

Taxa detection periods based on environmental DNA in DFO's Arctic, Gulf, Maritimes, Newfoundland, and Québec Regions

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2023

**Canadian Data Report of
Fisheries and Aquatic Sciences 1379**



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by

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Department of Fisheries and Oceans, 2023.

Cat. No. Fs97-13/1379E-PDF ISBN 978-0-660-68771-1 ISSN 1488-5395

Correct citation for this publication:

Morrison, M. K., Howland, K., LeBlanc, F., Bernier, R., Chevrinai, M., DiBacco, C., Gagné, N., Grey, E. K., Hamilton, L., Jeffery, N., Kinnison, M. T., Parent, G. J., Simard, N., Trudel, M., and Lacoursière-Roussel, A. 2023. Taxa detection periods based on environmental DNA in DFO's Arctic, Gulf, Maritimes, Newfoundland, and Québec regions. *Can. Data Rep. Fish. Aquat. Sci.* 1379: ix + 308 p.

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ABSTRACT

Morrison, M. K., Howland, K., LeBlanc, F., Bernier, R., Chevrinai, M., DiBacco, C., Gagné, N., Grey, E. K., Hamilton, L., Jeffery, N., Kinnison, M. T., Parent, G. J., Simard, N., Trudel, M., and Lacoursière-Roussel, A. 2023. Taxa detection periods based on environmental DNA in DFO's Arctic, Gulf, Maritimes, Newfoundland, and Québec regions. *Can. Data Rep. Fish. Aquat. Sci.* 1379: ix + 308 p.

With its power to increase marine survey coverage, environmental DNA (eDNA) detections are increasingly employed to monitor ecosystems and support decision-making. There is currently limited guidance on optimal timing for taxon detection in eDNA-based studies and monitoring programs; consequently, projects may fail to detect species of interest or spend more time and resources than needed. The lack of guidance on optimal eDNA sampling practices has been highlighted by regional, national, and international groups as a critical limitation to management and regulatory uptake. This data report is a first step towards providing guidance on optimal timing for eDNA sampling by presenting seven years (2015-2022) of eDNA-based observation data for 1476 taxa across multiple regions of Canada. eDNA detections are reported at the genus (DNA metabarcoding) and species (quantitative polymerase chain reaction) level from water samples collected along the Canadian Atlantic and the Eastern Arctic coast. End-users of eDNA (*e.g.*, aquatic invasive species and species at risk managers) looking to use the data herein to optimize sampling strategies for target taxa and regions should ensure frequent communication with eDNA experts for guidance on optimal field and lab methods.

RÉSUMÉ

Morrison, M. K., Howland, K., LeBlanc, F., Bernier, R., Chevrinai, M., DiBacco, C., Gagné, N., Grey, E. K., Hamilton, L., Jeffery, N., Kinnison, M. T., Parent, G. J., Simard, N., Trudel, M., and Lacoursière-Roussel, A. 2023. Taxa detection periods based on environmental DNA in DFO's Arctic, Gulf, Maritimes, Newfoundland, and Québec regions. *Can. Data Rep. Fish. Aquat. Sci.* 1379: ix + 308 p.

Grâce à son pouvoir permettant d'augmenter l'étendue de l'échantillonnage marin, l'utilisation de l'ADN environnemental (ADNe) est en croissance pour la surveillance d'écosystèmes et le support dans les prises de décision. Actuellement, les directives sur la temporalité optimale de l'ADNe pour les projets et les programmes de surveillance des taxons sont limitées; risquant ainsi d'échouer la détection des espèces d'intérêt ou prendre plus de temps et de ressources que nécessaire. L'absence de directives sur les pratiques optimales d'échantillonnage d'ADNe a été soulignée par des groupes régionaux, nationaux et internationaux comme limitation critique pour la gestion et l'adoption au sein de la réglementation. Ce document présente la première tentative de fournir des conseils sur la période optimale pour échantillonner l'ADNe. Il rapporte la compilation de données d'ADNe s'étalonnant sur sept années (2015-2022) couvrant 1 476 taxons à partir d'échantillons d'eau prélevés le long de la côte Nord-Ouest Atlantique Canadienne et de l'est de l'Arctique Canadien. Les détections sont rapportées au genre pour les données de métacodage à barres de l'ADN (ou métabarcoding) et à l'espèce pour les données recueillies à l'aide de la réaction en chaîne par polymérase en temps réel (qPCR). Les utilisateurs de détections d'ADNe (p.ex. gestionnaires des espèces aquatiques envahissantes, des espèces en péril) intéressés par l'utilisation de ces données pour l'optimisation des protocoles d'échantillonnage pour un taxon et une région donnés, devraient s'assurer de communiquer continuellement avec des experts en ADNe afin d'être conseillés sur les méthodes de terrain et de laboratoire optimales.

1. INTRODUCTION

Approaches for non-invasive ecosystem monitoring are becoming increasingly important with the rapid habitat degradation and global biodiversity loss that is continuing to occur with Earth's changing climate (WWF 2022). Detection of environmental DNA (eDNA) is one of these approaches. Environmental DNA is defined as DNA found in the environment from any biological source, including DNA from degradation of dead organisms (*e.g.*, dead larvae, digested remains from predators), and from the release of biological materials of live organisms (*e.g.*, epithelial cells, mucus, gut contents, urine, feces, biofilms; Beng and Corlett 2020), as well as microorganisms. Over the last decade, eDNA data have become more prevalent as a tool for monitoring species of conservation and management concern (*i.e.*, aquatic invasive species [AIS], species at risk [SAR], biodiversity assessments) as environmental sampling for eDNA does not require physical capture of the target species (Deiner et al. 2017). Moreover, the relative ease of collecting environmental samples (*e.g.*, water or sediments) compared to traditional species capture methods allows for greater accessibility of survey sites and the possibility of increased sampling frequency (Morrison et al. 2023).

A better understanding of factors altering eDNA detection rates is fundamental to optimize eDNA programs and interpret eDNA temporal trends within and among regions (Lacoursière-Roussel and Deiner 2021). It is well-known that detection rates vary depending on the sampling periods, field methods, and lab protocols (*e.g.*, Buxton et al. 2017; Deiner et al. 2018; Di Muri et al. 2022). The former is due to the fact that eDNA detection rates vary with both biotic (*e.g.*, organism abundance, metabolism, life-stage, distribution) and abiotic environmental factors (*e.g.*, water temperature, humic/tannic acid content, water flow; reviewed in Abbott et al. 2021; Rourke et al. 2021). While national and global efforts towards eDNA standardization are underway (Canadian Standards Association 2019), guidance is needed for end-users who currently use, or are considering using eDNA results to support day-to-day decision-making. The recent eDNA guidelines from DFO's Canadian Science Advisory Secretariat (CSAS; Abbott et al. 2021) was a first step toward general guidance for designing, conducting, reporting, and interpreting eDNA studies, but advice is general and does not provide information for specific ecosystems and taxa. Here we propose to complement the recent generalized eDNA guidelines by providing an overview of existing eDNA observations to guide decisions on appropriate sampling periods and interpretation of time-series for specific taxa and regions.

Currently, eDNA management decisions within DFO focus only on targeted eDNA approaches (*i.e.*, methods that selectively detect the DNA of a single species or taxon; DFO 2020), which is more established and less experimentally complex than semi-targeted approaches (*i.e.*, methods assessing whole communities, such as eDNA metabarcoding). Multi-species methods, however, are of growing interest for DFO. This data report is the first attempt to provide guidance on optimal sampling periods for both qPCR and metabarcoding eDNA studies. Like qPCR eDNA methods, metabarcoding can detect species presence in aquatic ecosystems, which can be useful in SAR assessments and tracking AIS range expansion (LeBlanc et al. 2020; Mahon et al. 2023); however, use of eDNA metabarcoding practices is still in its infancy. Primer and sequencing depth biases are difficult to quantify with metabarcoding as primers may bind better for specific taxonomic groups and skew the relative abundance of sequences in the sample (Deiner et al. 2017) and low sequencing depth could minimize the detection of less abundant species. Different protocols (*i.e.*, sample volume, filter type, primers, bioinformatic analyses) may also lead to different detection rates in the same environment (*e.g.*, Lacoursière-Roussel et al. 2016; Bourret et al. 2023). Thus, for a given species, comparing data obtained from different detection methods is not necessarily straightforward and should be done with caution.

This document aims to guide the development of eDNA-based research and/or monitoring programs based on observed optimal detection period(s) for taxonomic group(s) of interest, thereby maximizing resources and detection power to facilitate integration of eDNA results in management decisions. This report provides an overview of compiled DFO eDNA data obtained with qPCR and metabarcoding methods from 2015 through 2022 along the Canadian Atlantic coast and Eastern Arctic. Described herein are taxa detected from eDNA samples collected in both marine inshore and offshore environments. The spatial and temporal ranges of the data are provided to allow visualization of potential patterns in detection rates. This report is a first attempt to provide large-scale empirical evidence of successful DFO eDNA protocols. The data presented are observation data only, intended to allow inference of optimal detection periods based on taxa presence/absence, with no intent to provide quantitative population assessments. Finally, data are combined only when they are obtained via the same protocols, as different methods may influence the likelihood of organism DNA detection.

2. SUMMARY OF CONTENTS

This document reports eDNA results obtained from seven years of field sampling across the five major coastal marine biogeographic regions of the Canadian Atlantic coast and Eastern Arctic. The data are obtained from qPCR and DNA metabarcoding projects and are used to visualize species detection periods across a latitudinal range. A broad overview of each method is provided in its respective section.

3. MARINE ECOREGION CLASSIFICATIONS

Section summary: This section provides information on how marine ecoregions were classified for the purposes of this data report, describing how ecoregions were subdivided to provide greater resolution of eDNA data for species detection and biodiversity monitoring.

Marine ecoregions have historically been defined by different biogeographical classifications (Spalding et al. 2007). Where the DFO Canadian Science Advisory Secretariat (CSAS) has defined bioregions at the coarsest scale (DFO 2009), the project herein has adopted a more fine-scale approach due to the sensitivity of biological and physical factors that affect eDNA detection (Beng and Corlett 2020). We have therefore subdivided the biogeographic units defined by DFO based on Environment Canada's *Marine Ecological Classification System for Canada* (1994; Table 1; Figures 1-6). Ecoregions are classified as having similar physiographic, oceanographic, and biological characteristics that are further subdivided into ecodistricts based on variation in shelf depth, seasonal ice cover, current direction, and surface temperature (Environment Canada 1994). Parsing out ecodistrict differences may be important when interpreting eDNA data, such as delineating potential regional differences in eDNA for species detection.

Table 1. Northwest Atlantic and Eastern Arctic marine biogeographic classifications adapted from Environment Canada (1994) and DFO (2009).

<i>Ecoregion</i>	<i>Ecodistrict</i>
Eastern Arctic Shelf	Baffin Bay/Davis Strait
	Jones Sound
	Lancaster Sound
	North Baffin Fjords
	Parry Channel
	Gulf of Boothia
Hudson Bay Complex	Foxe Basin
	Hudson Strait
	Ungava Bay
	Hudson Bay
	James Bay
Newfoundland-Labrador Shelves	Newfoundland-Labrador Shelves
	Grand Banks
Gulf of St. Lawrence	North Gulf Shelf
	Laurentian Channel
	St. Lawrence River
	Magdalen Shallows
Scotian Shelf	Scotian Shelf
	Bay of Fundy

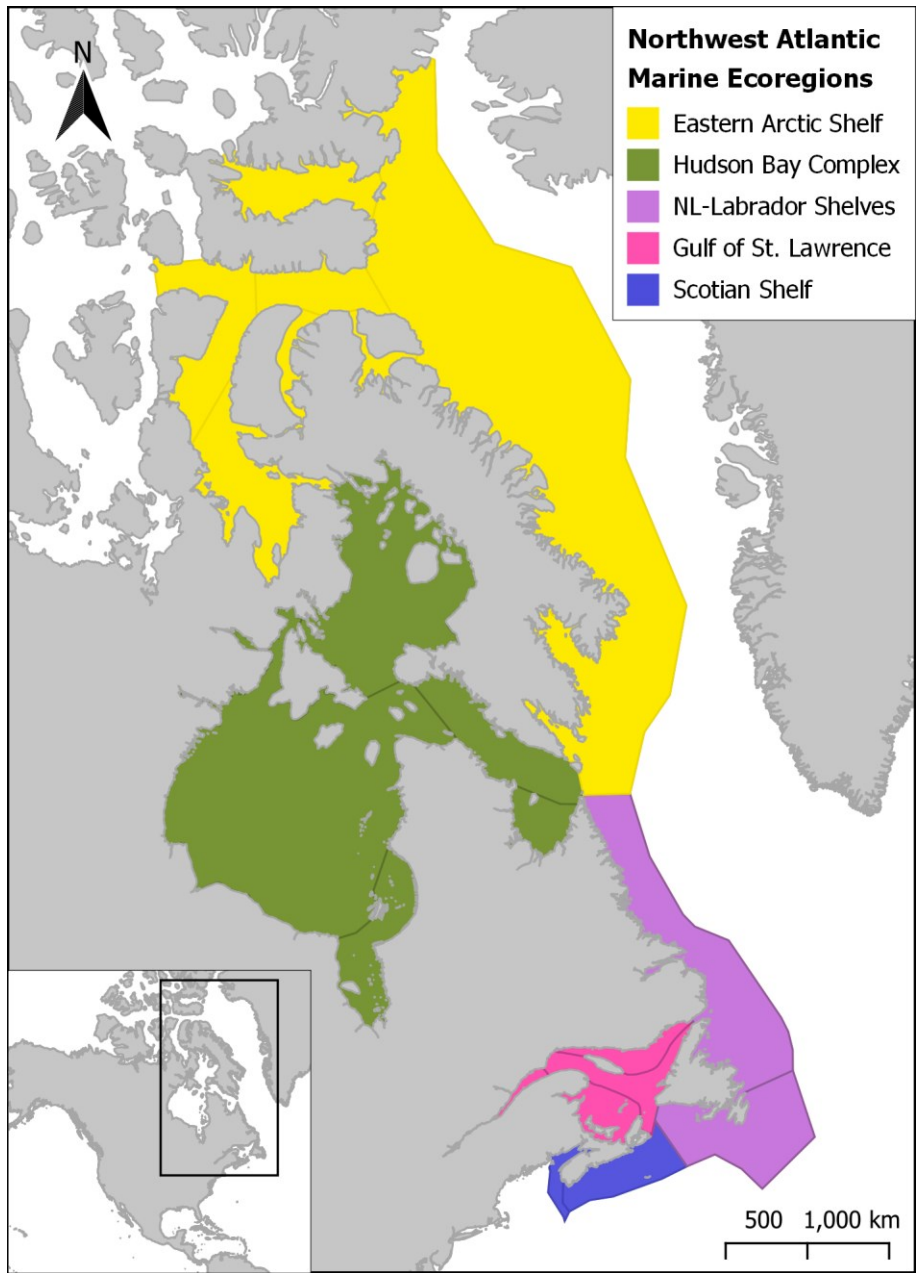


Figure 1. Broad marine ecoregions for the Northwest Atlantic and Eastern Arctic as per the DFO (2009) marine biogeographic classification system. The lines within each ecoregion represent the boundaries of the ecodistricts, with details shown in Figures 2-6.

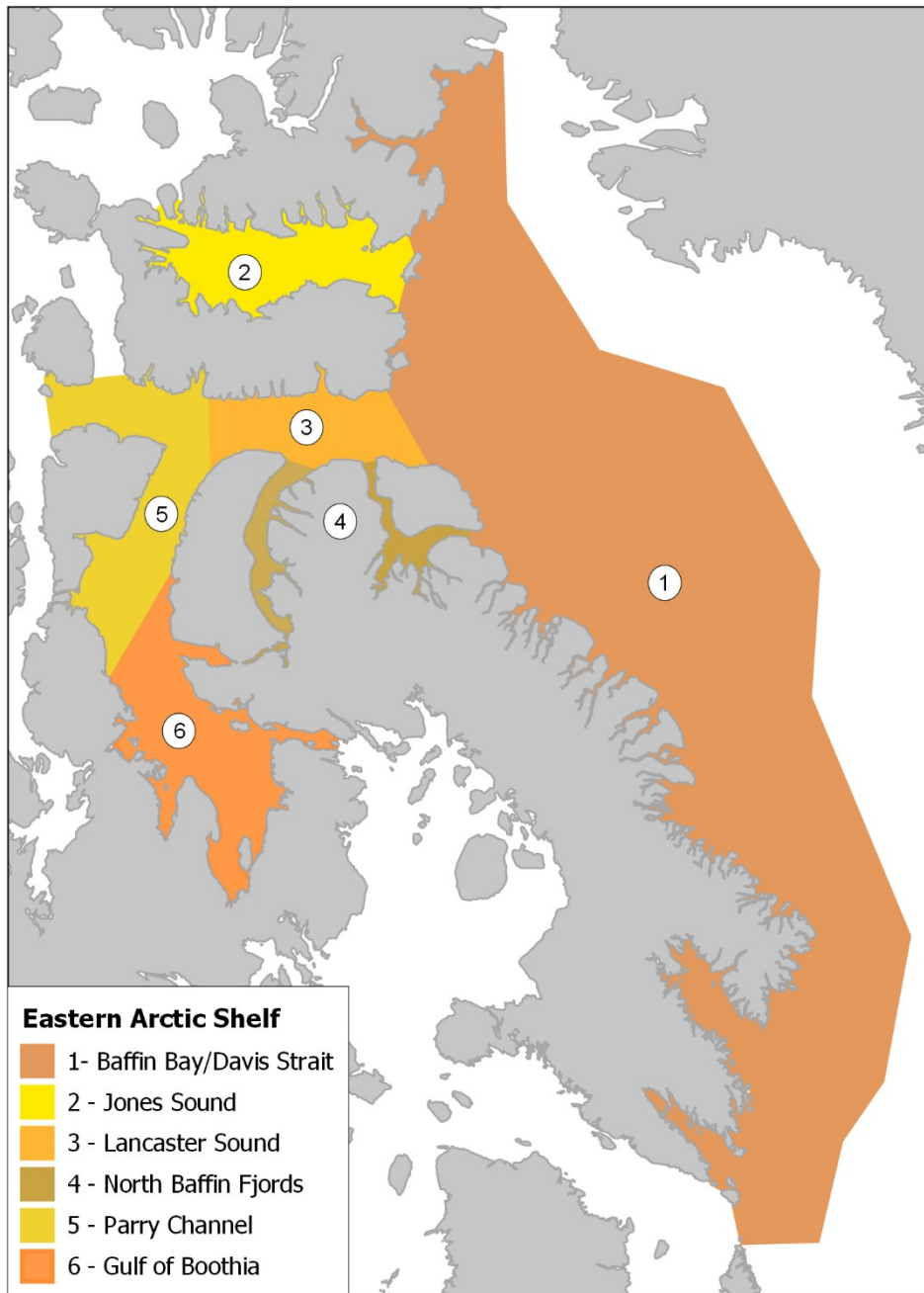


Figure 2. Ecodistricts of the Eastern Arctic Shelf ecoregion.

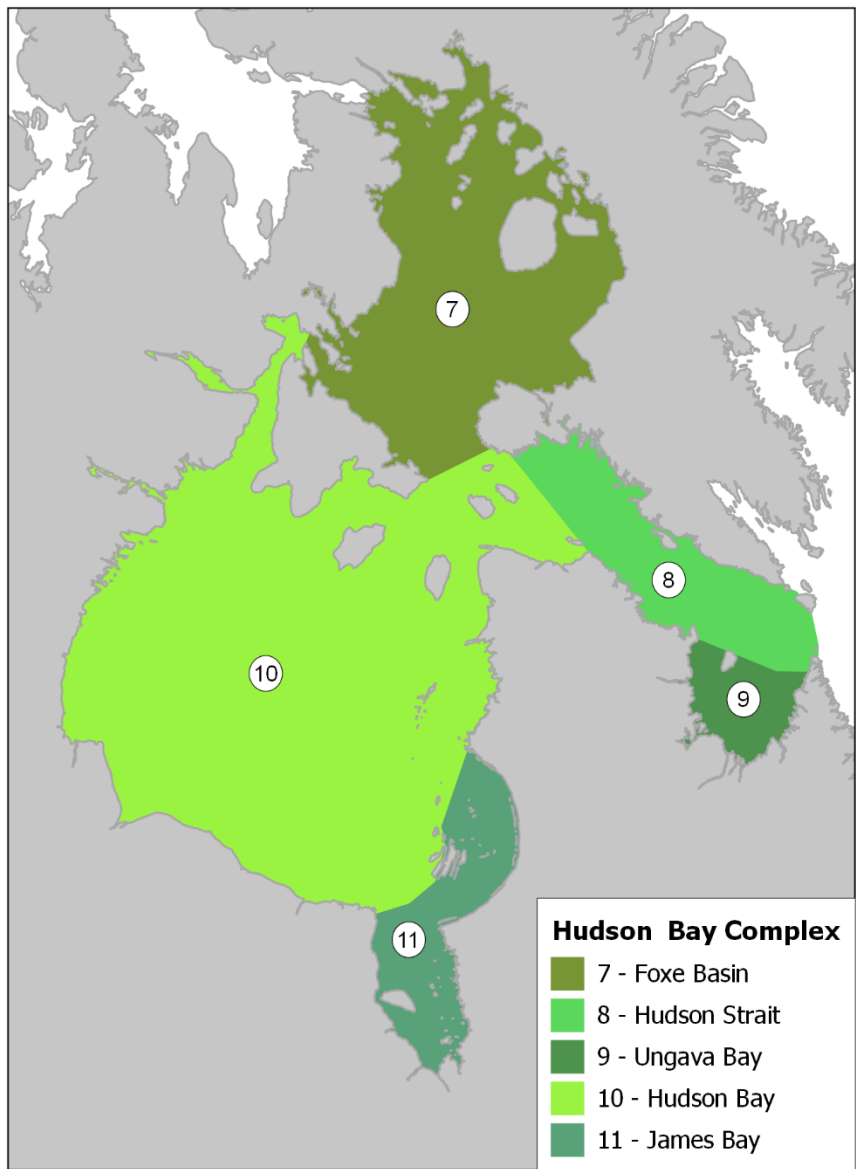


Figure 3. Ecodistricts of the Hudson Bay Complex ecoregion.

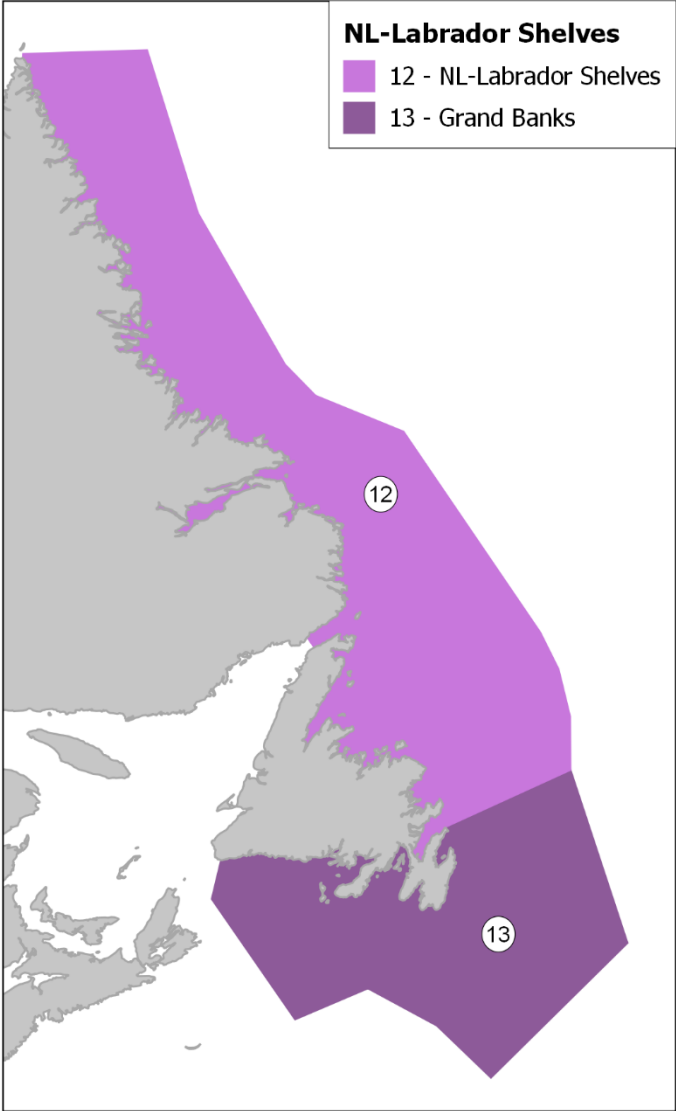


Figure 4. Ecodistricts of the Newfoundland-Labrador Shelves ecoregion.

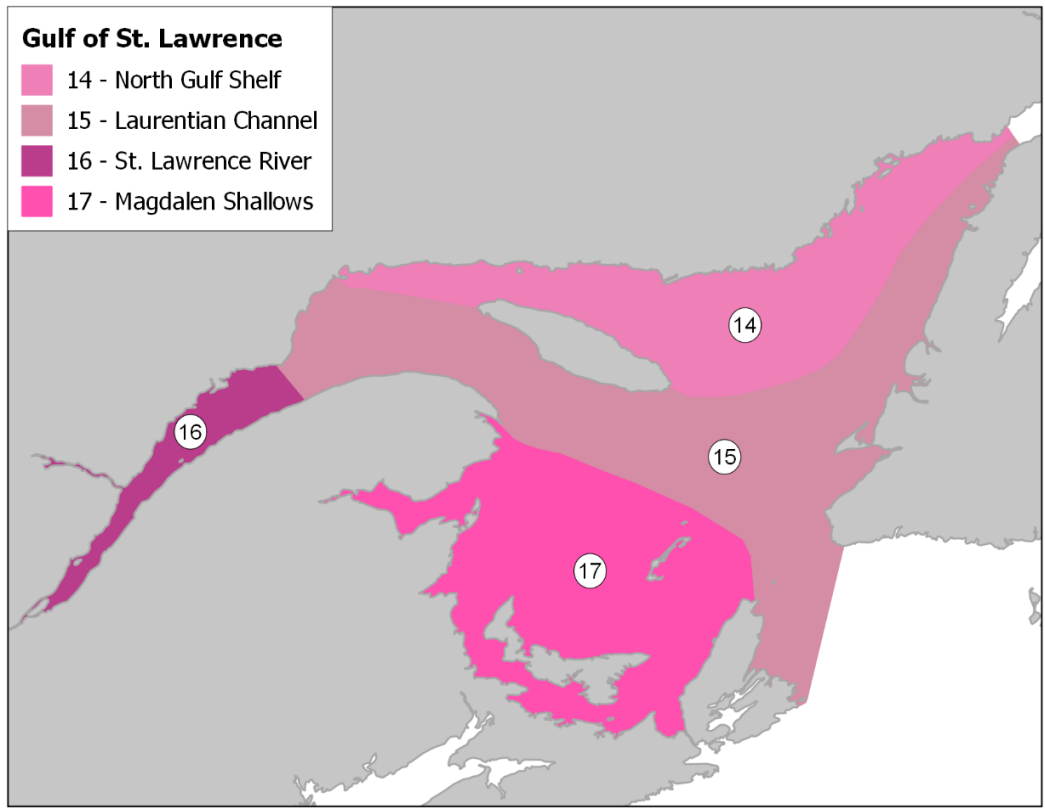


Figure 5. Ecodistricts of the Gulf of St. Lawrence ecoregion.

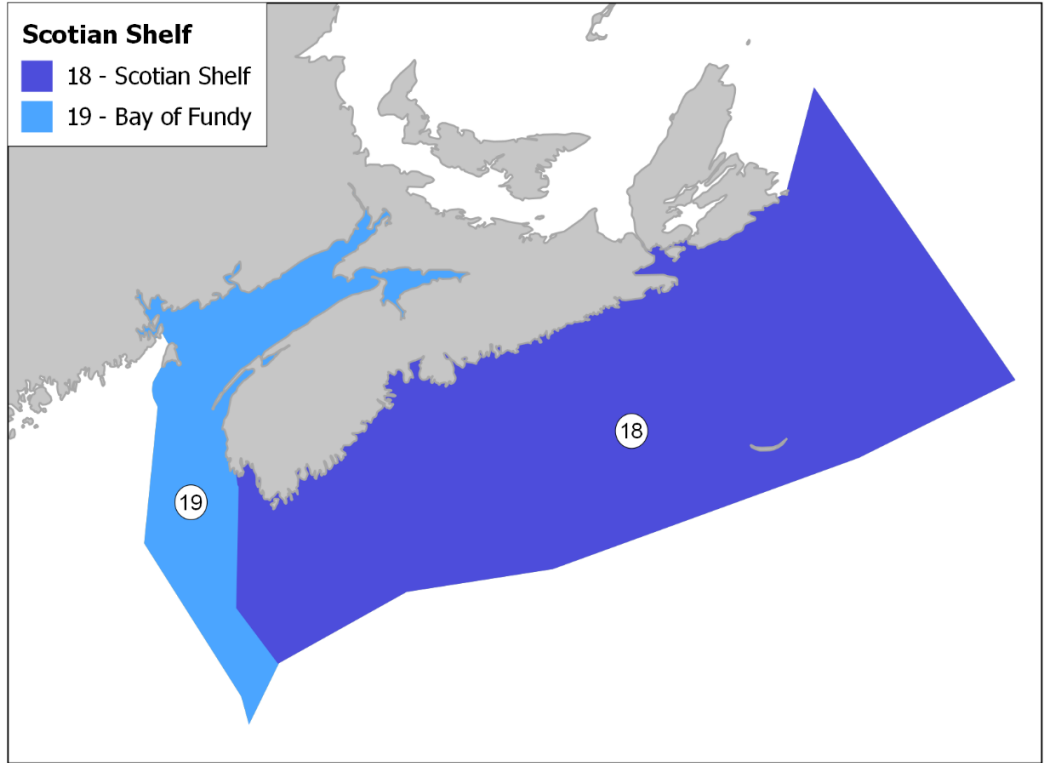


Figure 6. Ecodistricts of the Scotian Shelf ecoregion.

4. DESCRIPTION OF DATA

Section summary: This section provides information on how data were compiled. The *project definition* subsection describes how an eDNA project (*i.e.*, method) is defined for the purposes of this report. Summaries of data and sampling effort for each qPCR and metabarcoding project are presented in Section 5 and Section 6, respectively.

4.1 PROJECT DEFINITION

The data herein originated from a total of 24 distinct DFO projects that have been compiled; metabarcoding ($n = 15$) and qPCR ($n = 9$) projects are compiled separately. For the purposes of this report, a “project” is defined when field and lab methods are exactly the same across surveys. In order to compare eDNA data, protocols need to be identical as changes in methods can alter the amount of eDNA that is captured at every step through sample collection, filtration, and DNA extraction (*e.g.*, Kumar et al. 2022). For information regarding project methods, please contact the project’s data steward (Tables 2, 3).

5. qPCR

Section summary: This section provides information on how eDNA data from qPCR projects were prepared for analysis. Following the data and sampling effort summary are the full qPCR species detection heatmaps for all projects.

Quantitative polymerase chain reaction-based methods have been successfully employed to detect AIS (LeBlanc et al. 2020) and SAR (Rojahn et al. 2021). The targeted species’ DNA is detected by amplifying a short sequence of DNA (*i.e.*, barcode) that, while highly conserved within taxa, generally diverges among closely related species (Hebert et al. 2003a). A commonly used barcode for eDNA qPCR methods is the mitochondrial cytochrome *c* oxidase subunit I (COI) gene (Hebert et al. 2003b), although other gene targets are also used (*e.g.*, cytochrome *b* [cytb], NADH dehydrogenase subunit 5 [NAD5]; Rourke et al. 2021).

In the present report, qPCR projects targeted the mitochondrial COI, cytb, and NAD5 genes. All qPCR assays were previously validated to ensure species specificity. Contamination was assessed in all projects during field collection, filtration, DNA extraction, and in qPCR with negative controls of distilled or ultrapure water. We are reporting only what is detected with confidence (*i.e.*, all qPCR replicates > limit of detection [LOD]). Species were classified as not detected if there was no successful amplification, if one or more of the qPCR replicates amplified but DNA copy numbers were below the LOD, or less than 100% of qPCR replicates had copy numbers above the LOD.

Relative success of detection was calculated as the percentage of positive samples for each taxonomic level, grouped by project, year, and ecodistrict. Finally, *no data* indicates that the sampling did not occur, whereas *not detected* indicates that the taxon was not detected with the method used; monthly sampling effort is included for each species, project, and year (Figures 7-12). Note that number of qPCR samples does not include field blanks nor positive and negative laboratory controls (*i.e.*, filtration, extraction, qPCR).

Table 2. Summary of environmental DNA (eDNA) quantitative polymerase chain reaction (qPCR) projects, sampling effort, and data collected. Note that number of qPCR samples does not include laboratory controls (*i.e.*, filtration, extraction, qPCR). See Figures 7-12 for visual representation of monthly sampling effort for each project and year.

Project ID	Data Provider	Data Steward	Reference (if applicable)	Sampling Year	Ecodistrict	Sampling Design Summary (<i>n</i>)				Target Species
						Sampling Sites	Field Samples	Samples Per Site (min-max)	qPCR Samples	
1	DFO Maritimes	Lacoursière, A. ^a		2018	Bay of Fundy	4	76	14 - 24	228	<i>Homarus americanus</i>
3	DFO Maritimes	Lacoursière, A.		2019	Bay of Fundy	18	18	1	378	<i>Carcharodon carcharias</i> <i>Halichoerus grypus</i> <i>Isurus oxyrinchus</i> <i>Lamna nasus</i> <i>Pagophilus groenlandicus</i> <i>Phoca vitulina</i> <i>Thunnus thynnus</i>
					Bay of Fundy	18	36	2	756	<i>Carcharodon carcharias</i> <i>Isurus oxyrinchus</i> <i>Halichoerus grypus</i> <i>Lamna nasus</i> <i>Pagophilus groenlandicus</i> <i>Phoca vitulina</i> <i>Thunnus thynnus</i>
				2020	Grand Banks	12	83	1 - 17	1743	<i>Carcharodon carcharias</i> <i>Isurus oxyrinchus</i> <i>Halichoerus grypus</i> <i>Lamna nasus</i> <i>Pagophilus groenlandicus</i> <i>Phoca vitulina</i> <i>Thunnus thynnus</i>
					Bay of Fundy	25	96	2 - 5	1608	<i>Carcharodon carcharias</i> <i>Halichoerus grypus</i> <i>Isurus oxyrinchus</i> <i>Lamna nasus</i> <i>Pagophilus groenlandicus</i> <i>Phoca vitulina</i> <i>Prionace glauca</i> <i>Thunnus thynnus</i>

Table 2. (Continued)

Project ID	Data Provider	Data Steward	Reference (if applicable)	Sampling Year	Ecodistrict	Sampling Design Summary (n)				Target Species
						Sampling Sites	Field Samples	Samples Per Site (min-max)	qPCR Samples	
3	DFO Maritimes	Lacoursière, A.		2021	Grand Banks	5	74	10 - 16	756	<i>Carcharodon carcharias</i> <i>Halichoerus grypus</i> <i>Isurus oxyrinchus</i> <i>Lamna nasus</i> <i>Thunnus thynnus</i> <i>Pagophilus groenlandicus</i> <i>Phoca vitulina</i> <i>Prionace glauca</i>
				2022	Bay of Fundy	24	215	5 - 10	916	<i>Carcharodon carcharias</i> <i>Lamna nasus</i> <i>Halichoerus grypus</i> <i>Phoca vitulina</i>
5	DFO Gulf	LeBlanc, F. ^b	LeBlanc et al. (2020)	2017	Magdalen Shallows	4	20	4	240	<i>Botrylloides violaceus</i> <i>Botryllus schlosseri</i> <i>Ciona intestinalis</i> <i>Styela clava</i>
				2018	Bay of Fundy	2	8	4	96	<i>Asciidiella aspersa</i> <i>Caprella mutica</i> <i>Ciona intestinalis</i> <i>Didemnum vexillum</i> <i>Diplosoma listerianum</i> <i>Membranipora membranacea</i> <i>Styela clava</i>

Table 2. (Continued)

Project ID	Data Provider	Data Steward	Reference (if applicable)	Sampling Year	Ecodistrict	Sampling Design Summary (n)				Target Species
						Sampling Sites	Field Samples	Samples Per Site (min-max)	qPCR Samples	
5	DFO Gulf	LeBlanc, F.	LeBlanc et al. (2020)	2018	Magdalen Shallows	5	65	4 - 34	792	<i>Asciidiella aspersa</i> <i>Botrylloides violaceus</i> <i>Botryllus schlosseri</i> <i>Caprella mutica</i> <i>Ciona intestinalis</i> <i>Didemnum vexillum</i> <i>Diplosoma listerianum</i> <i>Membranipora membranacea</i> <i>Styela clava</i>
					Scotian Shelf	2	8	4	48	<i>Asciidiella aspersa</i> <i>Caprella mutica</i> <i>Didemnum vexillum</i> <i>Diplosoma listerianum</i> <i>Membranipora membranacea</i> <i>Styela clava</i>
6	DFO Québec	Parent, G. ^c	Chevriniais et al. (2023; for <i>C. maenas</i> only)	2021	Magdalen Shallows	3	15	3 - 6	120	<i>Ciona intestinalis</i>
				2022	Magdalen Shallows	8	81	6 - 15	461	<i>Carcinus maenas</i> <i>Ciona intestinalis</i> <i>Styela clava</i>

^a Lacoursière, A., anais.lacoursiere@dfo-mpo.gc.ca, Fisheries and Oceans Canada, Maritimes Region

^b LeBlanc, F., francis.leblanc@dfo-mpo.gc.ca, Fisheries and Oceans Canada, Gulf Region

^c Parent, G., genevieve.parent@dfo-mpo.gc.ca, Fisheries and Oceans Canada, Québec Region

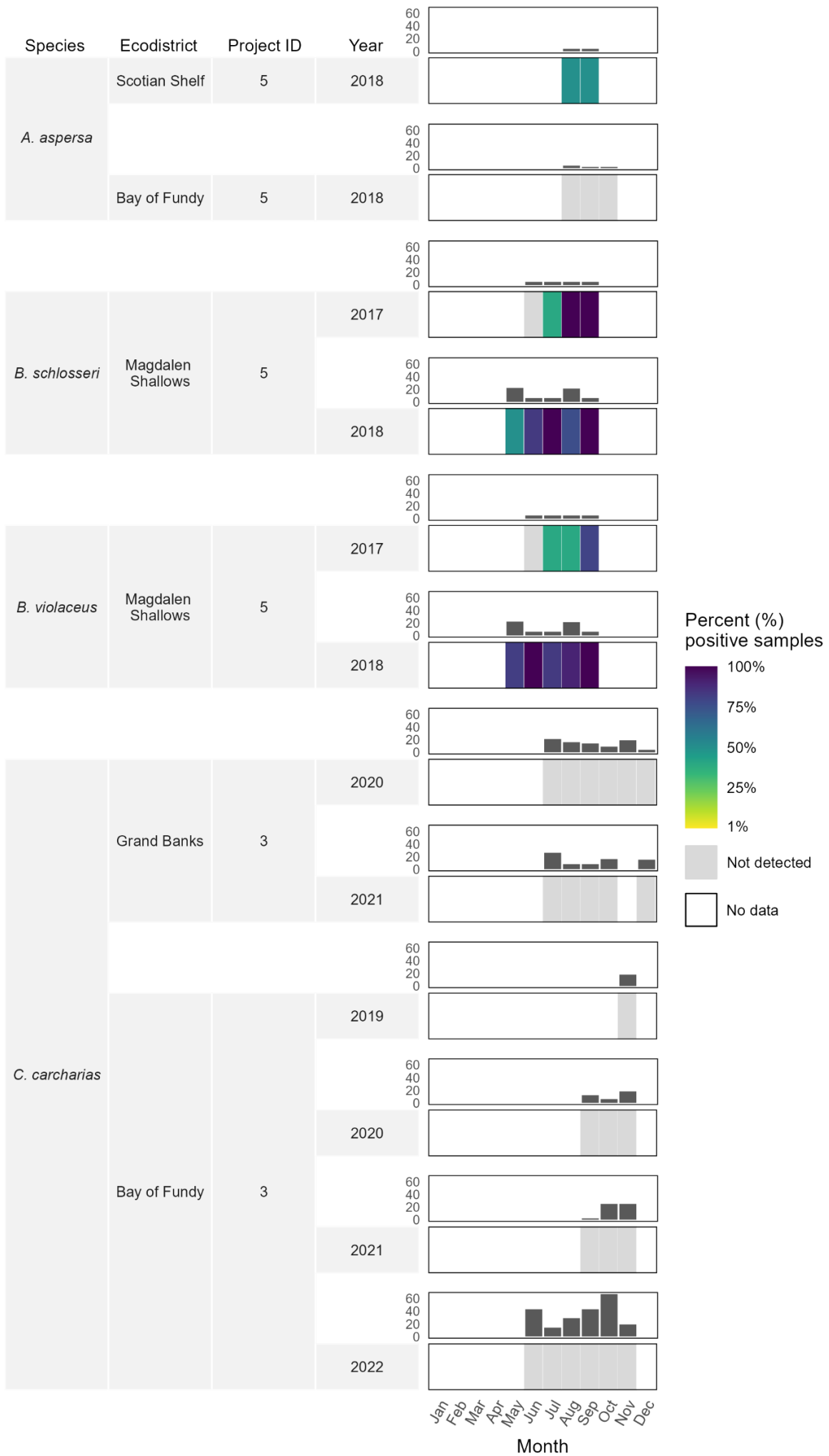


Figure 7. Proportion of positive environmental DNA (eDNA) detections per month from quantitative polymerase chain reaction (qPCR) projects for marine species *Ascidiella aspersa*, *Botryllus schlosseri*, *Botrylloides violaceus*, and *Carcharodon carcharias*. Species are distinguished by sample year and ecodistrict. Two panels are presented for each year. The number of samples collected per month is presented in the upper panel while the proportion of positive samples is presented in the lower panel. In the lower panel, the light grey indicates that the qPCR assay can successfully amplify the taxon and resulted in non-detection or concentrations below the limit of detection whereas white indicates no sampling.

