

## **Do Tuna Farms Impact the Benthos? A Malta Case Study**

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### **Abstract**

Environmental monitoring of tuna penning in Malta was initiated in 2000 and is still ongoing. The impact of the activity on benthic macrofauna in the vicinity of offshore tuna cages was assessed using polychaetes and amphipods as indicators. Grab samples of sediment for faunal studies were collected on a 'bare sand' bottom from stations located: (i) adjacent to the tuna pens, (ii) some 100 m away from the cages; and (iii) at two reference sites located 1 km and 2 km away, for three offshore tuna farms situated in waters of circa 50 m depth. Samples were collected before initiation of the tuna penning activities and

afterwards on a six-monthly or annual basis. Results from analyses of data collected over a period of ten years (2000 – 2010) indicated that the tuna farming activities influenced the biotic assemblages associated with the sedimentary bottom habitat up to 1-2 km away from the cages, possibly due to transport of particulate organic matter via sea currents; this exceeds the area of influence previously reported in studies of single tuna farms carried out over short temporal scales, as well as for other types of Mediterranean fish farms such as those culturing seabass and seabream. The level and spatial extent of impact differed between the three tuna farms and seemed to depend on the farm size and the number of years of operation, as well as on local environmental factors. The level of adverse impact on benthic habitats was highest at the tuna farm located off the north-eastern coast (the largest in terms of holding capacity) compared to the other two farms located off the south-eastern coast. Results from the north-eastern farm during its first year of operation indicated an elevated abundance of capitellid polychaetes, below the cages and at a distance 100 m from the cages, while values of the polychaete/amphipod ratio indicated 'Bad' ecological quality status (EQS) which increased significantly during the fallow periods to 'High'/'Good' EQS during the first years of operation, with no significant difference till the end of the study period. It was concluded that changes in the macrofaunal assemblages resulted from accumulation of uneaten feed-fish on the seabed. Feed management at the north-eastern farm improved following the first years of operation, and was sufficient to mitigate the benthic impact of the tuna penning activities over several years of operation. On the other hand, the spatial extent of impact appeared to be largest at one of the farms located off the south-eastern coast, while the temporal pattern of impact over several years of operation reflected a press-type of disturbance at both farms located off the south-eastern coast. Such disturbance seems to have resulted from the cumulative effects of nutrient enrichment from the tuna farms and a higher nutrient loading of coastal waters in the southern parts of the Maltese Islands as a result of more intense coastal zone use, compared to the north of the islands. The two farms located off the south-eastern coast may also have had an additive effect, given that they are relatively close to each other (1 km apart); this highlights the importance of good spatial planning for coastal aquaculture activities. Finally, the high spatio-temporal variation of the influence of the tuna farms on benthic habitats highlights the importance of including multiple impacted and reference areas as well as replicated temporal sampling in assessing the environmental impacts of the activity.

## **Introduction**

Environmental monitoring of the aquaculture industry which causes deterioration of water quality and benthic habitats in the vicinity of fish cages (e.g. Wu, 1995; Hargrave et al., 1997), is important to ensure its sustainability. The uneaten feed and fish faeces which accumulate on the seabed below fish cages form a decomposing mass of organic matter that results in reducing sediment conditions (Giles, 2008). Crustacean infauna are the first to be adversely affected, and are replaced by a high abundance of deposit-feeding opportunist taxa, resulting in reduced benthic diversity (Giles, 2008). The periodic cessation of tuna farming activities (fallowing) is a sustainable aquaculture practice that allows recovery of the benthos to take place between production cycles (Macleod et al., 2006; 2007). However, long recovery times are reported for benthic assemblages following cessation of

aquaculture activities (Borja et al., 2010), indicating that the benthic community may return to the pre-fallowed state as soon as production is resumed (Pereira et al., 2004).

