

## Carbon sources supporting Australia's most widely distributed freshwater fish, *Nematalosa erebi* (Günther) (Clupeidae: Dorosomatinae)

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The authors of the above-mentioned paper regret to inform readers that, in the Online Early version of their paper, the fifth author was incorrectly included as 'Tze Wai Ho'. The correct name is Tsz Wai Ho and the ORCID ID is <https://orcid.org/0000-0001-5096-3954>.

# Carbon sources supporting Australia's most widely distributed freshwater fish, *Nematalosa erebi* (Günther) (Clupeidae: Dorosomatinae)

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**Abstract.** Both brown (detrital-based) and green (algal-based) food pathways support freshwater food webs, although the importance of either source may vary within species, regions and different phases of the flow regime. The bony bream (*Nematalosa erebi* Clupeidae: Dorosomatinae) is one of Australia's most widely distributed freshwater fish species and is a key component of freshwater food webs, especially in northern Australia. We sought to better define the feeding habits of this species, previously classified as a detritivore, algivore or zooplanktivore (or combinations thereof), by undertaking meta-analyses of published accounts based on stomach content analysis and <sup>13</sup>C and <sup>15</sup>N stable isotope analysis. Stomach content analysis clearly indicated that detritus was the dominant food item, although benthic algae could be an important dietary component in some habitats (inland river flood plains) and during the wet season. Zooplankton were important for small fish (i.e. juveniles <100 mm in length). When data were pooled across a large number of locations, stable isotope analysis indicated that detritus derived from terrestrial vegetation was better aligned isotopically with values for both adult and juvenile bony bream, whereas algae were comparatively <sup>13</sup>C enriched, indicating the latter source was not the dominant contributor to the biomass of this species. However, using site-specific data and a regression approach, a significant relationship was revealed between algal carbon and that of large fish, suggesting that carbon derived from benthic algae contributed ~20% of the carbon of adult bony bream. Zooplankton contributed a similar amount. Zooplankton provided the majority of carbon for small fish. We contend that detritus derived from terrestrial vegetation is the likely remaining carbon source for large bony bream, and this interpretation was supported by the outcomes of multiple regression analyses. Although previous studies of aquatic food webs in northern Australia have emphasised the importance of high-quality algal basal resources, this study indicates that terrestrial sources may be important for some species and demonstrates the need to better consider the circumstances that cause biota to switch between different food sources.

**Additional keywords:** algivory, aquatic food webs, detritivory, northern Australia, zooplanktivory.

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## Introduction

Most plant matter ends up as detritus and most community food webs contain both detrital and living primary producer energy channels (brown and green food chains respectively; Moore *et al.* 2004; Rooney *et al.* 2006). Early models of aquatic ecosystem function emphasised the importance of terrestrial or aquatic vascular plant material in supporting the biomass of aquatic consumers via a detrital breakdown pathway (Vannote *et al.* 1980; Junk *et al.* 1989). Qualification of this viewpoint has

included the inclusion of microbiota as both conditioners of detritus that make nutrients and energy more available and as constituents, which are themselves consumed (e.g. France 2011). By contrast, while not discounting the importance of terrestrial inputs, Thorp and Delong (1994) emphasised the importance of algal production in supporting consumer biomass. The use of stable isotopes and fatty acid markers in food web studies has largely confirmed the importance of autochthonous algal production in aquatic food webs (Lewis *et al.* 2001;

Bunn *et al.* 2003; Guo *et al.* 2016a, 2016b; Brett *et al.* 2017). Algal carbon is easier to digest and assimilate than that of vascular plant material (Brett *et al.* 2017). Moreover, algae contain higher quantities of polyunsaturated fatty acids (PUFA), which are essential for metazoan growth (Guo *et al.* 2016a).

Douglas *et al.* (2005) proposed that most biomass of tropical northern Australian rivers was ultimately derived from algal production. This hypothesis is largely supported by subsequent research, although other sources, such as terrestrially derived detritus, may also be important (Bunn *et al.* 2013; Pettit *et al.* 2017). Elsewhere, several experimental and field-based studies have revealed that some aquatic consumers are supported by carbon derived from detritus and attached microbes (e.g. McGoldrick *et al.* 2008; Reid *et al.* 2008; Brett *et al.* 2009; Solomon *et al.* 2011; Belicka *et al.* 2012). Further, fatty acid profiles of some primary consumer organisms indicate a detrital origin by microbial processors (Belicka *et al.* 2012), and some aquatic organisms may possess the capacity to convert some fatty acids into more physiologically active forms (Murray *et al.* 2014; Guo *et al.* 2016b). Brett *et al.* (2017) suggested that the extent to which terrestrial carbon supports upper trophic level production may depend on the simultaneous availability of essential biomolecules derived from algae and concluded that there is no doubt that terrestrially derived carbon is ingested and assimilated by herbivores, but that it is done so at much reduced efficiency. Clearly, an algal–detrital dichotomy oversimplifies the complex relationships present within aquatic food webs (Taylor and Batzer 2010; Jardine *et al.* 2015).

Detritivorous fishes are an important component of tropical aquatic food webs (Lowe-McConnell 1975; Goulding *et al.* 1988; Flecker 1996), transferring basal production to higher trophic levels and frequently forming the major prey of piscivorous fishes of socioeconomic importance (Winemiller 2004). Although detritivorous fishes are common in tropical regions globally, detritivory is less evident in temperate regions (Egan *et al.* 2018). Coates (1993) stated that truly detritivorous fishes are absent from freshwaters of the Australasian region, tropical or otherwise, but subsequent studies have revealed that detritus may comprise a significant fraction of the stomach contents of several benthic foraging species, particularly in tropical northern Australia (Pusey *et al.* 2000; Bishop *et al.* 2001; Kennard *et al.* 2001).

The bony bream (*Nematalosa erebi* (Günther)), a clupeid gizzard shad, is one of Australia's most widely distributed freshwater fish species. It has been previously characterised as a detritivore, algivore or planktivore (or combinations thereof; for a review of its biology, see Pusey *et al.* 2004). This species is primarily tropical or subtropical, although its distribution extends as far south as 35°S in the Murray–Darling River system (Fig. 1). The species occurs in a great variety of perennial and intermittent aquatic habitat types and may achieve very high levels of abundance. *N. erebi* is itself consumed by many higher-order consumers, including piscivorous fishes, crocodiles and birds such as cormorants and pelicans; thus, it is an important component in the food webs of Australian rivers, particularly those of northern Australia.

In this study, we sought to better define the feeding habits and role of *N. erebi* in aquatic food webs by reference to published dietary information and more recent stable isotope analysis of

food web structure. We specifically sought to determine whether *N. erebi* was reliant on allochthonous production (i.e. terrestrial vegetation, TVEG, via a detrital pathway) or autochthonous algal production. We hypothesised that algae were the main source of carbon for this species.

