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Unpublished Mediterranean and Black Sea records of marine alien, cryptogenic, and neonative species (III)

Jennifer C.A. Pistevos[1], Argyro Zenetos[2], Yolanda Stranga[1], Adrian Teaca[3], Tatiana Begun[3], Domen Trkov[4], Joachim Langeneck[5], Mihaela Muresan[3], Robert Comas-González[6], Juan Sempere-Valverde[7,8], Andrea Toso[5,9], Sahar Chebaane[7,10], Carlos Navarro-Barranco[8], Antonietta Rosso[11], Lydia Png-Gonzalez[6], Sylvaine Giakoumi[12], Maite Vázquez-Luis[6], José Manuel Cañizares-González[6], Chiara Siddiolo[11,13], Gemma Donato[11], Salvatore Giacobbe[14], Aina Carbonell[6], Rym Zakhama-Sraieb[15,16], Emanuela Di Martino[11], Izdihar Ali Ammar[17], Andrea Cosentino[18], Intissar Mnasri Afifi[16], Rossana Sanfilippo[11], Ghazi Bitar[19], Eleni Kytinou[1,20], Gokhan Tari[21], Mehmet Fatih Huseyinoglu[21], Vasilis Gerakaris[20], Yiannis Issaris[20], Polytimi-Ioli Lardi[1,20], George Oikonomidis[20], Maria Salomidi[20], Konstantinos Tsirintanis[20], Periklis Kleitou[22], George Constantinou[22], Christina Michail[22], Michel Bariche[23], Alan Deidun[24], Alessio Marrone[24], Michail Ragkousis[1], Manuela Falautano[25], Luca Castriota[25], Carla Morri[26], Carlo Nike Bianchi[26], Grigorios Skouradakis[27], Fiona Tomas Nash[28], Markos Digenis[29], Emanuele Mancini[11,30], Elitsa Slaveva Stefanova[131], Adriana Vella[32], Noel Vella[32], Vesselina Vasileva Mihneva[33], Evangelos Papadimitriou[1], Marius Skolka[34], Kremena Stefanova[31], Ana Fortič[4], Maria Garcia[35], Thanos Dailianis[27], Laura Tamburello[12], Victor Surugiu[36], Bessy Stancanelli[37], Vincenzo Di Martino[38], Angeliki Bosmali[1], Borut Mavrič[4], Vasilis Gerovasileiou[27,29], Demetris Kletou[22,39], Fabio Crocetta[40,41], Ioannis Rallis[27,29], Julian Evans[42], Maria Altamirano[43], Leda Liyue Cai[44], Ergün Taşkın[45], Giulio Franzitta[12], Yassine Ramzi Sghaier[46], Abigail Kennedy[47], Angela G. Bartolo[48], Frithjof C. Küpper[47,49], Maria Rousou[50], Víctor Orenes-Salazar[51], Andrea Bonifazi[52], Louis Hadjioannou[44], Zois Vlantaridis[29], Hany El Shaer[53], Mülkibar Çiftçioğlu[54], Hasan Cerim[54], Lovrenc Lipej[4], Daniele Grech[55], Maria Zotou[1], Ernesto Azzuro[56], Patrick J. Schembri[42], Francesco Tiralongo[11,57], Georgios Chatzigeorgiou[27], Manfredi Di Lorenzo[12], Maria Vigo Fernandez[12], Antonio Di Franco[12], Martina Orlando-Bonaca[4], Antonio Calò[58], Christina Malefaki[59], Jose Antonio García-Charton[51], Orestis Papadakis[1], Sercan Yapıcı[60], Valentina Tanduo[40], Ilaria D'Aniello[61], Marco Munari[40,62], Thodoros E. Kampouris[63,64], Matteo Putignano[5,9], Riccardo Virgili[40,41,65], Sandra Agius Darmanin[66], Furkan Bilgiç[67], Giacomo Sardo[68], Monica Calabrò[68], Athanasios Nikolaou[1], Zinovia Erga[64], Ali Zogheib[69], Paolo Balistreri[70], Senem Çağlar[71], Ferhat Yalgın[72], David Osca[73], Eirini Gratsia[27,59], Giorgio Aglieri[12], Melih Ertan Çinar[74], Mirko Mutalipassi[40], Salvatore Coco[75], Andromachi Gkoulia[44], Alfonso A. Ramos-Esplà[76], Anna Maria Mannino[77], Desirèe Dimichele[5,9,78], Simone Spinelli[4], Elisa Arroyo Martínez[79], Andrea Tursi[80], Annalisa Capasso[81], Georgios Christidis[82], Michele Solca[9,134], Mouna Rifi[83], Radhouan Belgacem El Zrelli[84], Ramla Bouhlel[10], Riccardo Scalera[85], Ada Bandi[86], Anna Reboa[86], Antonios Mazaris[87], Hocein Bazairi[88,89], Marco Bertolino[86], Mohamed Selfati[90], Benedetto Sirchia[91], Constantinos G. Georgiadis[92], Donat Petricioli[93], Eleonora Macaluso[91], Enrico D'Aniello[94], Nikolaos Fotiadis[82], Rifat Tezel[54], Slavica Petović[95], Tatjana Bakran-Petricioli[96], Valentina Pitacco[4], Alberto Colletti[41,97], Bilel Bensari[98], Lamia Bahbah[98,99], Salvatore D'Aniello[100], Spiros Psailas[101], Stefanos Kalogirou[102], Tarik Kaikai[88], Aitor Forcada[103], Alessandro Manfredonia[40], Antonella Bottalico[80], Carlos Valle[103], Coline Bedoret[104], Dimitrios Damalas[105], Domenico Sacco[106,107], Ernesto Mollo[108], Foteini Iatrou[102], Frederique Cerqueira[109], Frédérique Viard[109], Guido Villani[110], Mathieu Foulquié[111,112], Michele Luca Geraci[68], Miljan Šiško[4], Oscar Ocaña[113], Renaud Dupuy de la Grandrive[114], Sergio Vitale[68], Tullio Rossi[115], Ziad Samaha[116], Ricardo Aguilar[117], Helena Alvarez[117], Selma Menabit[3], Ana Bianca Pavel[3], Annalisa Azzola[86], Arthur R. Bos[118], Athanasios Anastasiadis[119], Jorge Blanco[117], Christophe Lejeusne[104], Daniela Coppola[120], Dominique Davoult[121], Daniel Faget[122], Danilo Scannella[123], Erica Keppel[124], Fabio Falsone[68], Fortunato Palma Esposito[120], Silvia García[117], Giulia Furfaro[5,9,41], Helmut Zibrowius[125], Ibrahim Iziani[126], Iro Anastasopoulou[102], Konstantinos Kavakakis[102], Leonardo Scavino[127], Lorenzo Merotto[128], Maria Yankova[31], Monica Montefalcone[41,86], Neža Leban[4], Nicola Ungaro[129], Paolo Mariottini[130], Allison L. Perry[117], Petros Stavrou[131], Valerio Mazzella[41,132], Violin Raykov[133], Jasmine Ferrario[75], Caterina Stamouli[82], Apostolos Siapatis[82], Emmanouela Vernadou[27], Claudia Bommarito[12], Giacomo Milisenda[12], Emanuele Somma[12], Stefania Russo[12], Stelios Katsanevakis[1]