Farming of Atlantic bluefin tuna (*Thunnus thynnus thynnus* Linnaeus 1758) (ABT) is a lucrative sector of the aquaculture industry. ABT is captured in the Mediterranean from the wild and transferred to floating offshore cages where it is fattened using whole bait-fish as feed (Vita and Marin, 2007). The uneaten feed fish that accumulate below the tuna cages are the main source of benthic pollution (e.g. Vita and Marin, 2007; Mangion et al., 2014). Several studies have addressed the impacts of tuna penning in the Mediterranean on benthic habitats in the vicinity of the fish cages (Matijević et al., 2006; Marin et al., 2007; Vita and Marin, 2007; Matijević et al., 2008; Vezzulli et al., 2008; Jahani et al., 2012; Moraitis et al., 2013; Kružić et al., 2014; Mangion et al., 2014; Dal Zotto et al., 2016). Since different levels of benthic impact are reported for different tuna farms (e.g. Jahani et al., 2012; Moraitis et al., 2013), it would be useful to adopt a design that includes multiple tuna farms and reference sites, as well as replicated sampling times, in studies on the benthic impacts of tuna penning, as this would help to better understand the impact of this activity on the marine environment. Polychaetes (e.g. Martinez-Garcia et al., 2013; Aguado-Giménez et al., 2015) and amphipods (e.g. Fernandez-Gonzalez and Sanchez-Jerez, 2011; Fernandez-Gonzalez et al., 2013) are good biological indicators of fish farming impacts on benthic habitats. The polychaete/amphipod (BOPA-Fish farming [BOPA-FF]) ratio (Aguado-Giménez et al., 2015) is a biotic index developed under the European Water Framework Directive (WFD, 2000/60/EC) that classifies water bodies into 'High', 'Good', 'Moderate', 'Poor' or 'Bad' Ecological Quality Status (EQS) classes (Dauvin and Ruellet, 2007).

Environmental monitoring of tuna penning in Malta was initiated in 2000 and is still ongoing. The present study was aimed at assessing the impact of the activity on benthic macrofauna in the vicinity of offshore tuna cages over a period of ten years, using polychaetes and amphipods as indicators. The following null hypothesis was tested: tuna penning activities do not have a significant influence on BOPA-FF (Aguado-Giménez et al., 2015) in the immediate vicinity of the cages, some 100 m away from the cages, and at two reference sites located 1 km and 2 km away. Three offshore tuna farms located in Malta in waters of circa 50 m depth, and differing in size, stocking density and feed management regime, were used in the present study.

## **Materials & Methods**

The three tuna farms considered in the present study are located circa 1 km off the northeastern to southeastern coast of Malta where the seabed consists of 'bare sand' habitat, and the water depth is between 42–53 m. The northernmost farm ('NEF') has eight cages and a maximum total annual capacity of around 2500 t, while the other two southeastern farms are smaller and have a maximum total annual capacity of around 1500 t each; one farm ('SEF 1') had three cages and the other ('SEF 2') had four cages. Tuna farming operations started in summer in 2001 at NEF and SEF 2, and in 2003 at SEF 1. The farming practice included an annual fallow period during the winter when the cages did not hold any tuna.

The sampling design consisted of four sampling plots which supported the same benthic habitat type at a similar water depth: (i) the 'Farm' plot, i.e. the seabed area below the tuna cages; (ii) 'Impact' plot, i.e. the seabed area located circa 100 m away from the cages; (iii) 'Control 1' plot, located circa 1 km away from the cages; and (iv) 'Control 2' plot, located circa 2 km away from the cages. This sampling design was replicated at each of the three farms. Four sampling sites were allotted to each of the NEF and SEF 2 plots, while three sampling sites were allotted to the SEF 1 plots, since the first two farms had a minimum of four tuna cages and the latter farm had three tuna cages. Samples were collected before initiation of tuna penning in Nov 2000 and Mar 2001 at NEF, in Oct 2002 at SEF 1, and in Jun 2001 at SEF 2; and thereafter on six-monthly or annual intervals, over a period of ten years. Three replicate grab samples for benthic macrofaunal studies were collected at each sampling site using a 0.1m<sup>2</sup> van Veen grab. The macrofaunal samples were sieved on a 0.5 mm mesh on board the vessel and afterwards temporarily preserved in 10 % seawater formalin. In the laboratory, the samples were sorted for polychaetes and amphipods after washing on a 0.5 mm mesh. Individuals were identified to the family level (see Karakassis and Hatziyanni, 2000; Olsford and Somerfield, 2000) and enumerated to obtain estimates of number of families and individuals per grab sample.

