atkinson 2011); (2) the physical and temporal boundaries of the appraisal are set; (3) activities capable of affecting valued ecosystem components are identified; (4) baseline condition is characterized using appropriate indicators; (5) cumulative effects of identified activities are modeled (and the significance of predicted effects is assessed); and (6) environmental monitoring is conducted to track realized outcomes and evaluate performance of the appraisal process (Damman et al. 1995; Ross 1998; Canter and Ross 2010; Connelly 2011; Seitz et al. 2011)

The inherent unpredictability of ecosystems and ecological effects makes uncertainty an unavoidable part of CEA (Chapman and Maher 2014). Monitoring is fundamental as a tool for mitigating the risks of uncertainty, because it indicates ecosystem form and function, allows the condition and variability of valued ecosystem components to be quantified (Ball et al. 2013), signals when conditions are changing (Cairns et al. 1993), and provides datasets that can be modeled to associate stressors with their ecological outcomes (e.g., Therivel and Ross 2007; Schultz 2012).

Biological (effect-based) and chemical (stressor based) indicators have complementary roles in CEA. Stressor-based indicators describe physico-chemical conditions and quantify stressor exposures (Roux et al. 1999). Biological indicators “integrate a cumulative response to environmental stress” (Munkittrick et al. 2000; Dubé 2003) by measuring ecosystem structure or function. Such indicators should be conceptually simple (so their meanings can be conveyed to diverse audiences), predictive, sensitive (to human and environmental factors relevant to CEA), precise enough that they can discriminate stressor-specific effects, and should have reasonable sampling and data requirements that make them economically feasible (Cairns et al. 1993; Niemi and McDonald 2004; Bonada et al. 2006).

CEA requires the effects of multiple stressors to be quantified, both singly and in combination (Judd et al. 2015). Descriptive or predictive models are vital tools for describing stressor effects, guiding the design of monitoring schemes, and predicting outcomes of alternative scenarios of human activity (Rubin and Kaivo-Oja 1999, Seitz et al. 2013, Russell-Smith et al. 2015).

Water is essential for life. For this reason, the ecosystem is commonly viewed as the primary user of water (Participants of Muskoka Summit for the Environment 2010), and access to sufficient clean water is commonly viewed as a human right (Gupta et al. 2010). Surface waterbodies, including lakes and rivers, are tightly coupled with chemical, physical, and biological processes that play out in their catchments (Hynes 1970, Wetzel 2001). They are exposed to (Nõges et al., 2016), and subsequently integrate effects of, a multitude of stressors (Lowell et al., 2000, Townsend et al., 2008, Ormerod et al. 2010, Jackson et al. 2016), which makes them some of the most threatened ecosystems on Earth (Schindler 2001, Carpenter et al. 2011) and a critical consideration for CEA. The Boreal region is well suited to research on cumulative effects (e.g., Sorensen et al. 2008, Houle et al. 2010) and cumulative effects assessment (e.g., Dubé et al. 2006, Seitz et al. 2011) because it contains many thousands of lakes, and is important globally as a vast relatively natural ecosystem, and as a source of timber, energy, minerals, and other natural resources (Luke et al 2007).

I selected a case-study boreal watershed, the Muskoka River Watershed, where I conducted a spatial survey of littoral benthic macroinvertebrate communities and water chemistry in 107 lakes and 112 rivers, and where a temporal survey of the same biological and chemical attributes had been underway since 1993. I selected the Muskoka River Watershed for several reasons: (1) Its gradients of geology, hydrology, and land-use provide variation that could be exploited to model community structure, community thresholds, and cumulative effects of multiple environmental factors. (2) The watershed has significant development, agriculture, and forestry — human activities known to alter a variety of physical, chemical, and biological properties of waterbodies, (Utz et al. 2009) — but is also rare in southern Canada for having a large number of lakes and streams that have no measurable development in their catchments, and are exposed only to atmospheric pollution, making them suitable for exploring minimally impacted (*sensu* Stoddard et al. 2006) reference conditions. (3) The watershed's lakes and rivers are changing physically, chemically and biologically — for example, air temperatures are rising and wind speeds are decreasing, which has led to phenological changes to lake ice cover and lake thermal stratification (Yao et al. 2013, Palmer et al. 2014); conductivity and the concentrations of calcium, phosphorus, and various metals are declining, while pH, and the concentrations of dissolved organic carbon, nitrogen, and chloride are increasing (Jeziorski et al. 2008, Eimers et al. 2009; Palmer et al. 2011); and many lakes have been invaded by the spiny water flea, which has resulted in reduced pelagic biodiversity (Yan et al. 2011). (4) Multiple ecological stressors are implicated in many of these changes (e.g., Watmough and Aherne 2008, Yao et al. 2013), which provides a suitable backdrop for cumulative effects research; and (5) one of Canada's primary urban and economic centres, the Greater Toronto Area (located within a 2-hour drive to the south) exerts considerable development pressure on the Watershed, makes the management of cumulative effects a fundamental concern, and suggests a variety of practical applications for research designed to monitor and model cumulative effects (Eimers 2016).

Problem Statement

Several challenges related to monitoring and modeling cumulative effects need to be overcome in order for cumulative effects assessment to be undertaken in boreal watersheds.

Our knowledge about stressors and effects on most ecosystems, including boreal lakes and streams, is rudimentary (Venier et al 2014). Integrating the information that does exist is problematic because cumulative effects assessment lacks straightforward and effective methods for associating stressors and effects, and for exploring the outcomes of development scenarios. Models capable of handling stressor interactions and non-linear responses are needed (Ball et al. 2013).

Great interest in monitoring cumulative effects has been generated in some locations (e.g., Eimers 2016), but robust monitoring systems generally don't exist (Ball et al. 2013, Dubé et al. 2013, Venier et al. 2014). Ecological condition can only be assessed relative to reference benchmarks and the normal range of natural variation (Kilgour et al. 1998, Hawkins et al. 2010, Mitchell et al. 2014, Clapcott et al. 2017), but chemical and biological reference conditions are not well understood for many areas, including Muskoka. Quantitative criteria based on measured exposures to stress are generally used to define what qualifies as a reference site; however, objective criteria for determining the level of stressor exposure that is acceptable are lacking[3].

From a theoretical perspective, the long-term stability of ecosystems suggests some capacity to resist disturbance; however, as human impacts or fluctuations in

[3]In fact, subjective judgement is the norm at multiple junctures in the process of ecological assessment, including the definition and selection of reference sites, selection of metrics and numerical methods for evaluating index performance (Feio et al 2016).

natural factors become extreme, a threshold may be reached where normal processes of community assembly are overridden and effects become apparent (Pardo et al. 2012, Roubeix et al. 2016). Such onset-of-effect thresholds have been hypothesized (Hilderbrand et al. 2010, Pardo et al. 2012), and could be used to objectively delineate the least-disturbed reference condition (Ciborowski et al. 2015), but there is little empirical evidence to support their existence (Pardo et al. 2012).

Chemical or biological condition is often summarized with indicator metrics but, for many Boreal areas, little information is available to describe how sensitive candidate indicators are to various natural environmental features and human stressors. So many questions remain about their accuracy and bias (i.e., how influenced they are by natural factors, and therefore what variables should be considered when matching reference sites and test sites; Hawkins et al. 2010), precision (i.e., how similar their scores are when measured in similar contexts), responsiveness (i.e., how predictably their scores change in response to stressor exposures), and sensitivity (i.e., how closely high and low scores correspond with high and low stressor exposures; Mazor et al. 2016). Perhaps the most critical questions for CEA pertain to how accurately metric values can be predicted, and how well suited they are for use in “futuring” analyses[4] (Therivel and Ross 2007; Canter and Atkinson 2011).

As an example of multiple environmental stressors and cumulative effects, declining calcium concentrations have been documented in many lakes in the southern Boreal Shield (Watmough and Aherne 2008, Jeziorski et al. 2008). Low calcium concentrations represent a stressor for Calcium-rich taxa, and (particularly when

[4] i.e., analyses undertaken to explore the range of outcomes possible from alternative scenarios of human activity

combined with other climate and development-related changes) cumulative effects on aquatic communities are possible (Jeziorski and Smol 2016). Declines of crayfish abundances (Edwards et al. 2009, Edwards et al. 2013, Hadley et al. 2015) and altered species composition of zooplankton communities (Jeziorski et al. 2008, Shapiera et al. 2012) have been demonstrated, but effects on amphipods (an abundant component of the benthic taxa that has high Ca demands) have not been investigated despite their potential to have important ecological consequences.

Research Objectives

This thesis begins with a philosophical chapter that synthesizes the theory behind cumulative effects assessment (Chapter 2 — Cumulative Effects Assessment: Theoretical Underpinnings and Big Problems). By way of a critical review, it clarifies societal aspirations for CEA, assesses agreement between its theory and practice, and articulates its foremost challenges and opportunities. Notable shortcomings associated with describing, predicting, and monitoring cumulative effects are addressed in three subsequent research chapters.

Chapter 3 (Random Forests as Cumulative Effects Models: A Case Study of Lakes and Rivers in Muskoka Canada) models cumulative effects and evaluates several candidate monitoring indicators using lake and river water chemistry and benthic-invertebrate community data from the case-study watershed in order to answer the following four questions: (1) which measures of lake and river water chemistry and benthic community structure can be modeled and predicted most accurately, and therefore show the most promise as cumulative effects indicators? (2) What are the combined and singular effects of human activities (e.g., urbanization, agriculture), hydrologic, physiographic, and morphometric factors on these indicators? (3) Can any chemical or biological attributes of lakes or rivers be predicted accurately enough, and are these models sensitive enough to land-use changes, to be used in scenario models that explore potential ecological consequences of increased development?

In this chapter, I propose that the suitability of any given candidate CEA indicator can be assessed according to how precisely and accurately it can be predicted from natural environmental features and how sensitively it responds to measures of human activities, and I demonstrate how random forests[5] (Breiman 2001) can be used to make these assessments and predict outcomes of alternative scenarios of human activity.

An approach for objectively distinguishing reference and impacted conditions is presented in Chapter 4 (Onset-of-effect Thresholds and Reference Conditions: A Case Study of the Muskoka River Watershed, Canada). In this paper, 107 lakes and 112 streams are classified as reference or impacted. According to the concept of minimal disturbance (Stoddard et al. 2006), sites having no exposures to land-use stress are designated as reference, and lakes or streams having measurable road density, urbanization, or agriculture in their catchments are considered impacted. Two complementary statistical methods (partial dependence plots from random forest models and TITAN [threshold indicator taxa analysis]) are then used to investigate the existence of chemical and biological onset-of-effect thresholds along stressor gradients. Where present, these thresholds — i.e., break-points where stressors begin to override

[5] Random forests are ensembles of classification or regression trees. They make no assumptions about the distributions of predictor or response variables. They can be used to assess datasets having a large ratio of predictors to observations. They can incorporate complex predictor interactions, without these interactions having to be pre-specified; and they can account for multicollinearity of predictors, and non-linear relationships between predictors and the response variable (Jones and Linder 2015).

natural processes such that effects become detectable — are used to make empirical adjustments to the criteria used to distinguish reference and impacted waterbodies. Normal reference ranges and typical impacted ranges of benthic community structure and water chemistry are then tabulated as critical percentiles and tolerance regions, the degree of interspersion of reference and impacted sites is assessed, and cumulative effects of land-use stress are described as spatial patterns of biological and chemical variation.

Chapter 5 (Declining Amphipod Abundance is Linked to Low and Declining Calcium Concentrations in Lakes of the south Precambrian Shield) investigates the biological effects of Ca decline (an emerging stressor in several parts of the Boreal where historical acid deposition, logging, and development has altered Ca weathering rates; Watmough and Aherne 2008) on amphipod abundances using data from the spatial survey of lakes described in Chapters 3 and 4, and also from a series of 19 long-term study lakes that were sampled between 1993 and 2016. From a spatial perspective, the study investigates whether amphipods (a crustacean with high calcium demands, relative to many other aquatic taxa; Cairns and Yan 2009) are more likely to be found in high-Ca lakes than in low-Ca lakes; whether there is a low-level Ca threshold, below which amphipods are unable to persist in lakes; and how important Ca is as a predictor of amphipod abundance, relative to the importance of other chemical, morphometric, or physical-habitat factors? From a temporal perspective, the study quantifies declines in amphipod abundance that have occurred over the last 23 years; assesses how important a role Ca has had in these declines, and forecasts how abundant amphipods will be in the future if calcium levels continue to fall, as predicted by several authors (e.g., Watmough and Aherne 2008).

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PART 2: CUMULATIVE EFFECTS ASSESSMENT — THEORETICAL UNDERPINNINGS AND BIG PROBLEMS

NRC Research Press

REVIEW

Cumulative effects assessment: theoretical underpinnings and big problems

F. Chris Jones

Abstract: Cumulative effects assessment (CEA) is a sub-discipline of environmental impact assessment that is concerned with appraising the collective effects of human activities and natural processes on the environment. Aspirations for CEA have been expressed by many authors since 1969, when the foundation of environmental appraisal was laid by the US National Environmental Policy Act. This paper's purposes are (*i*) to review aspirations for CEA, relative to current practice; and (*ii*) to fully explain and critique the logic that connects CEA's operational steps and underlying philosophies. A literature review supports the following statements: Some conceptualizations emphasize the delivery of information to support decision making as the key purpose of CEA; others deem collaboration, debate, and learning as most important. Consensus on CEA's operational steps has been reached, but each step requires practitioners to make analytical decisions (e.g., about the scope of issues to include or the time horizon to consider) and objective rules for how to approach those decisions are lacking. Numerical methods for assessing cumulative effects are largely available, meaning that CEA's biggest problems are not scientific. CEA cannot succeed without substantive public engagement, monitoring, and adaptive management. CEA is best undertaken regionally, rather than project-by-project. CEA and planning are complementary, and should be merged. In its most enlightened form, CEA is a useful tool for ensuring that human undertakings ultimately conform to Earth's finite biosphere, but current practice falls short of the ideal, and CEA's logical derivation is not entirely sound. As regards CEA's big problems, sustainability has not been defined clearly enough to make criteria for judging the significance of cumulative effects indisputable; legal, regulatory, and institutional frameworks are poorly aligned for CEA; and objective criteria for judging the adequacy of CEA's scope, scale, and thresholds do not exist, which makes the question of how to provide general guidance to practitioners intractable. Recommendations call for sustainability goals to be clearly expressed as measurable targets. Furthermore, precaution in human enterprise should be exercised by avoiding, minimizing, restoring, and offsetting negative cumulative effects. CEA can assist by quantifying and optimizing trade-offs.

Key words: environmental impact assessment, planning, environmental management, sustainability, conceptual model, precautionary principle.

Résumé : L'évaluation des effets cumulatifs (EEC) est une sous-discipline de l'évaluation de l'impact sur l'environnement qui porte sur l'évaluation des effets collectifs des activités humaines et des processus naturels sur l'environnement. De nombreux auteurs ont exprimé leurs aspirations quant à l'EEC depuis 1969, le moment où les fondements de l'évaluation environnementale ont été jetés par le « National Environmental Policy Act » aux États-Unis. Les buts de cette étude sont (*i*) d'examiner les aspirations quant à l'EEC, relativement aux pratiques courantes, et (*ii*) d'expliquer en détail et d'analyser la logique qui lie les étapes opérationnelles et les philosophies sous-jacentes de l'EEC. Un examen de la littérature appuie les énoncés suivants : certaines conceptualisations soulignent la diffusion de l'information qui soutient la prise de décision comme étant le but principal de l'EEC; d'autres jugent que la collaboration, le débat et l'apprentissage sont les plus importants. On a atteint un consensus sur les étapes opérationnelles de l'EEC, mais chaque étape requiert que des spécialistes prennent des décisions analytiques (ex., à propos de la portée des enjeux à inclure ou l'horizon temporel à prendre en compte) et il manque des règles objectives à savoir comment aborder ces décisions. Il y a des méthodes numériques généralement disponibles pour évaluer les effets cumulatifs, ce qui signifie que les plus importants problèmes de l'EEC ne sont pas scientifiques. L'EEC ne peut pas être une réussite sans un important engagement de la population, la surveillance et une gestion adaptative. Il est préférable que l'EEC se fasse régionalement, plutôt que projet par projet. L'EEC et la planification sont complémentaires et devraient être fusionnés. L'EEC, sous sa forme la plus éclairée, constitue un outil utile afin d'assurer que les entreprises humaines sont en conformité avec les limites de la biosphère terrestre, cependant la pratique actuelle ne répond pas à l'idéal, et la dérivation logique de l'EEC n'est pas tout à fait solide. En ce qui concerne les gros problèmes de l'EEC, on n'a pas assez clairement défini la durabilité pour que les critères d'évaluation de l'importance des effets cumulatifs soient évidents; les cadres juridique, réglementaire et institutionnel sont mal alignés pour l'EEC; et des objectifs d'évaluation du caractère adéquat de la portée, de l'ampleur et du seuil de l'EEC n'existent pas, ce qui rend la question de comment fournir des directives générales aux spécialistes difficile. Les recommandations préconisent que les objectifs de durabilité soient clairement exprimés en tant que cibles mesurables. De plus, on devrait user de la précaution en matière d'entreprise humaine en évitant, en minimisant, en restaurant et en compensant les effets cumulatifs négatifs. L'EEC peut aider en quantifiant et en optimisant les critères de choix. [Traduit par la Rédaction]

Mots-clés : évaluation de l'impact sur l'environnement, planification, gestion de l'environnement, durabilité, modèle théorique, principe de precaution.

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Introduction

Environmental Impact Assessment (EIA) is a process by which the outcomes of a project are identified, predicted, evaluated, and (where necessary) mitigated before major decisions and commitments are made (Cashmore 2004; Glasson et al. 2013): a way to explore "options for more-sustainable (i.e., less environmentally damaging) futures" (Duinker and Greig 2007). The EIA concept was introduced legislatively in the USA via the 1969 National Environmental Policy Act (Senate and House of Representatives of the USA 1969). By 1996, EIA had spread to more than 100 countries (Benson 2003), and by 2012, some form of EIA had been mandated in 191 of the world's 193 nations (Morgan 2012; Pope et al. 2013).

Senécal et al. (1999) listed four objectives of EIA: to ensure that the environment is considered in the decision-making process surrounding new developments; to minimize or offset adverse effects; to protect ecological processes and functions; and to promote sustainability and optimal resource use. Gibson (2012) extended these objectives, by recommending that EIA should apply to all potentially significant undertakings; give equal attention to "biophysical, social and economic considerations"; begin at the outset of the decision process; have clear requirements and a predictable process; focus "attention on the most significant undertakings"; expedite "public engagement and learning"; render the decision-making process more consistent, impartial, transparent, and accountable; mesh easily with extant regulatory, planning, or policy instruments; and provide an authoritative basis for enforcement, monitoring, and adaptation.

Over time, EIA practice has been continually scrutinized, and has diversified into many distinct and specialized sub-disciplines (Pope et al. 2013; Morrison-Saunders et al. 2014). This diversification has coincided with our growing acknowledgement of (and desire to better manage) the widespread influences that human activities have on the environment (Morgan 2012). This EIA scrutiny soon illustrated that planning, legal, policy, and regulatory processes were not considering the combined environmental effects of multiple human activities (Damman et al. 1995), and that the EIA process was failing to explicitly consider that "all effects are cumulative" (Duinker and Greig 2006; Gunn and Noble 2011). In particular, the common practice of assessing short-term effects, project by project, was deemed ineffective because it provided little scope for managing the effects of more than one project that may be occurring in an area (Brundtland 1987; Spaling et al. 2000; Connelly 2011). As a response to such criticism, cumulative effects assessment (CEA) was proposed as a way to evaluate the collective effects of actions on the environment (Seitz et al. 2011). CEA promises to improve EIA by considering how a given "receptor is affected by the totality of plans, projects and activities, rather than on the effects of a particular plan or project" (Therivel and Ross 2007). CEA is considered to be a sub-discipline of EIA, because it derives from EIA's principles, methods, and tools, and is broadly applicable across the diversity of EIA practice (Baxter et al. 2001; Canter and Ross 2010; Bond and Pope 2012; Morrison-Saunders et al. 2014).

Case studies of CEA practice, critiques (e.g., Duinker and Greig 2006), and "lessons learned" (Canter and Ross 2010) are abundant in the published literature[1]. Although some retrogression (i.e., return to a former, less effective state) of CEA practice has occurred (e.g., Gibson 2012), published articles have stimulated and documented a limited refinement of CEA over time (Canter and Ross 2010).

The evolution of EIA thinking has been driven by practice (i.e., the need to satisfy legislative and regulatory requirements), not by theory (Cashmore 2004; Retief 2010), which remains inconsistent and incomplete (Cashmore 2004; Judd et al. 2015). Despite its 45 year history, the discipline is still viewed as young and unproven (Bérubé 2007): a diverse and confusing assortment of methods (Cormier and Suter 2008; Seitz et al. 2011; Pope et al. 2013), uncoupled from theory (Lawrence 2000; Cashmore 2004). Illuminating the conceptual or theoretical basis for EIA and CEA, merging theory and practice (e.g., Pope et al. 2013), and establishing how performance is to be assessed (e.g., Retief 2010; Morgan 2012) are essential if we are to answer Senécal's (and co-authors' 1999) call for EIA practice that is "purposive, rigorous, practical, relevant, cost-effective, efficient, focused, adaptive, participative, interdisciplinary, credible, integrated, transparent, and systematic". There is some urgency to these tasks, given the presently difficult economic times, in which governments seek to stimulate economic activity by encouraging development and cutting green tape (e.g., Bond and Pope 2012; Gibson 2012; Middle et al. 2013; Morrison-Saunders et al. 2014); and "the severity of many cumulative effects – global warming, plummeting world fish stocks, decline in biodiversity – means that we have to get (CEA) right, and fast." (Therivel and Ross (2007).

This paper explores aspirations for CEA that have been expressed by many authors at different times throughout CEA's history. Its purposes are to synthesize a formal understanding of what CEA is *supposed to* achieve, to explain how its components are conceptually linked together (i.e., as a "theory" or conceptual model[2]), to expose inconsistencies (both in CEA's underlying logic, and as incongruences between current practice and aspiration), and to recommend some pathways for improvement.

Logical connections among CEA's components

The following is intended as a comprehensive description of CEA, and an illustration of logical connections among its components. The description proceeds as a deductive argument (e.g., Michalos 1970), based on a series of premises (denoted P1–P95) and conclusions (denoted C1–C14). Conclusions build on one another, and each one is positioned in the sequence to immediately follow its full set of supporting premises. The format will appear unorthodox to scientific readers, but it highlights incomplete or inconsistent logic, and invites debate over problems and solutions.

P1: Earth's biosphere and ecosystems provide the essentials for life, and enable our survival as a species (MEA 2005)[3].

P2: Earth's biosphere is finite, as are its ecosystems.

P3: As a foundation for human life and well-being, there is no substitute for Earth's biosphere.

P4: Human undertakings influence Earth's biosphere (and its ecosystems) in a variety of ways, and changes to the biosphere have occurred as a result of human activities (Vitousek et al. 1997; Wackernagel and Rees 1998; MEA 2005; Foley et al. 2005; Steffen et al. 2007; Rockström et al. 2009; Ellis 2011; Hegmann and Yarranton 2011).

P5: EIA and CEA began in the United States, as legislative requirements under the 1969 National Environmental Policy Act (Pope et al. 2013). Since 1969, CEA's concepts and methodologies have been developed, refined, and entrenched worldwide as laws, policies, and regulations (Benson 2003; Pope et al. 2013); however, CEA's evolution has not been linear, and setbacks have been documented (e.g., Gibson 2012).

[1]For example, a relatively restrictive keyword search of Thomson-Reuters' Web of Science, using the search term "(cumulative NEAR/1 effects) AND assessment" to query the "environmental science/ecology" and "sociology" research domains, returned 788 citations on 23 September 2015.

[2]As per Lawrence (2000), the terms *theory* and *model* are used to describe loosely affiliated conceptual themes.

[3]Social, cultural, and economic attributes of the societies we live in do enhance our standard of living, but this can only happen after the basics for life (e.g., water, air, food) are provided by the biosphere.

C1: Human enterprise (e.g., land-use, economic growth) must conform to the limitations of Earth's biosphere (Boulding 1966; Daly 1977; Wackernagel and Rees 1998; Schnaiberg and Gould 2000; Moldan et al. 2012) and CEA is an important tool for making this happen (e.g., Morgan 2012; Morrison-Saunders et al. 2014).

P6: Ecosystems have three domains: physical habitat, biota, and interactions between habitat and biota (Tansley 1935). Ecosystems are characterized by non-equilibrium and irreversible phenomena, and are open, complex, adaptive, hierarchical systems that are highly integrated by matter and energy flows (Jørgensen et al. 2007; Häyhä and Franzese 2014).

P7: An *effect* is a change to the structure or function of the ecosystem (i.e., a change to the habit, biota, and (or) their interactions).[4] Effects may be measured against the normal range of variability (Kilgour et al. 1998; Scherer 2011) associated with a specified spatial and temporal baseline (Glasson et al. 2013; Pavlickova and Vyskupova 2015); however, deviation from a statistically defined normal range does not necessarily signify ecological significance.

P8: Effects can arise because of natural processes or because of "stressors"[5] related to human activities.

P9: Land-uses or environmental issues (e.g., urban development, agriculture) often implicate multiple stressors (Chapman and Maher 2014); that is, they cause environmental change by a variety of mechanisms.

P10: Effects accumulate (i.e., *become cumulative*) as repeated insults of a single stressor — that is, when exposures to the stressor "take place so frequently in time or so densely in space that the effects of individual insults cannot be assimilated" (Damman et al. 1995; Pavlickova and Vyskupova 2015). They also accumulate via the interaction of multiple stressors (Smit and Spaling 1995), the nature of the interaction potentially varying over time or space.

P11: Cumulative effects can be enigmatic. They include individually minor but collectively significant changes that take place over a period of time (US-Ceq 1978; Cocklin et al. 1992; Therivel and Ross 2007); they include changes that may take place remotely from the location where the stressor is created; they include changes that may be undetectable by any specific monitoring method; they include secondary effects facilitated by, but not directly caused by, a given undertaking; and they include changes caused by interactions between multiple stressors and (or) multiple undertakings (Raiter et al. 2014). They may be direct, indirect, antagonistic, synergistic, linear, or nonlinear, and they may manifest following a complex chain of events (CEARC 1988; Scherer 2011).

P12: Legislative, policy, and regulatory interpretations of cumulative effects vary, but they are approximately defined as ecosystem changes that result from the incremental, accumulating, and interacting impacts of an action when added to other past, present, and reasonably foreseeable future actions (e.g., US-Ceq 1978; Spaling et al. 2000; Dubé 2003).

C2: Cumulative effects[6] are aggregated, collective, accruing, and (or) combined ecosystem changes that result from a combination of human activities and natural processes (Scherer 2011); however, they are commonly defined in a more restrictive sense in policies, laws, and regulations — for example, as effects of an undertaking and its interaction with other activities that occur at the same time and in the same area (e.g., EU 1985; Damman et al. 1995).

P13: CEA is the assessment of ecosystem changes that accumulate from multiple stressors, both natural and manmade (Dubé et al. 2013).

P14: CEA is a process by which the potential environmental effects of one or more alternate visions for a particular place, sector, or other entity are systematically assessed (Gunn and Noble 2009; Seitz et al. 2013).

P15: CEA seeks to understand changes in environmental condition from past to present, and to predict potential outcomes associated with proposed developments (Dubé et al. 2013) and other foreseeable developments that may be subsequently induced. As is the case with EIA, CEA's emphasis is on exploring the future (Rubin and Kaivo-Oja 1999; Duinker and Greig 2007).

C3: CEA is a sub-discipline of EIA; it is "the process of predicting the consequences of development, relative to an assessment of existing environmental quality" (Dubé 2003), or "the process of systematically analyzing cumulative environmental change" (Smit and Spaling 1995; Dubé 2003). As per Judd et al. (2015), it is a systematic way to evaluate the significance of effects from multiple activities and to inform resource managers about these effects. Local differences in methods exist, and are necessitated by location-specific laws, policies, regulations, and planning systems (e.g., Gunn and Noble 2009)[7].

P16: How CEA is understood to work, how much policy significance is attributed to it, and how its efficacy is to be measured is determined largely by its theoretical frame of reference (e.g., Bartlett and Kurian 1999).

P17: As a sub-discipline of EIA, CEA shares EIA's "basic principles", which Senécal et al. (1999) suggest require a process that is "purposive, rigorous, practical, relevant, cost-effective, efficient, focused, adaptive, participative, interdisciplinary[8], credible, integrated, transparent, and systematic".

P18: CEA is, at least partially, a token bureaucratic requirement for development consent, which serves as a symbolic gesture to pacify environmentalists or reinforce conservation values (e.g., Bartlett and Kurian 1999; Karkkainen 2002).

[4]One can argue that the definition of *effect* should be broadened, for example, to include changes to social or economic factors, because of the many ways in which human and biophysical factors interact, and because social, economic, and environmental factors are equally considered in sustainability assessments; however, one can also argue that, despite their political salience in the decision-making process, social and economic factors ultimately depend on the environment. The contemporary metaphor for sustainability is a series of nested bowls (e.g., Wikimedia Commons 2015), with the environment bowl containing society, and society containing the economy; it is no longer a three-legged stool (e.g., Dawe and Ryan 2003) or triple bottom line (e.g., Elkington 1997). Regardless, laws and historical practice show that CEA is chiefly concerned with ecological effects, and, as Sheate (2010) wrote, we should not lose sight of the environmental origins and purpose behind EIA (hence CEA) as an instrument for environmental advocacy.

[5]A stressor is "any physical, chemical, or biological entity that can induce and adverse response" (US EPA 2000).

[6]A hypothetical cumulative effects scenario: roads are built on unstable terrain during a time of above-average rainfall. Landslides and avulsions in a local stream become more frequent, thus erosion is exacerbated and the stream channel is destabilized. Gravel beds used by fish during spawning are clogged with sediment, and, after several years, populations of several fish species are outside of historical limits (adapted from Scherer 2011).

[7]One may argue, on one hand, that inconsistent methods allow for "flexibility and context-specific approaches" (Pope et al. 2013), which are presumably strengths of EIA and CEA practice; on the other hand, pluralism may be viewed as a weakness (Morrison-Saunders et al. 2014) that has resulted from poor theoretical grounding and lack of a unified conceptual model (Bartlett and Kurian 1999; Wärnbäck and Hilding-Rydevik 2009; Retief 2010; Morgan 2012).

[8]One defence of the call for interdisciplinarity was provided by Rubin and Kaivo-Oja (1999), who cited "fundamental problems related to the idea of dividing contemporary human behaviour into social behaviour (sociology), economic behaviour (economics), personal behaviour (psychology), and political behaviour (policy sciences)". These authors considered holistic analytical approaches more fruitful for "complex political, economic, social, technological and ecological systems".

P19: CEA's purposes and objectives require participants' values and ethics to be considered explicitly (e.g., Cashmore 2004).

P20: Being a sub-discipline of EIA, CEA can be viewed, at least partially, as an applied science, because it both generates knowledge and employs the scientific method. For example, hypothesis tests are used to evaluate the significance of effects. Knowledge resulting from such tests enhances our understanding of stressor–effect relationships, and ultimately reduces uncertainty in future appraisals (e.g., Cashmore 2004).

P21: Appraisal itself derives from the belief that the decision making process can be made more rational if options are carefully analyzed. CEA is chiefly a method of "generating, organizing, and communicating information" (e.g., Bartlett and Kurian 1999; Dubé 2003; Morgan 2012), which is then delivered to decision makers (Senécal et al. 1999; Hegmann and Yarranton 2011). Such information is intended to improve public decisions (Karkkainen 2002; Sheate 2010) about proposed actions (Rubin and Kaivo-Oja 1999) by solidifying where the public interest lies, and by permitting one to estimate the probability that a given action will align with this interest (Wright 1986; Etzioni 1988; Hegmann and Yarranton 2011). "Decision makers are assumed to be acting in an objective and value-free manner, and their decisions are assumed to arise logically from a systematic and largely technical assessment of the evidence" (Benson 2003). Having access to information about the choices being considered, allows decision makers to overcome their personal biases, and more objectively consider issues germane to the decision (Adelle and Weiland 2012). Knowledge is expected to translate directly into decision outcomes, and a separation of powers is deemed to exist between neutral, authoritative experts and the decision makers they advise (Owens et al. 2004; Pielke 2007). CEA is a logical, consistent, and systematic process that uses reason, science, and technical knowledge as a basis for, and justification of, decision making in a society that has an articulated singular ("unitary") interest or goal (e.g., Lawrence 2000). As such, it is strongly rooted in positivism[9] and rational decision theory[10] (e.g., Benson 2003).

P22: As a basis for CEA, the technical–rational model of appraisal (which emphasizes the delivery of information to facilitate evidence-based decision making) is inadequate theoretically (because it fails to account for observed relationships between assessments and decisions[11]; Pope et al. 2013; Russell-Smith et al. 2015), politically (because, in practice, decisions are influenced by ethical and political judgments; Bond 2003), and practically (because exposing its logical fallacies jeopardizes the legitimacy of both CEA appraisals and the courses of action brought about by particular decisions; Owens et al. 2004).

P23: Rationality is limited in human decision making (Doyle 1999; Nooteboom 2000; Cashmore 2004; Partidário and Arts 2005; De Martino et al. 2006); the decision process is open to behavioural, political, and institutional influence (Bartlett and Kurian 1999); and the complexity and information demands of CEA problems habitually exceed our capacity for strict rationality[12] (Doyle 1999; Sheate 2010). Elements of the decision-making process that tend to reduce rationality include participants' ideological or political biases, poor governance of the process itself, corruption[13], and misinformation. Furthermore, human decisions are influenced by context (i.e., the manner in which options are presented; Gunn and Noble 2009) because of how decision-making tasks are allocated to different loci in the brain — logical and rational thought being concentrated in the prefrontal cortex, and intuitive or emotional thought being concentrated in the amygdala (De Martino et al. 2006).

P24: "Post-positivist philosophies" (Adelle and Weiland 2012) stress the relativity of knowledge and the political context in which decisions are made. Improvements to the decision-making process are made not primarily by better informing it, but by maximizing opportunities for debate and learning, by anticipating biases, and by managing the way power and influence are distributed among decision makers (Adelle and Weiland 2012).

P25: Although it is often viewed as a largely technocratic process, CEA still provides a forum for dialogue and learning (Owens et al. 2004; Owens and Cowell 2011), which can extend as spin-offs well beyond individual decisions to "influence the values and behaviours of organisations and society at large" (Pope et al. 2013).

P26: Deliberative rationality views the decision-making process as an opportunity to debate difficult choices, demonstrate that opposing views have been taken seriously, and learn from the outcomes. A distinction can be made between the instrumental type of learning that characterizes the technical rational decision-making model and the deliberative conceptual learning that more generally enlightens decision makers by introducing new ideas and perspectives (Adelle and Weiland 2012).

P27: CEA is, at least partially, a process that facilitates deliberation as a way of advancing environmental (also social and economic) justice. It represents a mechanism by which diverse competing interests and values are ratified by negotiation in a complex political and antagonistic arena that is often highly constrained by legislation, policy, and entrenched practices (e.g., Lawrence 2000).

P28: CEA is, at least partially, rooted in socio-ecological idealism (e.g., Lawrence 2000). By requiring bureaucratic organizations to employ environmentally minded staff (who generate or review CEA appraisals), CEA constitutes a way to import environmental virtue into, and ultimately make the culture, institutional values, and routine operations of these organizations more environmentally centred (e.g., Bartlett and Kurian 1999; Lawrence 2000).

P29: CEA is, at least partially, a strategy for advancing ecological economics (e.g., Costanza 1992) because it integrates environmental goals into economics, and provides incentives for innovations that maximize environmental and economic returns (Bartlett and Kurian 1999); however, in practice, this strategy is often thwarted because mitigation is emphasized to the point that alternatives are not properly explored.

P30: CEA is, at least partially, a way of enriching democracy because it establishes a transparent process by which consensus is sought via citizen involvement (e.g., Bartlett and Kurian 1999; Karkkainen 2002; O'Faircheallaigh 2010), communication, and collective visioning (Lawrence 2000).

C4: CEA has multiple objectives (e.g., Culhane 1990; Schultz 2012), which are loosely arranged around the themes of formalizing environmental values, establishing an ecologically viable socio-economy, and improving knowledge and governance (Pope et al. 2013). As a form of environmental appraisal, CEA is not just a tool for informing and influencing decision makers; it also changes the views and attitudes of its participants, and its influ-

[9]Positivism establishes logical verification, or mathematical proof, as a criterion for rational thought (Macionis and Gerber 2011).

[10]Rational decision making is defined as "choosing among alternatives in a way that … accords with the preferences and beliefs of an individual decision maker or those of a group making a joint decision" (Doyle 1999). Common criteria for rationality demand the decision-making process to employ logically sound and deductively complete reasoning (Doyle 1999).

[11]For example, experts cannot be neutral because of the adversarial way that the CEA plays out among proponents and opponents.

[12]For these reasons, many authors substitute the term *bounded rationality* (e.g., Leknes 2001; Sheate 2010).

[13]Dominant interests tend to be insulated from the eventualities of bad decisions. In general, the greater the power, the lesser the rationality (Owens et al. 2004).

ence can propagate outward through participants' social networks to influence entire institutions (Bond and Pope 2012).

P31: Ecosystems have a complex array of attributes that, taken together, are intractable: they are too complex to be fully considered within the scope of CEA (Damman et al. 1995).

P32: CEA evaluates effects on specific "valued ecosystem components" (VECs) or significant ecological features[14] (e.g., Beanlands and Duinker 1983; Fisheries and Oceans Canada 2004, 2011, 2014), which are considered to be[15] important ecosystem attributes (Bérubé 2007; Gunn and Noble 2009; Canter and Ross 2010; Canter and Atkinson 2011; Dubé et al. 2013). VECs can be "physical things (e.g., a fish population), ecological processes (e.g., C sequestration), and even abstract concepts, such as ecological integrity or water quality (Damman et al. 1995; Dubé et al. 2013).

P33: VECs focus appraisal on a small number of important, valuable, or significant ecosystem features, to provide a relevant and practically scoped reference point (Canter and Atkinson 2011).

P34: Consensus on which VECs to include in CEA may be achieved collaboratively according to social, cultural, economic, scientific, or aesthetic concerns that arise from a professional scoping exercise, or from more inclusive procedures, such as public hearings, questionnaires, interviews, workshops, or media reports (Beanlands and Duinker 1983; Canter and Ross 2010).

P35: Indicators (or indices; e.g., Janetos and Kenney 2015) can be used as measurable surrogates for VEC condition or status (Gunn and Noble 2009), and they can be used to provide a numerical framework for CEA analyses (Canter and Atkinson 2011).

P36: VEC selection trades-off parsimony, which makes CEA tractable, with comprehensiveness, which makes it realistic (Duinker and Greig 2007).

C5: VECs (or their index-based surrogates) must be used to provide a tractable scope for CEA, and to ensure that attention is paid to "the most important environmental features and processes" (Damman et al. 1995; Canter and Ross 2010).

General guidance about indicators applies to CEA: indicators should be conceptually simple enough that their meaning can be conveyed to diverse audiences; they should be predictive; they should be sensitive and sufficiently accurate and precise to discriminate stressor-specific effects; and their sampling and data requirements should not be overly onerous and costly (e.g., Cairns et al. 1993; Niemi and McDonald 2004; Bonada et al. 2006). Furthermore …

P37: The value of a cumulative-effects indicator may be judged on the soundness of its surrogacy for one or more VECs (i.e., the degree to which it reflects the VEC's condition).

P38: Given equal VEC surrogacy, the relative merits of candidate cumulative effects indicators may be assessed by how well they represent form and function of each ecosystem domain (i.e., habitat, biota, and interactions), by their sensitivity to changing conditions in each of those domains, and by their ability to diagnose causes of change.

P39: Stressor-based indicators evaluate an ecosystem's exposure to stress, but leave unanswered questions about the ecological relevance of that stress (Roux et al. 1999).

P40: Effect-based biological indicators measure ecosystem structure or function directly, because biota "integrate a cumulative response to environmental stress" (Munkittrick et al. 2000; Dubé 2003); but they leave unanswered questions about which stressor, or stressors, are being responded to (Roux et al. 1999; Gunn and Noble 2009).

P41: Stressor-based indicators measure habitat condition, and therefore integrate over only a single ecosystem domain; biological indicators integrate across all three ecosystem domains (habitat, biota, and their interrelationships), and they integrate across multiple stressors and their interactions.

C6: In principle, biological (effect-based) indicators are superior to stressor-based indicators for CEA. In practice, if ecological effects are detected, the stressors causing the effects must typically be identified (US EPA 2000; Dubé 2003); therefore both indicator types may be required to diagnose causes of, and model, ecosystem changes (Smit and Spaling 1995; Jones et al. 2002; Dubé 2003; Dubé et al. 2006).

P42: As a minimum, CEA must determine who specified VECs matter to, why they matter, how much they matter, how VECs will be effected by a specified set of activities, whether effects will be important given the past and present condition of the VEC, and what, if anything, could make up for any loss or damage to the VEC (Therivel and Ross 2007).

P43: In its most enlightened form, CEA constitutes a series of methods that assess the condition of the environment, describe causal pathways that link stressors and cumulative effects, predict risks and benefits associated with alternative management futures, and evaluate whether intended outcomes of the CEA decision were realized (Cormier and Suter 2008). This concept of CEA is rarely approached in practice.

