



**MEMORANDUM OF UNDERSTANDING  
ON THE CONSERVATION AND  
MANAGEMENT OF MARINE TURTLES  
AND THEIR HABITATS OF THE INDIAN  
OCEAN AND SOUTH-EAST ASIA**

CMS/IOSEA/AC9/Doc.23/Rev.1  
15 March 2021

Original: English

9<sup>TH</sup> MEETING OF THE ADVISORY COMMITTEE  
*online*, 15-19 March 2021  
Agenda Item 23

**GENETIC RESEARCH ON MARINE TURTLES IN THE IOSEA REGION**

*(Prepared by AC Members)*

Action Requested:

- Take note and develop a concrete plan how to proceed

## GENETIC RESEARCH ON MARINE TURTLES IN THE IOSEA REGION

Activity #44 of the IOSEA Work Programme 2020-2024 addressed Signatory States, the Advisory Committee and the Secretariat with the following tasks:

44. *Conduct genetic analyses to identify genetic stocks (management units) for Marine Turtles in the IOSEA region:*
- a) *compile and assess available information to identify needs and opportunities for regional analyses to inform genetic stock assessment, starting with nesting beach sampling*
  - b) *develop maps for each genetic stock indicating rookery locations and habitat use in pelagic and benthic environments, rather than relying on the Regional Management Unit (RMU) maps given in Wallace et al. (2011)<sup>1</sup>*
  - c) *incorporate genetic sampling into ongoing monitoring activities (subject to budget and resources available)*
  - d) *standardize protocols for collection and storage of samples to ensure they will remain usable in the long term*
  - e) *address capacity building needs to enable in-country analyses, upon request*
  - f) *AC and Secretariat to facilitate contacts with laboratories and CITES permits to conduct genetic analyses, upon request*

The present document is a follow up from the document “MARINE TURTLE GENETIC STOCKS OF THE INDO-PACIFIC: IDENTIFYING BOUNDARIES AND KNOWLEDGE GAPS” (FitzSimmons and Limpus), made available as [AC9/Inf.23](#), which addresses an action point identified by the Sixth Meeting of IOSEA Signatory States, and that was reported on during MOS7 (September 2014).

### **Brief history of genetic research on marine turtles**

Genetics research is a powerful way for understanding species population structure and connectivity at different geographic scales and time; phylogeography, population overlap (e.g., mixed stocks in feeding habitats), investigations of hybridisation between species, mating systems (e.g. multiple paternity in offspring) to mention a few. Of particular importance is the ability to identify and characterise different “genetic stocks” (or genetic management units). For example, individuals encountered at the same foraging aggregation may not necessarily originate from the same genetic stock: although they may share the same foraging area, they often nest at different genetically distinct rookeries across long distances– a concept known as “mixed stocks.” These overlapping genetic stocks add complexity to effective conservation planning and actions.

The use of genetic analysis of marine turtles began in the late 1970s, and since then, genetic methods have become increasingly important for answering critical biological questions for marine turtles. A wide range of “genetic markers” (distinct sequences of DNA from known locations on a chromosome, mitochondria, ribosome, or other DNA structure in a cell) are now available to answer a broad range of questions. The maternally inherited mitochondrial DNA (mtDNA) has been the marker of choice for detecting population structure among rookeries and is the preferred tool for conducting mixed stocks in foraging areas. However, mtDNA does not provide the complete picture and does not include any information about male-mediated gene flow that might connect rookeries that are considered isolated though maternally inherited mtDNA. Therefore, the combination of mtDNA and nuclear DNA (nDNA – inherited

---

<sup>1</sup> Wallace et al. (2011) Global Conservation Priorities for Marine Turtles. PLoS ONE 6(9): e24510. <https://doi.org/10.1371/journal.pone.0024510>

from both parents) studies should be encouraged to get a complete understanding of the genetic population structure.

The first genetic studies on marine turtles revealed low genetic diversity, possibly because of low metabolic rate and long generation time on these species; also, the early studies sampled relatively small numbers of turtles. This low level of genetic variation prompted most researchers working population structure to rely on the mtDNA from the 'control region' (that part of the mitochondria with the greatest variation in genetic diversity). Early studies using nDNA focused on microsatellite markers, (spans of nuclear DNA characterised by repeating sequences) were limited to studies on paternity, with only a few studies looking at population structure (compared to mtDNA). With the reduction in the cost of next-generation sequencing, these new genomic tools are now becoming accessible for the study of non-model organisms like marine turtles. These new approaches allow for the analysis of 10s of thousands of single nucleotide polymorphism markers (SNPs) (compared to 10s of markers for microsatellites) and will undoubtedly reveal new insights into fine-scale global population structure and connectivity. New approaches such as environmental DNA (eDNA) allow for detection of target species from environmental samples (e.g. water sample) and represent important advances in revealing the presence of particular species in any area where taxonomic or genetic samples cannot be physically collected. Such research may facilitate the definition of geographic distributions for particular species.