Data analyses were carried out separately for each farm, since sampling dates differed between the farms. Three-factor permutational analysis of variance (PERMANOVA) (Anderson, 2001) was run (with  $\alpha$  set at 0.05) on a Euclidean similarity matrix to test the hypothesis of no significant difference in BOPA-FF (as defined by Aguado-Giménez et al. [2015]) over time under the influence of tuna farming activities, based on a model with two fixed, orthogonal factors 'Date' (Da; 14 levels, Nov'00, Mar'01, Nov'01, Apr'02, Jan'03, Apr'03, Nov'03, Mar'04, Nov'04, Nov'05, Apr'06, Jun'07, May'08 and Apr'09, at NEF; 8 levels, Oct'02-'05 and Jun'06-'09, at SEF 1; 9 levels, Jun'01-'09, at SEF 2) and 'Plot' (Pl; 4 levels, Farm, Impact, Control 1 and Control 2), and a random factor 'Site' (Si; 4 levels, S1, S2, S3 and S4, at NEF and SEF 2; 3 levels, S1, S2 and S3, at SEF 1) nested within the 'Da x Pl' interaction. A total of 9999 unrestricted permutations of the raw data were carried out (Anderson, 2005). When the ANOVA term 'Da x Pl' was significant, pair-wise tests were carried out to investigate significant differences among groups (with  $\alpha$  set at 0.05). The analyses were implemented using PRIMER v.7.0.11 (Clarke and Gorley, 2006) with the PERMANOVA+ v.1.0 add-on package (Anderson et al., 2008).

## **Results**

PERMANOVA indicated significant difference in BOPA-FF for 'Da x Pl' ( $p < 0.001$ ) at NEF, SEF 1 and SEF 2 (Table 1). Pair-wise tests showed that BOPA-FF was significantly higher below the NEF tuna cages in Nov'01 and Nov'03 compared to Nov'00 and/or Mar'01, and subsequent sampling dates, and indicated 'Bad' EQS there (Fig. 1). BOPA-FF indicated 'High' or 'Good' EQS below the NEF tuna cages from Mar'04-Apr'09, and at NEF impacted and control plots during the study period; with the exception of Nov'05, when 'Moderate' EQS was detected below the cages. At SEF 1, BOPA-FF indicated 'Moderate'/'Poor' and 'Bad' EQS below tuna cages in Oct'03 and Oct'05,