[1]*Department of Marine Sciences, University of the Aegean, Lofos Panepistimiou, 81100 Mytilene, Greece;* [2]*Institute of Marine Biological Resources and Inland Waters, Hellenic Centre for Marine Research (HCMR), 19013 Anavissos, Greece;* [3]*National Institute for Research and Development on Marine Geology and Geoecology - GeoEcoMar, Bucharest, Romania;* [4]*Marine Biology Station Piran, National Institute of Biology, Fornače 41, 6330 Piran, Slovenia;* [5]*Consorzio Nazionale Interuniversitario per le Scienze del Mare (CoNISMa), U.L.R. di Lecce, Campus Ecotekne, strada provinciale Lecce-Monteroni, 73100, Lecce, Italy;* [6]*Instituto Español de Oceanografía (IEO-CSIC). Centro Oceanográfico de Baleares, Muelle de poniente s/n, 07015, Palma de Mallorca, Spain;* [7]*Biological and Environmental Sciences and Engineering (BESE), Red Sea Research Center (RSRC), King Abdullah University of Science and Technology (KAUST), Thuwal 23955-6900, Saudi Arabia;* [8]*Laboratorio de Biología Marina, Facultad de Biología, Universidad de Sevilla, Avenida Reina Mercedes, Sevilla, 41012, Spain;* [9]*Department of Biological and Environmental Sciences and Technologies (DiSTeBA) - Via Monteroni, 165 - 73100 Lecce, Italy - University of Salento;* [10]*Association Notre Grand Bleu (NGB, NGO), QRHM+566، Résidence Kairouan Cap marina Monastir, Monastir 5000, Tunisia;* [11]*Department of Biological, Geological and Environmental Sciences, University of Catania, Corso Italia, 57 – 95129 and Via Androne, 81 - 95124, Catania, Italy;* [12]*Stazione Zoologica Anton Dohrn, Department of Integrative Marine Ecology (EMI), Sicily Marine Center, Lungomare Cristoforo Colombo, 4521, 90149, Palermo, Italy;* [13]*University School for Advanced Studies IUSS Pavia, Pavia, Italy;* [14]*Department of Chemical, Biological, Pharmaceutical and Environmental Sciences, University of Messina, Via Stagno d'Alcontres 31 – 98166 Messina, Italy;* [15]*University of Manouba, High Institute of Biotechnology of Sidi Thabet, BiotechPôle, BP-66, 2020 Sidi Thabet, Ariana, Tunisia;* [16]*University of Tunis El Manar, Faculty of Sciences of Tunis, Laboratory of Diversity, Management and Conservation of Biological Systems, LR18ES06, Tunis, Tunisia;* [17]*Department of Marine Biology, High Institute of Marine Research, Latakia University, Latakia, Syria;* [18]*ANISN, Associazione Nazionale Insegnanti Scienze Naturali, Naples, Italy;* [19]*Department of Marine Biology, Faculty of Sciences, Lebanese University, Hadath, Beirut, Lebanon;* [20]*Hellenic Centre for Marine Research (HCMR), Institute of Oceanography (IO), Anavyssos 19013, Attica, Greece;* [21]*University of Kyrenia, Cyprus;* [22]*Marine and Environmental Research (MER) Lab, 4533, Limassol, Cyprus;* [23]*Department of Biology, FAS, American University of Beirut, Lebanon;* [24]*Oceanography Malta Research Group, Department of Geosciences, University of Malta, Msida, Malta MSD 2080, Malta;* [25]*Italian Institute for Environmental Protection and Research (ISPRA), Lungomare Cristoforo Colombo n.4521, Località Addaura, 90149 Palermo, Italy;* [26]*Department of Integrative Marine Ecology (EMI), Stazione Zoologica Anton Dohrn - National Institute of Marine Biology, Ecology and Biotechnology, Genoa Marine Centre (GMC), Villa del Principe, Piazza del Principe 4, 16126 Genova, Italy;* [27]*Hellenic Centre for Marine Research (HCMR), Institute of Marine Biology, Biotechnology and Aquaculture (IMBBC), 71500 Heraklion, Greece;* [28]*Instituto Mediterráneo de Estudios Avanzados (UIB-CSIC), C/ Miquel Marques 21, 07190, Esporles, Islas Baleares, Spain;* [29]*Department of Environment, Faculty of Environment, Ionian University, 29100 Zakynthos, Greece;* [30]*Dipartimento di Scienze e Tecnologie Biologiche e Ambientali (DiSTeBA), Campus Ecotekne, Strada Provinciale Lecce-Monteroni, 73100 Lecce, Italy;* [31]*Institute of Oceanology - Bulgarian Academy of Sciences,Varna, Bulgaria;* [32]*Conservation Biology Research Group, Department of Biology, University of Malta, Msida, Malta;* [33]*Institute of Fish Resources - Agricultural Academy, Varna, Bulgaria;* [34]*Faculty of Natural and Agricultural Sciences, Department of Natural Sciences Ovidius University of Constanta, Constanţa, Romania;* [35]*Center for Advanced Studies of Blanes (CEAB, CSIC), Carrer Accés Cala Sant Francesc, 14, 17300 Blanes, Girona, Spain;* [36]*Department of Biology, Faculty of Biology, "Alexandru Ioan Cuza" University of Iași, 700507 Iași, Romania;* [37]*Aqua Stories; via Vampolieri, 25/31 int. 27, 95022 Aci Catena (Catania), Italy;* [38]*CNR/ISAFoM (National Research Council of Italy/Institute for Agricoltural and Forestry System in the Mediterranean) via Empedocle, 58, 95125 Catania, Italy;* [39]*Frederick University, Limassol, Cyprus;* [40]*Department of Integrative Marine Ecology (EMI), Stazione Zoologica Anton Dohrn, Villa Comunale, I-80121 Naples, Italy;* [41]*NBFC, National Biodiversity Future Centre - Palermo, Piazza Marina 61, I-90133 Palermo, Italy;* [42]*Department of Biology, University of Malta, Msida, Malta;* [43]*Departamento de Botánica y Fisiología Vegetal, Facultad de Ciencias, Campus de Teatinos s/n, 29080 Málaga, Spain;* [44]*CMMI - Cyprus Marine & Maritime Institute, 6023, Larnaca, Cyprus;* [45]*Department of Biology, Faculty Engineering and Natural Sciences, Manisa Celal Bayar University Muradiye, Manisa 45140, Türkiye;* [46]*SAP/RAC, Boulevard du Leader Yasser Arafat, B.P. 337 - 1080 Tunis Cedex – Tunisie;* [47]*School of Biological Sciences, University of Aberdeen, Aberdeen AB24 3UU, Scotland, UK;* [48]*Institute of Earth Systems, University of Malta, Msida, Malta;* [49]*Oceanography Center, University of Cyprus, 1 Panepistimiou Av., 2109 Aglandjia, Nicosia, Cyprus;* [50]*Department of Fisheries and Marine Research (DFMR), Ministry of Agriculture, Rural Development and the Environment of the Republic of Cyprus, 101 Vithleem Street, 2033 Strovolos, Nicosia, Cyprus;* [51]*Departamento de Ecología e Hidrología, Universidad de Murcia, Murcia, Spain;* [52]*Dipartimento di Biologia, Roma Tor Vergata, 00133 Roma, Italy;* [53]*IUCN ROWA , Amman 11194 Jordan;* [54]*Mugla Sıtkı Koçman University Faculty of Fisheries, Mugla/Türkiye;* [55]*IMC - International Marine Centre, Loc. Sa Mardini, Torregrande - 09170 Oristano, Italy;* [56]*Institute for Marine Biological Resources and Biotechnology National Research Council of Italy (IRBIM-CNR) Largo Fiera della Pesca, 2, 60125 Ancona AN, Italy;* [57]*Italy Ente Fauna Marina Mediterranea, Avola 96012, Siracusa, Italy;* [58]*Department of Earth and Marine Sciences (DiSTeM), University of Palermo, Via Archirafi 20-22, 90123 Palermo, Italy;* [59]*Department of Biology, University of Crete, 70013, Heraklion, Greece;* [60]*Faculty of Fisheries, Muğla Sıtkı Koçman University, Muğla, Turkey;* [61]*Department of Biology, Stazione Idrobiologica Umberto D'Ancona, University of Padova, Chioggia, Venice, Italy;* [62]*Department of Biology, University of Padova, Via U. Bassi 58/B, 35121, Padova, Italy;* [63]*Astrolabe-Marine Research, Thessaloniki, 55132, Greece;* [64]*Oceanides-Institute of Marine Research & Education, Kalymnos Island, 85200, Greece;* [65]*Department of Biology, University of Naples "Federico II", Via Cintia 26, I-80126 Naples, Italy;* [66]*Conservation Biology Research Group, Department of Biology, University of Malta, MSD2080 Msida, Malta;* [67]*Eser Deniz Ecological, Environmental Company, Technocity of Manisa Celal Bayar University, Muradiye-Manisa, Türkiye;* [68]*Institute for Marine Biological Resources and Biotechnologies (IRBIM), National Research Council (CNR), Mazara del Vallo, Italy;* [69]*National Institute for Scuba Diving, Zeytouna bay, Beirut;* [70]*Arpa Sicilia UOC Area Mare, Lungomare Cristoforo Colombo 4521, Loc. Addaura, 90149 Palermo, Italy;* [71]*Istanbul University, Faculty of Science, İstanbul/Türkiye;* [72]*Bandirma Onyedi Eylul University, Maritime Vocational School, Balıkesir, Türkiye;* [73]*Department of Biology, Faculty of Marine Sciences, Instituto Universitario de Investigación en Estudios Ambientales y Recursos Naturales IUNAT, Universidad de Las Palmas de Gran Canaria, 35017 Las Palmas, Spain;* [74]*Ege University, Faculty of Fisheries, Department of Hydrobiology, 35100 Bornova, İzmir, Türkiye;* [75]*Department of Earth and Environmental Sciences, University of Pavia, Italy;* [76]*Department Marine Sciences and Applied Biology, University of Alicante, Alicante, Spain;* [77]*Department of Biological, Chemical and Pharmaceutical Sciences and Technologies, University of Palermo, 90123 Palermo, Italy;* [78]*Dipartimento di Scienze della Terra e del Mare (DiSTeM), Via Archirafi 22, 90123 Palermo, Italy;* [79]*Department of Marine Science and Applied Biology, University of Alicante, 03690 Alicante, Spain;* [80]*Department of Biosciences, Biotechnologies and Environment, University of Bari Aldo Moro, Via E. Orabona 4, 70125, Bari, Italy;* [81]*Department of Biology, University of Padova, Via Ugo Bassi 58/B, 35131, Padova, Italy;* [82]*Hellenic Centre for Marine Research, Institute of Marine Biological Resources and Inland Waters, 576 Vouliagmenis Avenue, Argyroupolis, 16452 Athens, Greece;* [83]*Laboratoire de Recherche Ecosystèmes et Ressources Aquatiques, Institut National Agronomique de Tunisie, 43 Avenue Charles Nicolle, 1082 Tunis, Tunisia;* [84]*Independent Researcher & International Environmental Consultant, 31400 Toulouse, France;* [85]*Department of Science, Roma Tre University, Viale Guglielmo Marconi 446, 00146 Rome, Italy;* [86]*Department of Earth, Environment and Life Sciences (DiSTAV), University of Genoa, Corso Europa 26, 16132 Genova, Italy;* [87]*Department of Ecology, School of Biology, Aristotle University of Thessaloniki, Thessaloniki, Greece;* [88]*Mohammed V University in Rabat, Faculty of Sciences, Laboratory of Biodiversity, Ecology and Genome, 4 Avenue Ibn Battouta, B.P. 1014 RP, Rabat, Morocco;* [89]*Natural Sciences and Environment Research Hub, University of Gibraltar, Europa Point Campus, Gibraltar;* [90]*Laboratoire Santé, Environnement et Biotechnologie, Faculté des Sciences Aïn Chock, Hassan II University of Casablanca, B.P 5366 Maarif, 20100 Casablanca, Morocco;* [91]*Arpa Sicilia UOC Area Mare, Lungomare Cristoforo Colombo 4521, Loc. Addaura, 90149 Palermo, Italy;* [92]*Benthic Ecology & Technology*

Laboratory, Fisheries Research Institute (ELGO-DIMITRA), 64007, Nea Peramos, Kavala, Greece; [93]*D.I.I.V. for marine, freshwater and subterranean ecology Sali IV 2, HR-23281 Sali, Croatia;* [94]*Department of Biology and Evolution of Marine Organisms, Stazione Zoologica Anton Dohrn (SZN), Naples, Italy;* [95]*Institute of Marine Biology, University of Montenegro, Kotor, Montenegro;* [96]*University of Zagreb Faculty of Science, Department of Biology, Rooseveltov trg 6, HR-10000 Zagreb, Croatia;* [97]*Department of Biology, University of Naples Federico II, Naples, Italy;* [98]*Biological Oceanography and Marine Environment Laboratory (LOBEM), Faculty of Biological Science, University of Science and Technology, Houari Boumediene. B.P 32 El Alia 16111 Bab Ezzouar, Algeria;* [99]*National Higher School of Marine Sciences and Coastal Management (ENSSMAL), 16320 Algiers, Algeria;* [100]*Department of Biology and Evolution of Marine Organisms (BEOM), Stazione Zoologica Anton Dohrn, Villa Comunale, 80121 Naples, Italy;* [101]*Corfu Scubanauts Diving Center, Kontokali Bay hotel, 49100 Corfu, Greece;* [102]*Agricultural University of Athens, School of Animal Sciences, Department of Animal Science, Laboratory of Applied Hydrobiology, Iera odos 75, 11855 Athens, Greece;* [103]*Department Marine Sciences and Applied Biology, University of Alicante, Alicante, Spain;* [104]*Aix Marseille Univ, CNRS, IRD, Avignon Univ, IMBE, UMR 7263, Station Marine d'Endoume, Marseille, France;* [105]*Hellenic Centre for Marine Research, Institute of Marine Biological Resources & Inland Waters, Former US Base at Gournes, P.C. 71500 municipality of Hersonissos, P.O. Box 2214 T.K. 71003 Heraklion Crete, Greece;* [106]*Department of Life and Environmental Sciences, Polytechnic University of Marche, Ancona, Italy;* [107]*Stazione Zoologica Anton Dohrn, Naples, Italy;* [108]*Institute of Biomolecular Chemistry, National Research Council of Italy, Pozzuoli, Naples 80078, Italy;* [109]*ISEM, Univ Montpellier, CNRS, EPHE, IRD, Montpellier, France;* [110]*Department of Research Infrastructures for Marine Biological Resources, Stazione Zoologica Anton Dohrn, Napoli, Italy;* [111]*Institut Océanographique Paul Ricard, Ile des Embiez, 83140 Six-Fours-les-Plages, France / and Université de Toulon, Aix Marseille Univ, CNRS, IRD, MIO, 83130 La Garde, France;* [112]*Université de Toulon, Aix Marseille Univ, CNRS, IRD, MIO, 83130 La Garde, France;* [113]*Museo del Mar, Ceuta, Spain;* [114]*Maison de la mer - Aire Marine Protégée de la côte agathoise, Rue de la Capitainerie, 34300 Cap d'Agde – France;* [115]*Animate your Science, Adelaide, Australia;* [116]*Lebanon project office (IUCN, ROWA), Beirut, Lebanon;* [117]*Oceana, Spain;* [118]*Biology Department, The American University in Cairo, P.O. Box 74, New Cairo 11835, Egypt;* [119]*Cretaquarium, Hellenic Center for Marine Research, P.O. Box 2214, 71003 Heraklion, Crete, Greece;* [120]*Department of Ecosustainable Marine Biotechnology, Stazione Zoologica Anton Dohrn, Via Ammiraglio Ferdinando Acton 55, 80133 Naples, Italy;* [121]*Sorbonne Université, CNRS, UMR 1744, Station Biologique, Roscoff, France;* [122]*Aix Marseille Univ, CNRS, IRD, Avignon Univ, TELEMME, UMR 7263, Station Marine d'Endoume, Marseille, France;* [123]*Institute for Marine Biological Resources and Biotechnology (IRBIM), National Research Council – CNR, Mazara del Vallo, Italy;* [124]*Smithsonian Environmental Research Center, Edgewater, MD, USA;* [125]*Le Corbusier 644, 280 Boulevard Michelet, 13008 Marseille, France;* [126]*Mohammed V University in Rabat, Faculty of Sciences, Laboratory of Biodiversity, Ecology and Genome, 4 Avenue Ibn Battouta, B.P. 1014 RP, Rabat, Morocco;* [127]*Aix Marseille Univ, CNRS, TELEMME, UMR 7303, Aix-en-Provence, France;* [128]*Portofino Marine Protected Area, Viale Rainusso 1, 16038 Santa Margherita Ligure, Italy;* [129]*Apulian Regional Agency for the Environmental Prevention and Protection, 70126, Bari, Italy;* [130]*Dipartimento di Scienze, Università 'Roma Tre', Viale G. Marconi 446, I-00146 Rome, Italy;* [131]*Department of Fisheries and Marine Research (DFMR), Ministry of Agriculture, Rural Development and the Environment of the Republic of Cyprus, 2033, Nicosia, Cyprus;* [132]*Department of Integrative Marine Ecology (EMI), Stazione Zoologica Anton Dohrn, Ischia Marine Centre, Ischia, Naples, 80077, Italy;* [133]*Institute of Oceanology - Bulgarian Academy of Sciences, 40 Parvi mai str, 9000 Varna, Bulgaria;* [134]*Marine Biology Museum "Pietro Parenzan", Via A. Vespucci, 13/17, 73010 Porto Cesareo (LE), Italia.*

Corresponding author: Jennifer C.A. Pistevos (jennie@aegean.gr)

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Abstract

To enhance regional knowledge of invasive alien species (IAS), cryptogenic, and neonative species, this study compiles previously unpublished records from the Mediterranean and Black Seas via contributions from 227 marine experts. The open-access dataset includes 7,422 records from 21 countries, representing 275 taxa: 222 Animalia, 44 Plantae, and 9 Chromista. Seventy-two percent of records were collected in 2021–2024 while most reports were from Lebanon, Greece, Cyprus, Italy, and Romania. The most frequently recorded species were *Siganus luridus*, *S. rivulatus*, *Callinectes sapidus*, *Caulerpa cylindracea* and *Parupeneus forsskali*. Several records are biogeographically notable: basin-level firsts include *Dynoides amblysinus* and *Symplegma rubra*, and the first confirmed occurrence of *Malmgrenia mcintoshi*. National firsts include *Mesanthura* cf. *romulea*, *Mitrella psilla*, and *Syllis ergeni* (Algeria); *Exogone breviantennata*, *Jassa marmorata*, *Leodice antennata*, and *Syllis schulzi* (Cyprus); *Botrylloides niger* and *Polyandrocarpa zorritensis* (Greece); *Microcosmus exasperatus, Phallusia nigra*, and *Stypopodium schimperi* (Italy); *Chaetozone corona* (Slovenia); *Pseudodiaptomus marinus* and *Symplegma brakenhielmi* (Spain); *Aquilonastra burtoni*, *Siphonaria crenata*, and *Sphaeroma walkeri* (Syria) and *Distaplia bermudensis* (Tunisia). In the Black Sea, *Hydroides dianthus*, *Ferosagitta* sp., and a zoea III of *Palaemon macrodactylus* are newly documented from Romanian waters. Beyond listing records, this compilation supports spatiotemporal analyses relevant to management. The sharp post-2021 increase reflects both species spread and intensified surveys, and habitat-labelled records include harbours/marinas and shellfish aquaculture settings, consistent with pathway hotspots. By revealing where and when detections concentrate—and which taxa dominate—these data can guide early detection, risk prioritization, and targeted biosecurity, while identifying under-reported coastlines.