### **Advances in genetic research on marine turtles in the IOSEA region**

Major advances in genetic research on marine turtles in the IOSEA region have progressed during the last 10-15 years. This has allowed greatly improved understanding of population structure of all six species; geographic and seasonal occurrence of numerous stocks of all species; and deeper understanding of the connectivity between different areas where certain stocks occur, particularly oceanic regions/areas where there has been a paucity of basic information on marine turtle occurrence, dispersal, and migration. This recently acquired information is essential for developing well-informed conservation and management plans for many stocks of all six species in the IOSEA region. An excellent example is the world/global phylogeographic work recently published for the Green Turtle (Figure 1).

The maps in Annex 1 drawn based on mtDNA results show the updated distributions of genetic units for all species in the IOSEA Region and the main gaps that still need to be investigated.

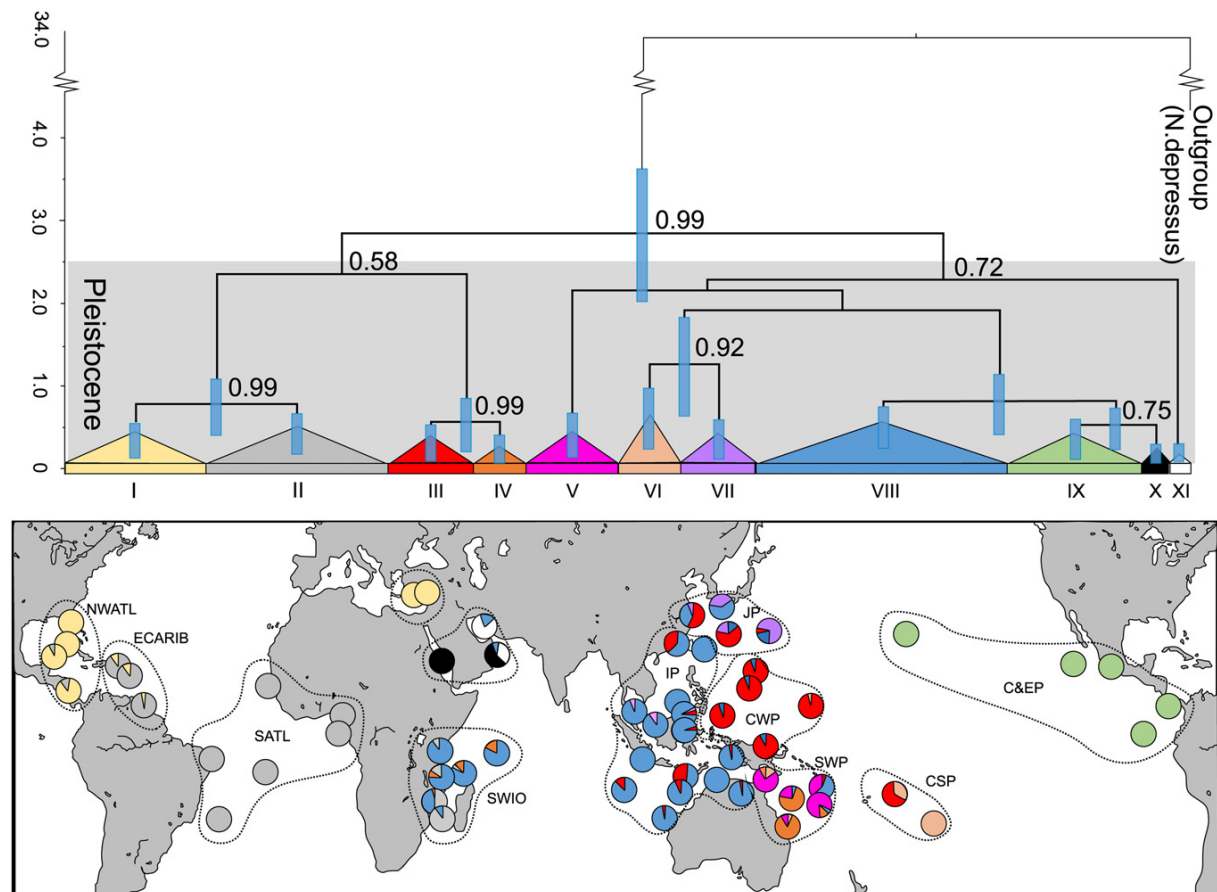
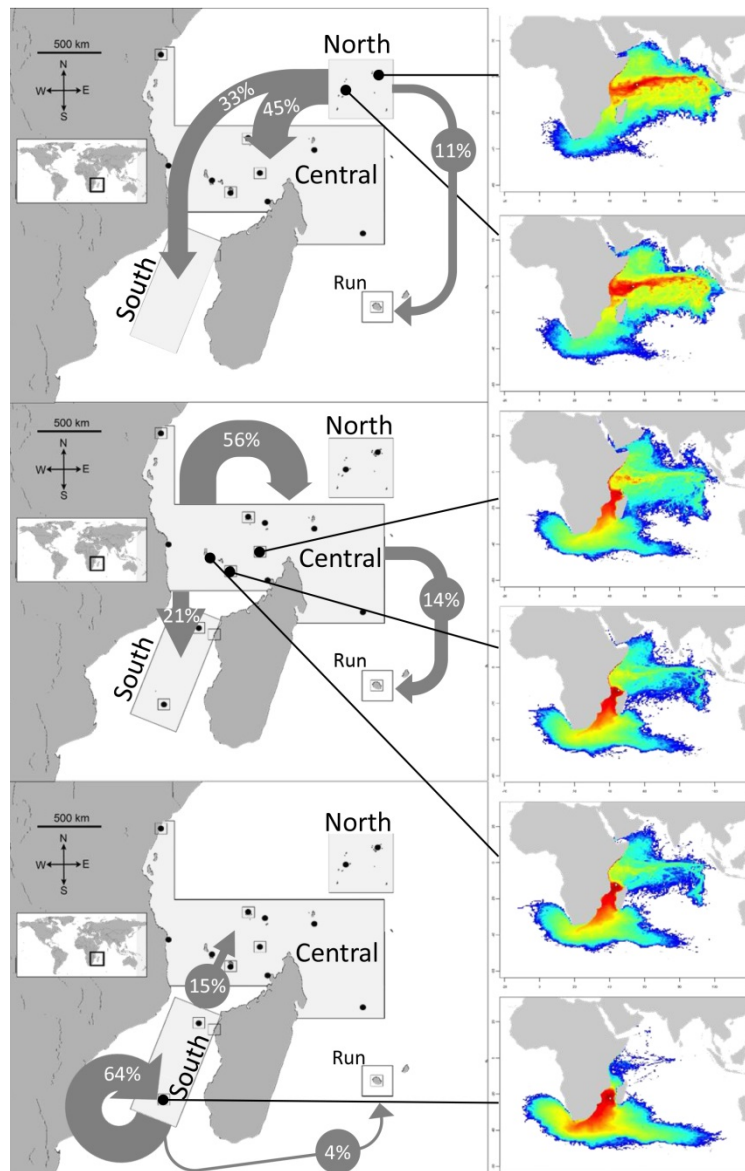