P44: CEA's operational practice includes the following seven steps (adapted from Damman et al. 1995; Ross 1998; Canter and Ross 2010; Connelly 2011; Seitz et al. 2011)[16]:

1. Select VECs.
2. Define the physical and temporal boundaries of the appraisal.
3. Identify activities (past, present, future) that may affect VECs.
4. Characterize baseline (this step includes the selection of indicators, description of environmental condition, and determination of stakeholders that should be involved in the process).
5. Analyze and predict cumulative effects on VECs.
6. Determine significance of effects (given planned mitigation).
7. Monitor outcomes and CEA performance.

P45: Decisions are made at each step in the CEA process, not just at the end (Benson 2003). CEA itself is a decision-making process.

P46: Although various decisions must be made as a CEA is undertaken, no objective, legitimate, and standardized criteria exist by which the soundness of these decisions can be evaluated. Illustrative examples include the lack of criteria for setting CEA's geographic and temporal scope (e.g., Schultz 2012), for establishing the breadth of issues to consider (e.g., which VECs, and which stressor–effect pathways to include (e.g., Duinker and Greig 2007)), for defining what constitutes an acceptable baseline against which present condition and future cumulative effects are gauged (e.g., McCold and Saulsbury 1996; Schultz 2012), for judging the significance of effects (Karkkainen 2002; Bérubé 2007; Scherer 2011), for determining who is responsible for carrying-out and paying for the appraisal, and for determining what function the appraisal is to have in the decision-making process.

[14]Given CEA's purpose as a sustainability instrument, one may argue that the term *valued ecosystem component* should be broadened as a measure of societal progress. In this scenario, indices could represent social, cultural, and economic dimensions of the "environment", thus allowing CEA to evaluate trade-offs among societal, not just ecological domains (Damman et al. 1995); however, some authors (e.g., Adelle and Weiland 2012) caution that more integrative CEA may backfire as environmental and social concerns are overwhelmed by more politically salient economic interests. For this reason, and to agree with the definition of *effect*, VEC is used here in its most restrictive sense.

[15]Typically this consideration is societally broad, universal, or reflects consensus among CEA participants (Canter and Ross 2010).

[16]Flexibility to adapt to cultural, political, institutional, and legal contexts is imperative (Bond and Pope 2012), and results in regional variation in these steps.

C7: Consensus on CEAs operational steps has been reached; however, objective criteria for guiding decisions that must be made at each step are lacking, and a degree of pluralism is permitted so each CEA is tailored to its context (e.g., Bond et al. 2012). CEA practice habitually falls short of the aspirational ideal.

P47: CEA is complex methodologically, scientifically, and institutionally (Canter and Ross 2010). It deals with the future ("a complex mixture of consequences" (Rubin and Kaivo-Oja 1999)) that unfolds because of present and past decisions, but also because of innumerable other determinants that cannot be wholly accounted for (e.g., natural environmental processes, demographics, economics, scientific and technological advancements, the evolving zeitgeist, and random events; Gunn and Noble 2011).

P48: Complex systems (e.g., ecosystems) are inherently unpredictable, which means that uncertainty is unavoidable in CEA (Hegmann and Yarranton 2011; Chapman and Maher 2014). As per Karkkainen (2002), "our hopes of accurately predicting all the impacts associated with a development action are virtually nil" (Karkkainen 2002). The larger the area a CEA considers, and the longer its time horizon, the less certain conclusions about cumulative effects will be (Hegmann and Yarranton 2011).

P49: Given CEAs limited predictive abilities, activities under investigation are best viewed as experiments[17] by which alternatives are explored (Duinker and Greig 2007; Canter et al. 2010; Schultz 2012).

P50: Monitoring indicates ecosystem form and function, allows the condition and variability of VECs to be quantified (Ball et al. 2013), signals when status is changing (e.g., Cairns et al. 1993; Jones et al. 2002), is used to test and refine predictive models (Davenport et al. 2008), and allows the performance of CEA and the decision-making process to be reviewed (e.g., Therivel and Ross 2007; Davenport et al. 2008; Schultz 2012), thereby making the CEA process more transparent and accountable (Karkkainen 2002).

P51: Monitoring facilitates adaptive management (Walters 1986) and "systemic learning" (Karkkainen 2002). It allows the CEA decision (about whether and how to proceed with one or more undertakings) to be retooled as a series of decision and adjustment points that are arranged along a timeline into the future (Karkkainen 2002).

P52: Monitoring can reduce the costs of environmental appraisals because it allows proponents to trade-off some of the exhaustive front-loaded information costs for potentially lesser incremental costs that are spread over the life of the project (Karkkainen 2002).

C8: Monitoring and adaptive management are essential to CEA (Karkkainen 2002; Davenport et al. 2008; Seitz et al. 2011; Schultz 2012; Ball et al. 2013).

P53: It is more likely that decisions will be rational if the decision-making process is formal and transparent, and if there is an opportunity for external review and input, as afforded by public participation[18]. These attributes modify how decision makers approach their job, by tempering the influence of their own personal biases and values, and promoting thoroughness and disciplined rationality (Karkkainen 2002; Gibson 2012).

P54: Public involvement alleviates some of CEA's operational challenges by providing access to local knowledge, broadening the consideration of social values and environmental issues, and reducing the likelihood of litigation (Baxter et al. 2001; O'Faircheallaigh 2010). It also helps to build consensus around sustainability goals by fostering cooperation and confronting "NIMBYism"[19] (Benson 2003).

P55: Public involvement adjusts the power relationships among participants in the decision-making process, making it more democratic, transparent, and accountable, and more responsive to societal values (Benson 2003; O'Faircheallaigh 2010; Sheate 2010). This, in turn, legitimizes the decision-making process, enhances public acceptance of its rulings, and stabilizes the participating institutions (Karkkainen 2002).[20]

P56: Public involvement lends CEA capacity to create more democratic and deliberative forms of decision making (Bartlett and Kurian 1999), which ultimately advances human rights and social justice (Benson 2003).

P57: Public involvement increases opportunities for reciprocal learning among CEA participants (Bond and Pope 2012); it provides a mechanism by which the community comes to understand CEA's goal and limits, and the questions it can answer (Gallagher et al. 2015); and it contributes organizational and institutional capacity by which CEA can be improved iteratively (Sheate 2010).

P58: The earlier in the CEA process that public engagement occurs, and the more extensive that engagement is, the more substantive its outcomes are (e.g., Canter and Ross 2010; Shirk et al. 2012; Gallagher et al. 2015).

C9: Public involvement in CEA is essential (e.g., O'Faircheallaigh 2010; Gallagher et al. 2015), regardless of how one considers CEA's value to partition among information-delivery (technical–rational) and deliberative (post-positivist) outcomes (Morgan 2012).

P59: Performance evaluation concerns effectiveness or efficacy; it provides a measure of how well an action or method meets its own objectives (Sheate 2010).

P60: Enabling legislation and operational guidance evolve, at least partly, based on assessments of efficacy (e.g., Bond and Pope 2012).

P61: Government priorities of stimulating economic growth and creating employment cause CEA to be subjected to intense scrutiny. Particularly during times of economic (thus political) upheaval, governments are prone to water-down environmental appraisal, and this is facilitated where performance has not been assessed or results have been seen as unfavourable (e.g., Gibson 2012; Morgan 2012; Middle et al. 2013; Morrison-Saunders et al. 2014).

P62: It is notoriously difficult to design a defensible study by which CEA's effectiveness can be judged, and our ability to generalize beyond a given CEA's political, cultural, legal, and institutional context is limited (Morgan 2012). It is tricky to specify what influence CEA ought to have on a given decision-making process (Cashmore 2004), and separating out the consequences of CEA from the many other factors that influence decision making is problematic (Sheate 2010). Furthermore, it is impossible to judge what would have happened in the absence of a given CEA appraisal, because controls are generally not available and decisions are not replicated. It is also impossible to determine whether any given decision (e.g., the granting of development consent) is correct because no objective criterion for correctness exists (Cashmore 2004), and because the totality of effects will not be realized until some future time.

P63: Performance evaluation is constrained by the lack of clarity with which CEA's nature and purpose is understood (e.g., Morgan 2012). CEA's pluralistic theoretical underpinnings mean that its performance may be assessed relative to ecological, social, political, economic, and learning outcomes (Lawrence 2000; Bond and Pope 2012). As Adelle and Weiland (2012) illustrated, a researcher with a positivist bent (presumably interested in the

[17]Preferably low risk and reversible experiments.

[18]*Public participation* is defined sensu O'Faircheallaigh (2010) as any interaction between government and corporate CEA parties and the public.

[19]That is, the "not-in-my-backyard" mindset.

[20]According to Birkeland (1999) public participation can be dangerous when emphasized to the extreme, because it can "mask the need for deeper institutional reforms".

soundness of assessment methods and the quality of predictions) may cite misuse of, or inadequate, monitoring or modeling tools as a CEA weakness. A researcher from the post-positivist tradition (perhaps interested in learning and how evidence is used in the decision-making process) may cast aside analytical methods as irrelevant, but single out inadequate deliberation on available options as a crucial inadequacy. Similarly, a researcher assessing CEA's operational steps may view any gap between goals and results as a critical weakness; whereas a researcher interested in the politics of appraisal may consider such gaps simply as evidence that CEA's underlying motivation is mainly symbolic.

P64: Pluralistic frameworks for evaluating performance have been proposed by several authors (e.g., Bond et al. 2012; Middle et al. 2013); however, translating these narrative frameworks into evaluative criteria is usually problematic because of conceptual difficulties associated with ill-defined terms, and because indicators are either not apparent, not routinely monitored, or difficult to quantify. For example, Bond et al. (2012) suggested evaluating the appraisal process based on its "procedural effectiveness" (i.e., by considering the degree to which implementation agrees with professional and institutional standards of practice)[21], "substantive" and "normative" effectiveness (i.e., by measuring to what extent the outcome of the appraisal aligns with the reasons for doing the appraisal)[22], "transactive effectiveness" (i.e., by assessing the degree to which time and money investments in the appraisal are justified by its outcomes)[23], engagement (i.e., by determining to what extent stakeholders are accommodated into, and satisfied by, the appraisal process)[24], and learning (i.e., by quantifying the degree to which the appraisal facilitates "instrumental and conceptual" learning[25]). Middle et al. (2013) suggested evaluating CEA performance based on the comprehensiveness of the CEA requirement (i.e., the proportion of proposed activities subject to CEA appraisal)[26], scope (i.e., how adequately VECs represent important ecological attributes)[27], "objectivity, openness, and transparency" (of both the CEA process and the decision-making process it is a part of)[28], quantity and quality of public participation, timeliness (i.e., how early in the decision-making process CEA occurs, earlier being better because decision makers have flexibility to more fully consider alternatives the earlier in the process the appraisal is situated (Gunn and Noble 2009)), the degree to which CEA outcomes guide the evolution of environmental policy, capacity for adaptation (i.e., how flexible the CEA process is, and how much learning and innovation results from it)[29], efficiency (i.e., money and time costs)[30], rationality (i.e., how predictable the decision outcome is, given the nature of information that surfaces during the CEA appraisal)[31], and follow-up (i.e., the degree to which the CEA outcome is legally binding, and how closely resultant cumulative effects match those predicted).

P65: CEA's ultimate goal is *sustainability or environmental protection*; holistic, quantitative, and objective surrogates for these embattled terms are the Holy Grail of CEA performance evaluation.

P66: Evaluative techniques from disciplines, such as results-based management and risk assessment, can be adapted for CEA. For example, logic models (borrowed from results-based management) allow one to create a simplified description of a program, initiative, or intervention that illustrates the logical relationships that connect invested resources with activities and outcomes (Taylor-Powell et al. 2003); bow-tie analysis, a risk-assessment technique (IEC/ISO 2009), can be adapted for CEA by elucidating what measures are in place to prevent cumulative effects associated with a given activity or policy, and by evaluating how effective those measures are, and how fully they are implemented (e.g., ICES 2014).

C10: Multifaceted evaluation of CEA's efficacy and efficiency is required if CEA is to be endorsed and enlightened in the future, but methods for evaluating CEA are embryonic.

P67: Alternative patterns of human activity can be examined in CEA, given a process that allows cumulative effects associated with these patterns to be assigned different probabilities (Rubin and Kaivo-Oja 1999).

P68: CEA requires the effects of multiple stressors to be understood, both singly and in combination (Ball et al. 2013; Judd et al. 2015).

P69: Processes within ecosystems can magnify minor localized effects, requiring a variety of interactions to be considered, for example, stressor interactions with time, with space, and with other stressors.

P70: Quantitative CEA requires large complex datasets (describing multiple actions, VECs and their surrogate indicators, and predictors that quantify stressor exposure), and these datasets must be analyzed from various spatial and temporal perspectives (Atkinson and Canter 2011).

P71: A variety of stressor-based and effect-based indicators have been described, such as habitat attributes (e.g., May et al. 1997 (watershed land cover), Jennings et al. 1999 (lake shoreline substrates), Shifley et al. 2008 (forest attributes)), chemical attributes (e.g., Smith and Owens 2014), metabolomic markers (e.g., Viant 2009; Van Aggelen et al. 2010), and indices of the taxonomic or functional structure of communities (e.g., Norris and Georges 1993; Dolédec et al. 2006; Borja et al. 2011). These indicators are available for adoption by CEA practitioners wishing to associate stressors with effects on VECs (Canter and Atkinson 2011)[32].

P72: Remote sensing and computerized systems for geospatial analysis provide CEA practitioners the ability to create, store, manipulate, analyze, and visually display large amounts of geographically referenced data (Atkinson and Canter 2011); these technologies can be used to quantify stressors, and to relate stressors and effects, in a "spatially explicit manner" (Davenport et al. 2008; Atkinson and Canter 2011; Ball et al. 2013; Seitz et al. 2013).

P73: Scientific methods for investigating stressor effects are well developed, and they range from experimentation to a variety of mechanistic and statistical modeling approaches.

P74: Modeling is used in CEA to associate indicators of ecosystem form or function with natural factors or stressors (Seitz et al.

[21]But how is agreement defined?

[22]But authors have not reached consensus about the importance of CEA's various motivations.

[23]But how can this justification be critiqued objectively?

[24]But what is the optimal amount of engagement?

[25]That is, instrumental and conceptual learning are not discrete, and both are difficult to quantify.

[26]But how can the optimal degree of comprehensiveness be established objectively?

[27]Importance is subjective.

[28]But what degree of openness and transparency is necessary? Can there be too much?

[29]But attributing changes to any particular CEA will be problematic.

[30]But how much *should* CEA cost?

[31]But how will predictability be assessed? Is predictability really a measure of efficacy?

[32]Indicators of natural capital and ecosystem services (which may be called for, given alternative concepts of VECs) are also available (e.g., Häyhä and Franzese 2014).

2013; Ball et al. 2013), to explore futures associated with different scenarios of human activities (e.g., Duinker and Greig 2007; Russell-Smith et al. 2015), and to guide the design of monitoring schemes (Davenport et al. 2008). Modeling can be as simple as hypothesis-of-effect diagrams or interactive matrices (e.g., Smit and Spaling 1995), which illustrate suspected causal pathways that link stressors and effects, or they can be as complicated as "meta-model" frameworks (e.g., Smit and Spaling 1995), which combine simulation models to investigate stressor–effect relationships at a variety of spatial and temporal scales, or across a variety of ecological and societal dimensions. Statistical models for quantifying cumulative effects include regression (which associates a single response with one or more predictors; e.g., Steel and Torrie 1980; Sorensen et al. 2008), redundancy analysis (which associates multiple response measures with multiple predictors; e.g., Legendre and Legendre 2012), and variance components analysis (which partitions the variation in one or more response variables that is associated with selected predictors and their interactions; e.g., Peres-Neto et al. 2006). Interactive (e.g., additive or multiplicative) terms can be included in such models to account for combined stressor effects.

P75: Modeling involves trade-offs between scale and precision, and between conceptual complexity and computational complexity (Shifley et al. 2008).

P76: Retrospective analyses that link VEC trends with past activities provide a foundation for prospective *futuring* (i.e., use of methods that predict future conditions; Therivel and Ross 2007; Canter and Atkinson 2011).

P77: Futuring attempts to describe a plausible range of outcomes, given the uncertainty of future actions (Duinker and Greig 2007; Therivel and Ross 2007; Canter and Ross 2010). Its point is to "explore risks and sensitivities", which can "profitably be done without pinpointing the exact development future that will unfold into reality" (Duinker and Greig 2007). Futuring methods include scanning, trend analysis, extrapolation, and projection (which characterize historic trajectories and project these trends into the future, based on the assumption that causal mechanisms will persist)[33], scenario building (which is the process of creating alternative depictions of the future that may play out as a consequence of different decisions being made), polling or brainstorming (which includes the popular Delphi method (e.g., Lang 1998, in which questionnaires are used to structure expert discussions and arrive at consensus about future ranges of key variables), simulation modeling (which creates mathematical extrapolations based on mechanistic relationships between stressors and effects), gaming, historical analysis, and visioning (Cornish 2004; Duinker and Greig 2007).

P78: Methods of human and ecological risk assessment, which examine risks associated with "aggregate exposures to multiple agents or stressors" (e.g., US EPA Risk Assessment Forum Cumulative Risk Technical Panel 2003; Gallagher et al. 2015) can be adapted for CEA.

P79: Weight-of-evidence methods (e.g., Burton et al. 2002; Chapman and Maher 2014) for integrating multiple (possibly conflicting) lines of evidence that associate multiple stressors and their cumulative effects are available, and they range from qualitative use of causal criteria and logic models to integrated quantitative statistical analyses (Linkov et al. 2009).

P80: CEA should be "rigorous" (i.e., it should use defensible scientific methods that are appropriate to the questions posed; Senécal et al. 1999); however, practical limitations to scientific methods become apparent in CEA (Seitz et al. 2011). For example, appropriately replicated factorial experiments designed to test multiple-stressor effects (and their interactions) on biological endpoints require very large experimental apparatuses and associated infrastructure, and become difficult to implement because of personnel, equipment, and space constraints (Fisher 1960; Thorngate 1976; Steel and Torrie 1980). Similarly, complex statistical models, including interactive terms (which describe shared variance among predictors) and cumulative terms (which represent additive or multiplicative combinations of predictors) require large datasets that may resist compilation.

P81: Our knowledge about stressors and their effects on most ecosystems remains insufficient as a starting point for CEA (Seitz et al. 2011; Venier et al. 2014).

P82: A rough description of cumulative effects is generally all that is needed to identify appropriate management options (Therivel and Ross 2007).

C11: Basic knowledge about stressors and effects is rudimentary. Nonetheless, "the ecological literature is rife with … reports outlining various predictive models for forecasting effects of particular developments on specific … VECs" (Duinker and Greig 2007). Numerical methods for quantifying, assessing, modeling, and predicting cumulative effects are available. On one hand, our capacity for prediction is often small, and there are practical limits to the number of factors that can be tested experimentally or modelled; on the other hand, crude approximation of cumulative effects is sufficient in most CEA contexts (Therivel and Ross 2007).

P83: The subject of CEA may be an existing, proposed, or hypothetical project or undertaking (i.e., project-based CEA; e.g., Gunn and Noble 2009; Seitz et al. 2011) or may be an idea, policy, regulation, law, or similar strategic instrument (i.e., strategic CEA; Harriman and Noble 2008; Noble 2010; Connelly 2011; Hegmann and Yarranton 2011).

P84: CEA may be undertaken with a restricted spatial and temporal frame of reference, as is common with project-based CEA, or it can be undertaken at a scale that is not bounded by the parameters of a specific undertaking (e.g., at the regional[34] or watershed scale (Spaling et al. 2000; Ball et al. 2013; Dubé et al. 2013; Smith and Owens 2014)[35], and with a longer-term perspective, as is common with strategic CEA (Gunn and Noble 2009))[36].

P85: Current practice in project-based CEA reflects a mindset of ecological abundance, with environmental assessment at the scale of individual projects, and from the perspective of regula-

[33]To address complexity and uncertainty, it is wise to combine backcasting (which specifies desirable future scenarios, and explores various means of attaining them) with forecasting (which specifies undesirable future scenarios, and explores various means of avoiding them).

[34]*Regional scale* is a rather ill-defined term that describes an area that is much larger than the footprint of a specific project, typically an area that is ecologically or jurisdictionally relevant (e.g., a watershed, ecoregion, or municipality).

[35]Given CEA's eco-centricity, several authors (e.g., Gunn and Noble 2009) assert that regional units should be defined ecologically (e.g., as watersheds), not politically or administratively (e.g., as municipalities).

[36]Some authors classify sub-disciplines of EIA (e.g., strategic environmental assessment, regional strategic environmental assessment, CEA) according to how "vertically integrated" or "tiered" they are (e.g., Pope et al. 2013). A project-based appraisal of development effects for the purpose of regulatory approval would be considered lower-tier, with restricted geographic scope, and little vertical integration; whereas, an appraisal of the cumulative effects of a regional development policy would be considered upper tier, because of its broad geographic scope and its integration with decision-making processes in planning and policy realms. The term *strategic* has also been used to describe the degree to which alternative options are considered in CEA, and how early in the decision process this consideration occurs (more comprehensive considerations of options, occurring earlier in the process, being considered more strategic; for example, Gunn and Noble 2009).

Table 1. Hallmarks of enlightened CEA, relative to conventional impact assessment (adapted from Gunn and Noble 2009, and Noble 2010).

	Conventional impact assessment	Enlightened CEA
Questions[a]	What are the likely additive or incremental effects of stressors associated with the alternative project(s)? Are these effects likely to be significant? Can effects be mitigated by altering project designs?	What are the potential cumulative effects associated with alternate future scenarios, relative to sustainability goals?
Planning perspective[a,b,c]	Abundance; project planning and prioritization	Limits (finite biosphere); societal futures, especially concerning regional development and sustainability
Management perspective[a]	Mitigating development impacts	Ensuring preferred societal outcomes; sustainability
Trigger[a,b]	Effects of a project, or a collection of projects, on the environment immediately surrounding the projects	Contemplated changes or undertakings (especially where outcomes have potential to be at odds with sustainability)
Proponent[a,b,d]	Developer, collective of developers, or sector advocate	Lead organization (often a regional planning authority) or cross-sector collaboration with sufficient mandate, finances, and capacity
Scope of stressors[a,b]	Project stressors	All stressors
Temporal Bounds[a,b,e]	Life cycles of past, present, and future projects	Long-term societal futures
Spatial bounds[a,b,f]	Project(s) vicinity	Ecologically based, often watersheds, ecoregions, planning authority jurisdictions, or areas that encompass a series of related projects (existing and (or) planned)
Criteria for evaluating effects[a,b]	Ecological significance	Ecological significance; significance relative to sustainability goals
Alternatives considered[a,b,e]	Whether to proceed; whether modifications are required before proceeding	Alternative future scenarios
Rationale for monitoring[a,b,g,h]	Project-based regulatory compliance; efficiency-based performance indicators common	Behooved by complexity and uncertainty (permits adaptation); promotes reciprocal learning between CEA and planning, particularly with respect to thresholds for judging the acceptability of effects and carrying capacity; performance assessed largely on the basis of effectiveness

[a]Gunn and Noble (2009).
[b]Noble (2010).
[c]Wackernagel and Rees (1998).
[d]Sheelanere et al. (2013).
[e]Duinker and Greig (2007).
[f]Spaling et al. (2000).
[g]Walters (1986).
[h]Owens et al. (2004).

tory compliance; its scientific integrity is generally limited to the extent necessary for project approval (Seitz et al. 2011); and it promises mitigation of effects, though performance is assessed predominantly with measures of efficiency, not efficacy (Gunn and Noble 2009; Noble 2010; Table 1). As a result, project-based CEA is unable to cure incremental effects like biodiversity loss (Connelly 2011) and other products of the present dogma in which continual development is pursued in a finite biosphere (Schnaiberg and Gould 2000). Furthermore, "…CEA operates in three silos. Project proponents operate in the silo of stressor-based approaches to identify and mitigate project stressors, with governments as gatekeepers. The scientific/academic community operate in the silo of effects-based science to understand ecosystem functioning and environmental effects in response to landscape disturbances. Land-use planners and managers are focused on broader environmental planning and social matters, while incremental impacts at the project level continue to accumulate" (Noble 2010).

P86: "Cumulative effects assessment ultimately is an attempt to describe environmental change driven by forces far larger than any one project" (Hegmann and Yarranton 2011), and to compare the sustainability of development alternatives, relative to the present-day status quo. There are many advantages of a regional approach to CEA: efficiencies associated with collective information gathering and sharing (including vital information about baseline conditions and historical trends); the ability to set regional thresholds for acceptable environmental condition; the opportunity to evaluate the collective effects of multiple projects or strategic instruments in a streamlined regulatory process; and empowerment of, and greater cooperation between, participating organizations who advance diverse visions of a locality's development (Spaling et al. 2000; Gunn and Noble 2009; Connelly 2011).

P87: Challenges associated with scaling-up CEA from the project scale to the regional scale partly arise from increased complexity of stressor–effect relationships, and partly from greater institutional complexity (Sheelanere et al. 2013). The most basic problem concerns "how to aggregate cumulative effects beyond the scale of the individual project" (Gunn and Noble 2011). Indeed, the transition to regional CEA thinking requires us to change our current perception of environmental issues as being specific to the location of a given undertaking. It also requires the dynamics of our social–ecological systems to be understood well enough that limits, targets, and indicators of change can be specified (Gunn and Noble 2011). One may also argue that a purely biophysical definition of *effect* may not be sensible for regional CEA, for which interactions among socioeconomic and ecological attributes are central considerations.

P88: The project-based approach to CEA is "narrow, reactive, and divorced from broader planning and decision-making contexts"; it fails to adequately predict and control continuous development and its effect on the environment" (Gunn and Noble 2011; Seitz et al. 2011).

P89: CEA has not advanced adequately to encompass regional and strategic perspectives. Beyond the individual project, CEA has focused on describing baseline environmental conditions, rather than on reporting trends, analyzing scenarios, and evaluating alternative futures, so it has provided limited advice to planners

Table 2. Core principles of CEA, and their logical soundness. The logical soundness of each principle is evaluated according to the degree of consistency or agreement among, and the completeness of, its supporting premises.

Theoretical element or operational principle	Logical status
(C1) To be sustainable, human enterprise must conform to the limitations of Earth's biosphere, and CEA is an important tool for making this happen.	Inconsistent/incomplete: Although it has been entrenched globally into laws and regulations, CEA has resulted in little progress toward sustainability, and some authors have argued it has been counterproductive (e.g., Duinker and Greig 2006). Furthermore, disagreement about precisely what CEA is supposed to achieve makes its performance difficult to assess unequivocally.
(C2) Cumulative effects are aggregated, collective, accruing, and (or) combined ecosystem changes that result from a combination of human activities and natural processes; however, they are commonly defined in a more restrictive sense in policies, laws, and regulations — for example, as effects of an undertaking and its interaction with other activities occurring at the same time and in the same area.	Inconsistent/complete: The chief inconsistency concerns the definition of *effect*, which can be argued either as purely biophysical, or as including a socio-cultural dimension.
(C3) Cumulative effects assessment is a subdiscipline of EIA; it is "the process of predicting the consequences of development, relative to an assessment of existing environmental quality", or "the process of systematically analyzing cumulative environmental change". It is a systematic way to evaluate the significance of effects from multiple activities and to inform resource managers about these effects. Local differences in methods exist, and are necessitated by region-specific laws, policies, regulations, and planning systems.	Consistent/complete
(C4) CEA has multiple objectives, which are loosely arranged around the themes of formalizing environmental values, establishing an ecologically viable socio-economy, and improving knowledge and governance. As a form of environmental appraisal, CEA is not just a tool for informing and influencing decision makers. It also changes the views and attitudes of its participants, and its influence can propagate outward through participants' social networks to influence entire institutions.	Inconsistent/incomplete: Evidence about precisely what CEA is supposed to achieve is incomplete and contradictory.
(C5) VECs are used to provide a tractable scope for CEA, and to ensure that attention is paid to "the most important environmental features and processes".	Inconsistent/incomplete: Controversy exists about whether the concept of a VEC, as significant ecological feature, should be more broadly defined as elements of sustainability or societal progress; objective criteria for determining which VECs ought to be included in a given CEA are lacking.
(C6) In principle, biological (effect-based) indicators are superior to stressor-based indicators for CEA. In practice, if ecological effects are detected, the stressors causing the effects must typically be identified; therefore both indicator types may be required to diagnose causes of, and model, ecosystem changes.	Consistent/complete
(C7) Consensus on CEA's operational steps has been reached; however, objective criteria for guiding decisions that must be made at each step are lacking, and a degree of pluralism is permitted so each CEA is tailored to its context. CEA practice falls short of the aspirational ideal.	Consistent/complete: To avoid confusion, note that this well-supported claim argues that inconsistencies in operational methods exist, largely because no objective guidance can be given about the particular scope, scale, and significance thresholds to use in any given CEA; furthermore, it argues that CEA implementation has fallen short of expectations, particularly as regards the exploration of development alternatives, use of scenario-based predictive models as futuring tools, and monitoring effectiveness of the CEA process itself.
(C8) Monitoring and adaptive management are essential to CEA.	Consistent/complete
(C9) Public involvement is essential, regardless of how one considers CEA's value to partition among information-delivery and deliberative outcomes.	Consistent/complete
(C10) Continued enlightenment and endorsement of CEA requires its efficacy and efficiency to be assessed.	Consistent/complete (but note that efficacy is difficult to evaluate because of disagreements about precisely what outcomes CEA ought to bring about; and it remains unclear how to implement evaluative frameworks that have been proposed)

Table 2 (*concluded*).

Theoretical element or operational principle	Logical status
(C11) Basic knowledge about ecological stressors and effects is rudimentary. Nonetheless, numerical methods for quantifying, assessing, modeling, and predicting cumulative effects are well described. On one hand, our capacity for prediction is often small, and there are practical limits to the number of factors that can be tested experimentally or modelled; on the other hand, crude approximation of cumulative effects is sufficient in most CEA contexts.	Consistent/complete: The most significant problems with CEA are not scientific.
(C12) CEA is best undertaken in a strategic and regional context, but there are few examples of this in current practice.	Consistent/complete
(C13) CEA and planning are inseparably linked, and communication between these disciplines allows for reciprocal learning.	Consistent/incomplete: The full case for integrating CEA and planning has not yet been compiled. To avoid confusion, note that the logical case for merging CEA and planning is consistent; however, examples of this marriage from current practice are rudimentary, in part because of institutional and jurisdictional mismatches and inertia.

and decision makers (Harriman and Noble 2008; Gunn and Noble 2009).

P90: Reciprocal learning can occur between project-based and more strategic and regional forms of CEA (Connelly 2011; Seitz et al. 2011). "A regional approach can set the context needed for scoping, assessing and managing cumulative effects attributable to individual projects, whereas project-specific CEAs should build on the regional understanding and suggest in some detail how to manage the cumulative effects…(of a)…specific project" (Baxter et al. 2001).

C12: CEA is best undertaken in a strategic and regional context (Baxter et al. 2001); however, the current practice falls short of ideology (Gunn and Noble 2011).

P91: Planning is concerned with articulating societal priorities and managing change via the allocation of resources (Hegmann and Yarranton 2011); it establishes the context within with sustainability is to be implemented" (Gunn and Noble 2011).

P92: Responsibilities for EIA and CEA are more commonly the jurisdiction of "agencies having a development mandate, rather than an environmental mandate" (Pope et al. 2013). Regional planning authorities are the "logical CEA proponents" (Gunn and Noble 2009) because of the "unprecedented need for the integration of sustainability principles in the development of regional policies, plans, and programs" (Gunn and Noble 2009).

P93: Instructive feedback loops exist between CEA and planning. Sustainability goals provide a benchmark against which the significance of cumulative effects may be judged (Hegmann and Yarranton 2011); whereas, CEA organizes, analyzes, and presents information in a way that allows the sustainability of various alternatives to be assessed (Hegmann and Yarranton 2011). Indeed, reciprocal feedback loops extend even more broadly across societal domains and academic disciplines, because more, and better, CEA means that the outcomes of human activities can be better predicted. The more formal, quantitative, and strategic the planning and regulatory process, the better CEA can be tuned, and the more meaningfully its performance can be evaluated (Duinker and Greig 2007; Chapman and Maher 2014).

P94: Interdisciplinary communication and reciprocal learning permit efficiency gains to accrue as CEA is iterated. For example, scenario development becomes more efficient and cost effective if premises, assumptions, and scenario types (e.g., "business as usual", "pessimistic", "disastrous", "optimistic", or "miracle"; Cornish 2004; Duinker and Greig 2007) are shared.

C13: CEA and planning are inseparably linked, and communication between these disciplines allows for reciprocal learning.

P95: The 96 premises and 13 conclusions listed here characterize what CEA is, how and why it is done, and how and why it *ought to be* done; however, these statements are incomplete and occasionally contradictory.

C14: As is true of EIA (Cashmore 2004), CEA's theoretical basis is inadequately unified and detailed as "… a series of somewhat nebulous models operating along a broad spectrum of philosophical beliefs and values"; Table 2).

CEA's big problems

CEA should be innate. Our lives proceed via a great number of decisions, the consequences of which are — or, at our own peril, are not — considered proactively. When the oven timer rings, signaling that the casserole is cooked, a plausible future scenario includes burnt hands and a trip to the emergency room. Burning the hands leads to pain and misery, infection, loss of function, and ultimately threatens survival (especially if done more than once). Prudent mitigation by the cook is achieved by donning oven mitts before dinner is removed from the oven. Difficulty with CEA arises when we scale up from the individual level to the collective, societal, and institutional levels. This scaling-up is most easily done given an ethos of ethical objectivism, in which the collective good is paramount, principles of conservation or preservation are highly valued, and individuals are held accountable to these morals. By contrast, many authors have argued that we are in an age of individualism, consumerism, cultural, and ethical relativism, and personal non-accountability (e.g., Rubin and Kaivo-Oja 1999; Truss 2005; Hamilton 2010). These social viewpoints ultimately influence CEA decisions because they influence how development-related trade-offs[37] are perceived (Coates and Leahy 2006; Nunneri et al. 2008). Fully harnessing the potential of EIA and CEA as sustainability instruments requires innovations that overcome, redirect, or reprogram these social norms.

Cumulative effects laws, regulations, and policies on their own are universally incomplete and underachieve (Duinker and Greig 2006). Furthermore, it is important to appreciate that in most democracies the supreme constitutional principle is individual liberty. When implementing a regulation or law, the onus is on ensuring protection of individual rights and freedoms (not the environment). For this reason, legislators often take an indirect approach to environmental issues by prohibiting certain impacts,

[37]Trade-offs arise commonly in CEA where a favourable response in one indicator can only be achieved via the adverse response of another.

rather than limiting human activities (R. Cormier. pers. comm. 2015). All the while, the blame-game circumvents meaningful confrontation of cumulative effects: plan-makers argue that effects are caused by individual actions, and individuals argue that their actions are constrained by government policies (Therivel and Ross 2007).

In addition, legal, regulatory, and institutional frameworks are largely insufficient to enable CEA's diverse outcomes to be realized (Ball et al. 2013), uncertainties about jurisdictional roles and responsibilities are pervasive (especially when CEA is undertaken at a regional cross-jurisdictional scale; Damman et al. 1995), and connections to science and planning are rudimentary. For example, the Chinese case study written by Wang et al. (2003) described a series of systemic institutional problems, including project-based institutional attention being focused on air, water, and soil pollution, with social and ecological cumulative effects being largely ignored; complex and confusing lines of responsibility and accountability that vary by jurisdiction, resulting in regional variation in the quality of appraisals; waning institutional capacity, caused by insufficient funding as departmental responsibilities and workloads have expanded; perverse inter-agency incentives that arose from environmental regulators being funded by pollution levies; and weak enforcement.

CEA does cultivate some obviously good practices. For example, it upholds informed, transparent, and accountable decision making, and it results in irreconcilable disputes being referred to the judicial system (Hegmann and Yarranton 2011); however, appraisal, and its role in the decision-making process, is inadequately conceptualized (Owens et al. 2004), and its ostensive purposes are difficult to translate into definable outcomes, so it is not clear how to use CEA to make demonstrably better decisions (Cashmore 2004). CEA's integration with the decision-making process stresses information delivery to decision makers, as a way of promoting rationality. But this is an unstable philosophical basis for CEA because it neither accounts accurately for the scientific assessment – decision nexus (i.e., CEA rarely informs decisions in a straightforward linear fashion, and decision makers fail to use vast quantities of information produced for their use; Russell-Smith et al. 2015), nor for the decision-making systems that exist in modern democracies (in which there is no singular societal vision; Owens et al. 2004). As a result, CEA is both naive and potentially dangerous — it allows "ethical and political choices (to) masquerade as technical judgments" (Owens et al. 2004), and the resulting tokenism tends to reinforce a society's norms and distribution of power (which are often at odds with CEA's promise of sustainability).

In recent years, CEA has been seen in a new light, its focus "less on an individual decision and more on long-term outcomes through processes of transformation" (Pope et al. 2013); then again, CEA (and EIA) have failed to realize their pledged deliberative outcomes. One of the main reasons for this deficiency is superficial public engagement, whereby members of the public are invited only to review materials and provide comment (usually too late in the CEA process to have much impact). For example, Sinclair et al. (2012) published (in the EIA literature) a particularly scathing account of citizens' experience in the Emera Brunswick natural gas pipeline hearings in New Brunswick, Canada, which showed that "most elements of meaningful participation were not satisfied": information sessions intended to brief public participants were overly complicated, confusing, and formal, and failed to prepare participants for the intimidating adversarial and quasi-judicial review process; funds provided to public participants were meager, arrived too late in the process, and carried too many conditions to level the playing field among parties to the appraisal; the review panel failed to adjust the scope of the assessment, despite repeated appeals from the public; time horizons for phases of the appraisal, and hearing schedules, did not accommodate the schedules of public participants (most of whom had full-time jobs and no staff to assist with preparation for hearings); open discussion about how issues raised during the process could be solved was discouraged by a review process that was based on cross-examination; and professionals in the process undervalued local public knowledge (all of which left participants feeling "bitter, disrespected, marginalized, and wasted". As several authors have reported (Adelle and Weiland 2012; Pope et al. 2013) more empowering and transformative models of engagement have been slow to catch on.

Aside from CEA's theoretical basis, "resource-based targets or limits are keys to stimulating careful thought about, and robust management of, cumulative effects" (Therivel and Ross 2007) because they allow *significance* and *acceptability* to be specified quantitatively. Yet societies pay only lip service to the goal of sustainability, and *sustainability* is not defined in a suitably operational sense to provide a sound basis for CEA (Spaling et al. 2000; Connelly 2011; Hegmann and Yarranton 2011; Dubé et al 2013). As a result, solutions to cumulative problems are often deemed unpalatable, their ownership is dismissed as someone else's remit (Therivel and Ross 2007), and economic interests easily outweigh environmental interests (e.g., Adelle and Weiland 2012). A variety of methods exist for setting indicator targets relative to reference levels or other baselines, nonlinear stressor–response relationships, and social norms[38] (e.g., Samhouri et al. 2011); but there are also a variety of challenges, as Samhouri et al. (2011) described with reference to marine-ecosystem targets for Puget Sound. For example, it is difficult to calibrate present ecosystem form or function to a historic or spatial reference condition in which human impacts were absent. Furthermore ecosystems vary dynamically over time and space, meaning that historical and spatial baselines are arbitrary.

Barriers to interdisciplinary research are another problem. As Rubin and Kaivo-Oja (1999) wrote, "the boundaries between natural sciences, social sciences, and the humanities…have blurred", and this blurring needs to take place given that complex sustainability problems have to be solved by changing the interactions between social and ecological systems (Klein 2004; Binder et al. 2013); however, methods of interdisciplinary research and collaboration are rudimentary (e.g., Sheate 2010; Morrison-Saunders et al. 2014), and disincentives abound — for example, obstructive prejudgments that social and natural scientists make about one another, the lack of opportunities for financing interdisciplinary work, differing discipline-specific publishing expectations, power differentials that exist in diverse research collaborations, and the need for time-consuming procedures to manage team dynamics and clearly articulate research goals and methods (Campbell 2005; Morse et al. 2007).

Many how-to documents have been written for CEA practitioners (e.g., Hegmann et al. 1999; Cooper 2004; Munier 2004; Glasson et al. 2013), but guidance on how to operationalize CEA remains incomplete and difficult to generalize, and a variety of methodological challenges remain. This is true because decisions have to be made at each step of the CEA process, but objective and universal criteria for guiding those decisions do not exist. Societal values and sustainability goals vary from place to place, which means that the scope of issues to include in CEA assessments is impossible to standardize, as are decisions about which VECs and indicators to use (e.g., Baxter et al. 2001; Therivel and Ross 2007, Ball et al. 2013). The outcome of this subjectivity is that VECs are often included only if significantly impacted by the undertaking(s) being appraised, so environmental components that are incrementally affected in a minor way are excluded, and deaths by a thousand cuts can continue unabated (Bérubé 2007). Further-

[38]Social norms define what is generally excepted within a given cultural setting, and can be quantified statistically from survey data (Samhouri et al. 2011).

more, the appropriate spatial and temporal scale for a given CEA depends on the subject of the CEA appraisal, and on selected VECs and indicators, among other factors; however, chosen surrogate indicators may not be predictable enough to be used in CEA scenario models, and ecological indicators that can be modeled with suitable precision may not be relevant to local VECs. Objective thresholds for the significance of effects are elusive given ambiguity of the term *threshold*, and the fuzziness of sustainability goals (McCold and Saulsbury 1996; Bérubé 2007; Ball et al. 2013). Methods of evaluating performance are difficult to standardize because CEA's pluralistic information-delivery and social outcomes are treated as more or less legitimate by different authors.

