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Are Indian Ocean tuna populations assessed and managed at the appropriate spatial scale? A brief review of the evidence and implications

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Abstract

Tuna species managed under the auspices of the Indian Ocean Tuna Commission have generally been assumed (explicitly or implicitly) to be highly mobile and consist of a single panmictic spawning population for the purposes of stock assessment and management. In this paper, we: i) briefly review evidence that questions this assumption (largely based on recent population genetics and tagging studies), ii) qualitatively discuss the implications of violating this assumption, and iii) outline some elements of a collaborative research plan to resolve these issues and mitigate the consequences of getting this assumption wrong. This paper is intended to stimulate discussion within the IOTC scientific community about the potential importance of population structure within the Indian Ocean and extent to which it should be considered a research and management priority.

1. Introduction

Each of the tuna species managed under the auspices of the IOTC is generally assumed to be highly migratory, with a single, highly mixed spawning population. This is the most parsimonious assumption that greatly simplifies stock assessment modelling, data collection and management. However, as early as the 1950s, the possibility of intra-specific population structure has been recognized. e.g. Kurogane and Hiyama (1958, and references therein) noted morphometric differences among yellowfin tuna sampled from longliners across the equatorial Indian Ocean. It was recognized that the observed differences (generally smaller sizes to the east) were consistent with a serial depletion effect, as the fleet expanded westward through the 1950s (CPUE was also observed to sequentially decline from east to west). They concluded that they could not distinguish whether there were discrete populations in the east and west or a mixture. The population structure of the Indian Ocean tunas remains poorly quantified half a century later. If the stock assessment advice is not based on appropriate structural assumptions, management may fail to achieve objectives related to conservation and optimal economic use of the resource. Specifically, if there are distinct spawning populations (or mixing rates are very slow across a geographically broad

panmictic population), some populations (or sub-regions) could be locally over-exploited and management measures might be directed toward the wrong populations.

Attempts have been made to partition some IOTC stock assessments into sub-populations. e.g. For several years, yellowfin tuna have been assessed (e.g. Langley et al. 2012) with a spatially-disaggregated assessment (shared spawning population, but region-specific recruitment deviations and migration among regions). The most recent Indian Ocean swordfish assessments have included a speculative scenario in which a south-west population was assessed independently of the remainder of the ocean, to admit the possibility that this might represent a reproductively isolated, highly depleted, population (e.g. IOTC 2009). The stock status concerns in this sub-region provided some of the impetus for an ambitious swordfish genetic stock structure project (Muths et al. 2012), however, initiatives of a similar scale have not been undertaken for the tuna species.

Due to the convergence of a number of factors, it seems timely for a concerted effort to resolve the tuna stock structure uncertainties in the Indian (and other oceans):

- For the main commercial tuna species, exploitation rates have increased dramatically over the last two decades, largely due to the increasing purse seine activities
- A number of small scale genetics projects (discussed below) have provided indications of population structure within surprisingly small regions.
- The recent Regional Tuna Tagging Progamme (RTTP-IO) provides new insights into tuna movement (discussed below).
- Rapid developments in high throughput, next generation sequencing technologies has greatly increased the power, and reduced the cost, of genetic analyses relevant to questions of population structure.
- Coastal and small island nations are actively attempting to take an active role in the management of the localized tuna populations on which they depend, and
- Market forces are demanding sustainable seafood products, which require assessment and management on appropriate scales.

In this paper, we briefly review some of the evidence that questions the panmictic spawning population assumption for the main tropical tuna species (yellowfin, bigeye and skipjack), discuss the implications of assessing and managing stocks with incorrect population structure assumptions, and propose a collaborative research plan to address these concerns.

2. Population structure inferences from population genetics

We are aware of five Indian Ocean genetic studies suggesting that the main commercial tuna species have a more fragmented population structure than typically assumed in the assessment and management process (Figure 1). These are briefly summarized:

1. Dammannagoda et al. (2008) examined yellowfin mitochondrial DNA (mtDNA) sequences and 3 nuclear DNA microsatellites from 6 locations around Sri Lanka, plus one from the Maldives. Spatial differentiation (and temporal stability) were observed for mtDNA and spatial AMOVA suggested that Sri Lankan samples from the north-west and south-east were distinct from each other and all other sites. It was noted that the NW and SE samples

consisted of larger fish (longline-caught) than the others, and hence were more likely to include individuals that had dispersed further from their spawning grounds. Evidence for spatial differentiation from the microsatellites was much weaker.

2. Swaraj et al. (2013) used yellowfin D-loop mtDNA sequences to compare 7 sample locations in Indian waters. Spatial AMOVA suggested that the north-western (Verava) and offshore western (Lakshadweep Islands) locations were distinct from each other and all other locations (from southern India and the Bay of Bengal).

3. Dammannagoda et al. (2011) used a single mtDNA gene and 6 nuclear DNA microsatellites to examine skipjack population structure from 7 sample locations (5 around Sri Lanka, 1 from Maldives and 1 from Lakshadweep). Phylogenetic reconstruction from mtDNA revealed 2 coexisting, divergent clades, with both clades present at all sample locations. AMOVA revealed significant differences among locations (stable across years), with Spatial AMOVA partitioning the mtDNA locations into 3 populations (Maldives, eastern Sri Lanka, and all others). Evidence for population differentiation was also found using microsatellite DNA markers, however, the relationships among the populations were not consistent between the mtDNA and microsatellite marker sets. The authors concluded that different evolutionary/demographic processes were responsible for the differentiation of the two marker types, and suggested that the fisheries around Sri Lanka probably represent a mixture of spawning populations converging on shared feeding grounds.

4. Menezes et al. (2012) examined skipjack D-loop mtDNA sequences from 6 locations in Indian waters (including Lakshadweep and Andaman Islands). Phylogenetic analysis indicated four haplotype clades without an obvious geographic pattern. Spatial AMOVA estimated 4 genetically-differentiated populations consisting of the 2 Arabian Sea locations (Veraval and Kakshadweep), Kochi (west coast of southern India), PondiCherry (east coast of southern India), and 2 locations further east in the Bay of Bengal (Vishakhapatnam and the Andaman Islands).