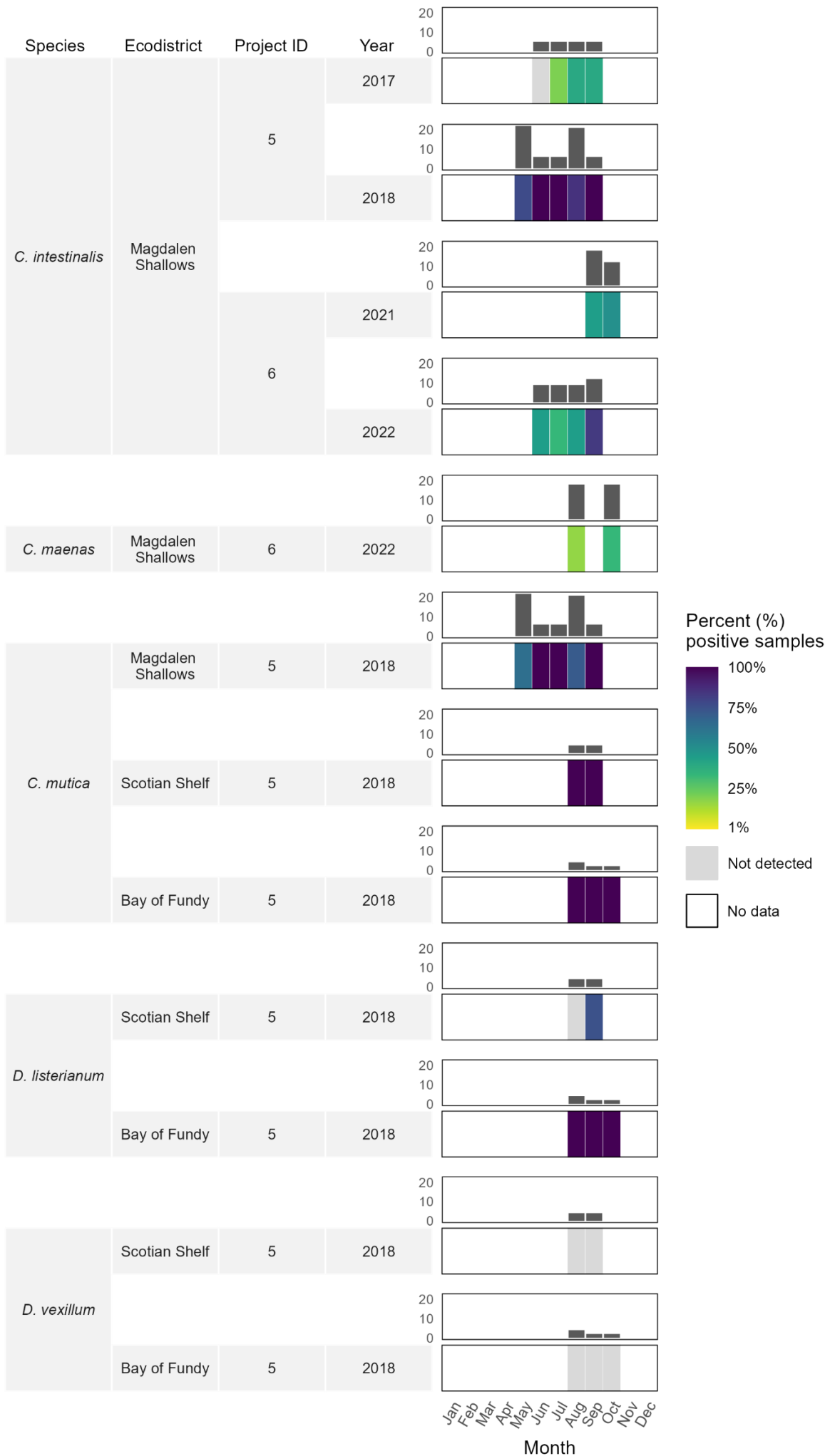


Figure 8. Proportion of positive environmental DNA (eDNA) detections per month from quantitative polymerase chain reaction (qPCR) projects for marine species *Ciona intestinalis*, *Carcinus maenas*, *Caprella mutica*, *Diplosoma listerianum*, and *Didemnum vexillum*. Species are distinguished by sample year and ecodistrict. Two panels are presented for each year. The number of samples collected per month is presented in the upper panel while the proportion of positive samples is presented in the lower panel. In the lower panel, the light grey indicates that the qPCR assay can successfully amplify the taxon and resulted in non-detection or concentrations below the limit of detection whereas white indicates no sampling.

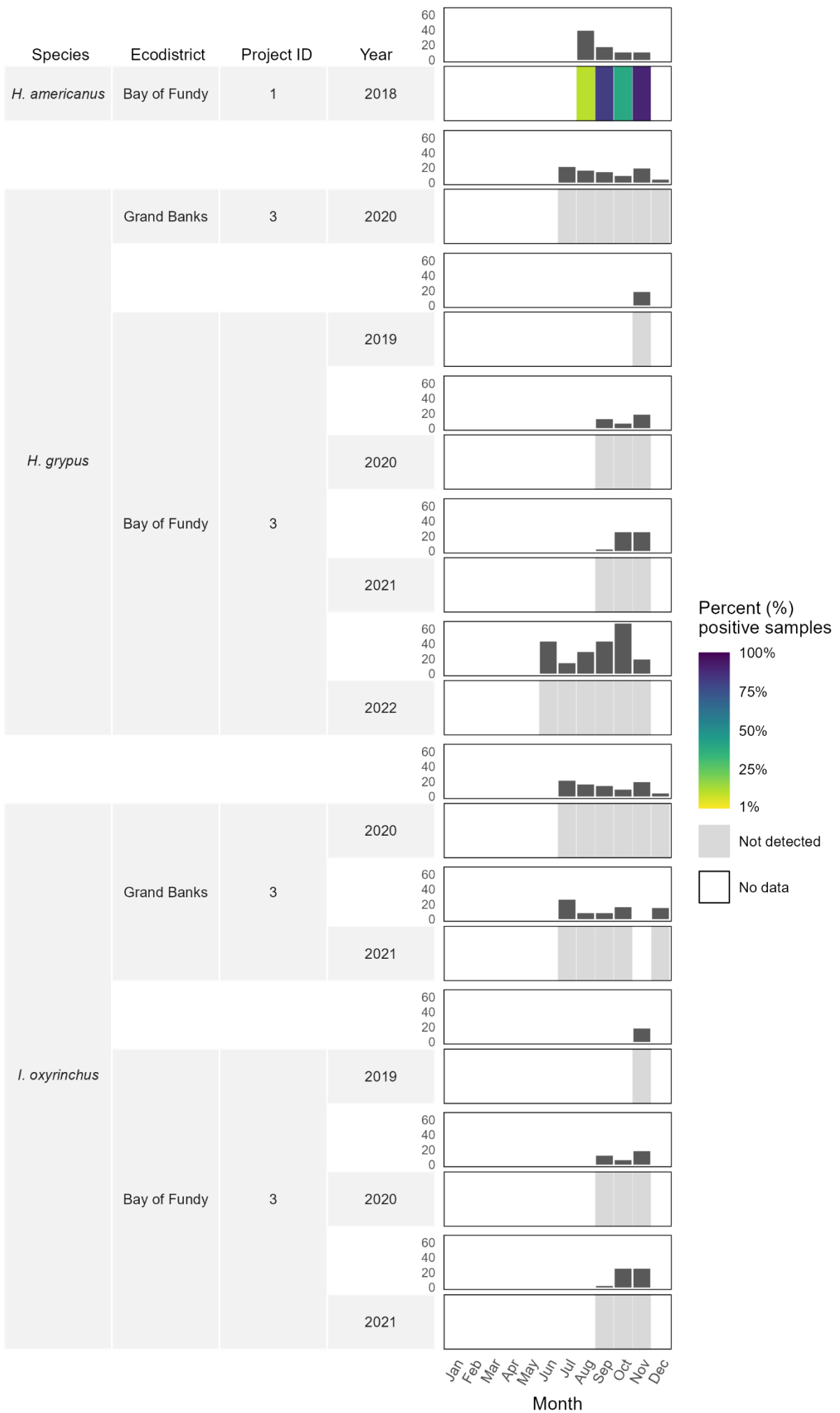


Figure 9. Proportion of positive environmental DNA (eDNA) detections per month from quantitative polymerase chain reaction (qPCR) projects for marine species *Homarus americanus*, *Halichoerus grypus*, and *Isurus oxyrinchus*. Species are distinguished by sample year and ecodistrict. Two panels are presented for each year. The number of samples collected per month is presented in the upper panel while the proportion of positive samples is presented in the lower panel. In the lower panel, the light grey indicates that the qPCR assay can successfully amplify the taxon and resulted in non-detection or concentrations below the limit of detection whereas white indicates no sampling.

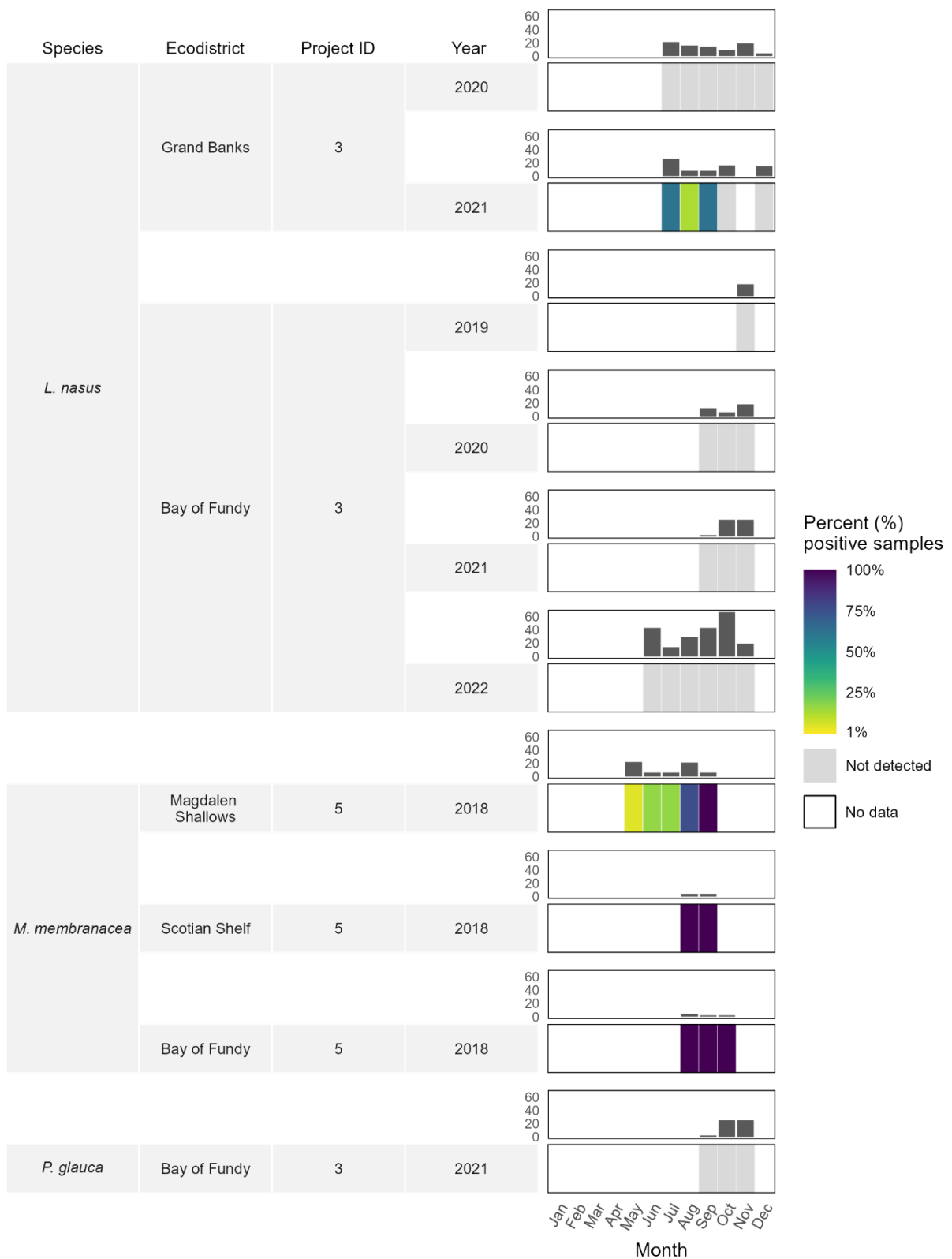


Figure 10. Proportion of positive environmental DNA (eDNA) detections per month from quantitative polymerase chain reaction (qPCR) projects for marine species *Lamna nasus*, *Membranipora membranacea*, and *Prionace glauca*. Species are distinguished by sample year and ecodistrict. Two panels are presented for each year. The number of samples collected per month is presented in the upper panel while the proportion of positive samples is presented in the lower panel. In the lower panel, the light grey indicates that the qPCR assay can successfully amplify the taxon and resulted in non-detection or concentrations below the limit of detection whereas white indicates no sampling.

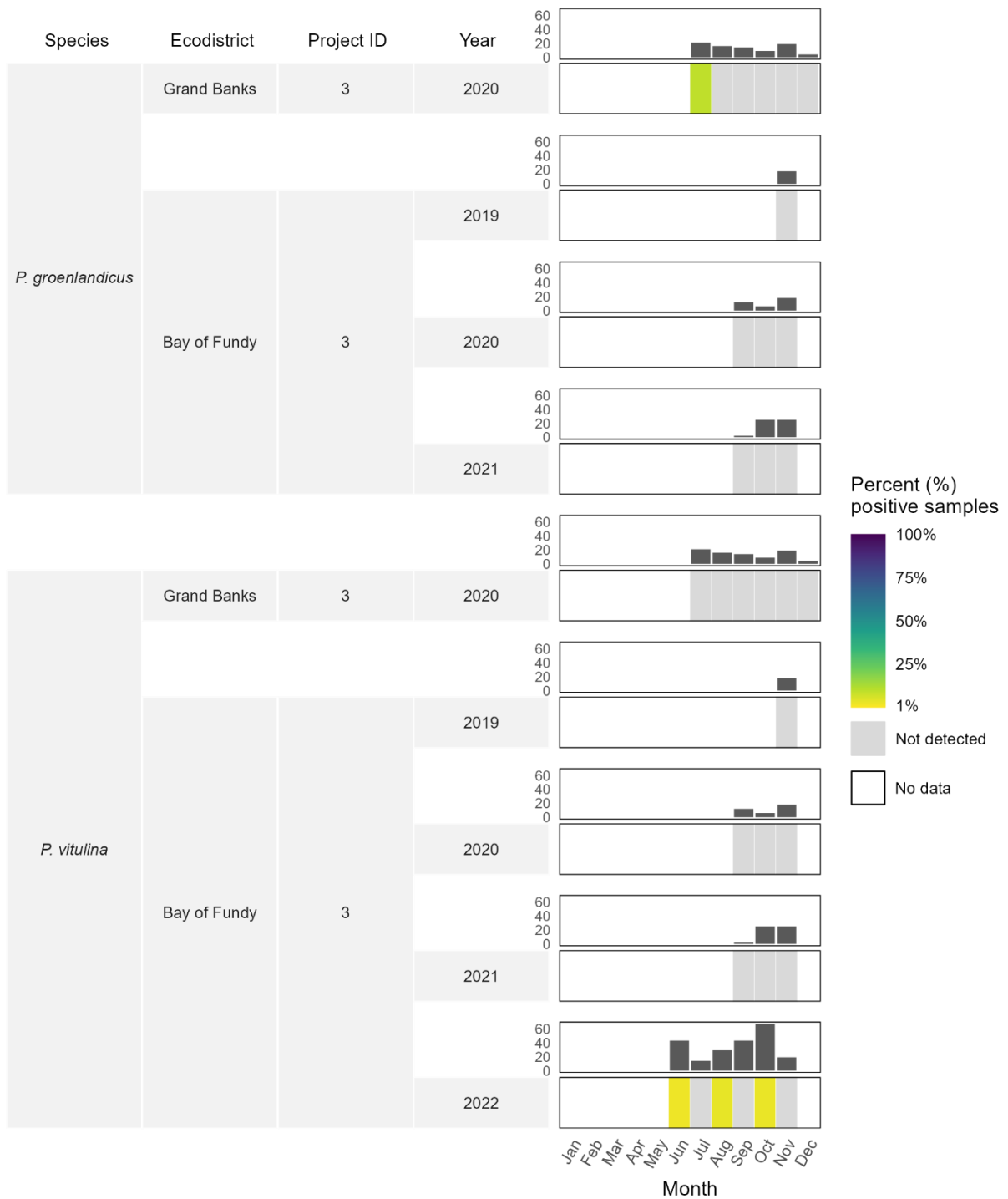


Figure 11. Proportion of positive environmental DNA (eDNA) detections per month from quantitative polymerase chain reaction (qPCR) projects for marine species *Pagophilus groenlandicus* and *Phoca vitulina*. Species are distinguished by sample year and ecodistrict. Two panels are presented for each year. The number of samples collected per month is presented in the upper panel while the proportion of positive samples is presented in the lower panel. In the lower panel, the light grey indicates that the qPCR assay can successfully amplify the taxon and resulted in non-detection or concentrations below the limit of detection whereas white indicates no sampling.

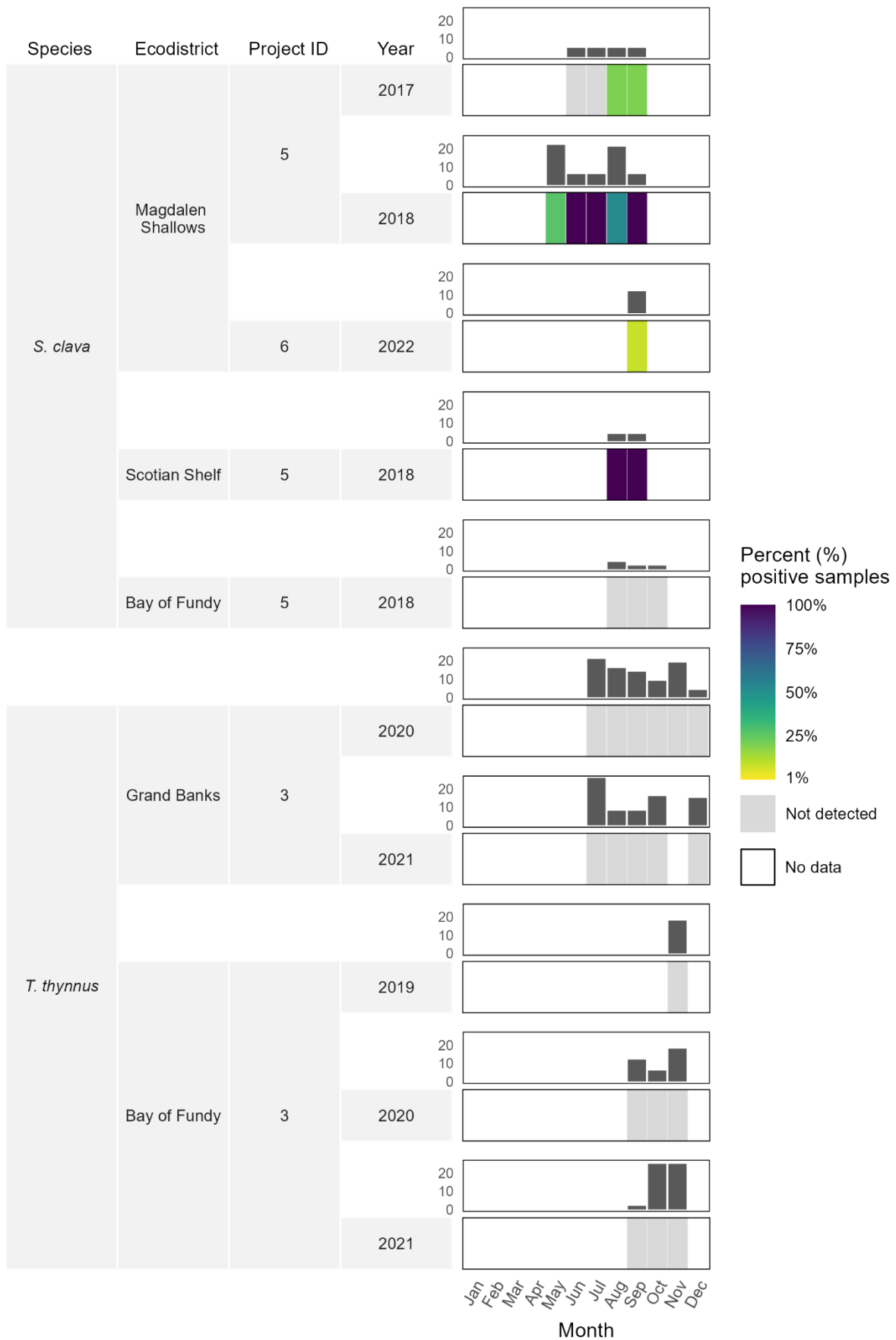


Figure 12. Proportion of positive environmental DNA (eDNA) detections per month from quantitative polymerase chain reaction (qPCR) projects for marine species *Styela clava* and *Thunnus thynnus*. Species are distinguished by sample year and ecodistrict. Two panels are presented for each year. The number of samples collected per month is presented in the upper panel while the proportion of positive samples is presented in the lower panel. In the lower panel, the light grey indicates that the qPCR assay can successfully amplify the taxon and resulted in non-detection or concentrations below the limit of detection whereas white indicates no sampling.

6. DNA METABARCODING

Section summary: This section provides information on the use of metabarcoding in eDNA biodiversity monitoring. Following the data and sampling effort summaries for each project are the full DNA metabarcoding genus detection heatmaps.

DNA metabarcoding relies on a series of bioinformatic steps to identify various species of multiple taxa from DNA samples, each of which require multiple quality assurance validations to minimize processing errors (Deiner et al. 2017). Metabarcoding targets a segment of DNA (*i.e.*, barcode) that is present and similar across most taxa. Similar to qPCR, the mitochondrial cytochrome *c* oxidase subunit I (COI) gene is the standard DNA barcode (Hebert et al. 2003b), though other genes are often targeted when COI is unable to differentiate taxonomic groups (Deagle et al. 2014). Differentiating marine invertebrates, for example, can be difficult because microscopic eukaryotes are often under-represented in public sequence databases (Bik et al. 2012). Efforts to improve taxonomic assignments in metabarcoding studies using curated reference databases are ongoing (*e.g.*, DeWaard et al. 2019; Bourret et al. 2023).

In the present report, metabarcoding projects targeted subfragments of the mitochondrial genome (COI, 12S ribosomal RNA [rRNA], and 16S rRNA) as well as subfragments of the nuclear gene encoding the 18S rRNA subunit to provide a good scope for biodiversity monitoring and visualize which primers performed best for each taxonomic group. Taxonomic assignments were performed for each project using either the Barque pipeline (v1.7.2, v1.7.3, v1.7.4; Mathon et al. 2021), Maurice Lamontagne Institute pipeline (DFO, unpublished data), or QIIME 2 (Bolyen et al. 2019; Table 3). The lowest taxonomic level reported is genus due to the possibility of species mismatches or identical DNA sequences among species within a genus, as the reference databases have not been vetted for accuracy of all sequence identifications (Leray et al. 2019). Note that for taxonomic groups having historically been less well-described, reporting at the phylum level may present stronger optimal detection periods (Bik et al. 2012). Finally, genus data for phylum Chordata are separated by class to represent more closely related taxonomic groups.

Relative success of detection was calculated as the percentage of positive samples for each genus, grouped by project, year, ecodistrict, and target gene subfragment (*i.e.*, primers). For the purpose of this report, genera were classified as detected when there was at least one sequence detected (*i.e.*, operational taxonomic units or amplicon sequence variants > 0). Finally, all metabarcoding occurrences containing multiple taxonomic assignments as determined by the reference database were removed from analysis due to sequence similarity (*i.e.*, more than one potential genus per DNA sequence). Detections are more reliable when results have been corroborated from different primers and when results show a high number of reads/sequences. Interpretation from metabarcoding data should thus be done with care and this report aims to provide only a high-level overview of taxonomic spatiotemporal trends for data that have been generated to date by DFO eDNA experts.

No data depicts that either 1) the sampling did not occur or 2) the genus was not detected in the project of the given primer. The latter might represent that the genus might not be present at that location, that the method is inefficient (*i.e.*, false negative), or the sequence was not available in the sequence reference database used within the bioinformatic pipeline. *Not detected* depicts that the genus has been detected with the same method and primers, but was not detected in the project at that specific time (Figure 13). Finally, a target subfragment being absent for the genus indicates that the primers were either not used or the method was not efficient for that specific genus within that specific project (see Table 3 for details).

Table 3. Summary of environmental DNA (eDNA) metabarcoding projects, sampling effort, and data collected. Sampling effort displayed is total number of monthly samples for each ecodistrict and year, excluding field blanks. Target genes: cytochrome c oxidase subunit I (COI; subfragments COI-1, COI-2), 12S ribosomal RNA (rRNA; 12S-248, 12S-MiFishU, 12S-MiMam), 16S rRNA (16S, 16S-chord), 18S rRNA (18S; subfragments 18S-1, 18S-2). Taxonomic hierarchies were obtained from the World Register of Marine Species (WoRMS Editorial Board 2022).

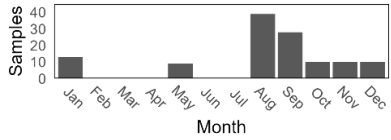
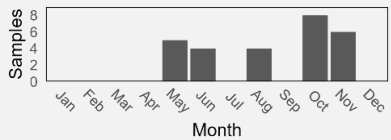
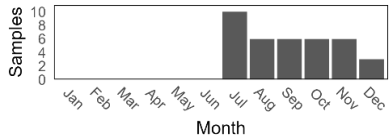
Project	Data Provider	Data Steward	Reference (if applicable)	Sampling Year	Ecodistrict	Sampling Design Summary (<i>n</i>)			Target Gene Subfragment	Phyla Detected
						Sampling Sites	Total Field Samples	Samples Per Site (min - max)		
1	DFO Maritimes	Lacoursière, A. ^a		2018	Bay of Fundy	9	133	5 - 24	COI-1	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Echinodermata, Mollusca, Nematoda, Nemertea, Platyhelminthes, Porifera, Priapulida, Rotifera
									COI-2	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Mollusca, Nemertea, Porifera, Rotifera
									18S-1	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Ctenophora, Echinodermata, Entoprocta, Hemichordata, Kinorhyncha, Mollusca, Nemertea, Platyhelminthes, Porifera, Rotifera
									18S-2	Annelida, Arthropoda, Brachiopoda, Bryozoa, Chordata, Cnidaria, Ctenophora, Echinodermata, Entoprocta, Gastrotricha, Hemichordata, Kinorhyncha, Mollusca, Nemertea, Platyhelminthes, Porifera, Rotifera, Tardigrada, Xenacoelomorpha
2	DFO Maritimes, DFO Arctic	Lacoursière, A., Howland, K. ^b		2019	Baffin Bay/Davis Strait	4	27	4 - 11	COI-1	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Echinodermata, Mollusca, Nemertea, Porifera, Priapulida, Rotifera
									12S-248	Chordata
									16S	Chordata, Cnidaria, Echinodermata
									18S-2	Annelida, Arthropoda, Brachiopoda, Bryozoa, Chordata, Cnidaria, Ctenophora, Echinodermata, Mollusca, Nematoda, Nemertea, Platyhelminthes, Porifera
				2019	Bay of Fundy	2	37	17 - 20	COI-1	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Echinodermata, Mollusca, Nemertea, Platyhelminthes, Porifera, Priapulida, Rotifera
									12S-248	Chordata, Echinodermata
									16S	Chordata, Cnidaria, Echinodermata
									18S-2	Annelida, Arthropoda, Brachiopoda, Bryozoa, Chordata, Cnidaria, Ctenophora, Echinodermata, Hemichordata, Mollusca, Nemertea, Platyhelminthes, Porifera, Rotifera

Table 3. (Continued)

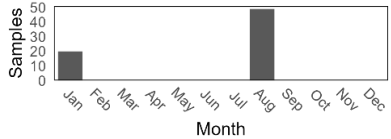
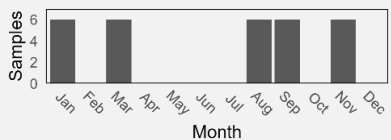
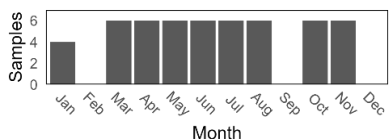
Project	Data Provider	Data Steward	Reference (if applicable)	Sampling Year	Ecodistrict	Sampling Design Summary (<i>n</i>)			Target Gene Subfragment	Phyla Detected
						Sampling Sites	Field Samples	Samples Per Site (min-max)		
2	DFO Maritimes, DFO Arctic	Lacoursière, A., Howland, K.		2019	Magdalen Shallows	6	69	9 - 20	COI-1	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Echinodermata, Mollusca, Nemertea, Platyhelminthes, Porifera, Rotifera, Xenacoelomorpha
										
						COI-2	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Mollusca, Platyhelminthes, Porifera			
								18S-1		
				18S-2	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Ctenophora, Echinodermata, Hemichordata, Mollusca, Nematoda, Nemertea, Platyhelminthes, Porifera, Rotifera, Xenacoelomorpha					
						2020	Bay of Fundy	2	30	15
										
				18S-2	Annelida, Arthropoda, Brachiopoda, Bryozoa, Chordata, Cnidaria, Ctenophora, Echinodermata, Hemichordata, Mollusca, Nemertea, Platyhelminthes, Porifera, Rotifera					
						2021	Bay of Fundy	2	52	26
										
18S-2	Annelida, Arthropoda, Brachiopoda, Bryozoa, Chordata, Cnidaria, Ctenophora, Echinodermata, Hemichordata, Mollusca, Nematoda, Nemertea, Platyhelminthes, Porifera, Rotifera									

Table 3. (Continued)

Project	Data Provider	Data Steward	Reference (if applicable)	Sampling Year	Ecodistrict	Sampling Design Summary (n)			Target Gene Subfragment	Phyla Detected	
						Sampling Sites	Field Samples	Samples Per Site (min-max)			
4	DFO Arctic	Howland, K.	Lacoursière et al. (2018); Leduc et al. (2019)	2015	Baffin Bay/Davis Strait	1	98	98		COI-1	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Echinodermata, Mollusca, Nemertea, Porifera, Priapulida, Rotifera
										COI-2	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Echinodermata, Mollusca, Nemertea, Porifera, Rotifera
										18S-1	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Ctenophora, Entoprocta, Gastrotricha, Gnathifera, Hemichordata, Mollusca, Nematoda, Nemertea, Platyhelminthes, Porifera, Rotifera, Tardigrada
										18S-2	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Ctenophora, Echinodermata, Entoprocta, Gastrotricha, Gnathifera, Hemichordata, Mollusca, Nematoda, Nemertea, Platyhelminthes, Porifera, Priapulida, Rotifera, Tardigrada, Xenacoelomorpha
					Hudson Bay	1	71	71		COI-1	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Echinodermata, Mollusca, Nemertea, Porifera, Priapulida, Rotifera
										COI-2	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Echinodermata, Mollusca, Nemertea, Porifera, Priapulida, Rotifera
										18S-1	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Ctenophora, Entoprocta, Gastrotricha, Gnathifera, Hemichordata, Mollusca, Nematoda, Nemertea, Platyhelminthes, Porifera, Priapulida, Rotifera, Tardigrada
										18S-2	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Ctenophora, Echinodermata, Entoprocta, Gastrotricha, Gnathifera, Hemichordata, Mollusca, Nematoda, Nemertea, Platyhelminthes, Porifera, Priapulida, Rotifera, Tardigrada, Xenacoelomorpha

Table 3. (Continued)

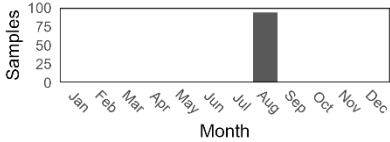
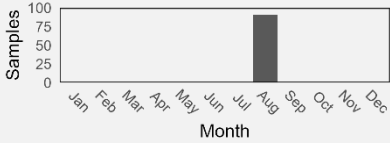
Project	Data Provider	Data Steward	Reference (if applicable)	Sampling Year	Ecodistrict	Sampling Design Summary (<i>n</i>)			Target Gene Subfragment	Phyla Detected				
						Sampling Sites	Field Samples	Samples Per Site (min-max)						
4	DFO Arctic	Howland, K.	Lacoursière et al. (2018); Leduc et al. (2019)	2016	Hudson Bay	1	94	94		COI-1	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Echinodermata, Mollusca, Nemertea, Porifera, Priapulida, Rotifera			
										COI-2	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Echinodermata, Mollusca, Nemertea, Porifera, Rotifera			
										18S-1	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Ctenophora, Entoprocta, Gastrotricha, Gnathifera, Hemichordata, Mollusca, Nematoda, Nemertea, Platyhelminthes, Porifera, Priapulida, Rotifera, Tardigrada			
										18S-2	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Ctenophora, Echinodermata, Entoprocta, Gastrotricha, Gnathifera, Hemichordata, Mollusca, Nematoda, Nemertea, Platyhelminthes, Porifera, Priapulida, Rotifera, Tardigrada, Xenacoelomorpha			
								Hudson Strait	1	91	91		COI-1	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Echinodermata, Mollusca, Nemertea, Porifera, Priapulida, Rotifera
									COI-2	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Echinodermata, Mollusca, Nemertea, Porifera, Rotifera				
									18S-1	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Ctenophora, Entoprocta, Gastrotricha, Gnathifera, Hemichordata, Mollusca, Nematoda, Nemertea, Platyhelminthes, Porifera, Priapulida, Rotifera, Tardigrada				
									18S-2	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Ctenophora, Echinodermata, Entoprocta, Gastrotricha, Gnathifera, Hemichordata, Mollusca, Nematoda, Nemertea, Platyhelminthes, Porifera, Priapulida, Rotifera, Tardigrada, Xenacoelomorpha				

Table 3. (Continued)

Project	Data Provider	Data Steward	Reference (if applicable)	Sampling Year	Ecodistrict	Sampling Design Summary (<i>n</i>)			Target Gene Subfragment	Phyla Detected
						Sampling Sites	Field Samples	Samples Per Site (min-max)		
4	DFO Arctic	Howland, K.	Sevellec et al. (2021)	2017	Hudson Bay	1	78	78	COI-1	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Echinodermata, Mollusca, Nemertea, Porifera, Priapulida, Rotifera
										COI-2
									18S-1	
									18S-2	Annelida, Arthropoda, Brachiopoda, Bryozoa, Chordata, Cnidaria, Ctenophora, Echinodermata, Entoprocta, Gastrotricha, Gnathifera, Mollusca, Nematoda, Nemertea, Platyhelminthes, Porifera, Priapulida, Rotifera, Xenacoelomorpha
					Lancaster Sound	1	54	54	COI-1	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Echinodermata, Mollusca, Nemertea, Porifera, Priapulida, Rotifera
										COI-2
									18S-1	
									18S-2	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Ctenophora, Gastrotricha, Gnathifera, Hemichordata, Mollusca, Nematoda, Nemertea, Platyhelminthes, Porifera, Priapulida, Rotifera, Xenacoelomorpha

Table 3. (Continued)

Project	Data Provider	Data Steward	Reference (if applicable)	Sampling Year	Ecodistrict	Sampling Design Summary (<i>n</i>)			Target Gene Subfragment	Phyla Detected			
						Sampling Sites	Field Samples	Samples Per Site (min-max)					
4	DFO Arctic	Howland, K.	Sevellec et al. (2021)	2017	North Baffin Fjords	1	80	80		COI-1	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Echinodermata, Mollusca, Nemertea, Porifera, Priapulida, Rotifera		
										COI-2	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Mollusca, Nemertea, Porifera, Rotifera		
										18S-1	Annelida, Arthropoda, Brachiopoda, Bryozoa, Chordata, Cnidaria, Ctenophora, Gastrotricha, Gnathifera, Mollusca, Nematoda, Nemertea, Platyhelminthes, Porifera, Priapulida, Rotifera, Tardigrada		
										18S-2	Annelida, Arthropoda, Brachiopoda, Bryozoa, Chordata, Cnidaria, Ctenophora, Echinodermata, Entoprocta, Gastrotricha, Gnathifera, Mollusca, Nematoda, Nemertea, Platyhelminthes, Porifera, Priapulida, Rotifera, Xenacoelomorpha		
						2018	Baffin Bay/Davis Strait	65	72	1 - 3		COI-1	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Echinodermata, Mollusca, Myzozoa, Nemertea, Porifera, Rotifera
								COI-2	Annelida, Arthropoda, Chordata, Cnidaria, Mollusca				
								18S-1	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Ctenophora, Echinodermata, Entoprocta, Mollusca, Nemertea, Porifera, Priapulida, Rotifera, Xenacoelomorpha				
								18S-2	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Ctenophora, Echinodermata, Mollusca, Nemertea, Porifera				

