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Altered transcription levels of endocrine associated genes in two fisheries species collected from the Great Barrier Reef catchment and lagoon

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ABSTRACT

The Great Barrier Reef (GBR) is chronically exposed to agricultural run-off containing pesticides, many of which are known endocrine disrupting chemicals (EDCs). Here, we measure mRNA transcript abundance of two EDC biomarkers in wild populations of barramundi (*Lates calcarifer*) and coral trout (*Plectropomus leopardus* and *Plectropomus maculatus*). Transcription levels of liver vitellogenin (*vtg*) differed significantly in both species amongst sites with different exposures to agricultural run-off; brain aromatase (*cyp19a1b*) revealed some differences for barramundi only. Exposure to run-off from sugarcane that contains pesticides is a likely pathway given (i) significant associations between barramundi *vtg* transcription levels, catchment sugarcane land use, and river pesticide concentrations, and (ii) consistency between patterns of coral trout *vtg* transcription levels and pesticide distribution in the GBR lagoon. Given the potential consequences of such exposure for reproductive fitness and population dynamics, these results are cause for concern for the sustainability of fisheries resources downstream from agricultural land uses.

1. Introduction

Exposure to endocrine-disrupting chemicals (EDCs) can alter endocrine processes and disrupt reproductive development in wildlife and humans (United Nations Environment Programme and the World Health Organization, 2013). The physiological responses of fish to EDCs have been well studied, mostly in freshwater environments (Jobling and Tyler, 2003) and to a lesser extent in coastal and marine systems (Matthiessen et al., 2002; Porte et al., 2006). Sources of EDCs that affect endocrine function in fish include sewage treatment plants (STPs) (Lavado et al., 2004), industrial waste (Colborn et al., 1993), pulp and paper mills (Milestone et al., 2012), livestock waste (Kolodziej et al., 2004), and agricultural land uses (Sellin et al., 2009). Recent studies have demonstrated that reproductive success is compromised in wild endocrine-disrupted fish (Harris et al., 2011; Lange et al., 2011), and can ultimately impact on the sustainability of wild fish populations (Kidd et al., 2007).

The Great Barrier Reef (GBR) World Heritage Area is exposed to fluxes of pesticides discharged from the GBR catchment (Kroon et al., 2012; Smith et al., 2012) as a result of large-scale agricultural development mainly in the central and southern zones (Brodie et al., 2012). Land uses in the adjacent 423,000 km^2

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catchment comprise rangeland cattle grazing (75%), conservation areas (13%), forestry (5%) and sugarcane (1.3%), with a small urban (0.5%) and mining (0.3%) footprint (Brodie et al., 2012). Transport of agricultural pesticides from Queensland's rivers to the GBR lagoon occurs predominantly in riverine flood plume waters during the summer wet season, though pesticide contamination of GBR waters is chronic, widespread and year-round (Kennedy et al., 2012; Smith et al., 2012). Many of the agricultural pesticides detected in GBR rivers are well-known EDCs and include herbicides (e.g. 2,4-D, atrazine and its metabolite desethyl atrazine, pendimethalin, simazine), organophosphates (e.g. chlorpyrifos), organochlorines (e.g. dieldrin, endosulfan, lindane), and insecticides (e.g. fipronil, permethrin) (Smith et al., 2012; United Nations Environment Programme and the World Health Organization, 2013). Given that low but chronically sub-lethal concentrations of agricultural pesticides persist in the GBR waters throughout the year, the potential for exposure of GBR fish populations to EDCs, and associated impacts on reproductive fitness and population dynamics, is likely to be high.

In contrast to gonochoristic development in well-established model fish species for testing and assessing EDCs (Knacker et al., 2010), functional hermaphroditism is common amongst tropical marine fish (Sadovy de Mitcheson and Liu, 2008). However, the effects of EDCs on patterns of adult sex change in fish, and more specifically on development pathways between testicular and ovarian differentiation post-zygotically, has not been examined. In fish, sex change in each direction is mediated by the aromatase pathway (Kroon et al., 2005), with the aromatase enzyme catalysing the irreversible conversion of testosterone (T) into estradiol (17β-E). Two isoforms of the aromatase gene have been documented in fish, namely the gonadal (*cyp19a1a*) and brain (*cyp19a1b*) isoform (Tchoudakova and Callard, 1998). Recent studies have reported that exposure to estrogenic compounds can alter aromatase *cyp19a1* activity in both brain and ovaries in fish (Chung et al., 2011; Guyon et al., 2012; Kroon et al., In review; Mills et al., 2014). Hence, in the GBR region exposure to estrogenic compounds in agricultural run-off may modulate aromatase *cyp19a1* expression and function, compromising natural patterns of sex change in tropical marine fish species.