5. Nugraha et al. (2011) identified differences between bigeye sampled (by longline observers) from west of Sumatra and south of Java, on the basis of Restriction Fragment Length Polymorphisms (RFLPs) on D-loop mtDNA.

Several other studies (many cited in the references above, e.g. Ely et al. 2005, Ward et al. 1997) have either not been able to identify genetic differences in tuna populations, or found differences at much larger scales (e.g. between oceans or across vast distances within the Pacific). In general, we would expect that genetic population structure studies would be more susceptible to false negative results (failure to identify structure that really exists) than false positives (identification of population structure when it does not exist). e.g. Populations may not have been reproductively isolated for long enough to develop distinct genetic differences, or the appropriate genetic markers might not be identified, or sample sizes may be too small to be conclusive. Given that these results show more population structure than has been generally assumed, it is worth considering the mechanisms that might cause false positives:

- There could be a fundamental error in species identification in some cases. Juvenile yellowfin and bigeye tuna can be difficult to distinguish if the fish are not fresh (e.g. Grewe and Hampton 1998). We cannot comment on the taxonomic skill of the fish samplers in these studies, however, it seems unlikely that this problem would affect all 3 of the bigeye/yellowfin studies. Furthermore, there is no equivalent species identification problem that can easily explain the skipjack studies.

- The origin of the fish samples might not be correct. Long range fleets or transhipment might introduce samples to local markets from distant waters. This might exaggerate the perception of population structure in some cases, but given the sampling descriptions, it seems unlikely that this would be a consistent problem or reflect a contamination of samples from other oceans.

- False positives could reflect publication bias (e.g. Goldacre 2008). Statistically significant false positive results occur by chance, and positive studies are far more likely to be published than negative studies. However, given the large number of highly significant results, and the limited number of researchers active in this field, this mechanism seems unlikely.

The high degree of differentiation reported in these small geographical scale studies is intriguing, and may have wide-ranging implications for local management of tropical tunas (not only in the Indian Ocean). Given that similar DNA investigations examining population differentiation at much broader geographical scales have not previously revealed such a high degree of differentiation for any highly migratory pelagic fish species (as far as we know), these results merit careful consideration. As we now have access to more powerful and cost effective genetic tools (e.g. next generation sequencing technologies), it should be a priority to revisit these studies, including markers for validated species identification (e.g. Vinas and Tudela, 2009), as a first step toward resolving the potentially complicated pattern of stock structure in this area.

3. Population structure inferences from tagging studies

Tagging studies provide different information from genetic analyses, and the two lines of evidence should be combined (ideally along with additional information from morphometrics, otolith microchemistry, parasitology, etc.) to provide the best synthesis of population structure and movement. Conventional tags might not be very useful for discriminating spawning populations, because fish must be of a minimum size before they can be tagged, and hence they could move a long distance from natal regions before being tagged. Furthermore, recovery locations depend on the presence of fishing effort, and accurate reporting of recovered tags, and rarely include observations about spawning condition. However, tags can reveal important dynamics that may not be evident from genetics. A very low continuous genetic exchange rate (or high but intermittent exchange rate) may be sufficient to prevent any genetic population distinction from developing. But from the perspective of fisheries management, population dynamics of the different sub-populations may be sufficiently independent to warrant spatially-structured management.

The recent Regional Tuna Tagging Programme (RTTP-IO) clearly demonstrated that there are long distance movements of yellowfin, skipjack and bigeye tunas within the Indian Ocean (e.g. Figure 2, and Hallier and Million 2009). Qualitatively, this is consistent with the notion that the Indian Ocean basin probably supports a single well-mixed population for each species. However, there are a number of contributing factors which do not necessarily mean that the tagging and genetic studies above are incompatible. The RTTP-IO focussed on the western Indian Ocean, with the vast majority of tags released near east Africa and recovered in the equatorial purse seine fishery (plus substantial numbers in the Maldivian pole and line fishery). Very few tags were returned from the eastern Indian Ocean. It is unclear the extent to which this reflects a lack of movement from west to east, and the extent to which this reflects poor reporting rates for recovered tags. To date, movement dynamics within the eastern Indian Ocean have not been adequately observed. And while there is evidence of large-scale movement, tags also demonstrate that mixing is not rapid and uniform:

- Langley and Million (2012) qualitatively demonstrated that tagged yellowfin show evidence of incomplete mixing a year after release within the western Indian Ocean.
- Kolody and Hoyle (2013) demonstrated that tags of all three tropical tuna species do not mix within spatially-disaggregated assessment regions at the expected rate in the Western Pacific. The same analysis applied to the Indian Ocean was less clear, however, it was evident that tuna do not mix at uniform rates across the western Indian Ocean, with evidence for different rates of tuna retention between the open waters of the western Indian Ocean and the archipelagic waters around the Maldives (e.g. reproduced in Figure 3).
- Tropical tuna tag displacements in the Pacific have been shown to be of relatively small magnitude on average, despite a number of very large displacements (e.g. Figure 4, Sibert and Hampton 2003, Hampton and Gunn 1998, Schaefer and Fuller 2005).
- Ianelli et al. (2012) demonstrated that western Pacific bigeye abundance (and stock status) estimates are very sensitive to the exclusion or inclusion of tagging data from the numerically minor SW Pacific region. It appears that there is some fundamental incompatibility between the spatial/temporal structure assumed in the assessment (and other data sources) and the dynamics inferred from tags.

This is not a comprehensive review of tagging studies, but sufficient to demonstrate that tuna movements are probably of a lower magnitude and more structured nature than generally assumed in tuna RFMOs. However, the large-scale tag movements that do occur would probably be sufficient to obscure genetic evidence of population structure within the (western) Indian Ocean, if there is no fidelity to natal spawning grounds. However, it is conceivable that there is spawning fidelity to natal regions, while foraging grounds are shared by multiple populations, in which case the possibility of multiple genetically distinct populations within the Indian Ocean cannot be easily dismissed.

4. Implications of Population structure for stock assessment and management

There are a number of ways in which tuna populations could be structured, and the implications for management could be very different. We briefly discuss four possibilities below:

- Rapidly mixing panmictic population. This represents the least risky management situation, and is the default assumption in the spatially aggregated assessments. If the stock is

assessed and managed effectively as a whole unit, then growth and recruitment overfishing should, on average, be avoided.