Table 3. (Continued)

Project	Data Provider	Data Steward	Reference (if applicable)	Sampling Year	Ecodistrict	Sampling Design Summary (<i>n</i>)			Target Gene Subfragment	Phyla Detected	
						Sampling Sites	Field Samples	Samples Per Site (min-max)			
4	DFO Arctic	Howland, K.		2018	NL-Labrador Shelves	12	106	3 - 15	<p>Detailed description: A bar chart showing the number of samples collected per month for the 2018 NL-Labrador Shelves project. The y-axis is labeled 'Samples' and ranges from 0 to 100 in increments of 25. The x-axis is labeled 'Month' and lists the months from Jan to Dec. A single bar for July reaches the 106 mark on the y-axis.</p>	COI-1	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Echinodermata, Mollusca, Myzozoa, Nemertea, Porifera, Rotifera
						COI-2	Annelida, Arthropoda, Chordata, Cnidaria, Mollusca, Nemertea, Porifera				
						18S-1	Annelida, Arthropoda, Brachiopoda, Bryozoa, Chordata, Ctenophora, Mollusca, Nematoda, Platyhelminthes, Porifera, Rotifera, Tardigrada				
						18S-2	Annelida, Arthropoda, Brachiopoda, Bryozoa, Chordata, Cnidaria, Ctenophora, Gastrotricha, Mollusca, Nematoda, Platyhelminthes, Porifera, Rotifera, Tardigrada, Xenacoelomorpha				
				2019	North Baffin Fjords	15	51	3 - 4	<p>Detailed description: A bar chart showing the number of samples collected per month for the 2019 North Baffin Fjords project. The y-axis is labeled 'Samples' and ranges from 0 to 50 in increments of 10. The x-axis is labeled 'Month' and lists the months from Jan to Dec. A single bar for August reaches the 51 mark on the y-axis.</p>	COI-1	Annelida, Arthropoda, Chordata, Cnidaria, Echinodermata, Mollusca, Porifera
						COI-2	Annelida, Arthropoda, Chordata, Cnidaria, Mollusca				
						18S-1	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Ctenophora, Echinodermata, Gastrotricha, Mollusca, Nematoda, Nemertea, Platyhelminthes, Porifera, Priapulida, Xenacoelomorpha				
						18S-2	Annelida, Arthropoda, Bryozoa, Cnidaria, Ctenophora, Gastrotricha, Mollusca, Nemertea, Platyhelminthes, Rotifera				
6	DFO Québec	Parent, G. ^c		2020	Magdalen Shallows	1	90	90	<p>Detailed description: A bar chart showing the number of samples collected per month for the 2020 Magdalen Shallows project. The y-axis is labeled 'Samples' and ranges from 0 to 80 in increments of 20. The x-axis is labeled 'Month' and lists the months from Jan to Dec. A single bar for July reaches the 90 mark on the y-axis.</p>	COI-1	Annelida, Arthropoda, Bryozoa, Chordata, Cnidaria, Echinodermata, Nematoda, Porifera
						12S-MiFishU	Chordata				
						12S-MiMam	Chordata				
						16S-chord	Chordata, Cnidaria, Echinodermata, Porifera				

Table 3. (Continued)

Project	Data Provider	Data Steward	Reference (if applicable)	Sampling Year	Ecodistrict	Sampling Design Summary (n)			Target Gene Subfragment	Phyla Detected
						Sampling Sites	Field Samples	Samples Per Site (min-max)		
6	DFO Québec	Parent, G.		2021	North Gulf Shelf	13	27	1 - 2	12S-248	Chordata, Echinodermata
									12S-MiFishU	Annelida, Chordata, Mollusca
									16S-chord	Chordata, Cnidaria, Echinodermata, Mollusca, Porifera
					Laurentian Channel	26	51	1 - 2	12S-248	Chordata, Echinodermata
									12S-MiFishU	Annelida, Chordata, Mollusca
									16S-chord	Chordata, Cnidaria, Echinodermata, Mollusca, Porifera
7	DFO Maritimes	Jeffery, N. ^d	He et al. 2022	2019	St. Lawrence River	4	164	2-158	COI-1	Arthropoda, Cnidaria, Echinodermata
									12S-248	Chordata, Echinodermata
					Scotian Shelf				12S-MiFishU	Annelida, Chordata, Echinodermata
									16S-chord	Chordata, Cnidaria, Echinodermata, Mollusca, Porifera
						10	30	3	12S-248	Chordata
									16S-Fish	Chordata

^a Lacoursière, A., anis.lacoursiere@dfo-mpo.gc.ca, Fisheries and Oceans Canada, Maritimes Region

^b Howland, K., kimberly.howland@dfo-mpo.gc.ca, Fisheries and Oceans Canada, Arctic Region

^c Parent, G., genevieve.parent@dfo-mpo.gc.ca, Fisheries and Oceans Canada, Québec Region

^d Jeffery, N., nick.jeffery@dfo-mpo.gc.ca, Fisheries and Oceans Canada, Maritimes Region

6.1 ANNELIDA

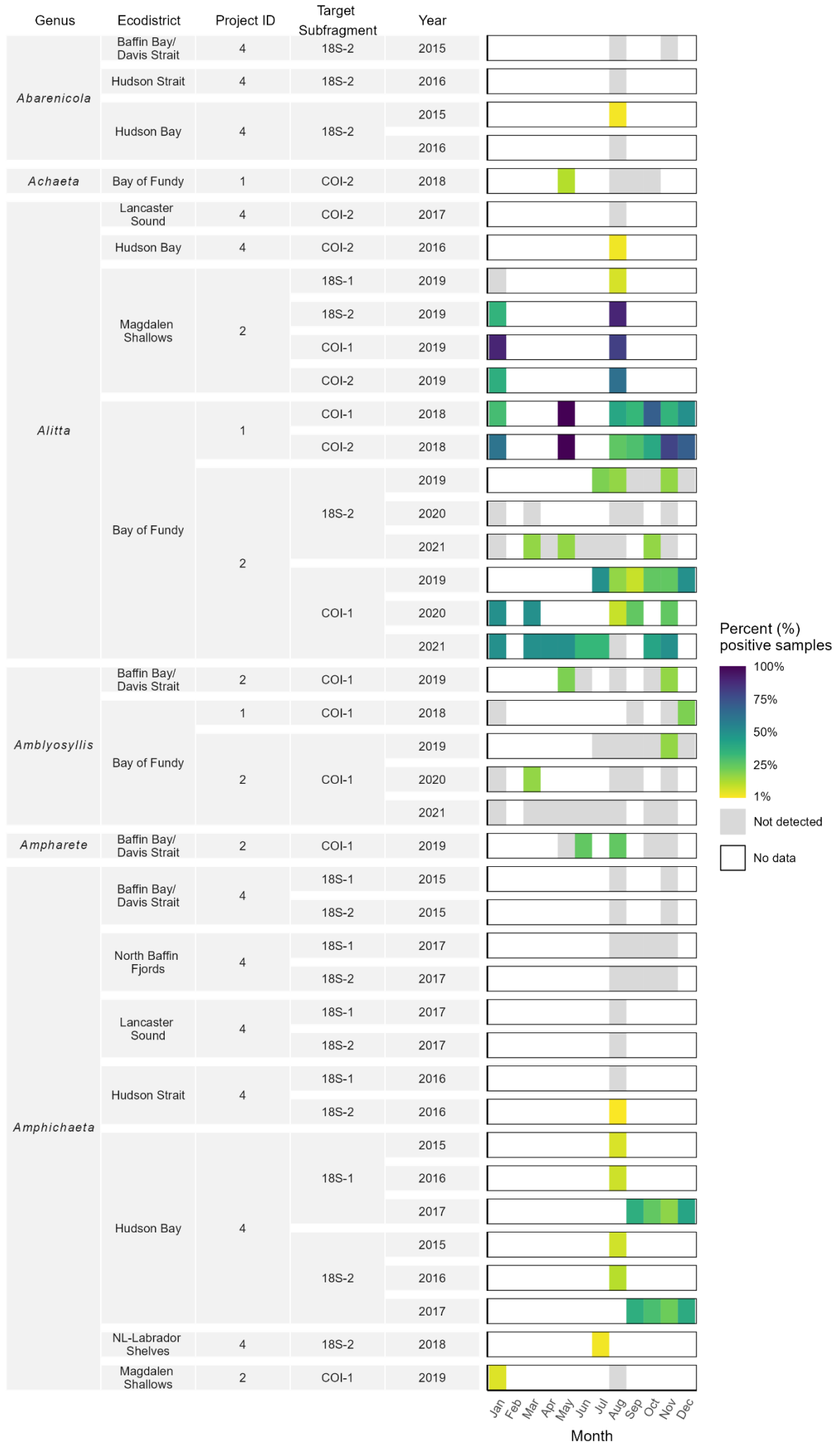


Figure 13. Proportion of positive environmental DNA (eDNA) detections per month from marine DNA metabarcoding projects. Genera are distinguished by sample year, ecodistrict, and target gene subfragment (*i.e.*, primers). See Table 3 for project summaries including monthly sampling effort. Grey indicates that the target subfragment can successfully amplify the taxon and resulted in non-detection. White indicates that sampling did not occur that month or the taxon was not detected within the dataset.

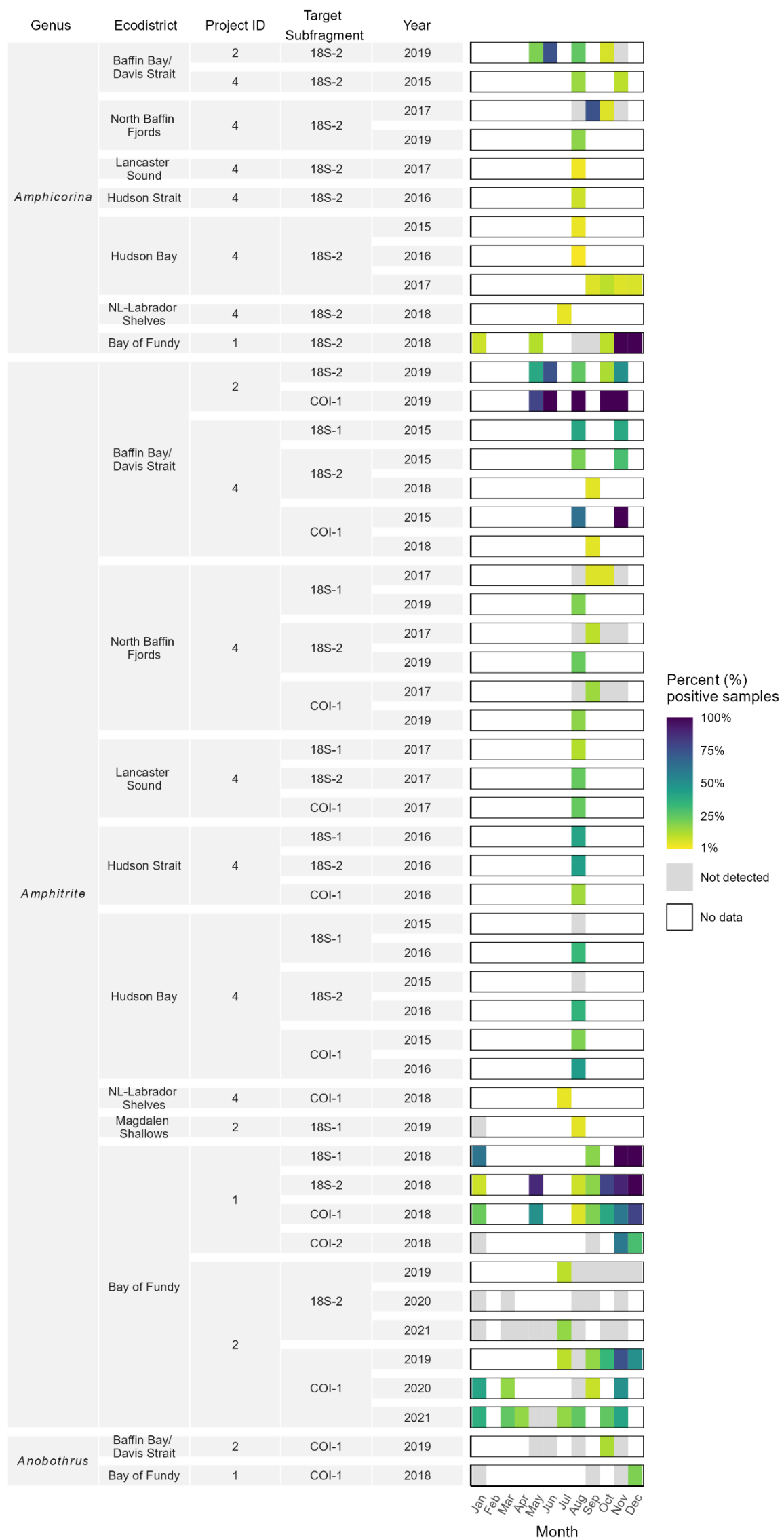


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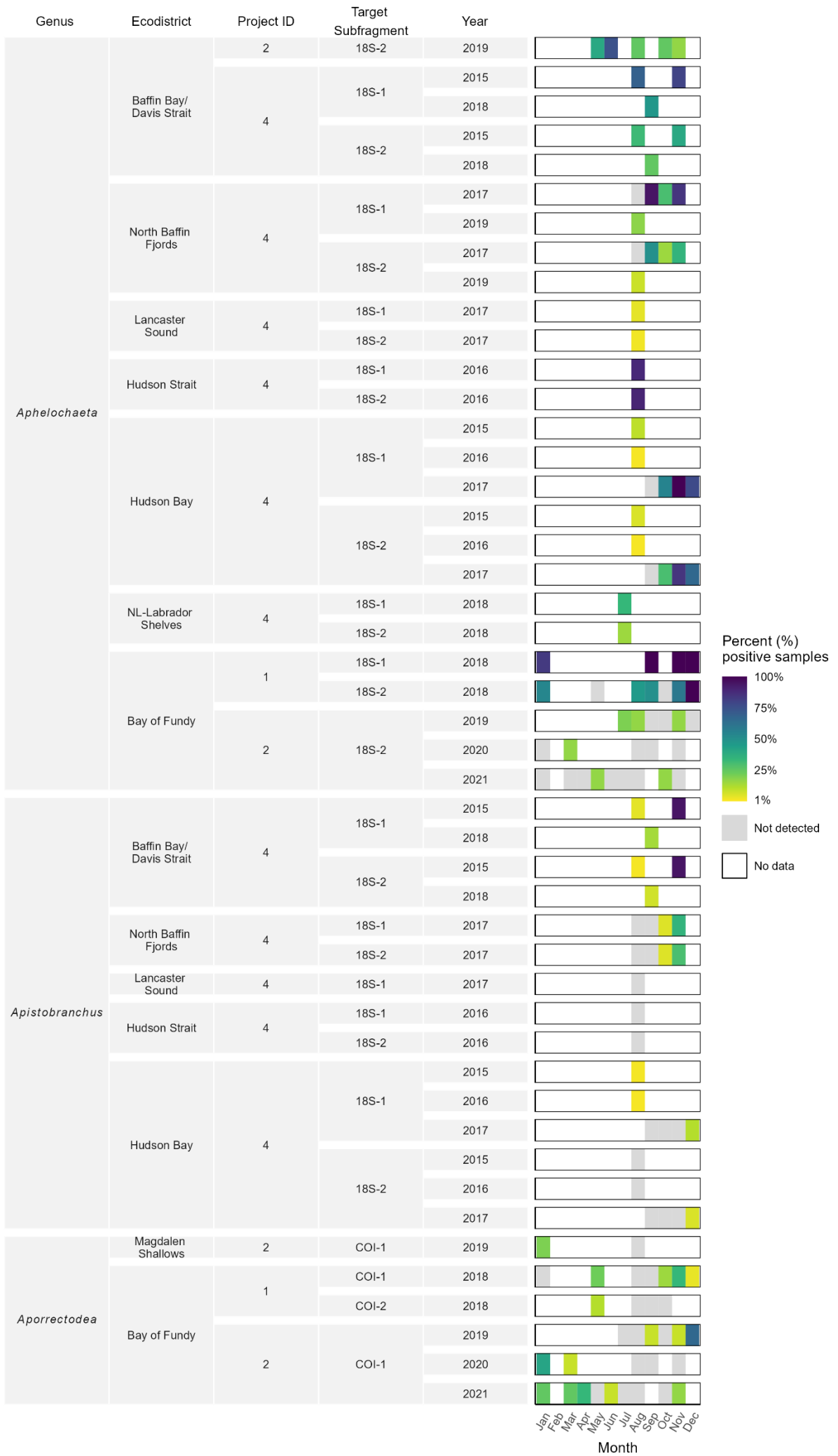


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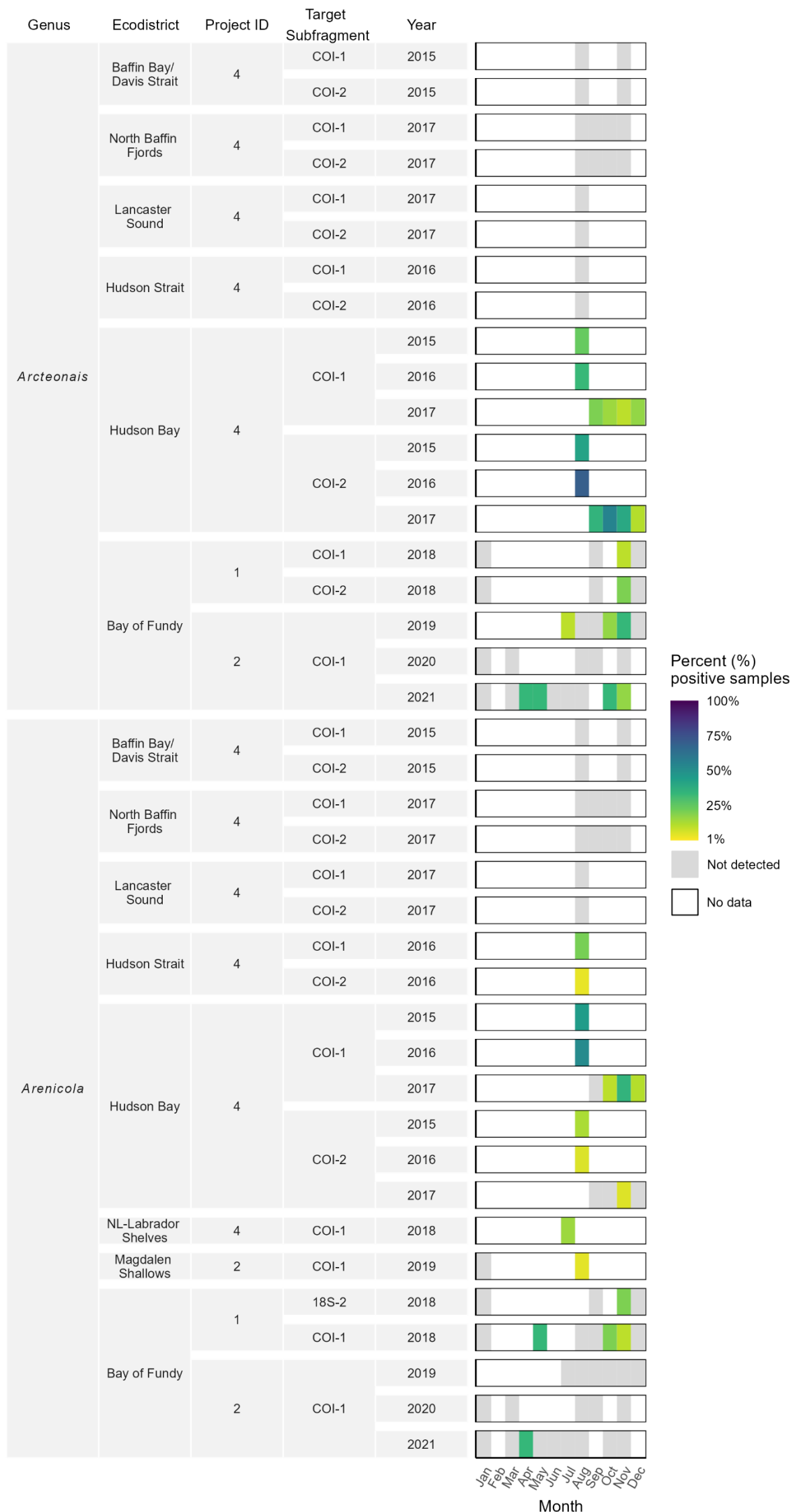


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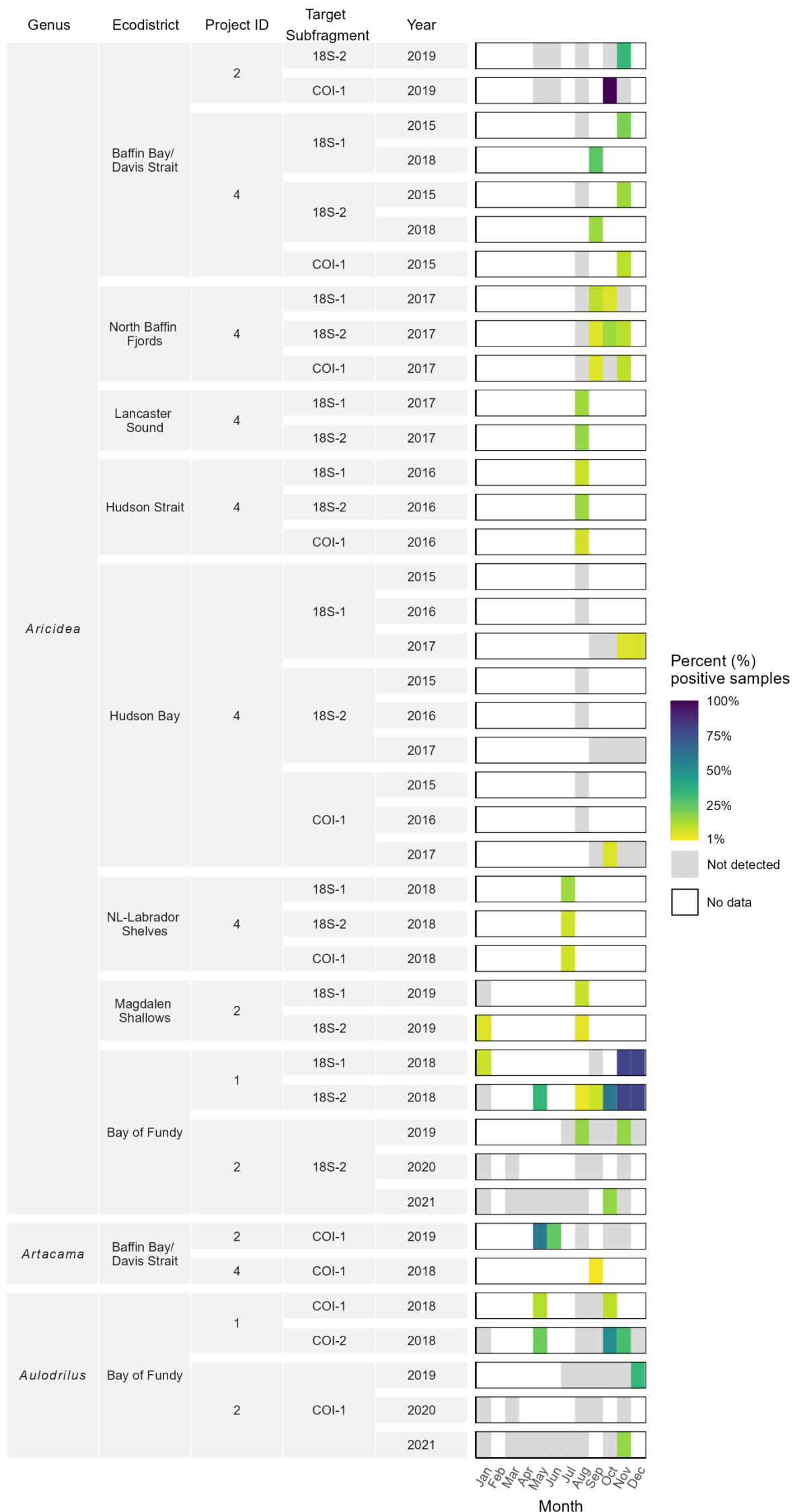


Figure 13. (Continued)



Figure 13. (Continued)

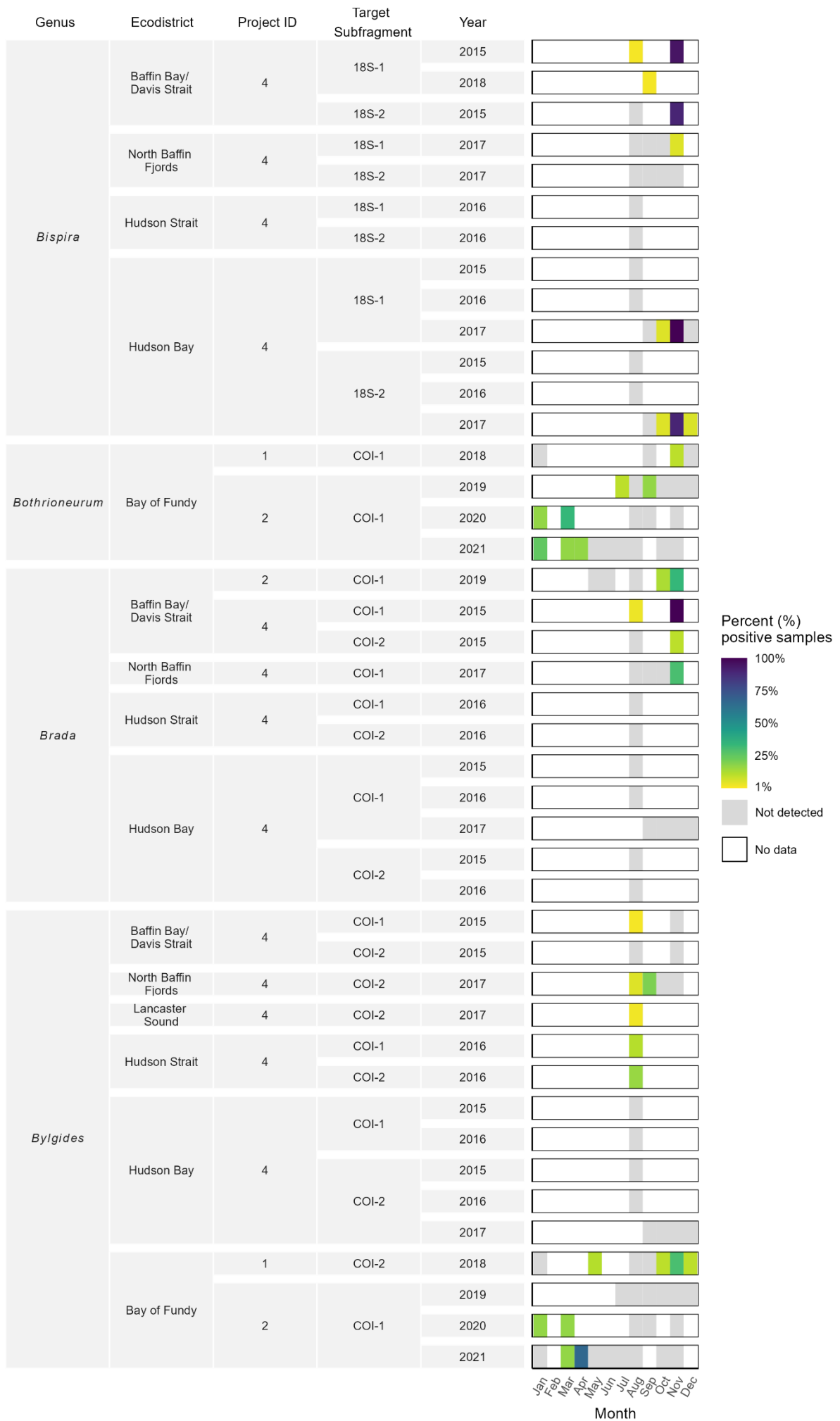


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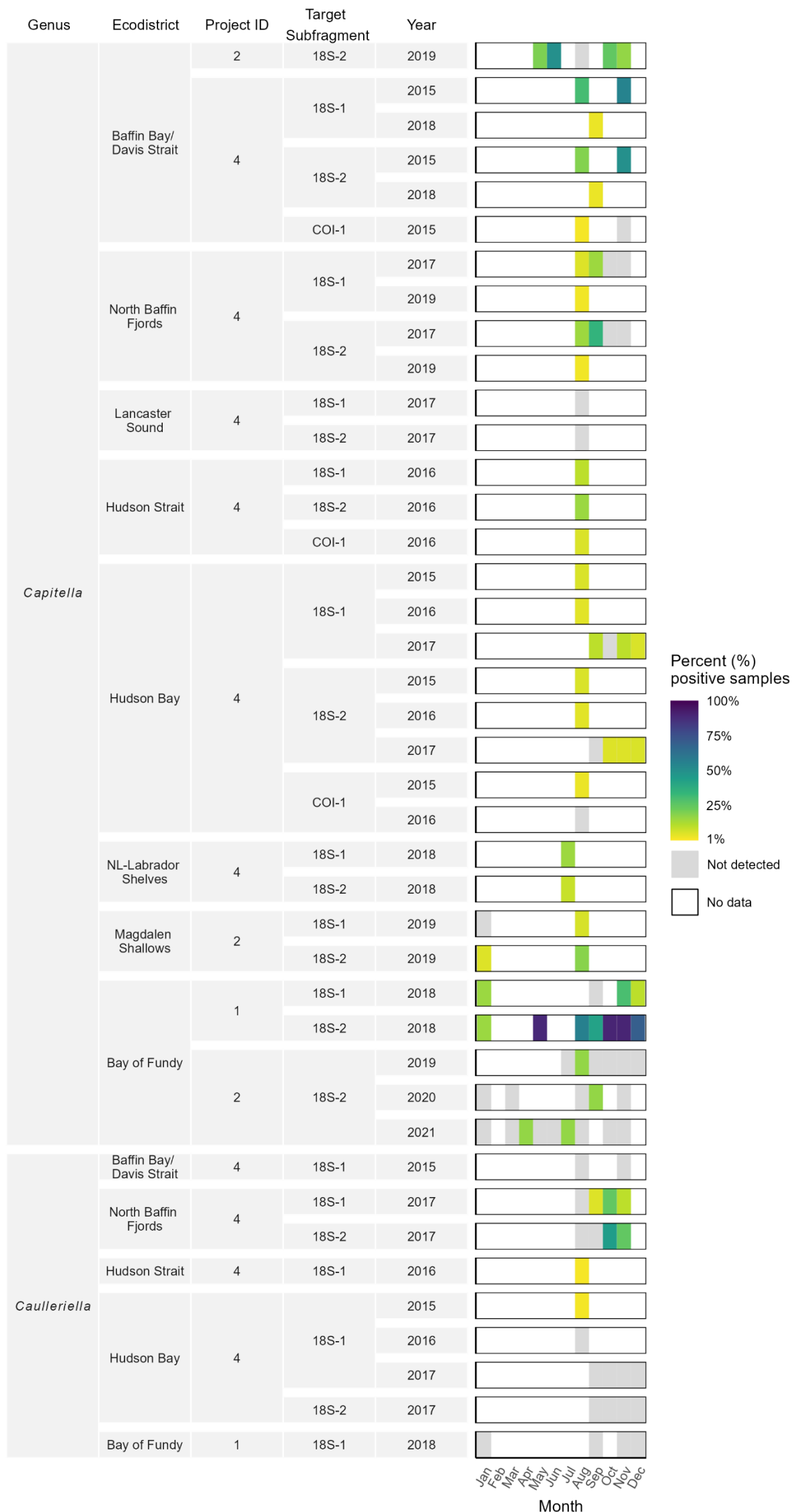


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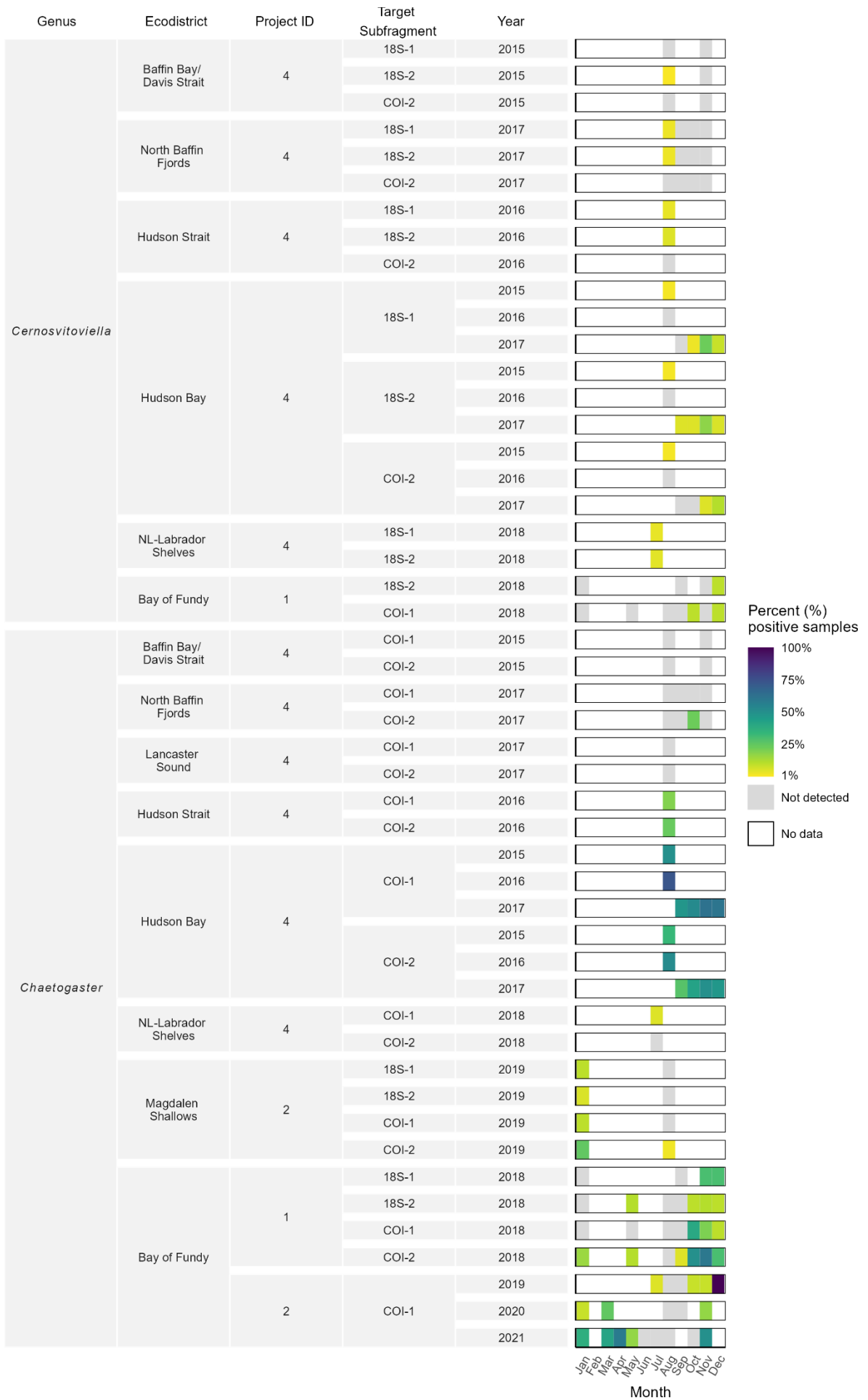


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Figure 13. (Continued)