In this study, we examine whether exposure to agricultural run-off could cause estrogenic effects in wild populations of the protandrous barramundi (*Lates calcarifer*) and the protogynous coral trout (*Plectropomus leopardus* and *Plectropomus maculatus*). Specifically, we test the prediction that exposure to agricultural run-off results in an increase in transcription levels of brain aromatase (*cyp19a1b*) and liver vitellogenin (*vtg*) in juvenile barramundi and coral trout. Vitellogenin, the precursor of fish egg-yolk protein, is a well-established biomarker of exposure to estrogenic compounds

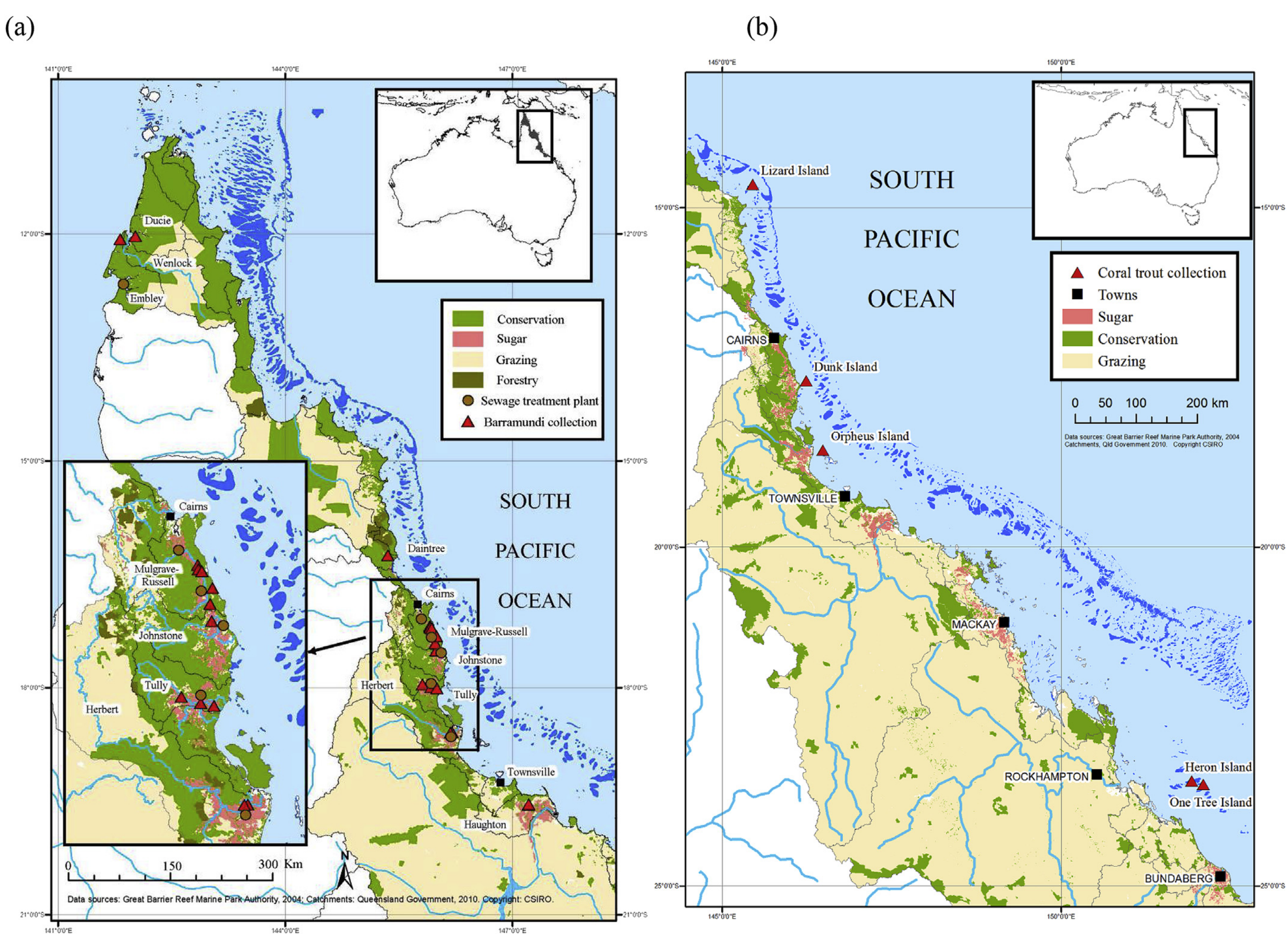

Fig. 1. Collection sites in the Great Barrier Reef region for (a) barramundi (*Lates calcarifer*) and (b) coral trout (*Plectropomus leopardus* and *P. maculatus*). Names of the nine river catchments, five reef islands and major towns are given. Agricultural land use in the catchments discharging into the collection sites ranged from 0% (Ducie and Embley rivers) to 98% (Haughton River) (see Supplementary material Table A.1 for more detail on main land uses).