- A spatially structured population with a common stock-recruitment process. This is the default assumption in the spatially-disaggregated Multifan-CL assessments (e.g. Langley et al. 2012). If the global spawning population is assessed and managed effectively, recruitment over-fishing should not occur. However, appropriate sub-regions and their connections need to be assessed and managed to avoid growth over-fishing. The shared stock-recruitment assumption is a parsimonious tool of convenience. It is unlikely anyone believes that spawning in the south Java Sea is going to have a strong and immediate effect on recruitment 7000km away in the Arabian Sea, but since this model estimates independent recruitment deviations by region, and there is little observational evidence for declining recruitment with decreasing spawning biomass for tuna populations (aside from the bluefins), this assumption may not be numerically unreasonable. This model also assumes that other biological characteristics are constant among regions (e.g growth, mortality and maturity, which may not be the case).

- Spatially-segregated populations. This represents a reasonably straightforward assessment and management situation. Provided that there are adequate data, each population can be assessed and managed as an independent unit.

- Spatially-segregated spawning populations with spawning site fidelity but shared foraging grounds and vulnerability in mixed stock fisheries. This potentially represents a more difficult management situation, because mixed-stock fisheries catch composition needs to be monitored to reliably assess the individual populations. Aside from the additional monitoring requirements, management issues are similar to mixed-species fisheries, i.e. the needs of high and low productivity stocks need to be considered simultaneously.

It is of course possible to define other potential complications, e.g. sex-specific philopatry (such as might lead to the reported divergences observed between genetic inferences from mtDNA and nuclear DNA), large but intermittent gene exchanges, or spasmodic sub-population formation due to irregular recruitment events driven by environmental fluctuations.

The major risk of ignoring the tuna population structure is the over-exploitation of some populations (most likely the smaller or less productive stocks). This could potentially result in serious food security problems and/or economic hardship, particularly for island/coastal nations with a strong tuna dependency and short-range fishing fleets. However, there may also be lost economic opportunities resulting from not selectively targeting the more productive, or lightly-exploited populations.

The increasing consumer demand for certified-sustainable products offers an additional motive for resolving these stock-structure uncertainties. Regardless of whether the population structure ultimately has disruptive implications for tuna management, the certification process may expect this issue to be resolved, and it is consistent with the precautionary approach to responsible fisheries management.

5. Proposed tuna population structure research and related projects

We (CSIRO Marine and Atmospheric Research, plus collaborators to be determined) are currently seeking to establish a broad-ranging initiative to improve the assessment and management of highly migratory tuna stocks, to take advantage of the high throughput, low cost, genetic technologies that are rapidly emerging. This involves linking together various inter-related studies, including:

- Stock structure studies of bigeye and yellowfin tunas in the Indonesian archipelago and adjacent populations in the Indian and Pacific Oceans. This work is currently underway with Indonesian collaborators (Proctor et al 2012). Understanding the tuna dynamics in this region is important for domestic management and understanding the link between Indian and Pacific Ocean populations.
- Expand tropical stock structure studies throughout the Indian Ocean, including temperate and neritic tuna species as opportunity allows. Attachment A is a draft concept proposal for this body of work, including a (probably incomplete) list of potential collaborators.
- Simulation study to understand the implications of complicated population stock structure and migration dynamics on assessment and management. Just because realistic dynamics are more complicated than we have traditionally assumed (or been able to quantify with the available data) does not mean that simple management is necessarily ineffective. We want to identify potentially risky situations, quantify the likely implications of getting the assumptions wrong, and explore the assessment data requirements and/or robust management strategies that would mitigate potential problems. This relates to the Management Strategy Evaluation initiatives overseen by the IOTC Methods Working Group.
- The issues are very similar for most tuna species and oceans, and this work has natural extensions into the Pacific and Atlantic Oceans.
- Exploration of genetic tagging methods for tuna. A mark-recapture study based on close-kin relationships has proven successful for southern bluefin tuna (Bravington et al. 2012), and we are examining marker development and scaling costs for similar applications to other populations. There are certain synergies to be realized by pursuing these sorts of studies in parallel with other genetic analyses.

We encourage feedback on the scope and methods of this ambitious research program, and seek expressions of interest from potential collaborators that are already active in this field, to minimize duplication of effort and realize synergies (e.g. in obtaining and analysing samples, developing genetic markers, participate in simulation studies, etc.).

6. References

Bravington, M., Grewe, P., and Davies, C., 2012. Report of the close-kin project: estimating the absolute spawning stock size of SBT using genetics. Commission for the Conservation of Southern Bluefin Tuna working paper CCSBT-ESC/1208/19.

Dammannagoda, S.T., Hurwood, D.A., and Mather, P.B. 2011. Genetic analysis reveals two stocks of skipjack tuna (*Katsuwonas pelamis*) in the northwestern Indian Ocean. Can. J. Fish. Aquat. Sci. 68: 210-223.

Dammannagoda, S.T., Hurwood, D.A., and Mather, P.B. 2008. Evidence for fine geographical scale heterogeneity in gene frequencies in yellowfin tuna (*Thunnus albacares*) from the north Indian Ocean around Sri Lanka. Fish. Res. 90: 147-157.

Ely, B., Vinas, J., Alvaredo-Bremer, J.R., Black, D., Lucas, L. Covello, K., Labrie, A.V., and Thelen, E. 2005.

Goldacre, B. 2008. Bad Science. Published by Fourth Estate. 338 p.

Grewe, P.M., and Hampton, J. 1998. An assessment of the bigeye (*Thunnus obesus*) population structure in the Pacific Ocean, based on mitochondrial DNA and DNA microsatellite analysis. Joint Institute for Marine and Atmospheric Research contribution 98-320. 1000 Pope Rd, Honolulu, HI, 96822. 29 p.

Hallier, J.P. and Million, M. The contribution of the regional tuna tagging project – Indian Ocean to Indian Ocean stock assessment. IOTC-2009-WPTT-24.

Hampton, J., and Gunn, J., 1998. Exploitation and movements of yellowfin tuna (*Thunnus albacares*) and bigeye tuna (*T. obesus*) tagged in the north-western Coral Sea. Mar. Freshwater Res., 49, 475–89.