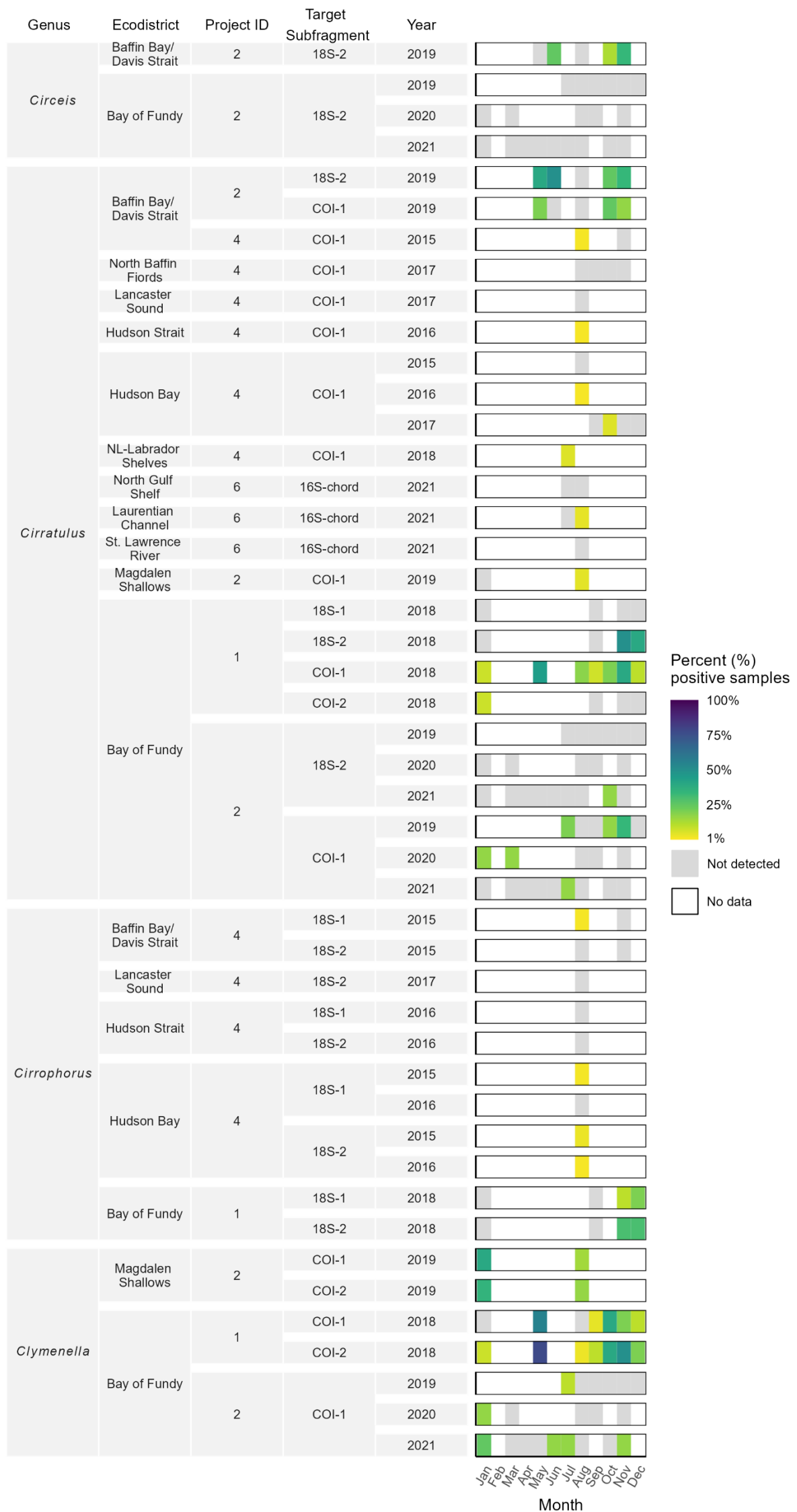


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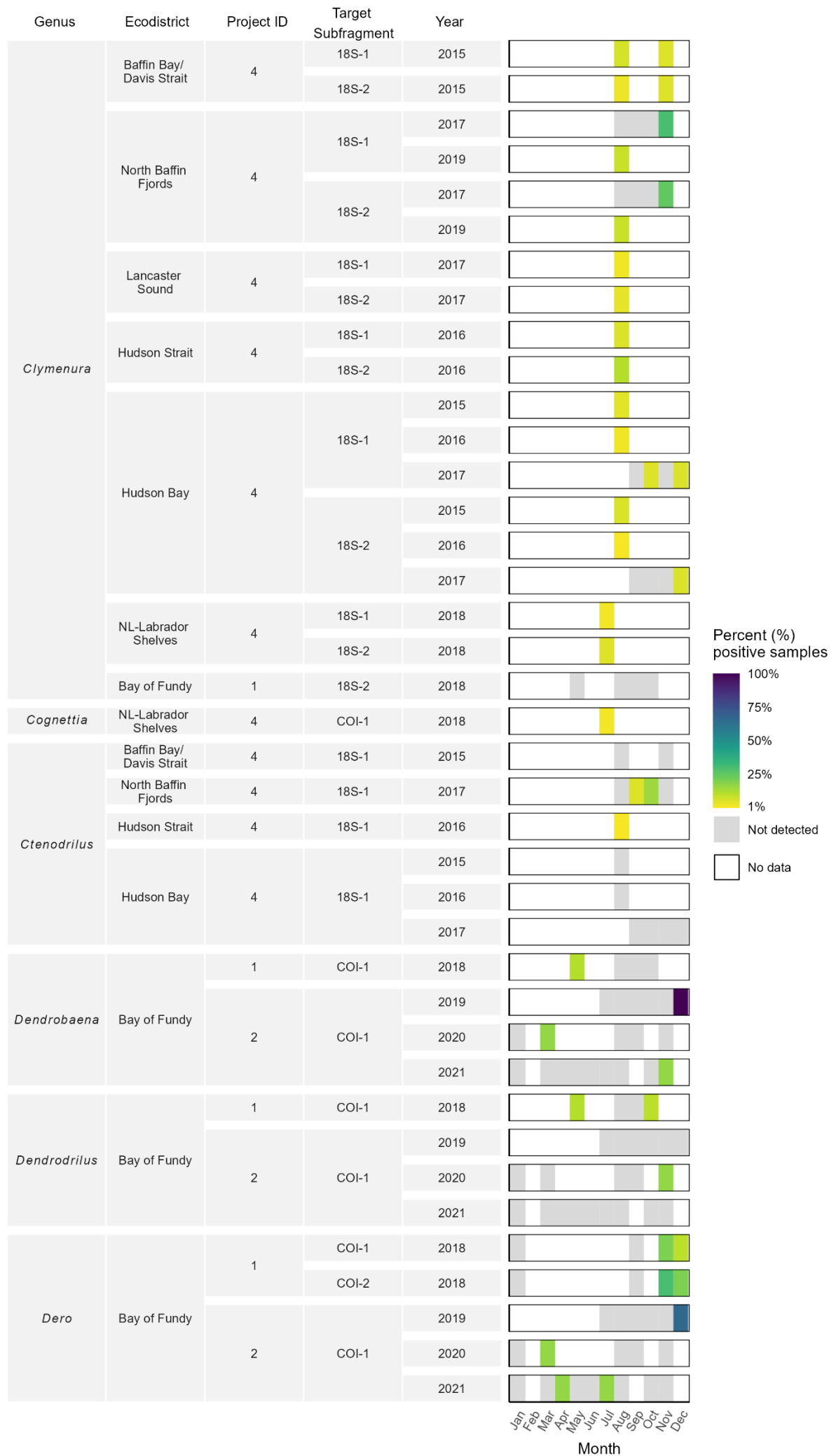


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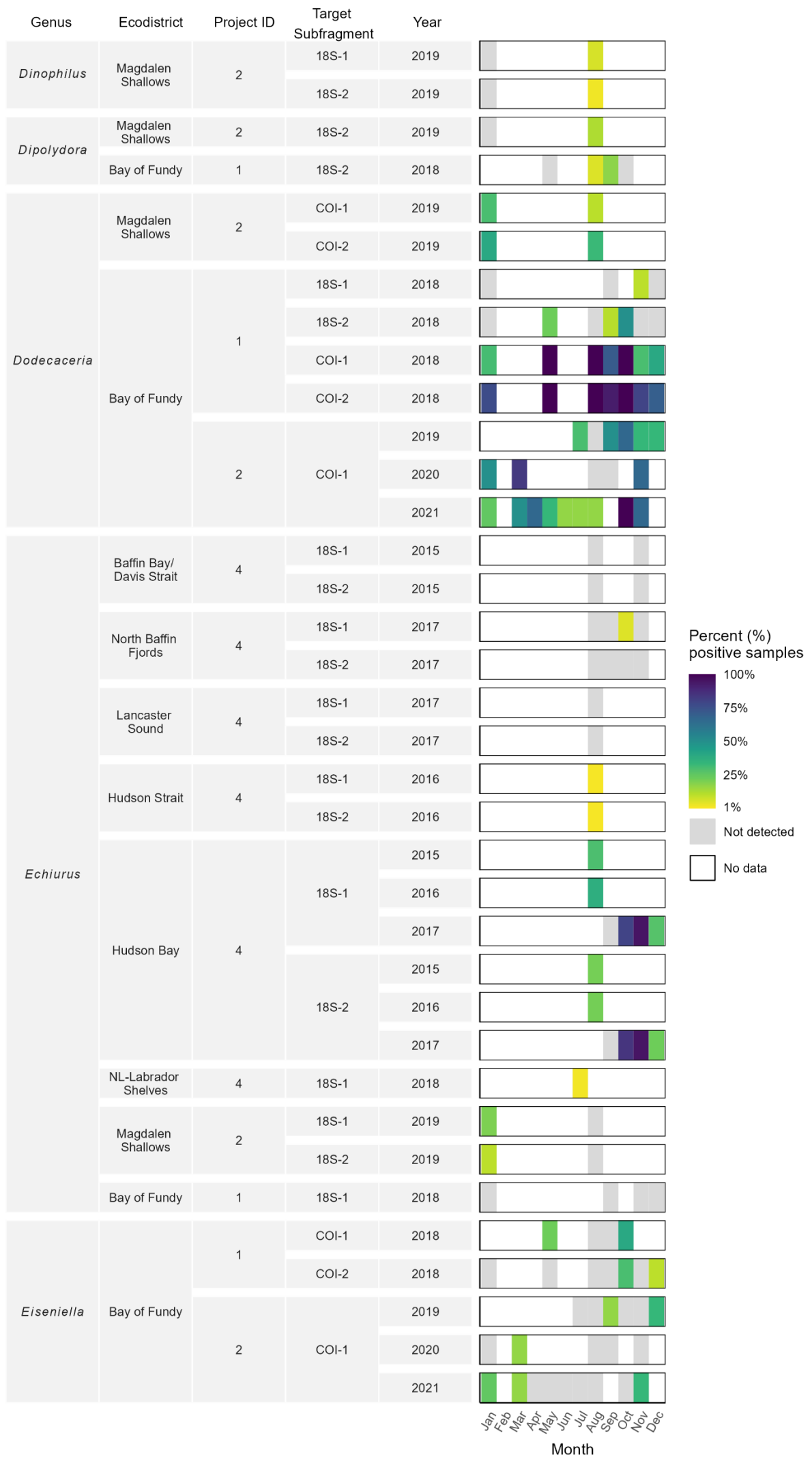


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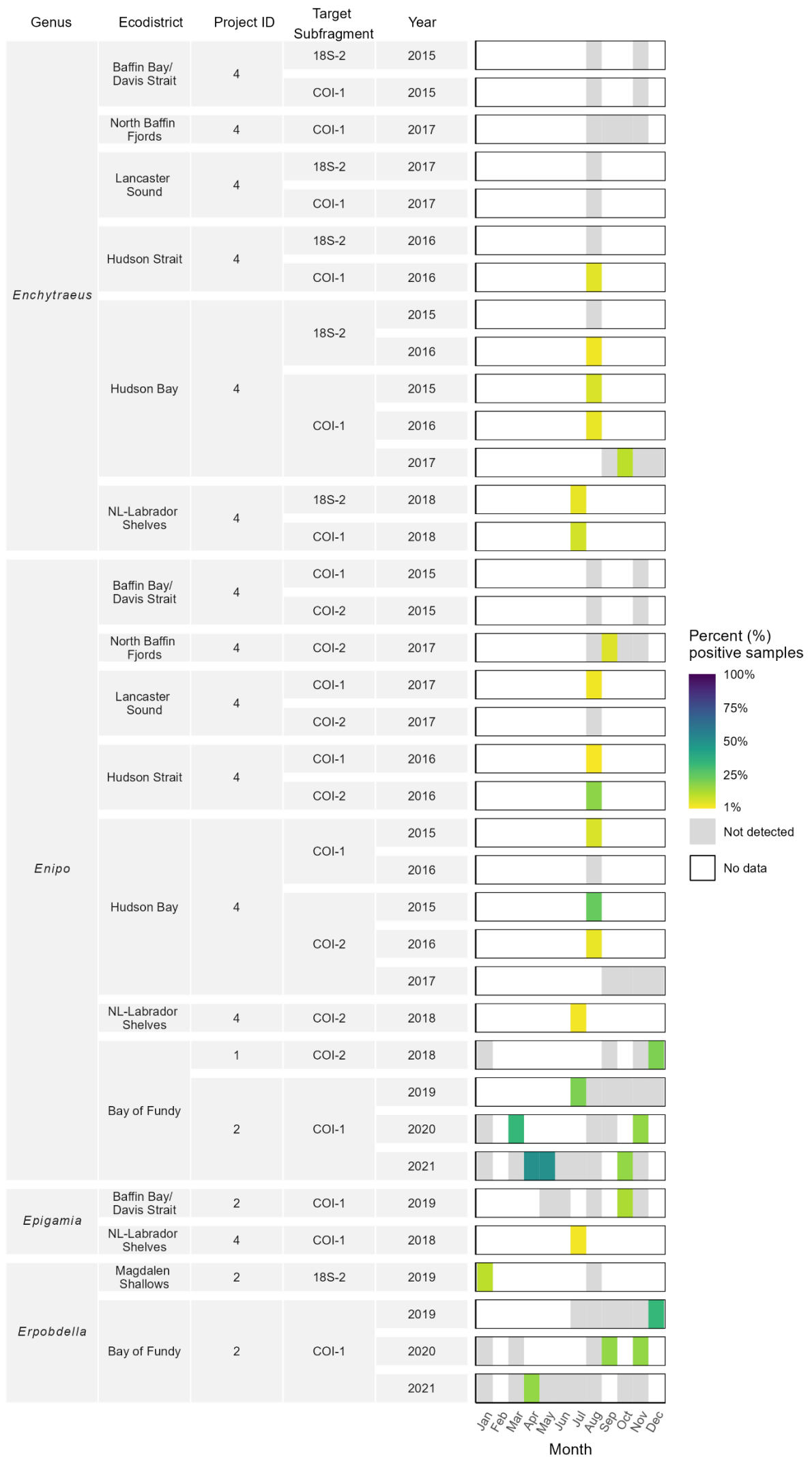


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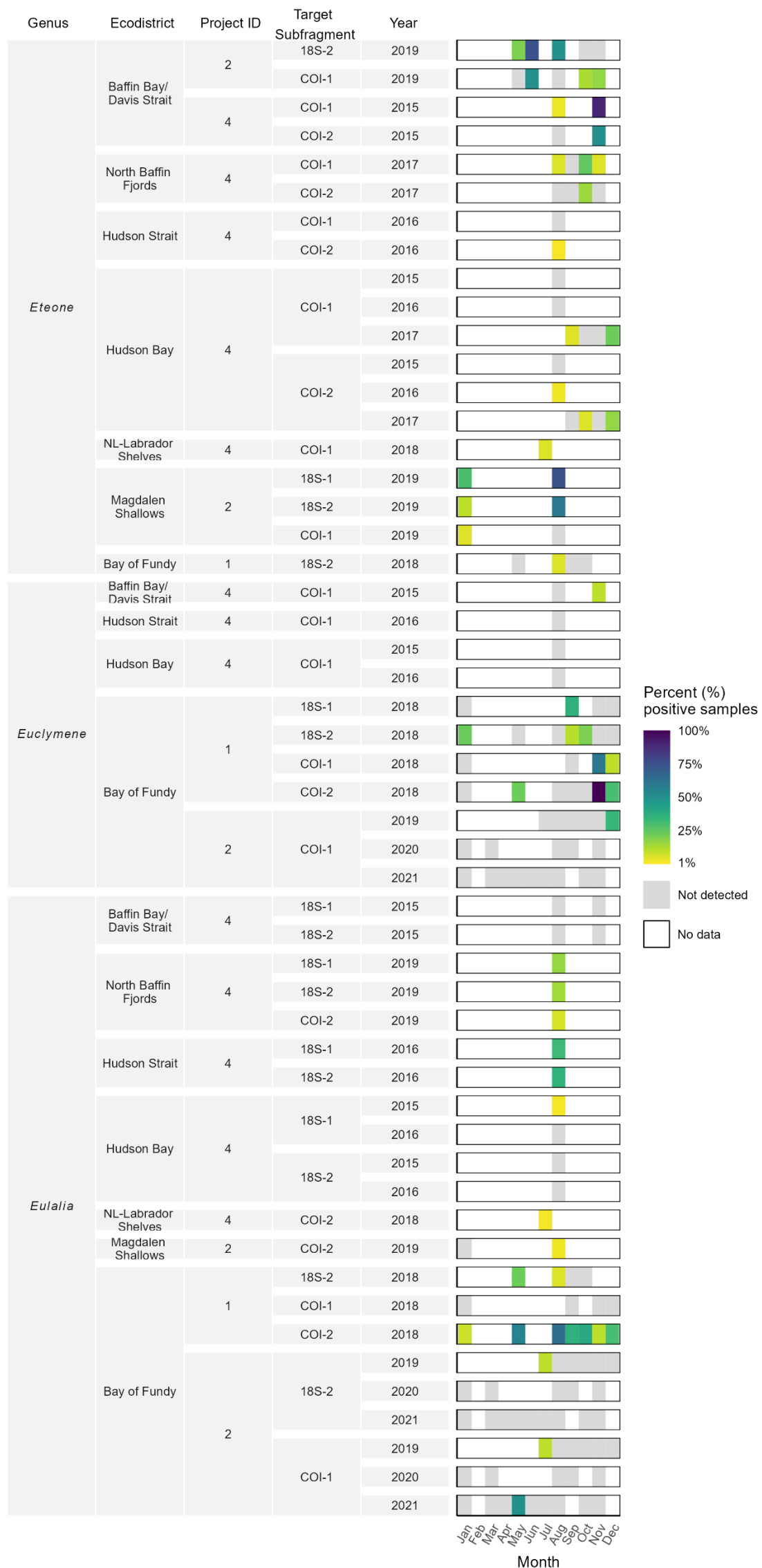


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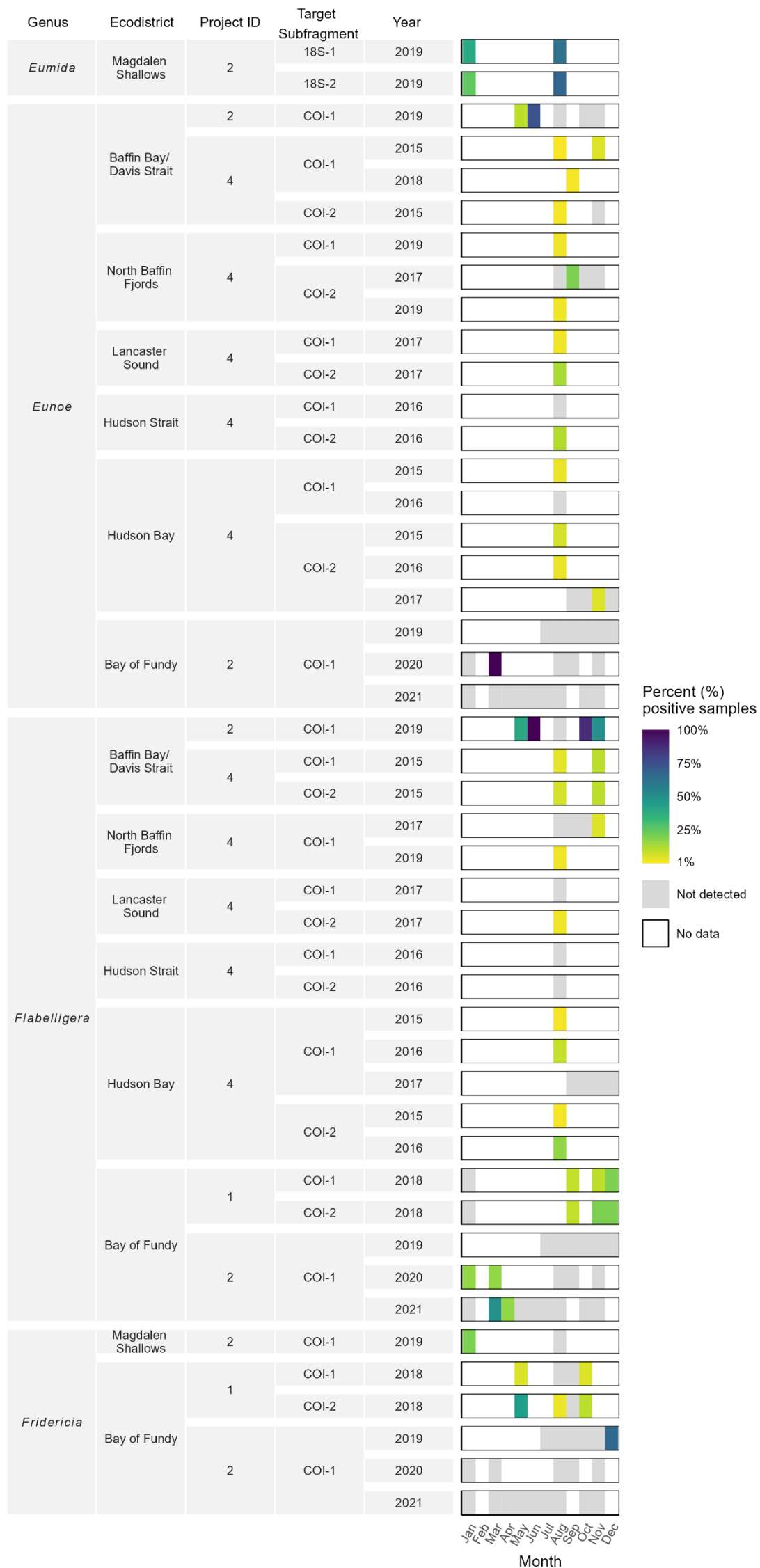


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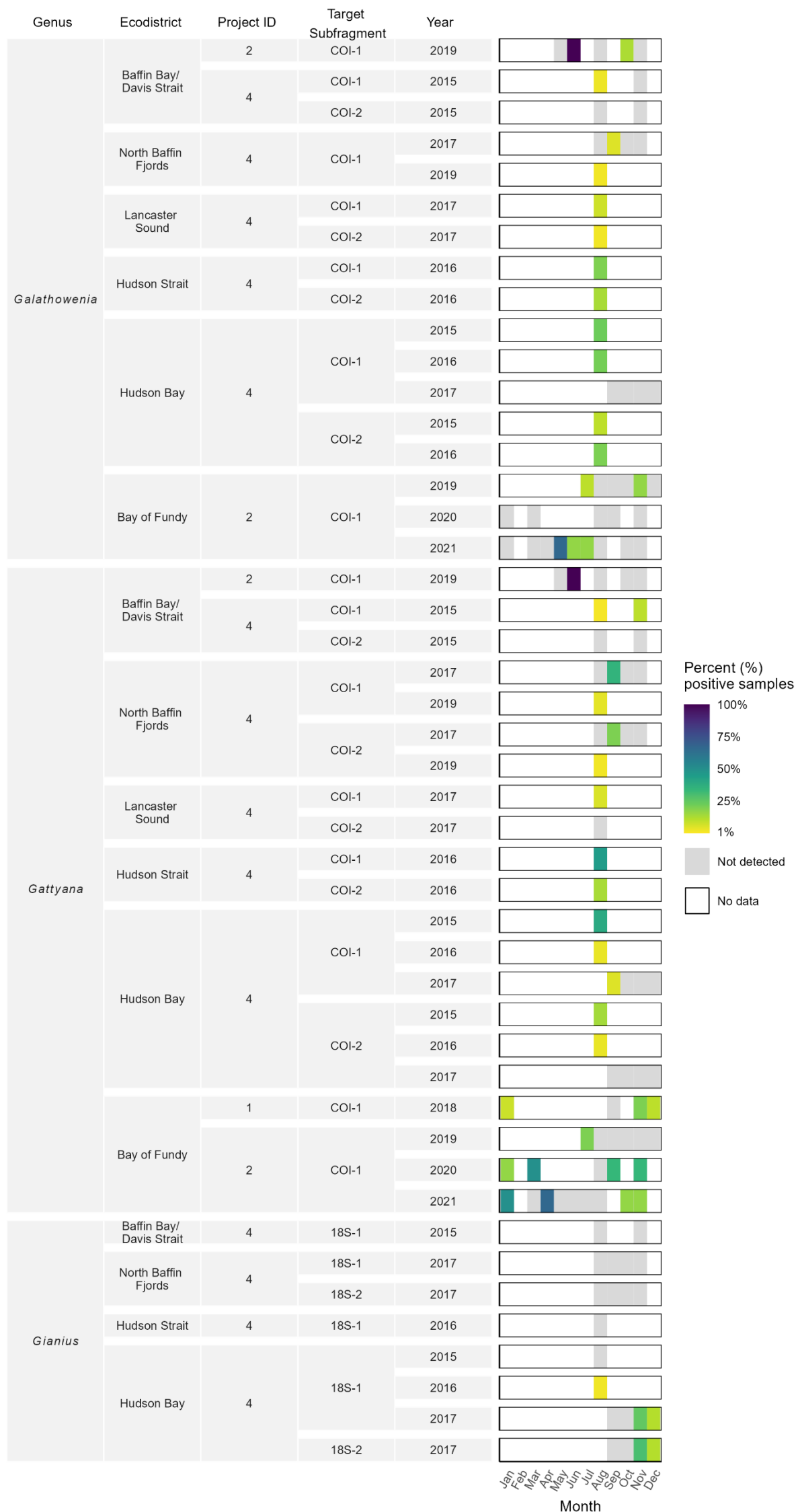


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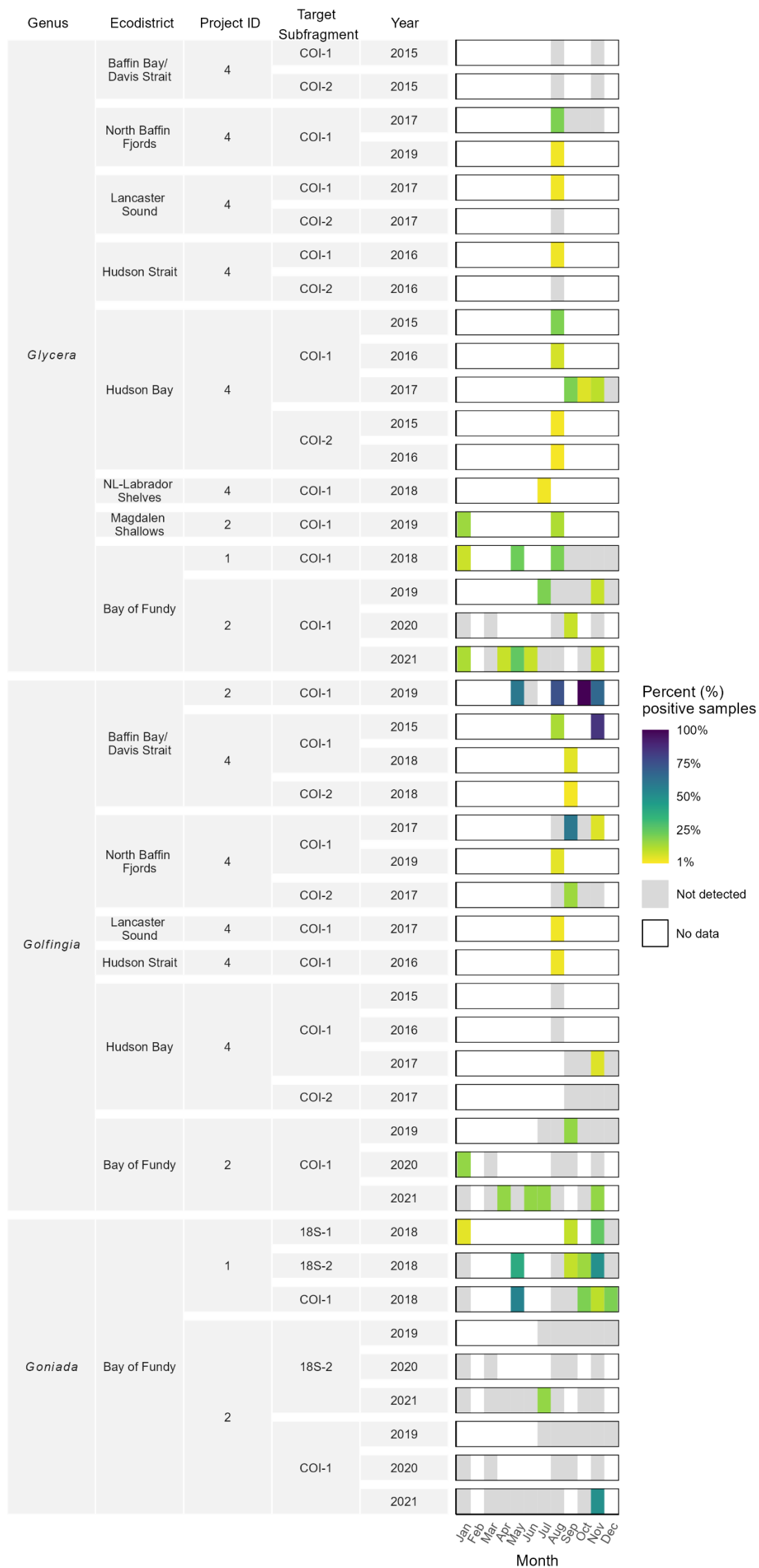


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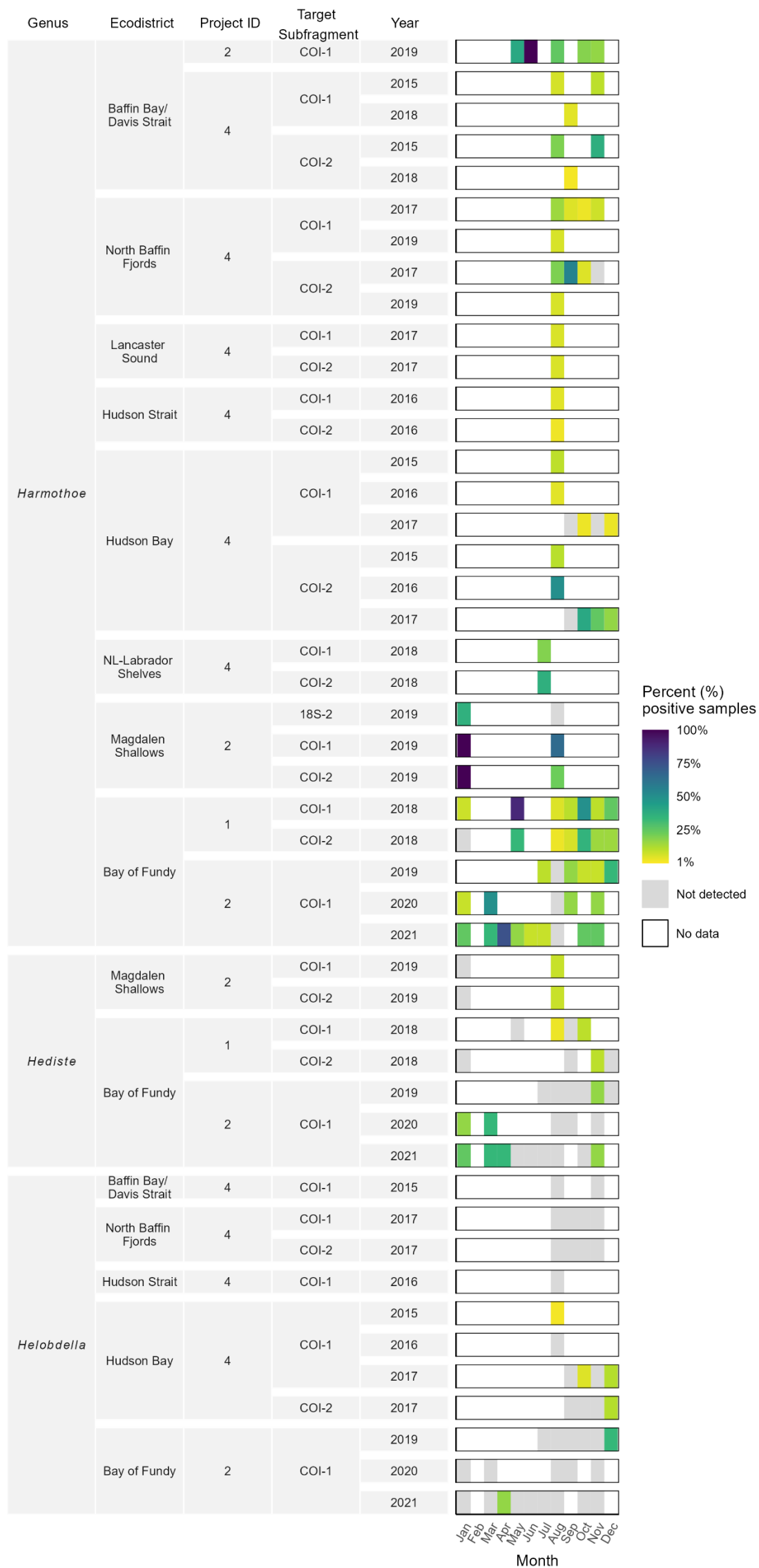


Figure 13. (Continued)



Figure 13. (Continued)



Figure 13. (Continued)

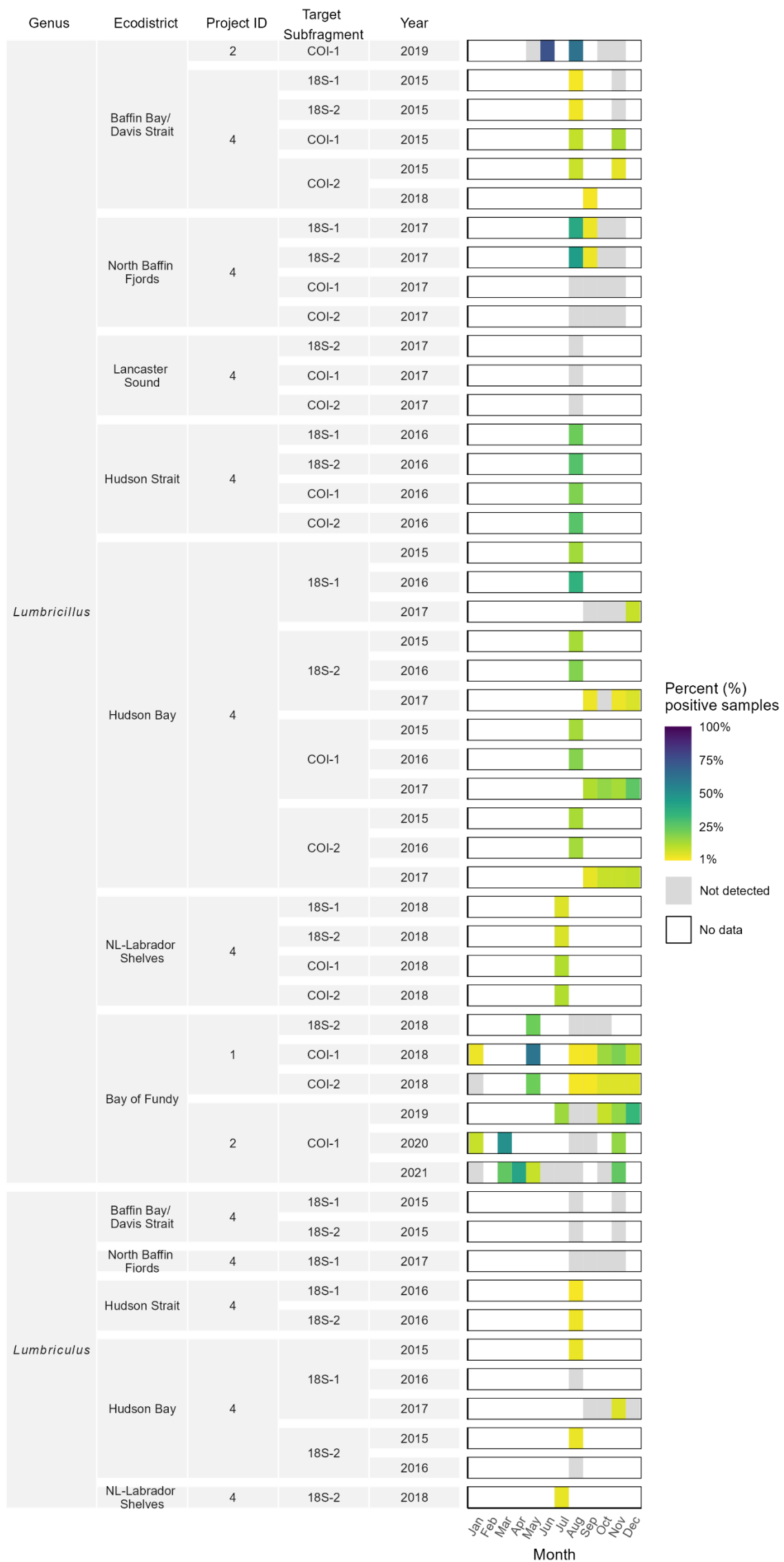


Figure 13. (Continued)



Figure 13. (Continued)

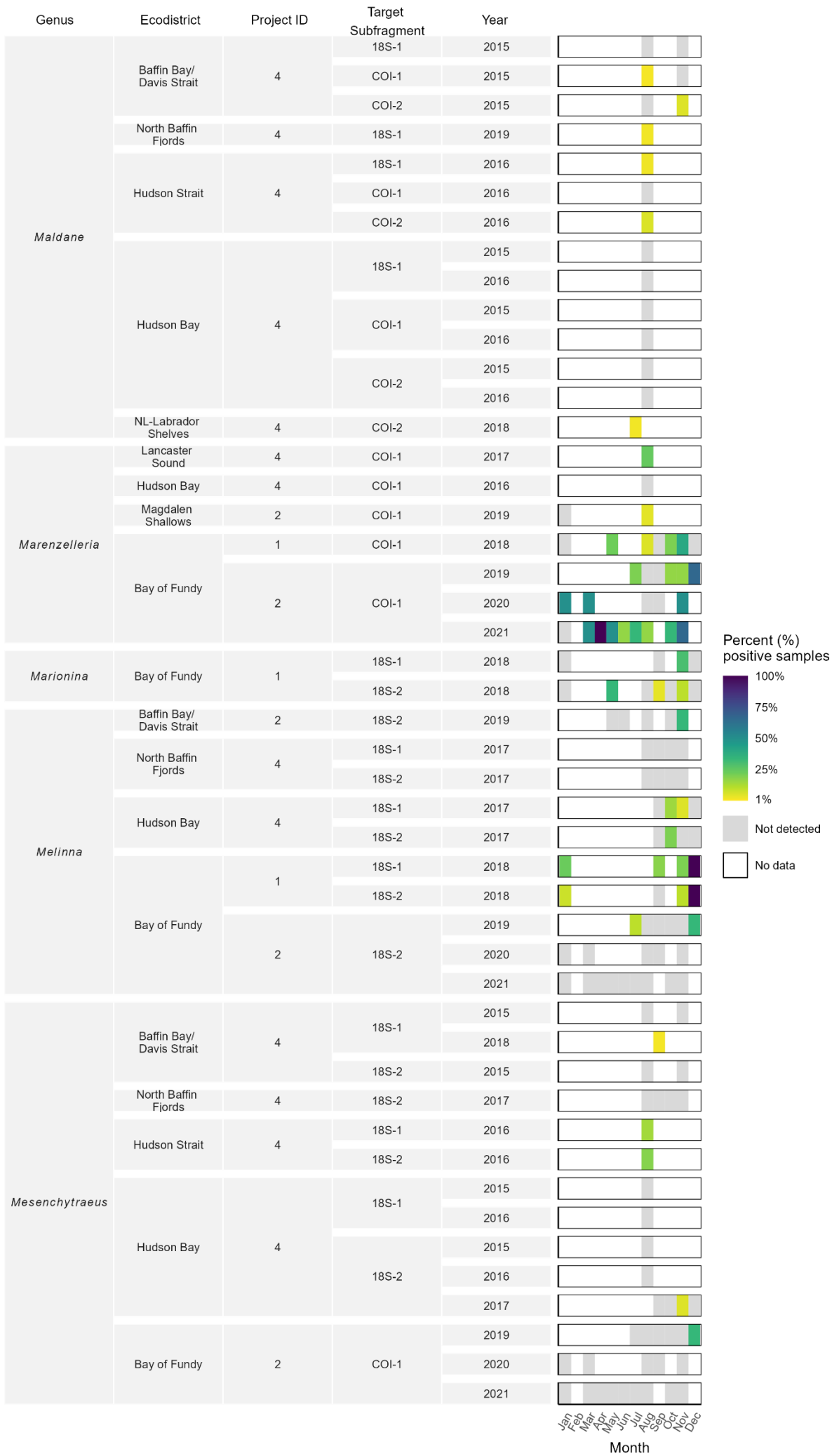


Figure 13. (Continued)



Figure 13. (Continued)