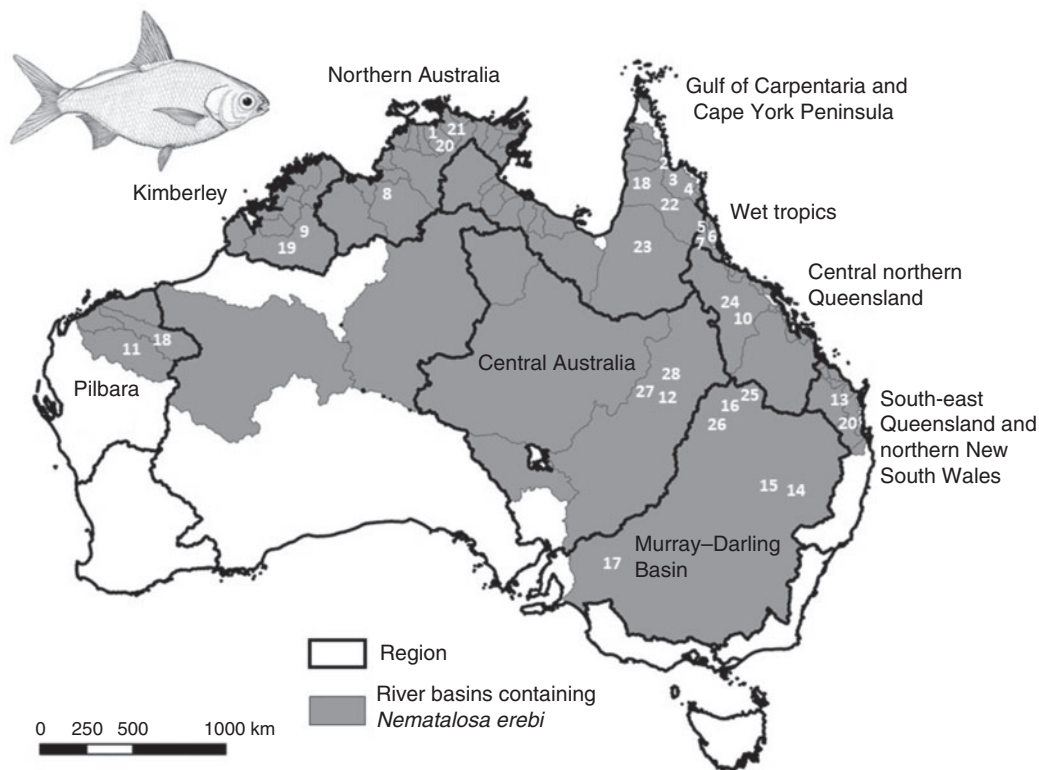
## Materials and methods

### Sources of dietary information: stomach content analysis

Dietary information for *N. erebi* was available from 17 individual studies, drawn from the published literature, 1 university thesis, 2 consultancy reports to state government and 1 unpublished dataset (mean  $\pm$  s.e.m. sample size  $191.5 \pm 61.6$ ; see Table 1; Fig. 1). The data used in this study were drawn from a larger dataset being used to examine the trophic ecology of Australian freshwater fishes in which diet was apportioned to proportional contributions within 15 categories (for details concerning diet summarisation and data treatment, see Kennard *et al.* 2001). Here we present information for only five categories, namely aquatic insects, aquatic macrophytes, detritus, algae (including filamentous algae, diatoms and biofilm) and zooplankton, because these collectively accounted for a mean  $\pm$  s.e.m.  $96.5 \pm 1.8\%$  of the diet across studies (Table 1). We also included some measure of the size of the fish examined in each study, either as the mean  $\pm$  s.e.m. size or size range. The lower limit of the size range of individuals included was  $>100$ -mm standard length (SL) for eight studies (i.e. adults; cf. Table 1), whereas four studies were largely dominated by individuals  $<100$  mm SL (i.e. juveniles; studies 9, 14, 15 and 17 in Table 1). These latter studies were undertaken within the Murray–Darling Basin, with the exception of one undertaken in the Burdekin River of Central North Queensland (Fig. 1). Most studies attempted to distinguish between detritus *per se* and microalgae mixed in with small detrital particles. Only 4 of the 17 studies reviewed (studies 1, 6, 8 and 11 in Table 1) included data from both wet season and dry season sampling periods. In the remaining 13 studies, dry season samples were collected as part of a one-off sampling event.

### Sources of stable isotope information

Information was sourced on carbon and nitrogen stable isotope (SI) values of bony bream tissue (fin or muscle) and three potential food sources (benthic algae (primarily periphyton), TVEG and zooplankton) from 11 separate food web studies undertaken in northern, eastern and central Australia and the northern portion of the Murray–Darling Basin (Fig. 1) in which the authors have been individually or collectively involved and that included *N. erebi* (Beesley 2006; Blanchette *et al.* 2014; Bunn *et al.* 2003; Jardine *et al.* 2012a, 2012b, 2013, 2015, 2017; L. S. Beesley, B. J. Pusey, M. M. Douglas, C. A. Canham, C. S. Keogh, O. P. Pratt, M. J. Kennard, and S. A. Setterfield, unpubl. data; S. E. Bunn's three unpublished data sets). These studies were intended to examine nutrient and energy transfer between a variety of basal sources, many organisms (including many species of fish) and trophic levels. Only three of these studies were undertaken in the dry season only (Blanchette *et al.* 2014; Jardine *et al.* 2012a; Bunn, unpubl. data). We excluded any data that did not allow us to distinguish between fish of different size classes (i.e.  $<100$  and  $>100$  mm SL). The manner



**Fig. 1.** Distribution of *Nematalosa erebi* within freshwater regions of Australia. Unshaded areas do not contain *N. erebi*. Region delineation is based on general similarities in climate and catchment physiography, as well as biogeographic variation in fish species distributions (Unmack 2013). The approximate location of studies used here is given and denoted by numbers as follows: 1, Bishop *et al.* (2001); 2, Pusey *et al.* (2000); 3, Kennard (1995); 4, Hurtle and Person (1990); 5, Pusey *et al.* (1995); 6, Rayner *et al.* (2009); 7, Morgan *et al.* (2004); 8, Thorburn *et al.* (2014); 9, Pusey *et al.* (2010); 10, P. M. Davies, unpubl. fish diet data from Robe River, Pilbara, Western Australia; 11, Balcombe *et al.* (2005); 12, Bluhdorn and Arthington (1994); 13, Arthington *et al.* (1992); 14, Medeiros and Arthington (2014); 15, Medeiros and Arthington (2008); 16, Sternberg *et al.* (2008); 17, Atkins (1984); 18, Beesley (2006); 19, L. S. Beesley, B. J. Pusey, M. M. Douglas, C. A. Canham, C. S. Keogh, O. P. Pratt, M. J. Kennard, and S. A. Setterfield (unpubl. data); 20, S. E. Bunn (unpubl. data); 21, Jardine *et al.* (2017); 22, Jardine *et al.* (2012b); 23, Jardine *et al.* (2013); 24, Blanchette *et al.* (2014); 25, Jardine *et al.* (2015); 26, S. E. Bunn (unpubl. data, collected as part of the DryLand Refugia Project; see [https://ewater.org.au/archive/crcfe/ewater/domino/html/Site-CRCFE/CRCFE\\_WebSite.nsf/pages/Program+C+Progress+2004.html](https://ewater.org.au/archive/crcfe/ewater/domino/html/Site-CRCFE/CRCFE_WebSite.nsf/pages/Program+C+Progress+2004.html), accessed 18 May 2020); 27, Bunn *et al.* (2003); 28, S. E. Bunn (unpubl. data, collected in the Cooper Creek but not included in Bunn *et al.* 2003).

in which samples were collected and analysed was largely consistent across studies (for detailed methods, see Jardine *et al.* 2012a) with the exception of particulate organic matter for which different particle sizes (i.e. coarse particulate organic matter, CPOM, and fine particulate organic matter, FPOM) were not consistently differentiated or collected. By contrast, all studies collected dead leaves of riparian trees (i.e. TVEG; primarily *Melaleuca* and *Eucalyptus* spp.), and these species contribute most allochthonous carbon inputs to freshwater systems in the study area. For those samples in which SI information was available for TVEG, CPOM and FPOM,  $\delta^{13}\text{C}$  values of CPOM and FPOM differed from TVEG by less than +1 and +2–3‰ respectively, and differences in  $\delta^{15}\text{N}$  were less than +1‰. These differences accord well with similar comparisons elsewhere (e.g. Finlay and Kendall 2007). In total, SI information was available for fish collected from 120 separate locations (i.e. sites).  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values for putative source material for

each site were estimates based on the mean of at least three samples. Similarly, information from at least 3, but often up to 20, individuals for each size class of *N. erebi* was used to estimate mean  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values of fish at each site.

We generated histograms of the frequency distributions for  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  for the three food sources and both size classes of *N. erebi* across all sites to assess the extent of spatial variation in isotope values and the extent of overlap in isotope values for different potential food sources. Broad distributions (i.e. high variance) indicate high spatial variation. We also assessed whether  $\delta^{13}\text{C}$  or  $\delta^{15}\text{N}$  of individual source materials varied independently using Pearson's correlation. Gradient-based approaches where isotope variation of producers and consumers is measured at multiple locations have proved useful for determining the importance of different food sources exhibiting large spatial variation in isotope values (Rasmussen 2010; Jardine *et al.* 2012a). This approach, in contrast to a mixing model

Table 1. Diet of *Nematolosa erebi* as described in individual studies

The studies included in this analysis are as follows: 1, Bishop *et al.* (2001); 2, Pusey *et al.* (2000); 3, Kennard (1995); 4, Horte and Person (1990); 5, Pusey *et al.* (1995); 6, Rayner *et al.* (2009); 7, Morgan *et al.* (2004); 8, Thorburn *et al.* (2014); 9, Pusey *et al.* (2010); 10, P. M. Davies, unpubl. data, 2000; 11, Balcombe *et al.* (2005); 12, Bluhdorn and Arthington (1994); 13, Arthington *et al.* (1992); 14, Medeiros and Arthington (2014); 15, Medeiros and Arthington (2008); 16, Sternberg *et al.* (2008); 17, Atkins (1984). The regions are shown in Fig. 1. Where appropriate, size data are presented as the mean  $\pm$  s.e.m. ALG, algae (including filamentous algae, diatoms and biofilm); AQINS, aquatic insects; CENT AUS, central Australia; CNQ, central northern Queensland; DET, detritus; FP, flood plain; GOC-CYP, Gulf of Carpentaria and Cape York Peninsula; KIMB, the Kimberley; L, lacustrine; MAC, aquatic macrophytes; MDB, Murray-Darling Basin; NTH AUS, northern Australia; PILB, the Pilbara; R, riverine habitat; SEQ-NEN, south-east Queensland and northern New South Wales; SL, standard length; TL, total length; CFL, length at caudal fork; WT, wet tropics; ZOOP, zooplankton