Ianelli, J., Maunder, M. and Punt, A.E. 2012. Independent review of 2011 WCPO bigeye tuna assessment. WCPFC-SC8-2012/SA-WP-01.

IOTC. 2011. Report of the Ninth Session of the IOTC Working Party on Billfish, Victoria, Seychelles.

Kolody, D., Herrera, M., and Million, J. 2010. Exploration of Indian Ocean Bigeye Tuna Stock Assessment Sensitivities 1952-2008 using Stock Synthesis (Updated to include 2009). IOTC-2010-WPTT-04 (Rev1).

Kolody, D., and Hoyle S. 2013. Evaluation of tag mixing assumptions for skipjack, yellowfin and bigeye tuna stock assessments in the western Pacific and Indian Oceans. WCPFC Working Paper: WCPFC-SC9-2013/SA-IP-11

Kurogane, K., and Hiyame, Y. 1958. Morphometric comparisons of the yellowfin tuna from the six grounds in the Indian Ocean. Bull. Fish. Soc. Sci. Fish. 24, 487-494.

Langley, A., Million, J., 2012. Determining an appropriate tag mixing period for the Indian Ocean yellowfin tuna stock assessment. Indian Ocean Tuna Commission 14th Working Party on Tropical Tuna Working paper IOTC-2012-WPTT14-31.

Langley, A., Herrera, M., and Million, J. 2012. Stock assessment of yellowfin tuna in the Indian Ocean using MULTIFAN-CL. IOTC–2012–WPTT14–38 Rev_1

Menezes, M.R., Kumar, G., and Kanal, S.P. 2012. Population genetic structure of skipjack tuna *Katsuwonas pelamis* from the Indian coast using sequence analysis of the mitochondrial DNA D-loop region. J. Fish. Biol. 80: 2198-2212.

Muths D., Le Couls S., Evano H., Grewe P., and Bourjea J. Microsatellite and mtDNA markers were unable to reveal genetic population structure of swordfish (Xiphias gladius) in the Indian Ocean. IOTC–2012–WPB10–15

Nugraha, B., Novianto, D., and Barata, A. 2011. Keragaman genetik ikan tuna matabesar (*Thunnus obesus*) di Samudera Hindia. [*Genetic diversity of bigeye tuna (Thunnus obesus) in Indian Ocean*]. Ind. Fish. Res. J. Vol.17 (4), 285 - 292 (In Bahasa Indonesia, with English abstract).

Proctor, C. 2009. Developing research capacity for management of Indonesia's pelagic fisheries resources. ACIAR project proposal.

Schaefer, K.M., Fuller, D.W., 2005. Conventional and archival tagging of bigeye tuna (*Thunnus obesus*) in the eastern equatorial Pacific ocean. Col. Vol. Sci. Pap. ICCAT, 57(2), 67-84.

Sibert, J., Hampton,J., 2003. Mobility of tropical tunas and the implications for fisheries management. Mar. Pol. 27, 87-95.

Swaraj, P.K., Kumar, G., M.R. Menezes and R.M. Meena. 2013. Mitochondrial DNA analysis reveals three stocks of yellowfin tuna *Thunnus albacares* (Bonnaterre, 1788) in Indian waters. Conserv. Genet 14: 205-213.

Vinas, J., and Tudela, S. 2009. A validated method for genetic identification of tuna species (Genus *Thunnus*). PLoS ONE 4(10): e7606.doi:10.1371/journal.pone.0007606.

Ward, R.D., Elliott, N.G., Innes, B.H., Smolenski, A.J. and Grewe, P.M. 1997. Global population structure of yellowfin tuna (*Thunnus albacares*) inferred from allozyme and mitochondrial DNA variation. *Fishery Bulletin US*, 95, 566-575.

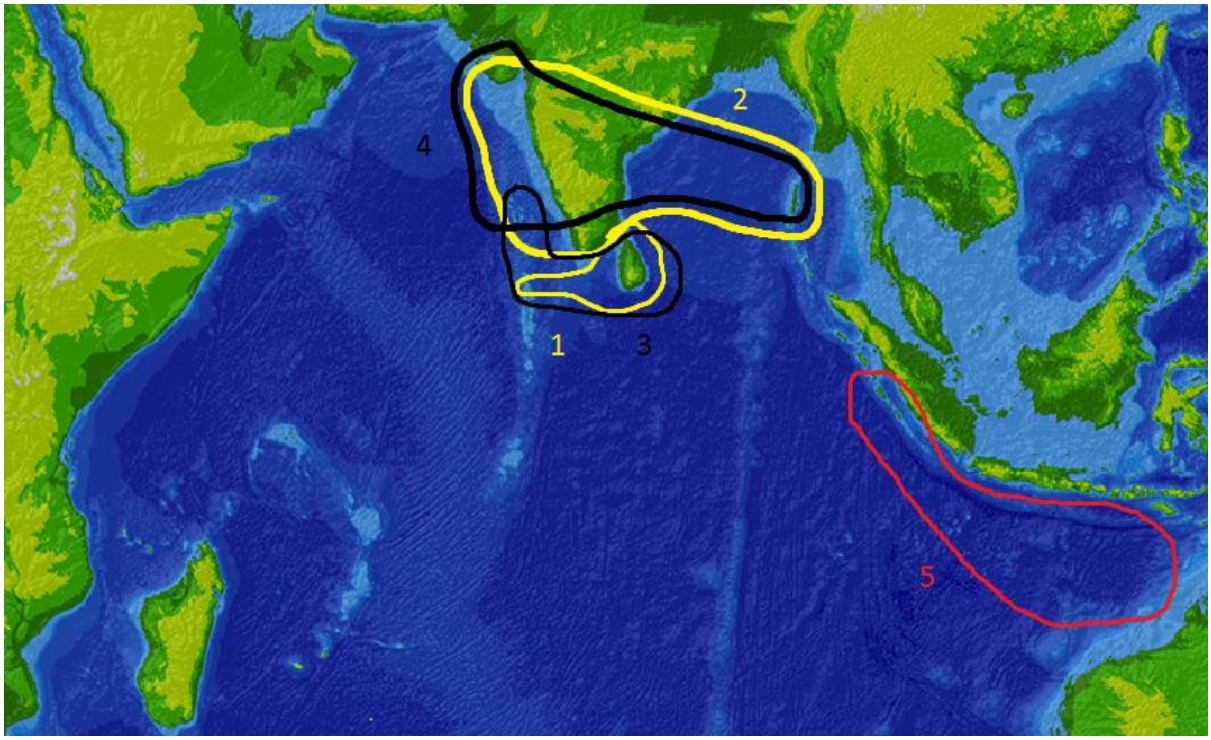

Figure 1. Genetic evidence for structured tuna populations has been reported for studies within each of the regions approximately outlined above (Numbers indicate each of the 5 studies cited in section 2). SKJ = black, YFT = yellow, BET = red. (Base map obtained under Wikipedia commons license)