Key words: non-native species, marine bioinvasions, inventories, distribution, geo-referenced records, Mediterranean Sea

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Introduction

Biological invasions represent one of the most significant and fast-growing threats to marine biodiversity worldwide, often leading to the disruption of native ecosystems and ecosystem services (Molnar et al. 2008; Katsanevakis et al. 2014; Tsirintanis et al. 2022). The Mediterranean Sea, despite its relatively small size, has emerged as a hotspot for marine invasive alien species (IAS) and hosts the highest number of introductions among European seas (Galil et al. 2014; Thomsen et al. 2016; Seebens et al. 2016; Zenetos et al. 2022a). This phenomenon is facilitated by a combination of intense anthropogenic pressures, including maritime traffic, aquaculture, and the Suez Canal, as well as regional warming that enhances the suitability of conditions for thermophilic species (Zenetos et al. 2012; Schroeder et al. 2016; Albano et al. 2021). Due to its geopolitical complexity, being bordered by 22 countries with varying policies and capacities for marine monitoring, the Mediterranean faces significant challenges in implementing cohesive biosecurity and biodiversity monitoring frameworks (Zenetos and Galanidi 2020). Meanwhile, the neighbouring Black Sea has experienced a steady rise in alien species introductions over the past five decades, driven both by the secondary spread of IAS from the Mediterranean via the Dardanelles and the Bosporus, and by primary introductions, mainly through shipping (Băncilă et al. 2022; Ragkousis et al. 2023).

Increasing attention is also being paid to neonative species—those undergoing climate-driven natural range expansions—which can also exert ecological pressures (Essl et al. 2019; Azzurro et al. 2022; Virgili et al. 2025). Whether alien or neonative, their expanding presence continues to drive processes such as tropicalization in the Mediterranean (Bianchi and Morri 2003; Peleg et al. 2020) and Mediterranization in the Black Sea (Kideys et al. 2000; Eyuboglu 2022).

Despite the ecological significance of these changes, a large volume of valuable distribution data remains unpublished or inaccessible, often stored in personal archives or institutional databases (Katsanevakis et al. 2020). This gap hinders comprehensive assessments of invasion dynamics, species trends, and the development of management responses under frameworks such as the European Marine Strategy Framework Directive (MSFD) and the Barcelona Convention/UNEP/MAP (Tsiamis et al. 2019; 2021). The need for frequent updates to alien species inventories has led to coordinated efforts to collect and disseminate such data via collaborative networks of marine scientists.

This paper builds upon two previous collaborative efforts to document unpublished records of alien species – including IAS in the region. The first effort compiled 5,376 records of 239 alien and cryptogenic marine taxa from the Mediterranean Sea, contributed by 126 scientists across 16 countries (Katsanevakis et al. 2020). A subsequent expansion incorporated

both the Mediterranean and Black Seas, collecting 12,649 records of 247 taxa, including neonative species, and substantially enriching the spatial and temporal resolution of available data (Ragkousis et al. 2023). The present study represents the third iteration of this initiative and aims to further strengthen regional inventories by collating newly gathered and previously unpublished records of NIS, cryptogenic, and neonative species. In doing so, it contributes to a growing open-access dataset that is essential for tracking bio-invasion dynamics and supporting efforts to monitor, manage, and mitigate the impacts of alien species in the region.

Dataset compilation

Following the approach of the previous two iterations of this initiative, the call for contributions was circulated by direct e-mail invitation to known marine experts across Mediterranean and Black Sea countries, using a standardized Excel template to harmonize submissions. Compared to the template used in our previous studies, we revised several terms to better align with the Darwin Core standards (http://rs.tdwg.org/dwc/doc/em/). Contributors were also invited to suggest additional expert contacts, enabling further targeted outreach through professional networks. Records originating from third parties (including citizen-science platforms) were included only when the identification was validated by a marine expert, typically supported by photographic documentation and/or reliable observer provenance as reported in Supplementary material Table S1. No records derived from fisher questionnaires were included in the present compilation.

Overall, a total of 227 contributors from 21 different countries provided data, resulting in a dataset of 7,422 individual records (see Table S1). Each entry in the spreadsheet corresponds to a unique observation, documented with spatial and temporal details. Key information included species name, biogeographic status, geographic coordinates (latitude and longitude), country of observation, year, name of the identifier, observation type, and documentation method.

Biogeographic status followed established references—Zenetos et al. (2022b) for the Mediterranean and Băncilă et al. (2022) for the Black Sea—supplemented by species-specific literature when classification was uncertain. Four categories were used to define status: "Alien" (as per Essl et al. 2018), "Neonative" (*sensu* Essl et al. 2019), "Cryptogenic" (unknown origin; Carlton 1996; Essl et al. 2018), and "Data deficient" (for taxa whose biogeographic status could not be determined due to lack of data; Essl et al. 2018). Neonatives were included as a distinct status category to capture climate-driven, natural range expansions (*sensu* Essl et al. 2019), and to avoid conflating such range recognition with human-mediated introductions. In the Mediterranean and Black Sea, neonatives may expand through natural corridors (e.g., through the Gibraltar Strait or into the Black Sea via

the Dardanelles–Sea of Marmara system), as discussed in the previous iteration of this initiative (Ragkousis et al. 2023). However, we do not attempt to reconstruct entry routes or quantify range-shift trajectories for each neonative taxon, as this would require standardized baseline distributions and longitudinal time-series beyond the scope of this record-mobilization dataset.

Record verification and inclusion criteria

The dataset comprises records submitted by participating marine experts (227 contributors from 21 countries). Only records with the required minimum information were retained (taxon name, biogeographic status, coordinates/country, year, identifier, observation type, and documentation method). "Observation type" describes the verification pathway and includes (i) direct expert visual identification (e.g., in situ surveys, including validated photographic material submitted to the expert by collaborators) and (ii) specimen-based identification following collection by scientific personnel, typically confirmed through morphological examination in the laboratory and, where necessary/available, DNA-based confirmation.

Optional fields were used when provided to capture additional context (e.g., specific or estimated observation date, depth, habitat type, estimated abundance or coverage, and supporting notes). The dataset is dominated by conspicuous and readily identifiable alien taxa; for biogeographically notable records (e.g., basin-level or national firsts) and/or taxa with higher identification uncertainty, supporting details are provided within the relevant species accounts. Where the available evidence was insufficient to confidently resolve taxonomic identity and/or biogeographic status, records were treated conservatively using the dataset categories (e.g., cryptogenic).

Taxonomic coverage

A total of 275 distinct taxa were recorded in the dataset, primarily Animalia (222; 80.58%), with smaller proportions of Plantae (44; 15.98%) and Chromista (9; 3.44%) (Figure 1). Within Animalia, records were dominated by Chordata (primarily fishes), followed by major invertebrate groups (notably Arthropoda and Mollusca) (Figure 1C). This dominance of conspicuous taxa—especially fishes—is consistent with previous iterations of this initiative and likely reflects both detectability and monitoring focus. Most of the records were classified as alien (6,438; 86.6%), with additional contributions from cryptogenic (752; 10.1%), neonative (84; 1.1%), and uncertain cases assigned as neonative/cryptogenic (161; 2.2%). The low proportion of neonative records (1.1%) should not be interpreted as evidence that climate-driven range expansions are rare in the region. Rather, it likely reflects the purpose and reporting bias of this expert-mobilized compilation, which was primarily designed to strengthen alien species inventories (the

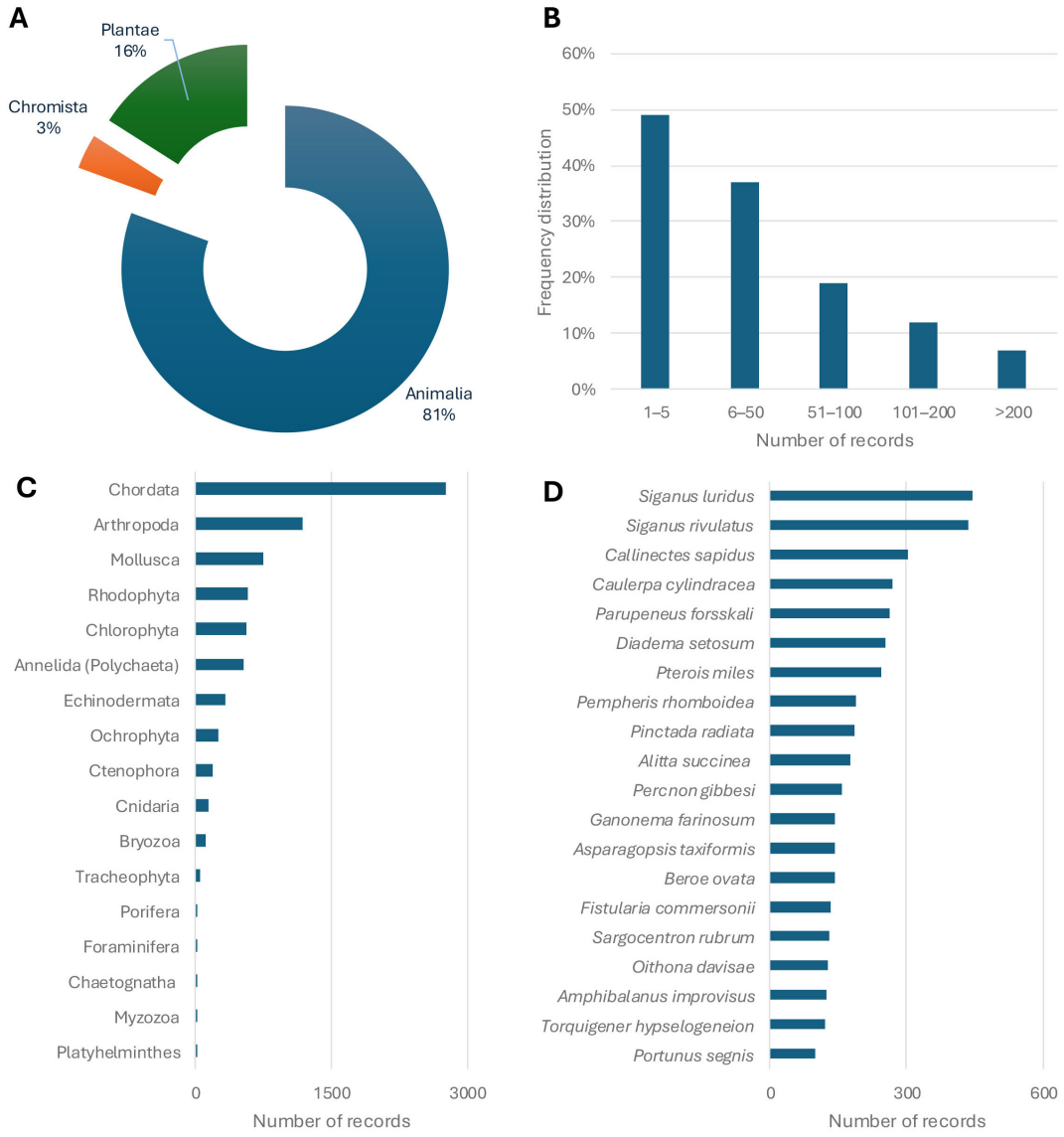

Figure 1. Summary of taxonomic information from the dataset: (A) proportion of records grouped by kingdom; (B) histogram showing the distribution of record counts per species; (C) breakdown of records by phylum; (D) species-level record frequency for the 20 taxa with the highest number of observations.

first iteration of this initiative targeted alien and cryptogenic species specifically) and therefore tends to elicit and prioritize reports of alien taxa. In addition, identifying a species as neonative requires stronger contextual information (baseline distribution knowledge and confidence that the expansion is natural rather than human-mediated), which increases uncertainty and can lead to conservative categorization. This is reflected in the additional 2.2% of records classified as "neonative/cryptogenic", i.e., cases where species status is uncertain.

Approximately 77% of the reported taxa come from five countries—Lebanon, Greece, Cyprus, Italy, and Romania. The five species with the highest number of records were *Siganus luridus* (Rüppell, 1829) (447), *Siganus rivulatus* Forsskål & Niebuhr, 1775 (435), *Callinectes sapidus* Rathbun, 1896 (303), *Caulerpa cylindracea* Sonder (269), and *Parupeneus forsskali* Fourmanoir & Guézé, 1976 (263).