Study	Region (habitat)	Season	Size	n	AQINS	MAC	DET	ALG	ZOOP	Total	Comments
1	NTH AUS (R, FP),	All seasonal flow phases	Seasonal means ranged from 76 to 216 mm; overall mean 120 mm CFL for entire sample	471	2.5	0.0	42.9	41.8	12.6	99.8	Distinguished between detritus, desmids or diatoms and other algae
2	GOC-CYP (R),	Dry season	120–270 mm SL	20	5.0	0.0	95.0	0.0	0.0	100.0	Distinguished between detritus, desmids or diatoms and algae
3	GOC-CYP (FP)	Dry season	200 $\pm$ 7 and 178 $\pm$ 7 mm SL for early and late dry season respectively	98	0.1	0.0	99.1	0.6	0.1	99.9	Distinguished between detritus and algae
4	GOC-CYP (R)	Dry season	187–262 TL	7	0.0	0.0	50.0	50.0	0.0	100.0	Distinguished between detritus and algae
5	WT (R)	Dry season	200–300 SL	7	0.0	0.0	100.0	0.0	0.0	100.0	Distinguished between detritus, desmids or diatoms and algae
6	WT (R)	Wet and dry season	209 $\pm$ 18 mm SL	66	0.1	0.2	97.2	0.8	0.2	98.4	Distinguished between detritus and algae
7	KIMB (L)	Dry season	47–330 mm TL	21	7.3	0.0	59.4	4.1	2.0	72.8	Distinguished between biofilm or silt and filamentous algae. Tables list the contribution by biofilm or silt, but summary figures and text refer to this component as detritus. Other plant material contributed to 23.4% of the diet
8	KIMB (R)	All flow phases	25–420 mm TL	132	0.3	0.0	58.6	15.7	12.9	87.2	Distinguished between biofilm or silt and filamentous algae. Sand contributed to 26.4% of the diet in the late dry season and to 9.7% of the overall total diet (not included here). Tables list the contribution by biofilm or silt, but summary figures and text refer to this component as detritus
9	CNQ (R)	Wet season	<100 mm	11			97.7	1.6			
10	PILB (R)	Wet season	>100 mm	22			86.1	9.2			
11	CENT AUS (WH, FP)	Early dry	<100 mm	19			68.0	24.0	87.3		
		Late dry	>100 mm	36			48.7	21.4			
12	SEQ-NEN (R)	Dry season	70 $\pm$ 2 mm SL	508	10.3	0.0	86.0	1.3	0.0	97.6	Distinguished between detritus and algae
13	SEQ-NEN (R)	Dry season	N/A	9	6.0	0.0	0.0	92.0	0.0	98.0	
14	MDB (WH)	Wet and dry season	29–260 mm (most <100 mm TL)	98	3.0	3.2	16.8	52.2	23.9	99.2	
15	MDB (WH)	Dry season	>100 mm	88	0.0	0.0	76.3	23.0	0.0	99.2	
16	MDB (WH)	Dry season	72% of sample <69 mm SL	411	8.0	6.7	40.1	13.4	20.1	88.3	
			Mean $\pm$ s.d. from 67 $\pm$ 7 to 100 $\pm$ 16 mm	948	0.0	0.9	93.8	0.4	4.9	100.0	Distinguished between detritus, filamentous alga, Volvox and 'algal matter'
17	MDB (R)	Dry season	34–396 mm TL (most <80 mm TL)	212	0.5	8.1	69.0	12.3	10.1	100.0	Distinguished between filamentous and non-filamentous alga (but here pooled); the detrital component was greatest during periods of low flow
Mean $\pm$ s.e.m.				61	0.0	0.0	54.1	45.9	0.0	100.0	
				98	2.5 $\pm$ 0.9	1.1 $\pm$ 0.6	63.0 $\pm$ 7.6	20.8 $\pm$ 6.6	9.0 $\pm$ 4.2	96.5 $\pm$ 1.8	

approach, does not require potential sources to be distinct at all sites, does not require *a priori* knowledge of the extent of isotope trophic discrimination and does not require spatial variation in isotope values of different consumers to be independent (Moore and Semmens 2008; Rasmussen 2010). From a practical viewpoint, a gradient-based approach can maximise the number of locations used in analyses because it does not require all three potential sources of carbon to have been measured at every site.

Such an approach is well suited to the present case, where data were collected from multiple locations within many rivers. We plotted  $\delta^{13}\text{C}$  values of each size class of *N. erebi* against  $\delta^{13}\text{C}$  values of algae, TVEG and zooplankton. We used simple linear regression to assess the strength of the relationship between consumer (i.e. *N. erebi*)  $\delta^{13}\text{C}$  values and food source (i.e. algae, TVEG and zooplankton)  $\delta^{13}\text{C}$  values and report statistical significance at the  $\alpha = 0.05$  level. We estimated whether the slopes of the relationship between isotope values were significantly different from 1 or 0 (i.e. not within the 95% confidence limits of the estimated slope). A close dependency on one source or the other should see *N. erebi* muscle  $\delta^{13}\text{C}$  values aligned with spatial variation in  $\delta^{13}\text{C}$  for that potential source (i.e. values should fall along a line denoting a 1:1 relationship or slope = 1). Conversely, if no significant relationship (i.e. slope = 0) is detected between consumer and food sources isotope values, then it is assumed that source is unlikely to be important. Slopes significantly different from both 0 or 1 indicate a mixed feeding model (i.e. more than one source contributes to the carbon or nitrogen assimilated into body mass; Jardine *et al.* 2012a). An important assumption in this approach is that isotope ratios in the consumer organism are in equilibrium with the measured source materials (i.e. isotope values do not reflect feeding at some other time or location). To a large degree, this assumption underpins most food web studies undertaken in natural environments. We similarly examined and tested the relationship between  $\delta^{15}\text{N}$  values of potential food sources and those of *N. erebi* tissue. We also performed multiple regression analyses for each size class and both isotopes for those locations for which information concerning all three potential sources was available ( $\delta^{13}\text{C}$ :  $n = 29$  and  $35$  for small and large fish respectively;  $\delta^{15}\text{N}$ :  $n = 28$  and  $29$  for small and large fish respectively). Finally, we estimated  $\delta^{15}\text{N}$  trophic enrichment factors (i.e.  $\Delta\delta^{15}\text{N} = \delta^{15}\text{N}_{\text{consumer}} - \delta^{15}\text{N}_{\text{source}}$ ) for each source and age class combination and, from these data, estimated overall trophic enrichment taking into account the estimated proportional contribution of each source derived from the slopes of the lines relating  $\delta^{13}\text{C}$  variation of source and consumers.

Where appropriate, data are given as the mean  $\pm$  s.e.m.

## Results

### *Stomach contents analysis*

Across all studies, the mean contribution of detritus to the diet was  $63 \pm 8\%$  and that of algae was  $20.8 \pm 9.0\%$  (Table 1). Zooplankton contributed a further  $9 \pm 4\%$ , whereas aquatic insect larvae and aquatic macrophytes formed only a minor fraction of the diet (2.5 and 1.1% respectively). Detritus was the dominant dietary component in most studies except two undertaken in arid zone rivers, where algae contributed 90% and 52% to the diet (Studies 10 and 11 respectively, Table 1) and

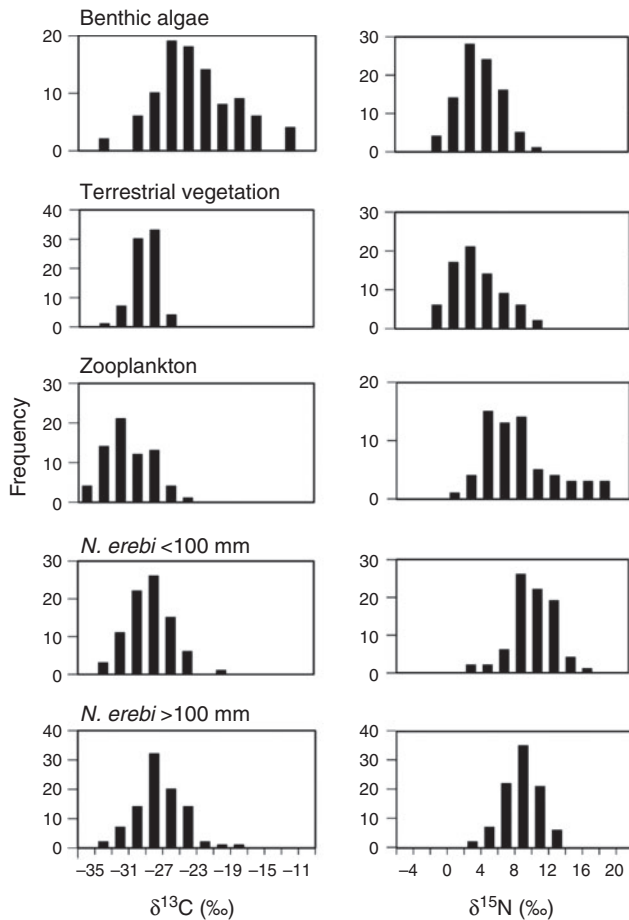
another undertaken in the Gulf Cape York Peninsula region (Study 4, Table 1) in which detritus and algae were codominant. Consumption of zooplankton was greatest in arid zone or southern regions (i.e. the Murray–Darling Basin); however, high consumption of zooplankton was also recorded in northern regions (i.e. the Kimberley and north Australia). All studies in which zooplankton contributed more than 1% of the diet (seven studies) were either dominated by or included fish <100 mm SL. For example, zooplankton comprised 87.3% of the diet in a seasonal subsample comprised entirely of small fish within one study undertaken in the Kimberley region (Study 8, Table 1). Similarly, a high contribution of zooplankton was recorded in the most southern study available (in the Murray–Darling Basin; Study 17, Table 1) and in which the sample was dominated by small individuals. Aquatic insect larvae (chironomid larvae) comprised  $\sim 10\%$  of the diet in another study (Study 9, Table 1) undertaken in a large shallow sand bed river. Individuals included in that study were also small (mean  $\pm$  s.e.m. SL  $70 \pm 2$  mm). Thus, consumption of zooplankton and, to a lesser extent, aquatic insect larvae was limited to individuals of small size. Consumption of detritus and algae was greatest in larger individuals (i.e. >100 mm SL).