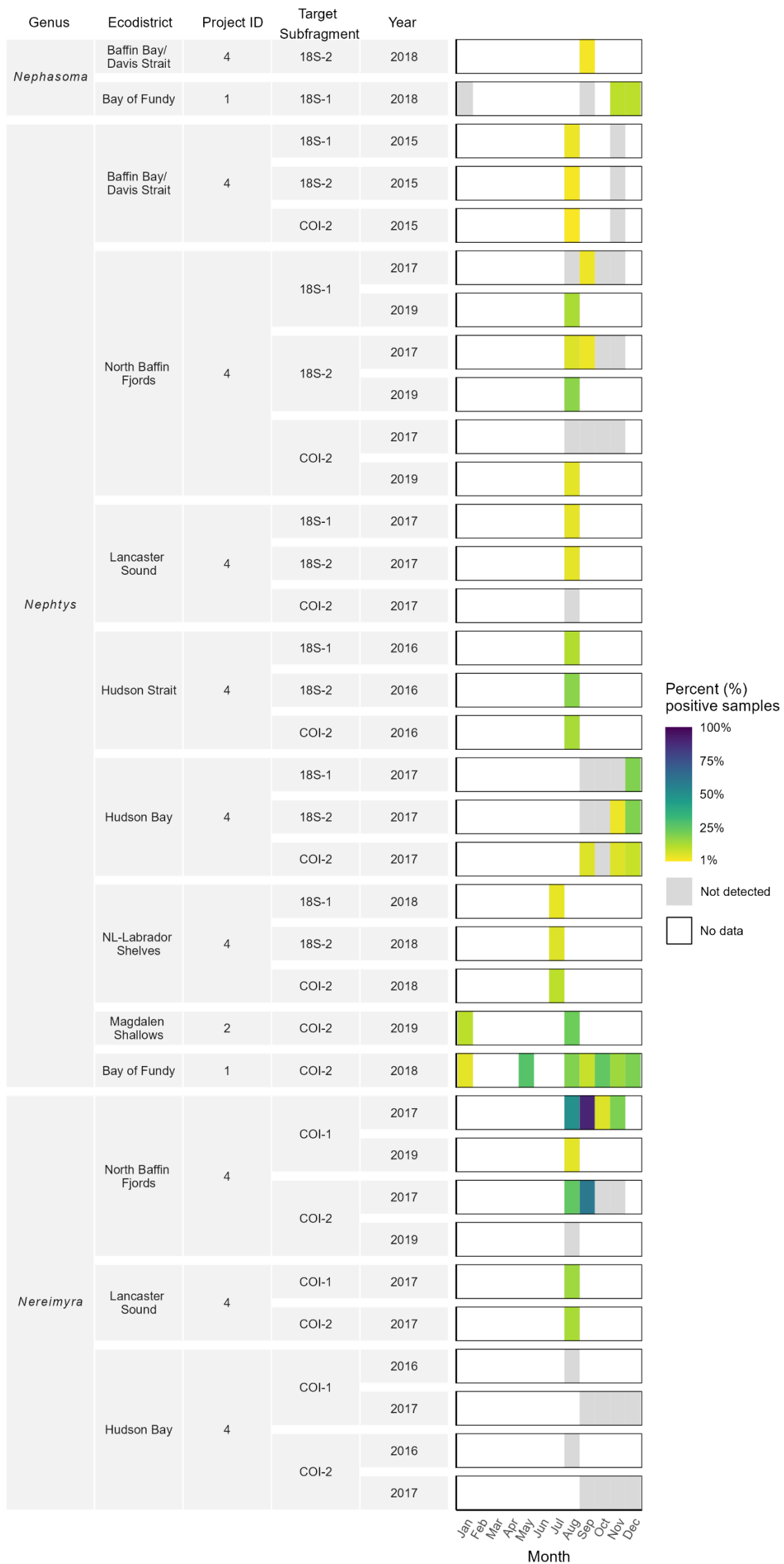


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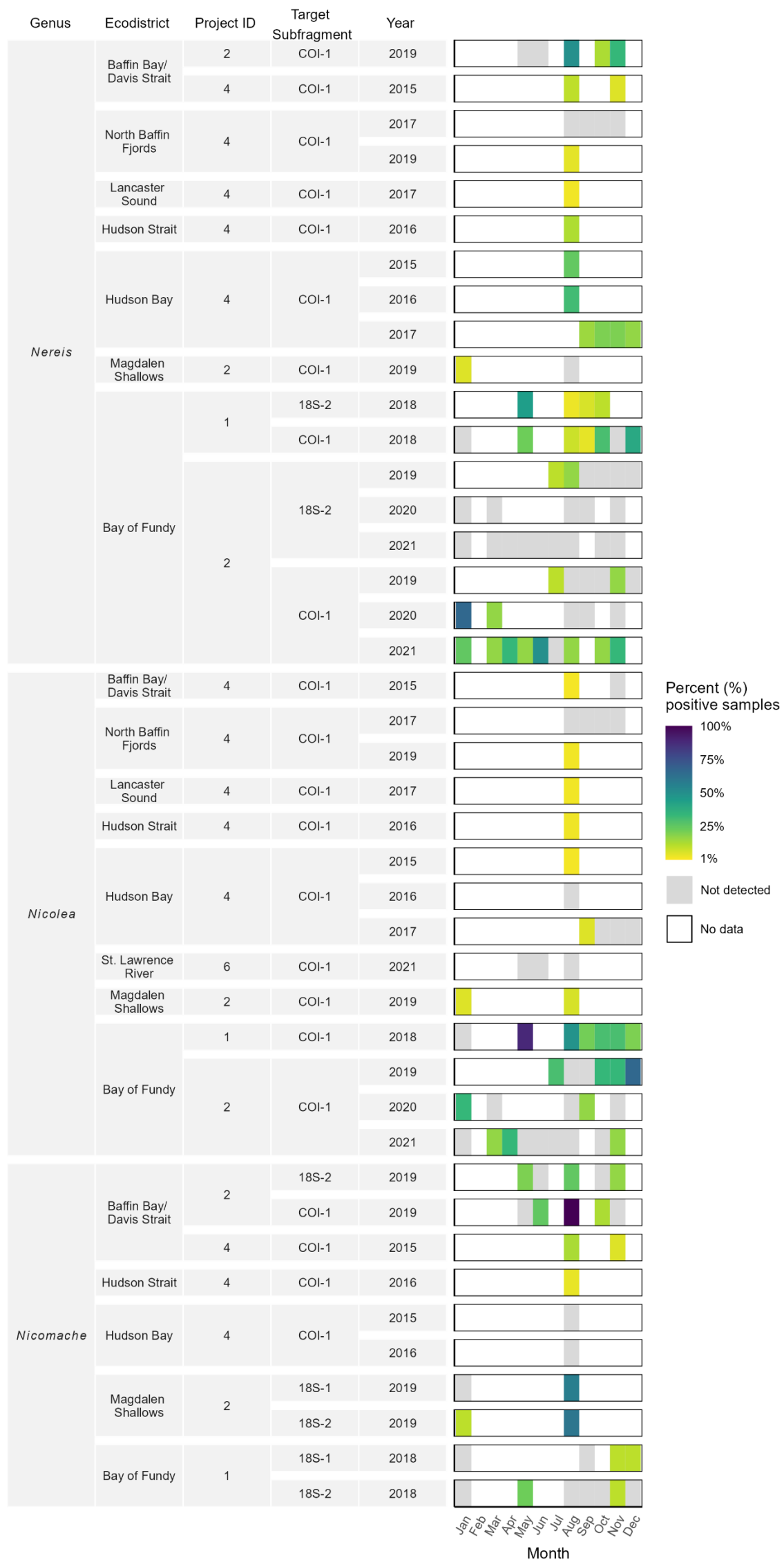


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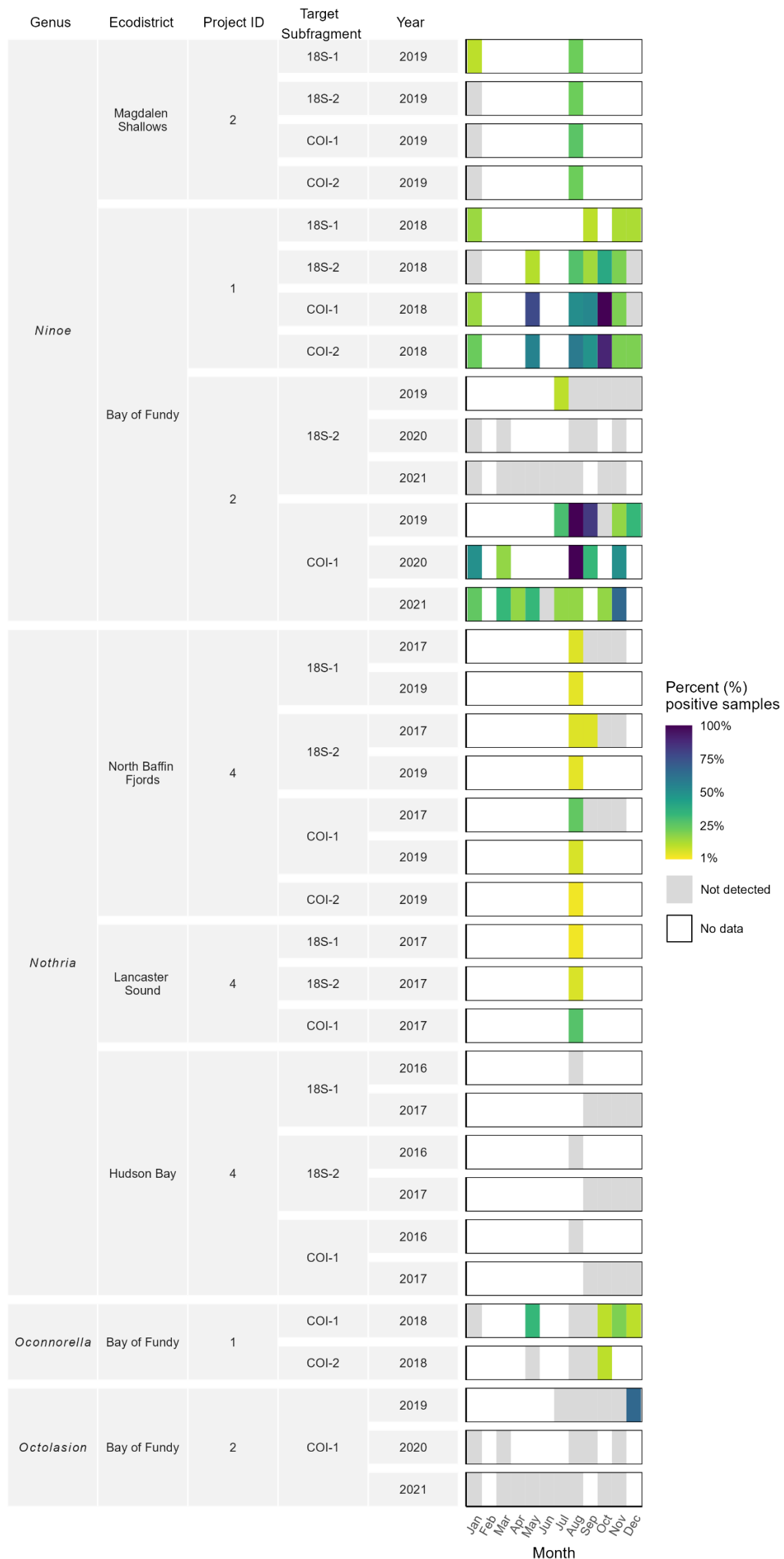


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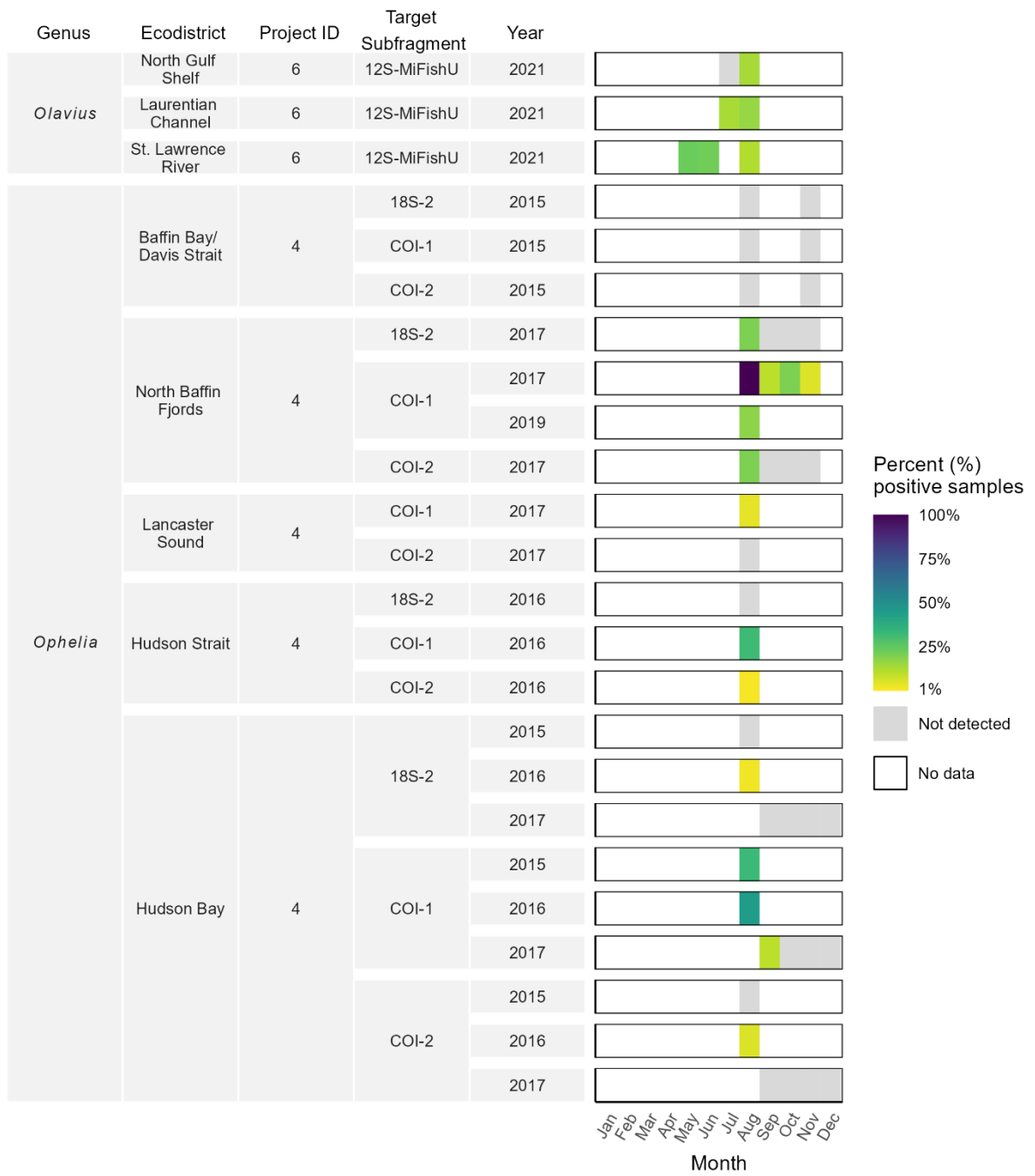


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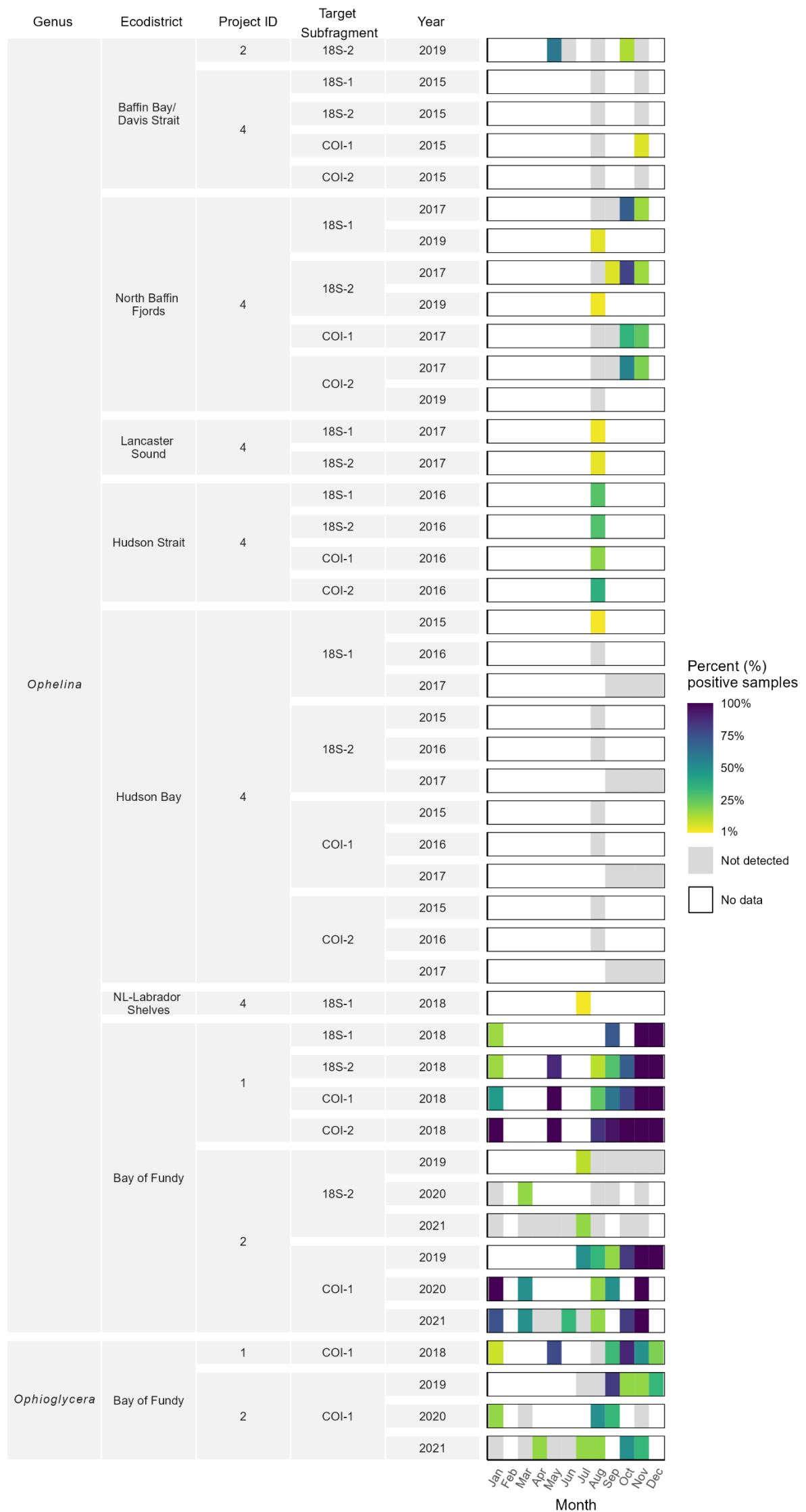


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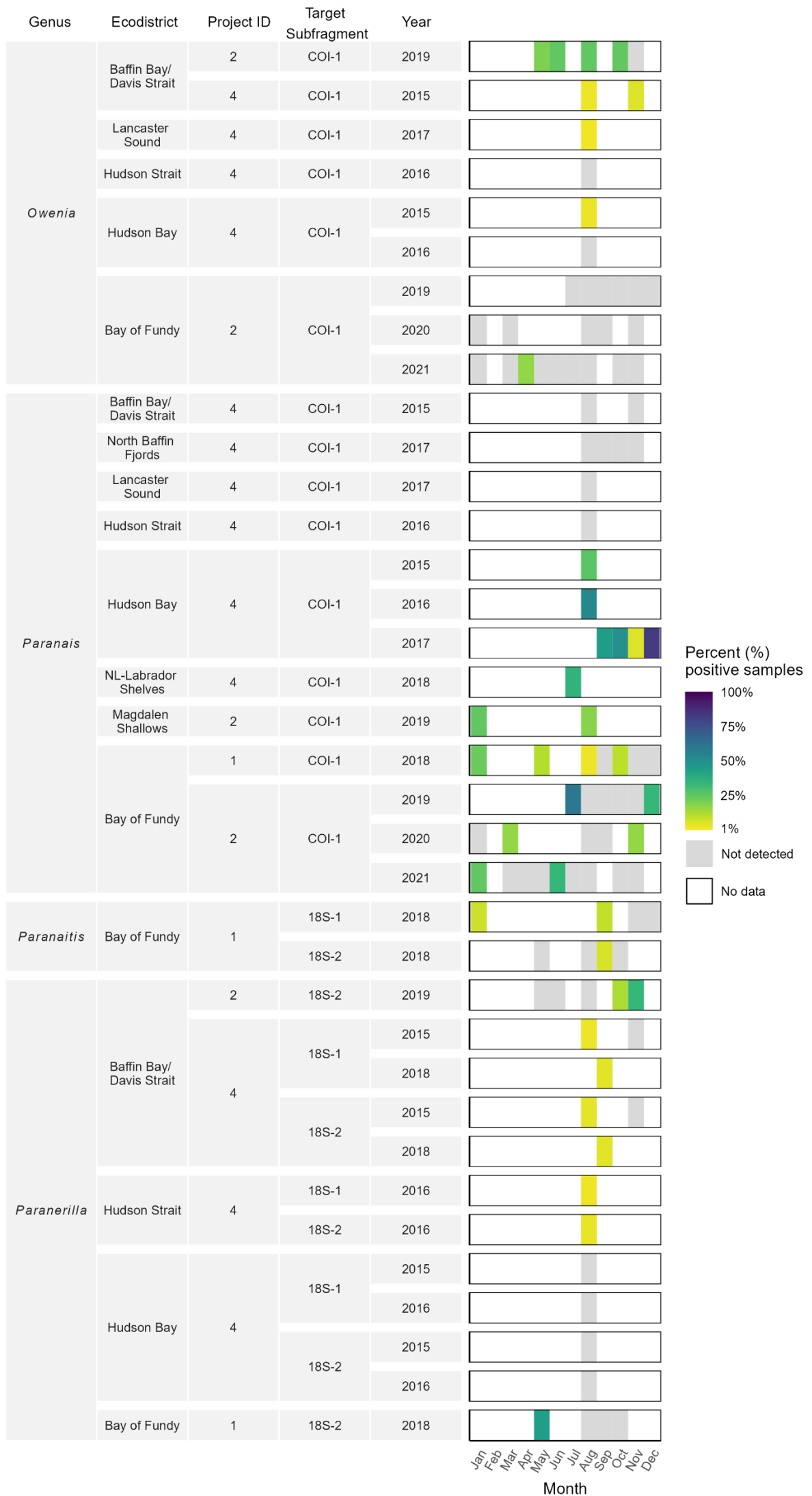


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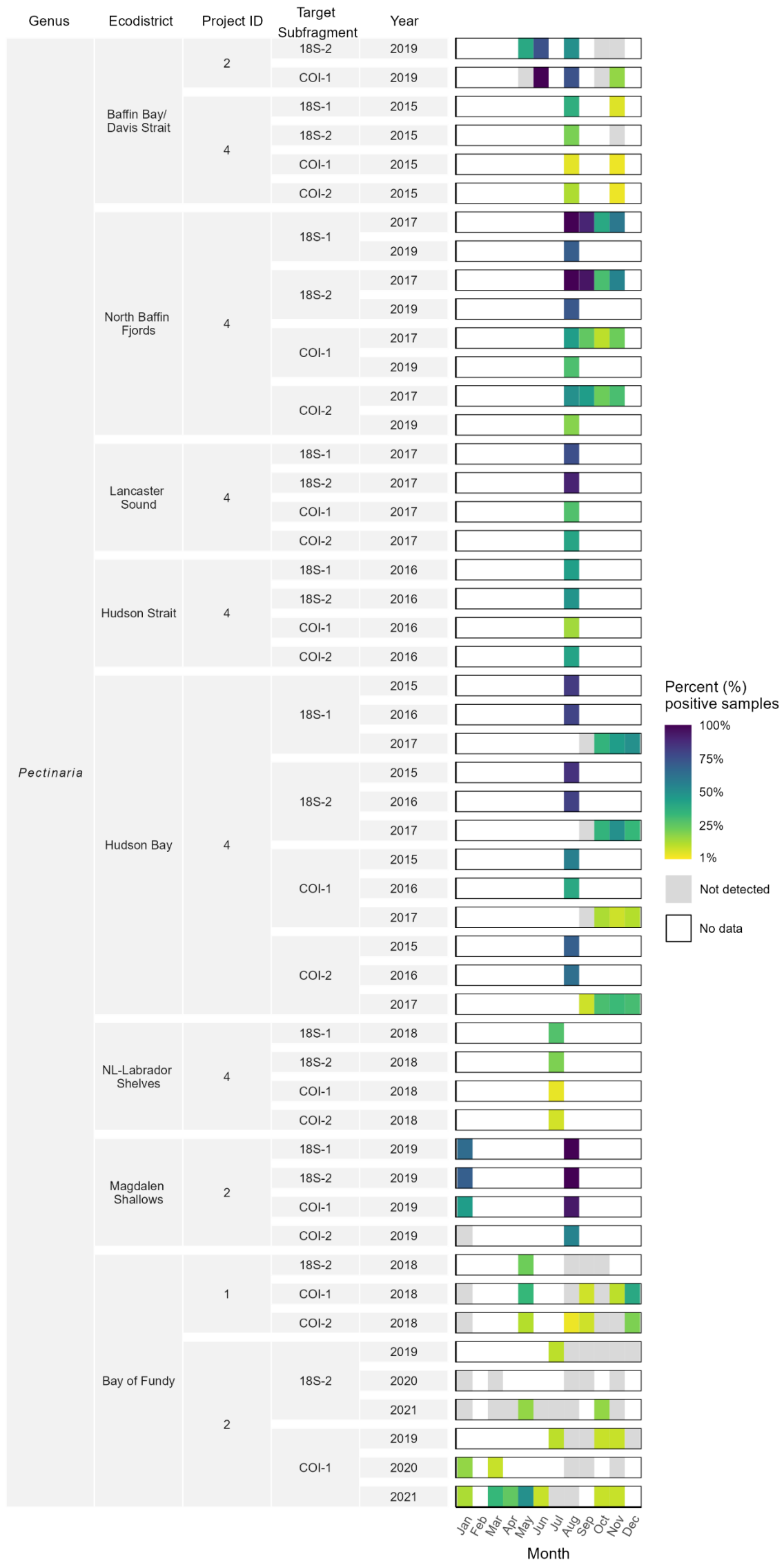


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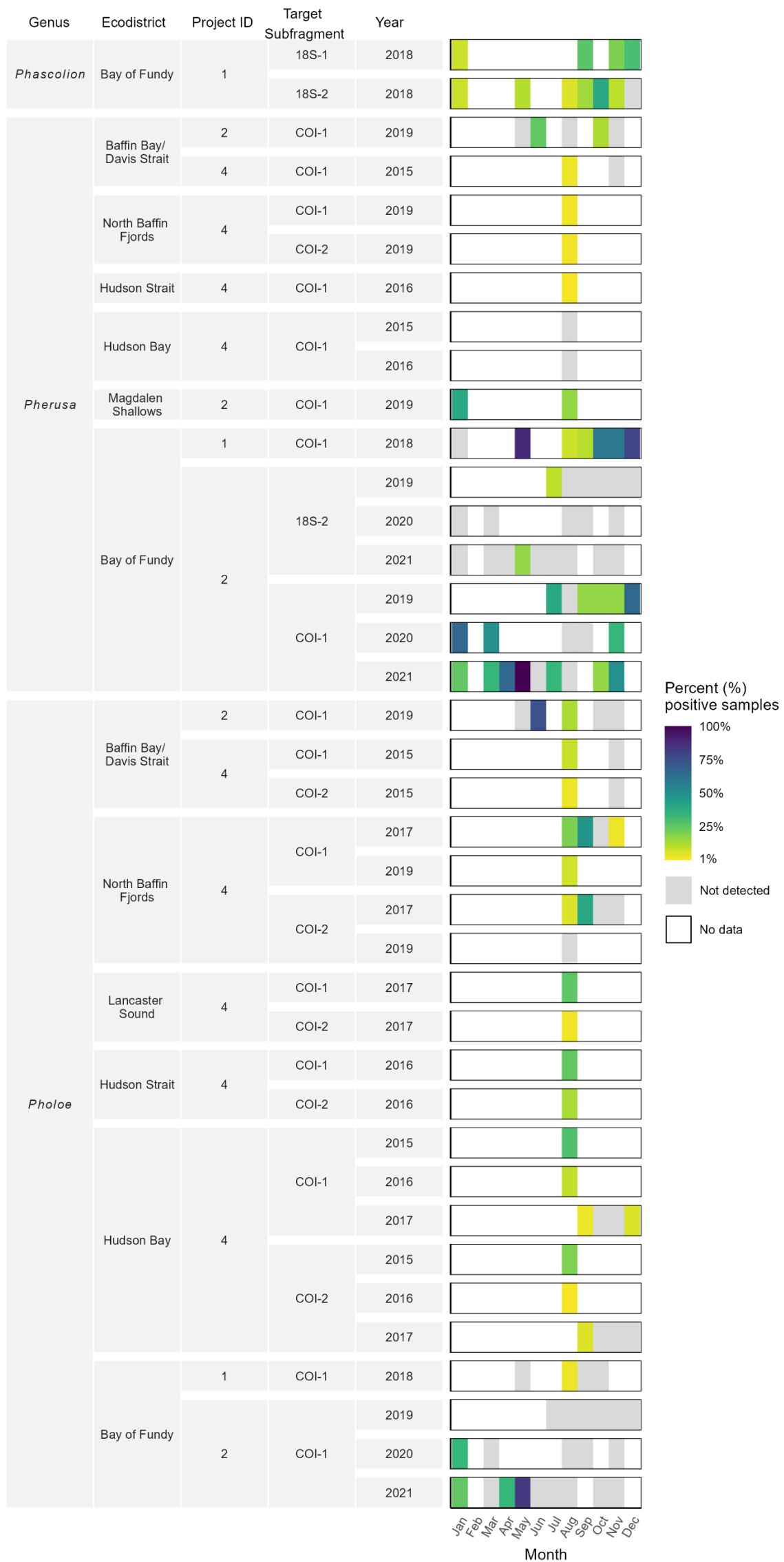


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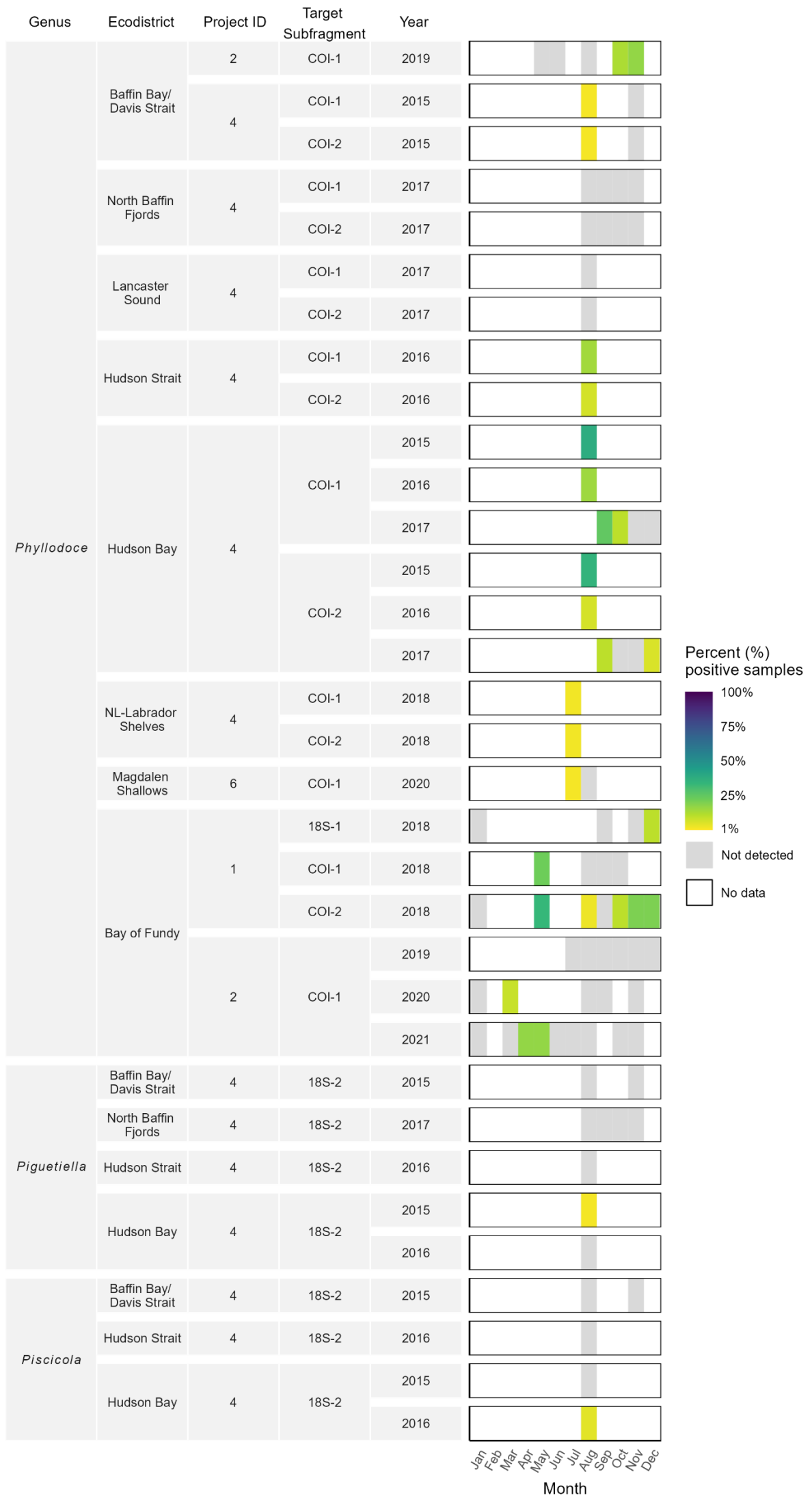


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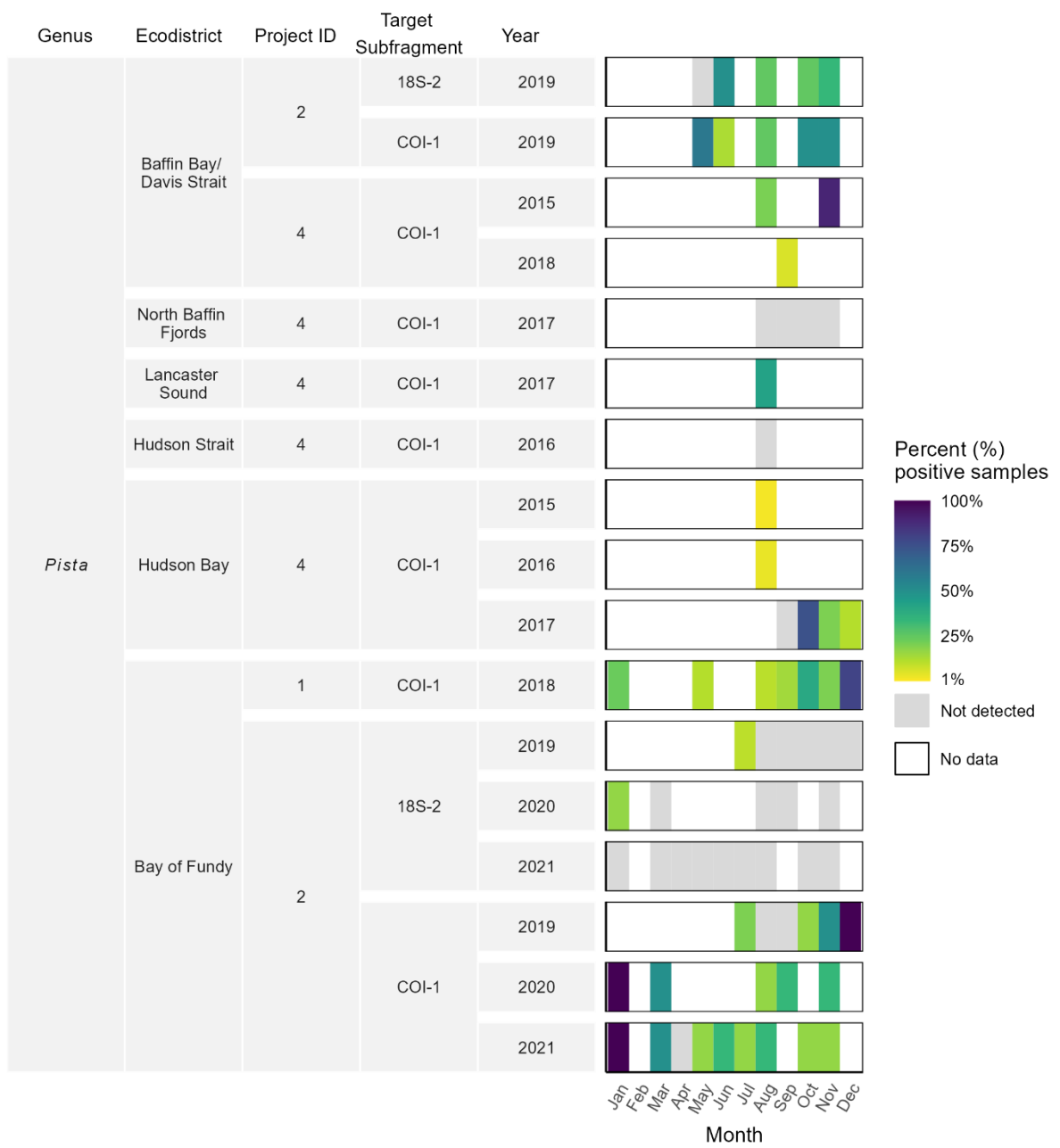


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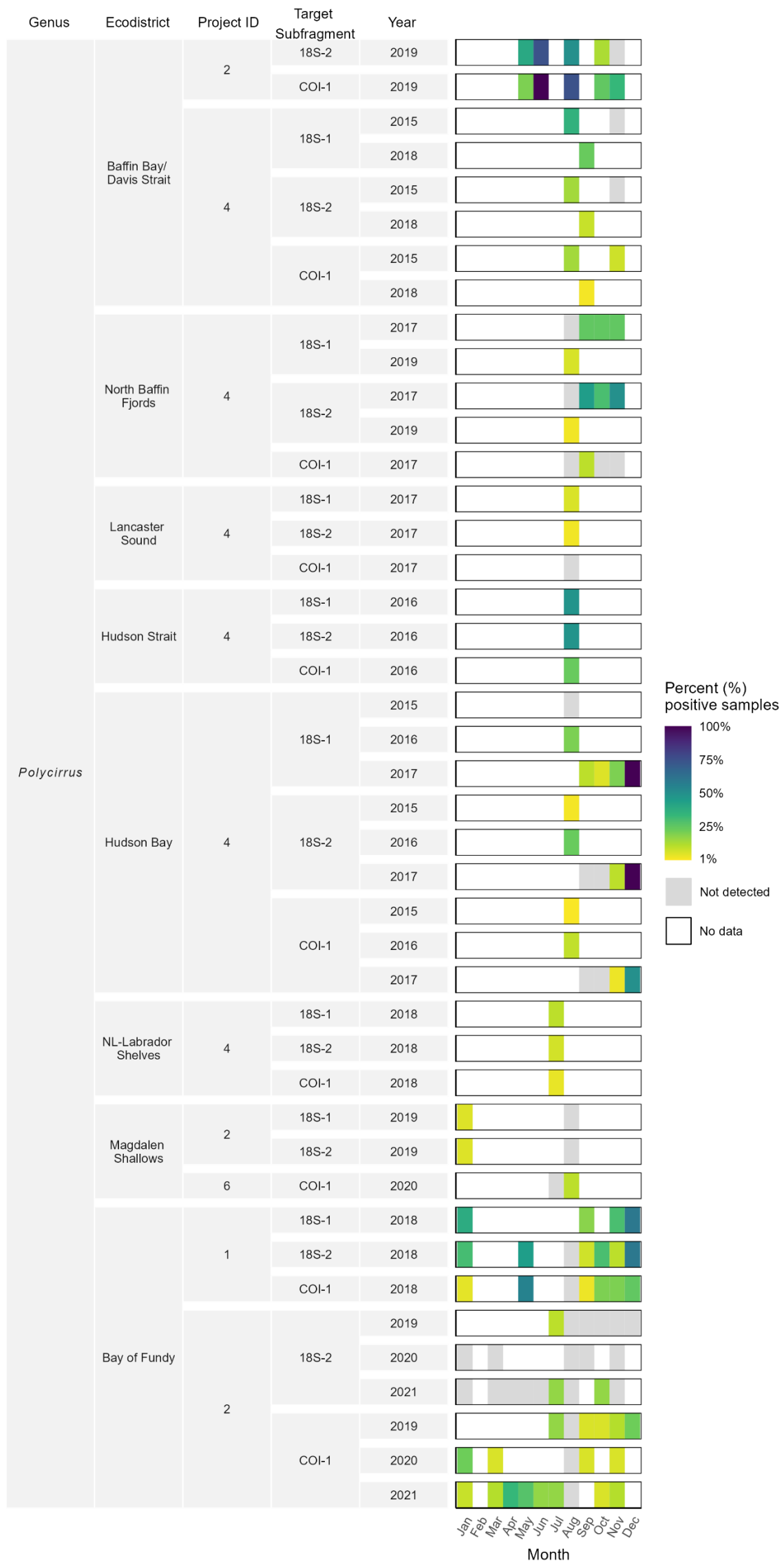