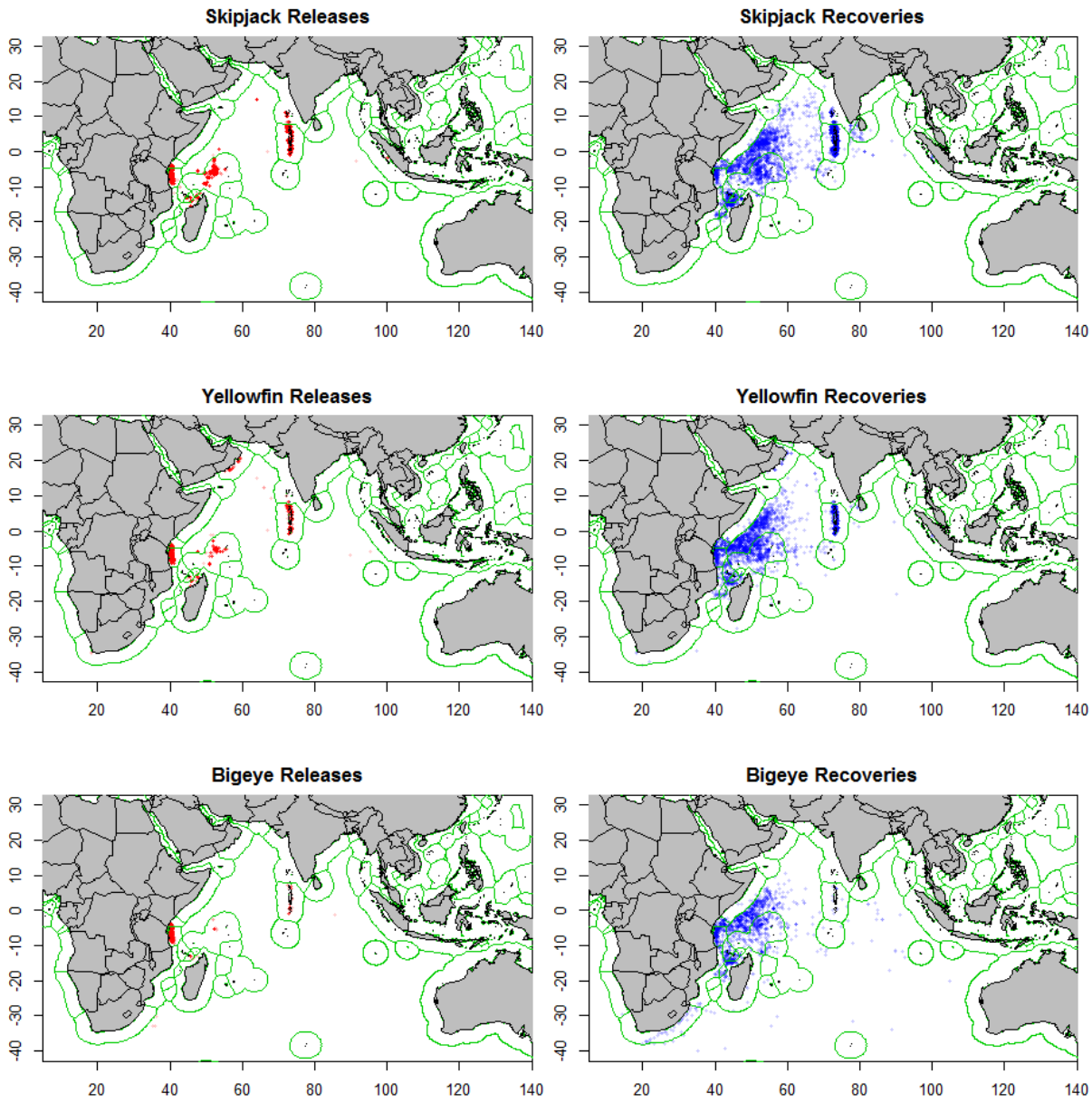

Figure 2. Tag release and reported recovery locations for the RTTP-IO and small-scale tagging programmes.

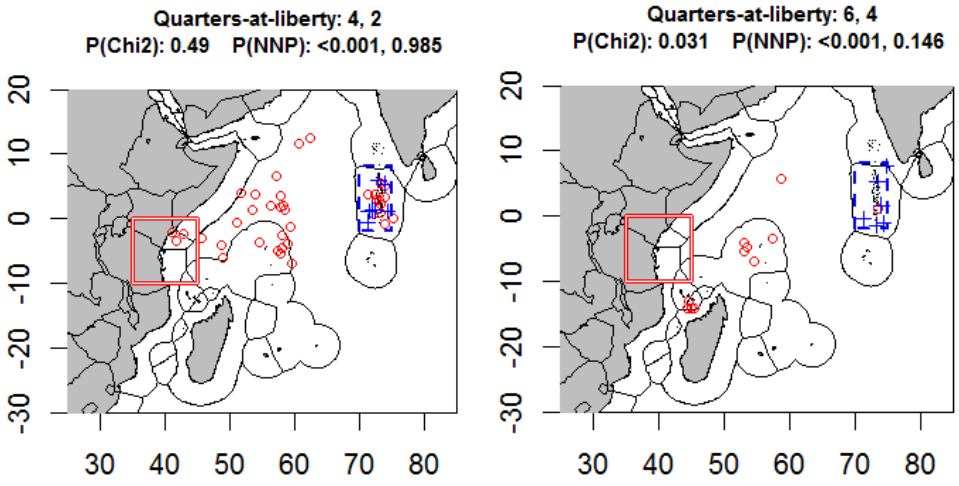

Figure 3. Some example tag recovery distributions for similarly sized Indian Ocean skipjack (see Kolody and Hoyle 2013 for interpretation of mixing statistics). Small coloured boxes represent release areas, points represent recovery locations from the same time window (colours correspond to the release areas, i.e. red circle = recovery from east African release event). The key point is that mixing dynamics appear to be slow and not uniform, i.e. east African releases appear to disperse eastward, while Maldives releases seem to remain resident around the Maldives (for at least 1 year after release).