### *Stable isotope analyses*

A wide range of  $\delta^{13}\text{C}$  values from  $-34.6$  to  $-12.0\%$  (mean  $-23.0 \pm 0.5\%$ ) was recorded for benthic algae (Fig. 2). TVEG was relatively depleted in  $^{13}\text{C}$  and varied little (mean  $\delta^{13}\text{C}$   $-29.1 \pm 0.2\%$ ; range  $-33.3$  to  $-26.5\%$ ). Zooplankton  $\delta^{13}\text{C}$  values ranged from  $-38.2$  to  $-23.5\%$  and were typically highly depleted in  $^{13}\text{C}$  (mean  $\delta^{13}\text{C}$   $-31.1 \pm 0.4\%$ ). Large *N. erebi* exhibited an intermediate range of  $\delta^{13}\text{C}$  values (ranging from  $-33.3$  to  $-18.5\%$ ; mean  $^{13}\text{C}$   $-27.4 \pm 0.3\%$ ) and were more depleted in  $^{13}\text{C}$  compared with algae (as were small *N. erebi*: range  $-33.7$  to  $-20.9\%$ ; mean  $\delta^{13}\text{C}$   $-28.4 \pm 0.3\%$ ). The  $\delta^{13}\text{C}$  values of algae, TVEG and zooplankton varied independently of one another ( $r < 0.20$ ,  $P > 0.05$  for all comparisons).

Algae and TVEG had similar mean values and variability in  $\delta^{15}\text{N}$  values (mean  $\delta^{15}\text{N}$   $4.5 \pm 0.3$  and  $4.3 \pm 0.2\%$  respectively; Fig. 2). By contrast, zooplankton were comparatively enriched in  $^{15}\text{N}$  (mean  $\delta^{15}\text{N}$   $9.2 \pm 0.5\%$ ) and some samples were highly enriched (maximum 18.9%). Large *N. erebi* were similarly enriched in  $^{15}\text{N}$  (mean  $\delta^{15}\text{N}$   $9.0 \pm 0.2\%$ ) and small *N. erebi* were slightly more enriched in  $^{15}\text{N}$  than larger fish and zooplankton (mean  $\delta^{15}\text{N}$   $10.8 \pm 0.3\%$ ). The  $\delta^{15}\text{N}$  values of algae, TVEG and zooplankton did not vary independently of one another ( $r = 0.60$ ,  $0.51$  and  $0.93$  for algae and TVEG, algae and zooplankton, and TVEG and zooplankton respectively;  $P < 0.001$  for all).

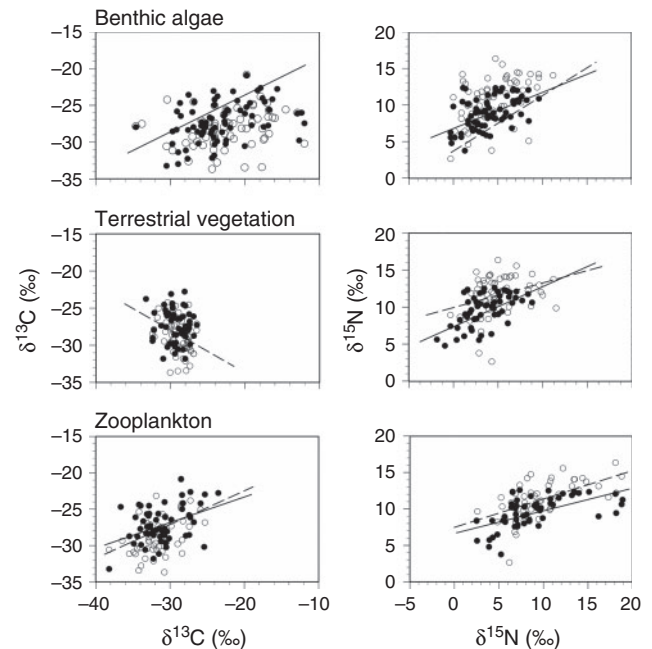
Fig. 3 plots isotope variation ( $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$ ) in large and small *N. erebi* against variation in isotope values of putative dietary components across a large number of sites (for sample sizes and regression statistics, see Table 2). Variation in  $\delta^{13}\text{C}$  of large *N. erebi* was significantly positively related to variation in  $\delta^{13}\text{C}$  of both algae and zooplankton, and the slopes for these relationships (0.22 and 0.33 respectively) were both significantly different from 0 and 1, suggesting a mixed feeding strategy with algae and zooplankton together accounting for approximately one-half of assimilated carbon. No significant relationship between  $\delta^{13}\text{C}$  of large *N. erebi* and terrestrial carbon was detected. Variation in  $\delta^{13}\text{C}$  of small *N. erebi* was



**Fig. 2.** Frequency distributions (%) of spatial variation in stable isotope values ( $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$ ) for potential source material (i.e. aquatic and terrestrial primary producers and zooplankton) and for the two size classes of the consumer *Nematalosa erebi*. Sample sizes are 94, 75, 68, 93 and 82 for algae, terrestrial vegetation, zooplankton, *N. erebi* >100 mm and *N. erebi* <100 mm respectively.

not significantly related to variation in benthic algal  $\delta^{13}\text{C}$  values, but was significantly positively correlated with zooplankton  $\delta^{13}\text{C}$ , with the slope of this relationship (0.52) being significantly different from 0 and 1 (Table 2), again suggesting a mixed feeding model with approximately one-half of assimilated carbon being derived from this source (Fig. 3; Table 2).

$\delta^{15}\text{N}$  variation in both large and small *N. erebi* was significantly positively correlated with variation in all putative food sources (i.e. algae, TVEG and zooplankton), perhaps not surprisingly given variation within sources was also significantly positively correlated. However,  $\delta^{15}\text{N}$  variation in large *N. erebi* was most strongly correlated with variation in  $\delta^{15}\text{N}$  of TVEG (Table 2). Trophic enrichment ( $\delta^{15}\text{N}_{\text{consumer}} - \delta^{15}\text{N}_{\text{diet}}$ ) in large *N. erebi* averaged  $4.9 \pm 0.3$ ,  $6.0 \pm 0.3$  and  $0.5 \pm 0.2$ ‰ for algae, TVEG and zooplankton respectively and  $6.2 \pm 0.4$ ,  $6.7 \pm 0.4$  and  $1.5 \pm 0.4$ ‰ respectively in small *N. erebi*. However, when scaled for the different proportional contributions by each putative food source, the estimated mean trophic enrichment for all sources was  $4.1 \pm 0.3$ ‰ and  $3.2 \pm 0.4$ ‰ for large and small *N. erebi* respectively. The results of multiple



**Fig. 3.** Carbon and nitrogen isotope values ( $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$ ) of muscle or fin tissue of *Nematalosa erebi* plotted against isotope values of potential food sources (algae, terrestrial vegetation and zooplankton). Each point represents the mean value from an individual site within each study. Closed symbols indicate large fish (>100-mm standard length, SL); open symbols indicate small fish (<100 mm SL). Unbroken lines represent significant regressions between consumer and producer values for large fish, whereas broken lines are for the smaller size class. Sample sizes and regression statistics are given in Table 2.

regression analyses based on data available from the reduced set of locations (Table 3) strongly supported the outcomes of simple linear regression analysis. That is,  $\delta^{13}\text{C}$  values of small fish (<100 mm SL) were most strongly related to those of zooplankton, whereas those of the larger size class were most strongly related to TVEG values and then benthic algae. The  $\delta^{15}\text{N}$  isotope information was less informative than that for  $\delta^{13}\text{C}$  (as was the case for simple linear regression) in that only zooplankton exhibited a significant relationship with  $\delta^{15}\text{N}$  values of small fish.