According to Schultz (2012), "determining the appropriate geographic and temporal scales of analysis is one of the most perplexing aspects of CEA". This is partially because stressors and effects commonly occur at different spatial and temporal scales (Seitz et al. 2011), but also because the larger the area covered by a CEA, the less important local issues (e.g., a new factory or shopping mall) will be, the more important landscape-scale phenomena (e.g., pervasive impacts associated with the manufacturing or commercial sector) will be, the less likely it will be that a particular effect will be singled out as significant, the less likely it will be that CEA boundaries will align with administrative ones, and the less clear it will be where the responsibility for follow-up action lies. Similarly, the longer the timeframe considered, the less certain CEA analyses will be (Therivel and Ross 2007; Seitz et al. 2011).

Mathematical procedures for associating stressors and effects are relatively well described, but the required monitoring data are generally lacking (Damman et al. 1995; Scherer 2011; Schultz 2012; Ball et al. 2013; Venier et al. 2014), and are "expensive and time consuming to gather" (Connelly 2011). Obviously, more complex models call for more complex input datasets and underlying monitoring programs, so a vicious cycle is apparent (Gallagher et al. 2015). The information that is available is "highly uncertain and variable in quality" (Karkkainen 2002), and is, for the most part, not efficiently shared among CEA participants (Damman et al. 1995).

Hutchinson's (1959) concept of the niche provides a strong theoretical basis for thresholds, because it posits that organisms can tolerate only a finite range of conditions relative to any environmental dimension; however, in practice, multiple definitions of the term *threshold* exist. For example, the term can be used to describe a fundamental shift in community structure or dynamics, or a nonlinear response of a population to one or more disturbances. Furthermore, there are many levels of biological organization at which thresholds may be evident; there are a limitless number of potential response variables for which thresholds could be derived; and different statistical approaches will result in different thresholds. Exploring only a small number of these is costly and requires an immense amount of data, but only a small subset of identified thresholds will be relevant to VEC conservation (Johnson 2013). Statistical significance may be easy to demonstrate, for example, as the value of a given indicator being outside of a statistically defined "normal" (e.g., Kilgour et al. 1998), but ecological significance is another matter. Furthermore, the societal significance and relevance of a cumulative effect is open to debate, given how vague the concept of sustainability is (Scherer 2011; Moldan et al. 2012).

In addition to the subjectivity of decisions made in the CEA process, complexity and uncertainty are fundamental difficulties, commonly invoked to describe, for example, the diversity of biological and physical land-use effects, stressor interactions, spatial and temporal lags between stressors and effects, nonlinear relationships, and positive and negative feedback loops (Spaling and Smit 1993; Therivel and Ross 2007; Seitz et al. 2011; Ball et al. 2013). In Hegmann and Yarranton's (2011) pessimistic assessment, "even if practitioners employ the most advanced and complex analysis and the best that analytical thought may offer (CEA) still delivers just words and numbers buried within limitations, assumptions and uncertainties" (Hegmann and Yarranton 2011).

It is clear that CEA's big problems are not discrete. Rather, they overlap and feed off each other. They are also replicated throughout the other subdisciplines of EIA, where they have caused confusion and exaggerated methodological incompatibilities (Canter et al. 2010; Morrison-Saunders et al. 2014).

Recommendations

CEA should be considered one part of a precautionary sustainability strategy, not as a standalone solution for dealing with development-related environmental effects

Earth can neither support our current resource demands nor assimilate the ecological stresses associated with further demands (Wackernagel and Rees 1998). Ecological trends suggest deepening unsustainability (Vitousek et al. 1997; Foley et al. 2005; Steffen et al. 2007). We are caught in a vicious cycle of ecological exploitation that leads to ecosystem damage, the exhaustion of resources, and a spiral of continuing degradation, as livelihoods are undermined (Schnaiberg and Gould 2000; Gibson 2012). Environmental conservation and development may both be seen as societal imperatives, but ecological considerations ultimately must be the trump card. Proposed human endeavors should move forward only if they are expected to pay long-term ecological dividends. A mitigative approach, which slows down incremental losses, is inadequate because it falls short of the principle of no net loss (e.g., Benson 2003), and the needed reversals of present ecological trajectories (Bond et al. 2012). As a starting point, the sustainability goals of the United Nations General Assembly (UNGE 2015), which are largely directed at the world's governments, may help to change societal values so they become more aligned with CEA. Furthermore, the influence of the UN targets could be maximized by also mobilizing extra-governmental advocates for change, which Hajer et al. (2015) suggested could be done by invoking complementary notions, such as "planetary boundaries" (which stress the urgency of environmental concerns and the need to take responsibility for ecosystem goods and services), "the safe and just operating space" (which highlights interconnectedness of social and environmental concerns), and "the energetic society" (which stimulates "green competition" and technological innovation). All of these concepts should be operationalized with relevant indicators and monitoring (e.g., Hák et al. 2016), some of which will be of use in CEA.

Recognize that people and political systems are not programmed to think in terms of long-term strategy (e.g., Diamond 2005), and do not allow CEA (or EIA) to usurp precaution[39]

Considerable societal inertia exists that is counter to the principles of sustainability, and EIA's forty-year track record is not stellar. There is, however, hope for CEA, which can be viewed as a way to enforce strategic thinking.

As regards development, logically sound precaution should be operationalized in the decision making process as: *avoid, minimize, restore, offset*. CEA traditionally encompasses the first two of these strategies. Compensatory restoration and offsets (e.g., Therivel and Ross 2007; Connelly 2011) are much-needed complements, which are consistent with the idea of no net loss, and can "make

[39]Precaution is used here in the legal sense, as a principle that "ensures that a substance or activity posing a threat to the environment is prevented from adversely affecting the environment, even if there is no conclusive scientific proof linking that particular substance or activity to environmental damage" (Cameron and Abouchar 1991).

room" for future developments (Canter and Ross 2010) where further consumptive expansion is justifiable because of current deficiency. CEA should therefore be used to transition from consumptive to non- or less-consumptive means of enhancing human expression and wellbeing.

In general, any measures to protect more areas and concentrate development in areas that are already disturbed warrant attention (e.g., Venier et al. 2014). New roads and development in previously remote areas should be avoided, because opening up access causes chain reactions of effects (Cochrane and Laurance 2008; Fraser 2014). Similarly, Bond and co-authors' (2012) rules about how to handle trade-offs warrant implementation: an acceptable trade-off is defined as delivering net long-term sustainability gains; no trade-off involving significant adverse effects is acceptable, and no displacement of adverse impacts into the future is tolerable "unless all other alternatives are worse"; when a trade-off is proposed, its justification is the responsibility of the proponent, and this justification must be reviewed in an open participatory process, relative to established decision criteria. Ultimately, planning and regulatory instruments should include limits[40] (e.g., Johnson 2013) on population and developed land (Hegmann and Yarranton 2011) as a way to stimulate innovation and prevent the death by a thousand cuts.

Societies should better integrate CEA and planning (i.e., CEA should be viewed as both a tool for plan creation and evaluation), and should set clear targets for social–cultural, economic, and ecological indicators

Better consolidating CEA and planning would create a "vertically integrated framework" (Pope et al. 2013) in which environmental and sustainability issues could be considered with appropriate detail at appropriate levels of decision making. It could also benefit both processes by helping to clarify sustainability goals and uncovering (and putting to practical use) meaningful thresholds for important indicators of ecological and social wellbeing (Hegmann and Yarranton 2011). Articulating sustainability goals, and implementing them as regulatory limits (e.g., Johnson 2013), would lend administrative efficiency to CEA. This would alleviate the tendency for organizations to demand onerous appraisals as a way to compensate for inadequate policies and conflicting views of sustainability (Hegmann and Yarranton 2011). It would also greatly simplify evaluations of CEA performance. Furthermore, beneficial spin-offs outside of CEA would accrue, because targets remove ambiguity from well-intended but vague policy statements, which facilitates strategic planning and provides antecedents for complementary laws and regulations (Samhouri et al. 2011).

Consider environmental appraisal and planning to be tools for managing change, not development

Encouraging development where it makes sense to do so, and constraining it where unacceptable trade-offs are likely to result should be pursued to the extent possible (Hegmann and Yarranton 2011). The goal should be "qualitative rather than quantitative growth"[41] (Häyhä and Franzese 2014).

If the chief purpose of CEA is to enforce evidence-based rationality, then formalize quantitative rules in the decision-making process

Evidence-based decision making demands trade-offs to be defined and revealed through a structured process (Johnson 2013), and targets are a critical part of this process. Even a vague, qualitative target can stimulate policy debate and research concerning desirable and undesirable end-points. Beyond this, evidence-based decision making can be formalized and quantified by measuring indicator distances to the target (Moldan et al. 2012). Control charts (i.e., time-series plots showing trends in VEC condition, relative to acceptable limits) should be hanging on boardroom walls. Referring to such charts would help to enforce rationality by drawing decision makers' attention to their own biases (e.g., Rubin and Kaivo-Oja 1999), while promoting "futures-oriented thinking" (Masini 1993), and focussing debate on trade-offs. By this method, human activities would be assessed on a distance-to-target basis (e.g., Moldan et al. 2012) and approved if their predicted effects did not exceed a pre-defined regulatory limit, which Johnson (2013) suggested could be set relative to models of ecological response, socioeconomic trade-offs, and technological best practices. Johnson's (2013) uncertainty-based guidance illustrated how such control charts could be interpreted in relation to "critical limits" (a point at which a VEC is nearing unacceptable change), "target limits" (which operationalize an acceptable amount of VEC change, relative to ecological and socioeconomic realities), and "cautionary limits" (which identify ecological or socioeconomic changes that require adaptive monitoring).

Maximize opportunities for deliberation in CEA, because its deliberative outcomes – which include and require debate, collaboration, creativity, learning, exposure to different world views, and consideration of the distribution of power among decision makers – have the greatest potential to revolutionize our approach to sustainability

Given damning critiques of the concept of rational information-based decision making, various authors have emphasized the need for "new thinking about planning and decision making processes in their wider social, cultural, political and economic contexts" (Morgan 2012). One of the major problems with the model of information-based decision making in CEA is that it subverts meaningful civic debate by disguising development options as being merely technical, rather than political. Unfortunately, no better model has come to light (Owens et al. 2004; Bond and Pope 2012): more political views of the decision-making process, for example, are called out as being naive because their proponents expect a difficult, time-consuming, expensive, and potentially inconclusive process of deliberation to result in clear recommendations and actions (Owens et al. 2004). Regardless, information-based appraisals, and appraisals that stress the deliberative process, are not mutually exclusive, and providence calls for CEA to derive its knowledge-based products from a process that is as inclusive as possible, and seeks to maximize creativity and opportunities for collaboration and open dialogue among its participants (e.g., Bartlett and Kurian 1999; Adelle and Weiland 2012; Folkeson et al. 2013; Ball et al. 2013). Novel forms of "ad hoc delib-

[40]Johnson (2013) defined *regulatory limit* as "the magnitude or extent of human disturbance that is permitted, after which unacceptable ecological change or …. risk is expected".

[41]This turn of phrase from Häyhä and Franzese (2014) was made while contrasting flows of different types of capital in what they referred to as "strong" and "weak" systems of sustainability. Re-stating their comment, strong sustainability considers natural capital (local ecosystems, biomes, sub-soil resources) to be irreplaceable, whereas weak sustainability assumes that technological substitutes can be found for lost natural capital. Strongly sustainable ("qualitative") development, then converts natural capital to manufactured capital (e.g., roads, buildings) so as to maximize positive spin-offs to human capital (e.g., knowledge), and social capital (institutional capacity); and this conversion of natural capital is constrained by its natural rate of regeneration. The Ecological Footprint concept of sustainability (i.e., Wackernagel and Rees 1998) takes a similar view, thus a city is sustainably developed so long as its footprint does not exceed the area of biosphere required to produce its raw materials.

eration" have been advocated as one promising approach to complement regulated consultation and boost deliberative outcomes (i.e., Folkeson et al. 2013).

Continue to reason through CEA's operational challenges, and update guidance documents where possible (Connelly 2011), as a way of ushering in a more enlightened CEA

Many operational challenges and shortcomings of CEA guidance documents arise because objective decision criteria (e.g., in relation to scope, scale, and thresholds) are nonexistent. The regional variability of CEA requirements is a further complication, because it precludes generalization. Regardless, there are still plenty of opportunities for innovation. Researching methods by which trade-offs can be optimized quantitatively may be a promising approach. For example, ways of optimizing spatial and temporal scale relative to the uncertainty and significance of effects would be helpful, as would procedures that align CEA's spatial boundaries with VECs (Baxter et al. 2001), and with the presumable geographic extent of stressors and effects (Seitz et al. 2011; Ball et al. 2013).

Improve monitoring systems

Lack of data is a universal complaint among CEA practitioners. Regional monitoring programs that are interwoven with research agendas, planning and regulatory processes, and are supported with long-term funding, robust data-management, and data-sharing systems should be implemented widely (Ball et al. 2013; Dubé et al. 2013). Such programs stand to benefit many elements of CEA, by characterizing baseline conditions, detecting and describing trends, permitting stressors and effects to be associated (Seitz et al. 2013), and enabling adaptive management (Karkkainen 2002).

Consolidate good practices among EIA disciplines

Returning to the basics with a refocusing on better scoping and the sustainability ideal would inspire practitioners intellectually, and would uncover different ways of looking at the same problem, which would benefit EIA and all of its subdisciplines (Canter et al. 2010; Sheate 2010; Morrison-Saunders et al. 2014).

Continue to work on critical science questions

In particular, continue to investigate and theorize how environmental appraisal and decision-making interrelate (Cashmore 2004; Bond and Pope 2012), and continue to elucidate links between stressors and effects, which will enhance our capacity for prediction.

Foster interdisciplinarity among CEA researchers and their institutions

Interdisciplinary study of CEA's role in socio-ecological systems is equally important as discipline-specific research (Leknes 2001; Cashmore 2004). EIA researchers should establish interdisciplinary teams, governed by accountability and communication strategies that maximize reciprocal learning and define expected outcomes and timelines. Academic institutions should provide training and mentorship opportunities in interdisciplinary research (Morse et al. 2007).

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PART 3: RANDOM FORESTS AS CUMULATIVE EFFECTS MODELS — A CASE STUDY OF LAKES AND RIVERS IN MUSKOKA CANADA

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Research article

Random forests as cumulative effects models: A case study of lakes and rivers in Muskoka, Canada

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ABSTRACT

Cumulative effects assessment (CEA) — a type of environmental appraisal — lacks effective methods for modeling cumulative effects, evaluating indicators of ecosystem condition, and exploring the likely outcomes of development scenarios. Random forests are an extension of classification and regression trees, which model response variables by recursive partitioning. Random forests were used to model a series of candidate ecological indicators that described lakes and rivers from a case study watershed (The Muskoka River Watershed, Canada). Suitability of the candidate indicators for use in cumulative effects assessment and watershed monitoring was assessed according to how well they could be predicted from natural habitat features and how sensitive they were to human land-use. The best models explained 75% of the variation in a multivariate descriptor of lake benthic-macroinvertebrate community structure, and 76% of the variation in the conductivity of river water. Similar results were obtained by cross-validation. Several candidate indicators detected a simulated doubling of urban land-use in their catchments, and a few were able to detect a simulated doubling of agricultural land-use. The paper demonstrates that random forests can be used to describe the combined and singular effects of multiple stressors and natural environmental factors, and furthermore, that random forests can be used to evaluate the performance of monitoring indicators. The numerical methods presented are applicable to any ecosystem and indicator type, and therefore represent a step forward for CEA.

1. Introduction

Cumulative effects are “aggregated, collective, accruing, and (or) combined ecosystem changes that result from a combination of human activities and natural processes” (Jones, 2016). Cumulative effects assessment (CEA; abbreviations and acronyms used in this article is provided in the Appendix) is a sub-discipline of environmental impact assessment (Jones, 2016). Its chief purpose is to inform resource managers about the likely effects of multiple activities (Judd et al., 2015), which are predicted “relative to an assessment of existing environmental quality” (Dubé, 2003).

Ecosystems are inherently complex and unpredictable. Uncertainty is, therefore, inexorable in CEA (Hegmann and Yarranton, 2011; Chapman and Maher, 2014), and monitoring is fundamental because it allows the condition of valued ecosystem components to be assessed (Ball et al., 2013), signals when condition is changing (e.g., Cairns et al., 1993; Jones et al., 2002), provides inputs for

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predictive models, and facilitates adaptive management (Walters, 1986; Karkkainen, 2002).

In principle, stressor-based indicators, such as water-chemistry analytes, evaluate an ecosystem's exposure to stress, but leave unanswered questions about the ecological relevance of that stress (Roux et al., 1999). Effect-based (i.e., biological) indicators measure organisms' cumulative responses to environmental stress (Dubé, 2003); they provide a direct measure of ecological condition, but leave unanswered questions about which particular stressor, or stressors, are being responded to (Roux et al., 1999; Gunn and Noble, 2009). Stressor- and effect-based indicators are, therefore, complementary, and CEA relies on both of them (Jones, 2016).

Random forests (Breiman, 2001) are an extension of the binary partitioning algorithm that is used to produce classification and regression trees (CART; e.g., Breiman et al., 1984). The random forest algorithm creates a large ensemble or "forest" of trees, each tree being built from a random subset of the observations, and each binary partition considering only a random subset of the predictors. By averaging predictions across all the trees, the random forest improves predictive accuracy relative to classification and regression trees (CART), and the randomizations incorporated into each of the trees overcomes CART's over-fitting problem and constitutes a form of built-in cross-validation, which improves the model's generality (Breiman, 2001; Liaw and Wiener, 2002; Jones and Linder, 2015).

Random forests have several characteristics that make them practical tools for expounding individual and combined effects, and for predicting outcomes associated with scenarios of human activity: they make no assumptions about the distributions of predictor or response variables; unlike multiple regression, they do not require one to maintain a certain ratio of predictors to observations (in fact, the input dataset can contain many more predictors than observations); they can incorporate complex predictor interactions, without these interactions having to be pre-specified; and they can handle multicollinearity of predictors, and non-linear relationships between predictors and the response variable (e.g., Cutler et al., 2007; Jones and Linder, 2015).

In order to demonstrate some of their applications in CEA and ecological monitoring, we used random forests to model cumulative effects and evaluate several monitoring indicators using data from case-study lakes and rivers in the Muskoka River Watershed (Ontario, Canada; Fig. 1). Because they integrate processes that play out in their catchments, lakes and rivers are often exposed to (Nõges et al., 2016), and are highly sensitive to, a multitude of stressors (Ormerod et al., 2010; Jackson et al., 2016). These stressors act on the environment through different mechanisms (Lowell et al., 2000) and interact with one another, which makes their cumulative effects difficult to predict (Townsend et al., 2008).

The Muskoka River flows 201 km from its headwaters in Algonquin Provincial Park to Georgian Bay, and its 5660 km^2 watershed (Fig. 1) includes more than 600 lakes, which constitute the heart of Ontario's cottage country. Local gradients of geography, hydrology, and land-use (Table 1) provide sources of variation that models can exploit, and the Watershed is situated in close proximity to the Greater Toronto Area, which is one of Canada's primary urban and economic hubs and therefore exerts a degree of development pressure on the region.

Monitoring data collected for the past 30$^+$ years, combined with research conducted at the Dorset Environmental Science Centre (e.g., Yan et al., 2008) demonstrates that the Watershed's lakes and rivers are changing physically, chemically and biologically, and that multiple stressors are implicated in these changes (Jeziorski et al., 2008; Eimers et al., 2009; Palmer et al., 2011; Yan et al., 2011; Kerr and Eimers, 2012; Yao et al., 2013; Palmer et al., 2014). Cumulative effects assessment has not been implemented in a substantive way, but several of its important building blocks are in place in the Watershed: a commitment to long-term ecological monitoring, as demonstrated by a public series of occasional lectures and "watershed report cards" (e.g., Muskoka Watershed Council, 2014) sponsored by the Muskoka Watershed Council; and open lines of communication between scientists and the regional planning authority, as demonstrated by the "Muskoka River Watershed Cumulative Effects Research Node" (a research partnership between scientists and the District Municipality of Muskoka; Persaud and Eimers, 2016).

We answer the following questions in this article:

1. Which measures of lake and river water chemistry and benthic community structure can be modeled and predicted most accurately by random forests (i.e., which measures would perform best as indicators of cumulative effects)? Are lake and river models equally accurate?
2. How much information is contributed to these models by different classes of predictors (e.g., predictors describing hydrologic or physiographic attributes, land-use, land cover, waterbody morphometry, or physical habitat conditions at the sampled locations), and by variables measured at different spatial scales (i.e., at the scale of the cumulative catchment, local catchment, or riparian zone, and immediate vicinity of the sampled locations)? What are the combined and singular effects of these various predictors?
3. To what degree are random forest models predictive (i.e., how well do they predict chemical or biological qualities of waterbodies that were not included in their training datasets)?
4. Can chemical or biological indicators be predicted accurately enough, and are they sensitive enough to land-use changes, to be used in scenario models that explore potential ecological consequences of increased development?

2. Methods

2.1. Site selection

The 647 lakes and 1879 candidate stream sampling locations (CSL's) in the Muskoka River Watershed were mapped, and an attribute table was created to describe each location's ecological context using variables that described land-cover/land-use (Ontario Ministry of Natural Resources, 2015), physiography (Ontario Ministry of Northern Development and Mines, 2012), hydrology or waterbody morphometry (Ontario Ministry of Natural Resources, 2011a; 2011b). This attribute table was used to implement a stratified random site selection procedure, by which 82 lakes (36 near-pristine, 46 impacted) and 112 streams (36 near-pristine, 76 impacted) were selected to be sampled (Fig. 2) in a way that ensured relatively even spatial coverage, proportional representation of waterbodies of different sizes, and inclusion of waterbodies that reflected the spectrum of environmental characteristics and exposures to human activity that existed in the Watershed.

2.2. Sampling procedures

Sampling activities were carried out during summers (July–August) of 2012 and 2013. Benthic invertebrates were sampled using methods proposed by Jones et al. (2007). Grab samples of water were collected at each stream location, and submitted to the laboratory at the Dorset Environmental Science Centre, where assays were performed (as per Ontario Ministry of Environment, 1983) for the following 8 chemical analytes or physical

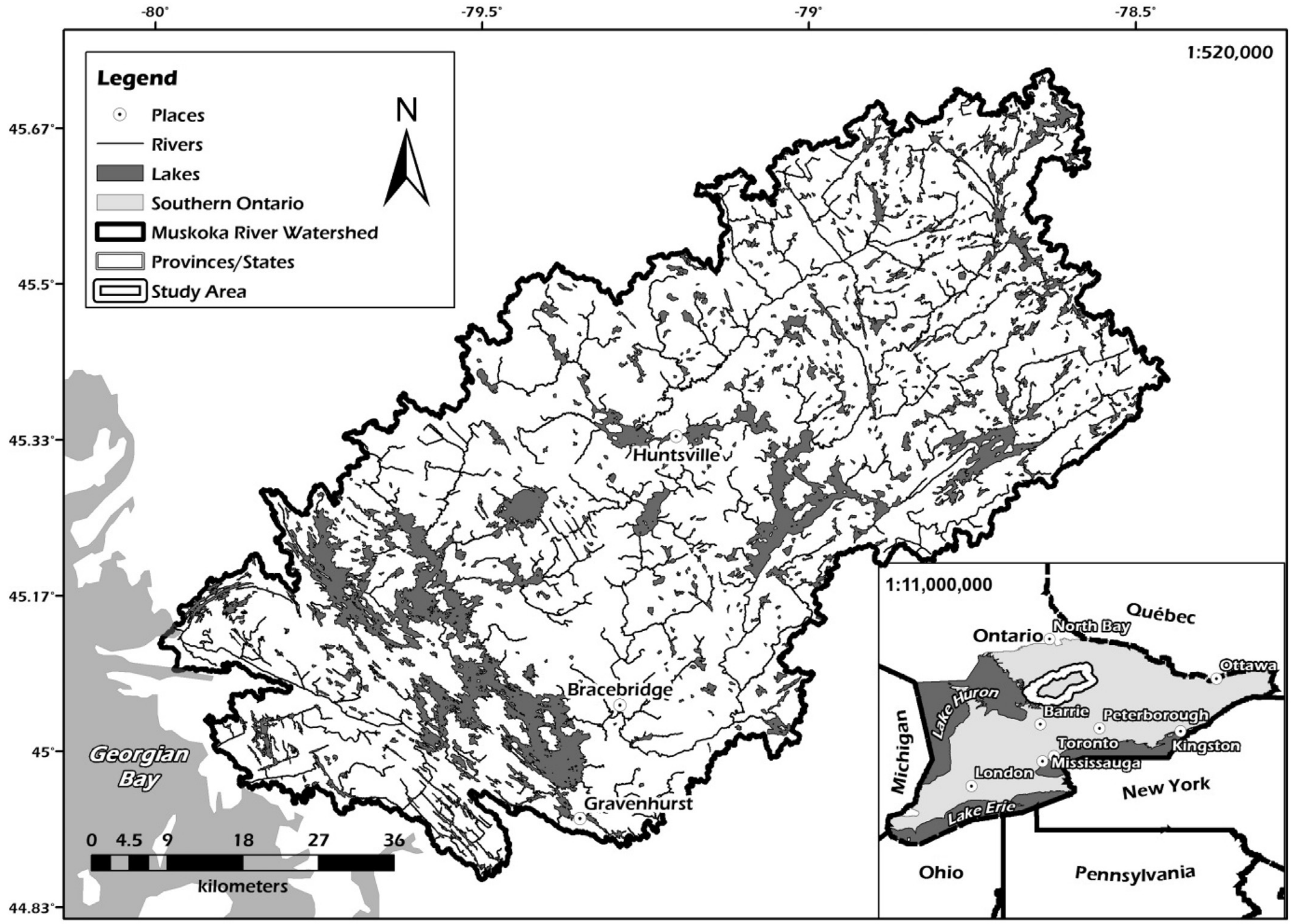

Fig. 1. Muskoka River Watershed, regional context.

properties: alkalinity (ALKT), calcium (Ca), chloride (Cl), conductivity (COND), dissolved organic carbon (DOC), total Kjeldahl nitrogen (TKN), pH, and total phosphorus (TP).

Lake sampling activities included secchi-depth, temperature, and depth measurements, which were made at the approximate centre of each lake's basin. From this central location, a random 3 of the 4 cardinal compass bearings (i.e., north, south, east, west) were sighted toward shore, and landmarks were recorded to demarcate their intersections with the shoreline. Benthic invertebrates were collected at each of these intersections (or immediately adjacent to them, in the case of the intersection itself being not wadeable or otherwise unsampleable; MacDougall et al., 2017). A composite water sample was pooled for each lake as 1-L collections of water from each sampling area (sample handling, lab, lab methods, and analytes same as for streams).

Approximate 100-counts of invertebrates from each sample were sorted, enumerated, and identified (with taxonomic precision that was approximately family-level) from random aliquots, extracted from a 100-cell Marchant-style (e.g., Marchant, 1989) sub-sampling box.

2.3. Datasets

The Hydrologic, geographic, morphometric, and land-use/land-cover attributes included in our database of sampled locations were measured at four different spatial scales. They characterized sampled lakes' and rivers' cumulative catchment areas (variables measured at this scale are denoted by the prefix "c_"), local catchment areas (variables denoted by "l_"), and 300-m riparian zones ("r_"), as well as the habitat conditions observed at the immediate locations where water samples and benthic invertebrates were collected (Plewes, 2015).

Hydrologic predictors (some of which were proposed by Martin et al., 2011) included the soil wetness index (swi), drainage density (dd), mean slope of the drainage network (slp), Strahler order (strahl; Strahler, 1957) local drainage area (l_da), cumulative drainage area (c_da), proportions of the l_, d_, or r_ areas made up of bogs (bog) or other waterbodies (wtr), drainage ratio (DR; lakes only: the ratio of the cumulative drainage area to lake area), water residence time (wrt; lakes only: the mean length of time water stays in a lake), cumulative water residence time (cwrt; lakes only: the average length of time water in a given lake will stay in lakes, including time spent in upstream lakes; Müller et al., 2013), and lake network number (LNN; the number of lakes in the lake of interest's lake chain [headwater lakes have LNN = 1]). Geographic predictors included the latitude (lat), longitude (lon), and elevation (elev) of the sampled locations, overburden thickness (over), and the proportions of c_da that were in each of the Watershed's three physiographic regions, as classified by Chapman and Putnam (1984): the Highway 11 Strip (X11Strp), Algonquin Highlands (AH), and Georgian Bay Fringe (GBF). Morphometric predictors were quantified for lakes only (Plewes, 2015), and included lake

Table 1
Summary of geographic, hydrologic, land-cover, chemical, and biological attributes of study lakes and streams (see Appendix for explanations of abbreviations and measurement units).

	elev	c_da	c_over	c_swi	c_dd	c_slp	strahl	c_fn	ALKT	Ca	Cl	COND	DOC	TKN	pH	TP	Insect	Rich100	EPT	HBI
Lakes (n = 107)																				
min	189.0	4.4E+05	0.0	5.4	0.0001	0.01	1	66.4	2.5	0.8	0.07	9.6	2.0	160.0	5.2	1.7	17.3	7.1	0.5	4.5
mean	329.3	2.5E+08	1.1	7.6	0.0014	0.06	3.9	95.7	8.3	2.6	4.60	45.8	5.1	296.1	6.7	7.0	62.9	14.4	9.4	6.7
max	528.0	4.6E+09	10.6	11.1	0.0033	0.15	8	100.0	23.7	9.0	52.79	232.0	16.1	971.0	7.5	30.2	99.1	22.5	35.7	7.6
sd	85.1	7.8E+08	1.3	1.3	0.0006	0.03	2.1	6.2	4.2	1.4	7.34	41.6	2.3	108.6	0.4	5.2	19.0	3.0	7.3	0.6
Rivers (n = 112)																				
min	184.0	4.1E+05	0.0	7.5	0.0010	0.02	2	32.1	0.1	0.8	0.04	13.6	1.6	145.0	4.2	4.3	13.5	5.6	0.0	4.1
mean	300.2	7.3E+07	1.6	14.3	0.0022	0.07	3.8	91.1	20.5	6.9	20.68	120.3	10.8	557.7	6.7	30.5	79.7	13.1	12.2	6.7
max	479.0	1.2E+09	10.7	17.5	0.0057	0.34	6	100.0	124.0	44.4	355.62	1320.0	71.2	1570.0	8.1	302.0	99.2	22.7	57.9	7.9
sd	61.7	1.8E+08	2.4	1.3	0.0008	0.04	1.1	13.8	25.0	9.2	57.04	215.3	8.9	303.0	0.6	34.4	21.0	3.2	13.4	0.8

area (area), perimeter (pmtr), maximum depth (zmax), mean depth (zmean), and volume (vol). Land-use predictors included areal percentages urbanized (urban), under agricultural land-use (agri), or maintained as golf course (glf). Un-paved (urd) and paved (rd) road densities were also calculated, as were the number of municipal waste disposal sites (wast) and dams (dam) in c_da, and the cumulative distance to all dams in the c_da (damdist). Land-cover predictors included areal percentages as exposed bedrock (rock), deciduous (dec) or coniferous (con) forest, and the total of forest and other natural land-covers (fn). Predictors describing aquatic habitats at the locations where benthic invertebrates were collected included the maximum water depth along each sample-collection transect (sdepth), dominant (DS) and second dominant (2DS) inorganic pavement-layer substrate particle types, median axis dimensions of randomly selected pavement-layer particles (PCmed; as per Stanfield, 2007), ordinal abundances of emergent macrophytes (Me), rooted floating macrophytes (Mrf) and submergent macrophytes (Ms), ordinal coverages of woody material (wood) and detritus (det), and ordinal abundances of filamentous (Afi) and attached (Aa) algae (refer to the Appendix for units of measure).

Response variables (i.e., candidate indicators, Y's) included the 8 chemical analytes, 106 benthic invertebrate taxa (listed in Electronic Supplement #1), and 18 indices of benthic community structure (Electronic Supplement #2). The benthic indices summarized the relative abundances of the different taxa, their tolerances to pollution, their habit or functional feeding designations, or the diversity of taxa represented, and included: percent of sample abundance accounted for by Amphipoda (Amph), percent accounted for by Chironomidae (Chir), percent of the sample accounted for by the combined abundances of Corixidae, Isopoda, Gastropoda, and Hirudinea taxa (CIGH), percent accounted for by Ephemeroptera, Plecoptera, and Trichoptera taxa (EPT), Ephemeroptera, Odonata, and Trichoptera taxa (EOT), Insects (Insect), aquatic earthworms (i.e., non-hirudinean Clitellata; NHC), animals with a burrowing habit (Bw), filtering collectors (FC), gathering collectors (GC), predators (P), scrapers (SC), or shredders (SH); Hilsenhoff's family biotic index (HBI; Hilsenhoff, 1988); taxonomic richness (the number of different taxa represented, standardized to a 100-count sample using rarefaction; Rich100); Axis-1 and Axis-2 scores from a principal coordinates analysis ordination of samples, based on Bray-Curtis distances among their taxa counts (PCoA1 and PCoA2); and Axis-1 and Axis-2 scores from a correspondence analysis ordination of samples, based on their $\log_{10}$-transformed taxa abundances (CA1 and CA2). Each index was calculated individually for each sample, and the mean was used to characterize the lake or stream site.

Predictors and response variables for the lakes and rivers were assembled into six datasets (Electronic Supplement #2): (1) 107LB (107 Lakes Biology: 83 X's, 57 Y's); (2) 107LC (107 Lakes Chemistry: 64 X's, 8 Y's); (3) 112RB (112 Rivers Biology: 74 X's, 56 Y's); (4) 112RC (112 Rivers Chemistry: 55 X's, 8 Y's); (5) 219LRB (219 Lakes or Rivers Biology: 75 X's, 72 Y's); (6) 219LRC (219 Lakes or Rivers Chemistry: 56 X's, 8 Y's).

2.4. Cumulative effects models (random forests)

Cumulative effects of the multiple predictors were modeled independently for each dataset and Y-variable using random forests (e.g. Breiman, 2001; Liaw and Wiener, 2002). The 209 cumulative effects models, along with their associated diagnostic plots and descriptive statistics were created using a script written in the R programming language (R Core Team, 2016; Electronic Supplement #3). The accuracy of each model (i.e., the proportion of the variance in Y explained by X) was assessed using the "pseudo-R^2" (Liaw and

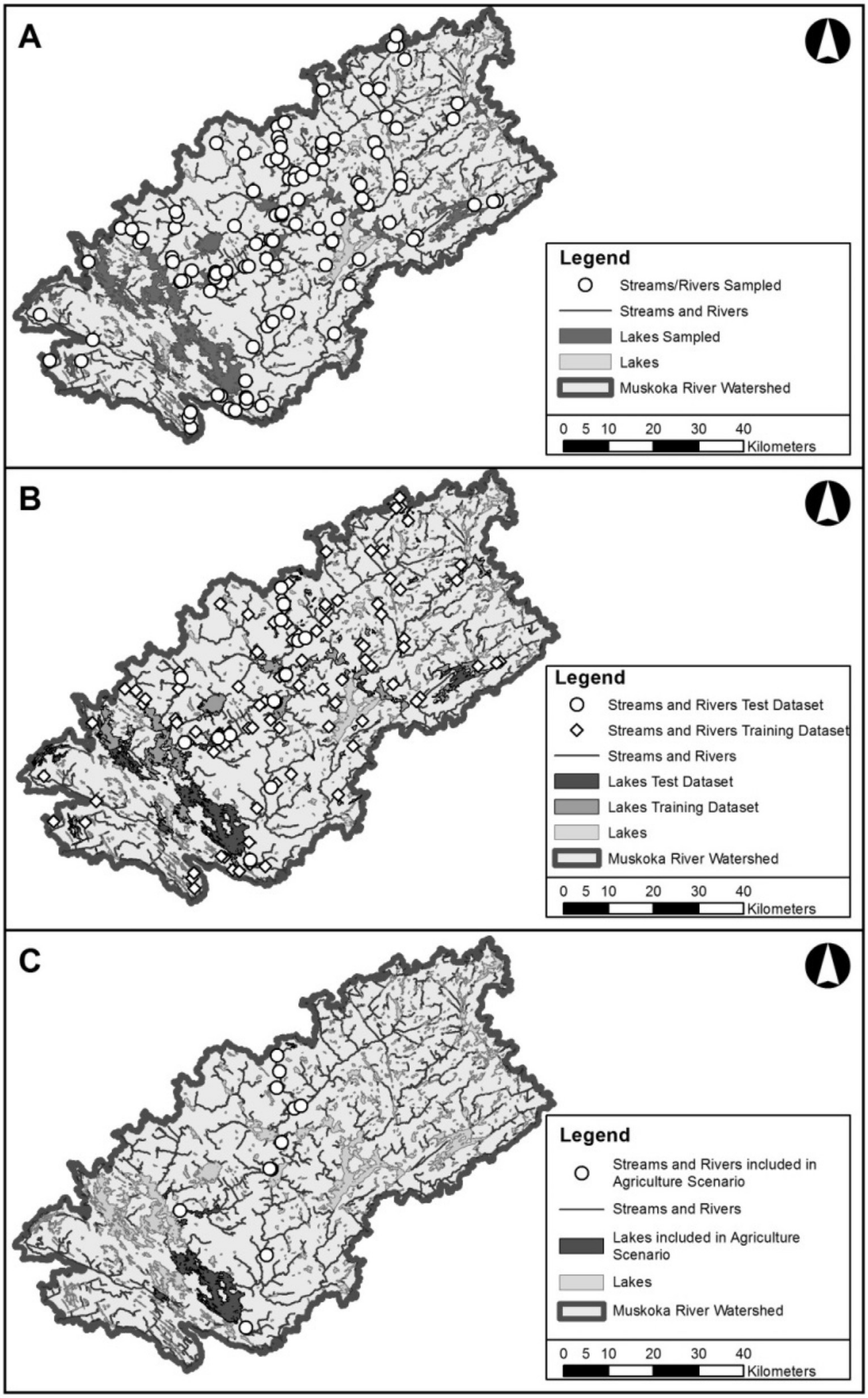

Fig. 2. Locations sampled to generate random forest input datasets.

Wiener, 2002; Smith et al., 2014) and the R^2 from a least squares regression of the random forest's predicted values against observed values. Each predictor's singular effect was demonstrated by how much it influenced the random forest's predictive accuracy (i.e., by its "variable importance"), and by the shape of its "partial dependence plot".

We used conditional variable importance plots (e.g., Hollister et al., 2016) to quantify the relative importance of the predictors included in each model. In these graphics, the conditional importance of each predictor was plotted against that predictor's rank, and the shape of the resulting curve provided context for interpreting the relative importances of all included predictors: a steeply declining curve indicated that the few most important variables overwhelmingly contributed predictive accuracy to the random forest, relative to what is contributed by the less important variables; whereas, a more gradual curve indicated that a diverse set of predictors contributed relatively evenly to the random forest's predictive accuracy.

2.5. Cross-validation

To evaluate the generality of the models, we recombined our 219LRB dataset into training datasets of 182 waterbodies (182BTrain: 75 X's, 13 biological Y's and 182CTrain: 56 X's, 8 chemical Y's) and test datasets of 37 waterbodies (37BTest: 75 X's, 13 biological Y's, and 37CTest: 56 X's, 8 chemical Y's; Fig. 2). These cross-validation datasets included all water-chemistry Y's, but only the 13 biological indices (Asell, Amph, CA1, CA2, Chir, CIGH, EPT, HBI, Insect, NHC, PCoA1, PCoA2, and Rich100) that previous models showed could be predicted with reasonable accuracy (as per Electronic Supplement #4). We then modeled all Y's in the training datasets, and used these random forests to predict values for the test datasets. Goodness of fit between predicted and observed values was assessed with R^2 statistics. Furthermore, each model's mean prediction error was calculated as the 95% confidence interval about the mean of its residuals (MAE_{test}; i.e., the mean absolute error of predicted values, relative to observed values).

2.6. Indicator sensitivity to simulated development

To determine whether any of the candidate biological indicators (Y's) would be sensitive enough for use in watershed- or region-scale CEA scenario analyses, we modified predictors in the 37BTest dataset to simulate a watershed-wide doubling of urbanization (2XUrb) or a doubling of agricultural land-use in the X11Strp physiographic region (2XAgri). Predicted values for the 13 Y's were generated for each of these scenarios (using random forests trained on 182BTrain dataset), and detectability of the resulting effect sizes was evaluated relative to model error (i.e., relative to MAE_{test}).

The scenario-specific watershed- or region-scale effect was quantified as the 95% confidence interval about its mean effect size, which we refer to as MAD_{2XUrb} or MAD_{2XAgri} — the mean absolute deviation of scenario predictions relative to predictions from 37BTest. The scenario was judged as detectable overall if its mean effect size was larger than model error (i.e., if MAD_{2XAgri} or MAD_{2XUrb} was significantly larger than MAE_{test}). Detectability was also evaluated on a site-by-site basis, by counting the number of sites having an effect size larger than its site-specific random-forest residual.