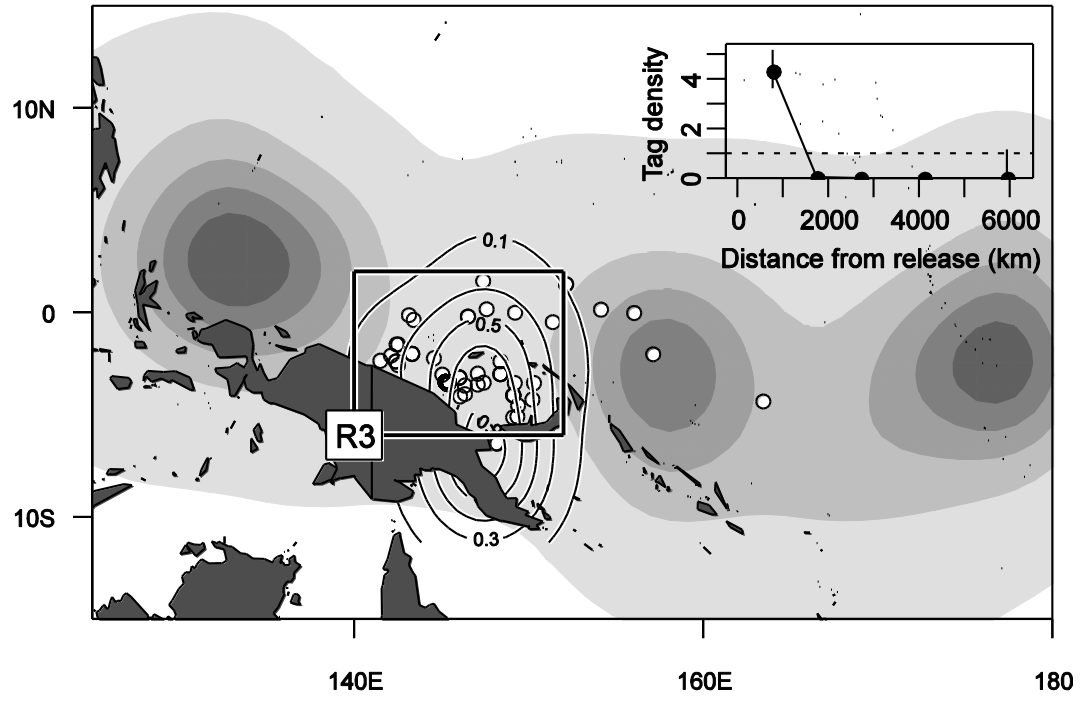

Figure 4. Map of skipjack catch, tag recoveries, and inferred tag density, illustrating that tags are not uniformly mixed with the untagged population as assumed in the stock assessment model (the whole region represents one region in the spatially-disaggregated western Pacific assessment). Tags released in area R3 were recaptured in the marked positions (circles) 4-6 months after release. Density of tags per unit of catch (of a similar size-class) is shown in the contour lines. The distribution of the catch for fish of the designated size class are represented as shaded contours. (from Hoyle et al. 2013)

Attachment 1. Draft concept proposal for an Indian Ocean tuna population study.

Indian Ocean Tuna Population Structure: Estimation with Next Generation Sequencing Technologies and Otolith Micro-chemistry

Concept proposal for discussion with IOTC, funding agencies and potential collaborators

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DRAFT for Discussion

Commercial-in-confidence

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PROJECT OVERVIEW

The project will seek to describe the population structure and connectivity of the most valuable (in terms of economics and food security) tuna and tuna-like species within the Indian Ocean (and adjacent Pacific and Atlantic waters) and outline the key stock assessment and management implications. Collaboration with regional partners will be sought to reduce costs and enhance the capacity for future monitoring and analysis within IOTC member nations.

BACKGROUND AND NEED

There are more than 10 tuna and tuna-like species of substantial commercial and food security value in the Indian Ocean. All of these species are assumed to be highly migratory, and straddle multiple coastal EEZs and international waters, necessitating a multi-national effort for effective fisheries management. The Indian Ocean Tuna Commission (IOTC) is responsible for the management of these species (with the exception of southern bluefin tuna). Some of these species have been assessed with modern, data-intensive, integrated population modelling techniques in recent years (yellowfin, skipjack, bigeye and albacore tunas), while many of the neritic tuna species have never been formally assessed. Attempts have been made to quantify movement within the IO for yellowfin (and to a lesser extent skipjack and bigeye) primarily on the basis of tag displacements observed in the Regional Tuna Tagging Programme (RTTP-IO). Unfortunately, constraints to the RTTP-IO release design and low tag reporting rates for many fleets has meant that movements to/from areas outside of the Western equatorial region are difficult to quantify, even for these tagged species. All assessments to date have assumed a single panmictic spawning population within the Indian Ocean. However, there have been studies suggesting that there may be distinct population structure at a much smaller scale than the IO basin (e.g. for yellowfin: Dammannagoda et al. 2008, Swaraj et al. 2013; skipjack: Dammannagoda et al. 2011, Menezes et al. 2012; and bigeye: Nugraha et al. 2011). Similarly, analyses of tagging data in the Indian Ocean and elsewhere (e.g. western Pacific) has suggested that movement/mixing rates may not be consistent with the large spatial regions that are typically assumed in tuna assessments. If the scientific stock assessment advice is based on invalid assumptions, management may fail to achieve stated objectives related to conservation and optimal economic use of the resource. Specifically, if populations are distinct (or mixing rates are very slow within a panmictic population), some populations (or sub-regions) could be locally over-exploited and management measures might be directed toward the wrong populations.