## Discussion

Stomach content analysis from multiple studies indicates that *N. erebi* is zooplanktivorous as a juvenile before transitioning to a primarily detrital diet with increasing size. These ontogenetic changes in diet mirror similar changes observed in a closely related clupeid, the American gizzard shad *Dorosoma cepedianum* (Smoot and Findlay 2010). When based on all samples, stable isotope information also suggested that detritus derived from TVEG, and zooplankton, provided a large fraction of the assimilated carbon. On average, the  $\delta^{13}\text{C}$  values of both small and large *N. erebi* ( $-28.5 \pm 0.3$  and  $-27.4 \pm 0.3$ ‰ respectively) were very closely aligned to that of TVEG, depleted in  $^{13}\text{C}$  with respect to algal  $\delta^{13}\text{C}$  values ( $-23.0 \pm 0.5$ ‰) and enriched compared with zooplankton ( $-31.1 \pm 0.4$ ‰). Collectively, these data do not support a significant contribution by benthic algae to carbon assimilation in large *N. erebi*. However,

**Table 2.** Regressions statistics (intercept, slope and *F* values) for comparisons of isotope values of sizes classes of *Nematalosa erebi* and putative food sources (see Fig. 3)Also given are the lower and upper 95% confidence limits (CLs) of the slope. n.s.,  $P > 0.05$ ; \*,  $P < 0.05$ ; \*\*\*,  $P < 0.001$ 

Source	Consumer size class (mm)	Isotope	<i>n</i>	Intercept	Slope	Lower 95% CL	Upper 95% CL	<i>F</i>
Benthic algae	<100	$\delta^{13}\text{C}$	64	-11.32	0.39	-0.07	0.84	2.95 <sup>NS</sup>
		$\delta^{15}\text{N}$	63	8.37	0.52	0.27	0.77	18.17***
	>100	$\delta^{13}\text{C}$	73	-22.16	0.22	0.10	0.35	12.71***
		$\delta^{15}\text{N}$	72	7.13	0.44	0.26	0.63	22.91***
Terrestrial vegetation	<100	$\delta^{13}\text{C}$	58	-44.85	-0.55	-0.96	-0.14	7.10*
		$\delta^{15}\text{N}$	58	10.15	0.29	0.03	0.56	4.98*
	>100	$\delta^{13}\text{C}$	55	-30.67	-0.10	-0.48	0.27	0.30 <sup>NS</sup>
		$\delta^{15}\text{N}$	55	7.74	0.53	0.32	0.74	26.80***
Zooplankton	<100	$\delta^{13}\text{C}$	54	-12.44	0.52	0.26	0.77	16.94***
		$\delta^{15}\text{N}$	52	7.54	0.38	0.25	0.52	32.45***
	>100	$\delta^{13}\text{C}$	56	-16.91	0.33	0.11	0.54	9.81***
		$\delta^{15}\text{N}$	54	6.75	0.31	0.19	0.43	28.39***

regression analyses using site-specific data revealed a significant positive relationship between algal carbon and that of large fish, suggesting that benthic algae may also form an important carbon source for this size class of *N. erebi*.

Important methodological considerations for both stomach content and stable isotope analyses must be considered before accepting the generality of these findings. First, most studies, particularly those undertaken in northern Australia, examined diet during the dry season only, whereas the consumption of algae was greatest in those few studies undertaken over a long period and that included either wet seasons or periods immediately following a wet season. Thus, the contribution of algae to the diet of *N. erebi* could conceivably be higher than reported here. Second, trituration of ingested material within the muscular gizzard renders most material to a fine paste and it is highly likely that, despite the best intentions of researchers, algal material, other than filamentous algae, may not be readily identifiable (i.e. distinguished from detritus) and reliably quantified when examined macroscopically. Third, in aggregating stable isotope information across studies and locations, any spatial variation in the relationship between algal isotope values and those of the consumer is likely obscured. As a consequence, conclusions regarding the importance of detritus derived from TVEG and a minimal contribution by benthic algae warrant further scrutiny.

Indeed, isotope values for the putative food sources of algae and zooplankton varied greatly and the range of values overlapped substantially for all sources. Such large variation in  $\delta^{13}\text{C}$  is not unexpected given the range of different potential pathways by which carbon is available for uptake (i.e. atmospheric v. dissolved) and the wide array of factors that affect carbon fractionation in aquatic systems (Finlay 2004; Barnes *et al.* 2007). Similarly, baseline values of  $\delta^{15}\text{N}$  vary extensively in space due to variation in the taxonomic composition of producers, isotope distinction between various sources (i.e.  $\text{N}_2$ ,  $\text{NO}_3$ ,  $\text{NH}_3$ ) and variation in the efficiency with which they are used (Akiyama *et al.* 1997). However, it is notable that  $\delta^{15}\text{N}$  values of TVEG (riparian species primarily within Myrtaceae) and benthic algae in the present study were highly correlated despite

their taxonomic distinctiveness. This finding suggests that both derived their nitrogen from the same source (i.e. that dissolved within the stream or groundwater).

Rather than being an impediment to interpreting relationships between sources and consumers, the presence of spatial variation in algal and zooplankton  $\delta^{13}\text{C}$  values helps identify the source of carbon sustaining *N. erebi*. Our data suggest a mixed feeding strategy in both large and small individuals. A significant relationship between  $\delta^{13}\text{C}$  values of algae and large fish with a slope (0.22) significantly different from both 0 and 1 indicates that algae contribute approximately one-quarter (range, as defined by confidence intervals, 10–35%) of the carbon assimilated by this size class. Carbon derived from benthic algal production is important in freshwater ecosystems globally (Roach 2013), including in Australia and especially in northern regions and arid zones (Bunn *et al.* 2003, 2006; Douglas *et al.* 2005; Leigh *et al.* 2010; Jardine *et al.* 2012a, 2012b). However the present study suggests that benthic algal carbon contributed little to the biomass of small *N. erebi*, which were, in contrast, more reliant on zooplankton carbon (52%; range 26–77%). Zooplankton also contributed substantially to the biomass of large *N. erebi* (33%; range 11–54%). Stomach content analysis for small *N. erebi* also identified zooplankton as an important dietary component. Medeiros and Arthington (2011) reported a significant correlation between spatial variation in  $\delta^{13}\text{C}$  values for *N. erebi* (and other fish species) and zooplankton that is consistent with the findings of stomach content analysis (Medeiros and Arthington 2008). Further, Jardine *et al.* (2015) found that zooplankton accounted for 50% of assimilated carbon in small (~1 g) *N. erebi*, declining to 25% in fish as large as 500 g. Phytoplankton are typically highly depleted in  $^{13}\text{C}$  (Vuorio *et al.* 2006) and are the most likely source of carbon for zooplankton in the present study. In contrast to benthic algal production, planktonic algal production likely contributes to carbon assimilation in small individuals by their consumption of zooplankton.

Thus, algae and zooplankton potentially contribute approximately one-half of the carbon assimilated by large *N. erebi*, whereas zooplankton contribute approximately one-half of the

**Table 3.** Summary of results of multiple regression analyses for comparisons of isotope values of sizes classes of *Nematalosa erebi* and putative food sourcesSmall, <100 mm; large, >100 mm. n.s.,  $P > 0.05$ ; \*,  $P < 0.05$ ; \*\*,  $P < 0.01$ ; \*\*\*,  $P < 0.001$ 

Isotope	Size class	Source	Estimated slope	<i>t</i>	<i>F</i>
$^{13}\text{C}$	Small	Benthic algae	0.024	0.236 <sup>n.s.</sup>	$F_{3,29} = 167.7^{***}$
		Terrestrial vegetation	0.198	1.163 <sup>NS</sup>	
		Zooplankton	0.699	5.294 <sup>***</sup>	
	Large	Benthic algae	0.278	3.257 <sup>**</sup>	$F_{3,31} = 193.8^{***}$
		Terrestrial vegetation	0.463	3.465 <sup>**</sup>	
		Zooplankton	0.218	2.027 <sup>NS</sup>	
$^{15}\text{N}$	Small	Benthic algae	0.069	0.322 <sup>NS</sup>	$F_{3,24} = 0.992^{NS}$
		Terrestrial vegetation	-0.276	-1.232 <sup>NS</sup>	
		Zooplankton	0.500	4.229 <sup>***</sup>	
	Large	Benthic algae	0.079	0.589 <sup>NS</sup>	$F_{3,25} = 5.967^{**}$
		Terrestrial vegetation	0.296	1.449 <sup>NS</sup>	
		Zooplankton	0.115	1.172 <sup>NS</sup>	

carbon assimilated by small *N. erebi*. What then accounts for the remaining fractions? Whereas spatial variation and correlation between source and consumer isotope values proved useful here for quantifying the contribution of algae and zooplankton, the minimal spatial variation in  $\delta^{13}\text{C}$  values of TVEG provided little scope for doing so. Nonetheless, stomach content analysis clearly indicates that detritus is the dominant food item, and the near absence of potential food items other than zooplankton or algae in stomach contents strongly suggests that we have not failed to consider or assess other potential sources. Moreover, the multiple regression analysis strongly supported a significant contribution by TVEG to *N. erebi* biomass. Thus, it seems most parsimonious to suggest that terrestrial detritus is, indeed, the missing source, despite the failure to detect a correlation between detrital  $\delta^{13}\text{C}$  values and those of fish, and the apparent poor nutritional quality of this food source (Brett *et al.* 2017). In addition, dead phytoplankton that have entered the detrital pool may have also contributed to the carbon assimilated by *N. erebi*.