Unless otherwise indicated, datasets (including 2XUrb and 2XAgri) were compiled in Microsoft Excel. Statistical analyses were performed using scripts (Electronic Supplement #3) written and executed in R Studio version 0.99.903, which was running R version 3.3.1. Maps were created, and geospatial analyses were performed, using ArcGIS Desktop 10.2 and 10.4.1 or System for Automated Geoscientific Analyses (SAGA) version 2.0.8 (Plewes, 2015). Additional methodological details are provided in Electronic Supplement #5.

3. Results

3.1. Environmental variation among sampled waterbodies

The lakes and streams included in our study varied hydrologically (e.g., cumulative drainage areas between 4.1×10^5 and 4.6×10^9 m^2, with soil wetness indices ranging from 5.4 to 17.5), geographically (e.g., elevations between 184 and 528 m asl, mean catchment overburden thicknesses ranging from 0 to 10.7 m), and according to land cover (e.g., forest and other natural land cover classes accounting for between 32 and 100% of catchment areas). This variation was reflected in waterbodies' chemical, attributes (ALKT ranged from 0.1 to 124 mg/L as $CaC0_3$; Ca concentrations from <1 to 44 mg/L; Cl from 0.04 to 355 mg/L; COND 9.6 to 1320 μS cm^{-1}; DOC 1.6–71.2 mg/L; TKN 145–1570 μg.L^{-1}; pH 4.2 to 8.1; and TP ranged from 1.7 to 302 μg.L-1), and also in their biological attributes (between 6 and 23 unique taxa were collected); 14–99% of invertebrate abundance was contributed by insects; EPT taxa accounted for between 0 and 58% of abundance, and HBI ranged from 4.1 to 7.9 (Table 1).

3.2. Accuracies of cumulative effects models

Pseudo-R^2 values for the 209 random forest models created from the 107LB, 107LC, 112RB, 112RC, 219LRB, and 219LRC datasets ranged from 0.00 to 0.75 (Table 2, Figs. 3–5, Electronic Supplement #4). Pseudo R^2 and R^2 values were in close agreement (pearson's correlation between the two statistics was 0.997, considering the 71 models included in Electronic Supplement #4 that had pseudo-R^2 values $\geq$ 0.20.) In general, chemical models had greater predictive accuracy than was achieved by biological models (Fig. 5). The best performing biological models created from the lake datasets had greater predictive accuracies than those created from the river datasets; however, the reverse was true for chemical models, i.e., the most accurate river models were better than the best lake models (Fig. 5).

The most accurately modeled biological Y's in the 107LB, 112RB, and 219LRB datasets were CA2 (107LB pseudo-R^2 = 0.75), Asell (112RB pseudo-R^2 = 0.35), and PCoA1 (219LRB pseudo-R^2 = 0.55; Table 2, Fig. 3, Electronic Supplement #4). The most accurately modeled chemical Y's were ALKT (107LC pseudo-R^2 = 0.62) and COND (112RC pseudo-R^2 = 0.76; 219LRC pseudo-R^2 = 0.73; Table 2, Fig. 4, Electronic Supplement #4).

3.3. Influential predictors

The most important predictors in the CA2 model (107LB dataset) were pmtr, pH, and strahl (−14% to −13% MSE); in the Asell model (112RB dataset) were c_fn, c_over, and r_urban (−6% to −5% MSE); in the PCoA1 model (219LRB dataset) were r_dd, c_swi, and strahl (−14% to −11% MSE); in the ALKT model (107LC dataset) were c_fn, c_agri, and c_rd (−19% to −8% MSE); in the COND model (112RC dataset) were c_rd, fn, c_agri (−9.5% to −9% MSE); and in the COND model (219LRC dataset) were c_fn, c_agri, and c_rd (−12% to −9% MSE). Further details are provided in Figs. 6–8. Variable importance curves for CA2 (107LB) and ALKT (107LC) declined sharply beyond the first few predictors (i.e., the few most important predictors contributed overwhelmingly to the model's accuracy, relative to what was contributed by less influential predictors); whereas the curves for Asell (112RB) and COND (112RC) declined much more gradually (i.e., a more diverse set of predictors

Table 2
Performance of best random forest models trained on each of eight datasets (Y = the number of response variables [i.e., the number of random forest models built]; X = the number of predictors included in the random forest models; n_1 = the number of observations included in the training dataset; n_2 = the number of observations included in the test dataset [i.e., for calculation of R^2]; mtry = the number of candidate splitting variables considered at each node; Y1 − Y10 represent the best modeled response variables), which are listed in descending order according to their pseudo-R^2 values (which are given in parentheses).

	107LB	107LC	112RB	112RC	219LRB	219LRC	182BTrain/37BTest	182CTrain/37CTest
waterbody type	L	L	R	R	L, R	L, R	L, R	L, R
Y	57	8	56	8	72	8	13	8
X	83	64	74	55	75	56	75	56
mtry	27	21	24	18	25	18	25	18
n_1	107	107	112	112	219	219	182	182
pseudo-R^2 range	0.00–0.75	0.30–0.62	0.00–0.35	0.00–0.76	0.00–0.55	0.16–0.73	0.15–0.51	0.04–0.59
n_2	107	107	112	112	219	219	37	37
R^2 range	0.00–0.77	0.33–0.63	0.00–0.35	0.07–0.78	0.00–0.57	0.22–0.76	0.13–0.67	0.46–0.92
Y1	CA2 (0.75)	ALKT (0.62)	Asell (0.35)	COND (0.76)	PCoA1 (0.55)	COND (0.73)	PCoA1 (0.51)	COND (0.59)
Y2	PCoA1 (0.67)	Ca (0.53)	CIGH (0.35)	ALKT (0.69)	CA2 (0.52)	ALKT (0.71)	CA2 (0.47)	ALKT (0.56)
Y3	Chir (0.64)	DOC (0.51)	CA1 (0.34)	Cl (0.68)	Insect (0.48)	Cl (0.65)	CA1 (0.42)	Ca (0.54)
Y4	Chiro (0.61)	TP (0.43)	Insect (0.32)	Ca (0.60)	CA1 (0.48)	Ca (0.62)	Insect (0.41)	TKN (0.45)
Y5	Insect (0.56)	pH (0.42)	Rich 100 (0.26)	pH (0.44)	Amph (0.41)	TKN (0.50)	Amph (0.38)	pH (0.43)
Y6	PCoA2 (0.37)	COND (0.36)	CA2 (0.23)	TKN (0.38)	Hyale (0.40)	pH (0.47)	Chir (0.35)	Cl (0.33)
Y7	CIGH (0.36)	TKN (0.33)	Hyale (0.22)	DOC (0.19)	Chir (0.40)	DOC (0.28)	PCoA2 (0.29)	DOC (0.22)
Y8	Rich100 (0.35)	Cl (0.30)	Arach (0.20)	TP (0.00)	PCoA2 (0.38)	TP (0.16)	Rich 100 (0.28)	TP (0.04)
Y9	Platy (0.34)	–	EPT (0.18)	–	Asell (0.37)	–	NHC (0.26)	–
Y10	Asell (0.31)	–	PCoA1 (0.17)	–	Chiro (0.35)	–	CIGH (0.20)	–

contributed to the models' accuracies).

Considering the three most important variables in each model, chemical and morphometric classes of predictors were most influential in models of lake biology; geographic predictors were most influential in models of lake chemistry; hydrologic and land-use/land cover variables were most influential in models of river biology; and land-use/land-cover variables were most influential in models of river chemistry. Similarly, predictors measured at the site scale were most influential in lake biology models, whereas predictors measured at the scale of the cumulative drainage basin had the most influence in models of chemistry or river biology (Fig. 9). Variable importances for each model having a pseudo-R^2 > 0.2 are provided in Electronic Supplement #4.

3.4. Singular effects

Singular effects of the most important predictor in each of the six datasets' most accurate models (Fig. 10) demonstrate that lake benthic communities shift up on axis 2 of the CA in response to increasing pmtr; that the alkalinity of lake water decreases along a gradient from low to high natural land cover; that the proportion of Asellidae in river samples decreases in response to increasing natural land cover in the cumulative drainage basin; that the conductivity of river water increases with the density of paved roads in the cumulative drainage basin; that lake and river benthic communities shift to the left on PCoA1 in response to increasing drainage density in the riparian zone; and that the conductivity of lake and river water decreases in highly naturalized watersheds, relative to watersheds with less forest and other natural land cover classes. Some of the singular effects were approximately linear (e.g., the effect of c_fn on Asell [112RB], of c_rd on COND [112RC], and r_dd on PCoA1 [219LRB]). Others were non-linear (e.g., the response of CA2 to pmtr [107LB]). Additional singular effects are plotted in Electronic Supplement #4 and summarized in Table 4. On the whole, singular effects were relatively small, ranging from 7.1% to 13.7% of range_Y (Table 3). Effect sizes were positively correlated with models' pseudo-R^2 statistics, and negatively correlated with variable importance (Electronic Supplement #4).

3.5. Model generality (cross-validation)

The 182BTrain/37BTest and 182CTrain/37CTest models performed similarly as their complementary 219LRB and 219LRC models: these two groups of random forest models had similar ranges of pseudo-R^2 values; similar ranges of R^2 statistics (the R^2 statistics for the training/test datasets were often much higher than expected, given the training datasets' pseudo-R^2 statistics); and their Y's were similarly ranked according to predictive accuracy (Table 2).

3.6. Indicator sensitivity (2XUrb and 2XAgri scenarios)

For each of the Y's modeled in the urbanization and agricultural intensification scenarios, 95% confidence intervals for MAD_{2XUrb} and MAD_{2XAgri} overlapped with MAE_{test}. Nonetheless, effects of the 2XUrb scenario were detected at one or more sites by all of the modeled Y's, and effects of the 2XAgri scenario were detected at one or more sites by 5 of the 13 modeled Y's (Table 5). The indicator most sensitive to the simulated urbanization was Asell, which detected lake effects at 10 of 22 sites and river effects at 3 of 15 sites. The least sensitive indicators were CA1 and Rich100, which detected only lake effects, at a single location each (Table 5). The indicator most sensitive to simulated agricultural development was CIGH, for which lake effects were detected at 2 of 8 locations and river effects were detected at 1 of 12 locations. Eight of the thirteen modeled indicators (CA2, NHC, Amph, Chir, Insect, HBI, CA1, and Rich100) did not detect simulated agricultural intensification at any sites (Table 5). The large R^2 values that described correlations between c_urban and the water-chemistry predictors suggested that chemical effects of the 2XUrb scenario would be detectable.

4. Conclusions and discussion

4.1. Random forests are cumulative effects models

Random forests are appropriately referred to as cumulative effects models because they describe (e.g., Shmueli, 2010) empirical relationships between interacting environmental factors and indicators of aquatic ecosystem condition — and they do so in a way that makes no assumptions about the distributions of the

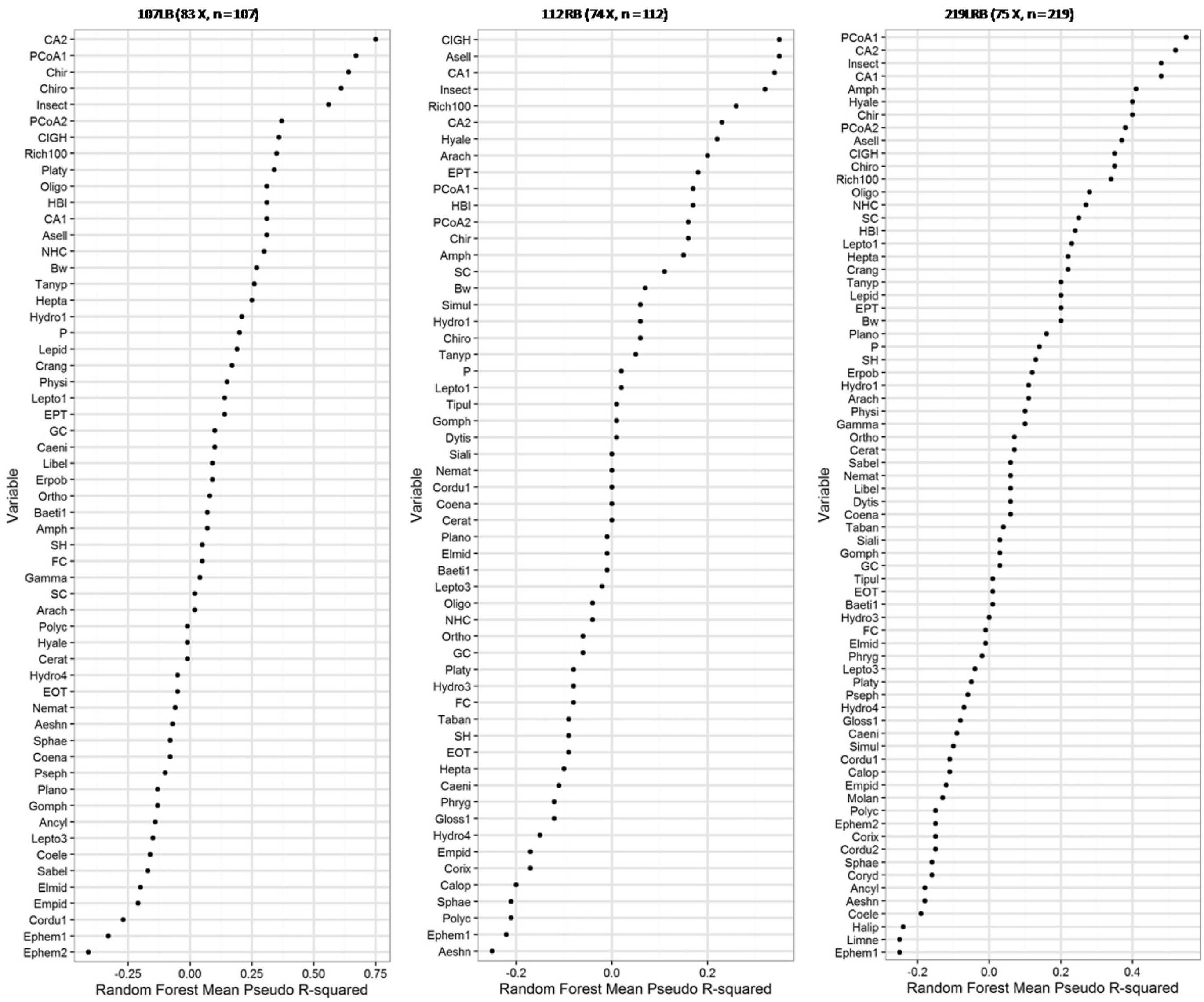

Fig. 3. Accuracies of biological cumulative effects models (abbreviations explained in Appendix and Electronic Supplement #1). Pseudo-R^2 values < 0 are interpreted as equivalent to zero.

predictors, is robust to over-fitting and inclusion of a large number of predictors, and accounts for predictor interactions, without these interactions having to be pre-specified (as in multiple regression).

4.2. *Which chemical and biological indicators were modeled most accurately?*

The most accurate biological model for lakes was CA2, for rivers was Asell, and for lakes and rivers was PCoA1. The most accurate chemical model for lakes was ALKT, and for rivers (and both types of waterbodies combined) was COND. In general, water-chemistry effects were predicted more accurately than biological effects. As a rule, biological effects were predicted more accurately for lakes than for rivers. The reverse was true for chemical effects. Although there were a large number of biological lake and river attributes that could not be modeled accurately, pseudo-R^2 statistics for our best models were quite high, and exceeded many of the accuracies reported in Nõges et al. (2016) literature review, which reported ranges of R^2 statistics from 0.3 to 0.7 (median = 0.47) for lake models and 0.3–0.59 (median of 0.42) for river models.

4.3. *How much information was contributed to cumulative effects models by different classes of predictors, and by variables measured at different spatial scales?*

Our case study demonstrated that chemical and biological attributes of lakes and rivers were typically influenced by a variety of predictors. Variable importance curves typically had relatively steep slopes in the vicinity of the most important predictors, and flatter slopes associated with lower ranked predictors. In other words, the several most important predictors generally contributed much more predictive power to their random forests than was provided by lower ranked predictors. Individual predictors typically had small singular effect sizes, as demonstrated by their partial dependencies.

Chemical, morphometric, hydrologic and land-use/land cover predictors were more important in biological models than geographic variables or measures of physical habitat at the sampled

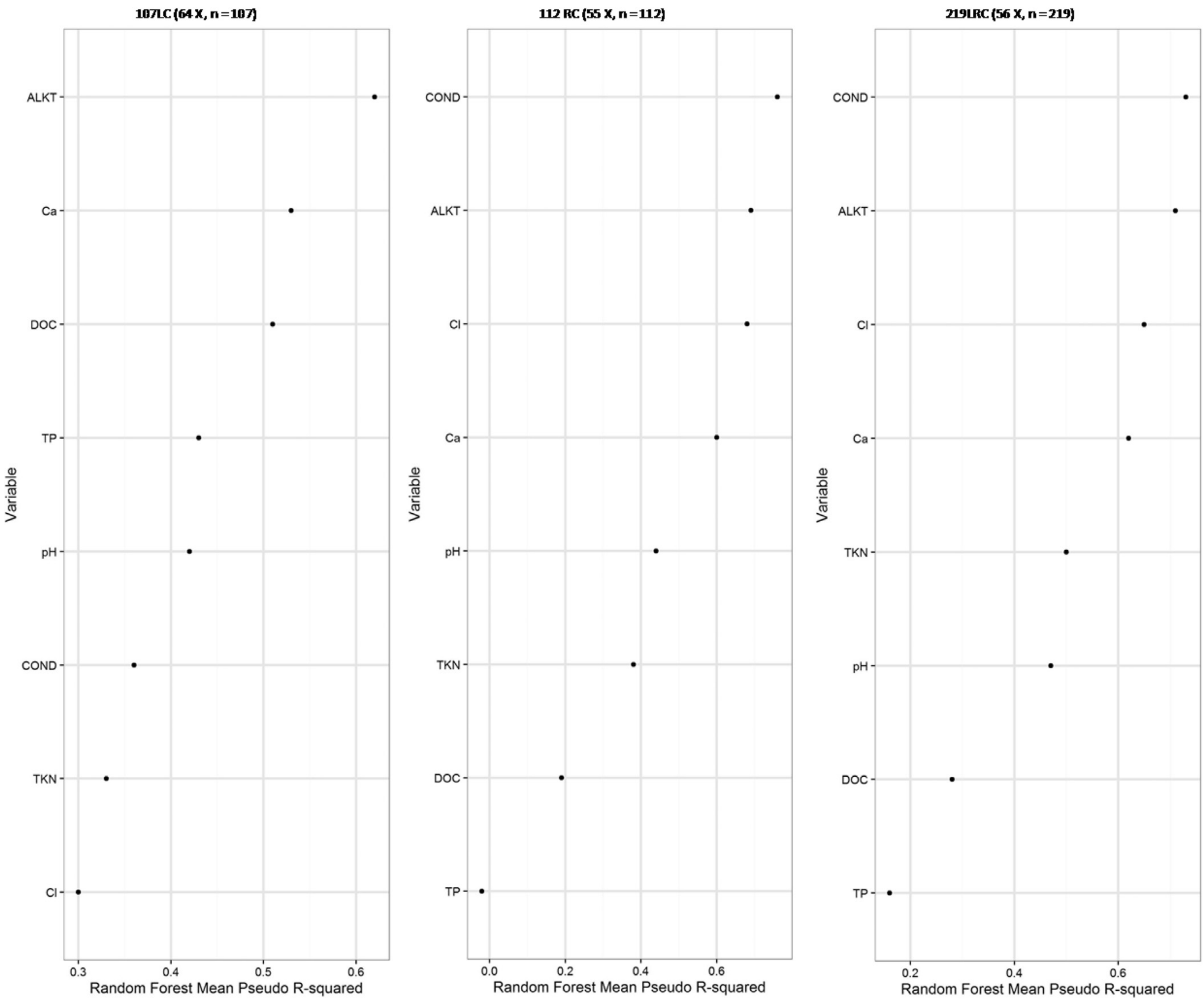

Fig. 4. Accuracies of chemical cumulative effects models (abbreviations explained in Appendix and Electronic Supplement #1). Pseudo-R^2 values < 0 are interpreted as equivalent to zero.

locations were. Measures of land-use and land cover were of greatest importance to the water-chemistry models. Regarding the spatial scale of the predictors, variables describing attributes of the cumulative catchment or physical characteristics of the sampled locations tended to be most important in biological models; whereas variables measured at the cumulative catchment scale were, for the most part, overwhelmingly important predictors in the water chemistry models.

4.4. How can random forest outputs be used to evaluate indicators?

CEA requires cumulative effects to be understood collectively and individually. An indicator becomes a candidate for inclusion in a monitoring program or cumulative effects modeling exercise once it is accepted as a measure of the condition of a valued ecosystem component. The usefulness of any candidate indicator is determined to a large degree by how well its value can be predicted, and by how sensitive it is to the human activity that is the subject of environmental appraisal. The random forest's pseudo-R^2 provides a direct measure of predictive accuracy. Variable importances and

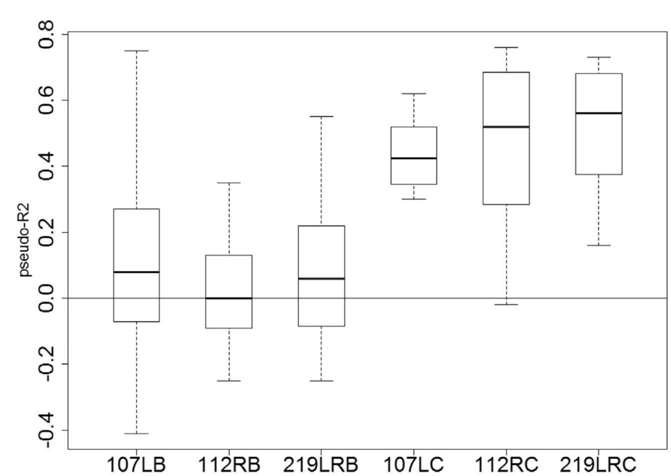

Fig. 5. Distributions of pseudo-R^2 values arising from all models generated from each of six datasets (n = 57, 56, and 72 for the 107LB, 112RB, and 219LRB datasets, respectively; and n = 8 for the 107LC, 112RC, and 219LRC datasets). Pseudo-R^2 values < 0 are interpreted as equivalent to zero (boxes enclose the central 50% of the distributions of pseudo-R^2 values; medians are represented as horizontal lines, and whiskers extend up to the maximum, and down to the minimum, values).

Fig. 6. Importances of predictors in the best random forest models (i.e., models with the highest pseudo-R^2) from the 107LB and 107LC datasets (model MSE's are given in parentheses). Each predictor's plotted point represents the percent change in MSE that resulted from its conditional permutation.

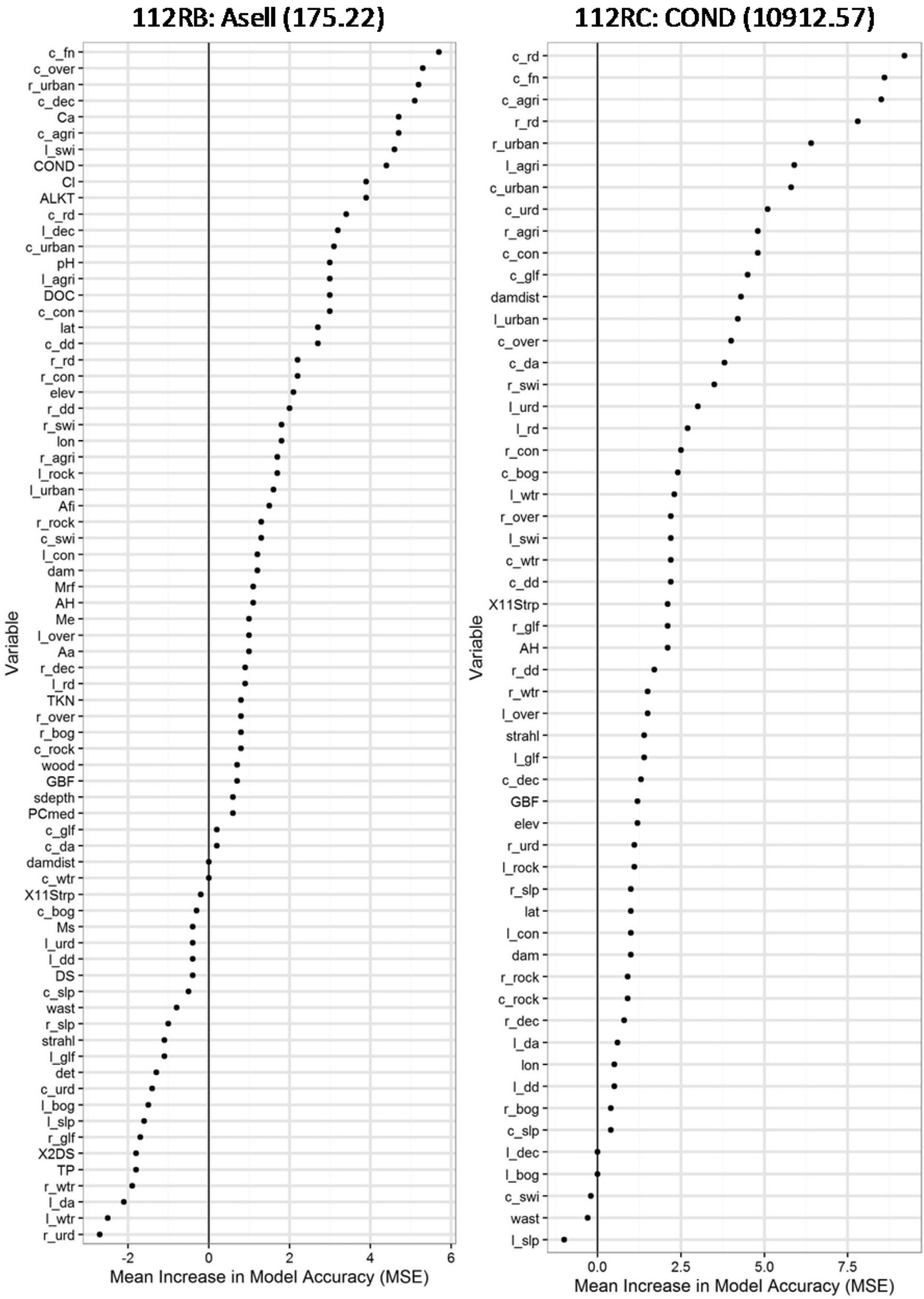

Fig. 7. Importances of predictors in the best random forest models (i.e., models with the highest pseudo-R^2) from the 112RB and 112RC datasets (model MSE's are given in parentheses). Each predictor's plotted point represents the percent change in MSE that resulted from its conditional permutation.

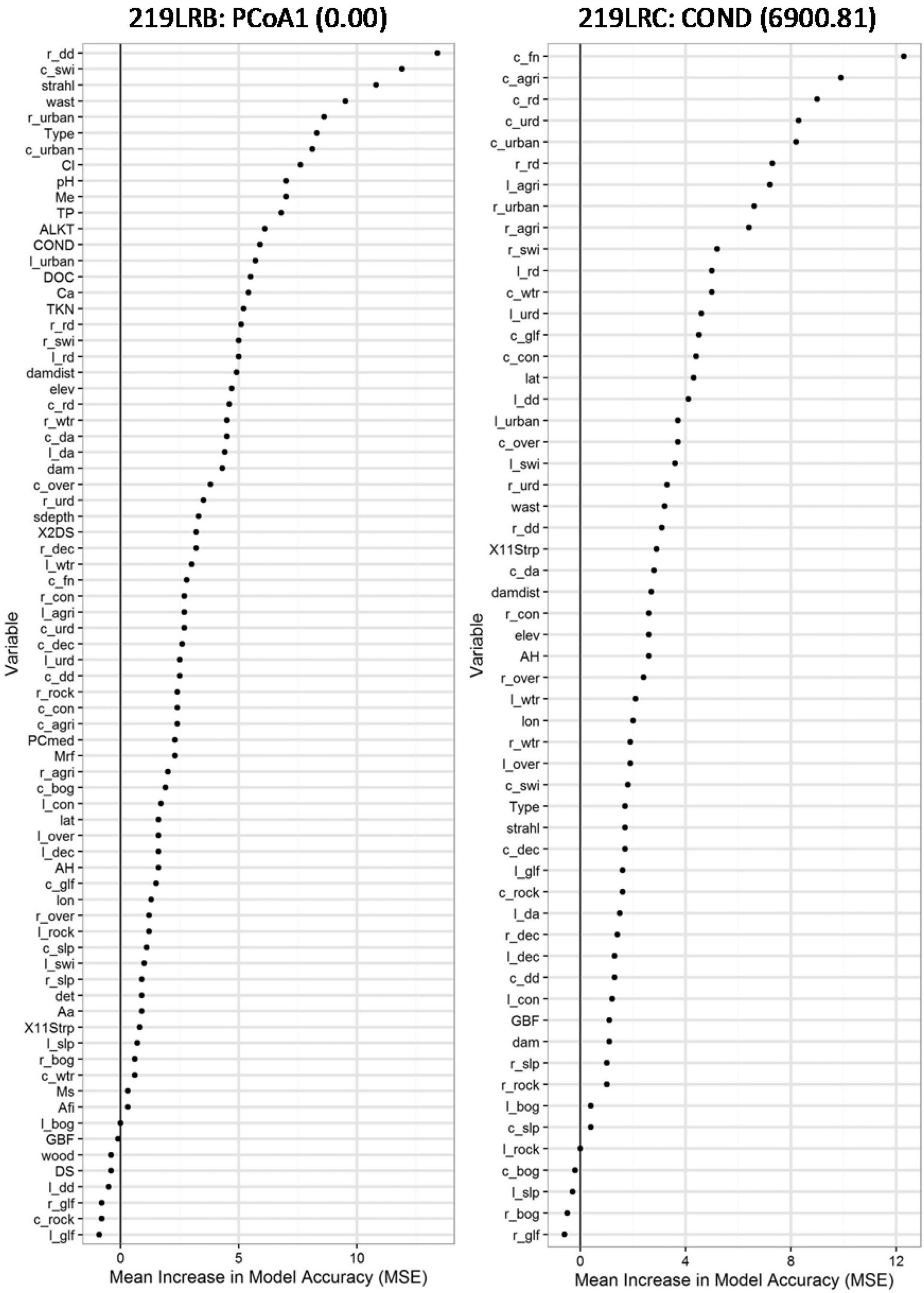

Fig. 8. Importances of predictors in the best random forest models (i.e., models with the highest pseudo-R^2) from the 219LRB and 219LRC datasets (model MSE's are given in parentheses). Each predictor's plotted point represents the percent change in MSE that resulted from its conditional permutation.

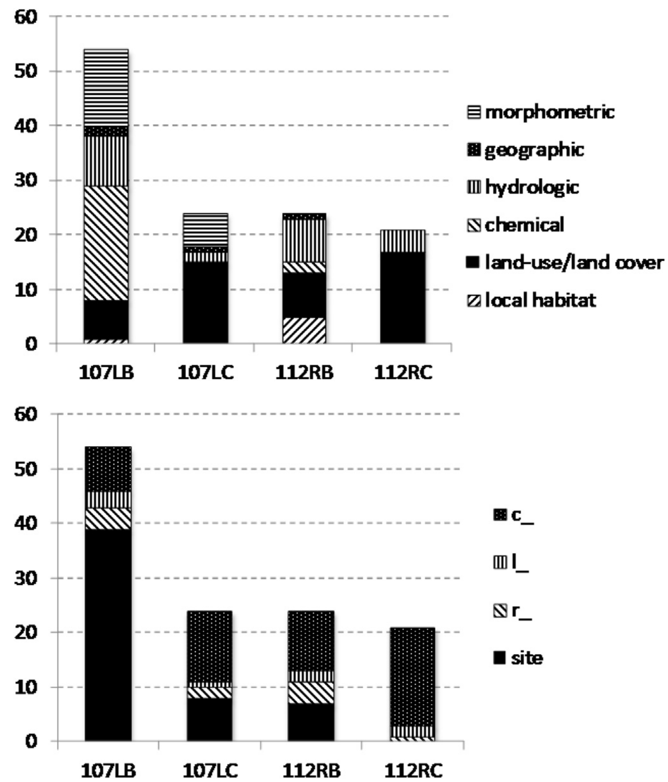

Fig. 9. Number of occurrences in which predictor classes (upper pane) or spatial scales of measurement (lower pane) were represented among models' three most important predictors. Plots summarize data only from random forest models having pseudo-$R^2 > 0.2$ (i.e., the 26 biological models and 15 chemical models included in Electronic Supplement #4).

partial dependencies describe sensitivity to stressors of interest, and identify natural sources of variation that also influence the indicator's value.

Our model for riverine Asellidae is a useful case in point. Predictive accuracy for this candidate indicator was moderate, given the random forest's pseudo-R^2 of 0.35, which indicated that the set of predictors included in the model accounted for 35% of the variation in the relative abundances of this taxon. Variable importances demonstrated this indicator to be sensitive to several land-use/land cover predictors that may be of interest to CEA (including coverage of forest or natural land cover in the cumulative catchment, urbanization in the riparian zone, and agriculture, road density and urbanization in the cumulative catchment); however, effect sizes observed across the Muskoka River Watershed's gradients of these land-use properties were relatively small (the land-use signal was small, relative to noise from natural variation), suggesting that it may be difficult to detect modest land-use changes with this indicator. Sources of natural variation, quantified by variable importances, included overburden thickness and soil wetness in the local catchment, and several interrelated chemical attributes of the river water (including calcium and chloride concentrations, conductivity, alkalinity, and pH). Controlling for these sources of natural variation when assessing or monitoring cumulative effects would maximize the indicator's signal and minimize noise due to natural variation (further discussion about indicator performance that is specific to the case study watershed is provided in Electronic Supplement #6).

4.5. Singular effects

Random forests illustrated a variety of singular biological and chemical effects in lakes and rivers. Many of these corroborate results of previous studies; others are potentially fruitful avenues for causal assessments. Our study demonstrated, for example, that the Chironomidae become more numerically dominant in lake samples as lake pH decreases, which reinforces results from earlier studies demonstrating the Chironomidae to be relatively insensitive to decreasing pH (Wiederholm and Eriksson, 1977) and to increase in abundance in acidified lakes (e.g., Schindler et al., 1985). Similarly, the abundances of aquatic earthworms (NHC) increased with increasing urban land-cover in the riparian zone (b_urban) — an effect of storm water and associated increased sedimentation? — and with increasing stream size (strahl) — which is expected under the river continuum concept (Vannote et al., 1980), given that larger rivers tend to have finer sediments and fine particulate organic matter that provides suitable habitat for these taxa (Verdonschot, 2001). Furthermore, we found conductivity, alkalinity, and chloride and calcium concentrations in lake and river water to increase with the intensity of human land-use in cumulative catchments, which is expected given the typical increase in solutes that accompanies many types of developments (Winter and Duthie, 1998; Paul and Meyer, 2001; Foley et al., 2005). Likewise, predictions of total phosphorus concentrations in lakes were best informed by drainage basin slopes (b_slope, l_slp) and agricultural intensity in the riparian zone (b_agri), which reinforces wetlands and farms as important nutrient sources (e.g., Sims et al., 1998; Reddy et al., 1999). Finally, given the well-known relationship between taxonomic richness and area (e.g., Palmer and White, 1994; Allen et al., 1999; Newman and Clements, 2008), and the correlation we observed between perimeter and shoreline development, we were unsurprised to find that lake perimeter strongly influenced the taxonomic richness of littoral lake benthic-invertebrates (perimeter was a more important predictor of richness than lake area was).

Given that random forests predict responses as step functions of the input predictors, it can be challenging to interpret the shape of partial dependencies as linear or non-linear. Notwithstanding this difficulty, many of the singular effects we observed appeared approximately linear (e.g., the singular effect of pH on Chir in lakes), but several appeared non-linear (e.g., the singular effect of c_dec on TKN in rivers; the effect of COND and pmtr on the percentage of Chironomidae in lake samples; and the relationships between zmean and zmax on lake DOC; Electronic Supplement #4).

Besides uncovering relationships between land-use and biological and chemical effects, our results contribute to landscape limnology's (e.g., Soranno et al., 2009) growing understanding of the natural environmental factors that control chemical and biological variation across time and space, and shed some light on how similar the drivers of chemistry and biology are for lakes and rivers, which are often studied in isolation (Lottig et al., 2011). For example, our identification of r_, l_ and c_slope, c_ and l_bog as key predictors in lake and river DOC models broadly agrees with various authors (e.g., Gergel et al., 1999; Mulholland, 2003) who reported that DOC concentrations in freshwaters largely derive from wetlands. Likewise, the importance of zmean, zmax and cwrt in our lake DOC model bolsters evidence (e.g., Hanson et al., 2007) of high rates of organic matter processing in lakes. Whereas Soranno et al. (2009) reported that differences in land cover or land-use had little effect on the alkalinity of Michigan lakes (which they reasoned are most responsive to geologic features), we found land-use to be the most important predictor of the alkalinity of Muskoka's soft-water lakes, presumably because human derived sources of base compounds represent a significant alkaline subsidy

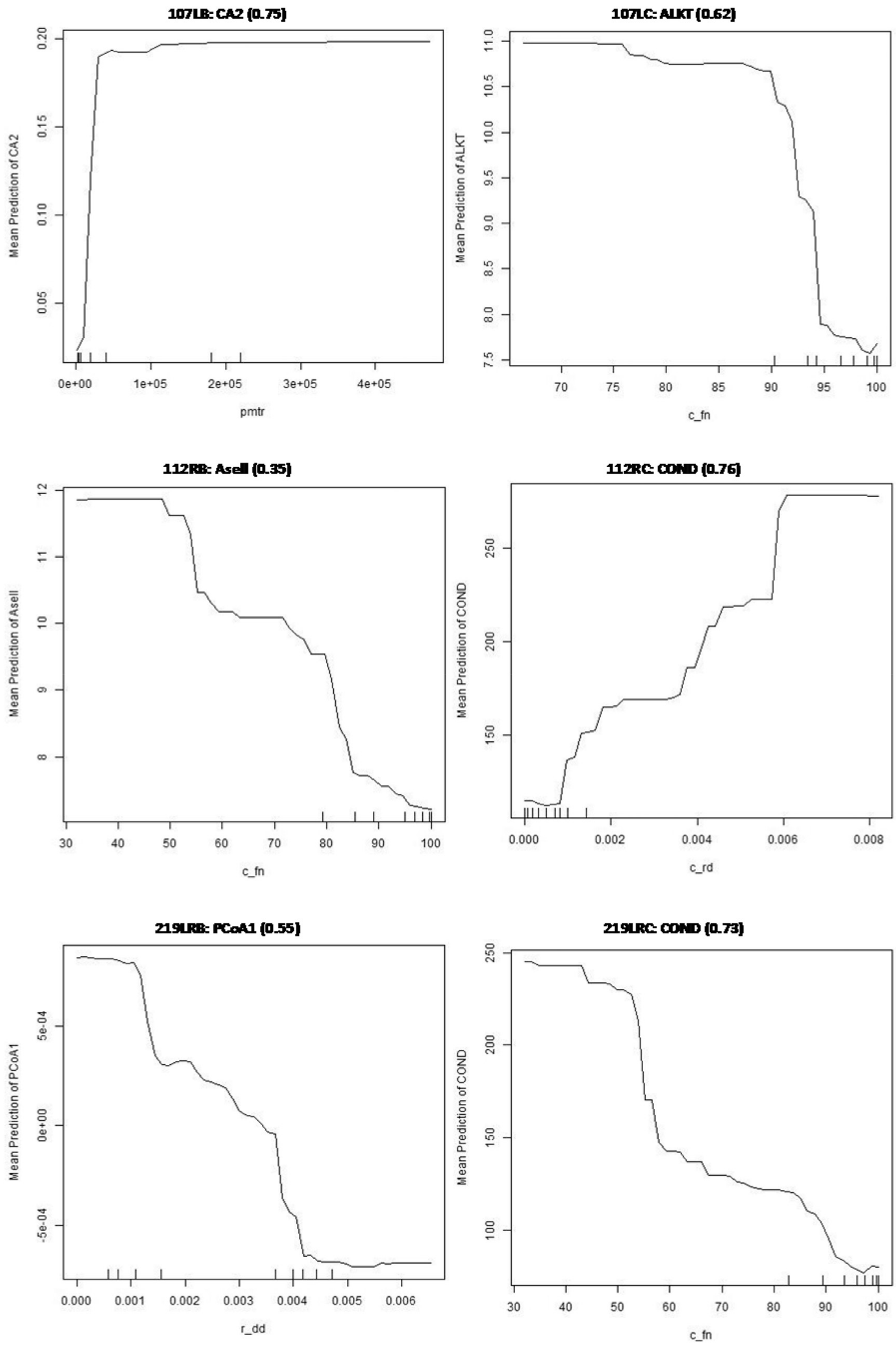

Fig. 10. Partial dependence plots describing singular effects of the most important predictors in the best random forest models (i.e., models with the highest pseudo-R^2 values) created from each of six datasets. Headers for each of the six panes in the figure describe the dataset and modeled Y, and include the model's pseudo-R^2 in parentheses. Hash marks along the horizontal axis mark deciles in the predictor's distribution.

Table 3
Effect sizes (ES) associated with the most important predictors (X) in the most accurate models (Y) yielded by each of six datasets.

Dataset	Y	X	ES as % of $Range_Y$
107LB	CA2	pmtr	10.9%
107LC	ALKT	c_fn	7.1%
112RB	CIGH	l_swi	7.2%
112RC	COND	c_rd	13.0%
219LRB	PCoA1	r_dd	7.2%
219LRC	COND	c_fn	13.7%

in our case-study watershed. Furthermore, Lottig et al. (2011) reported that median Ca concentrations in a series of streams in northern Wisconsin were approximately ten times higher than in lakes; whereas in the Muskoka River Watershed we found only a two-fold difference, possibly reflecting that region's shallower overburden, shorter flow paths, and lesser groundwater contact with calcareous bedrock.