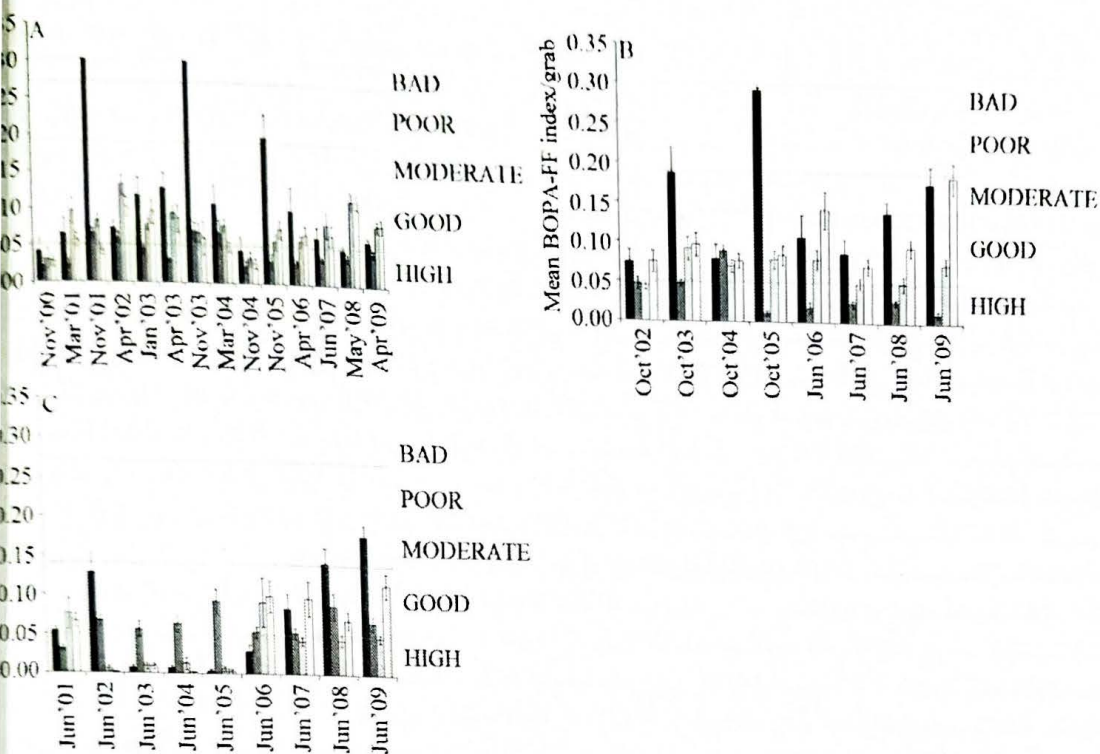


Fig. 1: Mean BOPA-FF per grab  $\pm$  SE recorded from farm (black bars), impacted (dark grey bars), 'Control 1' (light grey bars), and 'Control 2' (white bars) plots at NEF (A), SEF 1 (B) and SEF 2 (C).

Table 1: Results of the three-factor PERMANOVA ( $\alpha$  set at 0.05) for BOPA-FF recorded at NEF, SEF 1 and SEF 2. Degrees of freedom (df).

Source	NEF		SEF 1		SEF 2	
	df	p-value	df	p-value	df	p-value
Date = Da	13	0.0001	7	0.0001	8	0.0001
Plot = Pl	3	0.0001	3	0.0001	3	0.0005
Date x Plot	39	0.0001	21	0.0001	24	0.0001
Date (Da x Pl)	168	0.0001	64	0.0026	108	0.0001
Residual	448		192		288	
Total	671		287		431	

respectively, and was significantly higher there compared to Oct'02, Jun'06 and Jun'07, when EQS was 'Good', but not compared to May'08 and Jun'09, when EQS was 'Moderate', below tuna cages. BOPA-FF indicated 'Good' or 'High' EQS at the SEF 1 impacted and 'Control 1' plots from Oct'02 to Jun'09, but increased significantly at the SEF 1 'Control 2' plot from 'Good' (Oct'02-Oct'05, Jun'07 & Jun'08) to 'Moderate' (Jun'09) during the study period. At SEF 2, BOPA-FF indicated 'Good' or 'High' EQS at farm, impacted and control plots, except in Jun'08 and Jun'09 below the tuna cages, where it was significantly higher compared to Jun'02-Jun'07 and indicated 'Moderate' EQS (Fig. 1).

## Conclusions

The present study assessed data collected over a period of ten years from three different ABT farms; as far as we are aware, this is the first time that a study on the influence of tuna farming on benthic assemblages incorporates such a complete set of data collected for any region worldwide. Present results indicate that tuna farming activities influenced the 'bare sand' habitat at reference areas 1-2 km away from the cages, suggesting that particulate organic matter was transported there via sea currents, exceeding the distance for such transport previously reported at single tuna farms over shorter temporal scales (e.g. Marin et al., 2007; Vita and Marin, 2007) and for other Mediterranean farms such as those rearing seabass and seabream (e.g. Karakassis et al., 2000, 2002). Benthic impact is generally limited to the immediate vicinity of the cages (e.g. Karakassis et al., 2000; Tomassetti et al., 2016), but may extend from several meters to hundreds of kilometres from the farm (e.g. Edgar et al., 2010; Fernandez-Gonzalez et al., 2013). Fish farms located in offshore sites, such as those included in the present study, have a wider dispersion of particulate fish farm wastes due to stronger sea currents, compared to farms located in more sheltered waters (Kutti et al., 2007; Macleod et al., 2007), which result in a lower level of benthic impact (e.g. Maldonado et al., 2005; Vezzulli et al., 2008) but potentially wider spatial footprint of effects (Hall-Spencer et al., 2006). The oligotrophic nature of the Mediterranean Sea may also render the benthic ecosystem more sensitive to the introduction of fish farm wastes, such that a distance of 1 km (Porello et al., 2005) is not enough to achieve reference conditions at a control area in the vicinity of a fish farm.