It is rare for detritus not to have attached or embedded bacteria and fungi (Bowen 1987; Findlay *et al.* 2002). Detrital  $\delta^{13}\text{C}$  values do not change greatly with conditioning, and thus the isotope value of detritus, and of the microbial community living upon it, reflects its source origin (Finlay and Kendall 2007). As a result,  $\delta^{13}\text{C}$  values alone are unlikely to differentiate between carbon derived from detritus and that derived from microorganisms feeding upon that detritus. Given the refractory nature of vascular plant detritus, its nutritive value may be derived mostly from these attached organisms (France 2011) despite their low biomass relative to their substrate (Bowen 1987). Smoot and Findlay (2010) showed that the ingesta of the closely related facultative detritivore *D. cepedianum* contained eightfold more low-density material and was nutritionally enriched than the detrital or sediment material upon which it foraged. Moreover, the microbial biomass in ingesta was sevenfold greater than sediment. Smoot and Findlay (2010) suggested this living component of detritus was used as a food source by *D. cepedianum*. A similar comparison has not been performed for *N. erebi*. If, however, *N. erebi* possesses the same capacity to winnow detrital particles of differing quality, then it is possible that assimilation of carbon and nitrogen derived from

microbiota feeding upon detritus is substantial. There is scant information on  $^{15}\text{N}$  fractionation by microorganisms, making any interpretation of enrichment patterns in consumers of this form of prey difficult (Vanderklift and Ponsard 2003); however, the high availability of microbial biomass within the detrital pool can exert a disproportionate effect on enrichment dynamics on higher-order consumers that feed from both brown and green food chains (Steffan *et al.* 2017). We estimated a trophic enrichment of 4.1 and 3.2‰ for large and small *N. erebi* respectively; these values are not dissimilar to the ~3‰ per trophic level increase reported by Vander Zanden and Rasmussen (2001) and Post (2002). Bunn *et al.* (2013) reported a trophic enrichment of  $3.9 \pm 1.4$ ‰ for a range of Australasian herbivorous fishes.

Although the quality of the fine detrital fraction may not be as high as that of algae, and certainly not that of zooplankton, it is nonetheless an abundant food source. Moreover, if the higher-value microbial fraction can be separated from lesser-quality larger fractions, then its value is increased further. Fish faced with a diet of low or reduced quality, particularly of protein, can compensate by increasing consumption rates to meet both energy and essential nutrient demands provided the food source is not limiting, which is not usually the case for detritus. Notwithstanding the constraint imposed by the absence of intestinal structures enabling the processing of algae or detritus (e.g. the muscular gizzard is largely absent in fish <60 mm in length), switching between algal, detrital and zooplankton sources to achieve a blended diet across green and brown food chains may enable juvenile *N. erebi* >60 mm in length to achieve and maintain high growth and the intake of essential nutrients such as limiting amino acids and PUFAs.

This study has shown that detritus (with or without associated microbiota), algae and zooplankton are all important sources of carbon and nutrients for *N. erebi*. This species is almost ubiquitous across northern Australia and may dominate fish biomass (Pusey *et al.* 2017). It is itself consumed by many higher-order predators, some of which can move great distances, even across catchment boundaries in the case of water birds (Kingsford *et al.* 2010). Thus, the contribution of terrestrially derived carbon to *N. erebi* biomass, albeit occurring with low efficiency, may be

translated up into higher trophic levels of aquatic food webs of northern Australia. Furthermore, the liberation of nutrients due to mass mortality of *N. erebi* in dry season waterholes of arid zone rivers contributes greatly to the production dynamics of dry season waters (Burford *et al.* 2008). *N. erebi* is clearly an important component of riverine food webs. Although not entirely dependent on detritus as a food source, detritus is an important component of the diet of *N. erebi*, and may thus contribute more to tropical and subtropical Australian aquatic food webs than previously considered. Our knowledge of the biology of *N. erebi* is scant, particularly in regard to the relationships among hydrological variation, reproduction and movement. Changes in flow regimes and connectivity between parts of the riverine landscape arising from the expansion of water resource use in northern Australia (Douglas *et al.* 2011; Pettit *et al.* 2017) and that affect the production dynamics of *N. erebi* have the potential to disrupt riverine food webs (Turschwell *et al.* 2019). The present study has shown that both detritus and algae are important sources of energy and nutrients for this common species, and hence for food web structure in general. Moreover, the findings support the assertion by Jardine *et al.* (2015) that a focus on an algal–detrital dichotomy is unhelpful and that a greater focus on the circumstances in which species switch between different food sources would provide a better appreciation of the way in which food webs are structured and how they may change in response to changes in hydrology. Furthermore, a greater focus on the carbon sources supporting zooplankton production is warranted because zooplankton are key to early life history development of *N. erebi*, and probably to that of most other freshwater fish species of the region.

### Conflicts of interest

The authors declare that they have no conflicts of interest.

### Declaration of funding

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### References

- Akiyama, T., Oohara, I., and Yamamoto, T. (1997). Comparison of essential amino acid requirements with A/E ratio among fish species. *Fisheries Science* **63**, 963–970. doi:10.2331/FISHSCI.63.963
- Arthington, A. H., Bunn, S. E., and Gray, M. (1992). Tully–Millstream hydroelectric scheme, final report on additional limnological studies. Griffith University. Brisbane, Qld, Australia.
- Atkins, B. (1984). Feeding ecology of *Nematalosa erebi* in the lower River Murray. B.Sc.(Hons) Thesis, University of Adelaide, Adelaide, SA, Australia.
- Balcombe, S. R., Bunn, S. E., McKenzie-Smith, F. J., and Davies, P. M. (2005). Variability of fish diets between dry and flood periods in an arid zone floodplain river. *Journal of Fish Biology* **67**, 1552–1567. doi:10.1111/J.1095-8649.2005.00858.X
- Barnes, C., Sweeting, C. J., Jennings, S., Barry, J. T., and Polunin, N. V. (2007). Effect of temperature and ration size on carbon and nitrogen stable isotope trophic fractionation. *Functional Ecology* **21**, 356–362. doi:10.1111/J.1365-2435.2006.01224.X
- Beesley, L. (2006). Environmental stability: its role in structuring fish communities and life history strategies in the Fortescue River, Western Australia. Ph.D. Thesis, The University of Western Australia, Perth, WA, Australia.
- Belicka, L. L., Sokol, E. R., Hoch, J. M., Jaffé, R., and Trexler, J. C. (2012). A molecular and stable isotopic approach to investigate algal and detrital energy pathways in a freshwater marsh. *Wetlands* **32**, 531–542. doi:10.1007/S13157-012-0288-6
- Bishop, K. A., Allen, S. A., Pollard, D. A., and Cook, M. G. (2001). Ecological studies on the freshwater fishes of the alligator rivers region, Northern Territory: autecology. Office of the Supervising Scientist Report 145, Supervising Scientist, Darwin, NT, Australia.
- Blanchette, M. L., Davis, A. M., Jardine, T. D., and Pearson, R. G. (2014). Omnivory and opportunism characterize food webs in a large dry-tropics river system. *Freshwater Science* **33**, 142–158. doi:10.1086/674632
- Bluhdorn, D. R., and Arthington, A. H. (1994). 'The Effects of Flow Regulation in the Barker–Barambah Catchment. Volume 2: Biotic Studies and Synthesis.' (Griffith University: Brisbane, Qld, Australia.)
- Bowen, S. H. (1987). Composition and nutritional value of detritus. In 'Detritus and Microbial Ecology in Aquaculture: Proceedings of the Conference on Detrital Systems for Aquaculture', 26–31 August 1985, Como, Italy. (Eds D. J. W. Moriarty and R. S. V. Pullin.) pp. 192–216. (ICLARM: Manila, Philippines.)
- Brett, M. T., Kainz, M. J., Taipale, S. J., and Seshan, H. (2009). Phytoplankton, not allochthonous carbon, sustains herbivorous zooplankton production. *Proceedings of the National Academy of Sciences of the United States of America* **106**, 21197–21201. doi:10.1073/PNAS.0904129106
- Brett, M. T., Bunn, S. E., Chandra, S., Galloway, A. W., Guo, F., Kainz, M. J., Kankaala, P., Lau, D. C., Moulton, T. P., Power, M. E., and Rasmussen, J. B. (2017). How important are terrestrial organic carbon inputs for secondary production in freshwater ecosystems? *Freshwater Biology* **62**, 833–853. doi:10.1111/FWB.12909
- Bunn, S. E., Davies, P. M., and Winning, M. (2003). Sources of organic carbon supporting the food web of an arid zone floodplain river. *Freshwater Biology* **48**, 619–635. doi:10.1046/J.1365-2427.2003.01031.X
- Bunn, S. E., Balcombe, S. R., Davies, P. M., Fellows, C. S., and McKenzie-Smith, F. J. (2006). Aquatic productivity and food webs of desert river ecosystems. In 'Ecology of Desert Rivers'. (Ed. R. Kingsford.) pp. 76–99. (Cambridge University Press: Cambridge, UK.)
- Bunn, S. E., Leigh, C., and Jardine, T. D. (2013). Diet–tissue fractionation of  $\delta^{15}\text{N}$  by consumers from streams and rivers. *Limnology and Oceanography* **58**, 765–773. doi:10.4319/LO.2013.58.3.0765
- Burford, M. A., Cook, A. J., Fellows, C. S., Balcombe, S. R., and Bunn, S. E. (2008). Sources of carbon fuelling production in an arid floodplain river. *Marine and Freshwater Research* **59**, 224–234. doi:10.1071/MF07159
- Coates, D. (1993). Fish ecology and management of the Sepik–Ramu, New Guinea, a large contemporary tropical river basin. *Environmental Biology of Fishes* **38**, 345–368. doi:10.1007/BF00007528
- Douglas, M. M., Bunn, S. E., and Davies, P. M. (2005). River and wetland food webs in Australia's wet–dry tropics: general principles and implications for management. *Marine and Freshwater Research* **56**, 329–342. doi:10.1071/MF04084