The singular effects described in our study illustrate that algorithmic analytical methods (like random forests) can have a stimulatory effect on discovery-based and applied science, since the correlations and predictor interactions they uncover can be synthesized into provisional theories that are ripe for testing (Evans et al., 2011).

4.6. Model generality

Several authors (e.g., Breiman, 2001; Liaw and Wiener, 2002; Jones and Linder, 2015) have claimed that the random forest's bagging procedure (i.e., the process by which many trees are assembled from a dataset by bootstrapping) minimizes overfitting and results in the random forest having reasonable generality. In our study, the ($n = 37$) test datasets used in the cross-validation had similar pseudo-R^2 statistics as were generated from the ($n = 182$) training datasets, and correlations between observed and predicted biological and chemical values were stronger than would be expected from the random forests' pseudo-R^2 values. These results suggest that trained relationships between predictors and response variables hold for other lakes and rivers in the region that were not included in the training set, and cause us to uphold the generality of the random forest.

4.7. Detectability of simulated development

According to the results of our simulation, all tested biological indicators (Amph, Asell, CA1, CA2, Chir, CIGH, EPT, HBI, Insect, NHC, PCoA1, PCoA2, and Rich100) are capable of detecting a doubling of

Table 4
Singular effects of predictors having high variable importances in the most accurate random forest models, as characterized by partial dependence plots (↓ = "decreases", ↑ = "increases", * denotes non-linear effects). As an example of how to interpret the notation in the table, the fourth row from the top reads as — on average, the percent of a benthic invertebrate sample composed of Asellidae increases in lakes as r_dec decreases, as r_agri increases, and as elevation decreases; and increases in rivers as c_over increases, r_urban increases, and c_fn decreases. Refer to Electronic Supplement #4 for details about the shape of the Y responses across the observed ranges of the listed predictors. Abbreviations are defined in the Appendix.

Y	Singular Effect: Lakes (107LB, 107LC)	Singular Effect: Rivers (112RB, 112RC)
CA1↑	as ALKT↓, COND↓, Ca↓	PCmed↑, DS↑*, c_dec↑*
PCoA1↑	as pmtr↑*, strahl↑, pH↑	
Chir↑	as pmtr↓*, COND↓*, pH↓	
Asell↑	as r_dec↓, r_agri↑, elev↓*	as c_over↑, r_urban↑, c_fn↓
Insect↑	as pH↓*, ALKT↓, wast↓	as c_dec↑, Me↓, r_con↑
CIGH↑	as COND↑, pH↑, ALKT↓	as l_swi↑, r_urban↑, c_dec↓
Rich100↑	as COND↑, pmtr↑ *, ALKT↑	as c_da↑, ALKT↓, l_agri ↑
NHC↑	as r_urban↑*, pmtr↑*, damdist↑	
COND↑	as c_fn↓, c_rd↑, c_urban↑	as c_rd↑, c_fn↓, c_agri ↑
TKN↑	as zmean↓, zmax↓, l_wtr↓	as c_dec↓*, c_wtr↓, r_bog↑
Cl↑	as c_fn↓, c_agri↑, c_rd↑	as c_rdt, c_urban↑, c_fn↓
ALKT↑	as c_fn↓, c_agri↑, c_rd↑	as c_f n ↓ *, c_agri ↑, c_con ↓
Ca↑	as c_fn↓, c_urban↑, c_agri↑	as c_agri↑*, c_fn↓, l_agri↑
DOC↑	as zmean↓*, zmax↓*, wrt↓	
pH↑	as c_fn↓, elev↓*, r_rd↑*	as c_fn↓, c_agri↑*, l_rd↑
TP↑	as zmean↓*, zmax↓*, r_agri↑	

Table 5
Detectability of a simulated doubling of urban land-use in the Highway 11 Strip region, or doubling of agricultural land-use in the Muskoka River Watershed. Numbers in the 2XUrb and 2XAgri columns are counts of sites (lakes, rivers) for which MAD_{2XUrb} or MAD_{2XAgri} was greater than MAE_{test} (i.e., simulated effect size was greater than model error).

Model	pseudo-R^2	R^2	MAE_{test}	MAD_{2xUrb}	2XUrb ($n = 37$)	MAD_{2XAgri}	2XAgri ($n = 20$)
Asell	0.17	0.69	6.60 (3.20, 10.00)	1.84 (1.07, 2.61)	(10, 3)	1.29 (0.40, 2.19)	(2, 0)
CIGH	0.17	0.58	8.23 (4.83, 11.62)	2.05 (1.54, 2.56)	(8,2)	1.29 (0.37, 2.21)	(2, 1)
PCOA2	0.30	0.58	0.0059 (0.0040, 0.0077)	0.0019 (0.0012, 0.0026)	(8, 1)	0.0008 (0.0005, 0.0011)	(1, 1)
CA2	0.47	0.70	0.146 (0.110, 0.182)	0.041 (0.029, 0.052)	(4, 2)	0.004 (0.003, 0.006)	(0, 0)
NHC	0.26	0.31	5.41 (3.78, 7.04)	1.25 (0.88, 1.62)	(5, 1)	0.15 (0.07, 0.23)	(0, 0)
PCoA1	0.49	0.53	0.00313 (0.00227, 0.00399)	0.00095 (0.00055, 0.00135)	(5, 0)	0.00006 (0.00004, 0.00009)	(1, 0)
Amph	0.37	0.45	6.30 (3.98, 8.63)	1.53 (1.03, 2.04)	(3, 2)	0.22 (0.10, 0.34)	(0, 0)
Chir	0.35	0.42	12.79 (9.72, 15.85)	3.25 (2.04, 4.47)	(5, 0)	0.38 (0.22, 0.54)	(0, 0)
Insect	0.44	0.46	13.77 (9.98, 17.56)	2.96 (1.93, 3.99)	(4, 0)	0.47 (0.25, 0.70)	(0, 0)
EPT	0.20	0.13	5.24 (4.08, 6.41)	0.39 (0.25, 0.53)	(3, 1)	0.22 (0.11, 0.33)	(1, 1)
HBI	0.20	0.33	0.39 (0.31, 0.47)	0.03 (0.02, 0.03)	(1, 1)	0.01 (0.01, 0.02)	(0, 0)
CA1	0.40	0.70	0.139 (0.094, 0.185)	0.015 (0.011, 0.019)	(1, 0)	0.005 (0.003, 0.007)	(0, 0)
Rich100	0.27	0.45	2.05 (1.59, 2.51)	0.28 (0.2, 0.35)	(1, 0)	0.1 (0.07, 0.14)	(0, 0)

urbanization in the catchment. Five of thirteen (Asell, CIGH, EPT, PCoA1, and PCoA2) are capable of detecting a doubling of agricultural intensity in the Highway 11 Strip physiographic region. All indicators except HBI are better able to detect urbanization or agricultural effects in lakes than in streams. The simulation suggests that modest changes to urban land-use in the Muskoka River Watershed would have more significant ecological consequences than would result from similar changes to agricultural land-use. The fact that chemical, and the more promising biological indicators, are sensitive enough to detect moderate land-use changes should give some confidence to practitioners that they can be used in watershed monitoring and CEA scenario analyses.

4.8. Future work

Although rarely addressed in research on machine learning techniques (i.e., Evans et al., 2011), opportunities exist to build more parsimonious models, which have less onerous data requirements (a benefit to cash-strapped resource management agencies), are more easily interpreted, and can even have greater predictive accuracies than result from models with larger numbers of predictors (e.g., Evans et al., 2011). We encourage future work that incorporates the most important predictors into simplified empirical models. Ultimately, attempts should be made to develop mechanistic models that causally link predictors and response variables.

4.9. Summary (key messages for environmental managers)

Cumulative Effects Assessment has potential as a tool for managing sustainability (Jones, 2016), but this potential can only be realized if scientists, environmental managers, and planners work together using effective techniques. We encourage these professionals to consider our three key findings:

1. Random forest models describe the combined and singular effects of multiple stressors and natural factors, and can be used to evaluate the suitability of candidate indicators for use in monitoring schemes and scenario analyses.
2. A variety of chemical and biological measures of ecosystem condition can be modeled accurately enough, and are sensitive enough to land-use, to be used in watershed-scale cumulative effects assessment.
3. The analytical methods presented in this paper are equally applicable to other ecosystems and indicator types

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Appendix A. Supplementary data

Supplementary data related to this article can be found at http://dx.doi.org/10.1016/j.jenvman.2017.06.011.

Appendix. Abbreviations and acronyms

Prefixes

c_ denotes that a given predictor has been calculated for the cumulative catchment area (i.e., the entire catchment that drains to the point of interest)

l_ denotes that a given predictor has been calculated for the local catchment area (i.e., the part of the cumulative catchment that extends upstream from the point of interest to the nearest upstream lake)

r_denotes that a given predictor has been calculated for the riparian buffer associated with the point of interest (i.e., that part of the cumulative catchment area that includes only areas within 300 m of the lake or stream of interest and its contributing drainage network).

Abbreviations and acronyms

2XAgri: dataset simulating doubling of agricultural land-use for sites in the 37LRBTest dataset lying within the Highway 11 Strip physiographic region ($n = 20$)

2XUrb: dataset simulating doubling of urban land-use for lakes and rivers in the 37LRBTest dataset

37LRBTest: 37 Lakes or Rivers Biology Test Dataset

37LRCTest: 37 Lakes or Rivers Chemistry Test Dataset

107LB: 107 Lakes Biology Dataset

107LC: 107 Lakes Chemistry Dataset

112RB: 112 Rivers Biology Dataset

112RC: 112 Rivers Chemistry Dataset

182LRBTrain: 182 Lakes or Rivers Biology Training Dataset

182LRCTrain: 182 Lakes or Rivers Chemistry Training Dataset

219LRB: 219 Lakes or Rivers Biology Dataset

219LRC: 219 Lakes or Rivers Chemistry Dataset

2DS: second dominant inorganic pavement-layer particle type (ordinal)

Aa: abundance of attached algae (ordinal)

Afi: abundance of filamentous algae (ordinal)

agri: percent of area as agricultural land-use

area: lake area (m^2)

AH: percent of cumulative catchment area in Chapman and Putnam's (1984) Algonquin Highlands physiographic region

ALKT: Alkalinity, total fixed endpoint (mg/L as $CaCO_3$)

Amph: percent of sample abundance accounted for by Amphipoda

Asell: percent of sample abundance accounted for by Asellidae

bog: percent of area as bog

Bw: Percent of sample abundance accounted for by animals that burrow in substrates

Ca: Calcium, unfiltered total (mg/L)

CA1: Axis-1 score from correspondence analysis of samples, based on their taxa abundances

CA2: Axis-2 score from correspondence analysis of samples, based on their taxa abundances

CART: Classification and Regression Trees

CEA: cumulative effects assessment

Chir: percent of sample abundance accounted for by Chironomidae

CIGH: percent of sample abundance accounted for by Corixidae, Isopoda, Gastropoda, and Hirudinea combined

Cl: Chloride (mg/L)
con: percent of area as coniferous forest
COND: conductivity (μS/cm)
CSL: Candidate Sampling Location
cwrt: cumulative water residence time (days)
da: drainage or catchment area (m^2)
dam: number of dams
damdist: cumulative distance to all dams in the cumulative catchment
dd: drainage density (m^{-1}; stream length [m]/cuca [m^2])
dec: percent of area as deciduous forest
det: coverage of detritus (ordinal)
DOC: dissolved organic carbon (mg/L)
DR: lake drainage ratio (dimensionless)
DS: dominant inorganic pavement-layer particle type (ordinal)
elev: elevation (m above sea level)
EOT: percent of sample made up of Ephemeroptera, Odonata and Trichoptera taxa combined
EPT: percent of sample made up of Ephemeroptera, Plecoptera and Trichoptera taxa combined
ES: effect size; the minimum and maximum predicted values of Y that occur across the observed range of X (estimated by conditional permutation of X)
FC: percent of sample made up of filtering collectors
fn: percent of cumulative catchment under forest or other natural land cover
GBF: percent of cumulative catchment area in Chapman and Putnam's (1984) Georgian Bay Fringe physiographic region
GC: percent of sample made up of gathering collectors
glf: percent of area maintained as golf course
HBI: Hilsenhoff's family-level biotic index (dimensionless; Hilsenhoff, 1988)
Insect: percent of sample made up of insect taxa
lat: latitude (decimal degrees)
lon: longitude (decimal degrees)
LNN: lake network number (dimensionless)
MAE_{test}: mean absolute error (i.e., mean absolute error of predicted and observed values) of random forest predictions for the test dataset (37BTest or 37CTest)
MAD_{2XAgri}: mean absolute difference of predicted values of 37BTest, relative to predicted values for 2XAgri scenario (a measure of scenario effect size)
MAD_{2XUrb}: mean absolute difference of predicted values of 37BTest, relative to predicted values for 2XUrb scenario (a measure of scenario effect size)
Me: abundance of emergent macrophytes (ordinal)
min: minimum value
max: maximum value
Mrf: abundance of rooted floating macrophytes (ordinal)
Ms: abundance of submerged macrophytes (ordinal)
MSE: mean square error
NHC: percent of sample made up aquatic earthworms (non-hirudinean Clitellata)
over: overburden thickness (depth of inorganic materials deposited over bedrock; m)
P: percent of sample made up of predators
PCmed: median axis dimension of randomly selected substrate particles (mm)
PCoA1: Axis-1 score from principal coordinates analysis of samples, based on Bray-Curtis distances among their taxa counts
PCoA2: Axis-2 score from principal coordinates analysis of samples, based on Bray-Curtis distances among their taxa counts
pmtr: lake perimeter (m)
R^2: coefficient of determination (dimensionless)
$range_Y$: predictor range; the range of a given predictor
Rich100: taxonomic richness (the number of unique taxa collected), standardized to 100-count sample using rarefaction
rd: paved road density (m/m^2)
rock: percent of area as exposed rock
SC: percent of sample made up of scrapers
sd: standard deviation
sdepth: maximum water depth sampled
SH: percent of sample made up of shredders
swi: soil wetness index, a function of channel slope and drainage area (dimensionless; Plewes, 2015)
slp: slope of flow path (dimensionless; over-land flow distance [m]/vertical distance [m] separating starting and ending nodes; Plewes, 2015)
strahl: Strahler Order (dimensionless; Strahler, 1957; Plewes, 2015)
TKN: total Kjeldahl Nitrogen (μg/L)
TP: total phosphorus (μg/L)
urban: percent of area urbanized
urd: unpaved road density (m/m^2)
Var: variance
vol: lake volume (m^3)
wast: number of waste disposal sites
wood: coveage of woody material (ordinal)
wrt: water residence time (days)
wtr: percent of area as water
X: a predictor variable
X11Strp: percent of cumulative catchment area in Chapman and Putnam's (1984) Highway 11 Strip physiographic region
Y: a response variable
zmax: maximum lake depth (m)
zmean: mean lake depth (m)

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PART 4: ONSET-OF-EFFECT THRESHOLDS AND REFERENCE CONDITIONS — A CASE STUDY OF THE MUSKOKA RIVER WATERSHED, CANADA

ONSET-OF-EFFECT THRESHOLDS AND REFERENCE CONDITIONS: A CASE STUDY OF THE MUSKOKA RIVER WATERSHED, CANADA

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Abstract

An extensive survey of lake and stream water chemistry and benthic macroinvertebrate communities was undertaken throughout the 5660 km^2 Muskoka River Watershed (Canada) to identify onset-of-effect thresholds in gradients of exposure to roads, urbanization, and agriculture. Two complementary statistical methods (partial dependence plots from random forests and TITAN [threshold indicator taxa analysis]) were used. No biological thresholds were observed in either lakes or streams; however, lake-chemistry thresholds for pH, alkalinity, conductivity, Ca, Cl, DOC, total Kjeldahl N, and total P were detected, as were stream-chemistry thresholds for alkalinity, conductivity and Ca and Cl concentrations. These thresholds — i.e., break-points where stressors begin to override natural biogeochemical processes — were used to make empirical adjustments to the criteria used to distinguish reference and impacted waterbodies. Impacted waterbodies had more variable taxonomic structure and water chemistry, and had higher alkalinities and conductivities, Ca, Cl, and phosphorus concentrations, and pH than reference waterbodies. Amphipoda, Asellidae, Corixidae, Gastropoda, and Hirudinea were more abundant, while Chironomidae were less abundant in impacted streams than in reference streams; and insects were less abundant in impacted lakes than in reference lakes. This study presents an objective and transferrable approach for distinguishing reference and impacted locations, proposes quantitative decision criteria for threshold identification in TITAN, and illustrates how a non-parametric method can be used to calculate central 95%/95% tolerance intervals for limnological parameters.

Keywords: onset-of-effect thresholds, reference criteria, biocriteria, water quality, biological condition

Introduction

Many approaches to ecosystem management, monitoring, and assessment make use of biological or chemical thresholds to evaluate the impact of environmental stressors (Groffman et al. 2006). For example, thresholds are used as interpretive criteria for various indicators of ecosystem condition (Roubeix et al. 2016, Samhouri et al. 2017), to establish regulatory standards (e.g., Brenden et. al 2008), restoration targets (White and Walker 1997, Roubeix et al. 2016) and reference conditions (Wang et al. 2006, Baker and King 2010, Clapcott et al. 2017). Stoddard et al. (2006) distinguished two different kinds of reference conditions: the minimally disturbed reference condition, which they defined as "the biological condition that exists in the absence of significant human disturbance" (i.e., the near pristine condition) and the least disturbed reference condition, which they defined as the biological condition associated with least exposure to stress and "best" available habitat (the least disturbed condition being more commonly used as an assessment benchmark).

Quantitative criteria based on measured exposures to stress are generally used to define what qualifies as a least-disturbed reference site; however few studies have specifically evaluated the level of stressor exposure that is acceptable, i.e. the level of stressor exposure at which biological effects become apparent (Pardo et al. 2012). Such onset-of-effect thresholds have been hypothesized (Hilderbrand et al. 2010, Pardo et al. 2012), and can be used to objectively delineate the least-disturbed reference condition (Ciborowski et al. 2015), but there is little empirical evidence to support their existence (Pardo et al. 2012).

Because the reference condition describes a distribution, not a single value (Stoddard et al. 2006), reference sites are used as replicates (Reynoldson et al. 1997) that collectively describe the "normal" range of variation (e.g., Kilgour et al. 1998, Kilgour et al. 2017). Numerical standards for assessment indicators, against which ecological condition is interpreted (i.e., biocriteria; Seegert 2000), can be derived from this normal range (Kilgour et al. 2017).

The Muskoka River Watershed is a 5660 km^2 catchment that drains to Lake Huron, one of North America's Laurentian Great Lakes (Figure 1). A long-term program, in which benthic macroinvertebrates are used as indicators to monitor the biological condition of this region's lakes and streams, has been underway for more than 20 years (e.g., David et al. 1998, Bowman et al. 2006), and the Muskoka Watershed Council publishes semi-annual report cards on watershed "health" (Muskoka Watershed Council 2017); however, biological and chemical reference conditions for the watershed's lakes and streams have not been characterized. The Watershed has a human population of approximately 62,000 (DMM 2016 [2015 estimate]), but much of the development is concentrated in three towns (Huntsville, Bracebridge, and Gravenhurst), leaving much of its area with no measurable exposures to land-use stress. The Watershed's is unusual in southern Canada, because minimally impacted reference sites can still be found, and this fact makes it an appropriate region in which to investigate onset-of-effect thresholds.

We characterized benthic community structure and water chemistry in a variety of lakes and streams that had no measurable stressor exposures, and in a series of impacted lakes and streams. The resulting dataset was used to answer the following questions:

1) Are there clear biological or chemical onset-of-effect thresholds that can be used to distinguish reference from impacted conditions?
2) Based on a survey of reference sites, what are the normal ranges of chemical and biological conditions for lakes and streams in the Muskoka River Watershed?
3) Are cumulative effects of land-use evident in the Watershed's lakes and streams?

Methods

<u>Survey Design and Sampling Methods</u>

We consulted government datasets (e.g., OMNR 2011a, 2012) to create a list of lakes and candidate stream sampling locations in the Muskoka River Watershed. "Lake" was defined as any waterbody having an outlet stream and an area of at least 8 ha. The individual basins of large multi-basin lakes (e.g., the 12,300-ha Lake Muskoka) were considered separate lakes. To ensure that our sampling design included a practical mix of accessible and remote locations, candidate stream sampling locations (CSL's) were identified on 2^{nd}-to 6^{th}-order watercourses, as stream-road intersections (accessible) and as tributary confluences (potentially remote).

Using methods detailed by Plewes (2015), we calculated a series of predictors to describe the morphometric and hydrologic (OMNR 2011a and 2011b), physiographic (OMNDM 2012), and land-use/land-cover attributes (OMNR 2015) of the resulting 647 lakes and 1879 CSL's, their cumulative catchment areas, and local catchment areas. By cumulative catchment area, we mean the gross drainage area contributing to a specific CSL or lake outlet (variables measured at this scale are denoted by the prefix "c_"). Local catchment areas were defined as those parts of cumulative catchments that were adjacent to the lake or CSL in question but extended only up to, and not beyond, any upstream lakes (variables measured at the local catchment scale are denoted by the prefix "l_"). Morphometric predictors were quantified for lakes only, and included lake area ("area"; measured in m^2), perimeter (pmtr; measured in m), maximum depth (zmax; measured in m), mean depth (zmean; measured in m), and volume (vol; measured in m^3). Hydrologic predictors (some of which were proposed by Martin et al. 2011) included the mean slope of the drainage network (c_slp, l_slp), Strahler order (strahl; Strahler 1957), drainage area (c_da, l_da; measured in m^2), drainage ratio (DR [lakes only]: the ratio of the c_da to lake area), water residence time (wrt; lakes only: the mean length of time water stays in a lake; measured in days), cumulative water residence time (cwrt; lakes only: the average length of time water in a given lake will stay in lakes, including time spent in upstream lakes; measured in days; Müller et al. 2013), and lake network number (LNN; the number of lakes in the lake of interest's lake chain [headwater lakes have LNN = 1]). Physiographic predictors included the latitude (lat), longitude (lon), and elevation (elev; measured in m above sea level) of the sampled locations, and mean overburden thickness in the c_da (c_over; measured in m). Land-use predictors included areal percentages of catchments urbanized (c_urban, l_urban), under agricultural land-use (c_agri, l_agri), or maintained as golf course (c_glf, l_glf). Un-paved (c_urd, l_urd) and paved (c_rd, l_rd) road densities (measured as m/m^2) were calculated, as were the number of municipal waste disposal sites (wast) and dams (dam) in c_da. The percentage of forest and other natural land-covers in the c_da was also determined (c_fn).

For the purposes of selecting waterbodies to be sampled, we operationally defined reference lakes and streams as waterbodies having no measureable road density, development, agriculture, golf courses or waste disposal facilities in their catchments (OME 2012 or OMNR 2006a), and no dams (OMNR 2006b). Waterbodies not satisfying these criteria were considered impacted.

All threshold-identification methods are sensitive to the distributions of sampled locations across the stressor gradient(s) of interest, and long gradients of stressor exposure, or concentrations of sites at high levels of impact could "obscure sharp, nonlinear patterns at low levels of the gradient" (King and Baker 2014). Given our interest in onset-of-effect thresholds, it was critical in our design to adequately represent minimally impacted conditions and sites with low levels of stressor exposure. We used a stratified random procedure to proportionally select 84 lakes and 112 streams by size: 26 minimally impacted, and 58 impacted lakes; 19 minimally impacted and 93 impacted streams (Figure 1). The lake-size stratum was area-based (bins were <10 ha, 10-100 ha, and >100 ha), and the stream-size stratum was Strahler-based (bins were 2nd – 3rd, 4th–5th, and 6th order). In addition to the randomly selected ones, the Watershed's five largest lakes (23 lake basins) — each having extensively developed shorelines and a special cultural and economic importance to the region — were also included in the study: Kawagama Lake (3 basins), Lake Joseph (6 basins), Lake Rosseau (5 basins), Lake of Bays (6 basins), and Lake Muskoka (5 basins). Including these large impacted lakes brought the total number of sampled lakes to 107 (Figure 1).

<<INSERT FIGURE 1 HERE>>

Benthic invertebrates were sampled from streams and lakes using the kick-and-sweep method of Jones et al. (2007). Three separate collections were made at each CSL and in each lake. Stream samples were collected along bank-to-bank transects, that were oriented perpendicular to the direction of flow. Lake sampling locations were determined by boating to the approximate centre of each lake's basin. From this central location, a random 3 of the 4 cardinal compass bearings (i.e., north, south, east, west) were sighted toward shore to determine the shoreline locations where invertebrate samples were collected. If the area where the compass bearing intersected the shoreline was not wadeable or was otherwise inaccessible, an alternative location, immediately adjacent to this intersection point, was selected (MacDougall et al. 2017). The dominant substrate particle type (DS) at each sampled location was recorded, as per Jones et al. (2007).

In streams, a single water sample was collected at each CSL. In lakes, one litre of water was collected at each sampling location, and these were combined into a 3-L composite, from which water was decanted into bottles for submission to the lab. Water samples were kept on ice or refrigerated until they were submitted to the laboratory (i.e., the Dorset Environmental Science Centre), which was done within 2 days of their collection. Assays were performed (as per OME 1983) for alkalinity (ALKT; mg/l as $CaCO_3$), calcium (Ca; unfiltered total, in mg/L), chloride (Cl; mg/l), conductivity (COND; µS/cm), dissolved organic carbon (DOC; mg/l), total Kjeldahl nitrogen (TKN; µg/l), pH, and total phosphorus (TP; µg/l).

Collected invertebrates were preserved (in alcohol) in the field, and then in the lab were randomly sub-sampled to obtain a minimum of 100 collected specimens[9], which were considered to characterize relative abundances of the different taxa that comprised the community at each sampled location. Randomizations were performed with the aid of a 100-cell Marchant-style (e.g., Marchant 1989) sub-sampling box. Invertebrates were searched from among collected sediments, identified and enumerated with the aid of a stereomicroscope (the magnification used depended on particle sizes of the collected sediments). The specified taxonomic precision was a "mixed-level taxonomic aggregation" (Jones 2008) in which insects, crustaceans, molluscs, and leeches were diagnosed as families, with the exception of the Chironomidae, which were assigned to their sub-families, and the Coelenterata, Platyhelminthes, Nemata, Hydrachnidia, and oligochaetous clitellata, which were assigned only to these coarse taxonomic ranks. All identifications were made by taxonomists certified under the Society for Freshwater Science's Taxonomic Certification Program.

Threshold Detection

Gradients of exposure to land-use stress were represented by the ranges of c_- and l_agri, c_- and l_urban, c_- and l_rd. We were principally interested in the existence of onset-of-effect thresholds in the least impacted tails of these variables, which could be interpreted as empirical breakpoints between reference and impacted conditions. Given that there is no single ideal threshold-detection method (King and Baker 2014), we conducted two complementary types of analyses: (1) we ran a series of random forest (Breiman 2001) models, and inspected the associated partial dependence plots for non-linearities in the modeled relative abundances of benthic taxa and the

[9] Aliquots containing the 100th animal were entirely searched, meaning that abundances for each sample were generally higher than 100

modeled values of chemical indicators that occurred across the ranges of the six stressor variables; (2) we used Threshold Indicator Taxa Analysis (TITAN; Baker and King 2010) to detect thresholds in the relative abundances and occurrence frequencies of the benthic taxa that comprised the lake and stream communities.

Similar to multiple regression, random forests are multivariate with respect to X but univariate with respect to Y (i.e., they predict the value of a single response variable according to the values of multiple predictors [Breiman 2001]). Partial dependence plots illustrate the mean predicted value of Y across the observed range of a single X, given the averaged effects of the other predictors included in the model (Cutler et al. 2007). In contrast to random forest, TITAN is multivariate with respect to Y, but univariate with respect to X (Baker and King 2010). We used it to identify the break-point in the range of X for which the strongest threshold change in occurrence frequencies or relative abundances was observed among the sampled communities' multiple taxa. TITAN identifies this break-point as the point on X that creates the two-group partition of the observations (samples) that has the maximum sum of taxon-specific z-transformed IndVal (Dufrene and Legendre 1997) scores[10]. In this process, taxa having abundances that increase as X increases (i.e., Z+ taxa) and taxa having abundances that decrease as X increases (Z- taxa) are considered separately. Predictor values that result in optimal IndVal scores are referred to as taxa-specific change-points, and the predictor value at which there is maximal synchrony among taxa-specific change-points is referred to as the community change-point. The algorithm includes a bootstrapping procedure that allows each taxon's "reliability"[11] and "purity"[12] to be calculated, and quantifies uncertainty around the predictor value where taxa-specific thresholds and the whole-community threshold occur (Baker and King 2010).

TITAN identifies a community threshold by considering each taxon's pattern of abundances and occurrence frequencies (Baker and King 2010, Roubeix et al. 2016); however, it can explore only a single stressor gradient. This can be problematic given that our ability to infer thresholds from single-stressor models can be confounded by interactions among stressors or between stressors and natural environmental variables (Wagenhoff et al. 2017). On the other hand, random forests allow multiple stressors and synergisms to be accounted for (Pardo et

[10] The z-score transformation is calculated using the mean and standard deviation of samples permuted with respect to X. It increases information contributed by taxa having low occurrence frequencies but high sensitivity to X; therefore, emphasizes the relative magnitude and synchrony of taxa-specific threshold effects (Baker and King 2010).
[11] Reliability is measured as the proportion of bootstrapped taxa-specific change-points that are significant at a user-defined critical p-value (Baker and King 2010).
[12] Purity is measured as the proportion of bootstrapped replicates in which the Z+ or Z- response direction agrees with the observed response; therefore, a pure taxon consistently exhibits the same Z+ or Z- response, regardless of its abundance or the bootstrap's frequency distribution (Baker and King 2010).

al. 2012); but they require taxa to be either modeled individually (making the concept of a community-level threshold difficult to operationalize) or aggregated into an index of community structure (which requires generalities or assumptions to be made about taxa-specific responses, and reduces the dataset's ecological information content; King and Baker 2014)

Random forest models were created independently for chemical and biological indicators, and for lakes and streams, one random forest model for each combination of waterbody type and indicator. This resulted in a total of 46 lake models and 45 stream models. We refer to the input datasets as follows (Online Resource 1): LXRFB (lake predictors, random forest, biological: 34 X, n = 107); LYRFB (lake responses, random forest, biological: 38 Y, n = 107); LXRFC (lake predictors, random forest, chemical: 26 X, n = 107); LYRFC (lake responses, random forest chemical: 8 Y, n = 107); SXRFB (stream predictors, random forest, biological: 25 X, n = 112); SYRFB (stream responses, random forest, biological: 37 Y, n = 112); SXRFC (stream predictors, random forest, chemical: 17 X, n = 112); SYRFC (stream responses, random forest chemical: 8 Y, n = 112). Y variables (taxa abundances or chemical analytes) were only assessed for threshold effects if their respective random forests had pseudo-R^2 values ≥ 0.3.

Random forests and partial dependence plots were created using randomforest() and partialPlot() functions from the extendedForest package (Smith et al. 2014) for the R language and environment for statistical computing (R Core Team 2016). Default values for "mtry" and "ntree" arguments of the randomForest() function were called, such that the number of candidate splitting variables considered at each node was equal to 1/3 the number of predictors, and the forest contained 500 trees (Online Resource 2).

TITAN analyses were conducted independently for lakes and streams, one TITAN analysis for each predictor of interest (one each for c_agri, c_rd, c_urban, l_agri, l_rd, l_urban). All lake analyses considered relative abundances of the same 38 taxa, and all stream analyses considered relative abundances of the same 37 taxa. We refer to the input datasets as follows (Online Resource 1): LXT (lake predictors, TITAN: 6 X, n = 107); LYT (lake responses, TITAN: 38 Y, n = 107); SXT (stream predictors, TITAN: 6 X, n = 112); SYT (stream responses, TITAN: 37 Y, n = 112).

We conducted the TITAN analyses using the titan() function in the TITAN2 package (Baker et al. 2015) for R (R Core Team 2016). The function's arguments were set to enforce a minimum group size of 5 during the partitioning routine (i.e., minSplt=5), 250 permutation replicates (numPerm=250) and 500 replicates of bootstrap resampling (nBoot=500), with taxon-specific change-points determined as z-score maxima (imax=FALSE), IndVal scores based on mean relative abundances (ivTot=FALSE), and the taxon purity threshold set at 95% (pur.cut=0.95). As suggested by Brenden et al (2008) and Dodds et al. (2010) graphical methods are useful for evaluating how appropriate it is to designate a response as a threshold effect. Two of TITAN's standard diagnostic plots are particularly useful in this regard: plots of the sum of taxon-specific Z+ and Z- scores (i.e., SumZ values) occurring at each candidate change-point along the gradient of X provided evidence about the strength and position of a community change-point; and plots of the positions of Z+ and Z- taxon-specific change points along X provided evidence about how synchronous taxa-specific change points were with the community change point. Plots of SumZ values were created using the plotSumz() function in the TITAN2 package (Baker et al. 2015) for R (R Core Team 2016), with arguments set to include only pure and reliable taxa (filter=TRUE), and with cumulative frequencies of sumZ maxima from bootstrap replicates shown (cumfrq=T). Plots of taxon-specific change-points were created using the plotTaxa() function in the TITAN2 package (Baker et al. 2015) for R (R Core Team 2016), with quantiles of bootstrapped change-points[13] plotted (interval=T), and with change-points plotted as observed values (prob95=F).

Our decisions about whether a chemical or biological threshold existed, and where that threshold was positioned in the range of the predictor, was based on the following line of evidence from the random forest:

1. The empirical "dose-response", which described (by visual inspection) the nature of any non-linearities in the Y response that occurred across the X gradient, the position of these non-linearities in the range of X, and their congruence among taxa or chemical analytes.

For the biological Ys, the following eight additional lines of evidence from TITAN were also considered (as suggested by Baker and King 2010):

1. "MaxSum Z" (the maximum value of the SumZ+/SumZ- trace along X, divided by the number of pure and reliable taxa): a measure of the degree of change in community structure that occurs at the community change-point, measured as the mean contribution to SumZ made by each pure and reliable taxon.
2. "SumZ Peak" (strength of the peaks of the SumZ+/ SumZ- traces along X): evaluated visually by considering the slope of the ascending and descending curves on either side of maxSumZ, and the number of pure and reliable taxa associated with any secondary peaks.

[13] Given reasonable sample sizes, these quantiles approximate 95% confidence intervals, and are referred to from this point forward as 95% CIs.

3. “CFDW” (cumulative frequency distribution width): the width of the steeply ascending region of the SumZ+/SumZ- cumulative frequency distribution, scaled as a proportion of the range of X.
4. “#ST” (number of synchronous taxa): the number of taxa having 95% CIs describing their taxon-specific change-points that include the X-value of the community change point.
5. “TSCPCIW” (taxon-specific change-point confidence interval width): The median width of the 95% taxon-specific change-point CIs, scaled as a proportion of the range of X (measures the degree of uncertainty about taxon-specific change-points).
6. “CCPCIW” (community change point confidence interval width): The median width of the 95% community change-point CI, scaled as a proportion of the range of X (a measure of uncertainty concerning the position of the community change-point in the X range).
7. “PRT” (the proportion of taxa that were pure and reliable).

“Thresholds can be more or less broad in a gradient, depending on the variability in response among taxa and uncertainty associated with detection methods” (Roubeix et al 2016). We derived a mix of qualitative and quantitative criteria by which the strength of the above lines of evidence could be judged. These decision rules enabled us to make conclusions about the existence of thresholds in a standardized way. Visual (qualitative) assessments of the shape of the indicators’ partial dependencies on the stressor variables were used to evaluate evidence from the random forest models. TITAN was deemed to indicate a community threshold only if the following four (quantitative) conditions were met: (1) its MaxSum Z was greater than 5 (see also Baker and King [2010] who interpreted the existence of thresholds with MaxSumZ between 4.5 and 13, and Wallace and Biastoch [2016] who considered a MaxSumZ of 10.8 to signal a threshold) ; (2) Its CFDW was less than 11%; (3) At least 4 taxa responded purely and reliably to the stressor of interest (PRT $\geq$ 4); and (4) #ST was greater than 3, and these synchronous taxa included at least 25% of the pure and reliable taxa.

Normal ranges (biocriteria)

Kilgour et al. (2017) defined the normal range as “a part of a reference distribution” (commonly the central 95%; Kilgour et al. 1998, Barrett et al. 2015) “deemed to represent an expected condition”. These authors argued that normal ranges are appropriate as generic biocriteria because they are conceptually intuitive, statistically defined (meaning that they constitute a rigorous foundation for bioassessments), and applicable to any response variable and many study designs.

Considering the random forest and TITAN evidence about the existence of thresholds, we assigned all sampling locations to one of 4 groups (LRef [reference lakes], LImp [impacted lakes], SRef [reference streams], and SImp [impacted streams]; Online Resource 1), from which normal reference ranges and typical impacted ranges were summarized: The LRef and LImp datasets included the same 84 variables: our 8 chemical analytes (ALKT, Ca, Cl, COND, DOC, TKN, pH, and TP), 20 indices of benthic community structure[14] (% [of sample abundance accounted for by] Amphipoda [Amph]; % Asellidae [Asell]; % Chironomidae [Chir]; % Corixidae, Isopoda, Gastropoda, and Hirudinea taxa combined[CIGH]; % Ephemeroptera, Odonata, and Trichoptera taxa combined [EOT]; % Ephemeroptera, Plecoptera, and Trichoptera taxa combined [EPT]; % Insects [Insect]; % aquatic earthworms [i.e., oligochaetous Clitellata; Oligo]; % animals with a burrowing habit [Bw]; % filtering collectors [FC]; % gathering collectors [GC]; % predators [P]; % scrapers [SC]; % shredders [SH]; Hilsenhoff's family biotic index [HBI]; taxonomic richness [the number of different taxa represented, standardized to a 100-count sample using rarefaction; Rich100]; Axis-1 and Axis-2 scores from a principal coordinates analysis ordination of samples, based on Bray-Curtis distances among their taxa counts [PCoA1 and PCoA2]; and Axis-1 and Axis-2 scores from a correspondence analysis ordination of samples, based on their log10-transformed taxa abundances [CA1 and CA2]); relative abundances of the 38 lake taxa, our 6 stressor variables (c_- and l_urban, c_- and l_agri, and c_- and l_rd), and 12 variables describing geographic position or habitat (area, pmtr, zmax, DR, cwrt, LNN, lat, lon, elev, c_over, and c_slp). The SRef and SImp datasets included the same biological, chemical, and stressor variables as were included in the lake datasets, plus the relative abundances of 37 stream taxa, and 8 variables describing geographic position or habitat (c_da, lat, lon, elev, c_over, DS, Strahl, and c_slp).

Normal ranges of our 8 chemical analytes, 20 biological indicators, and relative taxa abundances were summarized for each site-group using key percentiles in their distributions (i.e., 5th, 10th, 25th, 50th, 75th, 90th, and 95th). They were also specified as two-sided central 95%/95% tolerance regions (i.e., the lower and upper limits of the variable's sample distribution that is estimated to contain at least 95% of that variable's population distribution with 95% confidence). We report tolerance limits because our non-normally distributed response variables mean that the standard definition of the normal range (i.e., mean ± 2 standard deviations) would not enclose 95% of the data (e.g., Barrett et al. 2015). Sample sizes in the reference-site groups were compared against required minima,

[14] Values of biological indices reported for each stream or lake were means from its three sampled locations.

estimated using the distfree.est() function (with $\alpha = 0.05$) in R (R Core Team 2016). This assessment indicated that group sizes were too small to allow tolerance limits to be precisely estimated by conventional methods. For this reason, we calculated them non-parametrically with interpolation/extrapolation, using the nptol.int() function in R (R Core Team 2016), an implementation of the methods proposed by Young and Mathew (2014).

In order to describe the geographic coverage of the four groups of waterbodies in the case-study watershed, to quantify their interspersion, and to gain insights about how cumulative effects have manifested themselves among the impacted lakes and rivers, we created density plots for all variables in the LRef, LImp, SRef, and SImp datasets. We also created an ordination biplot from the PCoA1 and PCoA2 scores associated with the 219 waterbodies, delineated the outer bounds of each site-group in the plot as convex hulls, and summarized biological variation about the site-group centroids as median absolute deviations among site scores on the horizontal and vertical axes. Similarly, we performed two principal coordinates analyses (PCAs) and summarized within-group variation in each ordination as median absolute deviations: the first PCA ordinated sites according to their chemical attributes (ALKT, Ca, Cl, COND, DOC, TKN, pH, and TP); the second, according to their geographic position (lat, lon, elev) and catchment characteristics (c_da, c_over, c_slp). When conducting the PCA analyses, input variables were standardized to z-scores (using the global mean and standard deviation) and eigenanalyses were performed on covariance matrices.

Unless otherwise indicated, datasets were compiled in Microsoft Excel. Statistical analyses were performed using scripts (Online Resource 2) written and executed in R Studio version 0.99.903, which was running R version 3.3.1 (R Core Team 2016). Maps were created, and geospatial analyses were performed, using ArcGIS Desktop 10.2 and 10.4.1 or System for Automated Geoscientific Analyses (SAGA) version 2.0.8 (Plewes 2015).