Fig. 1: Bayesian phylogenetic tree inferred from the mitochondrial control region (384 bp) of green turtles, *Chelonia mydas*. Numbers above branches correspond to posterior probabilities calculated in BEAST. The axis of branch lengths is given in millions of years. Vertical blue bars denote the 95% highest posterior density intervals values estimated for tree nodes. *Natator depressus* was the outgroup for this analysis. The map shows the proportions of population ancestry from each of 11 green turtle mtDNA lineages (I–XI). Pie charts on the map indicate the relative proportion of individuals from each MU that belong to each lineage with each color corresponding to a genetic lineage. Major evolutionarily distinct regions (grouped by dashed lines on the map) are NWATL = NW Atlantic, ECARIB = Eastern Caribbean, MED = Mediterranean, SATL = South Atlantic, SWIO = SW Indian Ocean, NWIO = NW Indian Ocean, IP = Indo-Pacific, JP = Japan, CWP = Central West Pacific, SWP = SW Pacific, CSP = Central South Pacific, and C&EP = Central and Eastern Pacific (From Jensen et al 2019).

Directly related to, and complementary with, the many studies on the population structure of nesting turtles, the studies on mixed stock composition of turtles of both sexes and different size classes from foraging grounds are also important. These have become more common in several IOSEA Sub-regions, and they emphasise how complex the mixing of individuals from different genetic stocks can be at the scale of the Indian Ocean and South East Asia (e.g., Figure 2).