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Figure 13. (Continued)



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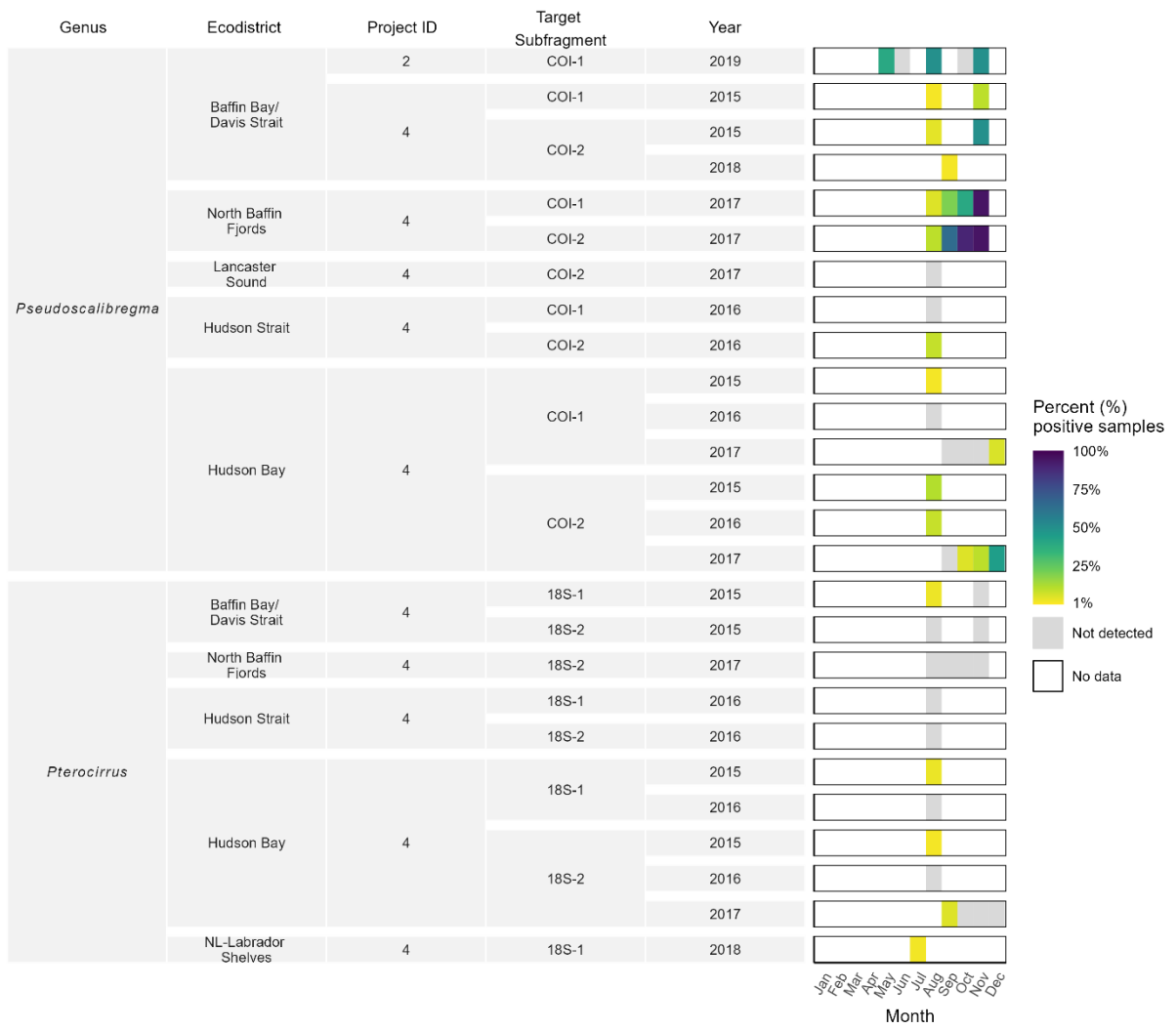


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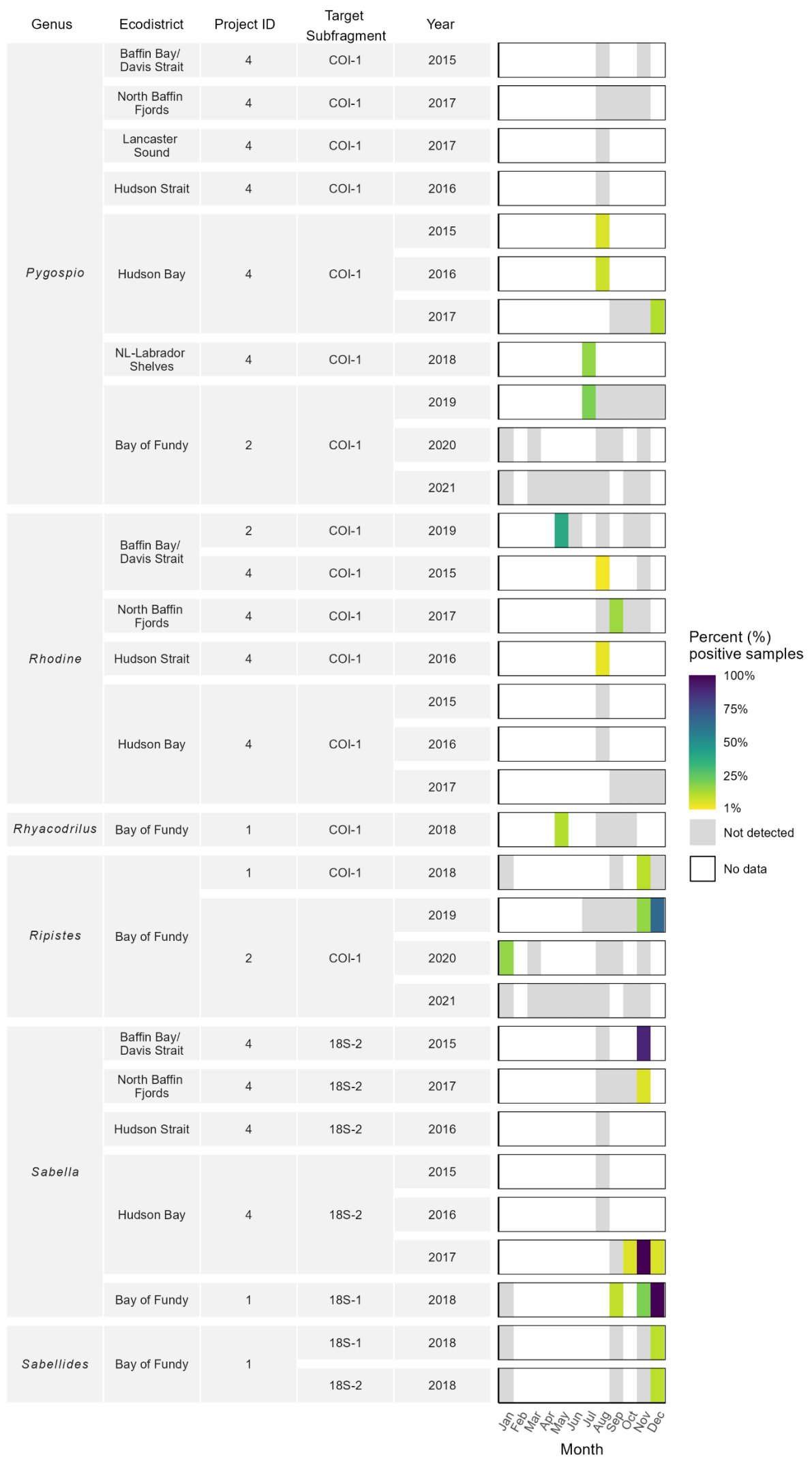


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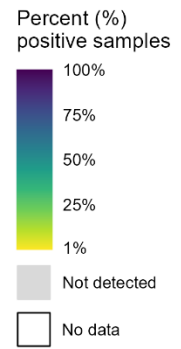
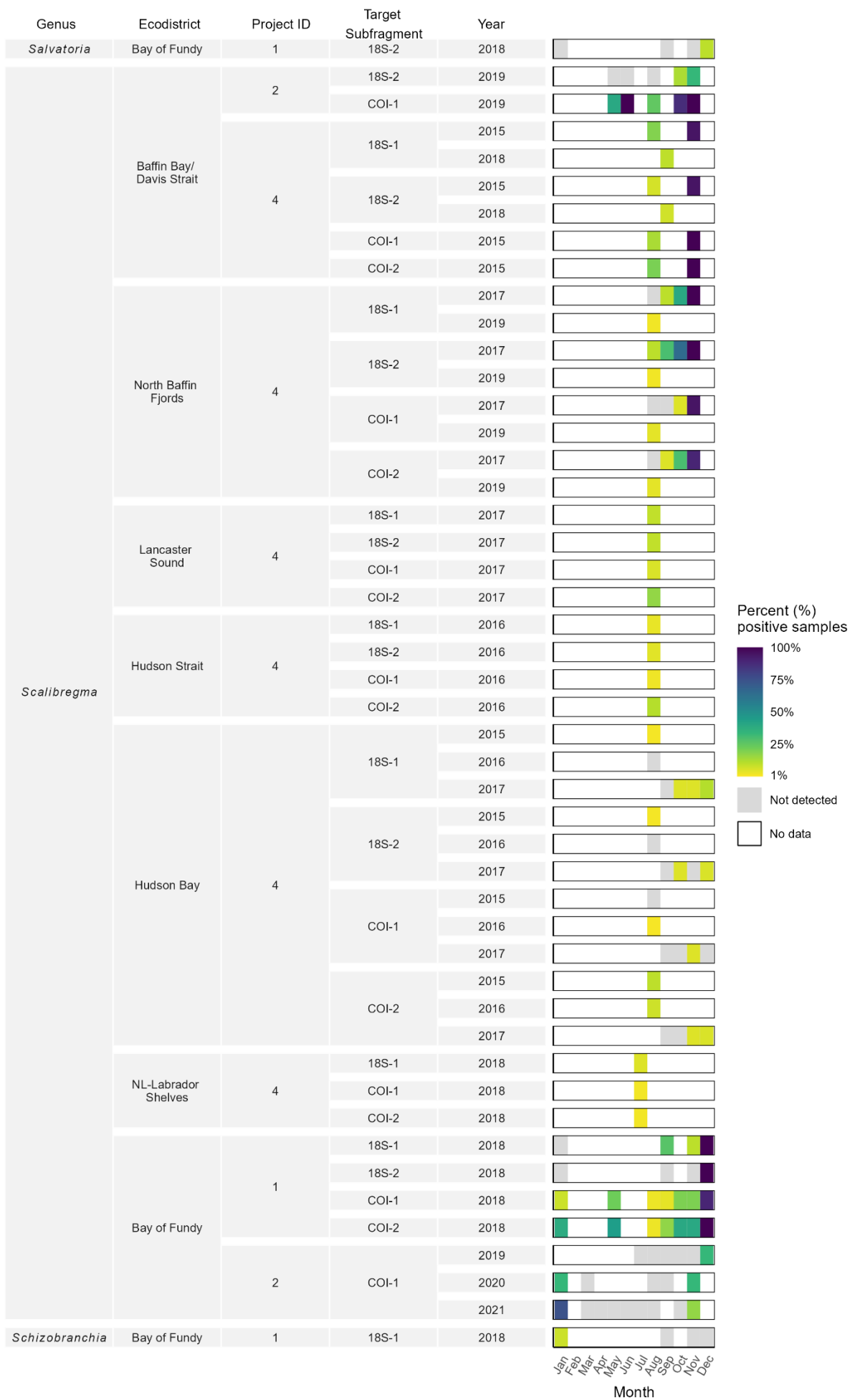


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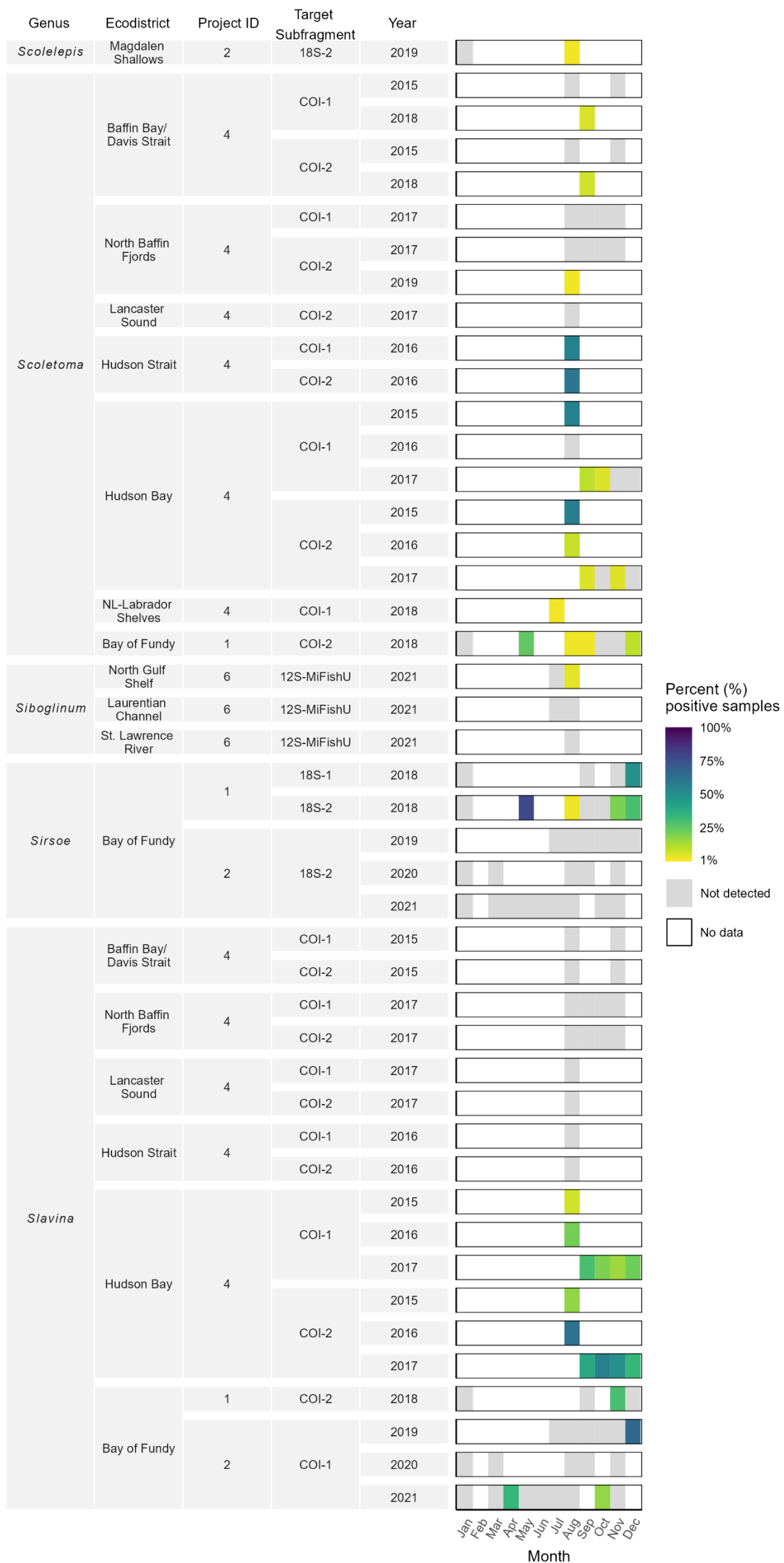


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Figure 13. (Continued)

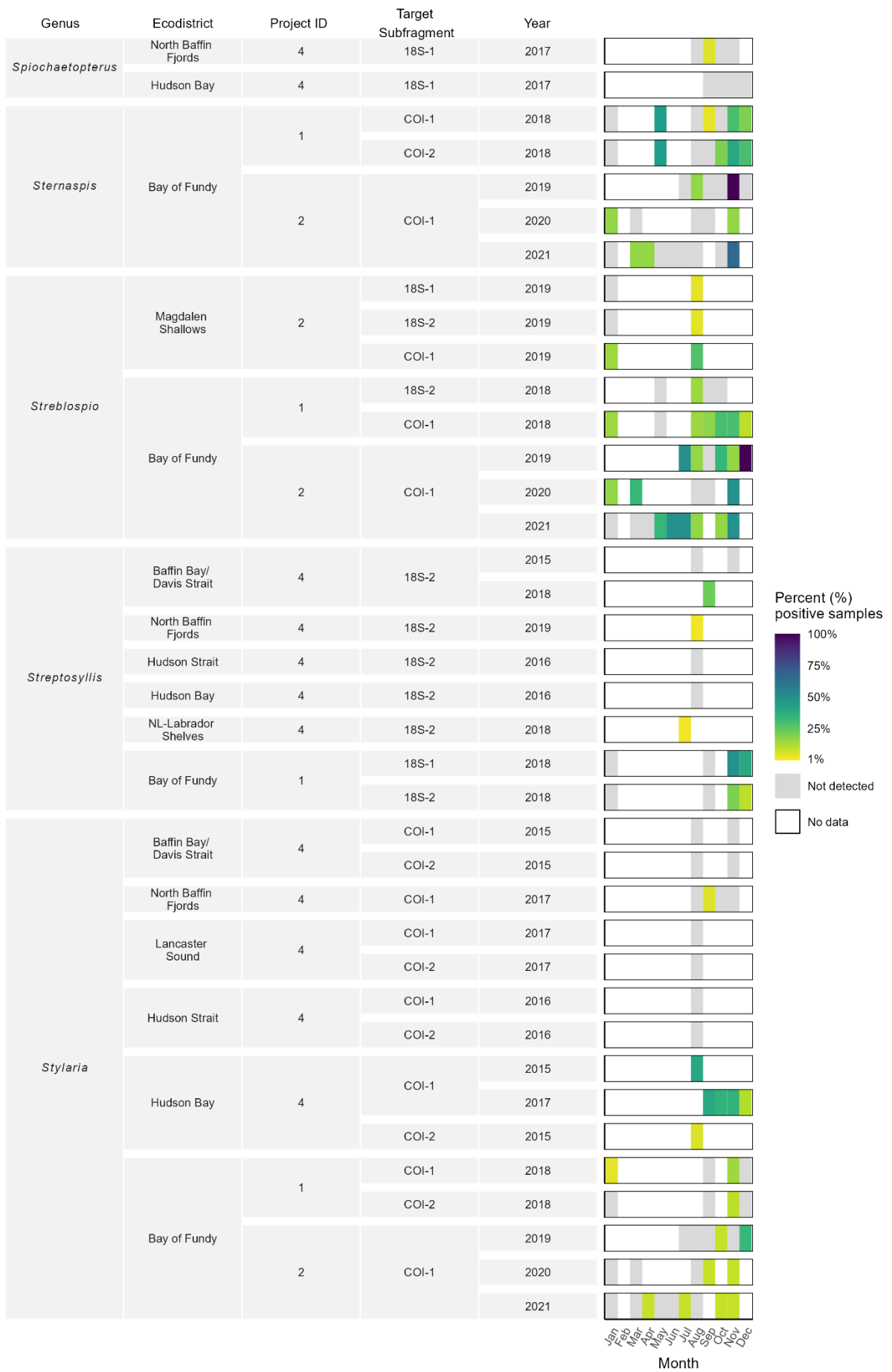


Figure 13. (Continued)



Figure 13. (Continued)



Figure 13. (Continued)

6.2 ARTHROPODA

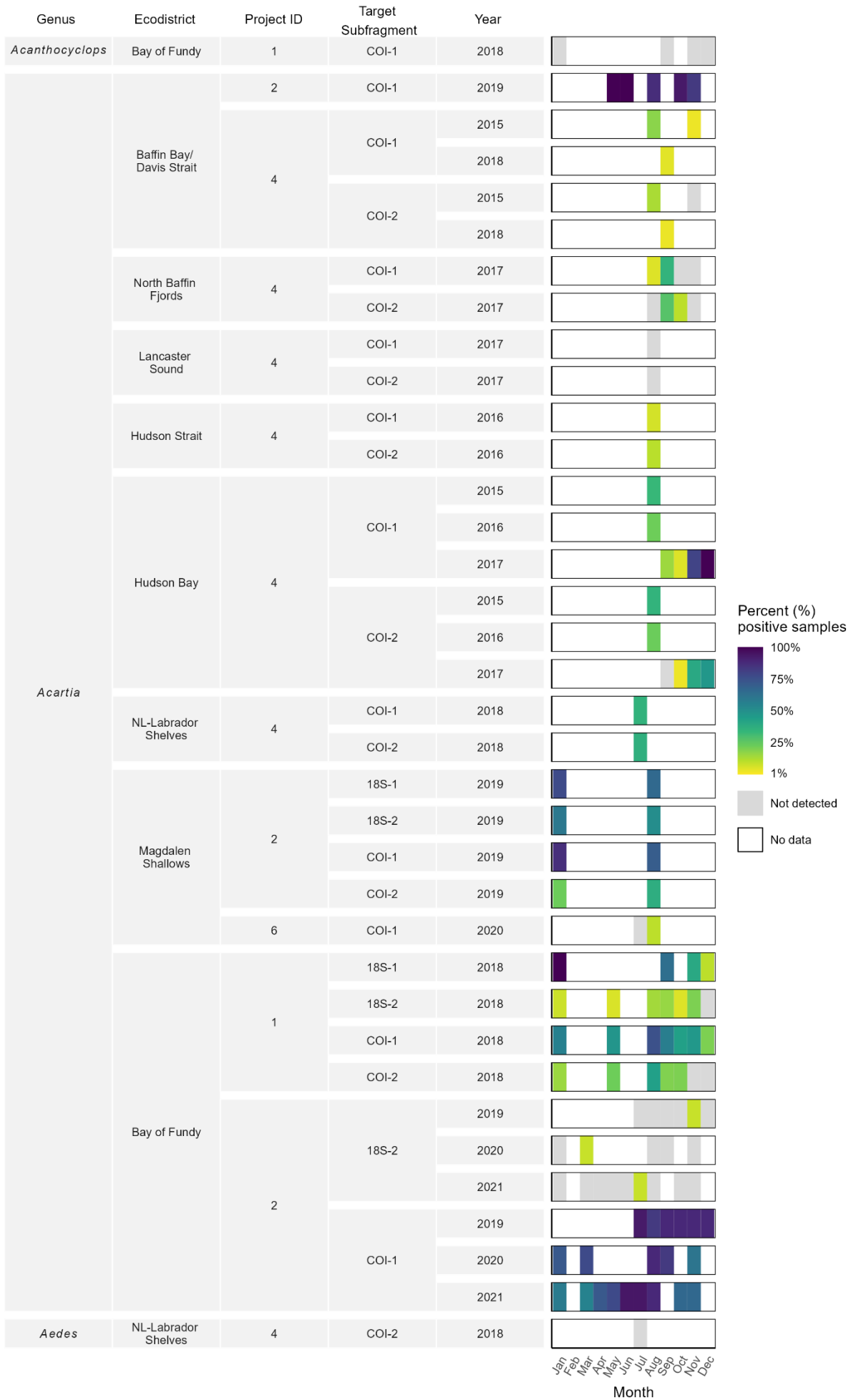


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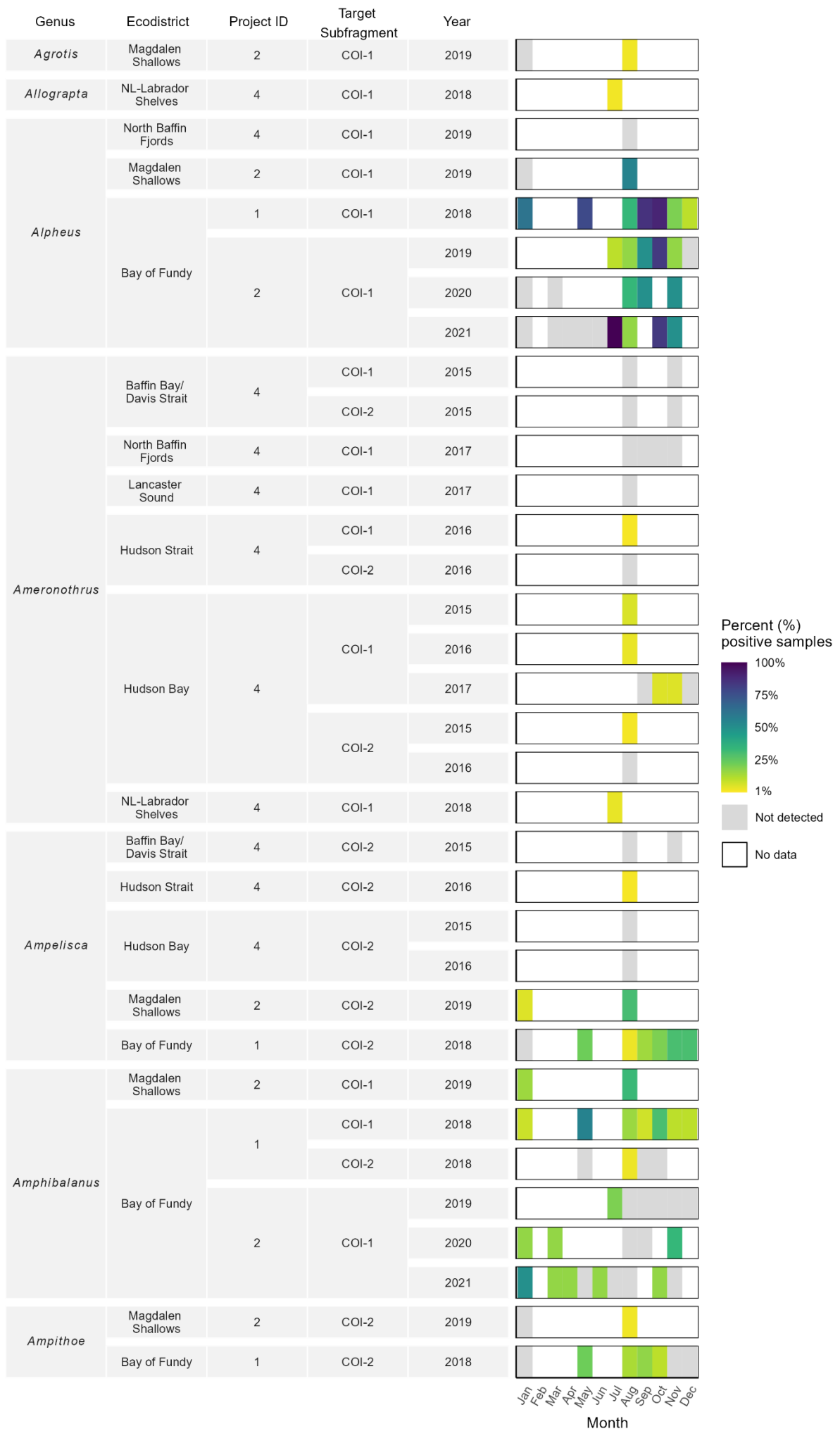


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Figure 13. (Continued)

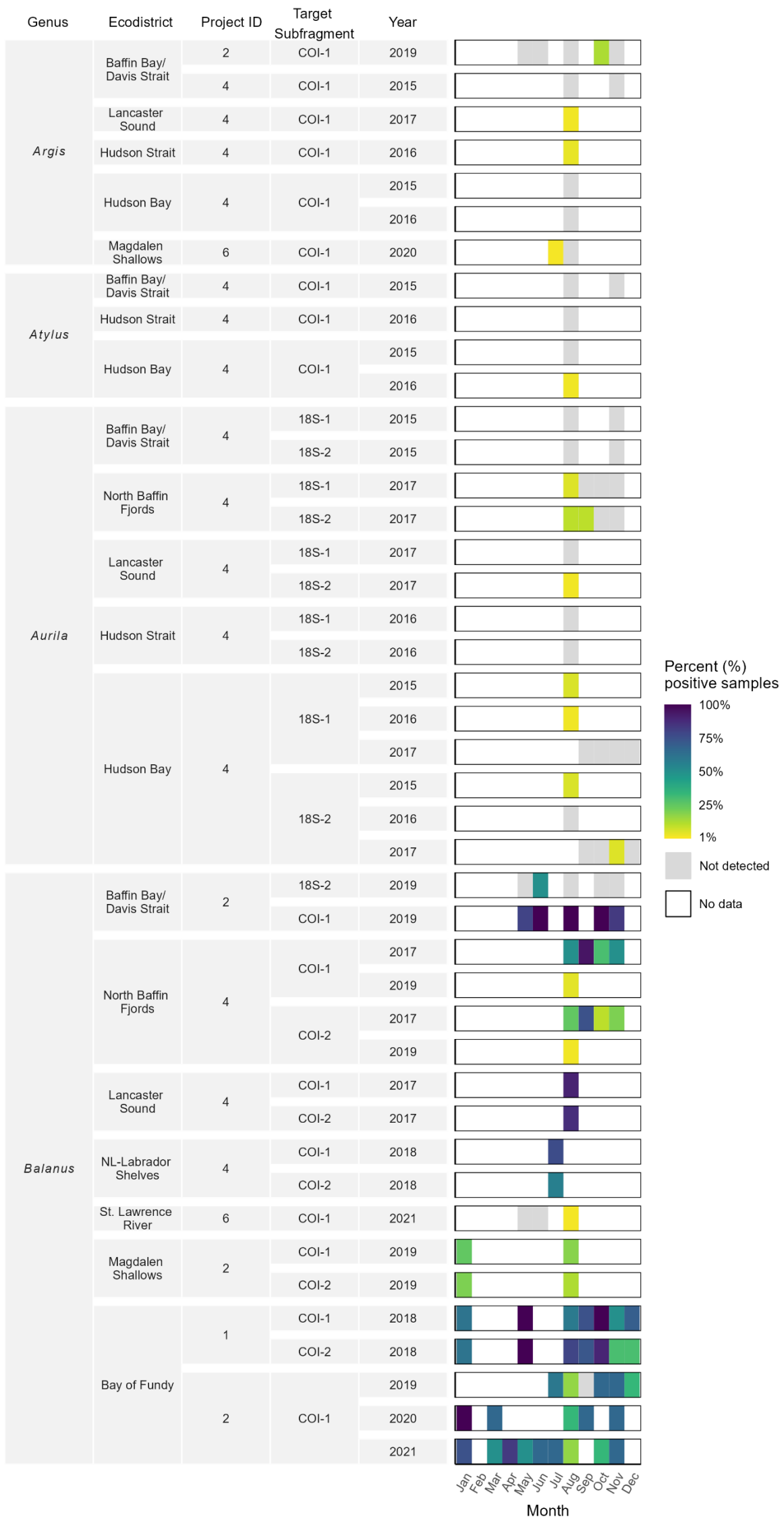


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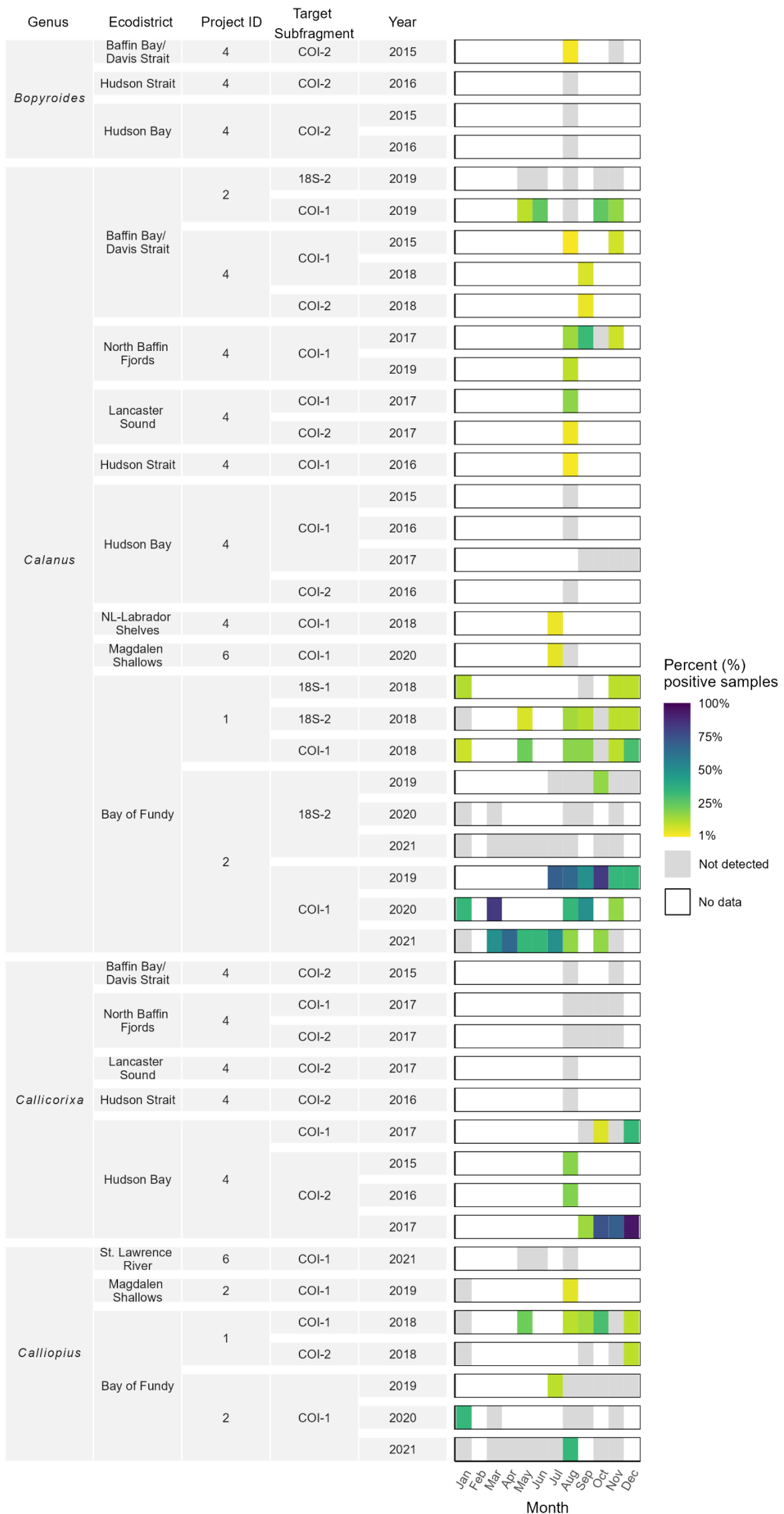


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Figure 13. (Continued)

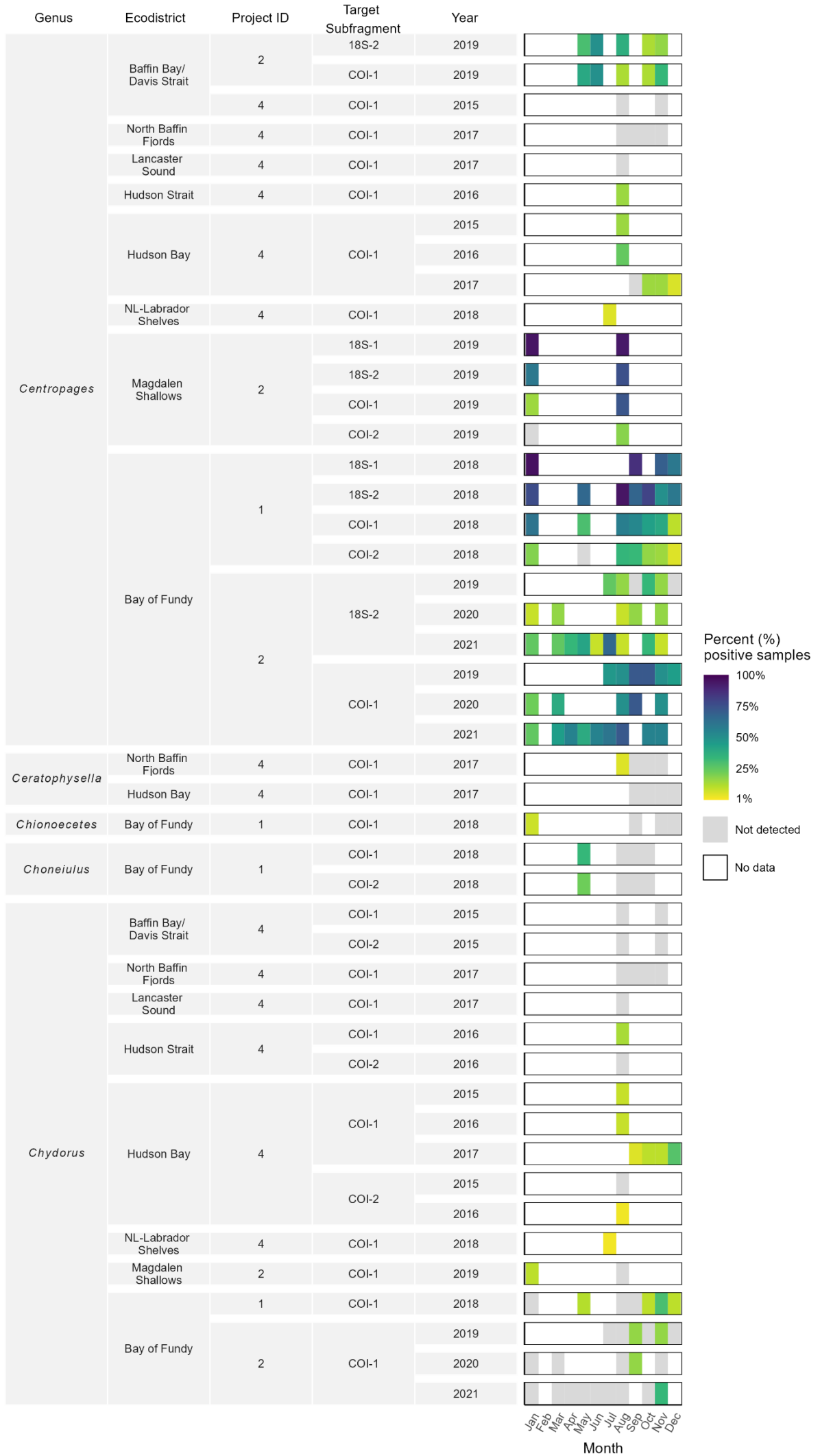


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Figure 13. (Continued)