Results

Evidence of Thresholds between Minimally Impacted and Disturbed Communities

Random forest models of lake biology explained between 0% and 62% of the variation in the relative abundances of benthic invertebrate taxa (Online Resource 3). Predictive accuracies of two models met our pseudo-$R^2 \geq 0.3$ threshold, thus partial dependencies of Chironomidae (pseudo-R^2 = 0.62) and Platyhelminthes (pseudo-R^2 = 0.35) abundances were assessed for evidence of threshold effects. In general, random forests modeled chemical responses more accurately — pseudo-R^2 values ranged between 0.33 (TKN) and 0.64 (ALKT) — so all eight chemistry variables were evaluated for threshold effects. Models of stream biology explained between 0% and 34% of the variance in taxa abundances, and only two taxa had pseudo-R^2 values that met our inclusion criterion: Asellidae (pseudo-R^2 = 0.34) and Hyalellidae (pseudo-R^2 = 0.32). Corresponding models explained between 0% and 72% of the variation in stream chemistry variables, and 5 models had pseudo-R^2 values that qualified them for inclusion in threshold evaluation: COND (pseudo-R^2 = 0.72), ALKT (pseudo-R^2 = 0.64), Cl (pseudo-R^2 = 0.63), Ca (pseudo-R^2 = 0.58), and pH (pseudo-R^2 = 0.43).

Random forest and TITAN analyses did not suggest clear onset-of-effect thresholds for littoral lake benthic communities associated with any of the six predictors of stressor exposure (c_agri, c_rd, c_urban, l_agri, l_rd, or l_urban; Tables 1 and 2, Online Resource 3). In the case of streams (Tables 3 and 4, Online Resource 3), the two analytical approaches provided consistent evidence suggesting the lack of an onset-of-effect threshold in relation to c_rd and l-urban; however partial dependencies suggested the presence of thresholds for c_agri (4-5%), l_agri (4-6%), and l_rd X 1000 (0.0002-0.0008 m/m^2) that were not corroborated by the TITAN analysis, and the TITAN analysis suggested a threshold for c_urban (0.1%) that was not corroborated by the random forest. We did not consider this evidence strong enough to unequivocally demonstrate biological onset-of-effects thresholds — save for the threshold effect of c_urban on stream benthic communities (which occurred so close to the zero-exposure level as to be practically indistinguishable from it, given measurement error). Biological reference criteria were, therefore, not relaxed relative to our default of zero stressor exposure (Online Resource 2).

Although TITAN did not conclusively demonstrate biological thresholds, it did provide insights regarding which taxa respond positively, and which respond negatively to urbanization, road density, and agriculture (Table 5). In the six TITAN analyses undertaken for lakes, Z+ taxa accounted for between 67% and 88% of the counts of pure and reliable taxa (representative Z+ taxa included Gamma, Asellidae, Oligochaeta, and Baetidae, and Z- taxa included Chironomidae, Coenagrionidae, and Corduliidae). Similarly, most (i.e., 43% to 90%) of the pure and reliable stream taxa (including Asellidae, Coenagrionidae, Corixidae, Elmidae, Hyalellidae, and Oligochaeta) also responded positively to the anthropogenic factors considered in our analysis (Table 5).

<< INSERT TABLES 1 – 5 HERE >>

Partial dependencies from the random forests suggested numerous lake-chemistry onset-of-effect thresholds (Table 6, Online Resource 3): threshold responses for ALKT were suggested for c_rd (0.0002 m/m^2), c_urban (0.2 %), and l_rd (0.0002 m/m^2); for Ca, were suggested for c_rd (0.00035 m/m^2) and l_rd (0.0003 m/m^2); for Cl, were suggested for c_rd (0.0004 m/m^2), c_urban (0.4%), l_rd (0.0002 m/m^2), and l_urban (2%); for COND, were suggested for c_rd (0.0003 m/m^2), c_urban (0.4%), and l_urban (2.5%); for DOC, were suggested for c_urban (0.4%); for TKN, were suggested for c_urban (0.4%), l_agri (5.5%), and l_urban (2.5%); for pH, were suggested for c_rd (0.0002 m/m^2); and for TP were suggested for c_agri (3%), c_urban (0.4%), l_agri (3%), and l_urban (3%). Stream-chemistry thresholds were also suggested (Table 7): for ALKT, these included c_urban (1%), l_rd (0.0005 m/m^2), and l_urban (2.5%); for Ca, these included c_urban (2%), l_rd (0.0005 m/m^2), and l_urban (4%); for Cl, these included c_agri (1%), c_rd (0.0005 m/m^2), and l_rd (0.0005 m/m^2); and for COND, these included c_rd (0.0009 m/m^2) and l_rd (0.0005 m/m^2). These thresholds were reflected as criteria for defining reference and impacted groups (Online Resource 2).

<< INSERT TABLES 6 & 7 HERE >>

Normal Reference Ranges and Typical Impacted Ranges of Chemical and Biological Lake and Stream Attributes

The geographic ranges (Figure 1) and ranges of natural environmental features associated with reference and impacted streams substantially overlapped in many cases (Figure 2; Online Resources 4 and 5), suggesting reasonable interspersion. The two groups of streams were, for example, similarly distributed in the Muskoka River Watershed (i.e., had a similar range of geographic coordinates and elevations); however, reference streams had shallower soils on average, and smaller catchment areas. Lesser interspersion was apparent for the two groups of lakes (Figures 1 and 2, Online Resources 4 and 5), reference lakes being, on average, smaller and shallower, with fewer up-gradient lakes in their drainage networks, and shallower soils in their catchments.

<<INSERT FIGURE 2 HERE>>

Although reference and impacted streams and lakes had similar DOC and TKN, the four groups' biological and chemical centroids differed (Figures 3, 4, and 5), and the impacted groups had more variable taxonomic structure and water chemistry than was observed among the reference groups (Table 8). Specifically, impacted lakes and streams had higher ALKT, Ca, Cl, COND, pH, and TP than was typical of reference lakes and streams (Tables 9 and 10, Figure 5, Online Resources 4 and 5). Impacted streams had more abundant Asellidae, Amphipoda, and CIGH taxa, and fewer Chironomidae than was typical of reference streams; and insects were less abundant in impacted lakes than in reference lakes (Tables 9 and 10, Figure 3, Online Resources 4 and 5).

<<INSERT TABLES 8 – 10 AND FIGURES 3, 4, AND 5 HERE>>

Conclusions and discussion

Do biological or chemical onset-of-effect thresholds distinguish reference and impacted sites?

Threshold detection methods are sensitive to the distribution of sites across the sampled gradient (King and Baker 2014), and different detection methods can provide conflicting results (King and Baker 2014). As suggested by Roubeix et al. (2016), we concentrated our sampling efforts in the lesser impacted tails of the Watershed's gradients of agricultural land use, urbanization and road density to provide optimal resolution for detecting onset-of-effect thresholds, and we used two complementary methods of threshold detection (TITAN and random forest partial dependencies). Because no objective criterion for defining a threshold exists, King and Baker (2014) suggested that researchers support their statistical results by examining the sensitivities, evolutionary relationships, and life-history characteristics of the taxa implicated in community change points. We assert that additional statistical rules of thumb would also be helpful. As a first approximation for TITAN, we suggest that reasonable evidence of a community threshold is provided when the following 4 criteria are satisfied: (1) MaxSum Z — a measure of the degree of change in community structure that occurs at the community change-point, expressed as the mean contribution to SumZ made by each pure and reliable taxon — is greater than 5 (see also Baker and King [2010] and Wallace and Biastoch [2016] for examples in which thresholds were inferred given MaxSumZ values between 4.5 and 13); (2) CFDW — the width of the steeply ascending region of the SumZ+/SumZ- cumulative frequency distribution — is less than 11% of the range of the stressor variable; (3) at least 4 taxa respond purely and reliably to the stressor of interest (PRT $\geq$ 4); and (4) #ST — the number of synchronous taxa having 95% CIs describing their taxon-specific change-points that include the X-value of the community change point — is both greater than 3, and represents at least 25% of the taxa shown to be pure and reliable.

Accordingly, we conclude that no onset-of effect thresholds for lacustrine and riverine benthic invertebrate communities were present in the Muskoka River Watershed — a finding that contrasts with Utz et al. (2009) who reported loss of sensitive taxa from streams in the Piedmont at 2.5% or more impervious cover, and with Pardo et al. (2012) who detected onset-of-effect thresholds for Maryland streams below 2% urbanization. Although no biological thresholds were demonstrated, TITAN did summarize taxa-specific responses to urbanization, road density, and agriculture. A majority of taxa responded positively to these "stressors". Given their largely stimulatory effects (as nutrient sources), using the term *stressor* to describe land-use factors in the Muskoka River Watershed can be misleading.

Numerous chemical thresholds were, however, apparent. For lakes, we detected thresholds for all 8 chemical indicators (ALKT, Ca, Cl, COND, DOC, TKN, pH, and TP) and for all six stressor variables (c_agri, c_rd, c_urban, l_agri, l_rd, l_urban). For streams, we detected thresholds for 4 chemical indicators (ALKT, Ca, Cl, and COND) and five of six stressor variables (c_agri, c_rd, c_urban, l_rd, l_urban). Thresholds were most commonly encountered in relation to urbanization (6 of 8 chemical analytes for lakes, and 2 of 8 for streams) and road density (5 chemical analytes for lakes and 4 for streams), but were also encountered in relation to agriculture (2 chemical analytes for lakes and 1 for streams). These thresholds typically occurred at very low levels of stressor exposure, as low as 1% agriculture (Cl, streams), 0.0002 m/m2 road density (e.g., ALKT, Cl, lakes) and 0.2% urban (ALKT, lakes), which is consistent with previously published results (e.g. Booth and Jackson 1997, Brabec et al. 2002). For lakes, thresholds were slightly more commonly detected for stressor variables measured at the cumulative-catchment scale than at the local-catchment scale; but for streams, thresholds were detected with approximately equal frequency in relation to stressor variables measured at the two different scales.

<u>Based on minimally impacted reference sites, what are the normal ranges of chemical and biological conditions for lakes and streams in the Muskoka River Watershed?</u>

The concept of the reference condition is ambiguous, and has been difficult to "translate from theory to practice" (Pardo et al 2012), given that the allowable level of anthropogenic impact is not clearly articulated (Stoddard et al. 2006, Pardo et al. 2012). Onset-of-effect thresholds represent empirical break-points where effects from stressor exposures become detectable (Utz et al. 2009). Objectivity of the reference condition approach can be improved by using such thresholds as criteria for defining reference and impacted sites.

Summarizing variation within reference and impacted groups allowed us to tabulate normal reference ranges of biological and chemical condition, as well as typical ranges of impacted conditions (Tables 9 and 10, Online Resource 4). We specified the boundaries of these ranges as critical percentiles and 95/95 tolerance ranges, non-parametric statistics that are easily calculated and simple to interpret as biocriteria (Smith et al. 2005). Local water managers may consult these tables when trying to understand how typical or atypical the chemical or biological attributes of different waterbodies are, or when seeking context in which to interpret the meaning of any changes in water quality or community structure that may be observed over time.

We included a variety of biological and chemical indicators in our normal-range tables because multiple indicators are required to represent the complexity of lake and stream ecosystems (Dale and Beyeler 2001), because different indicators respond differently to different stressors (Jones et al. 2017), and because multiple indicators are relevant to the Muskoka Region's water management goals and policies. The normal ranges reported in this paper should be considered a first approximation. Efforts should be made to sample additional reference lakes and streams, because having access to additional reference data would allow tolerance regions to be estimated more precisely. A larger pool of reference sites would also facilitate sophisticated model-based approaches for matching test-sites with the most appropriate set of reference sites (e.g., Norris and Hawkins 2000).

Relaxing our chemical reference criteria from our default definition of zero-stress, to the onset-of-effect threshold levels resulted in only very minor changes to the populations of qualifying reference sites, and therefore very minor changes to tabulated reference and impacted ranges. This minimal effect may be partially explained by the precipitous decline in the number of qualifying reference sites that occurs as one adds more stressor-variable-specific criteria into the definition of *reference site* (Jones 2009), and by correlations among stressor variables. For example, in a situation where a threshold effect is detected for c_urban but not c_rd, the effect of relaxing the c_urban criterion on the population of qualifying reference sites will be moderated because increases in c_urban are generally accompanied by increased c_rd.

<u>Are cumulative effects of land-use evident in the Watershed's lakes and streams?</u>

The chemical and biological conditions of reference and impacted waterbodies were not discrete; however, impacted lakes and streams have higher alkalinities and conductivities, and higher calcium and chloride concentrations on average than are found in reference lakes and streams. Biologically, the Chironomidae and other insects account for a smaller proportion of the abundance of benthic animals in impacted lakes and streams than they do in reference lakes and streams; and the relatively tolerant Asellidae and CIGH taxa are more abundant in impacted streams than in reference streams. Whole-community differences were evident from ordinations, in which site-group centroids occupied different positions on the horizontal and vertical axes; and, in general, chemical and biological attributes of impacted waterbodies were more variable than they were in reference waterbodies.

Some overlap between reference and impacted site groups was expected because community structure and water quality is determined by a mix of natural and anthropogenic factors, and stressor exposures in the Muskoka River Watershed were modest, relative to more developed areas of southern Canada. Differences in site-group means and within-group variation can be attributed to a combination of cumulative effects of human activities and natural factors. It is difficult to distinguish these sources of variation because land-uses are not dispersed evenly or randomly across watersheds, and imperfect interspersion of reference and impacted sites result in these factors being unequally distributed among reference and impacted groups.

Defining what constitutes a reference lake or stream, and characterizing normal ranges of chemical and biological condition based on these reference sites, should benefit local long-term monitoring and reporting programs. More generally, the methods we demonstrate for detecting community-level thresholds and summarizing normal ranges are also easily transferrable to other community types, other indicators, and other study regions. Threshold assessments can also have considerable academic and practical value. From the perspective of environmental monitoring and management, identifying onset-of-effect thresholds can be important for early detection and mitigation, and for setting planning and conservation targets and limits (Utz et al. 2009, Mitchell et al. 2014, Jones 2016). Heuristically, hypotheses about the mechanisms behind stressor effects can be generated by combining patterns of congruence among taxa-specific thresholds with knowledge describing the autecology and traits of component taxa (Wagenhoff et al. 2017).

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Tables

Table 1: Random forest-based evidence regarding the occurrence and position of community-level biological thresholds along gradients of lacustrine stress (Chiro = Chironominae, Platy = Platyhelminthes, I = inconclusive, IR = inconsistent response types, NT = non-threshold response [may be linear/complex, quadratic, or may have no response], T = threshold response [approximate position on X given in parentheses], US = under sampled; supporting details in Online Resource 3).

X	Platy	Chiro	Conclusion	Remark
c_agri	T (2.0)	NT	I	IR
c_rd X 1000	T (0.5)	NT	I	IR
c_urban	NT	NT	NT	
l_agri	T (20)	NT	I	IR, US
l_rd X 1000	T (1.5)	NT	I	IR
l_urban	NT	NT	NT	

Table 2: TITAN-based evidence about the existence of thresholds of stressor exposure in the taxonomic structure of lacustrine benthic macroinvertebrate communities. Upper pane concerns decreasing (Z-) and lower pane increasing (Z+) taxa. In each pane, the 7 upper rows describe community responses to land-use stressors; middle 3 rows describe responses to morphometric and hydrological attributes; and lower 3 rows describe community responses to chemical and physical properties of lake water. Abbreviations are explained in the appendix. The 5th and 95th percentiles from bootstrapped replicates are provided in parentheses to characterize uncertainty about the position of the community change-point in the range of the predictor (abbreviations as per Appendix).

	X	MaxSum Z	SumZ Peak	CFDW	PRT	# ST	TCPCIW	CCPCIW	Threshold	CCP
Z- taxa	c_agri	4.5	strong	9%	5	5	20%	8%	no	
	c_rd X 1000	6.0	weak	32%	2	2	70%	27%	no	
	c_urban	5.2	weak	9%	3	3	33%	8%	no	
	l_agri	3.8	weak	15%	7	6	19%	13%	no	
	l_rd X 1000	5.0	weak	14%	4	4	24%	8%	no	
	l_urban	8.5	weak	17%	7	7	11%	14%	no	
Z+ taxa	c_agri	5.0	strong	11%	15	15	21%	8%	no	
	c_rd X 1000	4.2	strong	24%	15	12	56%	15%	no	
	c_urban	4.5	strong	14%	15	10	20%	7%	no	
	l_agri	5.2	weak	14%	14	12	21%	12%	no	
	l_rd X 1000	4.4	weak	19%	17	13	14%	13%	no	
	l_urban	4.5	strong	15%	16	9	16%	6%	no	

Table 3: Random forest-based evidence regarding the occurrence and position of community-level biological thresholds along gradients of riverine stress (Asell = Asellidae, Hyale = Hyalellidae, I = inconclusive, IR = inconsistent response types, NT = non-threshold response [may be linear/complex, quadratic, or may have no response], T = threshold response [approximate position on X given in parentheses]; supporting details in Online Resource 3).

X	Asell	Hyale	Conclusion	Remark
c_agri	T (5)	T (4)	T (4 – 5)	
c_rd X 1000	T (0.4)	NT	I	IR
c_urban	NT	NT	NT	
l_agri	T (4)	T (6)	T (4–6)	
l_rd X 1000	T (0.8)	T (0.2)	T (0.2–0.8)	
l_urban	NT	NT	NT	

Table 4: TITAN-based evidence about the existence of thresholds of stressor exposure in the taxonomic structure of stream benthic macroinvertebrate communities. Upper pane concerns decreasing (Z-) and lower pane increasing (Z+) taxa. In each pane, the 7 upper rows describe community responses to land-use stressors; the middle 2 rows describe responses to drainage area and substrate type; and lower 3 rows describe community responses to chemical and physical properties of stream water. Abbreviations are explained in the appendix. The 5th and 95th percentiles from bootstrapped replicates are provided in parentheses to characterize uncertainty about the position of the community change-point in the range of the predictor (abbreviations as per Appendix).

	X	MaxSum Z	SumZ Peak	CFDW	PRT	# ST	TCPCIW	CCPCIW	Threshold	CCP
Z- taxa	c_agri	4.1	weak	30%	1	1	35%	13%	no	
	c_rd X 1000	2.8	weak	37%	4	4	21%	50%	no	
	c_urban	4.2	strong	3%	4	4	15%	62%	no	
	l_agri	3.9	weak	51%	1	1	22%	47%	no	
	l_rd X 1000	3.4	weak	14%	3	3	18%	41%	no	
	l_urban	2.5	strong	30%	4	2	19%	33%	no	
Z+ taxa	c_agri	3.7	weak	18%	9	7	38%	14%	no	
	c_rd X 1000	4.4	weak	9%	5	4	13%	6%	no	
	c_urban	5.2	strong	3%	7	7	12%	1%	yes	0.1 (0.09, 1.02)
	l_agri	4.0	weak	21%	9	6	43%	16%	no	
	l_rd X 1000	4.0	weak	14%	6	6	17%	5%	no	
	l_urban	4.4	strong	30%	3	2	29%	18%	no	

Table 5: Counts of positively responding Z+ and negatively responding Z- pure and reliable taxa from 12 TITAN analyses (abbreviations as per Appendix).

		l_urban	l_agri	l_rd	c_urban	c_agri	c_rd	Example Taxa
Lakes	Z^-	7	7	4	3	5	2	Chiro, Coena, Cordu1
	Z^+	16	14	17	15	15	15	Gamma, Asell, Oligo, Baetidae
Streams	Z^-	4	1	3	4	1	4	Chiro, Ortho, Gomph
	Z^+	3	9	6	7	9	5	Corix, Elmid, Oligo, Asell, Coena, Hyale

Table 6: Random forest-based evidence regarding the occurrence and position of chemical thresholds along gradients of lacustrine stress (NT = non-threshold response [may be linear/complex, quadratic, or may have no response], T = threshold response [approximate position on X given in parentheses]; supporting details in Online Resource 3).

X	ALKT	Ca	Cl	COND	DOC	TKN	pH	TP
c_agri	NT	NT	NT	NT	NT	NT	NT	T (3)
c_rd X 1000	T (0.2)	T (0.35)	T (0.4)	T (0.3)	NT	NT	T (0.2)	NT
c_urban	T (0.2)	NT	T (0.4)	T (0.4)	T (0.4)	T (0.4)	NT	T (0.4)
l_agri	NT	NT	NT	NT	NT	T (5.5)	NT	T (3)
l_rd X 1000	T (0.2)	T (0.3)	T (0.2)	NT	NT	NT	NT	NT
l_urban	NT	NT	T (2)	T (2.5)	NT	T (2.5)	NT	T (3)

Table 7: Random forest-based evidence regarding the occurrence and position of chemical thresholds along gradients of riverine stress (NT = non-threshold response [may be linear/complex, quadratic, or may have no response], T = threshold response [approximate position on X given in parentheses]; supporting details in Online Resource 3).

X	ALKT	Ca	Cl	COND	pH
c_agri	NT	NT	T (1)	NT	NT
c_rd X 1000	NT	NT	T (0.5)	T (0.9)	NT
c_urban	T (1)	T (2)	NT	NT	NT
l_agri	NT	NT	NT	NT	NT
l_rd X 1000	T (0.5)	T (0.5)	T (0.5)	T (0.5)	NT
l_urban	T (2.5)	T (4)	NT	NT	NT

Table 8: Variation in benthic community structure, water chemistry and geographic/catchment attributes within groups of lake and stream reference and impacted sites, expressed as horizontal (axis-1) and vertical (axis-2) median absolute deviations (MAD) about site-group centroids (abbreviations as per Appendix).

	Biology PCoA		Chemistry PCA		Geographic/Catchment PCA	
	MAD_{PCoA1}	MAD_{PCoA2}	MAD_{PCA1}	MAD_{PCA2}	MAD_{PCA1}	MAD_{PCA2}
LRef	0.015	0.011	0.191	0.398	0.946	0.822
LImp	0.016	0.015	0.307	0.364	1.675	1.613
SRef	0.008	0.009	0.544	1.323	2.058	1.131
SImp	0.015	0.018	1.177	1.382	1.291	1.015

Table 9: Normal ranges of selected chemical and biological attributes of minimally disturbed (reference) and impacted lakes (indicators set in bold typeface were recommended by Jones et al. 2017, based on accuracy with which indicator values could be modeled and sensitivity to stressors of interest in the case-study watershed). The full table, showing all predictors and response variables is provided in Online Resource 4 (abbreviations as per Appendix).

	Reference Lakes										Impacted Lakes									
attribute	n	5th	10th	25th	mean (SE)	median	75th	90th	95th	95 TI	n	5th	10th	25th	mean (SE)	median	75th	90th	95th	95 TI
l_urban	26	0.0	0.0	0.0	0.0 (0.00)	0.0	0.0	0.0	0.0	(0.0, 100.0)	81	0.0	0.0	1.0	5.1 (1.84)	2.6	5.7	10.3	18.2	(0.0, 100.0)
l_agri	26	0.0	0.0	0.0	0.0 (0.00)	0.0	0.0	0.0	0.0	(0.0, 100.0)	81	0.0	0.0	0.0	4.8 (1.41)	1.9	6.5	14.7	20.8	(0.0, 100.0)
l_rdX1000	26	0.000	0.000	0.000	0.000 (0.0000)	0.000	0.000	0.000	0.000	(0.000, 0.000)	81	0.176	0.282	0.676	1.402 (0.2398)	1.271	1.775	2.605	3.069	(0.000, 10.163)
c_urban	26	0.0	0.0	0.0	0.0 (0.00)	0.0	0.0	0.0	0.0	(0.0, 100.0)	81	0.0	0.0	0.5	2.3 (0.52)	1.5	3.3	5.4	7.0	(0.0, 100.0)
c_agri	26	0.0	0.0	0.0	0.0 (0.00)	0.0	0.0	0.0	0.0	(0.0, 100.0)	81	0.0	0.0	0.0	3.5 (1.04)	1.7	4.0	12.5	17.2	(0.0, 100.0)
c_rdX1000	26	0.000	0.000	0.000	0.000 (0.0000)	0.000	0.000	0.000	0.000	(0.000, 0.000)	81	0.085	0.189	0.439	0.907 (0.1246)	0.705	1.386	1.826	2.037	(0.004, 2.535)
c_fn	26	99.9	100.0	100.0	99.9 (0.09)	100.0	100.0	100.0	100.0	(92.4, 100.0)	81	81.6	87.4	93.5	94.4 (1.29)	96.1	98.6	99.7	100.0	(66.1, 100.0)
ALKT	26	2.75	2.80	3.10	4.97 (0.459)	4.58	5.68	7.05	8.39	(2.41, 25.24)	81	4.35	5.10	6.50	9.37 (0.812)	8.75	10.80	15.20	17.70	(2.38, 24.46)
Ca	26	0.93	0.96	1.12	1.42 (0.131)	1.27	1.46	1.92	2.20	(0.44, 9.17)	81	1.26	1.72	2.04	2.92 (0.270)	2.60	3.52	3.86	5.66	(0.75, 9.51)
Cl	26	0.08	0.10	0.16	0.21 (0.020)	0.19	0.26	0.35	0.38	(0.00, 0.76)	81	0.27	0.51	1.22	6.00 (1.558)	3.91	7.39	9.88	16.90	(0.11, 56.91)
COND	26	13.1	14.2	16.5	18.8 (1.00)	17.9	19.4	24.6	28.9	(8.7, 48.0)	81	16.8	22.2	25.6	54.5 (8.70)	40.0	63.4	108.0	163.0	(9.6, 239.4)
DOC	26	2.8	3.1	3.6	4.7 (0.35)	4.6	5.5	6.6	7.1	(1.1, 20.8)	81	2.8	3.0	3.7	5.2 (0.47)	4.4	6.1	7.8	9.3	(1.9, 16.8)
TKN	26	190.00	208.00	234.25	284.00 (14.523)	270.50	322.00	375.00	424.75	(178.16, 575.64)	81	185.00	206.00	228.00	299.99 (23.087)	275.00	336.00	418.00	508.00	(157.22, 1062.84)
pH	26	5.67	5.78	6.04	6.27 (0.081)	6.30	6.56	6.82	6.85	(4.24, 7.17)	81	6.10	6.45	6.70	6.83 (0.073)	6.92	7.01	7.23	7.28	(5.43, 7.48)
TP	26	3.03	3.10	4.00	5.95 (0.556)	5.15	7.10	11.15	11.73	(1.29, 14.19)	81	2.40	3.20	3.80	7.31 (1.115)	5.30	8.20	16.10	18.70	(1.63, 31.40)
Asell	26	0.0	0.0	0.0	0.1 (0.14)	0.0	0.0	0.0	0.0	(0.0, 12.3)	81	0.0	0.0	0.0	3.1 (1.10)	0.0	3.1	14.4	16.8	(0.0, 24.4)
Amph	26	0.1	0.3	1.0	11.3 (2.49)	6.6	16.6	29.8	34.2	(0.0, 100.0)	81	0.6	2.1	8.6	14.8 (1.92)	14.0	20.5	26.4	33.1	(0.2, 100.0)
Chir	26	37.9	43.1	50.9	62.0 (3.15)	62.5	72.3	84.5	86.5	(22.8, 100.0)	81	12.1	16.8	23.0	38.0 (3.73)	32.5	54.2	63.9	71.0	(4.4, 100.0)
CIGH	26	0.0	0.0	0.2	2.5 (0.71)	1.0	3.7	7.3	8.1	(0.0, 100.0)	81	0.9	1.2	2.6	8.3 (1.47)	5.8	10.9	20.6	25.0	(0.0, 100.0)
EOT	26	0.0	0.1	0.9	2.1 (0.33)	1.5	3.3	4.3	4.8	(0.0, 100.0)	81	0.0	0.3	0.6	1.4 (0.26)	1.0	1.8	2.6	3.6	(0.0, 100.0)
Insect	26	56.4	62.5	69.8	78.6 (2.76)	80.6	91.6	94.5	96.2	(24.0, 100.0)	81	30.1	36.0	45.7	57.9 (3.45)	57.4	72.0	79.4	89.1	(15.8, 100.0)
Rich100	26	8.3	8.7	9.8	12.1 (0.53)	12.3	13.9	14.9	15.5	(4.2, 26.5)	81	11.1	11.8	12.8	15.2 (0.54)	15.1	17.2	18.7	19.1	(9.2, 22.8)
PCoA1	26	-0.008	-0.008	-0.006	-0.003 (0.0008)	-0.004	0.002	0.002	0.004	(-0.011, 0.005)	81	-0.004	-0.002	0.001	0.005 (0.0010)	0.006	0.009	0.010	0.011	(-0.008, 0.014)
PCoA2	26	-0.012	-0.012	-0.009	-0.005 (0.0009)	-0.005	-0.002	0.001	0.001	(-0.018, 0.011)	81	-0.012	-0.011	-0.009	-0.003 (0.0016)	-0.005	0.000	0.010	0.012	(-0.015, 0.023)
CA1	26	-0.332	-0.287	-0.216	-0.177 (0.0192)	-0.191	-0.099	-0.053	-0.037	(-0.663, 0.039)	81	-0.459	-0.391	-0.329	-0.260 (0.0224)	-0.252	-0.182	-0.131	-0.084	(-0.667, -0.043)
CA2	26	-0.519	-0.469	-0.313	-0.183 (0.0437)	-0.178	-0.034	0.058	0.121	(-1.202, 0.599)	81	-0.303	-0.198	-0.073	0.181 (0.0697)	0.068	0.447	0.652	0.767	(-0.568, 1.027)
EPT	26	1.2	2.0	3.9	7.0 (0.94)	6.3	9.2	13.2	15.6	(0.0, 100.0)	81	1.9	2.4	4.4	10.2 (1.53)	7.5	14.2	22.2	23.3	(0.5, 100.0)
HBI	26	6.1	6.4	6.7	7.0 (0.08)	7.1	7.3	7.5	7.5	(5.8, 7.9)	81	5.4	5.7	6.2	6.6 (0.12)	6.7	7.0	7.2	7.4	(4.4, 7.6)
Bw	26	45.8	48.1	57.2	67.2 (2.81)	64.8	75.7	87.6	89.6	(25.1, 100.0)	81	32.0	33.9	43.3	55.1 (3.17)	55.5	68.2	76.5	79.1	(17.7, 100.0)
FC	26	0.0	0.0	0.3	1.3 (0.25)	0.7	1.9	2.9	3.4	(0.0, 100.0)	81	0.3	0.5	1.2	3.0 (0.50)	2.4	3.9	7.0	8.6	(0.0, 100.0)
GC	26	56.2	58.1	69.6	73.4 (1.78)	74.2	80.1	84.3	85.0	(50.0, 100.0)	81	49.7	53.7	62.3	68.0 (1.91)	68.9	74.4	79.8	82.7	(40.0, 100.0)
P	26	7.0	9.1	11.2	13.7 (0.86)	12.7	16.3	19.5	21.8	(6.4, 100.0)	81	5.2	5.9	8.8	12.8 (1.17)	11.6	15.5	19.9	23.4	(1.5, 100.0)
SC	26	0.0	0.0	0.1	6.6 (1.63)	4.0	9.7	17.6	19.8	(0.0, 100.0)	81	1.2	2.4	4.7	8.3 (1.01)	7.3	11.5	14.4	18.7	(0.0, 100.0)
SH	26	0.0	0.0	0.0	0.3 (0.07)	0.3	0.4	0.7	1.1	(0.0, 1.3)	81	0.0	0.0	0.0	2.6 (1.14)	0.5	1.6	9.4	13.0	(0.0, 38.8)

Table 10: Normal ranges of selected chemical and biological attributes of minimally disturbed (reference) and impacted streams (indicators set in bold typeface were recommended by Jones et al. 2017, based on accuracy with which indicator values could be modeled and sensitivity to stressors of interest in the case-study watershed). The full table, showing all predictors and response variables is provided in Online Resource 4 (abbreviations as per Appendix).

	Reference Streams											Impacted Streams										
attribute	n	5th	10th	25th	mean (SE)	mean (SE)	median	75th	90th	95th	95 TI	n	5th	10th	25th	mean (SE)	median	75th	90th	95th	95 TI
l_urban	19	0.0	0.0	0.0	0.0	0.0 (0.00)	0.0	0.0	0.0	0.0	(0.0, 100.0)	93	0.0	0.0	0.0	6.7 (3.06)	0.8	5.2	18.4	32.9	(0.0, 100.0)
l_agri	19	0.0	0.0	0.0	0.0	0.0 (0.00)	0.0	0.0	0.0	0.0	(0.0, 100.0)	93	0.0	0.0	0.0	7.6 (3.17)	1.2	4.9	19.3	54.3	(0.0, 100.0)
l_rdX1000	19	0.000	0.000	0.000	0.000	0.000 (0.0000)	0.000	0.000	0.000	0.000	(0.000, 0.000)	93	0.073	0.167	0.354	1.270 (0.3998)	0.767	1.260	2.542	3.121	(0.023, 14.268)
c_urban	19	0.0	0.0	0.0	0.0	0.0 (0.00)	0.0	0.0	0.0	0.0	(0.0, 100.0)	93	0.0	0.0	0.0	5.8 (2.73)	1.0	4.6	10.5	40.0	(0.0, 100.0)
c_agri	19	0.0	0.0	0.0	0.0	0.0 (0.00)	0.0	0.0	0.0	0.0	(0.0, 100.0)	93	0.0	0.0	0.1	4.1 (1.36)	1.1	4.8	12.3	15.8	(0.0, 100.0)
c_rdX1000	19	0.000	0.000	0.000	0.000	0.000 (0.0000)	0.000	0.000	0.000	0.000	(0.000, 0.000)	93	0.070	0.155	0.290	0.996 (0.2933)	0.662	0.970	1.564	3.740	(0.017, 8.189)
c_fn	19	99.5	100.0	100.0	99.8	99.8 (0.21)	100.0	100.0	100.0	100.0	(76.6, 100.0)	93	54.8	76.4	85.5	89.3 (2.84)	95.2	99.2	99.9	100.0	(32.1, 100.0)
ALKT	19	2.53	3.35	4.73	7.47	7.47 (0.977)	6.25	7.95	12.00	19.43	(1.56, 30.57)	93	5.71	6.47	8.55	23.13 (5.211)	14.60	23.20	52.28	94.88	(0.05, 124.00)
Ca	19	1.16	1.23	1.55	2.28	2.28 (0.209)	2.08	2.53	4.30	4.32	(0.00, 5.09)	93	1.96	2.22	2.74	7.90 (1.933)	4.70	7.14	13.46	36.82	(1.44, 44.40)
Cl	21	0.06	0.08	0.12	0.35	0.35 (0.162)	0.15	0.19	0.41	0.51	(0.00, 15.45)	91	0.21	0.36	0.87	25.38 (12.238)	5.20	18.40	43.30	96.87	(0.15, 356.11)
COND	21	15.0	16.4	17.8	23.3	23.3 (1.82)	20.2	22.8	41.4	44.8	(8.9, 44.8)	91	22.4	25.6	34.2	142.6 (45.77)	69.0	135.0	266.0	480.5	(17.5, 1321.6)
DOC	19	4.1	5.3	7.1	11.8	11.8 (1.32)	11.8	14.0	23.4	24.9	(0.0, 31.7)	93	3.2	4.0	5.0	10.6 (1.82)	7.6	14.0	19.8	24.1	(1.6, 71.2)
TKN	19	237.30	280.80	424.50	670.47	670.47 (63.800)	652.00	896.50	1,094.00	1,237.00	(0.00, 1, 575.97)	93	203.00	238.40	300.00	534.69 (57.809)	449.00	720.00	858.40	1,140.00	(145.00, 1569.94)
pH	19	5.19	5.50	5.94	6.14	6.14 (0.098)	6.23	6.49	6.64	6.66	(3.74, 6.95)	93	6.11	6.34	6.63	6.84 (0.099)	6.83	7.13	7.35	7.52	(4.24, 8.05)
TP	19	7.53	9.06	17.70	30.41	30.41 (3.148)	34.50	38.35	42.72	48.04	(0.00, 166.20)	93	6.42	7.36	11.70	30.55 (7.270)	24.20	36.80	53.64	65.88	(4.30, 301.97)
Asell	19	0.0	0.0	0.0	2.6	2.6 (1.88)	0.0	0.0	1.6	9.5	(0.0, 183.2)	93	0.0	0.0	0.0	8.5 (3.41)	0.0	9.3	30.1	45.2	(0.0, 79.7)
Amph	19	0.0	0.0	0.0	0.2	0.2 (0.07)	0.0	0.3	0.6	0.7	(0.0, 100.0)	93	0.0	0.0	0.0	2.0 (0.86)	0.2	1.7	7.2	11.1	(0.0, 100.0)
Chir	19	49.9	52.7	60.9	71.0	71.0 (2.61)	73.7	80.2	86.7	90.0	(26.5, 100.0)	93	14.7	20.6	37.8	52.8 (4.14)	57.8	68.7	79.0	81.7	(7.0, 100.0)
CIGH	19	0.0	0.0	0.0	3.0	3.0 (1.87)	0.3	1.0	3.8	10.4	(0.0, 100.0)	93	0.0	0.0	0.0	10.5 (3.47)	2.0	11.4	34.2	50.6	(0.0, 100.0)
EOT	19	0.0	0.0	0.3	1.4	1.4 (0.41)	0.8	1.3	3.6	5.6	(0.0, 100.0)	93	0.0	0.0	0.3	2.0 (0.59)	1.1	2.3	4.3	7.1	(0.0, 100.0)
Insect	19	80.6	88.0	92.3	92.0	92.0 (2.16)	94.8	97.0	98.5	98.7	(0.0, 100.0)	93	27.6	48.0	67.0	77.2 (4.26)	85.1	94.1	97.1	98.3	(13.5, 100.0)
Rich100	19	8.1	8.4	9.1	11.7	11.7 (0.57)	11.5	14.0	14.9	15.3	(6.6, 27.3)	93	8.1	9.0	11.2	13.4 (0.62)	13.4	15.5	17.2	18.1	(5.6, 22.7)
PCoA1	19	-0.010	-0.010	-0.009	-0.007	-0.007 (0.0006)	-0.007	-0.006	-0.002	0.000	(-0.010, 0.004)	93	-0.009	-0.008	-0.006	-0.002 (0.0010)	-0.003	0.001	0.006	0.007	(-0.010, 0.010)
PCoA2	19	-0.005	-0.004	-0.002	0.002	0.002 (0.0012)	0.001	0.003	0.007	0.009	(-0.020, 0.072)	93	-0.008	-0.005	-0.002	0.005 (0.0022)	0.002	0.008	0.020	0.028	(-0.014, 0.044)
CA1	19	-0.347	-0.209	-0.088	0.216	0.216 (0.0925)	0.063	0.384	0.821	1.239	(-0.706, 1.570)	93	-0.412	-0.302	-0.173	0.242 (0.1139)	0.069	0.524	0.991	1.445	(-0.572, 2.018)
CA2	19	-1.045	-0.785	-0.614	-0.511	-0.511 (0.0530)	-0.487	-0.362	-0.263	-0.195	(-1.565, 1.026)	93	-0.515	-0.426	-0.342	-0.131 (0.0717)	-0.167	0.054	0.210	0.505	(-1.706, 1.109)
EPT	19	0.0	0.4	3.1	9.2	9.2 (1.59)	6.4	15.5	18.4	19.9	(0.0, 100.0)	93	0.1	0.6	3.1	12.8 (2.78)	7.9	16.3	38.8	45.3	(0.0, 100.0)
HBI	19	6.3	6.3	6.5	7.0	7.0 (0.09)	6.9	7.3	7.6	7.7	(6.3, 7.9)	93	5.1	5.5	6.3	6.7 (0.16)	6.9	7.3	7.5	7.7	(4.1, 7.9)
Bw	19	55.0	55.9	64.0	75.8	75.8 (2.53)	78.2	85.5	90.3	91.4	(49.1, 100.0)	93	25.9	34.7	50.5	63.9 (3.87)	70.7	80.0	85.2	87.5	(13.0, 100.0)
FC	19	0.0	0.0	0.8	3.9	3.9 (1.42)	1.3	3.4	7.8	12.7	(0.0, 100.0)	93	0.3	0.5	1.4	8.2 (2.10)	3.6	11.1	22.0	31.4	(0.3, 100.0)
GC	19	53.4	57.0	62.4	69.7	69.7 (2.72)	71.5	80.7	82.2	83.5	(0.0, 100.0)	93	41.6	51.5	61.5	68.2 (2.65)	69.7	77.4	83.5	88.9	(26.8, 100.0)
P	19	8.4	9.2	14.1	20.2	20.2 (2.66)	15.7	26.3	29.3	33.8	(0.0, 100.0)	93	4.2	6.3	8.9	14.6 (1.55)	13.5	18.6	22.9	27.1	(1.5, 100.0)
SC	19	0.0	0.0	0.0	0.8	0.8 (0.25)	0.5	0.9	2.0	4.2	(0.0, 100.0)	93	0.0	0.0	0.5	3.4 (0.85)	2.1	3.9	8.3	11.7	(0.0, 100.0)
SH	19	0.0	0.0	0.4	1.6	1.6 (0.38)	0.9	1.8	4.3	4.9	(0.0, 16.8)	93	0.0	0.0	0.3	2.2 (0.77)	0.9	2.0	5.3	9.1	(0.0, 26.0)

Figures

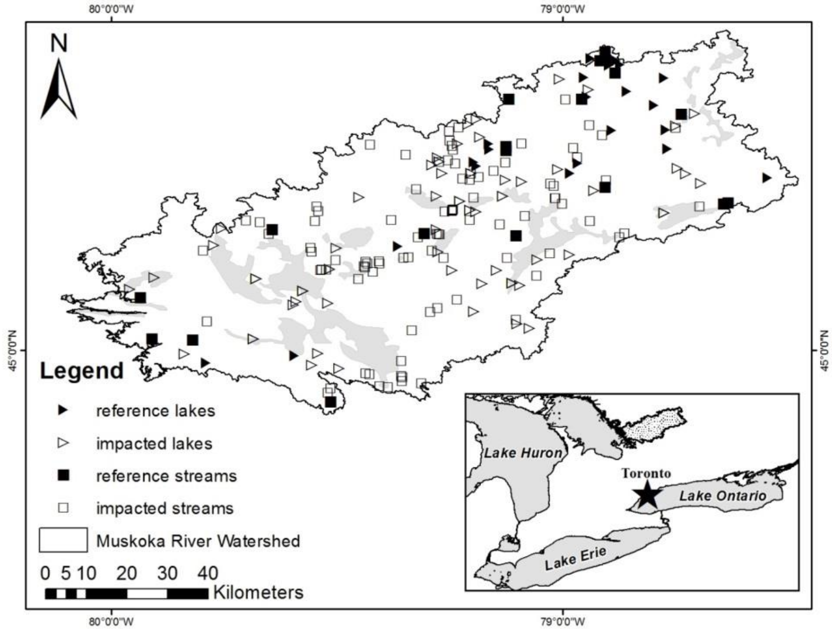

Figure 1: Muskoka River Watershed, showing sampled lakes and rivers.

