Journal of FISH BIOLOGY
Published for the Fisheries Society of the British Isles

Journal of Fish Biology (2012) **80**, 225–231
doi:10.1111/j.1095-8649.2011.03163.x, available online at wileyonlinelibrary.com

Stable isotopes challenge the perception of ocean sunfish *Mola mola* as obligate jellyfish predators

J. Syväranta*†, C. Harrod‡§, L. Kubicek‖, V. Cappanera¶
and J. D. R. Houghton‡**††

**Department of Biological and Environmental Science, University of Jyväskylä, Jyväskylä, Finland, †EcoLab, UMR 5245, CNRS/Université de Toulouse, 118, route de Narbonne, 31062 Toulouse Cedex 9, France, ‡School of Biological Sciences, Queen's University Belfast, MBC Building, 97 Lisburn Road, Belfast BT9 7BL, Northern Ireland, U.K., §Facultad de Recursos del Mar, Instituto de Investigaciones Oceanológicas, Universidad Antofagasta, Chile, ‖Whale Observation Project, W.O.P. Centre, Aussermatt, 3532 Zäziwil, Switzerland, ¶Portofino Marine Protected Area, V. le Rainusso, 1-16038 S. Margherita Ligure (GE), Italy and **Queen's University Marine Laboratory, 12-13 The Strand, Portaferry, Co. Down, BT22 1PF, Northern Ireland, U.K.*

(Received 10 January 2011, Accepted 14 October 2011)

Evidence is provided from stable isotope analysis that aggregations of small ocean sunfish *Mola mola* (total length <1 m) feed broadly within coastal food webs and their classification as obligate predators of gelatinous zooplankton requires revision.

Key words: food web; Mediterranean Sea; SIA; trophic ecology.

The ocean sunfish *Mola mola* (L. 1758) (Molidae) is the world's heaviest bony fish (maximum mass = 2235 kg; Carwardine, 1995) with a distribution spanning all tropical and temperate seas (Pope *et al.,* 2010). Significant gaps exist in knowledge of the species through a lack of commercial interest and exclusion from regional and international conservation strategies that focus research efforts upon certain target groups (Sims *et al.,* 2009). These problems are compounded by a historical perception that *M. mola* are solitary and rare pelagic vagrants that impact very little upon the marine systems they drift through (Pope *et al.,* 2010). A series of studies have shown, however, that large aggregations of small *M. mola* total length (L_{T}, typically <1 m) can be highly abundant in coastal waters (Silvani *et al.,* 1999; Sims & Southall, 2002; Houghton *et al.,* 2006) with the overall inference that such locale might serve some purpose as a developmental habitat.

There is an urgent need to reconsider the trophic position of *M. mola* aggregations as the species is removed *en masse* each year as fisheries by-catch with little or no understanding of the broader ecological consequences (Pope *et al.,* 2010). For

---

††Author to whom correspondence should be addressed. Tel.: +44 28 90972297; email: j.houghton@qub.ac.uk

example, the species contributed between 70 and 93% of the total fish catch in Spanish drift gillnet fisheries (targeted at swordfish *Xiphias gladius* L. 1758) within the Mediterranean Sea between 1992 and 1994 (Silvani *et al.,* 1999). Whilst the Spanish fishery was closed in 1994 (Silvani *et al.,* 1999; Tudela *et al.,* 2005), an illegal Moroccan drift net fleet continued to operate until 2007 with an estimated annual by-catch of *c*. 37 000 *M. mola* (Tudela *et al.,* 2005). When considering the overall implications of such wholesale removal from marine systems, it is vital that the putative prey groups that underpin these coastal aggregations are identified. *Mola mola* are typically considered obligate or primary consumers of gelatinous zooplankton (Fraser-Brunner, 1951) and whilst this view has pervaded the literature, it has little empirical support. Indeed, field guides and reference books provide tantalizing insights that the diet of *M. mola* might extend beyond gelatinous zooplankton by listing various stomach contents including algae, crustaceans, ophiuroids, molluscs, hydroids and fishes (Norman & Fraser, 1949; Clemens & Wilby, 1961; Hart, 1973), although how these observations were obtained is rarely specified (Pope *et al.,* 2010).