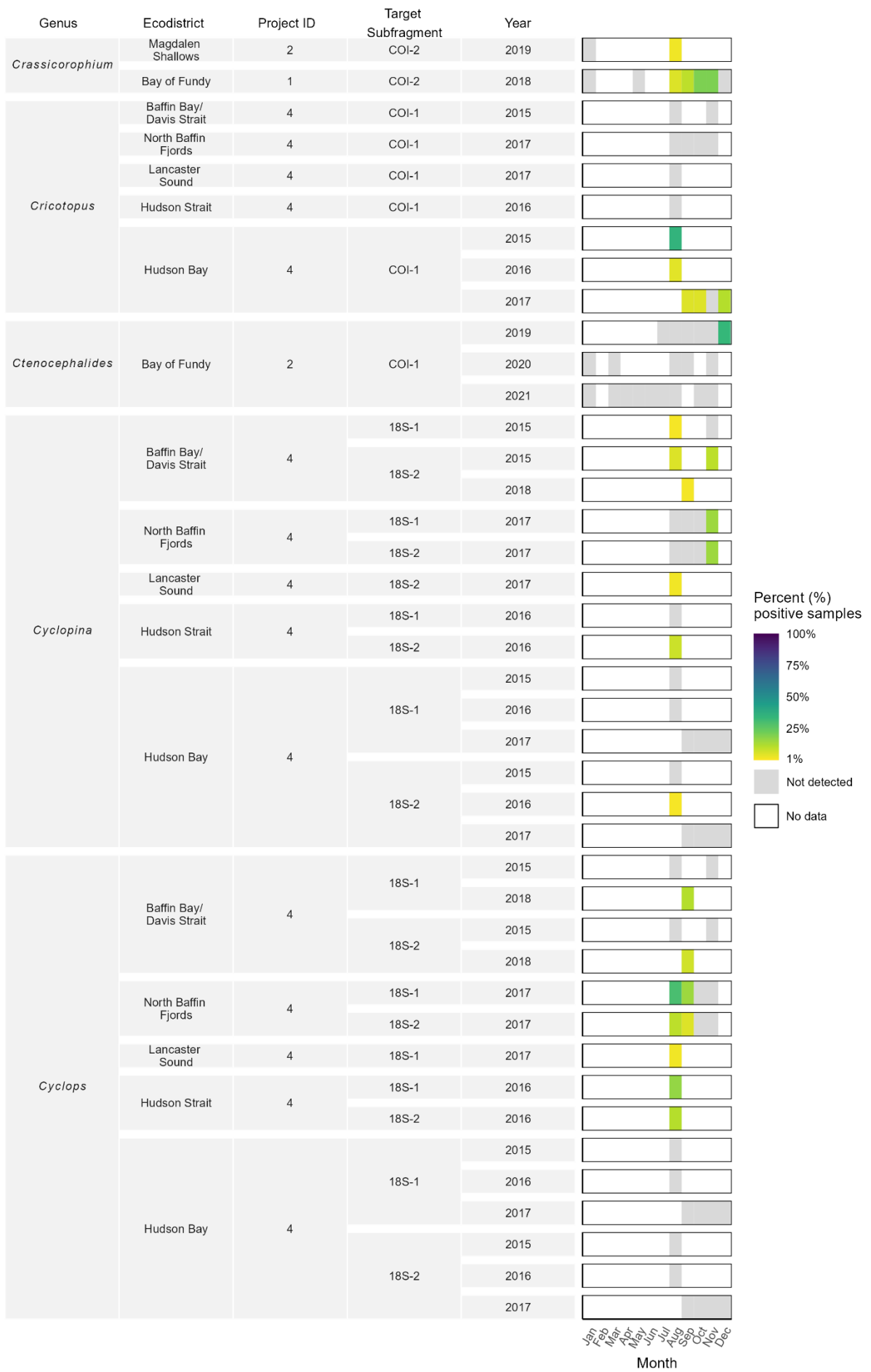


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Figure 13. (Continued)

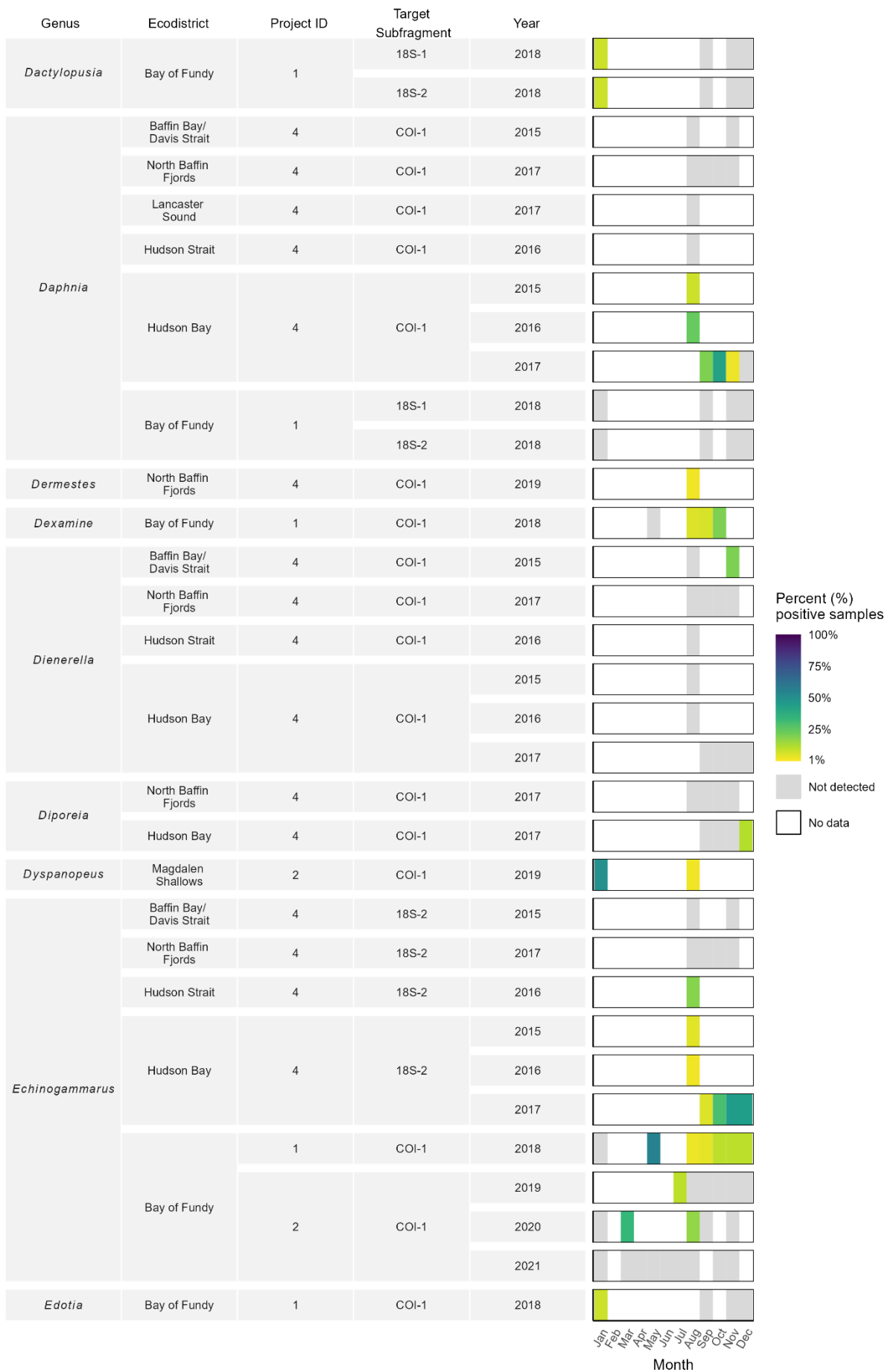


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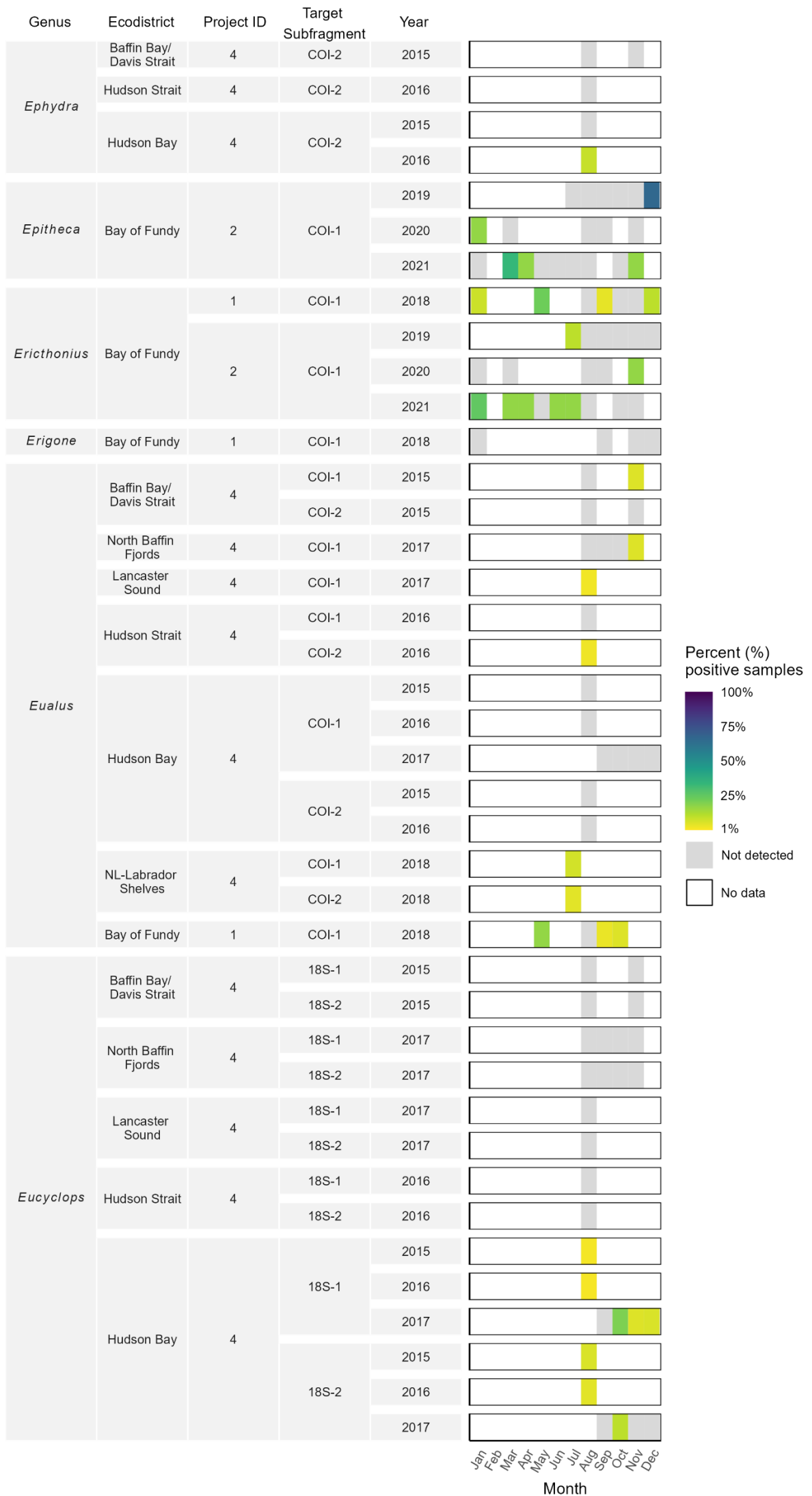


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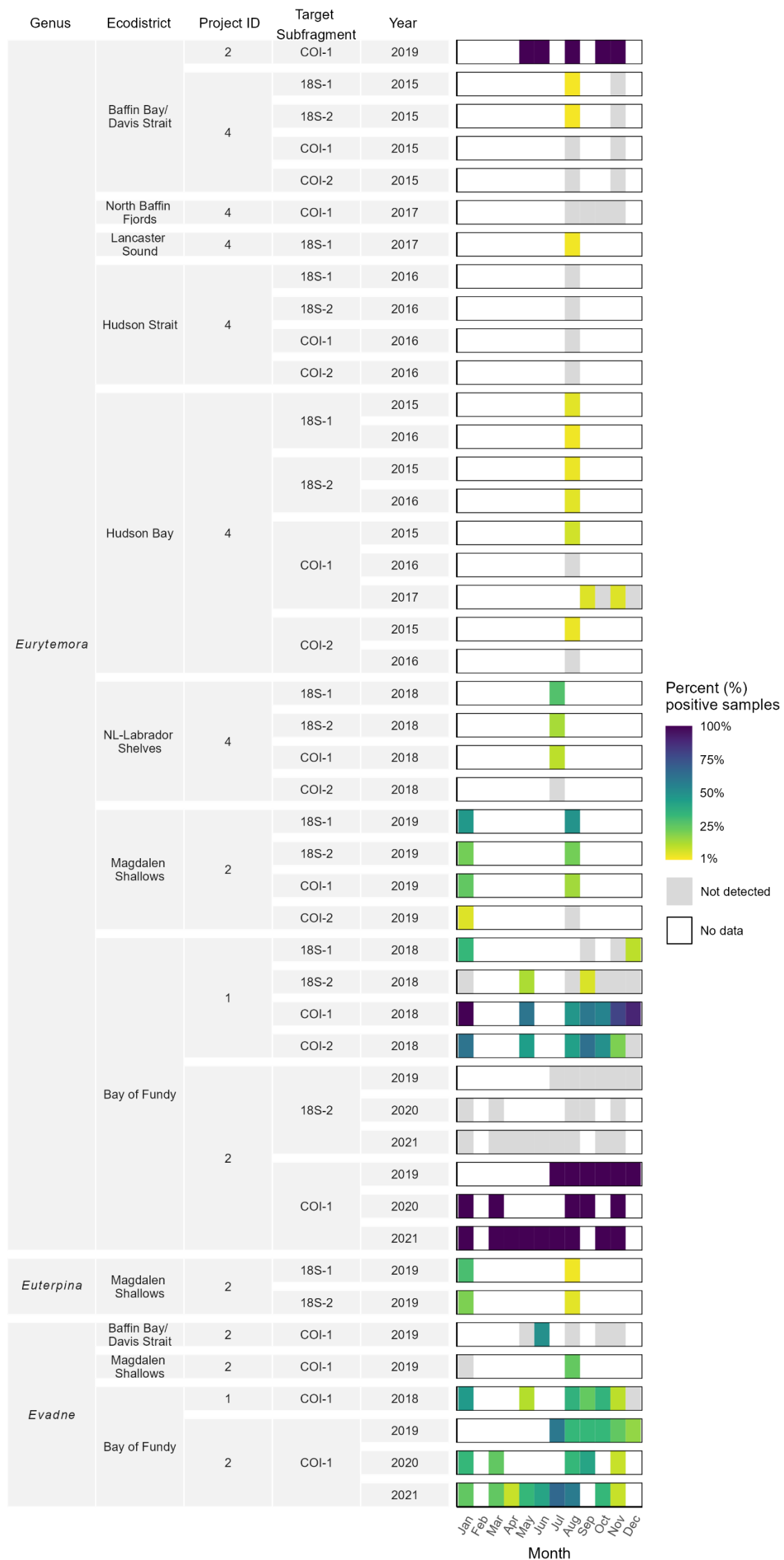


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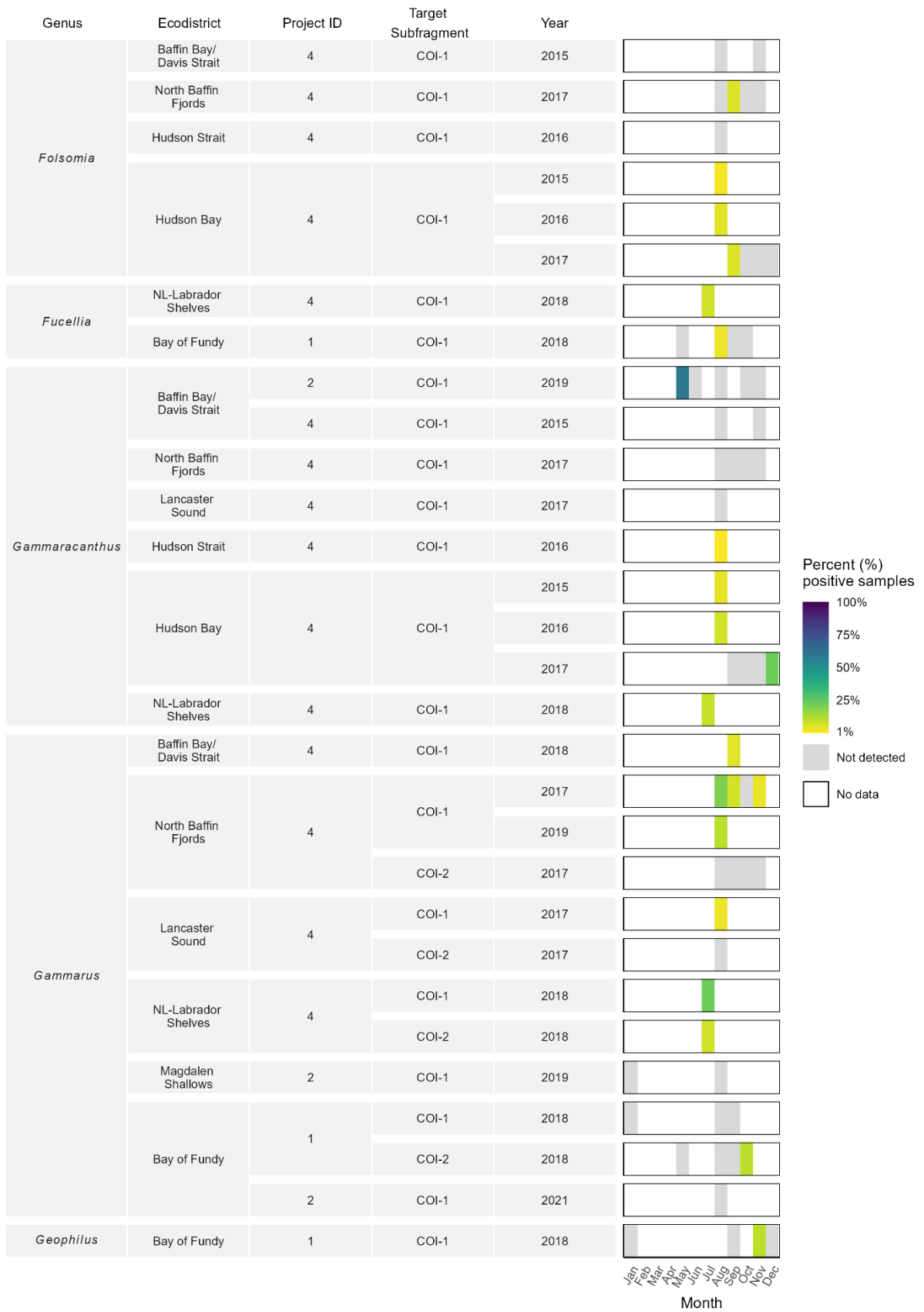


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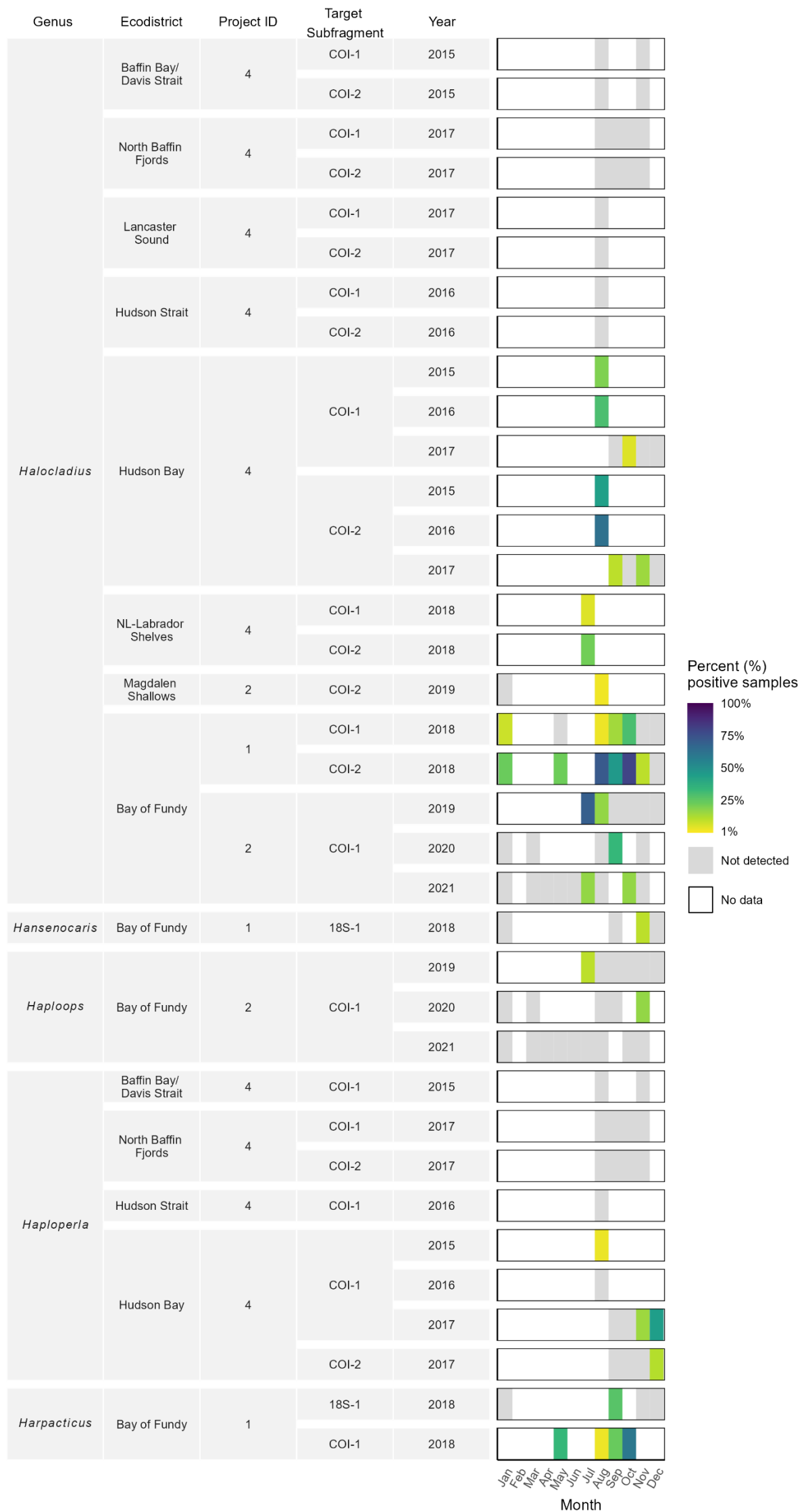


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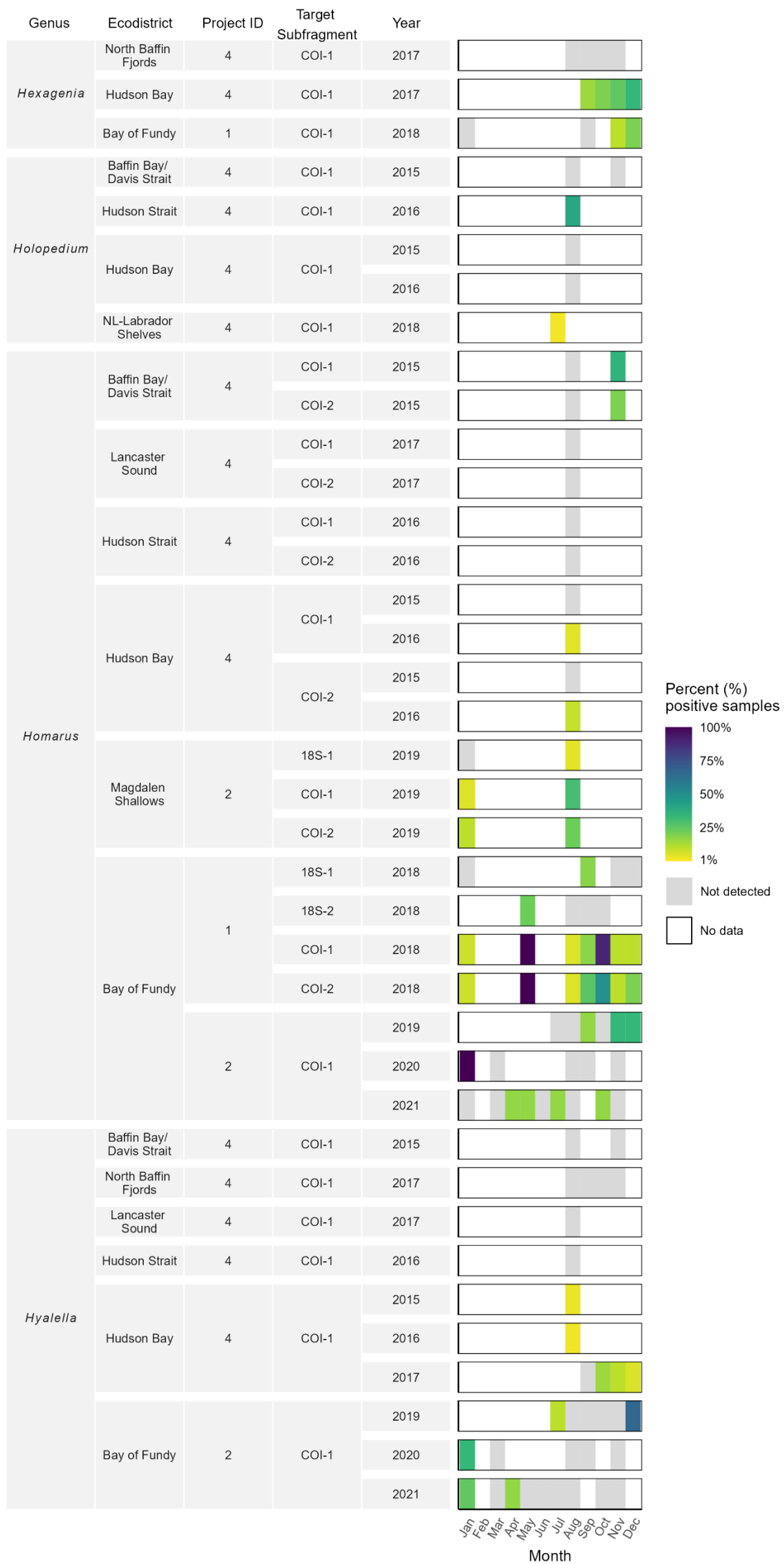


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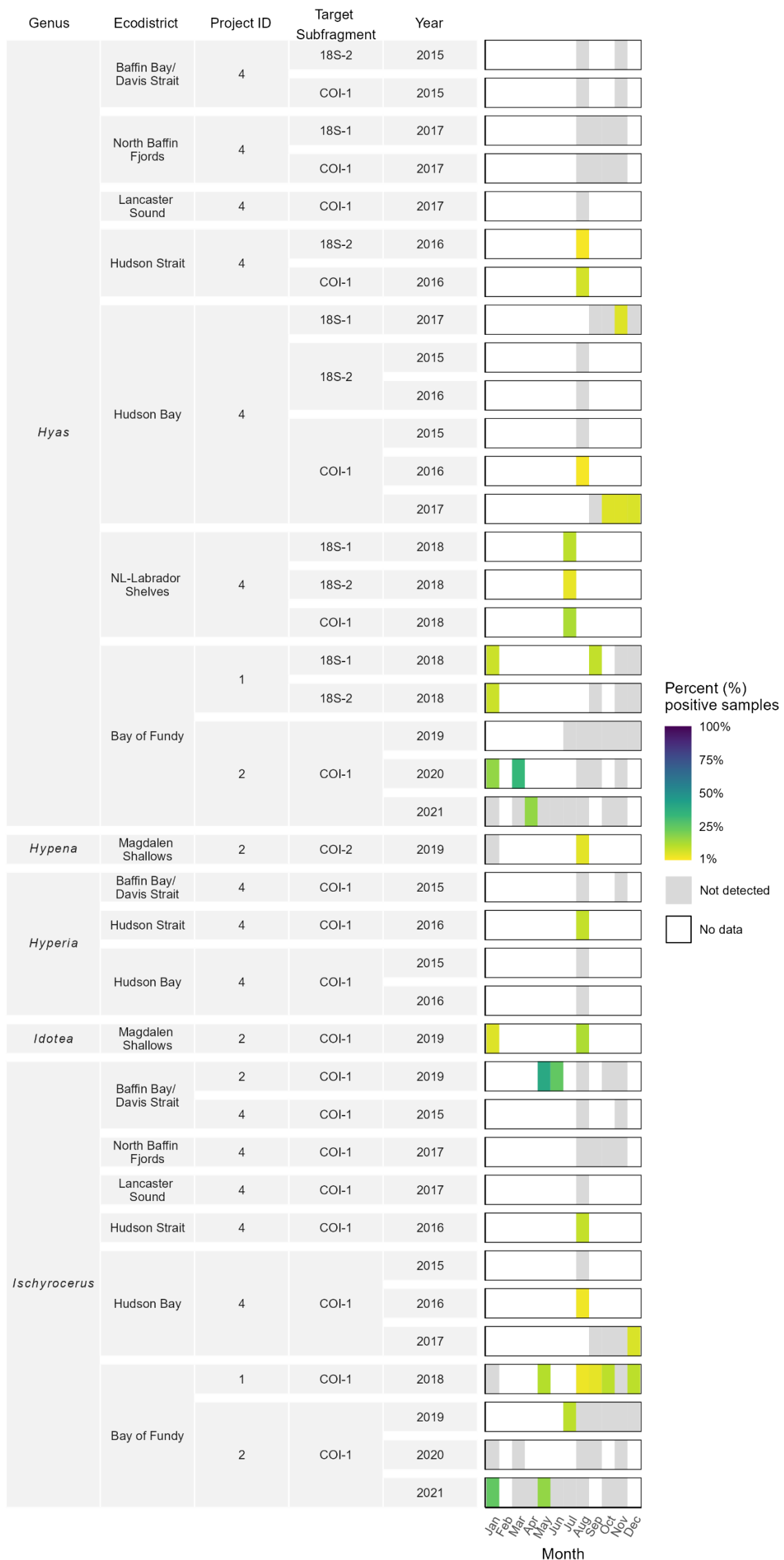


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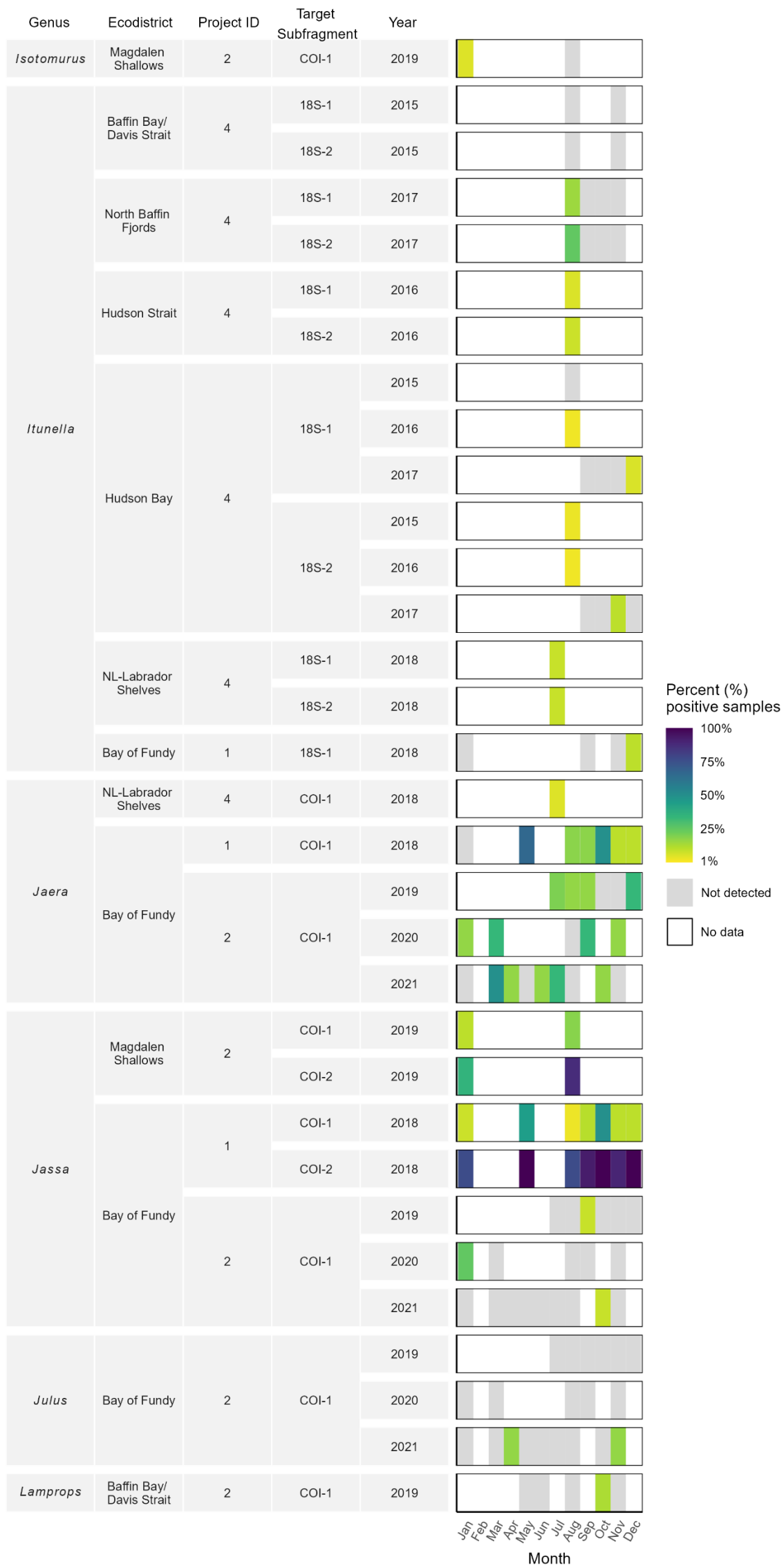


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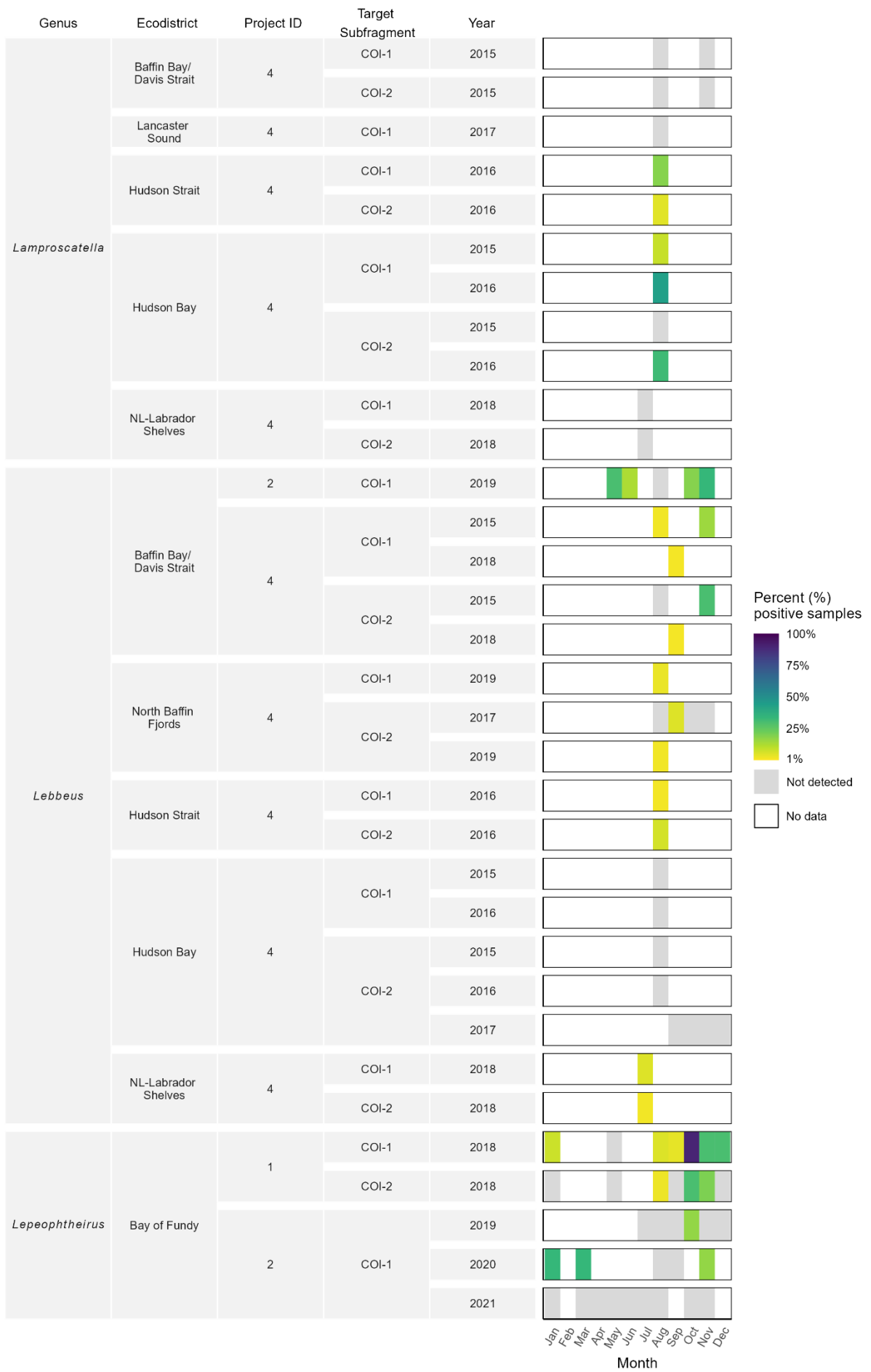


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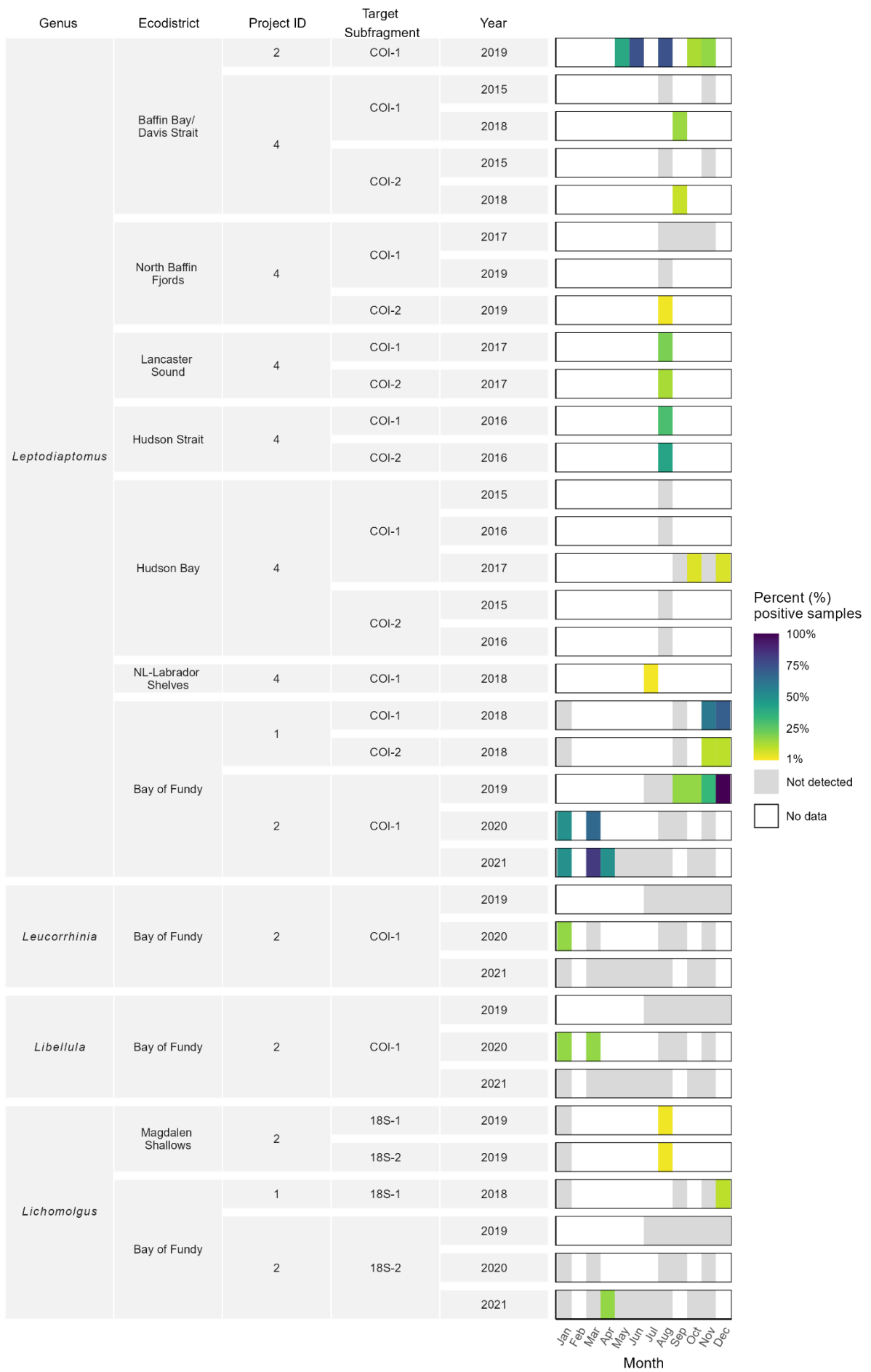