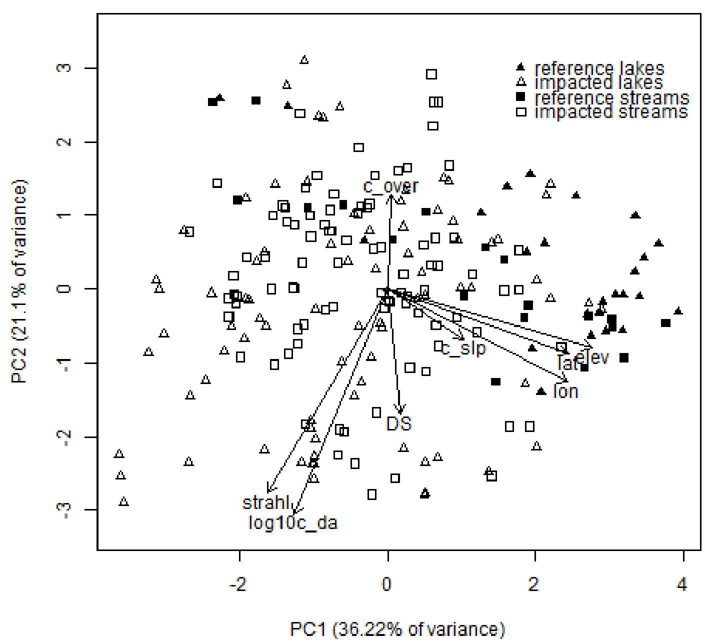

Figure 2: Principal components analysis ordination of reference and impacted lakes and streams, based on covariance among variables describing geographic position (lat, lon, elev) and catchment characteristics (c_da, c_over, c_slp). Vectors illustrate input-variable correlations with the principal axes.

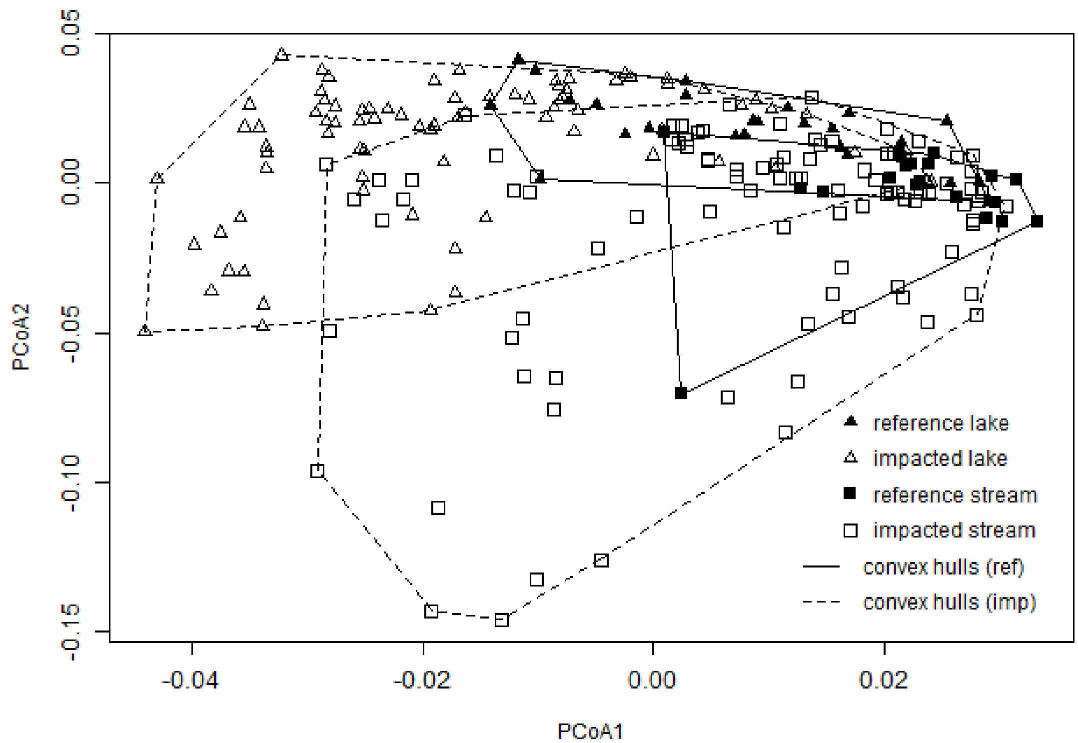

Figure 3: Principal coordinates analysis ordination of reference and impacted lakes and streams, based on Bray-Curtis distances among their taxa counts. Convex hulls (represented as solid or dashed lines) delineate each group's outer bounds in the plot.

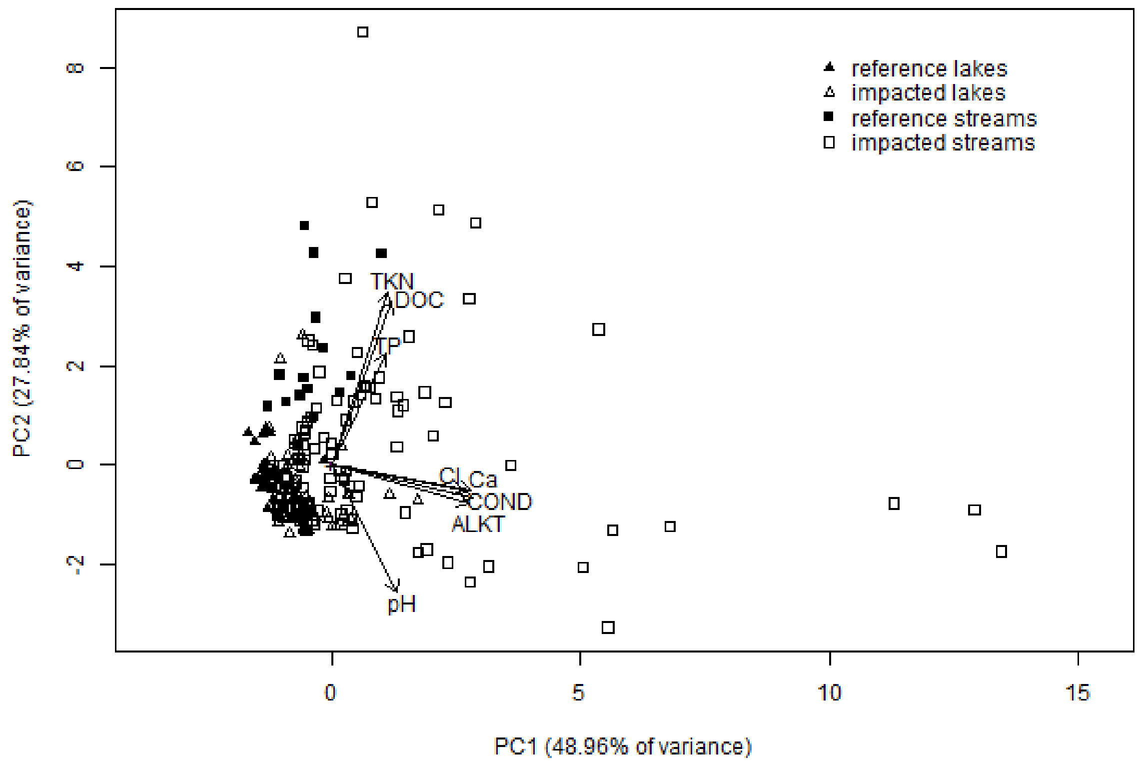

Figure 4: Principal components analysis ordination of reference and impacted lakes and streams, based on covariance among their chemical attributes (ALKT, Ca, Cl, COND, DOC, TKN, pH, and TP). Vectors illustrate input-variable correlations with the principal axes.

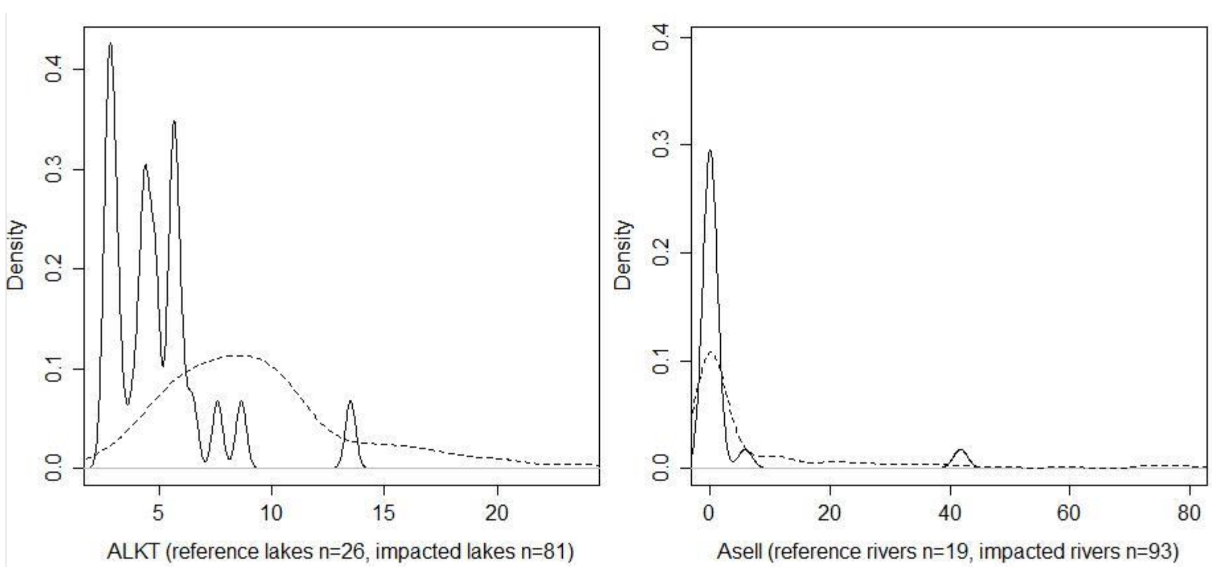

Figure 5: Probability density functions showing normal reference and typical impacted ranges (reference = solid lines, impacted = dashed lines) for an example chemical attribute (lakes, left pane) and biological attribute (rivers, right pane).

Electronic Supplementary Material

Online Resource 1: Datasets

Online Resource 2: R Scripts

Online Resource 3: Random forest and TITAN diagnostic plots

Online Resource 4: Ranges of stressor exposures, natural environmental features, taxa abundances, and biological index values for reference and impacted lakes and streams

Online Resource 5: Density plots showing variable distributions for reference and impacted waterbodies

Online Resource 6: Author contributions

PART 5: DECLINING AMPHIPOD ABUNDANCE IS LINKED TO LOW AND DECLINING CALCIUM CONCENTRATIONS IN LAKES OF THE SOUTH PRECAMBRIAN SHIELD

Declining Amphipod Abundance is Linked to Low and Declining Calcium Concentrations in Lakes of the South Precambrian Shield

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Abstract:

Calcium decline is an emerging stressor in boreal Shield lakes, with previously unknown implications for amphipod populations. Surveys of lakes in the Muskoka region (Ontario, Canada) have demonstrated both declining calcium concentrations and reduced abundances of amphipod taxa (Gammaridae and Hyalellidae) in low-calcium lakes. Temporally, amphipod abundances and calcium concentrations strongly declined between 1993 and 2016, and calcium was the most important chemical predictor of amphipod abundance. Spatially, general linear models showed calcium to be a moderately important predictor of amphipod presence/absence and abundance, however, associations between amphipods and calcium were complex. Amphipods were present in some lakes with calcium concentrations below 1 mg/l (the putative lower lethal threshold for *Gammarus*), and no threshold response was evident. This study corroborates evidence of widespread lake-water calcium declines reported by many authors, suggests that calcium levels have reached critically low levels in some lakes, and forecasts an average 57% decrease in amphipod abundances by the time calcium concentrations reach their predicted minima, which is expected to occur in most lakes between years 2023 and 2100.

Introduction

Amphipods are important in freshwater ecosystems as abundant members of several functional feeding groups (e.g., detritivores, predators; MacNeil et al 1999), as prey for fish and other animals (MacNeil et al. 1997), and as intermediate hosts for parasites (Kennedy 2006). These crustaceans have a heavily calcified integument and a regular moult cycle (Cairns and Yan 2009), which results in a high demand for calcium (Ca).

Ca is obtained directly from ambient water (Wheatley 1999, Cairns and Yan 2009) via active transport, which maintains internal Ca concentrations at levels that are elevated (Sutcliffe 1984) relative to the animal's dilute surroundings (Robertson 1960, Evans 2008). The rate of Ca uptake increases logarithmically with ambient concentration, up to a saturation point, above which the rate of influx is constrained physiologically (Greenaway 1974, Wright 1979). When Ca concentrations decline, metabolic rates must increase to satisfy the energy demands of Ca homeostasis. Very low Ca concentrations result in metabolic stress, reduced abundances and body sizes, slow growth and delayed maturity, reduced tolerance of environmental stressors, and mortality (Meyran 1997, Rukke 2002, Jeziorski et al. 2008, Cairns and Yan 2009, Edwards et al. 2016).

Declining Ca has been reported for many freshwater lakes, particularly those with a history of high acid deposition and intensive logging (Keller et al. 2001, Jeziorski et al. 2008, Jeziorski and Smol 2017, Lamothe et al. 2018). For example, Jeziorski et al. (2008) reported an average 13% decline in Ca concentrations in 36 lakes sampled between 1985 and 2005 in the Muskoka Region of the south Precambrian Shield. In a

2012-2013 survey of lakes from this same region, Jones (2017) observed a maximum Ca concentration of 9 mg/l, which is well below the saturation point for *Gammarus* (~20 mg/l; Cairns and Yan 2009). Furthermore, 93% of the lakes were below 5 mg/l, and 5% were below 1 mg/l (the estimated lower lethal limit for *Gammarus*; Cairns and Yan 2009). These results raise concerns that amphipods may be living under chronic metabolic stress in virtually all the area's lakes, and may be under threat of extirpation from a small proportion of them.

There is well-documented evidence that declining Ca is affecting crustaceans such as daphniids (Jeziorski et al. 2008, Shapiera et al. 2012) and mounting evidence for crayfish (Edwards et al. 2009, Edwards et al. 2013, Hadley et al. 2015); but, prior to our study, little evidence links low and declining lake-water Ca concentrations with declining abundances of amphipods (Zehmer et al. 2002, Cairns and Yan 2009). Bowman et al. (2014), did find the abundance of *Hyalella azteca* to be more strongly correlated with Ca than with pH in a series of acidified lakes in Nova Scotia, Canada, and Zehmer et al. (2002) showed that low Ca concentrations at the time of ecdysis limited the ability of *Gammarus pseudolimnaeus* to colonize and persist in dilute headwater streams draining Virginia's coastal plain.

We use a spatial survey of 107 lakes in the Muskoka region of the south Precambrian Shield, Canada (Figure 1), to address the following questions: Are amphipods more likely to be found in high-Ca lakes than in low-Ca lakes? Is a low-level Ca threshold suggested, below which amphipods are unable to persist in lakes? What is the relative importance of Ca as a predictor of amphipod abundance, relative to the importance of other chemical, morphometric, or physical-habitat factors? We use a long-term monitoring study of 19 lakes in the same region to evaluate whether amphipod abundances have declined over the last 23 years, and if so, to quantify the role of calcium in these declines, and to forecast how abundant amphipods will be in the future should Ca trends continue.

Methods

Spatial Survey

Amphipods were collected as part of a benthic macroinvertebrate survey of the 5660 km^2 Muskoka River Watershed. This region of shallow glacial till and Precambrian Shield bedrock contains 647 lakes greater than 8 ha in area (Ontario Ministry of Natural Resources 2011), many of which are dilute and nutrient-poor, with low acid-neutralization capacity (Dillon et al. 1987).

In July and August of 2012 and 2013, we sampled water and benthic invertebrates in 107 study lakes (Figure 1), selected using the partially randomized design detailed by Jones et al. (2017). Invertebrates and their associated sediments were collected (then immediately preserved in alcohol) at three randomly selected shoreline areas in each lake using the kick-and-sweep method of Jones et al. (2007). A surface sample of lake water was also collected at each sampled location. These three water samples were combined into a single composite and analyzed for Ca, total phosphorus (TP), dissolved organic carbon (DOC), and pH (Ontario Ministry of Environment 1983). Estimates of the median sizes of pavement-layer substrate particles (PCmed) present on transects where benthic invertebrates were collected were quantified as median-axis lengths of 10 randomly selected particles, as per Stanfield (2007). The relative abundances of submerged aquatic macrophytes (Ms), emergent aquatic macrophytes (Me), and rooted floating aquatic macrophytes (Mrf) were characterized visually using an ordinal classification (*absent, present, or abundant*). Lake areas were estimated using digital maps (Ontario Ministry of Natural Resources 2011).

Approximate 100-counts of invertebrates (including hydras, flatworms, nematodes, aquatic earthworms and leeches, crustaceans, mollusks, mites, and insects) were obtained from each sample. These animals were sorted, identified (family-level taxonomy), and counted (with the aid of a stereo microscope) from random aliquots, which were extracted from a 100-cell Marchant-style (e.g., Marchant, 1989) sub-sampling box. For each sample, the aliquot containing the 100th animal was entirely processed, meaning that total counts were often higher than the nominal 100. For the sake of brevity, we refer in this article to *abundances* of amphipods; however, these abundances are more properly described as *relative abundances*, because counts of these animals were quantified from fixed-count subsamples, relative to the counts of other major groups comprising multi-taxa benthic assemblages. Additional details of sampling methods were described by Jones et al. (2017).

Temporal Survey

Consistent methods (i.e., David et al. 1998) were used to sample littoral benthic macroinvertebrate communities annually at five shoreline locations in each of 19 Lakes (Figure 1; site selection details are provided by Reid et al. 1995): Blue Chalk (BC), Bigwind (BW), Chub (CB), Clear (CL), Crosson (CN), Cradle (CRDL), Dickie (DE), Delano (DO), Harp (HP), Hamer (HR), Heney (HY), Plastic (PC), Pincher (PNCR), Red Chalk East (RCE), Red Chalk Main (RCM), Ridout (RDT), Westward (WD), and Young (YG). Sampling occurred annually during October or November from 1993 to 2016, although not all lakes were sampled each year (Figure 4 and Figure 5). Lake areas and abundances of aquatic plants in the benthic-invertebrate collection areas were estimated as in the Spatial Survey. Dominant substrate particle types were classified on an ordinal scale, with 7 size-based classes, ranging from clay to bedrock.

Sweep-net samples of invertebrates were transported live and were kept cool for a maximum of 48 hrs until they could be washed in a 500-micron sieve. Teaspoon-sized aliquots of the sieved sediments were then transferred to a white enamel pan, from which they were searched through without visual aid until ~100-count subsamples of invertebrates were obtained.

In most cases, water samples for the 19 temporal survey lakes were collected during spring turn-over (i.e., April or early May) as mid-lake depth composites; however, three remote lakes (Cradle, Delano, and Pincher) could not always be accessed at this time of year. Where necessary, we augmented the missing springtime Ca data with data from surface grabs collected in July or October.

Additional details of chemical sampling methods are described by Ingram et al. (2017).

Statistical Analyses

To investigate whether amphipod families were more likely to be found in high-Ca lakes than in low-Ca lakes, we transformed amphipod abundances in the 107-lake spatial dataset (Electronic Supplement #1) to presences/absences, and modeled them using binomial generalized linear models (GLMs). These models allowed us to test whether Ca was a significant determinant of the presence of Gammaridae, Crangonyctidae, and Hyallelidae. The GLMs also allowed us to estimate the minimum Ca concentration at which amphipod absence was predicted to occur.

We also used binomial GLMs to investigate the importance of Ca as a predictor of amphipod abundances, relative to several other predictors we hypothesized could influence amphipod abundance: Cl (a surrogate for the density of paved roads and urbanization in the catchment), DOC (influences water clarity and lake thermal properties, such as depth to thermocline), TP (a measure of lake productivity), pH (a Ca correlate and indicator of chemical recovery from acid rain), lake area (a general hydrologic and morphometric predictor, and surrogate for lake depth and watershed position), PCmed (often a critical habitat attribute for benthic organisms), and Me, Mrf, and Ms (various measures of abundance of aquatic macrophytes, which are a food source for benthic detritivores and provide cover from predators). Lake area was log-transformed to normalize its extremely right-skewed distribution, and all predictors were range standardized (0 –1) so parameter estimates (i.e. regression coefficients) could be directly compared.

Thomson Lake was excluded from all the GLMs described above, because its high and outlying Ca levels (>9 mg/l) biased parameter estimates and weakened model fit. Confidence intervals about estimates of model parameters were calculated using the Profile Likelihood method (Stryhn and Christensen 2003), as implemented in the confint.glm() function from the MASS package (version 7.3-47) for R (R Core Team 2016). An R^2 statistic was calculated for each model as per Zhang (2016), using the rsq() function in the rsq package (version 1.0.1). The significance of parameter estimates was assessed by comparing z-ratios of the parameter estimates against a normal reference distribution (confidence limits were also calculated, as a measure of precision). GLM models were fitted, and significance tests were conducted, in R 3.4.2 (R Core Team 2016).

We used binomial generalized linear mixed effects models (GLMMs; e.g., Bolker et al. 2008) to investigate changes in amphipod abundances that occurred in the 19 lakes sampled between 1993 and 2016 (Electronic Supplement #1), and to estimate the effects of Ca and other water quality variables as predictors of these changes. We used linear mixed effects models (LMMs) to investigate changes in Ca concentrations over time. All predictors were range standardized, and models were fitted using lme4 (version 1.1-15; Bates et al. 2014).

Mixed-effects models allowed us to quantify potentially important sources of random variation that were expected in our hierarchical sampling design. These sources of variation included repeated sampling of lakes over time, multiple sampled locations within each lake, and habitat differences between sampled locations. The structure of the random effects was identical in all GLMM models: lakes and sampled locations (which were nested within lakes) were used as grouping factors; and within each level of these grouping factors, time (represented as monitoring years 1 – 23), percent of quadrat floor as organic material (qf.organic), and percent of quadrat floor as fine inorganic particles (i.e., silt, sand or clay; qf.small) were treated as random slope parameters. To alleviate overdispersion, we included an observation-level random effect (i.e., a supplementary random variation term; Harrison 2015) in each GLMM. The random effect in our LMM of Ca concentrations was time, which was grouped only by lake (i.e., not by lake and within-lake sampled location), given that chemistry-samples were whole-lake composites.

Two analogues to the coefficient of determination (R^2) were calculated: one as a marginal R^2 (R^2_m, which quantified variance explained only by fixed effects); and one as a conditional R^2 (R^2_c, which quantified variance explained collectively by fixed *and* random effects; Nakagawa and Schielzeth 2013). Calculating both statistics allowed us to estimate variance components associated with random and fixed effects, so the importance of within- and between-lake variation (random effects) could be assessed relative to the importance of Ca concentration or time (fixed effects).

To investigate whether amphipod abundances and Ca concentrations changed over time in the temporal series of lakes, GLMM and LMM models, respectively, were fit with time as both a fixed and random variable, and with the same random effects as described above. Fitting time as both a fixed and random effect allowed us to simultaneously investigate whether regional (all-lake) patterns in amphipod abundance and Ca were evident (i.e., time as a fixed effect) and which specific lakes demonstrated the greatest changes (i.e., time as random effect). Because the significance of individual random effect levels cannot be tested, we alternatively investigated lake-specific changes by scrutinizing the 95% confidence intervals of their slope parameters. To do so, a lake-specific mean slope was calculated as the fixed effect slope parameter, to which that lakes' conditional mean random-effect slope was added. The standard error for each lake was calculated as the standard error of the fixed effect slope parameter, plus the standard error of the conditional mean of the lake-specific random effect slope. The 95% confidence interval for each lake was calculated as that lake's mean slope plus and minus 1.96 times its standard error. Amphipod populations were considered to have changed in any lake having slope-parameter confidence intervals that did not include 0. Amphipods were present in so few of the samples from Pearceley Lake (the lowest Ca lake in the temporal dataset) that it had to be excluded from the GLMMs (including it biased parameter estimates and weakened model fit).

We used the Ca LMM and a simplified amphipod GLMM to predict future Ca levels and corresponding amphipod abundances under a scenario in which observed lake-specific 1993-2016 Ca trends (i.e., Ca slopes reported in Table 4) were projected into the future. One of two alternative stopping rules was used when projecting each lake's future Ca concentrations: the final Ca concentration was deemed to have been reached either when the projected Ca concentration declined to a level that was 40% below the mean value observed between 1993 and 2016, or when the length of the time series reached 84 years (i.e., extended out to 2100). Notwithstanding uncertainties around future development, acid deposition, and forest harvesting, these stopping rules were intended to make the projection as realistic as possible considering local weathering rates and the 10 % – 40 % declines in lake-water Ca concentrations expected in Muskoka-area lakes (Watmough and Aherne 2008). Projected Ca concentrations from the LMM were used as inputs for the GLMM so that corresponding amphipod abundances could be predicted. The GLMM included Ca as both a fixed and random effect (as a random effect, Ca was grouped by sampled location, and these sampled locations were nested within lakes) nested within lakes also as random effects). Pearceley lake was omitted from this analysis because no amphipods had been collected from that lake since 2004.

R scripts used for all statistical analyses are reproduced in Electronic Supplement #2.

Results

Based on the 107-lakes dataset, the amphipod families Gammaridae and Hyalellidae were significantly more likely to be found in high-Ca lakes than low-Ca lakes (Table 1, Figures 2 and 3); however, the Hyalellidae were absent from only 7 lakes (Figure 3), so it is unlikely that the model accurately described low-Ca stress in this family. All three families were found in at least some lakes with Ca levels below 1 mg/l (Figures 2 and 3).

The GLMs of amphipod presence/absence yielded no evidence that any of the three amphipod families exhibited a low-level Ca threshold. Despite Ca being a significant predictor in models of Gammaridae and Hyalellidae, low R^2 statistics, which ranged from 0.009 (Crang) to 0.039 (Hyale), suggested that Ca was, on its own, too weak a predictor of amphipod presence to have a threshold effect (Table 1). Furthermore, the shapes of the curves describing these families' predicted occurrences in lakes at different positions along the Ca gradient were not consistent with a threshold response (Figure 3). A threshold response would be suggested, for example, by a sigmoidal curve with a steeply rising limb, straddled at upper and lower limits by relatively flat tails; however the curve for Crangonyctidae was approximately linear, for Hyalellidae was broadly asymptotic, and for Gammaridae was sigmoidal with a gradually ascending middle portion.

Notwithstanding lack of evidence for a low-level Ca threshold, GLMs of amphipod abundances demonstrated Ca to be a reasonably important predictor (Table 2). For example, out of the ten predictors included in the Gammaridae GLM, the absolute value of the parameter estimate for Ca ranked 7th (and was statistically significant). The other more important (and also significant) predictors suggested that abundances of Gammaridae decreased with increasing coverage of rooted floating macrophytes, and increased with increasing lake area, PCmed, pH, and Cl. The only predictors included in the Gammaridae model that were not significant were Me and TP. Ca was the second ranked predictor in the Crangonyctidae model, after TP (TP was the most important factor for Crangonyctidae but the least important for Gammaridae). In addition, Crangonyctidae abundances were negatively related to TP and Mrf, and positively related to DOC and area. Hyalellidae was the only amphiphod family for which abundances were negatively related to Ca concentrations (marginally significant, $p = 0.05$). Estimates for DOC, PCmed, and Mrf were also significant and negative, whereas Cl, pH, area, and Me were significantly and positively related to Hyalellidae abundance.

Our analysis of the 18-lake temporal dataset illustrated that Ca concentrations and amphipod abundances have both decreased during the 1993 to 2016 time period (Figures 4 and 5, and Table 3). The LMM showed that, on average, Ca has declined by 0.023 mg/l each year in the studied lakes (all lakes had negative Ca slopes, and confidence intervals for 14 of the 18 lakes did not include zero). GLMMs demonstrated that the odds of encountering an amphipod decrease each year by 3.4% year over year (confidence intervals for the amphipod slope parameter were less than zero for 8 of the 18 lakes; Table 4).

Besides demonstrating declining Ca and amphipod abundances, two peculiarities in the GLMM results were instructive. First, in the fully parameterized GLMM, Ca and Year were the only significant predictors, and estimates of fixed effects did not change significantly after additional chemical predictors (Cl, DOC, TP, and pH) were added to the model. Nonetheless, Akaike's Information Criteria was lowest for the fully parametrized model, which means that the additional chemical variables did contribute some information that was useful to the prediction of amphipod abundances. Second, in Harp lake, Ca declined over time (Figure 4), amphipod abundances increased over time (Figure 5), but the relationship between amphipod abundance and Ca was positive (Figure 6). These patterns illustrate that the year factor in the GLMM was not just a surrogate for time, but included variation introduced by different sampling crews, and probably represented multiple factors that influence interannual variation in amphipod abundances.

The models provided additional insights into the way that explained variation in Ca and amphipod abundances partitioned between fixed and random effects (Table 3), fixed effects characterizing the generalized regional pattern across all lakes included in the study, and random effects characterizing local within- and between-lake variation. In the Ca model, 8% of the explained variation was attributed to the year fixed effect (e.g., changing climate, human development and forestry, changing acid deposition or other factors driving lake Ca declines regionally; Watmough and Aherne 2008), and 77% of the variation was attributed to localized random effects (i.e., lake-specific processes). In the amphipod model, 2% of the variation was attributed to regional fixed effects (Year and Ca), and 11% was attributed to within- and between-lake effects.

Ca concentrations ranged from 1.22 to 2.68 mg/l, and amphipod abundances ranged from 11% to 44% during the 1993 – 2016 baseline period of the temporal monitoring study (Table 5). Although confidence intervals around future estimates of Ca and amphipod abundances were wide, 40% Ca declines from 1993-2016 baselines were predicted to occur in 13 of the 18 lakes by 2100 (or earlier). These reduced Ca concentrations were predicted to correspond with a mean 57% decrease in amphipod abundances (lake-specific means were forecasted to be between 3% and 25% by the end of the simulation period).

Discussion

We conclude that lake-water Ca levels declined between 1993 and 2016 in 14 of 18 lakes in the Muskoka region of the south Precambrian Shield, and a simultaneous reduction in amphipod abundances occurred in 8 of 18 lakes. Ca explained 13% of the variation in amphipod abundances, making it the most important chemical, morphometric or physical-habitat predictor of the changing abundances of this taxon. Nevertheless, the (fixed-effect) “year” parameter in our model was a more important predictor of amphipod abundance, which indicated that inter-annual processes (and potentially inter-crew variation and other unmeasured factors that co-varied with time) were also important drivers of amphipod abundance in the region’s lakes..

Models of amphipod presence/absence generated from a 107-lake spatial survey conducted in the Muskoka region demonstrate that amphipods in the families Gammaridae and Hyalellidae are more commonly present in Precambrian Shield lakes with high Ca concentrations than in lakes with low Ca concentrations. But, in contrast to these results, models of amphipod abundance using the same 107-lake dataset demonstrated Ca to rank between 3rd (Crangonyctidae) and 8th (Gammaridae) most important in a list of ten chemical, morphometric, or physical-habitat predictors, suggesting it is only a moderately important driver of amphipod abundance. We also demonstrate that all three common amphipod families, Crangonyctidae, Gammaridae, and Hyalellidae, can be found in some lakes with Ca concentrations below 1 mg/l — the lower lethal limit inferred for *Gammarus*, based on laboratory bioassays (Cairns and Yan 2009) — and none exhibited a threshold response to this critical nutrient.

Our analyses were undertaken using data from a spatial survey and a long-term monitoring study of mixed-taxa littoral lake benthic communities. Neither of these studies was specifically designed to investigate the relationship between amphipods and Ca; however, several lines of evidence indicate a complex association: random between- and within-lake effects were very important in our models, and many other variables were significant predictors (e.g. lake area, pH, Cl, TP, DOC, and the coverage of different types of macrophytes). These results suggest that environmental heterogeneity among lakes is an important factor mediating amphipod responses to changing Ca levels. Unexpected contradictions were also instructive. For example, the presence of Hyalellidae in our spatial-survey lakes was positively related to Ca concentration but its abundance was negatively related; Ca was not a significant predictor of the presence/absence of Crangonyctidae, but was among the most important predictors of its abundance; and amphipod abundances increased in Harp Lake between 1993 and 2016, despite declining Ca concentrations[1].

Our amphipod sampling methods were similar in the spatial and temporal surveys, but the season in which this sampling was undertaken differed between surveys (spatial sampling done in summer, temporal in fall). Incongruence of results from our spatial and temporal analyses (e.g., Ca being the most important chemical, morphometric or physical-habitat predictor of amphipod abundances in temporal models, but ranking only 3rd to 8th in the spatial models) could indicate that low-Ca stress varies seasonally. Although lake-water Ca concentrations are fairly consistent

[1] Our ability to investigate the association between Ca and amphipod abundance in Harp Lake may be partially confounded by invasion of this lake by *Bythotrephes longimanus* in the mid 1990s (i.e., Yan and Pawson 1997).

throughout the year, it is feasible that amphipod populations may not respond to low-Ca until it coincides with sensitive life history stages (e.g., post-moult; Rukke 2002), or when colder lake temperatures reduce metabolic rates, and limit organisms' abilities to maintain Ca homeostasis (Greenaway 1974, Wright 1979). Additional work to investigate the potential seasonality of Ca stress is appropriate.

Moreover, the mechanics and mitigating factors of Ca-limitation in amphipods could be illuminated by more focussed surveys or experiments aimed at evaluating a series of additional working hypotheses: (1) Inputs from streams or groundwater (e.g., Lottig et al. 2011) could establish Ca-rich littoral refuges that allow amphipods to persist in lakes with low pelagic Ca levels. Given that amphipods are also present in riverine habitats (Peckarsky 1990) and are common in stream drift (Elliott and Corlett 1972), it is also possible that colonization pressure from lake-inflow streams could substantially mitigate population effects of Ca decline in some lakes. (2) Many environmental changes are occurring in the region (Palmer et al. 2011), and stressor interactions are possible (Holmstrup et al. 2010, Altshuler et al. 2011, Fischer et al. 2013, Rollin et al. 2017). Amphipod responses to low Ca could be strongly mediated by temperature, food availability, food quality, or other environmental variables. (3) Lakes' historical contexts could also be important. Short-term increases in lake-water Ca occurred due to logging, development, acid rain (Watmough and Aherne 2008), and local use of Ca-rich dust suppressants on some roadways (Shapiera et al. 2012), and these increases have been followed by gradual declines. This begs the question of whether eco-evolutionary dynamics (e.g., Lallensack 2018) could be important, such that populations that have experienced relatively large fluctuations are more tolerant of Ca decline than populations that have experienced modest changes. (4) A question remains about whether low-Ca toxicity can be proven experimentally in any of the region's lakes. For example, Pearceley Lake — the lowest-Ca lake in our temporal dataset, where amphipods have not been collected in annual monitoring efforts since 2004 — would be a good candidate for Ca-addition or mesocosm experiments. (5) Differences in body sizes suggest that *Crangonyx, Gammarus,* and *Hyalella* could have different Ca requirements, and useful insights could be gained from the relative abundances of species in these genera.

Notwithstanding factors that mitigate low-Ca stress, should lake-water Ca levels continue to decline as expected (i.e., Watmough and Aherne 2008), we forecast that the relative abundances of amphipods in Precambrian Shield lakes will decline, on average, by 57% (range 7% [CRDL] to 81% [PC]), with many of these declines occurring before the end of the 2030s. A marked reduction in the abundances of amphipod crustaceans, which were once a numerically dominant component of the littoral fauna (mean abundance in the 1993-2016 period was 28%) signals a major restructuring of benthic communities; therefore, this study not only corroborates evidence of widespread lake-water Ca declines reported by Jeziorski et al. (2008), but also reinforces warnings by these (and other) authors regarding declining Ca levels being a threat to freshwater biota. As animals with high Ca demands (crustacean zooplankton, amphipods, crayfish) decline in abundance, the structural and functional effects of these changes could propagate through foodwebs to substantially affect softwater boreal ecosystems (Jeziorski et al. 2017).

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Figures

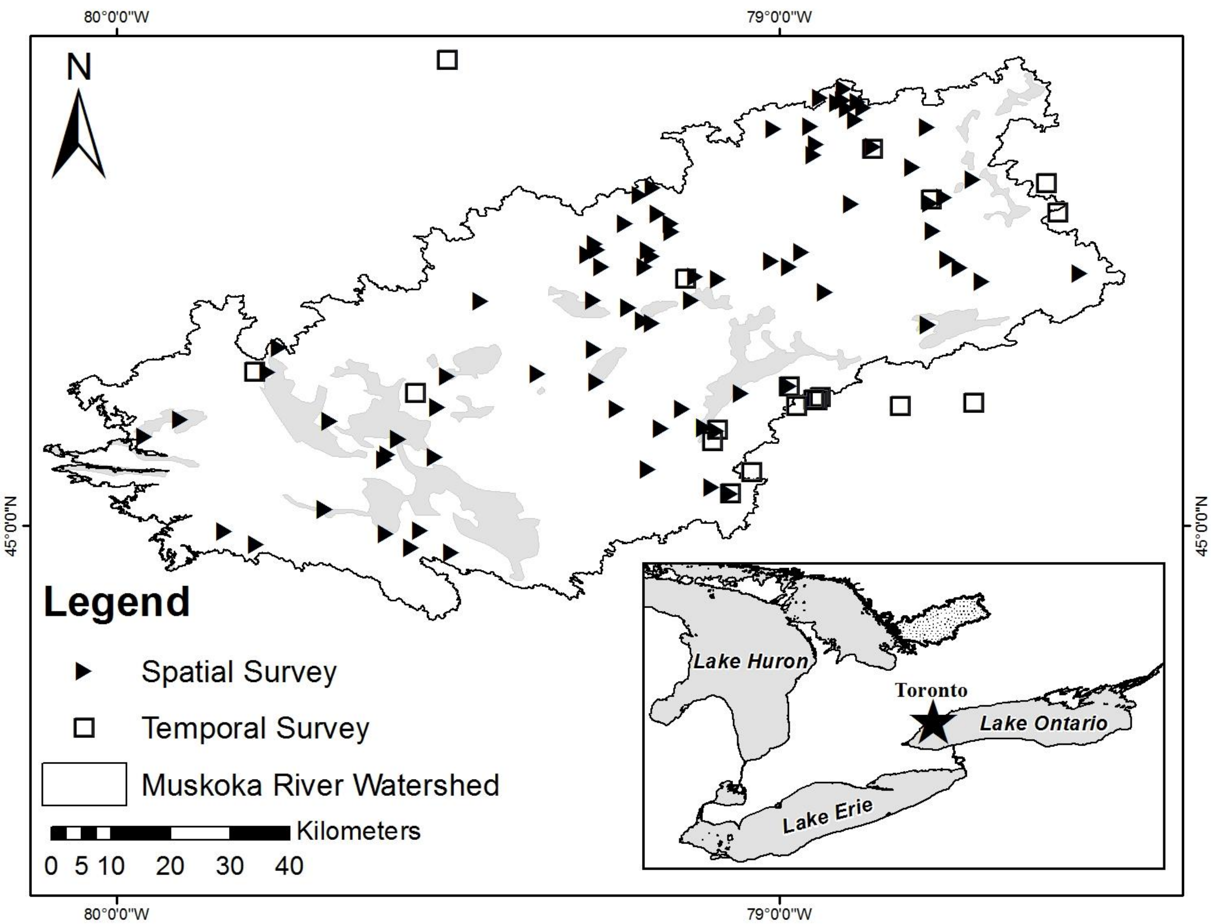


Figure 1: Study lakes, regional context.