Spatial and temporal coverage

The temporal span of the dataset ranges from 1973 to 2025. A substantial majority of records (approximately 72%) were collected between 2021 and 2024, with the highest contributions in 2024 (1,859 records), followed by 2023 (1,665), and 2022 (1,286) (Figure 2A). The dataset includes records from 21 countries, including 6,384 records from the Mediterranean Sea and 1,046 from the Black Sea (Figure 2B). The species recorded in the highest number of countries were *Callinectes sapidus* Rathbun, 1896 (10), *Bursatella leachii* Blainville, 1817 (8), *Fistularia commersonii* Rüppell, 1838 (8), *Amathia verticillata* (delle Chiaje, 1822) (7), *Caulerpa cylindracea* Sonder (7), *Pinctada radiata* (Leach, 1814) (7), and *Pterois miles* (Bennett, 1828) (7) (Figure 2C). Establishment status was not evaluated systematically in this study because it is scale-dependent and not uniquely attributable at the record level; readers are referred to the most recent basin-level assessments for the Mediterranean and Black Sea (Zenetos et al. 2022b; Magliozzi et al. 2025).

The spatial distribution of records remains uneven across the region (Figure 3A), influenced by species occurrence patterns, variability in sampling effort, and contributor distribution. In addition to uneven sampling effort and data-sharing, spatial gaps can also reflect uneven availability of taxonomic expertise among countries and taxa; absence of records should therefore not be interpreted as evidence of absence. A substantial portion of the older unpublished records in our dataset originated from Lebanon, which also accounts for the highest overall number of records (Figure 2B). The inclusion of historical observations (1970s–1990s) does not reflect a timeline inconsistency, but rather the purpose of this initiative to mobilize and georeference information that is often unpublished or inaccessible (e.g., retained in personal archives or institutional databases). As noted in previous iterations, alien-species observations are often published only when they constitute "first records", whereas many subsequent detections (and older observations) remain scattered and unpublished, despite their value for reconstructing invasion trajectories and temporal dynamics. In addition, some historical findings—particularly from Lebanon—have been mentioned previously without sufficient spatial detail for reuse; the present compilation provides the missing georeferenced locality information where available and includes backdated historical records not previously published (e.g., the three macroalgae reported as backdated first country records for Lebanon).

High reporting volumes were also observed in Greece, Cyprus, Italy, and Romania. Conversely, record contributions were scarce in Albania, Egypt, Georgia, Ukraine, France, and parts of North Africa, with only isolated records submitted from these regions (Figure 3A). This pattern likely reflects differences in sampling effort and data sharing rather than the actual absence or low presence of alien and other species targeted in the present study. The combination of the mapping points originating from all the three studies

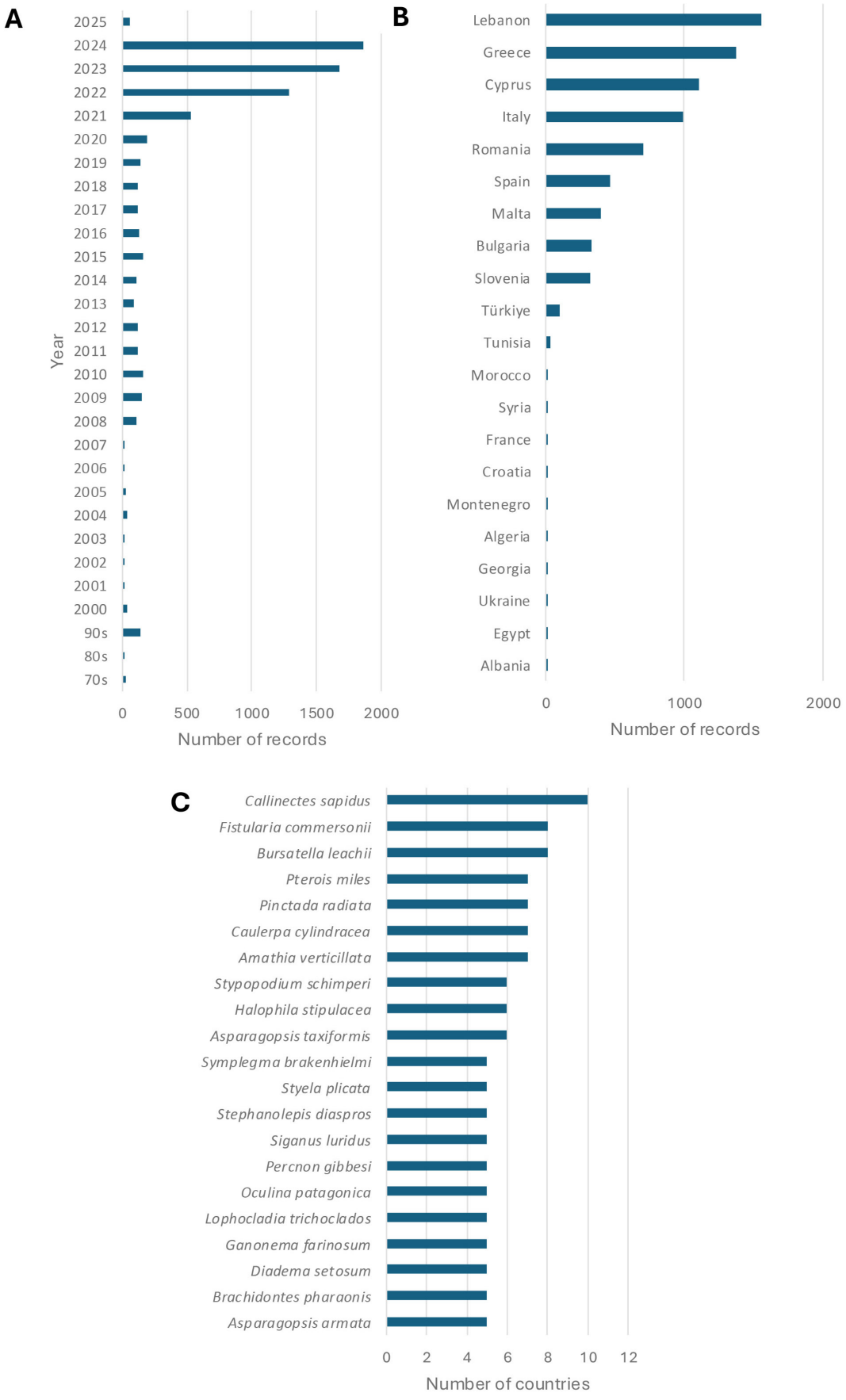

Figure 2. Spatial and temporal distribution characteristics of the NIS dataset: (A) yearly distribution of recorded observations; (B) number of records contributed by each country; (C) number of countries reporting each species (shown for species recorded in five or more countries).

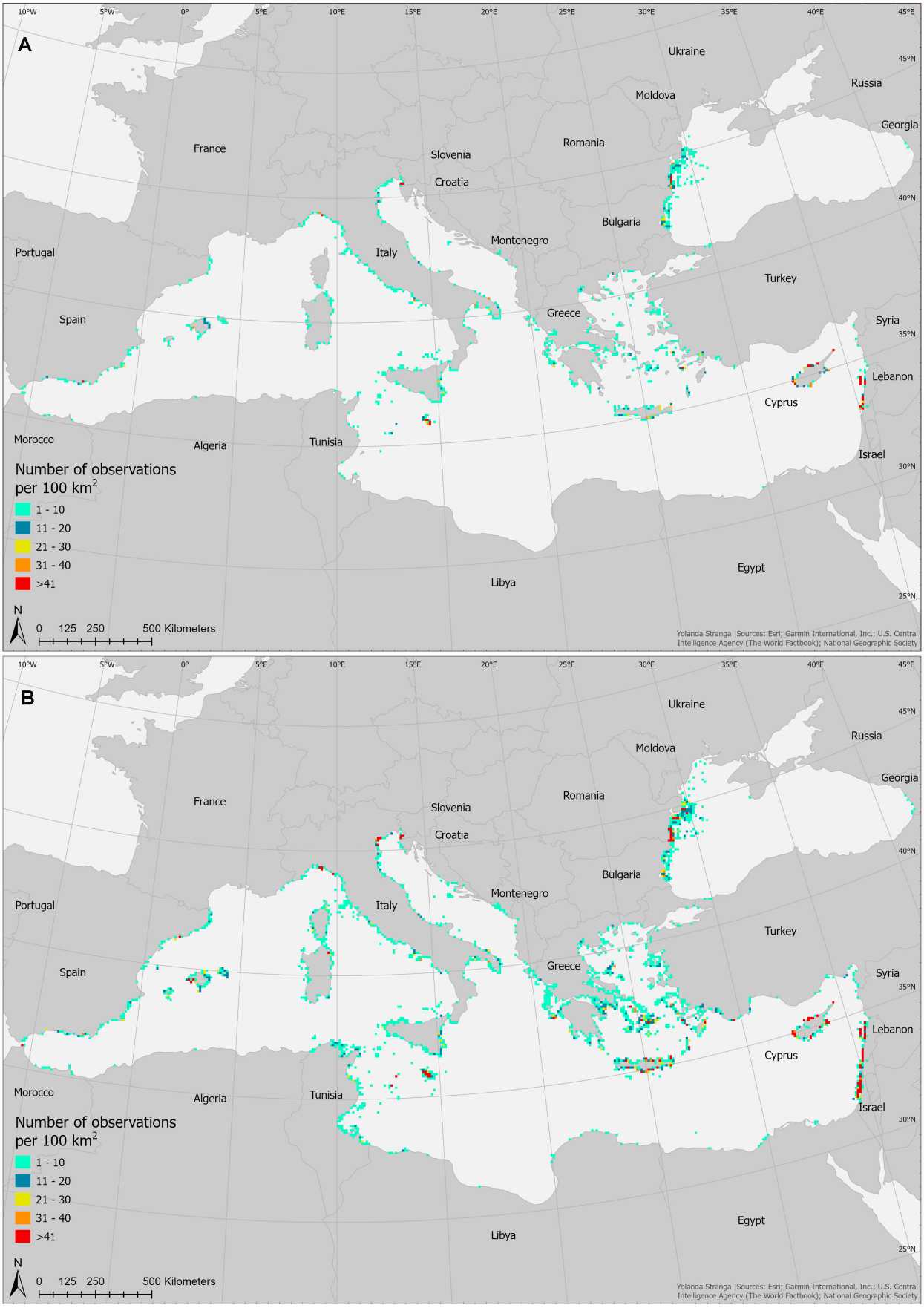

Figure 3. (A) number of alien, cryptogenic, and neonative records per 10 km × 10 km grid cell, included in the dataset of this study; (B) cumulative number of alien, cryptogenic and neonative records with the same grid cell from this study plus Katsanevakis et al. (2000) and Ragkousis et al. (2023).

(Katsanevakis et al. 2020; Ragkousis et al. 2023; present data) highlights both the extent of species spread and the areas where fewer records are evident (Figure 3B), which may reflect either information gaps or genuinely lower presence of IAS (e.g., parts of Sardinia and the Tyrrhenian coastline).

Remarkable new records of alien species

This dataset includes several records of particular significance, including first basin-level, national, and sub-regional occurrences, as well as confirmations of species establishments. Notably, the isopod *Dynoides amblysinus* Pillai, 1954, is recorded for the first time in the Mediterranean Sea, based on material collected from the Port of Catania (Italy). Other basin-level firsts include the colonial ascidian *Symplegma rubra* Monniot, 1972, newly observed in the Mediterranean (off the coast of Tunisia), and the polychaete *Malmgrenia mcintoshi* (Tebble & Chambers, 1982), which is here reported for the first time from the Ionian Sea, confirming its presence in the central Mediterranean Basin. *Paracalanus tropicus* Andronov, 1977 is recorded for the first time in Mediterranean waters in Algeciras Harbour, Spain. *Stypopodium schimperi* (Kützing) Verlaque & Boudouresque, a brown alga previously limited to the eastern Mediterranean, is reported for the first time in southern Italy, specifically from Lampedusa and Pantelleria in the Sicily Strait.