The best empirical evidence of jellyfish predation comes from Hooper *et al*. (1973) who considered the distribution of fatty acids in the lipids of four *M. mola* caught off Nova Scotia, Canada. High percentages of 18:0 and $20{:}4\omega6$ were found in the tissues which had previously been documented in the tissue of leatherback turtles *Dermochelys coriacea*, which unambiguously feed on jellyfish (Ackerman *et al.,* 1971). These findings in conjunction with numerous anecdotal observations (Pope *et al.,* 2010) provide confidence that *M. mola* do indeed feed on jellyfish. The issue which requires clarification is the nature of this predator and prey relationship (*i.e.* is it obligate or facultative, forming part of a more catholic diet?). To provide empirical data for dietary composition, the trophic position of eight small *M. mola* ($L_T < 1m$) caught as by-catch in fixed tuna nets [targeted at Atlantic bluefin tuna *Thunnus thynnus* (L. 1758)] near the Italian fishing port of Camogli on the Ligurian coast (44° 20′ 50·27 ″ N; 9° 09′ 21·78 ″ E) was opportunistically investigated during the summers of 2009 and 2010. This historical fishery dates back to the 17th century, with fishermen typically releasing *M. mola* unharmed in their efforts to minimize by-catch (Cappanera *et al*., 2010).

Using stable isotope analysis (SIA), the general hypothesis that aggregations of small *M. mola* may be better integrated into classical marine food webs than previously thought was examined. Analyses of carbon (δ^{13}C) and nitrogen (δ^{15}N) stable isotope ratios of consumers and their putative prey provide time-integrated information about energy sources, food web structure and consumer trophic positions within ecosystems (Peterson & Fry, 1987). Generally, δ^{15}N values increase by 2–4‰ per trophic level and can be used to define trophic levels of consumers, whereas δ^{13}C values change only little in trophic transfer (McCutchan *et al*., 2003). In marine systems, benthic and pelagic sources of C differ in terms of ^{13}C enrichment, with pelagic-derived C being depleted in ^{13}C relative to C derived from benthic or inshore primary producers (France, 1995; Mallela & Harrod, 2008).

A range of both invertebrate and fish species was used to characterize the study site and to place the trophic ecology of small *M. mola* into context. Taxa were collected to provide isotopic data from both putative pelagic- and benthic-associated consumers. Invertebrate samples included indicators of pelagic-derived C and N (crustacean zooplankton, mussels *Mytilus edulis*, jellyfish *Pelagia noctiluca* and two benthic consumers, the limpet *Patella caerulea* and gastropod *Nucella* sp.). Crustacean zooplankton were collected during horizontal surface tows using a 50 μm plankton net deployed for 30 min., with samples later sorted into 200–500 μm and >500 μm size fractions. Jellyfish medusae (>80 mm bell diameter) were collected with hand-nets from the boat while all benthic invertebrate samples were collected by Scuba diving. Tentacles, oral arms and gonads were removed from jellyfish and only the bells were used for SIA (Pitt *et al.,* 2009). Similarly, only the foot tissue of mussels and gastropods was dissected and used for SIA. Samples of the following fish species were obtained from local fishermen: barracuda *Sphyraena* spp., black scorpionfish *Scorpaena porcus* L. 1758, conger eel *Conger conger* (L. 1758), Atlantic chub mackerel *Scomber colias* Gmelin 1789, anchovy *Engraulis encrasicolus* (L. 1758) and *M. mola*. A small piece of muscle tissue was removed from the dorsal side of each fish, and subsequently stored in ethanol (*M. mola* samples in 2009) or frozen (all other fish samples). The samples were then transferred to the laboratory for further processing for SIA.