- Douglas, M., Jackson, S., Pusey, B., Kennard, M., and Burrows, D. (2011). Northern futures: threats and opportunities for freshwater ecosystems. In 'Aquatic Biodiversity of the Wet-Dry Tropics of Northern Australia: Patterns, Threats and Future'. (Ed. B. J. Pusey.) pp 203–220. (Charles Darwin University Press: Darwin, NT, Australia.)
- Egan, J. P., Bloom, D. D., Kuo, C. H., Hammer, M. P., Tongnunui, P., Iglésias, S. P., Sheaves, M., Grudpan, C., and Simons, A. M. (2018). Phylogenetic analysis of trophic niche evolution reveals a latitudinal herbivory gradient in Clupeoidei (herrings, anchovies, and allies). *Molecular Phylogenetics and Evolution* **124**, 151–161. doi:10.1016/J.YMPEV.2018.03.011
- Findlay, S., Tank, J., Dye, S., Valett, H. M., Mulholland, P. J., McDowell, W. H., Johnson, S. L., Hamilton, S. K., Edmonds, J., Dodds, W. K., and Bowden, W. B. (2002). A cross-system comparison of bacterial and fungal biomass in detritus pools of headwater streams. *Microbial Ecology* **43**, 55–66. doi:10.1007/S00248-001-1020-X
- Finlay, J. C. (2004). Patterns and controls of lotic algal stable carbon isotope ratios. *Limnology and Oceanography* **49**, 850–861. doi:10.4319/LO.2004.49.3.0850
- Finlay, J. C., and Kendall, C. (2007). Stable isotope tracing of temporal and spatial variability in organic matter sources to freshwater ecosystems. *Stable Isotopes in Ecology and Environmental Science* **2**, 283–333. doi:10.1002/9780470691854.CH10
- Flecker, A. S. (1996). Ecosystem engineering by a dominant detritivore in a diverse tropical stream. *Ecology* **77**, 1845–1854. doi:10.2307/2265788
- France, R. (2011). Leaves as 'crackers', biofilm as 'peanut butter': exploratory use of stable isotopes as evidence for microbial pathways in detrital food webs. *Oceanological and Hydrobiological Studies* **40**, 110–115. doi:10.2478/S13545-011-0047-Y
- Goulding, M., Carvalho, M. L., and Ferreira, E. G. (1988). 'Rio Negro, Rich Life in Poor Water. Amazonian Diversity and Foodchain Ecology as Seen Through Fish Communities.' (SPB Academic Publishing: Amsterdam, Netherlands.)
- Guo, F., Kainz, M. J., Sheldon, F., and Bunn, S. E. (2016a). The importance of high-quality algal food sources in stream food webs – current status and future perspectives. *Freshwater Biology* **61**, 815–831. doi:10.1111/FWB.12755
- Guo, F., Kainz, M. J., Sheldon, F., and Bunn, S. E. (2016b). Effects of light and nutrients on periphyton and the fatty acid composition and somatic growth of invertebrate grazers in subtropical streams. *Oecologia* **181**, 449–462. doi:10.1007/S00442-016-3573-X
- Hortle, K. G., and Person, R. G. (1990). Fauna of the Annan River system, far north Queensland, with reference to the impact of tin mining. I. Fishes. *Marine and Freshwater Research* **41**, 677–694. doi:10.1071/MF9900677
- Jardine, T. D., Pettit, N. E., Warfe, D. M., Pusey, B. J., Ward, D. P., Douglas, M. M., Davies, P. M., and Bunn, S. E. (2012a). Consumer–resource coupling in wet–dry tropical rivers. *Journal of Animal Ecology* **81**, 310–322. doi:10.1111/J.1365-2656.2011.01925.X
- Jardine, T. D., Pusey, B. J., Hamilton, S. K., Pettit, N. E., Davies, P. M., Douglas, M. M., Sinnamoni, V., Halliday, I. A., and Bunn, S. E. (2012b). Fish mediate high food web connectivity in the lower reaches of a tropical floodplain river. *Oecologia* **168**, 829–838. doi:10.1007/S00442-011-2148-0
- Jardine, T. D., Hunt, R. J., Faggotter, S. J., Valdez, D., Burford, M. A., and Bunn, S. E. (2013). Carbon from periphyton supports fish biomass in waterholes of a wet–dry tropical river. *River Research and Applications* **29**, 560–573. doi:10.1002/RRA.2554
- Jardine, T. D., Woods, R., Marshall, J., Fawcett, J., Lobegeiger, J., Valdez, D., and Kainz, M. J. (2015). Reconciling the role of organic matter pathways in aquatic food webs by measuring multiple tracers in individuals. *Ecology* **96**, 3257–3269. doi:10.1890/14-2153.1
- Jardine, T. D., Rayner, T. S., Pettit, N. E., Valdez, D., Ward, D. P., Lindner, G., Douglas, M. M., and Bunn, S. E. (2017). Body size drives allochthony in food webs of tropical rivers. *Oecologia* **183**, 505–517. doi:10.1007/S00442-016-3786-Z
- Junk, W. J., Bayley, P. B., and Sparks, R. E. (1989). The flood pulse concept in river–floodplain systems. *Canadian Special Publication of Fisheries and Aquatic Sciences* **106**, 110–127.
- Kennard, M. J. (1995). Factors influencing freshwater fish assemblages in floodplain lagoons of the Normanby River, Cape York Peninsula: a large tropical Australian river. Ph.D. Thesis, Griffith University, Brisbane, Qld, Australia.
- Kennard, M. J., Pusey, B. J., and Arthington, A. H. (2001). Trophic Ecology of freshwater fishes in Australia. CRC Freshwater Ecology Scoping Study SCD6, CRC for Freshwater Ecology, Brisbane, Qld, Australia.
- Kingsford, R. T., Roshier, D. A., and Porter, J. L. (2010). Australian waterbirds – time and space travellers in dynamic desert landscapes. *Marine and Freshwater Research* **61**, 875–884. doi:10.1071/MF09088
- Leigh, C., Burford, M. A., Sheldon, F., and Bunn, S. E. (2010). Dynamic stability in dry season food webs within tropical floodplain rivers. *Marine and Freshwater Research* **61**, 357–368. doi:10.1071/MF09107
- Lewis, W. M., Jr, Hamilton, S. K., Rodriguez, M. A., Saunders, J. F., III, and Lasi, M. A. (2001). Foodweb analysis of the Orinoco floodplain based on production estimates and stable isotope data. *Journal of the North American Benthological Society* **20**, 241–254. doi:10.2307/1468319
- Lowe-McConnell, R. H. (1975). 'Fish Communities in Tropical Freshwaters: Their Distribution, Ecology, and Evolution.' (Longman: London, UK.)
- McGoldrick, D. J., Barton, D. R., Power, M., Scott, R. W., and Butler, B. J. (2008). Dynamics of bacteria–substrate stable isotope separation: dependence on substrate availability and implications for aquatic food web studies. *Canadian Journal of Fisheries and Aquatic Sciences* **65**, 1983–1990. doi:10.1139/F08-109
- Medeiros, E. S., and Arthington, A. H. (2008). The importance of zooplankton in the diets of three native fish species in floodplain waterholes of a dryland river, the Macintyre River, Australia. *Hydrobiologia* **614**(1), 19–31. doi:10.1007/S10750-008-9533-7
- Medeiros, E. S., and Arthington, A. H. (2011). Allochthonous and autochthonous carbon sources for fish in floodplain lagoons of an Australian dryland river. *Environmental Biology of Fishes* **90**, 1–17. doi:10.1007/S10641-010-9706-X
- Medeiros, E. S., and Arthington, A. H. (2014). Fish diet composition in floodplain lagoons of an Australian dryland river in relation to an extended dry period following flooding. *Environmental Biology of Fishes* **97**, 797–812. doi:10.1007/S10641-013-0180-0
- Moore, J. W., and Semmens, B. X. (2008). Incorporating uncertainty and prior information into stable isotope mixing models. *Ecology Letters* **11**, 470–480. doi:10.1111/J.1461-0248.2008.01163.X
- Moore, J. C., Berlow, E. L., Coleman, D. C., de Ruiter, P. C., Dong, Q., Hastings, A., Johnson, N. C., McCann, K. S., Melville, K., Morin, P. J., and Nadelhoffer, K. (2004). Detritus, trophic dynamics and biodiversity. *Ecology Letters* **7**, 584–600. doi:10.1111/J.1461-0248.2004.00606.X
- Morgan, D. L., Rowland, A. J., Gill, H. S., and Doupé, R. G. (2004). The implications of introducing a large piscivore (*Lates calcarifer*) into a regulated northern Australian river (Lake Kununurra, Western Australia). *Lakes and Reservoirs: Research and Management* **9**, 181–193. doi:10.1111/J.1440-1770.2004.00247.X
- Murray, D. S., Hager, H., Tocher, D. R., and Kainz, M. J. (2014). Effect of partial replacement of dietary fish meal and oil by pumpkin kernel cake and rapeseed oil on fatty acid composition and metabolism in Arctic charr (*Salvelinus alpinus*). *Aquaculture* **431**, 85–91. doi:10.1016/J.AQUACULTURE.2014.03.039
- Pettit, N. E., Naiman, R. J., Warfe, D. M., Jardine, T. D., Douglas, M. M., Bunn, S. E., and Davies, P. M. (2017). Productivity and connectivity in tropical riverscapes of northern Australia: ecological insights for management. *Ecosystems* **20**, 492–514. doi:10.1007/S10021-016-0037-4