Differences in the level and spatial extent of impact of tuna farming activities on the seabed were recorded between the three ABT farms in the present study. Studies at other Mediterranean tuna farms report low diversity of macroinvertebrate assemblages below fish cages (e.g. Marin et al., 2007; Vita and Marin, 2007; Mangion et al., 2014) and elevated values of the polychaete/amphipod (BOPA) ratio (Jahani et al., 2012), while other studies report no significant benthic influence of tuna farming due to the exposed location of the farms (Moraitis et al., 2013). The impact of fish farm wastes on the seabed depends on farm characteristics, such as the farmed species, the number of years of operation, the feed management regime and the total output of the farm, as well as exposure, bottom currents, water depth and sediment type (e.g. Borja et al., 2009; Tomassetti et al., 2009). The three ABT farms included in the present study differed in size, stocking density and feed management, as well as in their location. As a result, one would expect differences in the level and spatial extent of potential adverse environmental impacts among the three tuna farms. Present results indicated a higher level of adverse impact on the seabed in the vicinity of the tuna farm located off the northeastern coast of Malta (the largest in terms of holding capacity), compared to the other two farms sited off the southeastern coast. These results corroborate the expectation that benthic ecological quality will be lower at fish farm sites that have a higher total annual production (Borja et al., 2009).

For the same farm considered in the present study, i.e. the northeastern farm, Mangion et al. (2014) reported a significant increase in POCC and PONC, and an elevated abundance of capitellid polychaetes in the vicinity of the tuna cages, during

Mangion et al.  
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its first year of operation, which appeared to have resulted from the uneaten feed-fish accumulated on the seabed (Holmer et al., 2008). Some of this organic matter was dispersed up to 100 m away from the cages, resulting in a significant increase in POCC and PONC (Mangion et al., 2014). The observed changes in sediment quality (Mangion et al., 2014) and BOPA-FF - which indicated 'Bad' EQS - were conspicuous during autumn, towards the end of the tuna penning season. It would seem that following cessation of tuna farming activities in winter, the uneaten feed-fish accumulated on the seabed below the cages during the production period start to decompose, and only fish bones and other organic matter persist on the seabed (Mangion et al., 2014). Storms and bottom currents disperse this organic waste, and there is some recovery of the state of the sediment and associated macrobenthic assemblages (e.g. Marin et al., 2007; Vita and Marin, 2007; Mangion et al., 2014). Full recovery of the sediment following each production cycle is not necessary for sustainability, as long as the sediment recovers sufficiently from fish farming activities to withstand additional organic loading without accumulative changes (Macleod et al., 2007). In the present study, values of BOPA-FF increased significantly during the fallow periods to 'Good' EQS during the first years of operation, with no significant difference till the end of the study period. These observations indicate that the alternate use of production and fallow periods, together with an improved feed management regime at the northeastern farm, were sufficient to mitigate the benthic impact of tuna penning activities over several years of operation, resulting in a 'pulse' rather than 'press' disturbance.

The influence of tuna farming activities on the seabed below the cages of both southeastern farms was indicated by the significant increase in BOPA-FF. 'Moderate'/'Poor' EQS recorded at the SEF 1 farm area indicated that the influence of tuna farming on the benthic habitat in the vicinity of SEF 2, which retained 'Good' EQS following the first year of operation, was not as large. On the other hand, the spatial extent of the impact appeared to be largest at one of the farms located off the southeastern coast - SEF1; significant decrease in BOPA-FF to 'Moderate' EQS at the end of the study period, was recorded at the reference area located 2 km away from this farm. Furthermore, the temporal pattern of impact over several years of operation at the two southeastern farms indicated a 'press' disturbance resulting from the cumulative effects of nutrient enrichment from the two southeastern tuna farms. 'Moderate' and 'Poor' EQS were recorded at the two southeastern farms at the end of the study period, which may reflect an additive effect given that the farms are located relatively close to each other (1 km apart). The higher nutrient loading found in the coastal waters off the southern half of the Maltese Islands (Axiak et al., 2000) that result from more intense coastal use compared to the north of the islands (Mallia et al., 2002), may also be effecting the coastal waters where these tuna farms are located. These latter observations have implications for spatial planning of coastal aquaculture activities, particularly since many countries are moving toward establishing allocated zones for aquaculture (Sanchez-Jerez et al., 2016). Finally, the high spatio-temporal variation in the influence of tuna farming activities on benthic habitats highlights the importance of including multiple impacted and reference areas, and good temporal replication, in such environmental impact monitoring studies.