All samples for SIA were oven-dried (at 60° C), except jellyfish samples which were freeze-dried, and ground into a fine homogeneous powder using a mortar and pestle. Invertebrate samples were divided into two subsamples: one was acid-washed in 1 M HCl to remove carbonates and re-dried in an oven (for δ^{13}C), and the other was analysed without acid washing (for δ^{15}N). All stable isotope analyses were carried out at the University of Jyväskylä stable isotope facility, using pike *Esox lucius* L. 1758 muscle tissue as an internal working standard to ensure accurate analysis and to allow any required correction for linearity and drift. Internal precision for standards was always better than 0.2‰ for both C and N in each run. *Mola mola* samples collected in 2009 were first preserved in ethanol and dried after careful rinsing in the laboratory. Preservation in ethanol is typically shown to have only little impact (<1‰) on sample δ^{15}N values (Barrow *et al*., 2008; Syväranta *et al*., 2008), while the impacts on δ^{13}C values can be more variable. This probably relates to removal of lipids by ethanol (Syväranta *et al*., 2008) and lipids are known to be depleted in ^{13}C leading to lower δ^{13}C values analysed from tissues with greater lipid content, particularly in fish and zooplankton samples (DeNiro & Epstein, 1977; Kiljunen *et al*., 2006; Syväranta & Rautio, 2010). The C:N ratios in fish muscle and zooplankton samples ranged between 3·4–4·4 and 5·9–6·2, and those of ethanol-preserved *M. mola* samples 2·8–3·5, indicating higher lipid contents in tissues with higher C:N ratios. Therefore, all fish δ^{13}C values to lipid-free muscle tissue were corrected using a lipid normalization model (Kiljunen *et al*., 2006) allowing the comparison of ethanol preserved and untreated *M. mola* samples. The zooplankton samples were corrected using a similar lipid normalization model, but specifically revised for zooplankton samples (Syväranta & Rautio, 2010). Nonetheless, it is conceded that preservation in ethanol might have changed the isotope values very slightly (Edwards *et al*., 2002; Sweeting *et al*., 2004; Kelly *et al*., 2006; Syväranta *et al*., 2008), but it is felt that the magnitude of such potential effects did not compromise the analysis.

On average, the small *M. mola* examined here were enriched in ^{13}C by 2.3‰ and in ^{15}N by 5·7‰ relative to that of their putative diet of gelatinous zooplankton (here represented by *P. noctiluca*; see Fig. 1). These values are approximately twice that of typical estimates of diet-tissue trophic fractionation for fish muscle tissue (McCutchan *et al*., 2003; mean ± S.D. $\delta^{13}\text{C} = 1.3 \pm 1.3$‰, $\delta^{15}\text{N} = 2.9 \pm 1.2$‰),

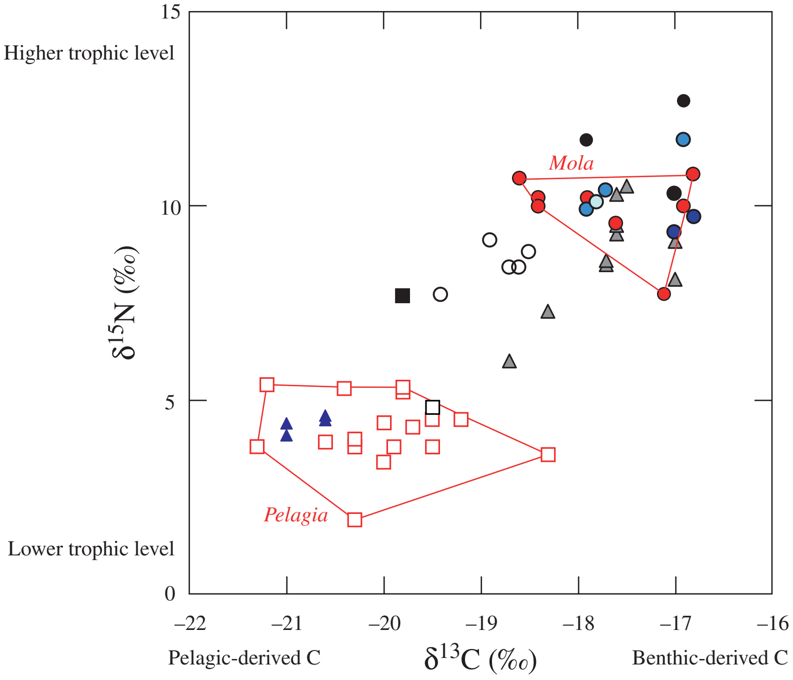

Fig. 1. Isotopic biplot showing the δ^{13}C and δ^{15}N values for *Mola mola* (●) and a selection of other fishes (●, *Scorpaena porcus*; ●, *Sphyraena* sp.; ●, *Conger conger*; ○, *Engraulis encrasicolus*; ●, *Scomber colias*). Also shown are values recorded from *Pelagia noctiluca* (□), small (□; 200–500 µm) and large (■; >500 µm) zooplankton collected at Camogli and molluscs (▲, *Nucella* sp.; ▲, *Mytilus edulis*). Note that δ^{13}C and δ^{15}N values of *M. mola* are enriched in both ^{13}C and ^{15}N relative to pelagic taxa, including their putative diet of jellyfish, indicating that they are unexpectedly utilizing C and N derived from a benthic source.