- Post, D. M. (2002). Using stable isotopes to estimate trophic position: models, methods, and assumptions. *Ecology* **83**, 703–718. doi:10.1890/0012-9658(2002)083[0703:USITET]2.0.CO;2
- Pusey, B. J., Read, M. G., and Arthington, A. H. (1995). The feeding ecology of freshwater fishes in two rivers of the Australian Wet Tropics. *Environmental Biology of Fishes* **43**, 85–103. doi:10.1007/BF00001820
- Pusey, B. J., Arthington, A. H., and Read, M. G. (2000). The dry season diet of freshwater fishes in monsoonal tropical rivers of Cape York Peninsula, Australia. *Ecology Freshwater Fish* **9**, 177–190. doi:10.1111/J.1600-0633.2000.EFF090307.X
- Pusey, B. J., Kennard, M. J., and Arthington, A. H. (2004). 'Freshwater Fishes of North-eastern Australia'. (CSIRO Publishing: Melbourne, Vic., Australia.)
- Pusey, B. J., Arthington, A. H., Stewart-Koster, B., Kennard, M. J., and Read, M. G. (2010). Widespread omnivory in freshwater fish assemblages of a hydrologically variable northern Australian river. *Journal of Fish Biology* **77**, 731–753.
- Pusey, B. J., Burrows, D. W., Kennard, M. J., Perna, C. N., Unmack, P. J., Allsop, Q., and Hammer, M. P. (2017). Freshwater fishes of northern Australia. *Zootaxa* **4253**, 1–104. doi:10.11646/ZOOTAXA.4253.1.1
- Rasmussen, J. B. (2010). Estimating terrestrial contribution to stream invertebrates and periphyton using a gradient-based mixing model for  $\delta^{13}\text{C}$ . *Journal of Animal Ecology* **79**, 393–402. doi:10.1111/J.1365-2656.2009.01648.X
- Rayner, T. S., Pusey, B. J., and Pearson, R. G. (2009). Spatio-temporal dynamics of fish feeding in the lower Mulgrave River, north-eastern Queensland: the influence of seasonal flooding, instream productivity and invertebrate abundance. *Marine and Freshwater Research* **60**, 97–111. doi:10.1071/MF08055
- Reid, D. J., Quinn, G. P., Lake, P. S., and Reich, P. (2008). Terrestrial detritus supports the food webs in lowland intermittent streams of south-eastern Australia: a stable isotope study. *Freshwater Biology* **53**, 2036–2050. doi:10.1111/J.1365-2427.2008.02025.X
- Roach, K. A. (2013). Environmental factors affecting incorporation of terrestrial material into large river food webs. *Freshwater Science* **32**, 283–298. doi:10.1899/12-063.1
- Rooney, N., McCann, K., Gellner, G., and Moore, J. C. (2006). Structural asymmetry and the stability of diverse food webs. *Nature* **442**, 265–269. doi:10.1038/NATURE04887
- Smoot, J. C., and Findlay, R. H. (2010). Microbes as food for sediment-ingesting detritivores: low-density particles confer a nutritional advantage. *Aquatic Microbial Ecology* **59**, 103–109. doi:10.3354/AME01383
- Solomon, C. T., Carpenter, S. R., Clayton, M. K., Cole, J. J., Coloso, J. J., Pace, M. L., Vander Zanden, M. J., and Weidel, B. C. (2011). Terrestrial, benthic, and pelagic resource use in lakes: results from a three-isotope Bayesian mixing model. *Ecology* **92**, 1115–1125. doi:10.1890/10-1185.1
- Steffan, S. A., Chikaraishi, Y., Dharampal, P. S., Pauli, J. N., Guédot, C., and Ohkouchi, N. (2017). Unpacking brown food-webs: animal trophic identity reflects rampant microbivory. *Ecology and Evolution* **7**, 3532–3541. doi:10.1002/ECE3.2951
- Sternberg, D., Balcombe, S., Marshall, J., and Lobegeiger, J. (2008). Food resource variability in an Australian dryland river: evidence from the diet of two generalist native fish species. *Marine and Freshwater Research* **59**, 137–144. doi:10.1071/MF07125
- Taylor, A. N., and Batzer, D. P. (2010). Spatial and temporal variation in invertebrate consumer diets in forested and herbaceous wetlands. *Hydrobiologia* **651**, 145–159. doi:10.1007/S10750-010-0283-Y
- Thorburn, D. C., Gill, H., and Morgan, D. L. (2014). Predator and prey interactions of fishes of a tropical Western Australia river revealed by dietary and stable isotope analyses. *Journal of the Royal Society of Western Australia* **97**, 363–387.
- Thorp, J. H., and Delong, M. D. (1994). The riverine productivity model: an heuristic view of carbon sources and organic processing in large river ecosystems. *Oikos* **70**, 305–308. doi:10.2307/3545642
- Turschwell, M. P., Stewart-Koster, B., Pusey, B. J., Douglas, M., King, A., Crook, D., Boone, E., Allsop, Q., and Kennard, M. J. (2019). Flow-mediated predator-prey dynamics influences fish populations in a tropical river. *Freshwater Biology* **64**, 1453–1466. doi:10.1111/FWB.13318
- Unmack, P. (2013). Biogeography. In 'Ecology of Australian Freshwater Fishes'. (Eds P. Humphries and K. Walker.) pp. 25–48. (CSIRO Publishing: Melbourne, Vic., Australia.)
- Vander Zanden, M. J., and Rasmussen, J. B. (2001). Variation in  $\delta^{15}\text{N}$  and  $\delta^{13}\text{C}$  trophic fractionation: implications for aquatic food web studies. *Limnology and Oceanography* **46**, 2061–2066. doi:10.4319/LO.2001.46.8.2061
- Vanderklift, M. A., and Ponsard, S. (2003). Sources of variation in consumer-diet  $\delta^{15}\text{N}$  enrichment: a meta-analysis. *Oecologia* **136**, 169–182. doi:10.1007/S00442-003-1270-Z
- Vannote, R. L., Minshall, G. W., Cummins, K. W., Sedell, J. R., and Cushing, C. E. (1980). The river continuum concept. *Canadian Journal of Fisheries and Aquatic Sciences* **37**, 130–137. doi:10.1139/F80-017
- Vuorio, K., Meili, M., and Sarvala, J. (2006). Taxon-specific variation in the stable isotopic signatures ( $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$ ) of lake phytoplankton. *Freshwater Biology* **51**, 807–822. doi:10.1111/J.1365-2427.2006.01529.X
- Winemiller, K. O. (2004). Floodplain river food webs: generalizations and implications for fisheries management. In 'Proceedings of the Second International Symposium on the Management of Large Rivers for Fisheries', 11–14 February 2003, Phnom Penh, Cambodia, (Eds R. L. Welcomme and T. Petr.) Vol. 2, pp. 285–309. (Food and Agriculture Organization of the United Nations and Mekong River Commission, FAO Regional Office for Asia and the Pacific: Rome, Italy.)

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