*Fig. 2: Dispersal from rookeries. Small maps show the density distributions of juveniles emerging from the seven simulated nesting areas (Colours indicate the number of particles). The three central maps show the proportional distribution of juvenile turtles from each area (Central North and South) based on Bayesian “rookery-centric” estimates using the many to many mixed stock analysis (from Jensen et al 2020)*

Therefore, it is recommended to continue supporting genetic approaches that use mtDNA markers and share results for global analysis. At the same time, innovating works such as genome-wide screening and eDNA should be strongly supported and developed on marine turtle species as those approaches may become a standard in the coming years.

### **Structuring the genetic work at the scale of the IOSEA Region**

The more recent advances mentioned above were made possible because active genetic working groups have been created in the region during the past few years. This includes the working group on hawksbill turtles that meets online once a month with the goal of obtaining samples and updating and expanding mtDNA data on hawksbill rookeries, with a focus on SE

Asia and SW Pacific (Indo-Pacific Hawksbill Genetics Working Group (IPHGWG)). A similar genetic expert group under the research project TIMOI (Tortue IMbriquée Océan Indien lead by CEDTM) focuses on the same species in the South West Indian Ocean, and links between the two groups have been created to share results for a broader analysis over the IOSEA Region.

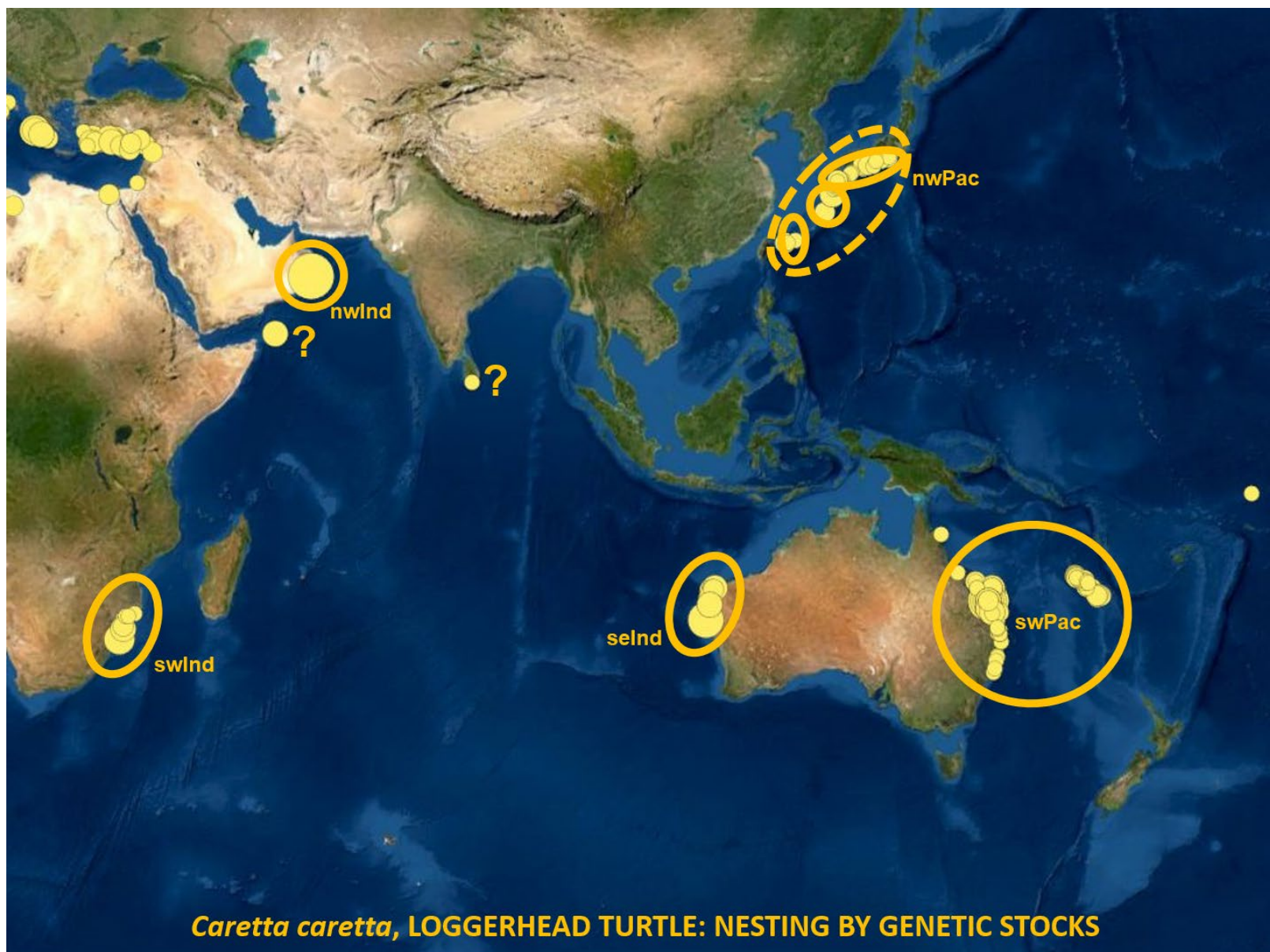
A third genetic working group is the Asia-Pacific Marine Turtle Genetic Working Group whose goals are to enhance technical capacity, standardise methodologies, identify research priorities, and establish a regional collaborative network to facilitate genetic studies that can support national and international marine turtle management and protection efforts.