which suggested that carbon and nitrogen assimilated by the *M. mola* sampled here originated from an alternative source. Of the other fishes examined, all apart from the zooplanktivorous, *E. encrasicolus* (Plounevez & Champalbert, 2000) were significantly ^{13}C and ^{15}N enriched relative to indicators of pelagic-derived C and N (see values for jellyfish, zooplankton and *M. edulis* in Fig. 1). From their characteristically enriched δ^{13}C values, these fishes were probably members of a benthic and inshore food web (Jennings *et al.*, 1997; Pinnegar & Polunin, 2000; Mallela & Harrod, 2008). The small *M. mola* examined here were not statistically distinguishable from these other fishes (with the exception of *E. encrasicolus*) (MANOVA of δ^{13}C and δ^{15}N data: Pillai's trace $= 0.23$, $F_{2,14} = 2.06$, $P > 0.05$).

The present estimates of δ^{13}C and δ^{15}N for taxa associated with the pelagic food web (jellyfish, zooplankton and mussels) were characteristic of Mediterranean Sea pelagic ecosystems. δ^{13}C and δ^{15}N values for both large and small crustacean zooplankton were similar to that for Mediterranean Sea zooplankton [mean ± s.d. $\delta^{13}\text{C} = -19.8 \pm 1.0‰$ ($n = 117$); $\delta^{15}\text{N} = 3.8 \pm 1.9‰$ ($n = 120$)] calculated from the literature (Pinnegar & Polunin, 2000; Vizzini & Mazzola, 2005; Koppelmann *et al.*, 2009; Denda & Christiansen, 2011; Fanelli *et al.*, 2011). Although only a single species of jellyfish was sampled (the putative diet of *M. mola*), their isotopic values were not significantly different (NPMANOVA, $F_{2,14} = 0.07$, $P > 0.05$) from values for a wide range of Mediterranean Sea gelatinous zooplankton [mean ± S.D. δ^{13}C $= -19.9 \pm 1.5‰$ ($n = 17$), δ^{15}N $= 4.3 \pm 1.7‰$ ($n = 16$)] published elsewhere (Malej *et al.*, 1993; Pinnegar & Polunin, 2000; Polunin *et al.*, 2001; Fanelli *et al.*, 2011).

The present results indicate that small-bodied *M. mola* feed within inshore food webs in the Mediterranean Sea, and that the trophic role of *M. mola* extends beyond obligate predation on jellyfish. It is tempting to suggest that the availability of such mixed prey types may be an important factor in the habitat preferences of *M. mola* during early life-history stages. Without comparative studies of larger individuals found in offshore areas (Pope *et al.,* 2010), however, this suggestion remains speculative. One potential caveat is also the proximity of the present sampling to commercial fishing activities which may grant *M. mola* access to discards of species that may not typically form part of their diet in other areas (Hooper *et al.,* 1973; Houghton *et al.,* 2000). Juvenile *M. mola* were not, however, enriched in ^{15}N relative to other fishes, suggesting that they fed at a similar trophic level to the other fishes sampled here (excluding *E. encrasicolus*). This matter aside, the long-standing perception of obligate jellyfish predation by *M. mola* clearly requires some revision.

We extend our thanks to the Cooperative Society of Camogli Fishermen and the Portofino Marine Protected Area for the facilities and the use of boats during the sampling and to C. Schwarz and M. Schwarz for their generous assistance with sample collection and initial processing. We further thank two anonymous reviewers and the Assistant Editor for their comments.

References

Ackerman, R. G., Hooper, S. N. & Frair, W. (1971). Comparison of the fatty acid compositions of depot fats from fresh-water and marine turtles. *Comparative Biochemistry and Physiology B* **40,** 931–944.