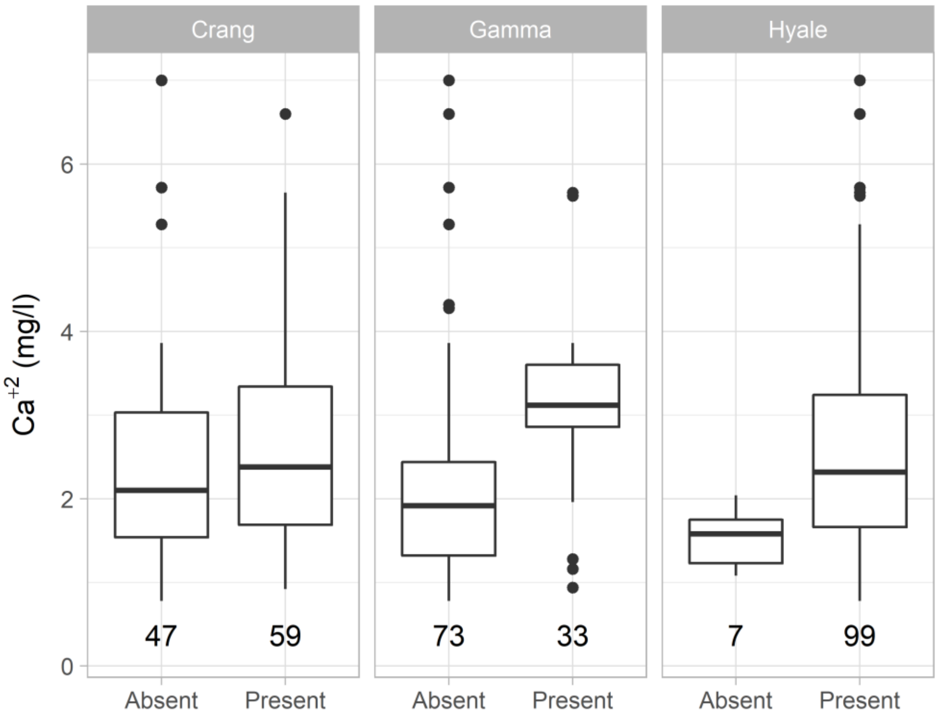


Figure 2: Ca levels in lakes from which amphipod families were collected (amphipods present) or were not collected (amphipods absent). In this box-and-whisker plot, boxes enclose the central 50% of each group's distribution, medians are demarcated as thick horizontal lines, and whiskers extend up and down to the highest and lowest values that are not more than 1.5 times the interquartile range above or below the box (more extreme values are shown as filled dots). Numbers represent the counts of observations in each series.

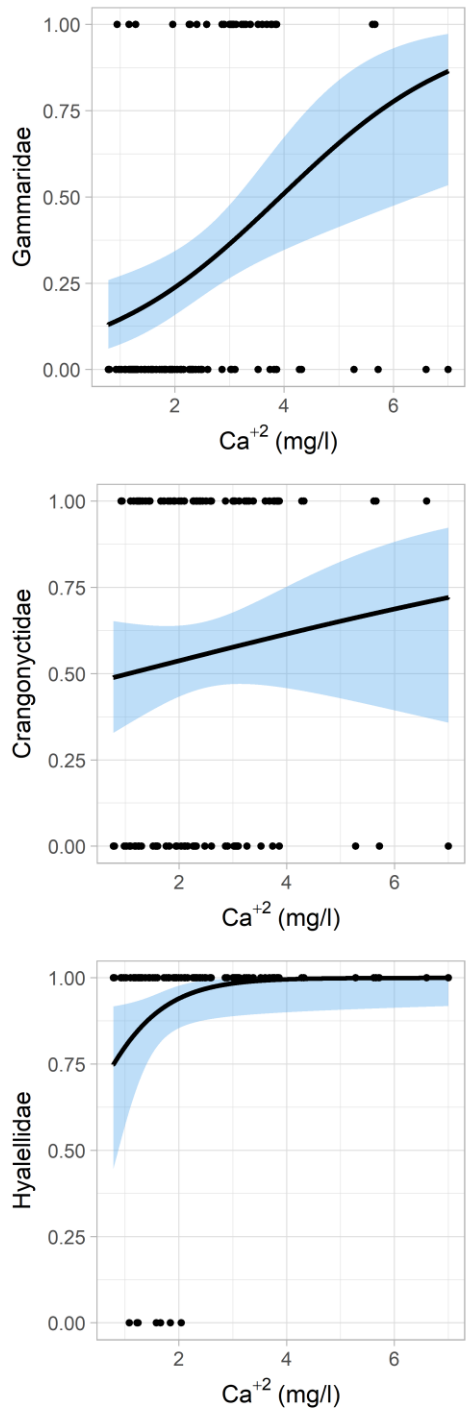


Figure 3: GLM-predicted probabilities of collecting three amphipod families in lakes relative to Ca concentrations represented in the 107-lakes spatial dataset. Blue shaded regions about the predicted values represent 95% confidence bounds around model estimates; dots represent individual data points across the sampled Ca gradient (those on the Y = 0 line represent lakes where the amphipod family was; those on the Y = 1 line represent lakes where the amphipod family was present).

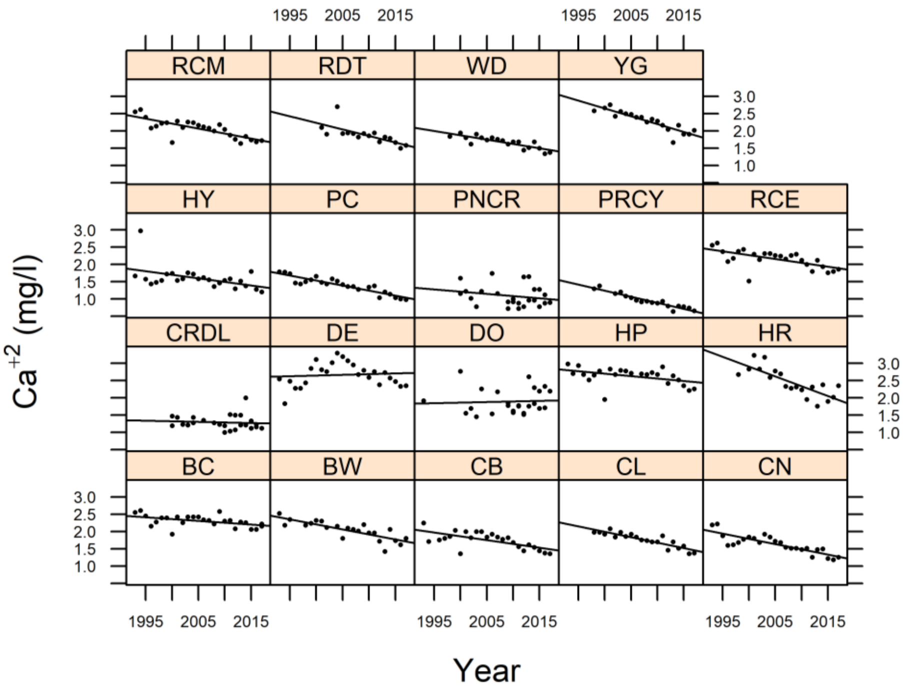


Figure 4: Relationship between lake-water Ca concentration and year, 19-lake temporal dataset.

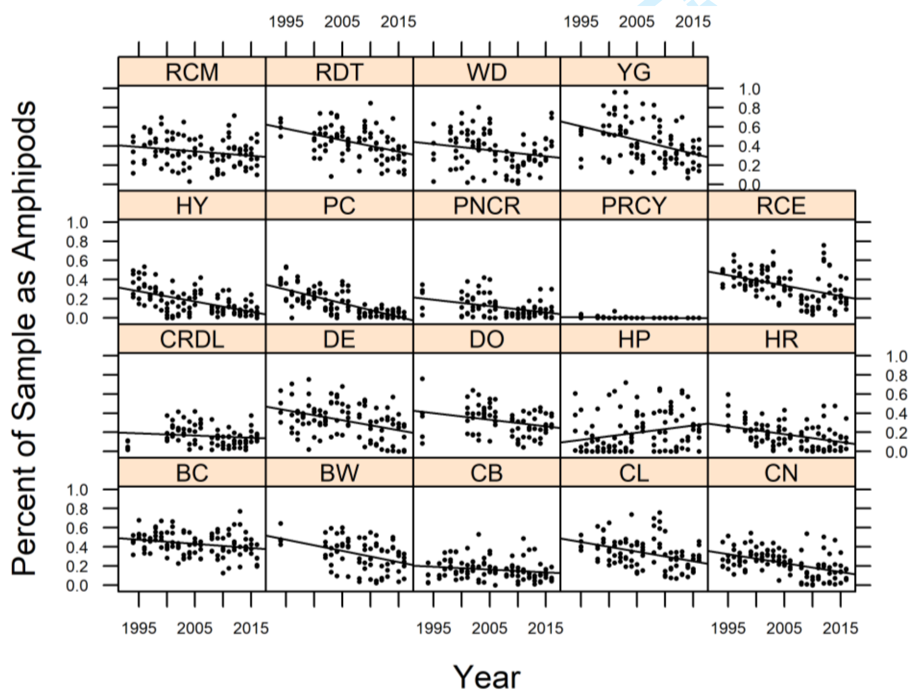
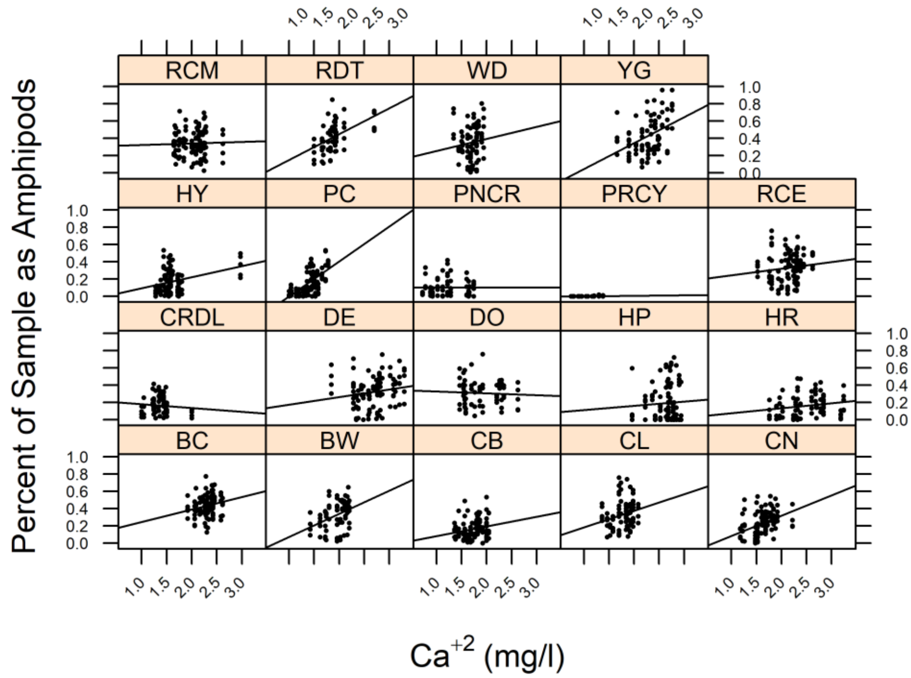


Figure 5: Relationship between amphipod abundance and year, 19-lake temporal dataset.



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592 Figure 6: Relationship between amphipod abundance and lake-water Ca concentration,
593 19-lake temporal dataset

Tables

Table 1: Summary of GLM statistics (presence/absence of amphipod families,107-lake spatial dataset; “.pa” = denotes presence/absence data; Gamma = Gammaridae, Crang = Crangonyctidae, Hyale = Hyalellidae; AIC = Akaike information criterion, SE = standard error; upper and lower confidence limits for parameter estimates are provided in parentheses).

Response	Predictor	Estimate	SE	p	AIC	R^2
Gamma.pa	Intercept	-1.894 (-2.782, -1.113)	0.422	<0.01	123.62	0.112
	Calcium	3.747 (1.555, 6.258)	1.187	<0.01		
Crang.pa	Intercept	-0.043 (-0.71, 0.615)	0.336	0.9	148.597	0.009
	Calcium	0.992 (-0.948, 3.073)	1.013	0.33		
Hyale.pa	Intercept	1.092 (-0.154, 2.457)	0.655	0.1	48.524	0.039
	Calcium	8.505 (1.821, 18.135)	4.082	0.04		

Table 2: Summary of GLM statistics (abundances of amphipod families,107-lake spatial dataset; Gamma = Gammaridae, Crang = Crangonyctidae, Hyale = Hyalellidae ; AIC = Akaike information criterion, SE = standard error; upper and lower confidence limits for parameter estimates are provided in parentheses).

Response	Predictor	Estimate	SE	p	AIC	R^2
Gamma	(Intercept)	-8.003 (-8.913, -7.144)	0.451	<0.01	1252.2	0.24
	Ca	1.11 (0.156, 2.073)	0.488	0.02		
	Cl	2.252 (1.037, 3.391)	0.599	<0.01		
	DOC	2.145 (0.594, 3.684)	0.788	<0.01		
	TP	0.726 (-0.225, 1.649)	0.478	0.13		
	pH	1.309 (0.587, 2.062)	0.376	<0.01		
	area	3.123 (2.444, 3.83)	0.353	<0.01		
	PCmed	1.51 (0.288, 2.685)	0.611	0.01		
	Me	-0.082 (-0.613, 0.452)	0.272	0.76		
	Mrf	-9.867 (-11.908, -8.045)	0.984	<0.01		
	Ms	1.96 (1.264, 2.673)	0.359	<0.01		
Crang	(Intercept)	-5.906 (-6.851, -5.02)	0.467	<0.01	576.1	0.27
	Ca	1.649 (0.424, 2.934)	0.638	<0.01		
	Cl	0.342 (-1.266, 1.692)	0.751	0.65		
	DOC	2 (0.048, 3.889)	0.981	0.04		
	TP	-2.21 (-3.803, -0.71)	0.789	<0.01		
	pH	0.726 (-0.251, 1.783)	0.518	0.16		
	area	1.03 (0.267, 1.805)	0.392	<0.01		
	PCmed	0.741 (-0.515, 1.907)	0.617	0.23		
	Me	-0.191 (-0.886, 0.496)	0.352	0.59		
	Mrf	-1.642 (-2.633, -0.719)	0.487	<0.01		
	Ms	-0.969 (-1.756, -0.188)	0.400	0.02		
Hyale	(Intercept)	-2.444 (-2.673, -2.218)	0.116	<0.01	2179.2	0.29
	Ca	-0.342 (-0.678, -0.008)	0.171	0.05		
	Cl	0.661 (0.378, 0.938)	0.143	<0.01		
	DOC	-1.26 (-1.758, -0.768)	0.253	<0.01		
	TP	0.218 (-0.14, 0.572)	0.182	0.23		
	pH	0.681 (0.366, 1)	0.162	<0.01		
	area	0.235 (0.013, 0.455)	0.113	0.04		
	PCmed	-1.997 (-2.385, -1.621)	0.195	<0.01		
	Me	0.737 (0.558, 0.917)	0.092	<0.01		
	Mrf	-0.87 (-1.085, -0.657)	0.109	<0.01		
	Ms	0.124 (-0.068, 0.317)	0.098	0.21		

Table 3: Summary of LMM (Calcium levels mg/l, 18-lake temporal dataset) and GLMM statistics (abundance of amphipods, 18-lake temporal dataset; AIC = Akaike information criterion, SD = standard deviation, SE = standard error, R^2_m = marginal R^2 [quantified variance explained only by fixed effects], R^2_c = conditional R^2 [quantified variance explained by fixed and random effects]; upper and lower confidence limits for fixed-effect parameter estimates are provided in parentheses).

		Fixed Effects			Random Effects					
Response	Parameter	Estimate	SE	p	Group	Parameter	SD	AIC	R^2_m	R^2_c
Calcium	Intercept	2.579 (2.283, 2.875)	0.151	<0.01	Lake	Intercept	0.62	35.0	0.08	0.85
	Year	-0.023 (-0.03, -0.016)	0.003	<0.01		Year	0.01			
Percent Amphipoda	Intercept	-1.087 (-1.331, -0.843)	0.125	<0.01	Rep:Lake	Intercept	0.24	13846.2	0.01	0.12
	Year	-0.822 (-1.195, -0.448)	0.191	<0.01		Year	0.70			
						qf.organic	0.25			
						qf.small	0.31			
					Lake	Intercept	0.47			
						Year	0.80			
						qf.organic	0.46			
						qf.small	0.30			
Percent Amphipoda	Intercept	-1.299 (-1.595, -1.004)	0.151	<0.01	Rep:Lake	Intercept	0.23	13083.5	0.02	0.12
	Year	-0.803 (-1.185, -0.421)	0.195	<0.01		Year	0.73			
	Ca	0.5 (0.149, 0.852)	0.179	<0.01		qf.organic	0.29			
						qf.small	0.34			
					Lake	Intercept	0.39			
						Year	0.79			
						qf.organic	0.74			
						qf.small	0.59			
Percent Amphipoda	Intercept	-1.138 (-1.56, -0.716)	0.215	<0.01	Rep:Lake	Intercept	0.20	12582.3	0.02	0.13
	Year	-0.843 (-1.255, -0.432)	0.210	<0.01		Year	0.74			
	Ca	0.478 (0.062, 0.893)	0.212	0.02		qf.organic	0.29			
	TP	-0.101 (-0.435, 0.234)	0.171	0.56		qf.small	0.34			
	pH	-0.272 (-0.658, 0.113)	0.197	0.17	Lake	Intercept	0.49			
	Cl	-0.208 (-0.833, 0.418)	0.319	0.52		Year	0.83			
	DOC	0.33 (-0.436, 1.097)	0.391	0.40		qf.organic	0.70			
						qf.small	0.62			

Table 4: Trends in amphipod abundances and lake-water Ca concentrations, 18-lake temporal dataset (upper and lower 95% confidence limits for slopes are provided in parentheses; where confidence limits for slopes do not include zero, the sign of the trend is indicated)

Lake	Amphipod Slope	Trend	Ca Slope	Trend
BC	-0.239 (-2.011, 1.533)		-0.013 (-0.027, 0.002)	
BW	-1.015 (-1.864, -0.165)	-	-0.028 (-0.043, -0.013)	-
CB	-0.550 (-1.373, 0.274)		-0.022 (-0.037, -0.007)	-
CL	-0.652 (-1.464, 0.159)	-	-0.029 (-0.044, -0.014)	-
CN	-1.218 (-2.107, -0.330)		-0.030 (-0.044, -0.015)	-
CRDL	-0.643 (-1.988, 0.703)		-0.009 (-0.024, 0.006)	
DE	-1.030 (-1.963, -0.098)	-	-0.001 (-0.016, 0.013)	
DO	-0.453 (-1.400, 0.494)		-0.003 (-0.018, 0.012)	
HP	1.264 (0.443, 2.085)	+	-0.016 (-0.031, -0.002)	-
HR	-1.330 (-2.179, -0.482)	-	-0.046 (-0.062, -0.030)	-
HY	-1.512 (-2.588, -0.435)	-	-0.021 (-0.036, -0.006)	-
PC	-2.425 (-4.295, -0.555)	-	-0.028 (-0.042, -0.013)	-
PNCR	-1.233 (-2.091, -0.374)	-	-0.016 (-0.031, -0.001)	-
RCE	-0.956 (-1.851, -0.060)	-	-0.023 (-0.037, -0.008)	-
RCM	-0.344 (-1.216, 0.528)		-0.030 (-0.043, -0.013)	-
RDT	-0.644 (-1.628, 0.341)		-0.031 (-0.048, -0.014)	-
WD	-0.526 (-1.880, 0.828)		-0.024 (-0.040, -0.008)	-
YG	-0.822 (-2.096, 0.451)		-0.038 (-0.053, -0.022)	-

Table 5: Future Ca concentrations and amphipod abundances projected from 18-lake temporal dataset ([Ca] = lake-water Ca concentration; upper and lower 95% confidence limits in parentheses)

Lake	1993 - 2016 mean observed [Ca] (mg/l)	[Ca] corresponding with 40% reduction from 1993-2016 mean (mg/l)	Year 40% [Ca] reduction reached	1993 - 2016 mean observed amphipod abundance (%)	Predicted amphipod abundance at minimum simulated [Ca] (%)
BC	2.30	1.38	2098 (2038, >2100)	43	23 (4, 70)
BW	1.93	1.16	2039 (2019, 2080)	32	14 (2, 53)
CB	1.73	1.04	2045 (2018, >2100)	16	9 (1, 42)
CL	1.78	1.07	2030 (2017, 2055)	34	18 (3, 62)
CN	1.63	0.98	2028 (2017, 2051)	23	11(2, 46)
CRDL	1.35	0.81	>2100 (2043, >2100)	16	15 (2, 59)
DE	2.68	1.61	>2100 (>2100, >2100)	32	19 (3, 64)
DO	1.90	1.14	>2100 (>2100, >2100)	31	24 (4, 70)
HP	2.63	1.58	>2100 (2046, >2100)	20	14 (2, 53)
HR	2.47	1.48	2023 (2017, 2031)	17	11 (2, 43)
HY	1.61	0.97	2035 (2017, 2077)	17	10 (1, 47)
PC	1.40	0.84	2025 (2017, 2048)	15	3 (0, 19)
PNCR	1.22	0.73	>2100 (2023, >2100)	11	9 (1, 44)
RCE	2.14	1.28	2047 (2022, >2100)	33	21 (3, 67)
RCM	2.04	1.23	2036 (2017, 2065)	34	25 (4, 72)
RDT	1.90	1.14	2029 (2017, 2050)	43	19 (3, 65)
WD	1.68	1.01	2036 (2017, 2077)	35	22 (3, 69)
YG	2.28	1.37	2026 (2017, 2037)	44	18 (3, 61)

Electronic Supplements

ES#1: Datasets

ES#2: R Scripts

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PART 6: SUMMARY, CONCLUSIONS AND DISCUSSION

Summary

A tense opposition is apparent in Earth’s boreal ecosystems. On one hand, the boreal region has expansive natural areas and valuable resources (such as timber, fossil fuels, minerals, fisheries, and waterfront real estate; Luke et al 2007). On the other hand, human influence is pervasive, and threats of additional development and resource exploitation are unrelenting (Dubé et al. 2006, Sorensen et al. 2008, Houle et al. 2010, Seitz et al. 2011).

Cumulative effects assessment (CEA) has been viewed as a way to promote precaution in the land use planning and development process and to facilitate sustainability (Benson 2003, Gunn and Noble 2009, Gunn and Noble 2011); however, its execution is frequently stalled due to a lack of clarity about what cumulative effects assessment is and how it is best implemented (Duinker and Grieg 2006, Gunn and Noble 2011), and because the necessary monitoring systems and cumulative effects models are rudimentary (Duinker and Greig 2007, Ball et al. 2013).

Water is essential for life, and access to sufficient clean water is commonly viewed as a human right (Gupta et al. 2010). Nonetheless, lakes and rivers are exposed to (Nõges et al., 2016), and integrate effects of, many stressors (Lowell et al., 2000, Townsend et al., 2008, Ormerod et al. 2010, Jackson et al. 2016). Indeed, several authors have considered aquatic ecosystems to be among the most threatened ecosystems on Earth (e.g., Schindler 2001, Carpenter et al. 2011). Given the universal and incomparable importance of water, the quality of lake and river water, and the biological condition of lake and river ecosystems are critical considerations for CEA.

The Muskoka River Watershed is a microcosm of the boreal region, and serves as a useful case-study. In this watershed, the regional planning authority (the District Municipality of Muskoka) and other regulatory agencies (for example Ontario's Ministries of Environment and Climate Change, and Natural Resources and Forestry), have been collaborating with several academic institutions to design a cumulative effects monitoring system and initiate a process for CEA (CWN 2015, Eimers 2016). My intent is to provide these agencies with requisite knowledge.

The thesis begins by synthesizing a conceptual model for CEA. Subsequent chapters describe and model lake and stream water quality, benthic community structure, and the cumulative effects of natural environmental factors and human land-use (urbanization and agriculture in particular). Specifically, I establish that monitoring and modeling are essential to CEA, and clarify where these techniques fit into the CEA process. I evaluate a series of candidate monitoring indicators by determining which ones can be best modeled, what human and natural factors they respond to, and whether they are sensitive enough to predict outcomes of alternative scenarios of human activity. From this assessment, a short list of chemical and biological indicators that should be incorporated in a regional cumulative effects monitoring program is suggested.

Furthermore, I objectively differentiate reference and impacted conditions, either using a criterion of no exposure to land-use stress or an empirical break-point where stressor exposures begin to cause measurable chemical or biological effects. Such criteria allow a series of assessment benchmarks to be reported as normal reference ranges for biological or chemical indicators of lake or stream condition.

I quantify cumulative effects by modeling spatial and temporal patterns of chemical and biological conditions across multiple gradients of natural environmental features and human activity, which identifies a variety of ecological issues that are relevant to CEA. As an example to illustrate the potential complexity of CEA in the boreal region, I also explore the association between lake-water calcium concentration, the abundance of amphipods (a freshwater crustacean with a high demand for calcium), and local factors that mediate the calcium-amphipod relationship.

Although the focus of this thesis is on lakes and streams of the southern Boreal Shield, its concepts and numerical methods are universal: they are equally applicable to regions outside the Muskoka River Watershed, even though human interests and environmental priorities may be quite different in these different localities.

Conclusions and Discussion

My review of the environmental-appraisal literature established 6 key themes that encapsulate CEA and its role as a precautionary instrument for sustainability: (1) Human wellbeing ultimately depends on the degree to which the human enterprise conforms to the limitations of Earth's biosphere (Wackernagel and Rees 1998, Schnaiberg and Gould 2000). (2) Cumulative effects are combined ecosystem changes that result from a combination of human activities and natural processes (Scherer 2011). (3) CEA is a sub-discipline of environmental impact assessment, and a complement to municipal planning (Morgan 2012, Morrison-Saunders et al. 2014). Its objectives are to formalize environmental values, establish a sustainable socio-economy, improve knowledge and governance (Culhane 1990, Schultz 2012, Pope et al. 2013), and predict the consequences of development, relative to an assessment of present ecological condition (Dubé et al. 2003). CEA is particularly well suited to forecasting trade-offs associated with alternative scenarios of human activity. Given these objectives, it is best undertaken in a strategic and regional context (e.g., at the watershed scale; Seitz et al. 2011), rather than at the conventional scale of individual development projects (Gunn and Noble 2011). (4) As objects of its assessment, CEA requires valued ecosystem components to be specified. These components set a tractable scope, and focus attention on the most important environmental features and processes (Damman et al. 1995, Canter and Ross 2010); however, objective criteria for guiding *other* decisions that need to be made in the CEA process (e.g., about the spatial or temporal bounds of the assessment, or the scope of issues to be considered) do not exist. (5) Monitoring and adaptive management are essential strategies within CEA for mitigating risks of uncertainty (Karkkainen 2002). And (6) our lack of basic knowledge about stressors and their effects is a barrier to the development of models that assess and predict cumulative effects (Duinker and Greig 2007).

Chapter 2 (Random Forests as Cumulative Effects Models: A Case Study of Lakes and Rivers in Muskoka, Canada) provided two key insights: (1) Random forest models (Breiman 2001) describe the combined and singular effects of multiple stressors and natural factors, and can be used to evaluate the suitability of candidate indicators for use in monitoring schemes and scenario analyses. (2) A variety of chemical and biological measures of ecosystem condition can be modeled accurately enough, and are sensitive enough to land-use, to be included in a cumulative effects monitoring system and considered in futuring exercises.

In the case-study watershed, the most accurately modeled biological indicator for lakes was an axis score from a principal coordinates analysis ordination of the relative abundances of the benthic taxa. The best modelled biological indicator for rivers was the relative abundance of Asellidae (a family of Isopod crustaceans, considered tolerant of a variety of chemical pollutants; Hilsenhoff 1988, Mandaville 2002, Oguma and Klerks 2017). The most accurately modeled chemical indicator for lakes was alkalinity, and for rivers was conductivity. In general, chemical effects were modeled more accurately than biological effects. As a rule, biological effects were modeled more accurately for lakes than for rivers, but chemical effects were modeled more accurately for streams than for lakes.

CEA practitioners should derive some confidence in indicators, and some optimism for the potential of futuring analyses, given that several chemical and biological indicators were sensitive enough to detect simulated land-use changes. All 13 biological indicators that I tested (including several measures of taxonomic composition, richness, taxa tolerances and whole-community ordination-based metrics) were capable of detecting a doubling of urbanization in the catchment. Five of thirteen (including measures of taxonomic composition and axis-scores from an ordination of taxa counts) were capable of detecting a doubling of agricultural intensity in the Highway 11 Strip physiographic region. These differences in the detectability of stressor exposures agree with several published studies that demonstrated urbanization effects as more severe than agricultural effects (e.g., Allan 2004, Poff et al. 2006). All indicators except Hilsenhoff's Family Biotic Index were better able to detect urbanization or agricultural effects in lakes than in streams.

Chemical and biological indicators were typically influenced by a complicated array of predictors, representing environmental factors operating at several different spatial scales, from the immediate vicinity of the sampled location, to the riparian zone, to the local catchment, to the cumulative catchment. Chemical, morphometric, hydrologic and land-use/land cover predictors were more important in biological models than geographic variables or measures of physical habitat at the sampled locations. Measures of land-use and land cover were of greatest importance to the water-chemistry models. Furthermore, variables describing attributes of the cumulative catchment or physical characteristics of the sampled locations tended to be most important in biological models; whereas those measured at the cumulative catchment scale were the most important drivers of chemical indicators. These findings should be considered when selecting monitoring indicators, and when choosing which effects to attempt to forecast. They also serve as a reminder that one's vantage point is consequential: our ability to understand the relative importance of local and regional effects on water chemistry and community structure depends both on how we define local and regional communities, and at what scale the regulatory mechanisms operate (Weiher et al. 2011).

My models of metric values demonstrated that random forests provide useful insights about the performance of candidate indicators. From the perspective of metric selection, the pseudo-R^2 statistic is informative because it provides a direct measure of predictive accuracy or explained variance. Variable importances and partial dependencies reveal what a given metric actually indicates — i.e., they provide information about which stressors and natural environmental factors the indicator responds to. Such evidence is needed so that reference-site-selection and bioassessment methods can be fine-tuned to judge ecological condition in a way that is minimally confounded by natural variability (Mazor et al. 2016). Aside from their use in indicator evaluations, random forest models could also be used as a basis for mapping present or expected waterbody condition, or to hindcast reference conditions (Clapcott et al. 2017).

Unexplained variation in my random forest models may be associated with unmeasured environmental features, sampling error, temporal variance (mazor et al. 2016), metacommunity dynamics (Leibold et al. 2004, Heino 2012), or neutral processes in community assembly that are inherently unpredictable (Hubbell 2001). The relative contribution of these factors should continue to be an active area of bioassessment research (Mazor et al. 2016).

A wide variety of cumulative effects were observed in the Muskoka River Watershed, and cross-validation demonstrated the underlying models to have reasonable generality[1]. These effects constitute environmental issues, and suggest hypothetical mechanisms that are worthy of consideration in CEA. For example, Chironomidae become more numerically dominant in lake samples as lake pH decreases, which reinforces results from earlier studies demonstrating the Chironomidae to be tolerant of acidification (Wiederholm and Eriksson 1977, Schindler et al. 1985). In rivers, aquatic earthworms (the oligochaetous Clitellata) become more abundant as the proportion of the riparian zone under urban land-cover increases (potentially an effect of storm water and associated sedimentation), and as stream size increases (which is consistent with predictions from the River Continuum Concept; Vannote et al. 1980). Conductivity, alkalinity, chloride, and calcium concentrations in lakes and rivers increase with the intensity of human land-use in their catchments, which isn't surprising given the typical increase in solutes that accompanies many types of developments (Winter and Duthie 1998, Paul and Meyer 2001, Foley et al. 2005). Likewise, models of lake total phosphorus concentrations were best informed by drainage basin slopes, and agricultural intensity in the riparian zone, which reinforces wetlands and farms as areas of elevated nutrient fluxes (e.g., Sims et al. 1998; Reddy et al.1999).

[1] probably owing to the random forest's "bagging" procedure, in which numerous trees are assembled by bootstrapping (Breimer 2001)

Although many of the singular effects I identified were linear (e.g. the effect of pH on the relative abundance of lake Chironomidae), several were non-linear (e.g., effect of catchment forest cover on total nitrogen in rivers; conductivity and lake perimeter effects on the relative abundance of lake Chironomidae; effects of lake depth on dissolved organic carbon). Therefore, the ability to handle non-linear responses should be considered a compulsory feature of cumulative effects models.

The concept of the reference condition is ambiguous, and has been difficult to "translate from theory to practice" (Pardo et al 2012), given that the allowable level of anthropogenic impact is not clearly articulated (Stoddard et al. 2006, Pardo et al. 2012). Onset-of-effect thresholds represent empirical break-points where effects from stressor exposures become detectable (Utz et al. 2009). As long as reasonable criteria for identifying thresholds can be derived[2], the reference condition approach can be made more objective by using such thresholds to differentiate reference and impacted sites. In Chapter 3 (Onset-of-effect Thresholds and Reference Conditions: A Case Study of the Muskoka River Watershed, Canada), I reported no biological onset-of-effect thresholds; however, lake alkalinities and electrical conductivities, calcium, chloride, dissolved organic carbon, total Kjeldahl nitrogen, and total phosphorus concentrations, and pH exhibited threshold responses to road density, urbanization, or agriculture, as did stream alkalinities, conductivities and calcium and chloride concentrations. For lakes, thresholds were slightly more commonly detected for stressor variables measured at the cumulative-catchment scale than at the local-catchment scale; but for streams, thresholds were detected with approximately equal frequency for stressor variables

[2] Chapter 3 proposes complementary use of random forest and TITAN assessments; however no objective criteria for threshold identification exist.

measured at the two different scales. Several authors (e.g., Booth and Jackson 1997, Brabec et al. 2002) have reported threshold responses at very low levels of stressor exposure, and my study provides further evidence of this phenomenon. I demonstrated onset-of-effect thresholds as low as 1% agriculture (chloride in streams), 0.0002 m/m^2 road density (e.g., alkalinity and chloride in lakes) and 0.2% urbanization (alkalinity in lakes).

When sampled locations were grouped as reference or impacted based either on these thresholds or the default zero-stress criterion, insights about cumulative effects from the random forests were reinforced. Impacted waterbodies had more variable taxonomic structure and water chemistry, and had higher alkalinities and conductivities, calcium, chloride, and phosphorus concentrations, and pH than was typical of reference waterbodies. Furthermore, Amphipoda, Asellidae, Corixidae, Gastropoda, and Hirudinea were more abundant, and Chironomidae were less abundant in impacted streams than in reference streams, and insects were less abundant in impacted lakes than in reference lakes.

Differences in means and within-group variation between the groups of reference and impacted lakes and streams can be attributed to a combination of cumulative effects of human activities and natural factors. It is difficult to distinguish these sources of variation because land-uses are not dispersed evenly or randomly across watersheds, and because the intensity and types of developments are typically correlated with environmental features, which results in these features being unequally distributed among reference and impacted groups[3]. Nonetheless, the degree of interspersion between reference and impacted waterbodies (i.e., how similar their geographic positions and natural environmental attributes are) is a critical consideration for the design of cumulative effects monitoring programs. I found the geographic locations and ranges of natural environmental features associated with reference and impacted streams do substantially overlap in many cases. The two groups of streams, for example, had similar ranges of geographic coordinates and elevations; however, reference streams had shallower soils on average, and smaller catchment areas. Lesser interspersion was apparent for the two groups of lakes, reference lakes being, on average, smaller and shallower, with fewer up-gradient lakes in their drainage networks, and shallower soils in their catchments. Imperfect interspersion of reference and impacted sites is a common problem in the reference-condition approach (Stoddard et al. 2006, Pardo et al. 2012), and continues to be an issue in the Muskoka River Watershed.

I tabulated normal ranges associated with a variety of chemical and biological indicators and environmental attributes, because these ranges are useful as assessment benchmarks (Kilgour et al. 2017). Multiple indicators were included because numerous indicators are required to represent the complexity of lake and stream ecosystems (Dale and Beyeler 2001), because different indicators respond differently to different stressors (Kilgour et al. 2004), because multiple indicators may be relevant to the Muskoka Region's water management goals and policies, and because

[3] For example, large lakes are preferred for cottage development, relative to small lakes, and areas with gradual land-surface slopes and deep overburden are preferred for agriculture, relative to areas with steep slopes and shallow soils

the degree of interspersion among reference and impacted sites is different for different indicators. My reported normal ranges should be considered a first approximation. Additional reference lakes and streams should be sampled, because supplementary reference data would allow tolerance regions to be estimated more precisely. A larger pool of reference sites would also facilitate sophisticated model-based approaches for matching test-sites with the most appropriate set of reference sites (e.g., Norris and Hawkins 2000).

Threshold assessments have several other practical applications besides their use in deriving assessment benchmarks. For example, the identification of onset-of-effect thresholds can be important for early detection and mitigation, and for setting planning and conservation targets and limits (Utz et al. 2009, Mitchell et al. 2014). Furthermore, hypotheses about the mechanisms behind stressor effects can be generated by combining patterns of congruence among taxa-specific thresholds with knowledge describing the autecology and traits of component taxa (Wagenhoff et al. 2017). Further progress on cumulative effects monitoring could be achieved by undertaking such analyses.

Chapter 4 (Declining Amphipod Abundance is Linked to Low and Declining Calcium Concentrations in Lakes of the South Precambrian Shield) corroborates evidence of widespread lake-water calcium declines (e.g., Jeziorski et al. 2008), suggests that calcium levels have reached critically low levels in some lakes (i.e., relative to calcium requirements reported by Cairns and Yan 2009), and forecasts an average 57% decrease in amphipod abundances by the time calcium concentrations reach their predicted minima (e.g., Watmough and Aherne 2008), which is expected to occur in most lakes between years 2023 and 2100.

I demonstrated that lake-water calcium levels declined between 1993 and 2016 in 14 of 18 lakes, and a simultaneous reduction in amphipod abundances occurred in 8 of 18 lakes. Calcium explained 13% of the variation in amphipod abundances in this time series, making it the most important chemical, morphometric or physical-habitat predictor of declining amphipod abundances. Furthermore, models of amphipod presence/absence generated from a 107-lake spatial survey demonstrated that amphipods in the families Gammaridae and Hyalellidae are more commonly present in lakes with high calcium concentrations than in lakes with low calcium concentrations. Nonetheless, all three common amphipod families, Crangonyctidae, Gammaridae, and Hyalellidae, can be found in some lakes with Ca concentrations below 1 mg/l — the lower lethal limit inferred for Gammarus, based on laboratory bioassays (Cairns and Yan 2009) — and none exhibited a threshold response to this critical nutrient.

The mechanics and mitigating factors of calcium limitation in amphipods could be illuminated by more focussed surveys or experiments aimed at evaluating a series of additional working hypotheses. First, inputs from streams or groundwater (e.g., Lottig et al. 2011) could establish calcium-rich littoral refuges that allow amphipods to persist in lakes with low pelagic calcium levels. Given that amphipods are also present in riverine habitats (Peckarsky 1990) and are common in stream drift (Elliott and Corlett 1972), it is also possible that colonization pressure from lake-inflow streams (so-called “mass effects”, Heino 2012) could substantially mitigate population effects of Ca decline in some lakes. Second, many environmental changes are occurring in the southern Boreal

Shield (Palmer et al. 2011, Yan et al. 2011, Yao et al. 2013, Palmer et al. 2014), and stressor interactions are possible (Holmstrup et al. 2010, Altshuler et al. 2011, Fischer et al. 2013, Rollin et al. 2017). Amphipod responses to low Ca could be strongly mediated by temperature, food availability, food quality, or other environmental variables. Third, lakes' historical contexts could also be important. Short-term increases in lake-water calcium occurred due to logging, development, acid rain (Watmough and Aherne 2008), and local use of calcium-rich dust suppressants on some roadways (Shapiera et al. 2012), and these increases have been followed by gradual declines; therefore, it is possible that eco-evolutionary dynamics (e.g., Lallensack 2018) could be important, such that populations that have experienced relatively large fluctuations are more tolerant of Ca decline than populations that have experienced modest changes. Fourth, a question remains about whether low-Ca toxicity can be proven experimentally in any of the region's lakes. For example, Pearceley Lake — the lowest-Ca lake in my temporal dataset, where amphipods have not been collected in annual monitoring efforts since 2004 — would be a good candidate for Ca-addition or mesocosm experiments. Fifth, body-size differences suggest that Crangonyx, Gammarus, and Hyalella could have different calcium requirements, and useful insights could be gained from the relative abundances of species in these genera, which calls for more detailed taxonomic diagnoses in routine monitoring programs.

Notwithstanding factors that mitigate low-calcium stress, should lake-water calcium levels continue to decline as expected (i.e., Watmough and Aherne 2008), we forecast that the relative abundances of amphipods in Precambrian Shield lakes will decline, on average, by 57%, with many of these declines occurring before the end of the 2030s. Historically amphipods have been a numerically dominant taxon in the benthic community (during the 1993-2016 period they accounted for 28% of the counts of littoral fauna, on average). A marked reduction in the abundances of these animals signals a major restructuring of benthic communities, and reinforces warnings about declining Ca levels being a threat to freshwater biota (e.g. Jeziorski et al. 2008; Jeziorski and Smol 2016). As animals with high Ca demands (such as crustaceans and mollusks) decline in abundance, the structural and functional effects of these changes could propagate through food webs to substantially affect softwater boreal ecosystems (Jeziorski and Smol 2016). Water managers should be mindful of this threat, particularly given the range of other changes presently being observed in the watershed. Strategies for mitigating calcium loss, or supplementing calcium in catchments (e.g., the wood-ash proposal by Azan and Yan 2017) warrant consideration.

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