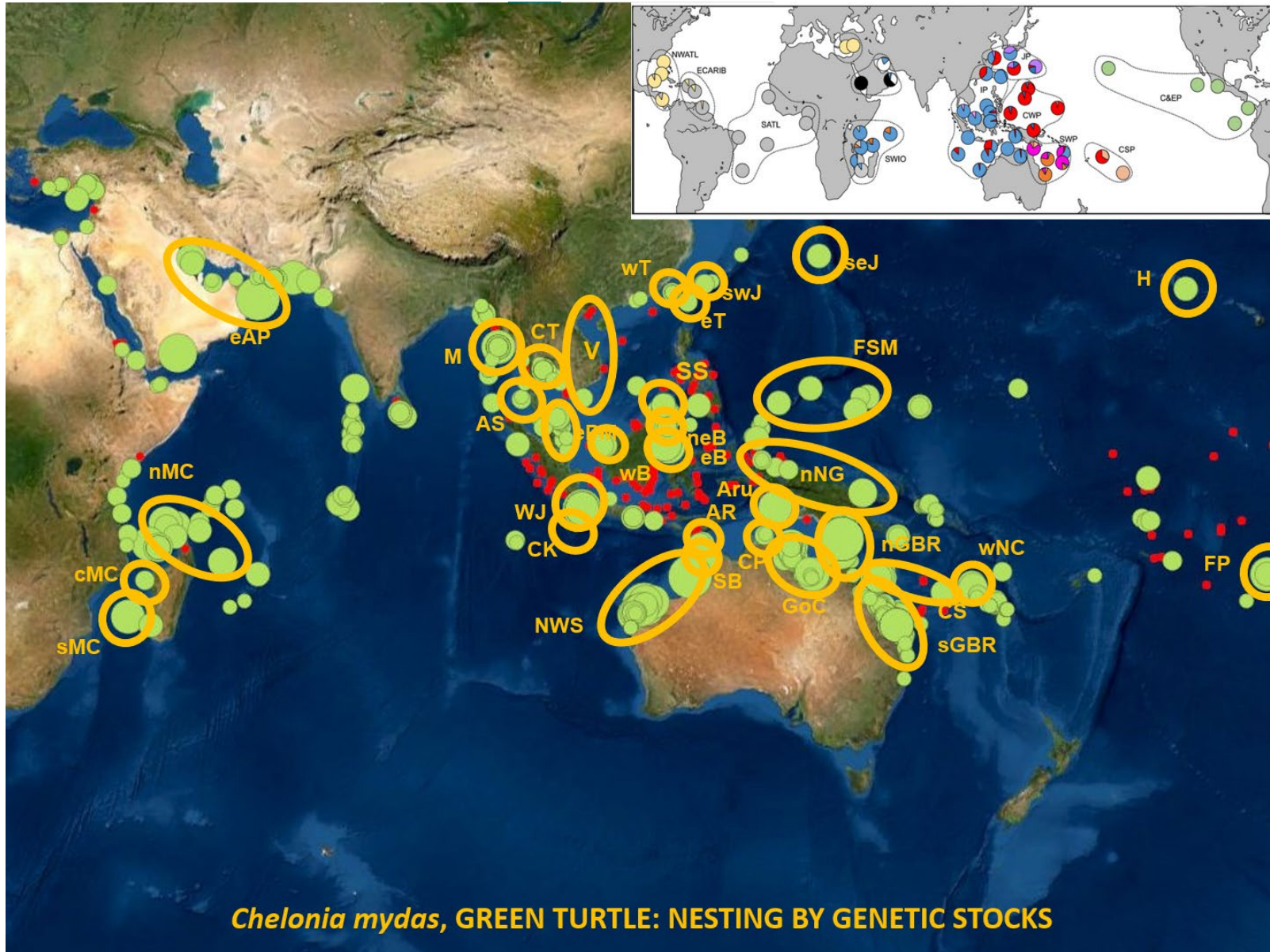
Another example is a recent group of expert created under an European Union Project (Development of new genome-wide SNP markers and Whole mitogenome sequencing for *C. mydas*). This project gather geneticists working in the Atlantic and Indian Oceans who are developing a fine scale population structure model of green turtles in the western Indian Ocean and also investigating past links between the Indian Ocean and the Atlantic populations.