Barrow, L. M., Bjorndal, K. A. & Reich, K. J. (2008). Effects of preservation method on stable carbon and nitrogen isotope values. *Physiological and Biochemical Zoology* **81,** 688–693.

Cappanera, V., Castellano, M., Bava, S., Povero, P., Fanciulli, G. & Cattaneo Vietti, R. (2010). Catch trend and meteo-marine condition inside the Camogli "Tonnarella" (Ligurian Sea). *Biologia Marina Mediterranea* **17,** 134–137.

Carwardine, M. (1995). *The Guinness Book of Animal Records.* London: Guinness Publishing.

Clemens, W. A. & Wilby, A. (1961). Fishes of the Pacific coast of Canada. *Bulletin of the Fisheries Research Board of Canada* **68**.

Denda, A. & Christiansen, B. (2011). Zooplankton at a seamount in the eastern Mediterranean: distribution and trophic interactions. *Journal of the Marine Biological Association of the United Kingdom* **91,** 33–49.

DeNiro, M. J. & Epstein, S. (1977). Mechanism of carbon isotope fractionation associated with lipid synthesis. *Science* **197,** 261-263.

Edwards, M. S., Turner, T. F. & Sharp, Z. D. (2002). Short- and long-term effects of fixation and preservation on stable isotope values (δ^{13}C, δ^{15}N, δ^{34}S) of fluid-preserved museum specimens. *Copeia* **2002,** 1106–1112.

Fanelli, E., Cartes, J. E. & Papiol, V. (2011). Food web structure of deep-sea macrozooplankton and micronekton off the Catalan slope: insight from stable isotopes. *Journal of Marine Systems* **87,** 79–89.

France, R. (1995). Carbon-13 enrichment in benthic compared to planktonic algae: foodweb implications. *Marine Ecology Progress Series* **124,** 307–312.

Fraser-Brunner, A. (1951). The ocean sunfishes (family Molidae). *Bulletin of the British Museum (Natural History) Zoology* **1,** 87–121.

Hart, J. L. (1973). Pacific fishes of Canada. *Bulletin of the Fisheries Research Board of Canada* **180**.

Hooper, S. N., Paradis, M. & Ackman, R. G. (1973). Distribution of trans-6-hexadecenoic acid, 7-methyl-7-hexadecenoic acid and common fatty acids in lipids of ocean sunfish *Mola mola*. *Lipids* **8,** 509–516.

Houghton, J. D. R., Woolmer, A. & Hays, G. C. (2000). Sea turtle diving and foraging behaviour around the Greek island of Kefalonia. *Journal of the Marine Biological Association of the United Kingdom* **80,** 761–762.

Houghton, J. D. R., Doyle, T. K., Davenport, J. & Hays, G. C. (2006). The ocean sunfish *Mola mola*: insights into distribution, abundance and behaviour in the Irish & Celtic Seas. *Journal of the Marine Biological Association of the United Kingdom* **86,** 1–7.

Jennings, S., Reñones, O., Morales-Nin, B., Polunin, N. V. C., Moranta, J. & Coll, J. (1997). Spatial variation in the ^{15}N and ^{13}C stable isotope composition of plants, invertebrates and fishes on Mediterranean reefs: Implications for the study of trophic pathways. *Marine Ecology Progress Series* **146,** 109–116.

Kelly, B., Dempson, J. B. & Power, M. (2006). The effects of preservation on fish tissue stable isotope signatures. *Journal of Fish Biology* **69,** 1595–1611.

Kiljunen, M., Grey, J., Sinisalo, T., Harrod, C., Immonen, H. & Jones, R. I. (2006). A revised model for lipid-normalizing δ^{13}C values from aquatic organisms, with implications for isotope mixing models. *Journal of Applied Ecology* **43,** 1213–1222.

Koppelmann, R., Böttger-Schnack, R., Möbius, J. & Weikert, H. (2009). Trophic relationships of zooplankton in the eastern Mediterranean based on stable isotope measurements. *Journal of Plankton Research* **31,** 669–686.

Malej, A., Faganeli, J. & Pezdič (1993). Stable isotope and biochemical fractionation in the marine pelagic food chain: the jellyfish *Pelagia noctiluca* and net zooplankton. *Marine Biology* **116,** 565–570.