Several records represent first observations at the national level. From Syria, the starfish *Aquilonastra burtoni* (Gray, 1840), the limpet *Siphonaria crenata* Blainville, 1827, and the isopod *Sphaeroma walkeri* Stebbing, 1905 are reported for the first time. From Cyprus, new records include the polychaetes *Exogone breviantennata* Hartmann-Schröder, 1959, *Leodice antennata* Savigny in Lamarck, 1818 and *Syllis schulzi* (Hartmann-Schröder, 1960), and the amphipod *Jassa marmorata* Holmes, 1905. In Algeria, new findings include the polychaete *Syllis ergeni* Çinar, 2005, the isopod *Mesanthura* cf. *romulea* Poore & Lew Ton, 1993, and the gastropod *Mitrella psilla* (Duclos, 1840). The cirratulid polychaete *Chaetozone corona* Berkeley & Berkeley, 1941 is reported for the first time from Slovenian waters. In Tunisia, first records include the colonial ascidian *Distaplia bermudensis* Van Name, 1902, and an occurrence of *Polyandrocarpa zorritensis* Van Name, 1931 from 2021, which backdates the first published Tunisian record by three years (Melki et al. 2026). This backdating is consistent with the generally short reporting lags documented for Tunisia and the comparatively longer (often near-decadal) lags reported for ascidians (Zenetos et al. 2019; Galanidi et al. 2025). In Italy, new national or remarkable records regard the confirmation of the ascidians: *Phallusia nigra* Savigny, 1816, which was only known in DNA samples (Varella et al. 2025), *Microcosmus exasperatus* Heller, 1878, *Distaplia bermudensis* Van Name, 1902, and *Polyclinum constellatum* Savigny, 1816; the mollusc *Lamprohaminoea ovalis* (Pease, 1868); and the polychaete *Eurythoe complanata* (Pallas, 1766),

the latter four recorded in the Adriatic Sea for the first time. The ascidians *Botrylloides niger* Herdman, 1886 and *Polyandrocarpa zorritensis* (Van Name, 1931) are documented for the first time in Greece. In Spanish Mediterranean waters, the tunicate *Symplegma brakenhielmi* (Michaelsen, 1904) is here reported from Alicante port while the copepod *Pseudodiaptomus marinus* Sato, 1913 is recorded for the first time from the Algeciras Harbour, representing the westernmost known occurrence of the species in the basin.

In the Black Sea, the present dataset provides the first records for Romania of the third-stage larvae of *Palaemon macrodactylus* Rathbun, 1902, the serpulid polychaete *Hydroides dianthus* (Verrill, 1873), and a new chaetognath of the genus *Ferosagitta*. It also provides further evidence of the establishment of the hydrozoan *Podocorynoides minima* (Trinci, 1903) in Romanian coastal and offshore habitats (Mureşan et al. 2021).

Additional significant range extensions include the bryozoans *Catenicella paradoxa* Rosso, 2009 and *Tricellaria inopinata* d'Hondt & Occhipinti-Ambrogi, 1985, the former first reported from the Sicily Strait and the latter observed for the first time in Sicily and the western Ionian Sea. The bryozoan *Arbopercula angulata* (Levinsen, 1909) is newly reported from the harbour of Lerici (Liguria, Italy), representing the northernmost known occurrence in both the Tyrrhenian and western Mediterranean. The spionid polychaete *Prionospio depauperata* Imajima, 1990 is also newly recorded from the Tyrrhenian Sea (western Mediterranean), extending its range beyond its previously known distribution in the eastern and central Mediterranean (Dağlı and Çinar 2009; Ragkousis et al. 2023; Langeneck et al. 2024).

Additionally, three macroalgae and one fish are here reported as backdated first country records for Lebanon: *Codium taylorii* P.C. Silva, *Galaxaura rugosa* (J.Ellis & Solander) J.V. Lamouroux, *Padina boergesenii* Allender & Kraft, and *Terapon puta* Cuvier, 1829, based on historical records not previously published.

A recent study of the structure of marine biofouling in two fishing and recreational ports in southern Syria has revealed more established alien species. In this report, we record the presence of the starfish *Aquilonastra burtoni* (Gray, 1840), the limpet *Siphonaria crenata* Blainville, 1827, and the isopod *Sphaeroma walkeri* Stebbing, 1905 for the first time in the country.

In the following taxon-focused subsections, we briefly present selected records of particular biogeographic importance (e.g., first records for the Mediterranean/Black Sea and/or first country records) and provide the available documentation supporting their identification, following the approach used in previous iterations of this initiative.

Polychaeta (Annelida)

- *Malmgrenia mcintoshi* (Tebble & Chambers, 1982) – first record in the Ionian Sea (central-eastern Mediterranean).

A single adult specimen of the polychaete *M. mcintoshi* has been recorded in July 2006 in shallow waters (2 m depth) in the Catania harbour area (Sicily, Italy). The specimen was found settled inside a living tube of *Chaetopterus variopedatus* (Renier, 1804), on a mixed sandy-rocky bottom, affected by temporary accumulation of pyroclastic sands. Identification was confirmed by Dr. Ruth Barnich. The specimen is stored at the Senckenberg Museum Polychaete Collection (code ID sesam-248664, catalogue no. 16988), preserved in 70% ethanol.

Malmgrenia mcintoshi is generally regarded as an Atlantic–Boreal species, with a main distribution in the northeastern Atlantic extending to the Barents Sea (Marin and Antokhina 2022). In a biogeographical revision of the genus *Malmgrenia*, its presence was generically stated for the northeastern Atlantic and the Mediterranean Sea (Barnich et al. 2017), whereas Marin and Antokhina (2022) considered it currently absent from the Mediterranean.

The present record therefore represents the first documented occurrence for Italian waters and confirms presence in the Mediterranean at least in the Ionian Sea (central-eastern basin). The apparent uncertainty in its Mediterranean distribution likely reflects its symbiotic lifestyle: species of *Malmgrenia* are commensals within tubicolous macrofauna (Martin and Britayev 1998; cited here for ecological context), and *M. mcintoshi* is typically associated with large *Chaetopterus* tubes. Such ecological specificity can lead to punctiform and irregular occurrences and may contribute to historical under-detection or misidentification. At the same time, its presence in a harbour setting with moderate anthropogenic disturbance means that passive transport (e.g., of juvenile stages) cannot be excluded. Accordingly, its status in the Mediterranean is best treated as cryptogenic rather than alien.

- *Exogone breviantennata* Hartmann-Schröder, 1959 – first record in Cyprus.

The polychaete *E. breviantennata* was first reported in the Mediterranean Sea from shallow-water vegetated environments off Lebanon (Aguado and San Martín 2007) and subsequently from Türkiye (Çinar 2009). This species is morphologically very similar to the native *Exogone verugera* (Claparède, 1868) and the alien *Exogone africana* Hartmann-Schröder, 1974. It can be distinguished from *E. verugera* by the presence of a dorsal cirrus at the second chaetiger, a character shared with *E. africana*, which however has a longer proventricle. The occurrence off Cyprus represents the first record of *E. breviantennata* for this island.

- *Leodice antennata* Savigny in Lamarck, 1818 – first record in Cyprus.

The polychaete *L. antennata* (Figure S1) is a Lessepsian species currently known with certainty for Türkiye, Egypt, and Lebanon (Kurt Şahin and Çinar 2009, 2017; Dorgham and Hamdy 2015; Fortič et al. 2023). The recorded specimens represent the first report from Cyprus. A COI

sequence obtained from one specimen (GenBank accession code: PV018846) has a 99% identity with a specimen from Lebanon sequenced by Toso et al. (2024) (Figure S1).

- *Syllis ergeni* Çinar, 2005 – first record in Algeria.

The polychaete *S. ergeni* (Figure S2) is a syllid species often associated with polluted and anthropized environments in the eastern Mediterranean Sea (Çinar et al. 2008, 2017; Rousou et al. 2023). Even if it was originally described from the Mediterranean Sea (Çinar 2005), it was subsequently recorded in the Red Sea and elsewhere in the Indo-Pacific region (Çinar et al. 2021; San Martín et al. 2024) and consequently considered alien in the Mediterranean basin (Çinar et al. 2017). The present records from Algeria considerably extend its distribution towards the western Mediterranean Sea.

- *Syllis schulzi* (Hartmann-Schröder, 1960) – first record in Cyprus.

The polychaete *S. schulzi* (Figure S3) was described for the Red Sea and later reported in the western Mediterranean Sea. Zenetos et al. (2010) and Çinar (2013) considered it as a questionable alien species. The distribution of *S. schulzi* encompasses the southern Mediterranean Sea as well as the Red Sea (Ba-Akdah et al. 2018; Toso et al. 2024), suggesting that it should be considered cryptogenic in the basin. It is here recorded for the first time from the coast of Cyprus.

- *Prionospio depauperata* Imajima, 1990 – westwards expansion.

Prionospio depauperata Imajima, 1990, (Figure S4) is a spionid polychaete originally described from Japan and widely reported from the eastern and central Mediterranean Sea (Dağlı and Çinar 2009; Ragkousis et al. 2023; Langeneck et al. 2024). The present data extend its distribution to the Tyrrhenian Sea, western Mediterranean.

- *Chaetozone corona* Berkeley & Berkeley, 1941 – first record in Slovenia.

Chaetozone corona (Figure S5) is a bitentaculate cirratulid polychaete that is considered an established alien species along the coasts of Europe (Le Garrec et al. 2017; Langeneck et al. 2020). The species can be easily distinguished from its congeners owing to its spines starting from the first chaetigers. It was originally reported from the Pacific Ocean (Berkeley and Berkeley 1941) and is currently known from several areas of the Mediterranean from where it was first recorded in muddy sands in the Bay of Izmir (Çinar and Ergen 2007), and later in the eastern coast of the Adriatic Sea (Grossi et al. 2017; Munari et al. 2017). This species was probably introduced from its original range via shipping (Le Garrec et al. 2017). In Slovenia, specimens of *C. corona* were found during benthic surveys as part of the national monitoring programme at four sites in the southern part of the Gulf of Trieste (Slovenia, northern Adriatic Sea) using

a Van Veen grab (0.1 m^2). The first specimen was found on June 2, 2022, on soft bottom at a depth of 7.5 m and three more specimens in 2023: one on June 2 at a depth of 7.5 m and the others at two sites at a depth of 8.1 m and 8.3 m.

- *Hydroides dianthus* (Verrill, 1873) – first record in Romania.

The serpulid polychaete *H. dianthus* is known for its role as a biofouler and invasive species across a wide range of marine and brackish environments globally (Sun et al. 2017). Initially described from the U.S. East Coast, its distribution has expanded significantly, now including the Mediterranean, Black Sea, Pacific, and Indian Oceans (Zibrowius 1973; Bastida-Zavala and ten Hove 2002; Streftaris et al. 2005), recently extending to northern China and Japan (Sun and Yang 2000; Link et al. 2009). In the Black Sea, it was first recorded along the Sevastopol coast, Crimea (Boltacheva et al. 2011); and later from the Sea of Marmara by Çinar et al. (2020, 2024). Here, we report the first documented occurrence of *H. dianthus* along the Romanian Black Sea coast, collected in September 2024 at a depth of 7 m on sandy-mud substrates (Figure S6). Morphological features of the specimen matched the original species description. The operculum exhibits a distinctive two-tiered structure functioning as a plug for the calcareous tube, comprising a basal funnel and a distal spinous verticil (Figure S6B). The funnel features 27 radii with pointed tips. Collar chaetae bear two short blunt teeth and a smooth distal blade. The tube is marked by two longitudinal ridges and lacks peristomes or transverse ridges. The branchial crown includes approximately 16 radioles on each side of the mouth, forming about one-quarter of the worm's total length.

The site of this finding in proximity to the Port of Constanța suggests introduction via maritime traffic. This aligns with recent first Romanian records of other alien polychaetes, such as *Laonome xeprovala* Bick & Bastrop, in Bick et al. 2018 and *Marenzelleria neglecta* Sikorski & Bick, 2004 (Teacă et al. 2021, 2022; Ragkousis et al. 2023), which are also likely shipping-related introductions. The ongoing spread of *H. dianthus* emphasizes the need for constant monitoring, research into its biology and invasion pathways, and the development of effective, environmentally sustainable control measures.

Crustacea (Isopoda, Copepoda and Decapoda)

- *Dynoides amblysinus* Pillai, 1954 –first record in the Mediterranean Sea (Port of Catania, Italy).

The isopod crustacean genus *Dynoides* Barnard, 1914 currently includes 19 species distributed across tropical and subtropical regions (Boyko et al. 2025; Espinosa-Pérez and Hendrickx 2002; Khalaji-Pirbalouty and Gagnon 2021). *Dynoides amblysinus* is native to tropical and temperate seas and has been reported from the western Indian Ocean to eastern Australia, Hong

Kong, and Japan (Bruce 1982; Harrison and Holdich 1984; Li 2000; Nunomura 2010; Kazmi and Yousouf 2013; Bruce and Wong 2015). In July 2024, during the vagile fauna sampling from two PVC panels, *D. amblysinus* was recorded at a depth of 1.2 m on the quay wall of the Levante Pier in the Port of Catania (Figure S7). This finding represents the first known record of the species in the Mediterranean Sea.

No specific information is currently available on the ecology of *D. amblysinus* but studies on other species of the same genus provide some general insights. For example, *D. crenulatus* Carvacho & Haasmann, 1984 and *D. saldanai* Carvacho & Haasmann, 1984, found along the Pacific coast of Mexico, inhabit rocky bottoms of lower intertidal zones thriving among algae such as *Jania adhaerens* J.V. Lamouroux; *Amphiroa misakiensis* Yendo; *Chaetomorpha linum* (O.F.Müller) Kützing and *Hypnea pannosa* J.Agardh (Espinosa-Pérez and Hendrickx 2002).

- *Sphaeroma walkeri* Stebbing, 1905 – first record in Syria.