Many other initiatives at smaller scales are ongoing with active and growing networks. We believe that any non-expert initiative in the IOSEA Region that would like to develop an initiative on marine turtle genetics will easily find a geneticist expert for support.

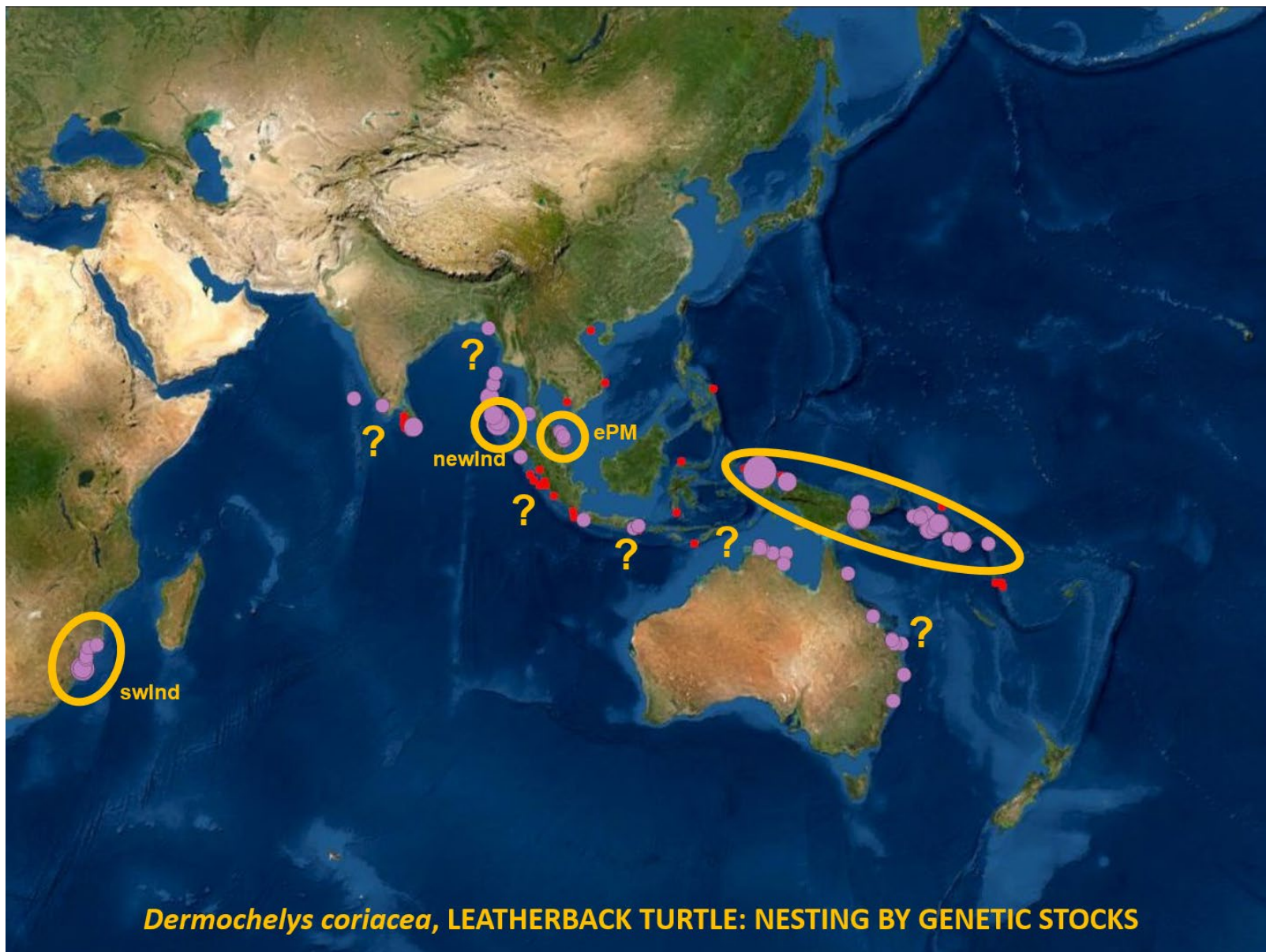
### **Main issues to highlight**

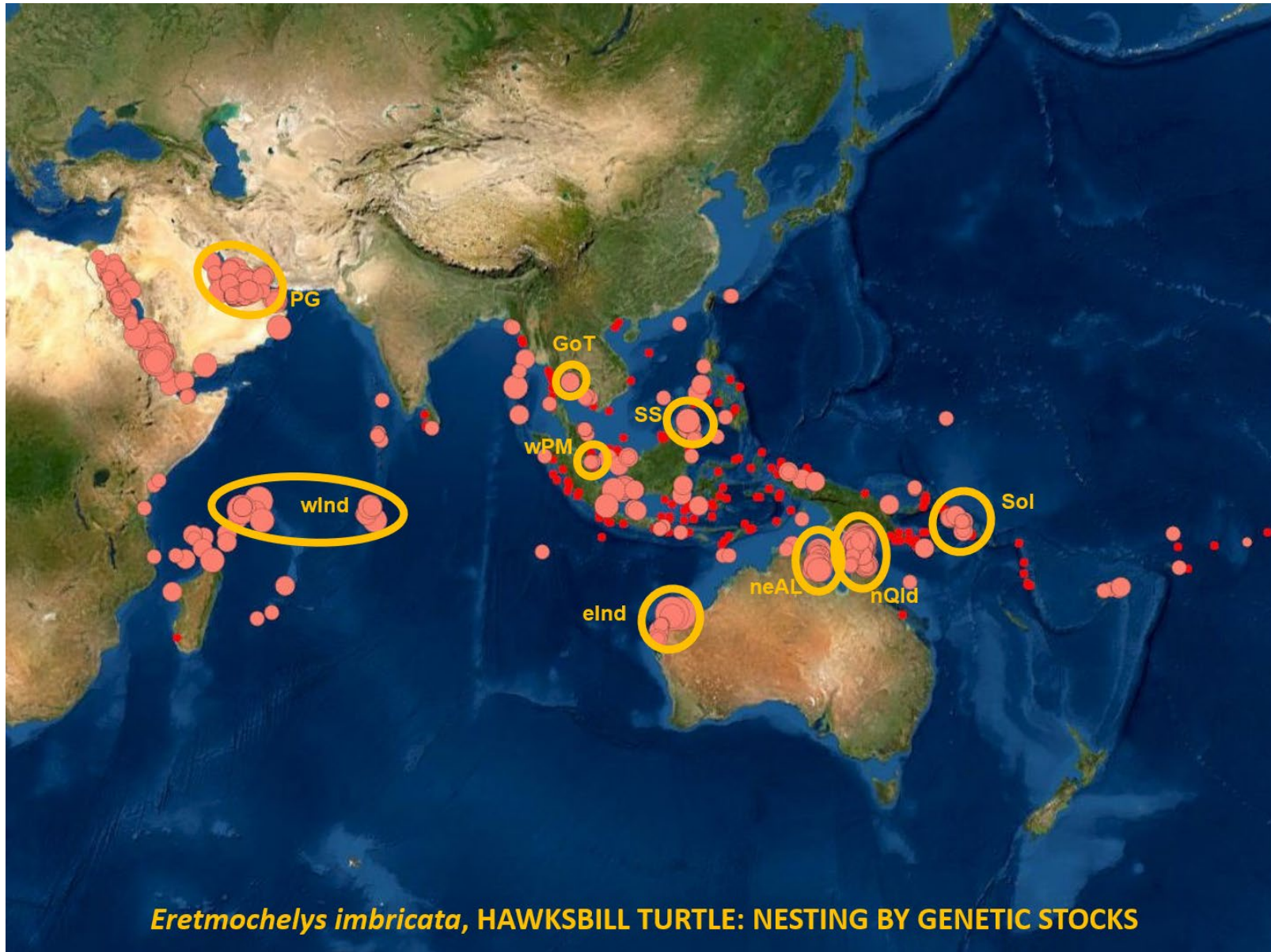
One of the biggest limitations for most genetic work is a lack of funding, not for networking, but for both supporting field collection and analysis of samples. Another important issue is obtaining required CITES permits. The national process for obtaining permits to import or export marine turtle samples is already complicated, but at least it works. However, when working with industrial fisheries and on-board observers (that can be a cost-effective way to routinely collect hundreds of genetic samples of marine turtles during their obscure open sea of life stage), the permit process becomes extremely complicated, if not impossible, with a cascade of importation/exportation demands that results in countless onerous and tedious administrative chores, which routinely result in making the project impossible. Several regional initiatives for expanding turtle sampling have failed because of enormous complications in obtaining required CITES permits. A CITES and IOSEA/CMS dialogue is urgently needed to clarify and facilitate the procedure for obtaining CITES permits to make it possible to carry out scientific research required for the conservation and management of these endangered species.