Mallela, J. & Harrod, C. (2008). δ^{13}C and δ^{15}N reveal significant differences in the coastal foodwebs of the seas surrounding Trinidad and Tobago. *Marine Ecology Progress Series* **368**, 41–51.

McCutchan, J. H. Jr., Lewis, W. M. Jr., Kendall, C. & McGrath, C. C. (2003). Variation in trophic shift for stable isotope ratios of Carbon, Nitrogen and Sulfur. *Oikos* **102,** 378–390.

Norman, J. R. & Fraser, F. C. (1949). *Field Book of Giant Fishes.* New York, NY: G.P. Putnam and Sons.

Peterson, B. J. & Fry, B. (1987). Stable isotopes in ecosystem studies. *Annual Reviews in Ecology & Systematics* **18,** 293–320.

Pinnegar, J. K. & Polunin, N. V. C. (2000). Contributions of stable-isotope data to elucidating food webs of Mediterranean rocky littoral fishes. *Oecologia* **122,** 399–409.

Pitt, K. A., Connolly, R. M. & Meziane, T. (2009). Stable isotope and fatty acid tracers in energy and nutrient studies of jellyfish: a review. *Developments in Hydrobiology* **206,** 119–132.

Plounevez, S. & Champalbert, G. (2000). Diet, feeding behaviour and trophic activity of the anchovy (*Engraulis encrasicolus* L.) in the Gulf of Lions (Mediterranean Sea). *Oceanologica Acta* **23,** 175–192.

Polunin, N. V. C., Morales-Nin, B., Pawsey, W. E., Cartes, J. E., Pinnegar, J. K. & Moranta, J. (2001). Feeding relationships in Mediterranean bathyal assemblages elucidated by stable nitrogen and carbon isotope data. *Marine Ecology Progress Series* **220,** 13–23.

Pope, E. C., Hays, G. G., Thys, T. M., Doyle, T. K., Sims, N., Queroz, N., Hobson, V. J., Kubicek, L. & Houghton, J. D. R. (2010). The biology and ecology of the ocean sunfish *Mola mola*: a review of current knowledge and future research perspectives. *Reviews in Fish Biology and Fisheries* **20,** 471–487.

Silvani, L., Gazo, M. & Aguilar, A. (1999). Spanish driftnet fishing and incidental catches in the western Mediterranean. *Biological Conservation* **90,** 79–85.

Sims, D. W. & Southall, E. J. (2002). Occurrence of ocean sunfish, *Mola mola* near fronts in the western English Channel. *Journal of the Marine Biology Association of the United Kingdom* **82,** 927–928.

Sims, D. W., Queiroz, N., Doyle, T. K., Houghton, J. D. R & Hays, G. C. (2009). Satellite tracking of the world's largest bony fish, the ocean sunfish (*Mola mola*) in the North East Atlantic. *Journal of Experimental Marine Biology and Ecology* **370,** 127–133.

Sweeting, C. J., Polunin, N. V. C. & Jennings, S. (2004). Tissue and fixative dependent shifts of δ^{13}C and δ^{15}N in preserved ecological material. *Rapid Communications in Mass Spectrometry* **18,** 2587–2592.

Syväranta, J. & Rautio, M. (2010). Zooplankton, lipids and stable isotopes: importance of seasonal, latitudinal, and taxonomic differences. *Canadian Journal of Fisheries and Aquatic Sciences* **67,** 1721–1729.

Syväranta, J., Vesala, S., Rask, M., Ruuhijärvi, J. & Jones, R. I. (2008). Evaluating the utility of stable isotope analyses of archived freshwater sample materials. *Hydrobiologia* **600,** 121–130.

Tudela, S., Kai Kai, A., Maynou, F., El Andalossi, M. & Guglielmi, P. (2005). Driftnet fishing and biodiversity conservation: the case study of the large-scale Moroccan driftnet fleet operating in the Alboran Sea (SW Mediterranean). *Biological Conservation* **121,** 65–78.

Vizzini, S. & Mazzola, A. (2005). Feeding ecology of the sand smelt *Atherina boyeri* (Risso 1810) (Osteichthyes, Atherinidae) in the western Mediterranean: evidence for spatial variability based on stable carbon and nitrogen isotopes. *Environmental Biology of Fishes* **72,** 259–266.