Sphaeroma walkeri (Figure S8) is a globally distributed isopod (WoRMS Editorial Board 2025), native to the Indian Ocean and the Red Sea. It is a widely ship-transported alien that entered the Mediterranean Sea as far back as 1924 (Omer-Cooper 1927). It is found on many Mediterranean coasts, including the eastern Mediterranean (e.g., the Aegean Sea, Greece) and Tunisia (Galil 2008; Molnar et al. 2008; Ounifi-Ben Amor et al. 2015; Zenetos et al. 2020). It thrives in crevices, under stones and wood, and is common in fouling communities. Many specimens were found among individuals of the invasive mussel *Brachidontes pharaonis* (P. Fischer, 1870) on the Latakia coast at the sites of Afamia, near the fishing port, and the southern Kornish, accompanied by many polychaetes. Small individuals, less than 5 mm long, were observed in densities of 10–30 individuals per m^2 from March to August 2021 in the intertidal zone. The body bears distinct tubercles on the telson and pods, and the telson is toothed (Figure S8A–C). This isopod appears to have been established on the Syrian coast long ago, but its discovery was delayed, probably due to its cryptic habit.

- *Mesanthura* cf. *romulea* Poore & Lew Ton, 1993 – first record in Algeria.

The cryptogenic *Mesanthura* cf. *romulea* (Figure S9) is currently widespread in the Mediterranean Sea, where it locally reaches considerable abundances (Ulman et al. 2017). While already known for most Mediterranean countries, it is here reported for the first time from Algeria.

- *Palaemon macrodactylus* Rathbun, 1902 – third-stage larvae, first record in Romania.

The decapod *P. macrodactylus* has been reported since 2002 in the Romanian Black Sea, and subsequently in the Varna Lake (2009), the Kerch Strait (Sea of Azov, Chushka Spit coast, July 2018) and the Azov–Don Basin (September 2022) by several authors (Micu and Nita 2009;

Raykov et al. 2010; Timofeev et al. 2019; Matishov et al. 2022). However, no larvae had been described so far in the natural environment. Herein, we report the first finding of zoea in the third stage in two samples collected in August 2024 in the Romanian Black Sea waters. The oriental shrimp third stage larvae were distinguished by the following characters: two dorsal spines on the carapace (one clearly visible in Figure S10A), not yet developed pleopods, the sixth somite of the pleon separated from the telson and uropods, and an upward dorsal spine on telson (Figure S10B). The specimens measured about 3 mm in length.

- *Pseudodiaptomus marinus* Sato, 1913 – first record in Spanish Mediterranean waters (Algeciras Harbour).

Pseudodiaptomus marinus (Figure S11) is a calanoid copepod characteristic of coastal waters and the lower parts of estuaries (Walter 1989), native to the Northwest Pacific, from Russia to Philippines (Fleminger and Hendrix Kramer 1988). It has been widely spread throughout the Mediterranean and neighbouring waters due to ballast water and surface currents (Uttieri et al. 2023), although last westernmost record was published in the Berre Lagoon in France, a very enclosed ecosystem, in 2012 (Delpy et al. 2012). The species was later found in the Guadalquivir Estuary (Gulf of Cadiz) on the Atlantic Spanish south-western coast in 2016 (Reyes-Martínez and González-Gordillo 2019). *Pseudodiaptomus marinus* has a very successful population dynamics, caused by a continuously high egg production from May to October (Uye et al. 1982) and tolerates a wide range of temperature and salinity, from 8 °C up to 37 °C (Liang and Uye 1997) and 4 to 44 PSU (Orsi et al. 1991). The present study found *P. marinus* peaking up to 17.43 ind/m^3 in two of the five sampling points of the Algeciras Commercial Harbour in April 2023 using 200 μm nets, under the Spanish Marine Strategy Framework Directive (MSFD) monitoring program (Descriptor 2: Non-indigenous species), indicating maritime transport as introduction vector. This is the first record in the Spanish Mediterranean waters and the westernmost locality recorded in the Mediterranean Sea.

- *Paracalanus tropicus* Andronov, 1977 – first record in Mediterranean waters (Algeciras Harbour, Spain).

Paracalanus tropicus (Figure S12) is an epipelagic calanoid copepod, present in coastal to offshore waters and endemic to the south Atlantic Ocean, Gulf of Guinea, Arabian Sea and Sea of Japan (Khelifi-Touhami and Ounissi 2023). *Paracalanus tropicus* is usually encompassed in the *Paracalanus parvus* Claus, 1863 species complex due to the cryptic differentiation of this group, including besides the nominal and present species also *P. indicus* Wolfenden, 1906, *P. quasimodo* Bowman, 1971, *P. nanus* Sars G.O., 1925, *P. intermedius* Shen & Bai, 1956, and *P. serrulus* Shen & Lee, 1963 (Cornils and Held 2014). Currently, taxonomic uncertainty limits species level monitoring efforts (Peters et al. 2025), usually needing genetical

analyses for ratification (Hidaka et al. 2016). Specimens of *P. tropicus* were found peaking up to 263.76 ind/m^3 in three of the five sampling points of the Algeciras Commercial Harbour in September 2023 using 200 µm nets, under the Spanish MSFD monitoring program, indicating a marine transport related introduction vector. Under the microscope, the diagnostic taxonomic trait distinguishing *P. tropicus* from *P. indicus* is the prosome:urosome ratio (Pr:Ur = 3.8–4.0) (Khelifi-Touhami and Ounissi 2023). Our specimens had a mean Pr:Ur ratio of 3.86 ± 0.46 (N = 10). Together with an eDNA match for *P. tropicus* (Alvite 2023) from the adjacent harbour of Málaga, obtained during the same monitoring program, confirms the species identification. Therefore, this is the first record of this species in Mediterranean waters.

Mollusca (Gastropoda)

- *Siphonaria crenata* Blainville, 1827 – first record in Syria.

The gastropod *S. crenata* is native to the Persian Gulf (Hubendick 1944) and previous studies indicate its presence as an alien species in the eastern Mediterranean Sea (Zenetos et al. 2004), after first record in Lebanon in 1967 (Crocetta et al. 2020). The species is characterized by a solid, thick shell, decorated with 10–12 irregularly strong radial ribs alternating with irregularly thin ribs (Figure S13A). The internal furrow is pale in colour (Figure S13B) and the cone is high (Figure S13C). There is no hole in the upper part of the shell. The outer surface is creamy or rusty brown, and the inner surface is marbled white, containing brown ripples in some individuals (Figure S13A, B). Seven small individuals, 10–20 mm long (Figure S13D), were collected in August 2021 from rocky substrates in the littoral zone of Ras al-Basit, northern Syria. We acknowledge that the taxonomy of the species belonging to the family Siphonariidae is subject to considerable confusion and requires further data and analysis of its genetic characteristics (Jenkins and Köhler 2024).

- *Mitrella psilla* (Duclos, 1846) – first record in Algeria.

The gastropod *M. psilla* (Figure S14), native to West Africa, was first recorded in the Mediterranean in Tunisia (Antit et al. 2009), and then in Italian waters in the Tyrrhenian and Ionian Sea (Nappo et al. 2019; Scuderi and Villari 2022), and in Spain, indicating its continued expansion into the Basin (Martínez-Ortí et al. 2020). Its presence in the port of Algiers further demonstrates its ongoing dispersal across the Mediterranean.

Bryozoa

- *Catenicella paradoxa* Rosso, 2009 – first record in Sicily/Sicily Strait.

The bryozoan *C. paradoxa* (Figure S15A) was originally described from a few colonies collected in July 2006 on the north side of Capo Passero Island

SE Sicily. These were found encrusting two algal species typical of the Infralittoral Algae biocenosis, at a depth of 11 m beneath a cliff overhang (Rosso 2009). The species was also recently reported from three stations within the Ciclopi Islands Marine Protected Area (W Ionian Sea, E Sicily), between 5 and 26 depth, again associated with Infralittoral Algae (Rosso et al. 2019; Mytilineou et al. 2016). *Catenicella paradoxa* forms inconspicuous, glassy, and transparent colonies composed of uniserial articulated branches, and remains the only catenicellid currently known from the present-day Mediterranean (Rosso and Di Martino 2023). Rosso (2009) suggested the cryptogenic status of *C. paradoxa*, proposing four different, some potentially concurrent, hypotheses to explain its presence in the Mediterranean Sea: the possible overlooking of an inconspicuous, hidden, resident species; the local differentiation of a new taxon; the human-mediated introduction of a non-native species; or the recent arrival of an undescribed species expanding its range through the Suez Canal, potentially facilitated by warming waters and global change. The discovery of a single colony in 2014, on an artificial panel deployed at approximately 20 m depth and a few meters inside a cave in the Plemmirio Marine Protected Area near Siracusa (Ionian Sea, SE Sicily), was interpreted by Rosso et al. (2018) as potentially supporting a non-native origin. Given the species' limited and localized records in the western Ionian Sea, the finding of a single colony of *C. paradoxa* on a sponge stranded on the Marina di Ragusa beach in the winter of 2022 represents the first documented occurrence in the Sicily Strait.

- *Tricellaria inopinata* d'Hondt & Occhipinti-Ambrogi, 1985 – first record in Sicily/western Ionian Sea.

The bryozoan *T. inopinata* (Figure S15B), was described based on colonies found attached to the byssus threads of the bivalve *Mytilus galloprovincialis* Lamarck, 1819, collected from the intertidal zone of the Giudecca Canal, near central Venice (northern Adriatic Sea) in 1982 (d'Hondt and Occhipinti Ambrogi 1985; Occhipinti Ambrogi 2000). This species forms small, erect, bush-like colonies a few centimetres high, composed of jointed, slightly mineralised biserial internodes, anchored to hard substrates by rhizoids in shallow waters. Colonies grow rapidly, intertwining to form dense, whitish mats/carpets. Although described from the Mediterranean, *T. inopinata* is considered an alien species in this basin. It was likely introduced accidentally, possibly through aquaculture or ship fouling, from its presumed native range in the Pacific Ocean (Occhipinti-Ambrogi et al. 2010). Since its original description, *T. inopinata* has been recorded from several western Mediterranean localities (e.g., Ben Suissi et al. 2006; Lodola et al. 2012; Ulman et al. 2017; Ferrario et al. 2018; Fortič and Mavrič 2018; Bensari et al. 2025), with its easternmost occurrence in the Old Venetian Harbor (Heraklion, Greece) (Ulman et al. 2017). The species has also spread globally (e.g., Dyrinda et al. 2000; Johnson et al. 2012; Micael et al. 2016).

Tricellaria inopinata has been classified as invasive following monitoring of its rapid spread and increase in colony abundance and surface coverage, often to the detriment of native species that decline or disappear after its arrival and establishment. For instance, this has been documented in the Venice lagoon (Occhipinti Ambrogi 2000; Corriero et al. 2007) and in some localities along the western Atlantic coast (Johnson et al. 2012). Based on expert evaluations and direct observations, *T. inopinata* has demonstrated moderate to high impacts on biodiversity and ecosystem services, primarily through competition and fouling (Tsirintanis et al. 2022). Moreover, according to Gavira-O'Neil et al. (2016), it may act as a driver of potential invasion meltdown.

Tricellaria inopinata was found in the channel connecting the Faro Lake (located in the NE corner of Sicily) to the Ionian Sea, based on samples collected in 2006 and 2008 at about 50 cm depth. This represents the first documented record for both Sicily and the western Ionian Sea; prior to this, *T. inopinata* was only known from the eastern Ionian sector, in the Old Venetian Harbor (Heraklion, Greece) (Ulman et al. 2017). At the time of collection, the species was by far dominating the community. However, subsequent targeted surveys conducted in summer 2021, and spring 2024 and 2025, failed to detect it, suggesting that its populations may undergo regression and even local disappearance, unlike in other localities. A focused investigation is needed to determine the environmental factors responsible for this short-term success.

- *Arbopercula angulata* (Levinsen, 1909) – first record in Liguria (northernmost Tyrrhenian/western Mediterranean).

The bryozoan *A. angulata* (Figure S15C) is a non-native species in the Mediterranean. It was first reported in this basin (Rosso 1994), misidentified as *Electra tenella* (Hincks, 1880), from the Ionian coast of southeastern Sicily, where it was found on stranded plastic debris in the Gulf of Catania and on experimental plastic containers deployed in Siracusa harbour at 5 m depth in meadows dominated by *Cymodocea nodosa* (Ucria) Ascherson and *Caulerpa prolifera* (Forsskål) J.V.Lamoroux. *Arbopercula angulata* was only found on containers retrieved after deployment periods of 3 to 6 months starting from 27 January 1990. The species forms thin, circular to lobate, encrusting, usually unilaminar colonies that can extend on broad surfaces. It often shows high intracolonial morphological variability. Over the past dozen years, the species, still often referred to as *E. tenella*, has been reported from additional sites, including Capo Rasocolmo near Messina (Sicily, February 2012), and the Livorno harbour (Tuscany, July 2011), both in the Tyrrhenian Sea (Thessalou - Legaki et al. 2012). More recently, it has been found on floating plastic debris collected in October 2020 off the Catalonia coast, where it was identified as *Arbopercula tenella* (Subías-Baratau et al. 2022). Other records also include floating ligneous cores near the Strait of

Gibraltar, where it was reported initially as *Electra* cf. *tenella* (López de la Cuadra and García-Gómez 1994), and plastic debris stranded in July 2012 in Menorca, Balearic Islands, subsequently identified as *A. angulata* (Reverter-Gil and Souto 2025). Collectively, these records indicate that *A. angulata* may have a wide distribution in the western Mediterranean. The numerous new occurrences documented in the present study suggest a constant presence of *A. angulata* at least all-around Sicily. The observation of colonies almost completely covering boat buoys in the harbour of Lerici (Liguria, Italy) represents the northernmost record of this species in both the Tyrrhenian and western Mediterranean.