## References

- Aguado-Giménez, F., Gairin J.I., Martínez-García E., Fernández-González, V., Ballester Moltó, M., Cerezo-Valverde, J. and Sánchez-Jerez, P. (2015), "Application of "taxocene surrogation" and "taxonomic sufficiency" concepts to fish farming environmental monitoring. Comparison of BOPA index versus polychaete assemblage structure", *Marine Environmental Research*, 103, 27-35.
- Anderson, M.J. (2001), "A new method for nonparametric multivariate analysis of variance", *Austral Ecology*, 26 (1), 32-46.
- Anderson, M.J. (2005), "*PERMANOVA: a FORTRAN computer program for permutational multivariate analysis of variance*", Department of Statistics, University of Auckland, New Zealand, 24 p.
- Anderson, M.J., Gorley, R.N. and Clarke, K.R. (2008), "*PERMANOVA+ for PRIMER: guide to software and statistical methods*", PRIMER-E, Plymouth, United Kingdom, 214 p.
- Axiak, V., Pavlakis, P., Sieber, A.J. and Tarch, D. (2000), "Re-assessing the extent of impact of Malta's (Central Mediterranean) major sewage outfall using ERS SAR", *Marine Pollution Bulletin*, 40 (9), 734-738.
- Borja, Á., Dauer, D.M., Elliott, M. and Simenstad, C.A. (2010), "Medium- and Long-term Recovery of Estuarine and Coastal Ecosystems: Patterns, Rates and Restoration Effectiveness", *Estuaries and Coasts*, 33 (6), 1249-1260.
- Borja, Á., Rodríguez, J.G., Black, K., Bodoy, A., Emblow, C., Fernandes, T.F., Forte, J., Karakassis, I., Muxika, I., Nickell, T.D., Papageorgiou, N., Pranovi, F., Sevastou, K., Tomassetti, P. and Angel, D. (2009), "Assessing the suitability of a range of benthic indices in the evaluation of environmental impact of fin and shellfish aquaculture located in sites across Europe", *Aquaculture*, 293 (3), 231-240.
- Clarke, K.R. and Gorley, R.N. (2006), "*PRIMER v6: User manual/tutorial*", PRIMER-E, Plymouth, United Kingdom, 190 p.
- Dal Zotto, M., Santulli, A., Simonini, R. and Todaro, M. A. (2016), "Organic enrichment effects on a marine meiofauna community, with focus on Kinorhyncha", *Zoologischer Anzeiger-A Journal of Comparative Zoology*, 265, 127-140.
- Dauvin, J.C. and Ruellet, T. (2007), "Polychaete/amphipod ratio revisited", *Marine Pollution Bulletin*, 55 (1), 215-224.
- Edgar, G.J., Davey, A. and Shepherd, C. (2010), "Application of biotic and abiotic indicators for detecting benthic impacts of marine salmonid farming among coastal regions of Tasmania", *Aquaculture*, 307 (3), 212-218.