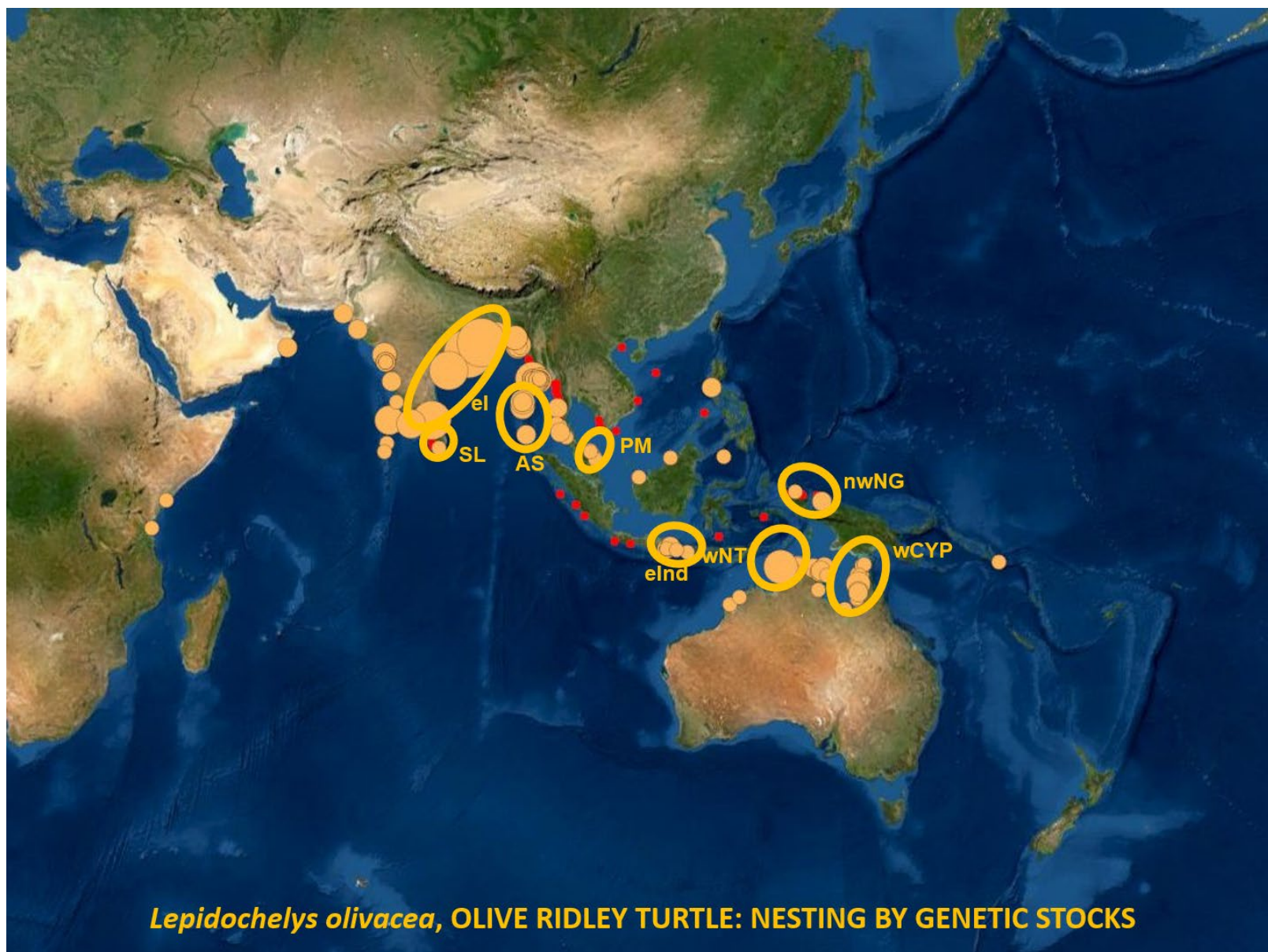












*Lepidochelys olivacea*, OLIVE RIDLEY TURTLE: NESTING BY GENETIC STOCKS