Echinodermata

- *Aquilonastra burtoni* (Gray, 1840) – first record in Syria.

The starfish *A. burtoni* is widely distributed in the Indian and Pacific Oceans, from the Hawaiian Islands and Australia to Madagascar, East Africa, and the Red Sea (WoRMS Editorial Board 2025). It is a Lessepsian migrant recorded in the Levantine Sea since the 1950s at depths of 8–10 m (Tortonese 1966; Koukouras et al. 2007), that later expanded its distribution to Cyprus and the Aegean Sea (Tzomos et al. 2011). Ten specimens of this species were collected in June 2024 by divers to a depth of 2 m, from marine fouling assemblages on the concrete wall of the fishing and recreational port in the city of Tartus, southern Syria. The abundance reached 6 individuals/m^2, and some additional individuals were found in empty oyster shells at the same site. The specimens showed variable shapes among individuals, rather than a uniform morphology (Figure S19A). The body was asymmetrical, with a diameter of approximately 15 mm (Figure S16B). The number of arms varied between three and eight, usually seven (Figure S16A). The arms were of unequal length and finger-like, wider at the base and narrowing and rounded at the end. The colour was greenish grey on the oral side, with a large, irregular, purple-brown spot in the middle, surrounded by red spots on the basal parts of each arm (Figure S16C, D). The asymmetrical shape of all individuals in the sample supports the idea of asexual reproduction by fission (Karako et al. 2002).

Chaetognatha

- *Ferosagitta* sp. – first record in Romania.

Ferosagitta sp. (Figure S17A) is a chaetognath species found in July 2024 in three stations sampled during a survey carried out across the Romanian shelf. Both juveniles and adults have been found in three out of 22 samples collected. The species differs from *F. galerita* (Dallot, 1971), the alien species reported in the Mediterranean Sea (Terbiyik et al. 2007) by the shape of the anterior fins (narrow, elongate in our specimens) (Figure S17B), the position of the seminal vesicles (touching the posterior fins in

Ferosagitta sp.) (Figure S17C), tail shape and length (short, about 1.6 mm long, pear-like, caudal fin fan-shaped, with radiating rays, symmetric) (Figure S17C). Apart from the prominent ventral ganglion (Figure S17D), the juveniles of our species resemble the adults' general conformation. The species was found across Mangalia and Sf Gheorghe transects in near-surface and deep-water layers (0–10 m; within thermocline: 10–35 m, and below thermocline: 150 m), with densities ranging from 0.98 to 13.69 ind. m^{-3}.

Tunicata (Ascidiacea)

- *Symplegma rubra* Monniot C., 1972 – first record in the Mediterranean.

Symplegma rubra (Figure S18) is a colonial ascidian with a tropical and warm-temperate distribution. In the Mediterranean, it has historically been confused with the congenerics *S. brakenhielmi* (Michaelsen, 1904) and *S. viride* Herdman, 1886 (Ramos-Esplá et al. 2020). However, *S. rubra* and *S. brakenhielmi* present lobed testes in contrast to *S. viride*, while *S. brakenhielmi* presents reticulate tunic, *S. rubra* has opaque tunic and characteristic red rings encircling both siphons, although this last character has also been observed in some varieties of *S. brakenhielmi* (Herdman 1906; Tokioka 1961). Moreover, specimens identified as either *Symplegma* sp., *S. brakenhielmi*, and *S. rubra* have been found to belong to different lineages, stressing the need of taxonomic revisions based on integrated data (Mastrototaro et al. 2019). Nonetheless, the specimens, here reported from Tunisia, off the coast of the island of Kuriat, morphologically match *S. rubra* and represent the first record of this species for the Mediterranean Sea.

- *Symplegma brakenhielmi* Michaelsen, 1904 – first record in Spanish Mediterranean waters (Figure S19).

Symplegma brakenhielmi is a colonial ascidian with a pantropical distribution, present in the Mediterranean from East to West (Ramos-Esplá et al. 2020), extending in the Tyrrhenian Sea (Ulman et al. 2017; Mastrototaro et al. 2019). The colonies observed in the Port of Alicante are red and yellow (Mastrototaro et al. 2019) with a reticulated tunic, without red rings encircling both siphons, and present lobed testes.

- *Phallusia nigra* Savigny, 1816 – first record in Italy.

The solitary ascidian *P. nigra* (Figure S20), originally considered a circumtropical species, was first reported in the Mediterranean Sea by Pérès (1958), in Israel. Later, Shenkar and Loya (2009) documented a wide distribution of the species along the Israeli coasts, from shallow waters down to 30 m, on both natural and artificial substrates. The species has also been reported in Lebanon (Zibrowius and Bitar 2003), Syria (Bitar et al. 2003), along the Turkish coasts (Çinar et al. 2006), Greece (Kondilatos et al. 2010; Latsoudis 2020), and in the Aegean Sea (Thessalou-Legaki et al. 2012).

In 2024, the first record of the species in Tunisian waters was reported by Mnasri-Afifi et al. (2024a), from buoys in the port of Bizerte (northern Tunisia), representing the westernmost sighting in the Mediterranean basin. In July 2024, during hard bottom sampling (scraping) the tunicate was found in two samples collected at depths of 2 m and 5 m on the quay wall of the Levante Pier in the Port of Catania. This record represents the first morphological evidence of the black tunicate in Italian waters, whose presence in Italian seas (ports of Trieste and Civitavecchia) was previously only inferred based on eDNA metabarcoding methods (Varrella et al. 2025). Following the Catania record, the species was sampled in the port of Augusta during a Consorzio Nazionale Interuniversitario per le Scienze del Mare (CoNISMa) survey and were further confirmed by DNA barcoding by the same team (sequence deposited in GenBank under the accession code PV018849, giving a 99% identity with deposited sequences of *P. nigra*).

- *Microcosmus exasperatus* Heller, 1878 – first record in Italy.

The identity and distribution of the ascidian *M. exasperatus* (Figure S21) in the Mediterranean Sea have been the object of some confusion, as this species was historically reported as a misidentification of another, more widespread, congeneric species, *M. squamiger* Michaelsen, 1927 (Ramos-Esplá et al. 2013). Revisions of Mediterranean records of this species confirmed its occurrence in the eastern and southern Mediterranean Sea (Turon et al. 2007; Ramos-Esplá et al. 2013). The specimens sampled in the port of Augusta represent the first record for Italian waters and might have arrived either by natural expansion from Tunisia, where the species is widespread, or aided by shipping from the eastern Mediterranean Sea. A similar situation explains the occurrence of the other alien ascidian *Phallusia nigra* in the same port.

- *Distaplia bermudensis* Van Name, 1902 – first record in Italy (Adriatic Sea) and Tunisia.

Distaplia bermudensis (Figure S22) is a small colonial ascidian originally described from Bermuda and widespread in the western Atlantic. It was first reported in the Mediterranean Sea in 2000 from the Gulf of Taranto (Italy), where subsequent studies on fouling assemblages indicate an established population (Mastrototaro and Brunetti 2006). A further record in 2019 in the Phlegraean lagoons (Gulf of Naples, Tyrrhenian Sea) also pointed to an established population in that area, with the Bacoli zone as the most likely introduction site, associated with marinas and mussel farming (Virgili et al. 2022). These two Italian populations appear geographically isolated and were never compared molecularly, so they may represent separate introduction events. Both sites are associated with marinas and mussel farming, suggesting these vectors may have facilitated its introduction (Orfanidis et al. 2021; Virgili et al. 2022). In 2021,

D. bermudensis was also recorded in three Tunisian marinas (first record for Tunisia), indicating a further southward expansion of this NIS in the Mediterranean. In 2024, an extremely small colony was recorded in the Brindisi port during a marina survey, representing the first report for the Adriatic Sea. Its introduction is likely linked to shipping or aquaculture activities.

- *Botrylloides niger* Herdman, 1886 – first record in Greece (Zakynthos Island, eastern Ionian Sea).

In October 2022, the colonial ascidian *B. niger* (Figure S23), was recorded for the first time on Zakynthos Island (Ionian Sea, Greece) during a rapid assessment survey of alien species. Colonies were observed at six of nine surveyed marinas, growing on artificial substrates such as ropes and tyres, and on the basal leaves and rhizomes of the invasive seagrass *Halophila stipulacea* (Forsskål) Ascherson. The same seagrass patch also hosted dense aggregations of the bryozoan *A. verticillata,* highlighting the role of *H. stipulacea* as a novel substrate for IAS. Although *B. niger* has previously been reported as an epiphyte on the surface of native seagrasses (Virgili et al. 2022), its occurrence on the invasive *H. stipulacea* raises questions about how interactions between co-occurring IAS may shape invasion dynamics. Given the seasonal shedding of *H. stipulacea* leaves in the Mediterranean, such associations may be temporary at the colony level, but detached leaves could also facilitate short-distance dispersal by rafting. In this case, the overgrowth of *H. stipulacea* by *B. niger* could reflect a form of invasional interference, where one IAS suppresses another, potentially altering invasion trajectories and ecosystem outcomes (Simberloff and Von Holle 1999). This contrasts with invasional meltdown, in which invaders facilitate each other's spread (Simberloff and Von Holle 1999). While such interactions may limit individual IAS, cumulative impacts can still be profound due to ecosystem-level disruptions (Gordon 1998).

The ability of *B. niger* to colonize both artificial structures and living macrophytes reflects its ecological plasticity and raises concern over its potential to displace native benthic assemblages. Its presence in Zakynthos expands its known range in the Mediterranean, where it has shown high morphological and genetic variability (Virgili et al. 2022; Temiz et al. 2023; Mnasri-Afifi et al. 2024b). Alongside a rise in introduced ascidian species from 14 to 20 in the last decade in the Mediterranean Sea (Muñoz and Ramos-Esplá 2019), this finding underscores the need for early detection, molecular surveillance, and ecosystem-based management in addressing marine bioinvasions. For example, the presence of *B. niger* has been previously hypothesized in Crete, South Aegean Sea, as early as 2018, on Artificial Reef Monitoring Structures (ARMS) via molecular screening, as manifested in the accompanying dataset of Obst et al. (2020). On the other hand, the analysis of *B. niger* colonies from the Mediterranean Sea has

already proved to be a potential biological resource for nutritional or functional purposes, reducing, in this way, the economic losses caused by this invasive organism (Della Sala et al. 2022). Therefore, its potential use for such purposes can be also tested in the new localities of occurrence reported here.

Macroalgae (Ochrophyta)

- *Stypopodium schimperi* (Kützing) Verlaque & Boudouresque – First record in southern Italy (Lampedusa & Pantelleria).

Stypopodium schimperi is a medium sized brown macroalga (up to 18 cm high) irregularly branched with fan-shaped blades (Figure S24). Its distinguishing characteristics are the flat growing margin, the absence of calcification, the hairs in concentric lines and the distinctive tetrasporangia external to the blade (Verlaque et al. 2015). It is distributed in shallow subtidal coral and rocky reefs in the Indian and Atlantic Oceans. It has been introduced in the Mediterranean Sea, most probably through the Suez Canal. It was first recorded in Israel in 1973 (Lundberg 1980) and is now very common in the Eastern Mediterranean with records in Libya (Nizamuddin and Godeh 1989), Syria (Mayhoub 1989), Egypt, Turkey, Cyprus (Verlaque and Boudouresque 1991), Lebanon (Bitar et al. 2000), Greece (Sartoni and De Biasi 1999; Tsiamis et al. 2008; Nicolaidou et al. 2012) and recently Croatia and Adriatic Sea (Žuljević et al. 2023). In summer 2024, it was observed for the first time in Southern Italy, in the islands of Lampedusa and Pantelleria. It is likely that the species is more widely distributed in the Mediterranean and maybe confused with *Lobophora variegata* (J.V. Lamouroux) Womersley ex E.C. Oliveira another brown algae with a similar morphology.

Discussion

This third collaborative compilation of unpublished records further consolidates an open, region-wide evidence base for alien, cryptogenic, and neonative species in the Mediterranean and Black Sea. By adding 7,434 geo-referenced records from 21 countries, it extends the temporal span (1973–2025) and complements two earlier datasets (Katsanevakis et al. 2020; Ragkousis et al. 2023) that collectively demonstrated the utility of mobilizing otherwise inaccessible information held by experts and institutions. Together, these efforts show that routine aggregation of unpublished records can rapidly enrich inventories, improve the spatial and taxonomic resolution of knowledge, and enable timely reassessments of invasion dynamics needed for policy and management (Katsanevakis et al. 2020; Ragkousis et al. 2023).

