Project Update: July 2021

#### Field studies across the Eastern Adriatic

During the past 3 months, we had seven short field trips lasting 3 to 7 days across the eastern Adriatic sea organised through National Geographic, Discovery Channel, Foundation Ensemble and Federal Fund for Environmental protection. The field studies combined technical diving, trawl analysis and the ROV filming on the continental shelves up to 100 m deep. I have filmed the breeding area of marbled electric torpedo rays and black-spotted smoothhound sharks in the south Adriatic. Those two breeding grounds have been carefully monitored by Sharklab ADRIA in past 3 years, with special focus on the prenatal disease diagnosis, health, reproductive biology, and population ecology. Furthermore, we have identified a likely breeding/nursery area for critically endangered common angel shark in the central Adriatic with several juvenile/subadult records – the original scientific paper is in press right now. Pressures such as solid waste and war waste were carefully mapped.

## Laboratories: Toxicology and Pathology

During the past 2 months, I have analysed 25 sharks (including 15 juveniles) from Rijeka and Pula (north Adriatic, Croatia), Rogoznica and Split (central Adriatic, Croatia), and Neum (south Adriatic, Bosnia and Herzegovina). All sharks were samples from an already dead by-catch. Random sections of liver and spleen were routinely processed, embedded in paraffin, sectioned at 5µm and stained with hematoxylin in order to analyse the presence of MMCs (melanomacrophages) as biomarkers for contaminants and pollution. Furthermore, samples of liver, brain, kidney, aills and reproductive system were analysed for the presence of pathological changes. Due to incompatibility with most of the mammalian CD markers, we stopped immunohistochemical analyses of diseases, especially tumours. The digestive system of each sample was isolated for further qualitative and quantitative micro- and nanoplastic studies for Discovery Channel. Upon isolation, organs were rinsed in hH<sub>2</sub>O, measured, weighed, and transferred into separate Erlenmeyer flasks. The biological material was digested via dilution with filtration, wherein 20 ml of 30% H<sub>2</sub>O<sub>2</sub> was added per 1 g of wet weight. Samples were incubated for 24h at 65 °C in a bain-marie a type of heated bath. The suspension was filtered through the Macherey-Nagel cellulose filter papers with retention capacity of 1 to 2 µm - grades MN 640 de (thickness 200 μm, weight 100 g/m<sup>2</sup>) and MN 619 (thickness 170 μm, weight 75 g/m<sup>2</sup>). Upon the filtration, filter papers were placed into the petri dishes and left to dry for 24h. Likely microplastic fibres, fragments, films, and pellets were identified using stereo microscope and secluded for Fourier-transform infrared (FTIR) and RAMAN spectroscopy. Results will be published with EC Discovery Channel and through several scientific papers which are under preparation.

# Media coverage and promotion

I was featured as one of the most successful scientists within the "30 under 30" program, with a mini docu-video about my field and laboratory studies, mostly conducted within the Rufford projects and my engagements in National Geographic and Discovery Channel. Furthermore, I had three other guest appearances at N1, regional CNN partner, discussing the effects of habitat loss, degradation and pollution on tangible aquatic taxa. I have spoken of the urgency of unique regional protection of sharks, skates and rays in the Adriatic sea – as the only effective way to save the threatened species. Even if not related to the project, I gave my full support and contributed to the saving of rivers in Bosnia and Herzegovina – which are highly

threatened by habitat loss and dozens of illegal mini hydropower plants. This resonated strongly in the media throughout the country and I will continue to provide full support for nature research and conservation.

## Recorded sharks and expertises in media:

https://morski.hr/2021/05/06/iznimno-rijedak-nalaz-ribar-ulovio-i-pustio-strogo-zasticenog-sklata-prvi-put-smo-ga-vidjeli-u-zivotu/

(Neonatal critically endangered Common Angel Shark from the Central Adriatic sea in Croatian waters)

https://morski.hr/2021/04/26/video-neobican-prizor-morski-pas-plivao-uz-samu-obalu-na-svega-pola-metra-dubine/

(juvenile Blue shark on the coast of Vis island, Croatia)

https://morski.hr/2021/05/27/strogo-zasticeni-pas-volonja-strucnjak-educirati-ljude-i-kroz-osudujuce-komentare-prema-pocinitelju/

(Six-gill shark landed in Croatia)

https://morski.hr/2021/07/03/morski-pas-u-tisnom-cijeli-dan-pliva-u-plicaku-i-nikoga-se-ne-boji-izgleda-osamuceno/

(juvenile Blue shark in Tisno, central Adriatic sea, Croatia)

### Links for webpage:

https://www.youtube.com/watch?v=pAukOrsvZb8&t=86s

(short docu-video for the "30 under 30" programe)

https://www.klix.ba/biznis/andrej-gajic-rukovodi-studijama-pri-national-geographicui-discovery-channelu-u-sad-u/210531160

(Article about my work for National Geographic and Discovery Channel)

https://www.klix.ba/vijesti/bih/borbu-za-rijeke-bosne-i-hercegovine-podrzao-je-i-naucnik-andrej-gajic/210724016

https://www.6yka.com/novosti/naucnik-andrej-gajic-podrzao-borbu-za-spas-rijeka-bih

https://radiosarajevo.ba/metromahala/teme/naucnik-andrej-gajic-podrzao-i-dao-doprinos-u-borbi-za-rijeke-bih/425029

(three articles about my support on the river conservation)



**PHOTO 1**: Field studies and ROV habitat filming across the Neretvanski channel, border line between Croatia and Bosnia and Herzegovina. © Sharklab ADRIA.



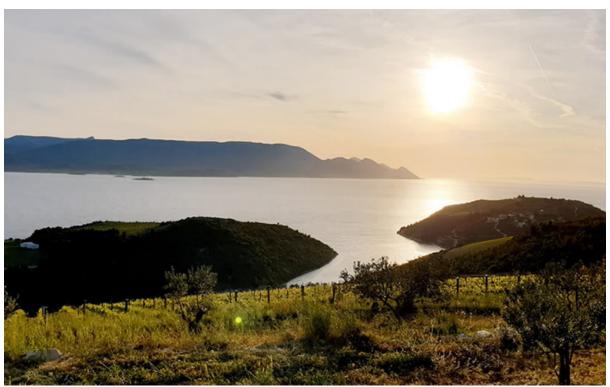
**PHOTO 2**: Morphological analysis and prenatal disease diagnostics in black-spotted smoothhound embryos from Split channel, Croatia. © E. Karalić/Sharklab ADRIA.



**PHOTO 3**: Andrej as featured scientist within the "30 under 30" project discussing the field and laboratory methodology and importance of long-term conservation. © Represent Communications.



**PHOTO 4**: Monitoring the Marbled electric ray breeding ground in Neum bay, Bosnia and Herzegovina. Popluation and health assessment. © E. Karalić/Sharklab ADRIA.



**PHOTO 5**: Beautiful Malostonki bay, highly important habitat for certain elasmobranch species in Croatia. © A. Gajić /Sharklab ADRIA