There is a clear need to underpin assessment and management advice with a basic understanding of population structure and connectivity among populations within the Indian Ocean (and potentially with adjacent populations in the Atlantic and Pacific Oceans). This is of particular importance for small island and developing states with short range fishing fleets. But responsible management is of course in the long-term interest of the distant water fishing nations as well, and addressing these fundamental concerns will assist with the attainment of sustainable product endorsements for all fisheries, regardless of whether the populations are revealed to be well-mixed or fragmented.

WORK PLAN

1. **Literature search** to identify prior (and current) population structure work for the target tuna and tuna-like species within the Indian and other oceans. The following list of target species is inferred from IOTC reports, and advice from the IOTC Secretariat:
    i. Yellowfin tuna (*Thunnus albacares*)
    ii. Bigeye tuna (*Thunnus obesus*)
    iii. Skipjack tuna (*Katsuwonus pelamis*)
    iv. Albacore tuna (*Thunnus alalunga*)
    v. Longtail tuna (*Thunnus tongol*)
    vi. Kawakawa (*Euthynnus affinis,* mackerel tuna)
    vii. Narrow-barred Spanish mackerel (*Scomberomorus commerson,* king mackerel)
    viii. Bullet tuna (*Auxis rochei rochei,* bullet mackerel)

ix. Frigate tuna (*Auxis thazard thazard, frigate mackerel*)
x. Indo-Pacific king mackerel (*Scomberomorus guttatus*)

2. **Pilot study to identify the most effective genetic tools for discriminating population structure and species identification**. High throughput, next generation sequencing (NGS) technologies represent a cost effective option (e.g. Restriction site Associated DNA markers or RADtags) for revealing population structure through examination of Single Nucleotide Polymorphisms (SNPs). SNP markers also lend themselves easily to routine and inexpensive ($10-$20 per fish) screening methodologies. Recently developed NGS approaches represent a major advancement over classical techniques (i.e. based on allozymes, DNA microsatellites, and mitochondrial DNA). These latter approaches, are more labour intensive in terms of ability to screen the quantity of suitable markers required to reveal subtle variation necessary to discriminate structure present in marine fish populations. Furthermore, limited screening of classical marker loci is less sensitive and results can potentially be misleading for management purposes when no stock structure is revealed due to limited genetic resolution. For a more broad scale Indian Ocean study, the methodological choice will be informed by an existing project analysing fine scale structure in bigeye and yellowfin samples collected from nine locations across the Indonesian archipelago, and two outlier populations in the Indian and Pacific Oceans(Proctor et al. 2009). At this time, investigation at CMAR examining yellowfin tuna in the Pacific have demonstrated the efficacy of using NGS SNP markers to reveal structure at a level not possible through classical microsatellite and mtDNA approaches. The samples proposed for a broader Indian Ocean pilot scale study will be drawn from our existing sample collections where species identification has been validated with a high level of confidence. If samples are readily accessible for the pilot study, we will seek to independently reproduce key studies which suggest that there is population structure around the Indian sub-continent at a much finer spatial scale than has generally been considered for highly migratory species (e.g. Dammannagoda et al. 2008,2011). If samples with reliable species identification are not readily available, this will be addressed as a priority from the main sampling program.

3. **Sample collection**. An adaptive sampling scheme is proposed, depending on i) existing sample collections held by collaborators, ii) availability of local sampling staff that can insure species identification reliability and low tissue contamination probability, iii) species priorities, iv) likelihood of obtaining a high proportion of multiple target species whenever possible, and v) budget.
   i. Priority locations will initially include approximate extremes of the population range and midpoint within the Indian Ocean (plus out populations in the Pacific and/or Atlantic for species that appear to form a continuum across oceans). Depending on the initial evidence for population structure indicated by the pilot study (and Proctor et al. 2009 study), additional intermediate populations will be added at the appropriate scale.
   ii. Target of 50-100 samples per species and location and time period.
   iii. Replication over two years is recommended as an initial assessment of marker stability
   iv. Muscle tissue will be extracted from reasonably fresh or frozen fish, otoliths extracted for microchemistry analysis and lengths taken to infer age (and likely proximity to natal spawning ground).
   v. The main sampling target for stock discrimination will be on juvenile tuna, as they are less likely to have moved far from their natal spawning grounds.
   vi. Samples of larger tuna from key fisheries will also be sought to understand the mixed-stock nature of the fisheries, i.e. for effective management you should know not only where the populations spawn, but also where they are captured. The two distributions could be very different if there is spawning ground site fidelity, but mixing on foraging grounds.

4. **Genetic analyses.** The most reliable and cost effective method identified in the pilot study will be applied to the samples obtained in 3. Samples will be analysed sequentially and adaptively - if there is no evidence of differentiation for the most distant samples, intermediate samples would not be analyzed, or given a low priority. Conversely, genetic differences at the finest sampling scale would encourage higher resolution sampling in the next iteration.

5. **Otolith microchemistry analysis.** Laser ablation mass spectrometry and solution-based inductively coupled mass spectrometry will be used as an independent assessment of early juvenile residence locations as an independent corroboration of genetic population differentiation.

6. **Statistical analyses and population structure summary synthesis.** Appropriate analyses will be undertaken to identify discrete spawning populations. For key species, population structure results will be synthesized in relation to other evidence of population structure and movement (e.g. from tagging data, fisheries catch, size composition and catch rates). Implications (for current) and recommendations (for future) IOTC stock assessment and management options will be described, and the need for mixed-stock fishery analyses will be evaluated.

DELIVERABLES - OUTPUTS

1. Genetic markers for population (stock) discrimination
2. Evaluation of usefulness of combining data from genetic markers and otolith microchemistry across multiple sampling years (i.e. uncertainty, sensitivity, spatial and temporal stability)
3. Population structure atlas based on combined otolith and genetics results
4. Peer-reviewed publications and IOTC Working Papers describing key results and implications for assessment and management.

DELIVERABLES – OUTCOMES/IMPACT

1. The genetic tools developed will provide the basis for ongoing monitoring of population structure, and will support emerging population estimation methods (e.g. genetics-based mark-recapture techniques based on the identification and re-identification of individuals or the observed frequencies of closely-related pairs).
2. The revised insight into population structure will permit more effective assessment and management tools to be applied to the Indian Ocean tuna populations (including the parameterization of operating models for management strategy evaluation).
3. Improved understanding of the population structure and connectivity will help small island and coastal states to more effectively negotiate their access rights in relation to local fisheries productivity, and all fisheries should gain market benefits associated with consumer demand for sustainable fisheries products (e.g. Marine Stewardship Council Certification).