- Fernandez-Gonzalez, V., Aguado-Giménez, F., Gairin, J.I. and Sanchez-Jerez, P. (2013), "Exploring patterns of variation in amphipod assemblages at multiple spatial scales: natural variability versus coastal aquaculture effect", *Aquaculture Environment Interactions*, 3, 93-105.
- Fernandez-Gonzalez, V. and Sanchez-Jerez, P. (2011), "Effects of seabass and seabream farming (Western Mediterranean Sea) on peracarid crustacean assemblages", *Animal Biodiversity and Conservation*, 34 (1), 179-190.
- Giles, H. (2008), "Using Bayesian networks to examine consistent trends in fish farm benthic impact studies", *Aquaculture*, 274 (2), 181-195.
- Hall-Spencer, J., White, N., Gillespie, E., Gillham, K. and Foggo, A. (2006), "Impact of fish farms on maerl beds in strongly tidal areas", *Marine Ecology Progress Series*, 326, 1-9.
- Hargrave, B.T., Phillips, G.A., Doucette, L.I., White, M.J., Milligan, T.G., Wildish, D.J. and Cranston, R.E. (1997), "Assessing benthic impacts of organic enrichment from marine aquaculture", *Water, Air, & Soil Pollution*, 99 (1), 641-650.
- Holmer, M., Hansen, P.K., Karakassis, I., Borg, J.A. and Schembri, P.J. (2008), "Monitoring of environmental impacts of marine aquaculture", In: Holmer, M., Black, K., Duarte, C.M., Marba, N. and Karakassis, I. (Eds.), *Aquaculture in the Ecosystem*, Springer, The Netherlands, 47-85.
- Jahani, N., Nabavi, S.N.B., Dehghan Madiseh, S., Mortezaie, S.R.S. and Fazeli, N. (2012), "The effect of marine fish cage culture on benthic communities using BOPA index in Ghazale Creek", *Iranian Journal of Fisheries Science*, 11 (1), 78-88.
- Karakassis, I. and Hatziyanni, E. (2000), "Benthic disturbance due to fish farming analyzed under different levels of taxonomic resolution", *Marine Ecology Progress Series*, 203, 247-253.
- Karakassis, I., Tsapakis, M., Hatziyanni, E., Papadopoulou, K.N. and Plaiti, W. (2000), "Impact of cage farming of fish on the seabed in three Mediterranean coastal areas", *ICES Journal in Marine Science*, 57 (5), 1462-1471.
- Karakassis, I., Tsapakis, M., Smith, C.J. and Rumohr, H. (2002), "Fish farming impacts in the Mediterranean studied through sediment profiling imagery", *Marine Ecology Progress Series*, 227, 125-133.
- Kružić, P., Vojvodić, V. and Bura-Nakić, E. (2014), "Inshore capture-based tuna aquaculture impact on *Posidonia oceanica* meadows in the eastern part of the Adriatic Sea", *Marine Pollution Bulletin*, 86 (1), 174-185.

- Kutti, T., Ervik, A. and Hansen, P.K. (2007), "Effects of organic effluents from a salmon farm on a fjord system. I. Vertical export and dispersal processes", *Aquaculture*, 262 (2), 367-381.
- Macleod, C.K., Moltschaniwskyj, N.A. and Crawford, C.M. (2006), "Evaluation of short-term fallowing as a strategy for the management of recurring organic enrichment under salmon cages", *Marine Pollution Bulletin*, 52 (11), 1458-1466.
- Macleod, C.K., Moltschaniwskyj, N.A., Crawford, C.M. and Forbes, S.E. (2007), "Biological recovery from organic enrichment: some systems cope better than others", *Marine Ecology Progress Series*, 342, 41-53.
- Maldonado, M., Carmona, M.C., Echeverría, Y. and Riesgo, A. (2005), "The environmental impact of Mediterranean cage fish farms at semi-exposed locations: does it need a re-assessment?", *Helgoland Marine Research*, 59 (2), 121-135.
- Mallia, A., Briguglio, M., Ellul, A.E. and Formosa, S. (2002), "Physical background, demography, tourism, mineral resources and land-use", In: Axiak, V., Gauci, V., Mallia, A., Mallia, E., Schembri, P.J., Vella, A.J. and Vella, L. (Eds.), *State of the environment report for Malta 2002*, Ministry for Home Affairs and the Environment, Malta, 42-161.
- Mangion, M., Borg, J.A., Thompson, R. and Schembri, P.J. (2014), "Influence of tuna penning activities on soft bottom macrobenthic assemblages", *Marine Pollution Bulletin*, 79 (1), 164-174.
- Marin, A., Montoya, S., Vita, R., Marín-Guirao, L., Lloret, J. and Aguado, F. (2007), "Utility of sea urchin embryo-larval bioassays for assessing the environmental impact of marine fishcage farming", *Aquaculture*, 271 (1), 286-297.
- Martinez-Garcia, E., Sanchez-Jerez, P., Aguado-Giménez, F., Avila, P., Guerrero, A., Sánchez-Lizaso, J.L., Fernandez-Gonzalez, V., González, N., Gairin, J.L., Carballeira, C., García-García, B., Carreras, J., Macías, J.C., Carballeira, A. and Collado, C. (2013), "A meta-analysis approach to the effects of fish farming on soft-bottom polychaeta assemblages in temperate regions", *Marine Pollution Bulletin*, 69 (1), 165-171.
- Matijević, S., Kušpilić, G. and Barić, A. (2006), "Impact of a fish farm on physical and chemical properties of sediment and water column in the middle Adriatic sea", *Fresenius Environmental Bulletin*, 15 (9), 1058-1063.
- Matijević, S., Kušpilić, G., Kljaković-Gašpić, Z. and Bogner, D. (2008), "Impact of fish farming on the distribution of phosphorus in sediments in the middle Adriatic area", *Marine Pollution Bulletin*, 56 (3), 535-548.