The spatiotemporal distribution of records highlights both strengths and gaps in current monitoring. Contributions were highest in Lebanon, Greece, Cyprus, Italy, and Romania, while Albania, Egypt, Georgia, Ukraine, France,

and parts of North Africa remained underrepresented. Similar geographic imbalances have been reported in the previous two "unpublished records" datasets, suggesting that uneven expert participation/sampling effort (alongside ecology) systematically shapes apparent hotspots and gaps (Katsanevakis et al. 2020; Ragkousis et al. 2023). This underrepresentation should not be interpreted exclusively as a lack of national expertise or monitoring programme; for example, Massé et al. (2023) provide an updated overview of marine alien (including cryptogenic) species across three metropolitan French regions. At the basin scale, expert-validated inventories (e.g., Galanidi et al. 2023) provide policy-oriented baselines, which our record-level, georeferenced compilation can complement by adding finer spatiotemporal resolution that supports inventory updates and monitoring prioritization.

Habitat was reported for a subset of records; within those, 453 records explicitly indicate observations in harbours/marinas or shellfish aquaculture settings (e.g., mussel/oyster farming). These represent a minimum count and are consistent with the role of artificial coastal infrastructures and aquaculture facilities as introduction gateways and detection hotspots for alien species. Because habitat/site type was not reported for all entries and does not allow a standardized classification of all records by site category, we do not attempt a full quantitative partition of the dataset by site type.

Temporally, reporting increased sharply after 2021, peaking in 2024 – a trend shaped not only by species spread but also by intensified surveys and improved contributor engagement. Such patterns caution against interpreting temporal increases as purely ecological signals. Instead, they flag locations where targeted effort, training, and data-sharing could most improve coverage (see also Katsanevakis et al. 2020; Ragkousis et al. 2023).

Taxonomic uncertainty is inevitable in a large, multi-source dataset spanning many groups and a large number of contributors, and some misidentifications are likely – particularly for taxa that require specialist examination or integrative taxonomy (e.g., cryptic species complexes and closely related congeners). Records should therefore be interpreted as best-available expert identifications at the time of reporting, and they may be refined in future as taxonomic knowledge improves, and as additional voucher or molecular evidence becomes available (see also Zenetos et al. 2017). For transparency and traceability, the identifier is reported for every record in Table S1, responsibility for correct identification and reporting rests with the observer of each record, as in previous iterations of this collective data initiative.

Several new basin-level records (e.g., *D. amblysinus*, *S. rubra*, *M. mcintoshi, P. tropicus*) and national firsts (e.g., in Algeria, Cyprus, Greece, Italy, Slovenia, Spain, Syria, Tunisia) show that introductions continue across taxa and pathways. Range extensions (e.g., *P. depauperata* in the Tyrrhenian; *A. angulata* northward; *S. brakenhielmi* westward) illustrate secondary

spread, often linked to shipping, marinas, or aquaculture. The isolated Italian populations of *D. bermudensis* highlight how repeated introductions may mimic continuity if molecular comparisons are lacking, while *P. marinus* in Algeciras emphasizes ballast water as a recurrent vector.

A complementary dimension not analysed quantitatively here is the donor-region structure of introductions (e.g., Atlantic vs Red Sea/Indo-Pacific sources), a topic discussed at basin scale by Tsiamis et al. (2018). Robust estimates would require standardized species-level attribution of native range for primary and secondary introductions at subregional level, which is not consistently captured as a structured field in Table S1 and would involve additional harmonization and literature-based validation for many taxa. We therefore treat donor-region partitioning as a priority for a subsequent synthesis, ideally integrating the three unpublished-record compilations with published inventories and origin/pathway classifications.

The dataset also sheds light on ecological dynamics. Cryptogenic cases such as *M. mcintoshi* illustrate how symbiotic lifestyles complicate origin assessments. Neonatives are expected to continue to expand under climate change, while interactions among invaders (e.g., *B. niger* overgrowing *H. stipulacea*) raise questions about "invasional interference" versus facilitation. These examples underline the need for an ecosystem-based perspective in monitoring and management, especially in port and marina environments.

As in earlier compilations, biases in sampling effort and taxonomic expertise limit quantitative inference, but do not reduce the dataset's value for early detection, range tracking, and prioritization of surveillance, including targeted molecular confirmation where appropriate. Continued coordination, standardization, and integration with official databases remain essential. Overall, this initiative confirms that collaborative mobilization of unpublished records is a cost-effective way to sharpen regional situational awareness and guide proactive responses to marine bioinvasions.

This work demonstrates how sustained collaboration and systematic mobilization of unpublished records can fill critical knowledge gaps on alien, cryptogenic, and neonative species in the Mediterranean and Black Seas. By improving the spatiotemporal resolution of regional inventories, the dataset supports early detection and prioritization of monitoring, contributing to EU-level obligations under the Marine Strategy Framework Directive (MSFD 2008/56/EC)—including Descriptor 2 (non-indigenous species)—and facilitating interoperability with MSFD-linked infrastructures such as EASIN (European Alien Species Information Network). It also supports region-wide commitments under the Barcelona Convention and UNEP/MAP, including the updated Action Plan concerning Species Introductions and Invasive Species in the Mediterranean Sea adopted in 2023 (Decision IG.26/5, Annex IV), which emphasizes coordinated monitoring and data sharing. In addition, regional practitioner networks such as

MedPAN can provide an operational pathway to strengthen ongoing acquisition and updating of alien-species occurrence information across Mediterranean MPAs.

Continued iterations of this effort will be essential to keep pace with ongoing introductions and shifting species distributions in a rapidly changing sea.

Authors' contribution

JCAP, SK research conceptualization; data analysis and interpretation; original draft; writing – review and editing; SK: funding provision; all authors: investigation and data collection, writing – review and editing.

Author ORCIDs (where available)

Jennifer C.A. Pistevos: 0000-0001-8081-7069; Argyro Zenetos: 0000-0003-2670-042X; Yolanda Stranga: 0009-0003-4231-856X; Athanasios Nikolaou: 0000-0003-2241-3984; Joachim Langeneck: 0000-0003-3665-8683; Mihaela Muresan: 0000-0002-8446-1263; Juan Sempere-Valverde: 0000-0001-5856-9214; Sahar Chebaane: 0000-0003-3700-8930; Lydia Png-Gonzalez: 0000-0001-7986-9654; Sylvaine Giakoumi: 0000-0002-3301-5538; Michail Ragkousis: 0000-0002-3886-4280; Adriana Vella: 0000-0003-2246-5354; Noel Vella: 0000-0003-0080-5709; Vesselina Vasileva Mihneva: 0000-0003-4581-7738; Thanos Dailianis: 0000-0002-4102-9331; Victor Surugiu: 0000-0003-4933-7678; Angeliki Bosmali: 0009-0003-4767-0300; Vasilis Gerovasileiou: 0000-0002-9143-7480; Fabio Crocetta: 0000-0002-2800-3735; Leda Liyue Cai: 0000-0001-7699-4601; Angela G. Bartolo: 0000-0001-9109-8979; Maria Rousou: 0000-0003-1234-2027; Jose Antonio García-Charton: 0000-0002-5973-9913; Sandra Agius Darmanin: 0000-0003-4489-9030; Riccardo Scalera: 0000-0002-4100-3959; Donat Petricioli: 0000-0002-0150-9322; Tatjana Bakran-Petricioli: 0000-0002-3368-4248; Alberto Colletti: 0000-0002-7027-6315; Stefanos Kalogirou: 0000-0002-3064-9236; Domenico Sacco: 0009-0003-6595-1931; Tullio Rossi: 0000-0003-0461-554X; Christophe Lejeusne: 0000-0002-8821-3849; Dominique Davoult: 0000-0001-8678-3998; Daniel Faget: 0000-0001-7122-7446; Petros Stavrou: 0009-0006-0670-0237; Jasmine Ferrario: 0000-0003-4462-4141; Stelios Katsanevakis: 0000-0002-5137-7540; Arthur R. Bos: 0000-0002-0708-314X; Grigorios Skouradakis: 0000-0002-8482-460X; Konstantinos Kavakakis: 0009-0003-0583-8304; Carlo Nike Bianchi: 0000-0003-1071-3285; Carla Morri: 0000-0003-4520-2741; Laura Tamburello: 0000-0003-1305-8802; Bessy Stancanelli: 0000-0001-7126-0633; Vincenzo Di Martino: 0000-0002-5987-0954; Julian Evans: 0000-0001-7837-5927; Abigail Kennedy: 0009-0006-8924-7588; Daniele Grech: 0000-0002-4936-5997; Maria Zotou: 0000-0002-9495-3907; Ernesto Azzuro: 0000-0002-9805-3164; Patrick J Schembri: 0000-0002-6723-7198; Ilaria D'Aniello: 0009-0001-7720-2335; Michele Solca: 0000-0002-3190-5844; Antonios Mazaris: 0000-0002-4961-5490; Frédérique Viard: 0000-0001-5603-9527; Michele Luca Geraci: 0000-0002-3143-4659; Giulia Furfaro: 0000-0001-8184-2266; Giacomo Sardo: 0000-0001-7997-637X; Sergio Vitale: 0000-0001-6063-4126; Fabio Falsone: 0000-0003-1754-4226; Monica Calabrò: 0000-0002-9972-0440; Louis Hadjioannou: 0000-0001-5864-5467; Michel Bariche: 0000-0001-6831-4311; Alfonso A. Ramos Esplá: 0000-0002-6886-218X; Luca Castriota: 0000-0002-3349-9501; Manuela Falautano: 0000-0001-7844-665X; Ana Fortič: 0000-0002-5715-3572; Borut Mavrič: 0000-0001-5488-0793; Domen Trkov: 0000-0002-6573-2822; Lovrenc Lipej: 0000-0002-7608-1631; Martina Orlando-Bonaca: 0000-0001-6717-0176; Valentina Pitacco: 0000-0003-2724-2838; Neža Leban: 0009-0009-5913-2693; Antonietta Rosso: 0000-0001-5565-9513; Emanuela Di Martino: 0000-0002-3892-4036; Gemma Donato: 0000-0003-1783-4688; Rossana Sanfilippo: 0000-0001-6154-8027; Chiara Siddiolo: 0000-0003-1871-5955; Rym Zakhama-Sraieb: 0000-0002-5174-9789; Intissar Mnasri: 0009-0004-7609-7950; Víctor Orenes-Salazar: 0000-0001-9291-1087; Antonio Di Franco: 0000-0003-3411-7015; Anna Maria Mannino 0000-0001-6419-7066.

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Data availability

Species georeferenced records are available at the European Alien Species Information Network: https://easin.jrc.ec.europa.eu/easin/RJD/Download/ca406ad8-fa04-42cf-825d-03232be3570d

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Supplementary material

The following supplementary material is available for this article:

Table S1. Dataset of unpublished Mediterranean and Black Sea records of marine alien, cryptogenic, and neonative species (III)

Figure S1. *Leodice antennata* from Lebanon

Figure S2. *Syllis ergeni* from Algeria

Figure S3. *Syllis schulzi* from Cyprus

Figure S4. *Prionospio depauperata* from Piombino, Tyrrhenian Sea

Figure S5. *Chaetozone corona* from the Gulf of Trieste

Figure S6. *Hydroides dianthus* from Romania

Figure S7. *Dynoides amblysinus* from Port of Catamia, Italy

Figure S8. *Sphaeroma walkeri* from Syria

Figure S9. *Mesanthura* cf. *romulea* from Algeria

Figure S10. *Palaemon macrodactylus* zoea from Romania

Figure S11. *Pseudodiaptomus marinus* found in Algeciras Harbour (Spain)

Figure S12. *Paracalanus tropicus* found in Algeciras Harbour (Spain)

Figure S13. *Siphonaria crenata* from Syria

Figure S14. *Mitrella psilla* from Algeria

Figure S15. (A) *Catenicella paradoxa* from Sicily Strait (B) Articulated biserial internodes of *Tricellaria inopinata* from Sicily (C) *Arbopercula angulata* from Liguria

Figure S16. (A, B) Regenerated specimens of *Aquilonastra burtoni* recorded from Syria

Figure S17. *Ferosagitta* sp. from the Black Sea

Figure S18. (A, B) *Symplegma rubra* off the coast of Tunisia

Figure S19. *Symplegma brakenhielmi* from Alicante pot (Western Mediterranean). Chromomorphs: red (A) and yellow (B)

Figure S20. *Phallusia nigra* from Italy

Figure S21. *Microcosmus exasperatus* from Italy

Figure S22. *Distaplia bermudensis*. (A) Bizerte, Tunisia; (B) Brindisi, Italy; (C) El Kantaoui, Tunisia

Figure S23. *Botrylloides niger* colony from Greece

Figure S24. *Stypopodium schimperi* in Lampedusa Island