CSIRO CAPABILITIES

CSIRO has the necessary capability and experience to lead and coordinate this project. This includes the technical and analytical skills to develop pioneering genetic techniques for tuna population analyses, otolith microchemistry experience, in-depth knowledge of tuna stock assessment methods and management options, ongoing engagement with tuna RFMOs (and IOTC in particular), and the management skills to coordinate large international collaborations and capacity building. Key staff:

- Peter Grewe – population geneticist
- Dale Kolody – population dynamics modeller
- Campbell Davies – fisheries scientist
- Craig Proctor – fisheries scientist

See references for some indicative project examples.

POTENTIAL COLLABORATIONS

There is potential for collaborations with other researchers to potentially reduce costs (e.g. in sample collection, marker development, marker validation, and analysis) and build wider confidence in the results and adoption of the outcomes. We have had discussions with the following institutions/individuals (or their colleagues) who have expressed interest in potential collaborations for work on tuna and tuna-like species. This list is not exhaustive, and detailed work plans have not been explored at time of submission of this concept note:

1. Fausto Tinti, Carlo Pecoraro – University of Bologna, associated with TROPTUNA project
2. Emanuel Chassot – IRD, Seychelles, associated with TROPTUNA project
3. Hilario Murua – AZTI, Spain – IO tuna otolith microchemistry
4. Lubna al Kharusi, Madjid Delghandi – Oman – longtail, kawakawa, frigate tuna and striped bonito population structure interest
5. BOBLME - (John Candy consultant) – Indian mackerel population structure project
6. Budi Nugraha – TFRI, Benoa, Indonesia – IO bigeye genetics
7. Hari Eko Irianto, Wudianto - RCFMC, Jakarta, Indonesia – high resolution yellowfin and bigeye samples within Indonesian waters.
8. Sudath Dammannagoda – Queensland Uni Tech. – IO skipjack & yellowfin genetics
9. Jérôme Bourjea – IFREMER, Ile de la Réunion – IO tropical and temperate tuna, neritic tuna

References

Bravington, M., Grewe, P., Davies, C., 2012. Report of the close-kin project: estimating the absolute spawning stock size of SBT using genetics. Commission for the Conservation of Southern Bluefin Tuna working paper CCSBT-ESC/1208/19.

Dammannagoda, S.T., Hurwood, D.A., and Mather, P.B. 2011. Genetic analysis reveals two stocks of skipjack tuna (*Katsuwonas pelamis*) in the northwestern Indian Ocean. Can. J. Fish. Aquat. Sci. 68: 210-223.

Dammannagoda, S.T., Hurwood, D.A., and Mather, P.B. 2008. Evidence for fine geographical scale heterogeneity in gene frequencies in yellowfin tuna (*Thunnus albacares*) from the north Indian Ocean around Sri Lanka. Fish. Res. 90: 147-157.

Kolody, D., Herrera, M., and Million, J. 2010. Exploration of Indian Ocean Bigeye Tuna Stock Assessment Sensitivities 1952-2008 using Stock Synthesis (Updated to include 2009). IOTC-2010-WPTT-04 (Rev1).

Kolody, D., Herrera, M., and Million, J. 2011. Indian Ocean Skipjack Tuna Stock Assessment 1950-2009 (Stock Synthesis). IOTC-2011-WPTT-13-31Rev_1.

Kolody, D., S. Hoyle. 2013. Evaluation of tag mixing assumptions for skipjack, yellowfin and bigeye tuna stock assessments in the western Pacific and Indian Oceans. WCPFC Working Paper: WCPFC-SC9-2013/SA-IP-11

Menezes, M.R., Kumar, G., and Kanal, S.P. 2012. Population genetic structure of skipjack tuna *Katsuwonas pelamis* from the Indian coast using sequence analysis of the mitochondrial DNA D-loop region. J. Fish. Biol. 80: 2198-2212.

Nugraha, B., Novianto, D., and Barata, A. 2011. Keragaman genetik ikan tuna matabesar (*Thunnus obesus*) di Samudera Hindia. [*Genetic diversity of bigeye tuna (Thunnus obesus) in Indian Ocean*]. Ind. Fish. Res. J. Vol.17 (4), 285 - 292 (In Bahasa Indonesia, with English abstract).

Proctor, C. 2009. Developing research capacity for management of Indonesia's pelagic fisheries resources. ACIAR project proposal.

Proctor,C., Purwanto, Budhiman, A., Nurhakim,S., Iskandar Prisantoso,B., Andamari,R., Davis,T.L., Dowling,N., Wudianto, Merta,G.S., Sadiyah,L., Triharyuni, S., Davies,C., Meta, B.S., Retnowati,D., Team observers and Team enumerators. 2010. Capacity development to monitor, analyse and report on Indonesian tuna fisheries. Final report for ACIAR Project FIS/2002/074. 69pp.

Proctor, C.H.; Thresher, R.E.; Gunn, J.S.; Mills, D.J.; Harrowfield, I.R.; Sie, S.H. 1995. Stock structure of the southern bluefin tuna Thunus maccoyii: an investigation based on probe microanalysis of otolith composition. Mar. Biol. 122: 511-526.

Swaraj, P.K., Kumar, G., M.R. Menezes and R.M. Meena. 2013. Mitochondrial DNA analysis reveals three stocks of yellowfin tuna *Thunnus albacares* (Bonnaterre, 1788) in Indian waters. Conserv. Genet 14: 205-213.

Ward, R.D., Elliott, N.G., Innes, B.H., Smolenski, A.J. and Grewe, P.M. 1997. Global population structure of yellowfin tuna (*Thunnus albacares*) inferred from allozyme and mitochondrial DNA variation. *Fishery Bulletin US*, 95, 566-575.

Williams, A.J., Farley, J.H., Hoyle, S.D., Davies, C.R., and Nicol S.J. 2012. Spatial and sex-specific variation in growth of albacore tuna (*Thunnus alalunga*) across the south Pacific Ocean. PLoS ONE 7(6): e39318.