- Moraitis, M., Papageorgiou, N., Dimitriou, P.D., Petrou, A. and Karakassis, I. (2013), "Effects of offshore tuna farming on benthic assemblages in the Eastern Mediterranean", *Aquaculture Environment Interactions*, 4 (1), 41-51.
- Olsgard, F. and Somerfield, P.J. (2000), "Surrogates in marine benthic investigations: which taxonomic unit to target?", *Journal of Aquatic Ecosystem Stress and Recovery*, 7 (1), 25-42.
- Pereira, P.M.F., Black, K.D., Mclusky, D.S. and Nickell, T.D. (2004), "Recovery of sediments after cessation of marine fish farm production", *Aquaculture*, 235 (1), 315-330.
- Porrello, S., Tomassetti, P., Manzueto, L., Finoia, M.G., Persia, E., Mercatali, I. and Stipa, P. (2005), "The influence of marine cages on sediment chemistry in the Western Mediterranean Sea", *Aquaculture*, 245 (1), 145-158.
- Sanchez-Jerez, P., Karakassis, I., Massa, F., Fezzardi, D., Aguilar-Manjarrez, J., Soto, D., Chapela, R., Avila, P., Macias, J.C., Tomassetti, P., Marino, G., Borg, J.A., Franičević, V., Yucel-Gier, G., Fleming, I.A., Biao, X., Nhhala, H., Hamza, H., Forcada, A. and Dempster, T. (2016), "Aquaculture's struggle for space: the need for coastal spatial planning and the potential benefits of Allocated Zones for Aquaculture (AZAs) to avoid conflict and promote sustainability", *Aquaculture Environment Interactions*, 8, 41-54.
- Tomassetti, P., Gennaro, P., Lattanzi, L., Mercatali, I., Persia, E., Vani, D. and Porrello, S. (2016), "Benthic community response to sediment organic enrichment by Mediterranean fish farms: Case studies", *Aquaculture*, 450, 262-272.
- Tomassetti, P., Persia, E., Mercatali, I., Vani, D., Marusso, V. and Porrello, S. (2009), "Effects of mariculture on macrobenthic assemblages in a western Mediterranean site", *Marine Pollution Bulletin*, 58 (4), 533-541.
- Vezzulli, L., Moreno, M., Marin, V., Pezzati, E., Bartoli, M. and Fabiano, M. (2008), "Organic waste impact of capture-based Atlantic bluefin tuna aquaculture at an exposed site in the Mediterranean Sea", *Estuarine, Coastal and Shelf Science*, 78 (2), 369-384.
- Vita, R. and Marin, A. (2007), "Environmental impact of capturebased bluefin tuna aquaculture on benthic communities in the western Mediterranean", *Aquaculture Research*, 38 (4), 331-339.
- Wu, R.S.S. (1995), "The environmental impact of marine fish culture: towards a sustainable future", *Marine Pollution Bulletin*, 31 (4), 159-166.