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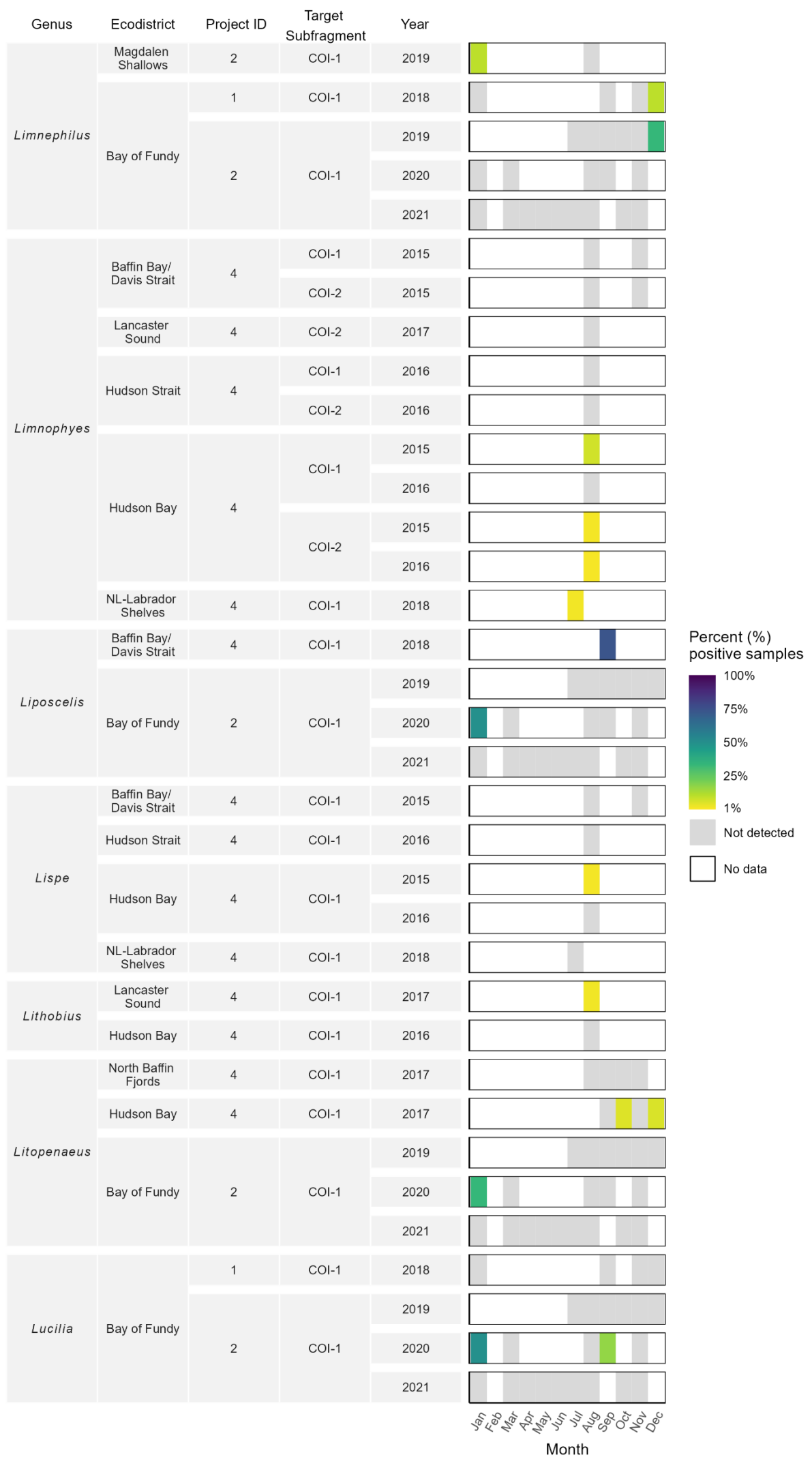


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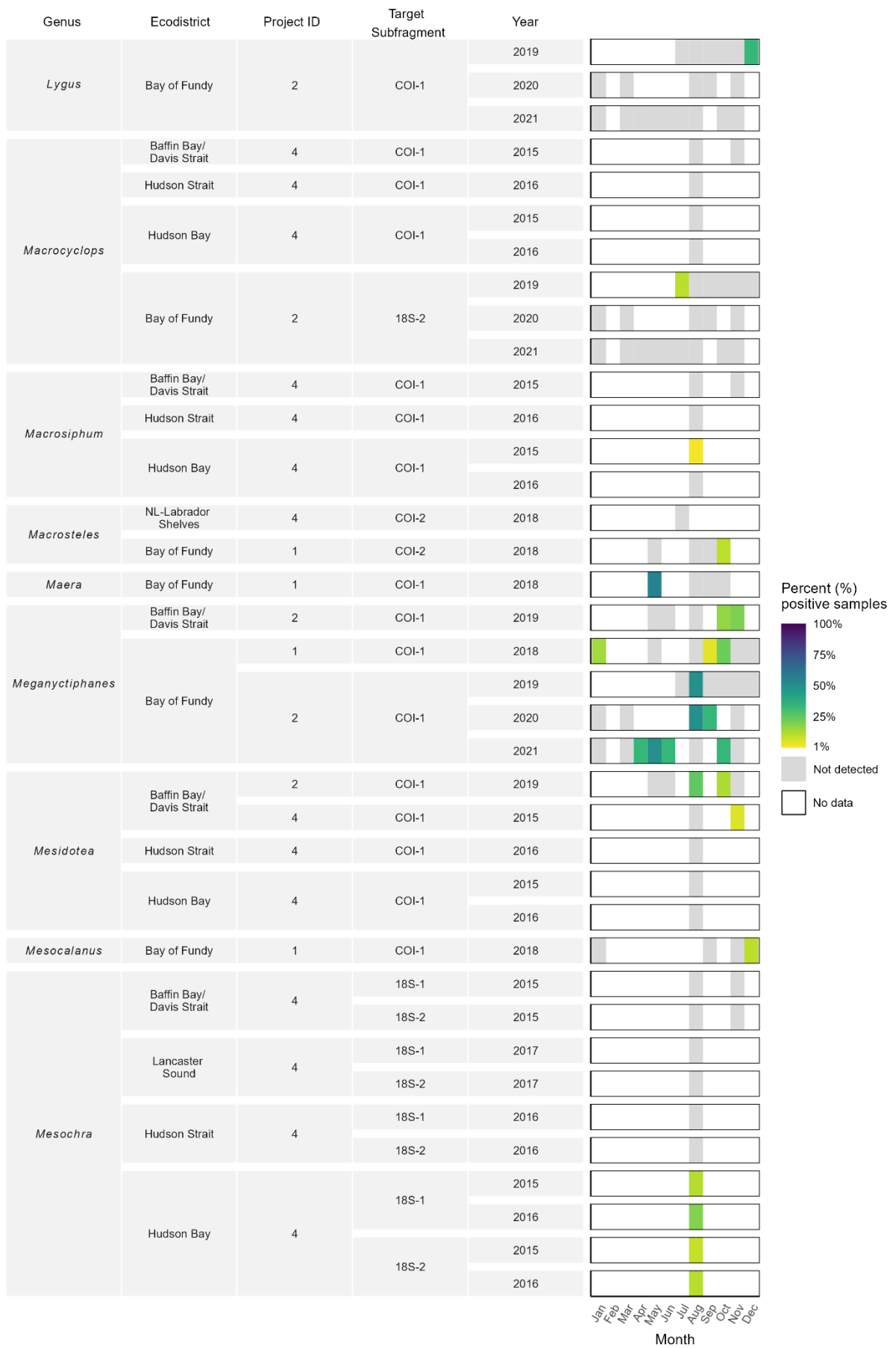


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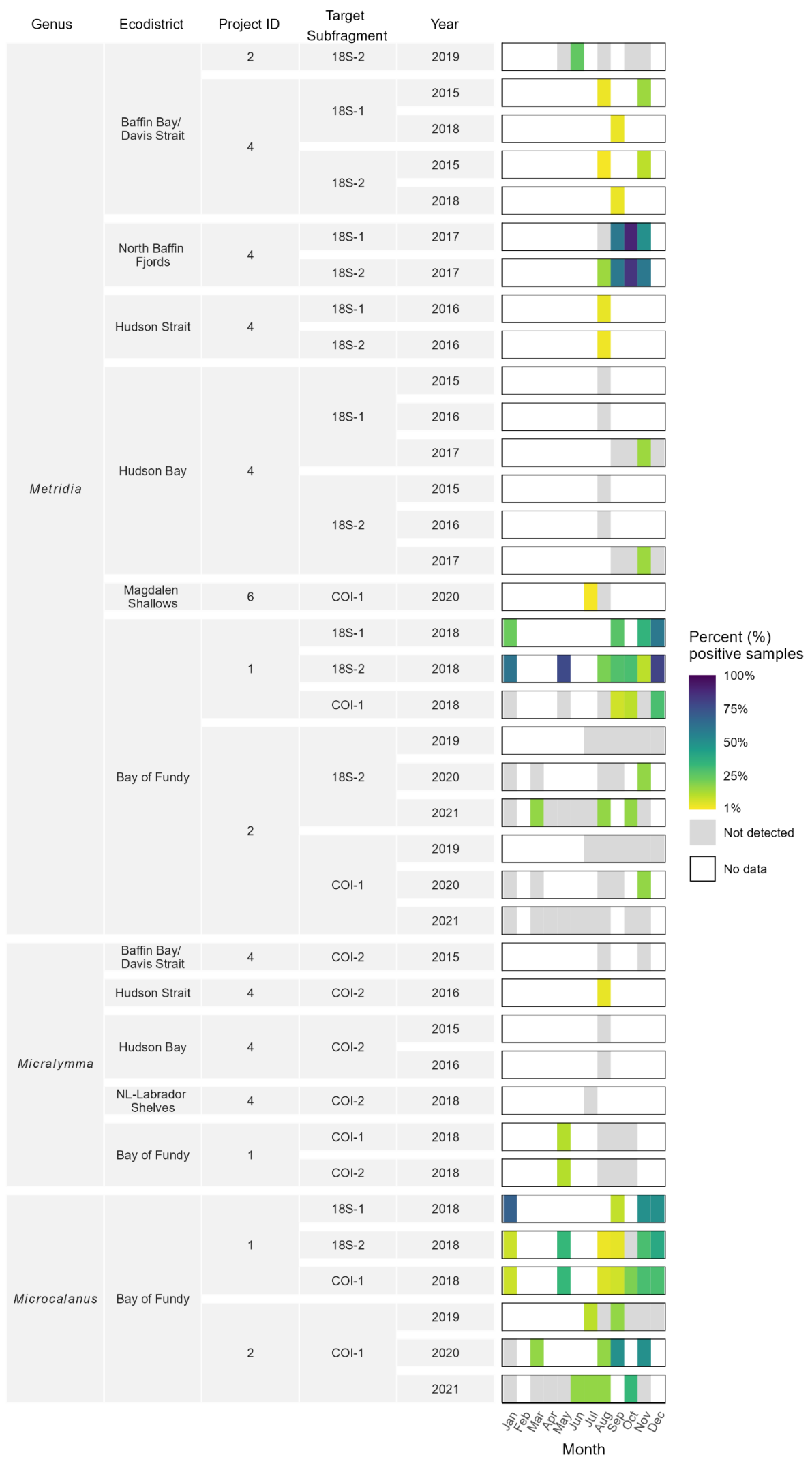


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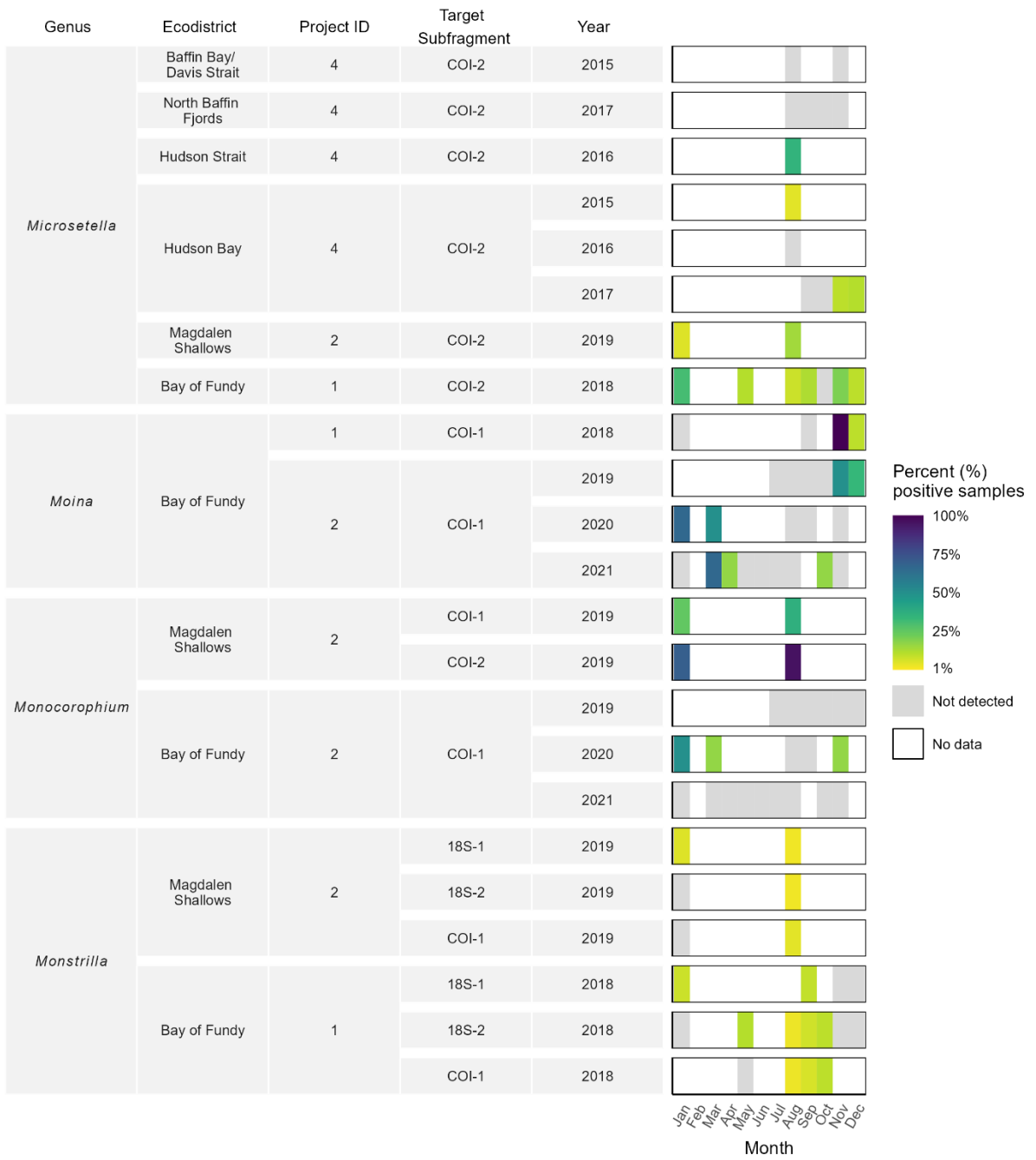


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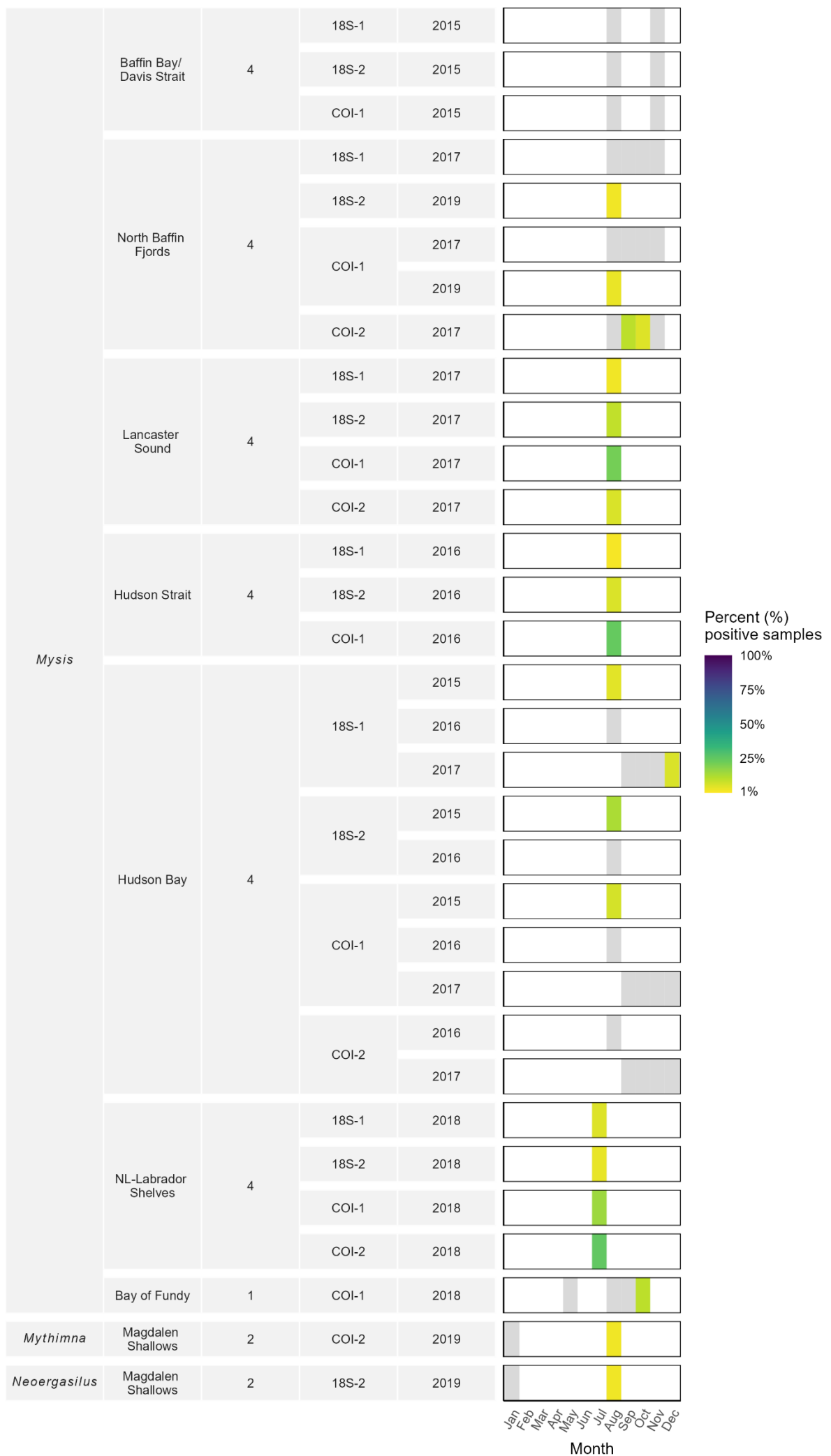


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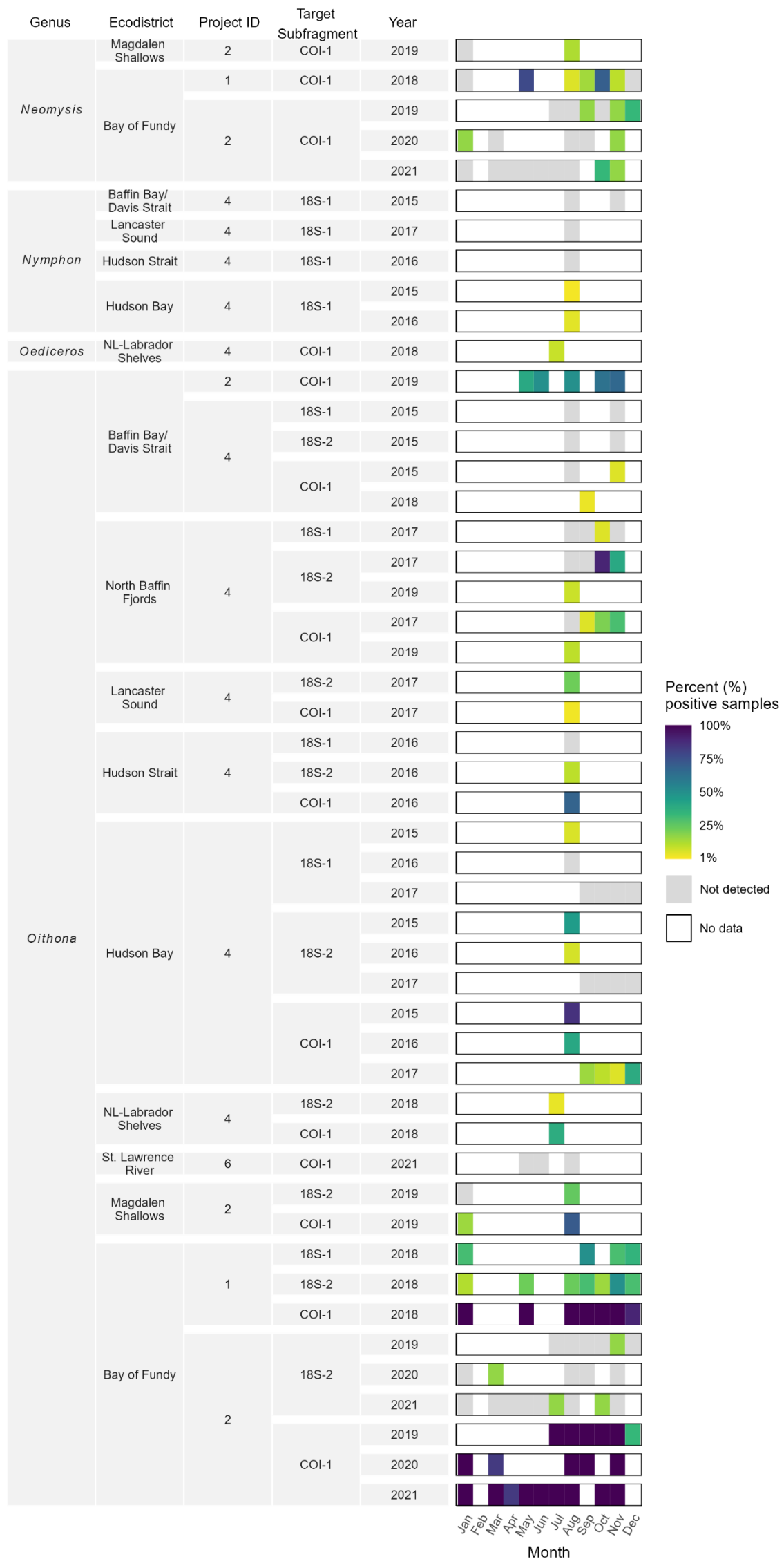


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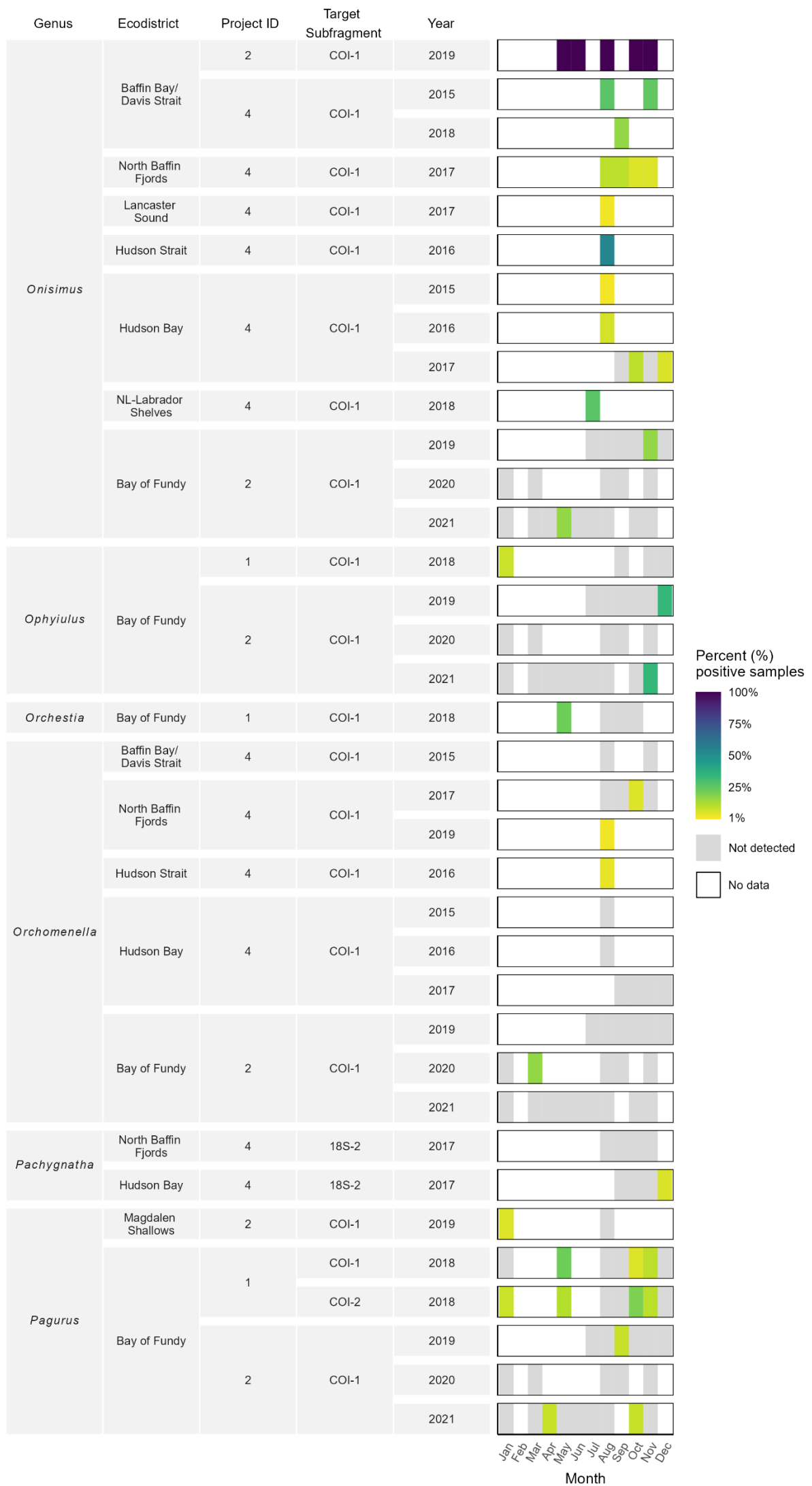


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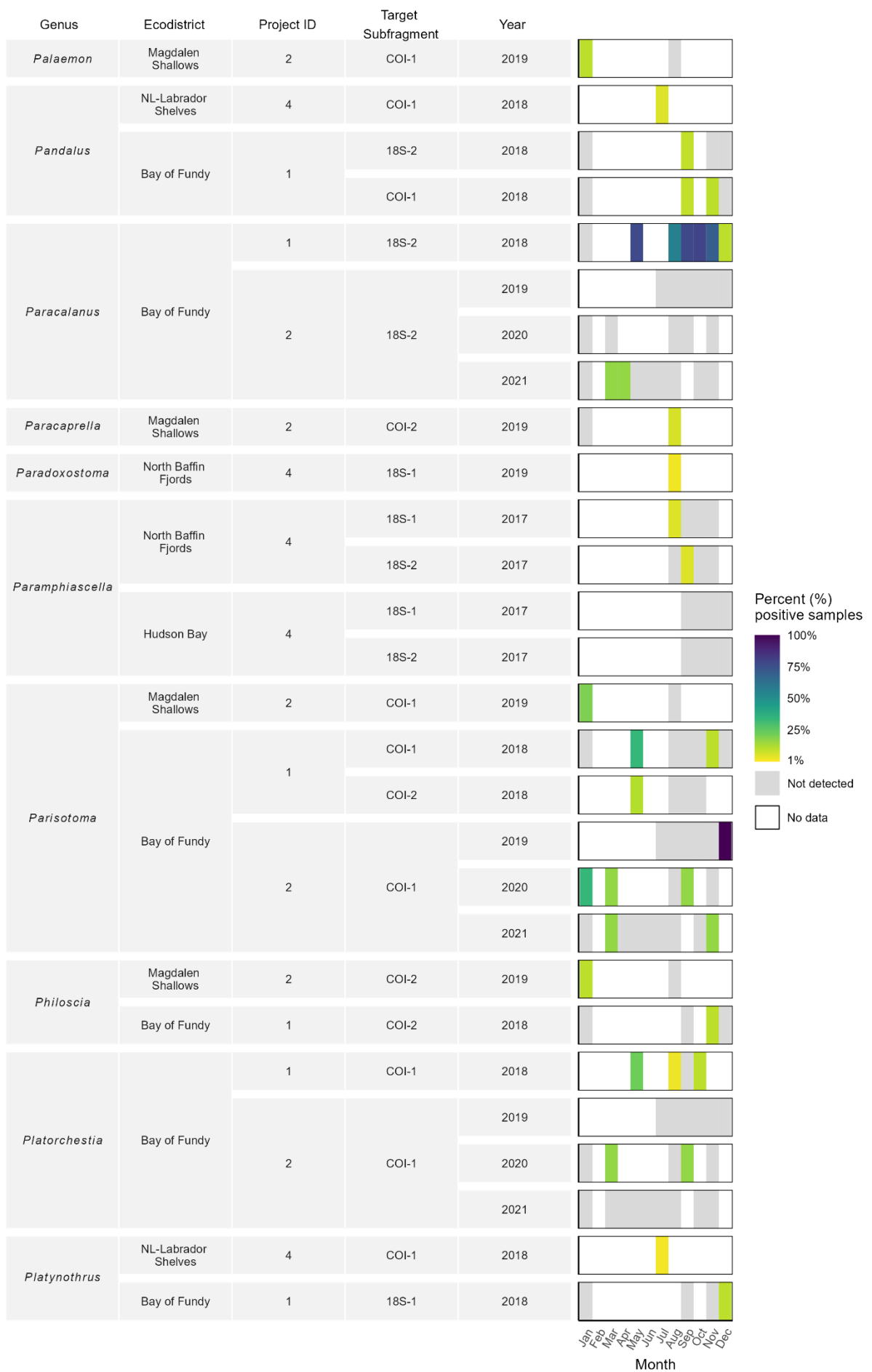


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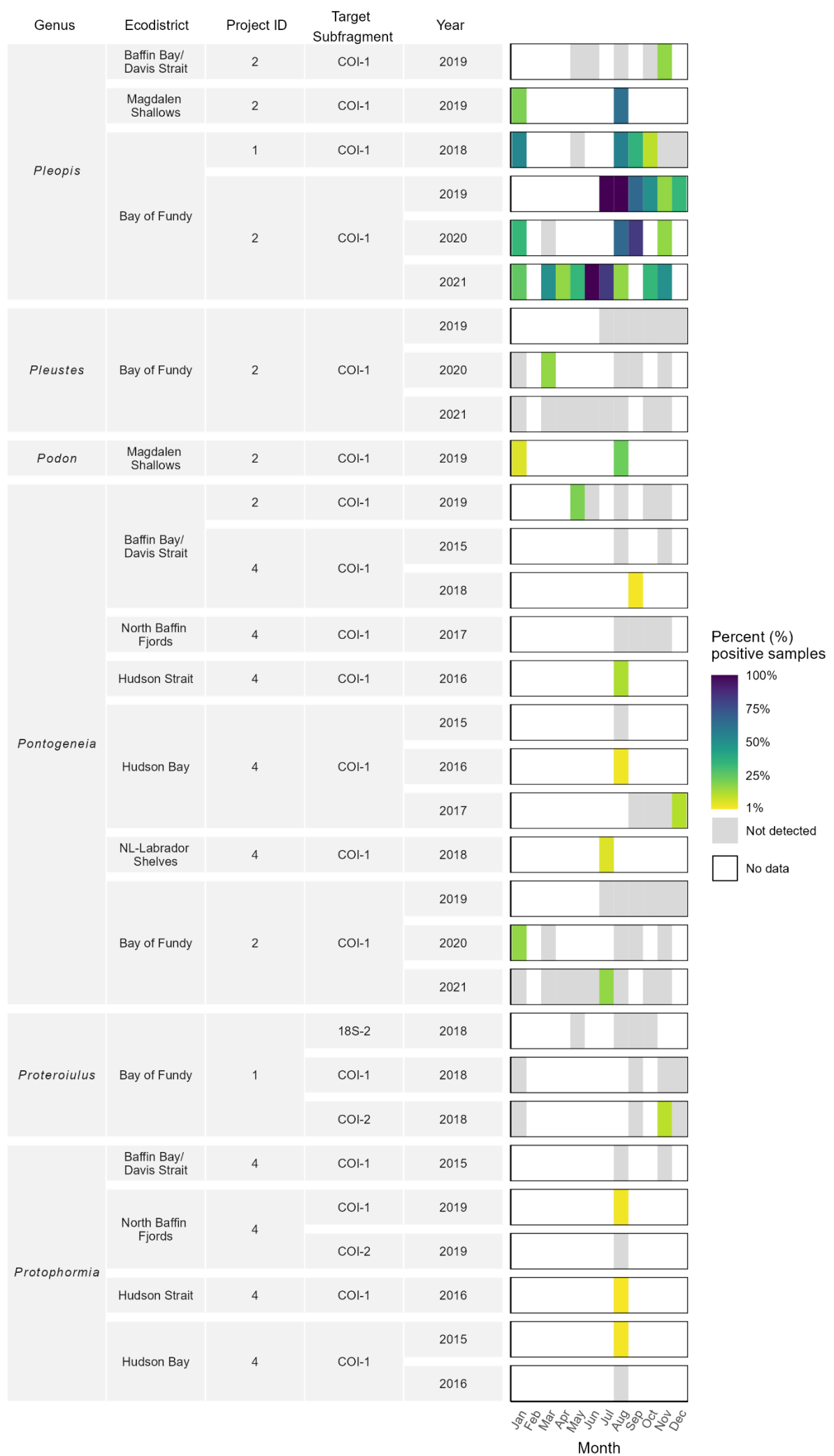


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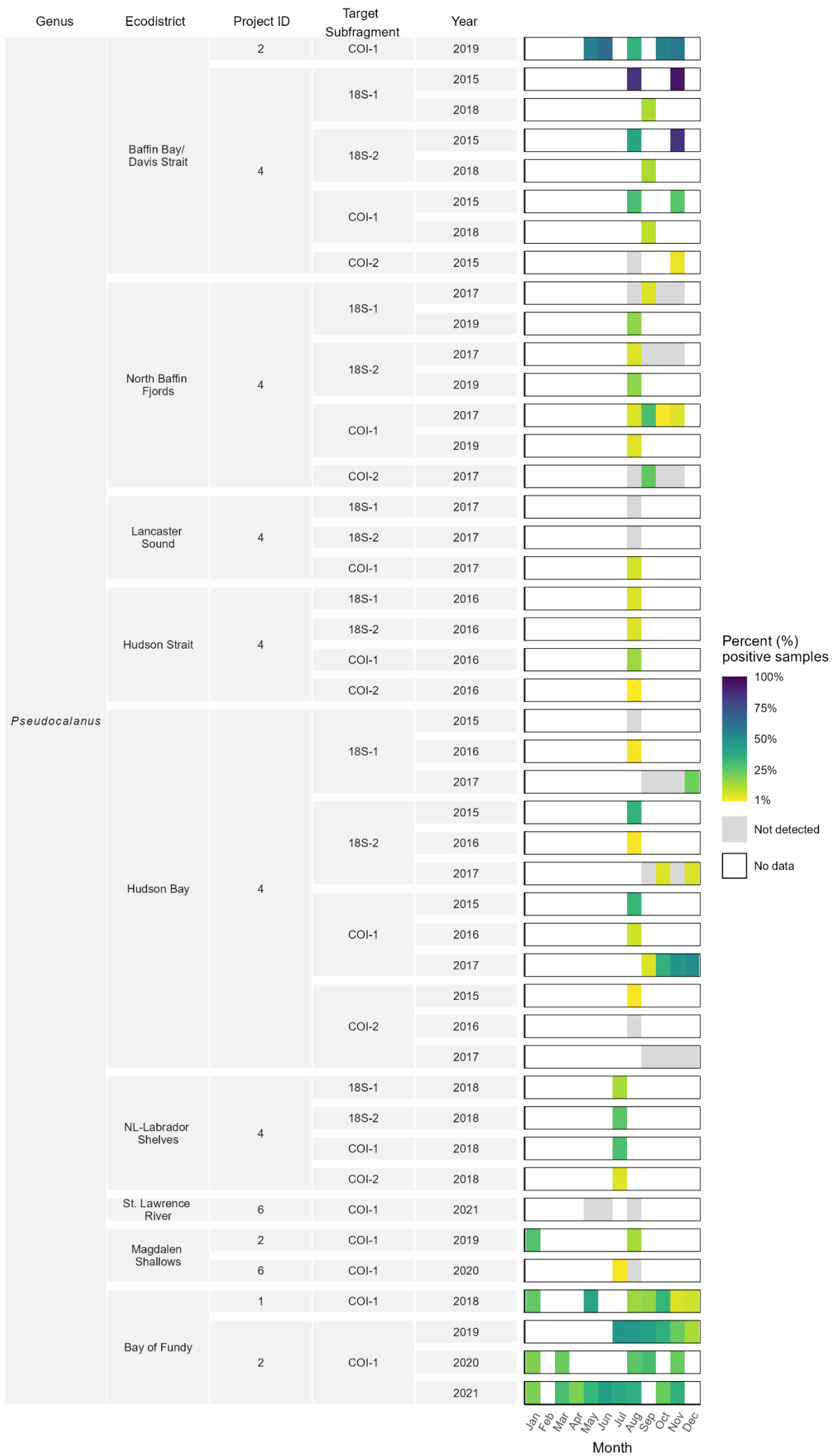


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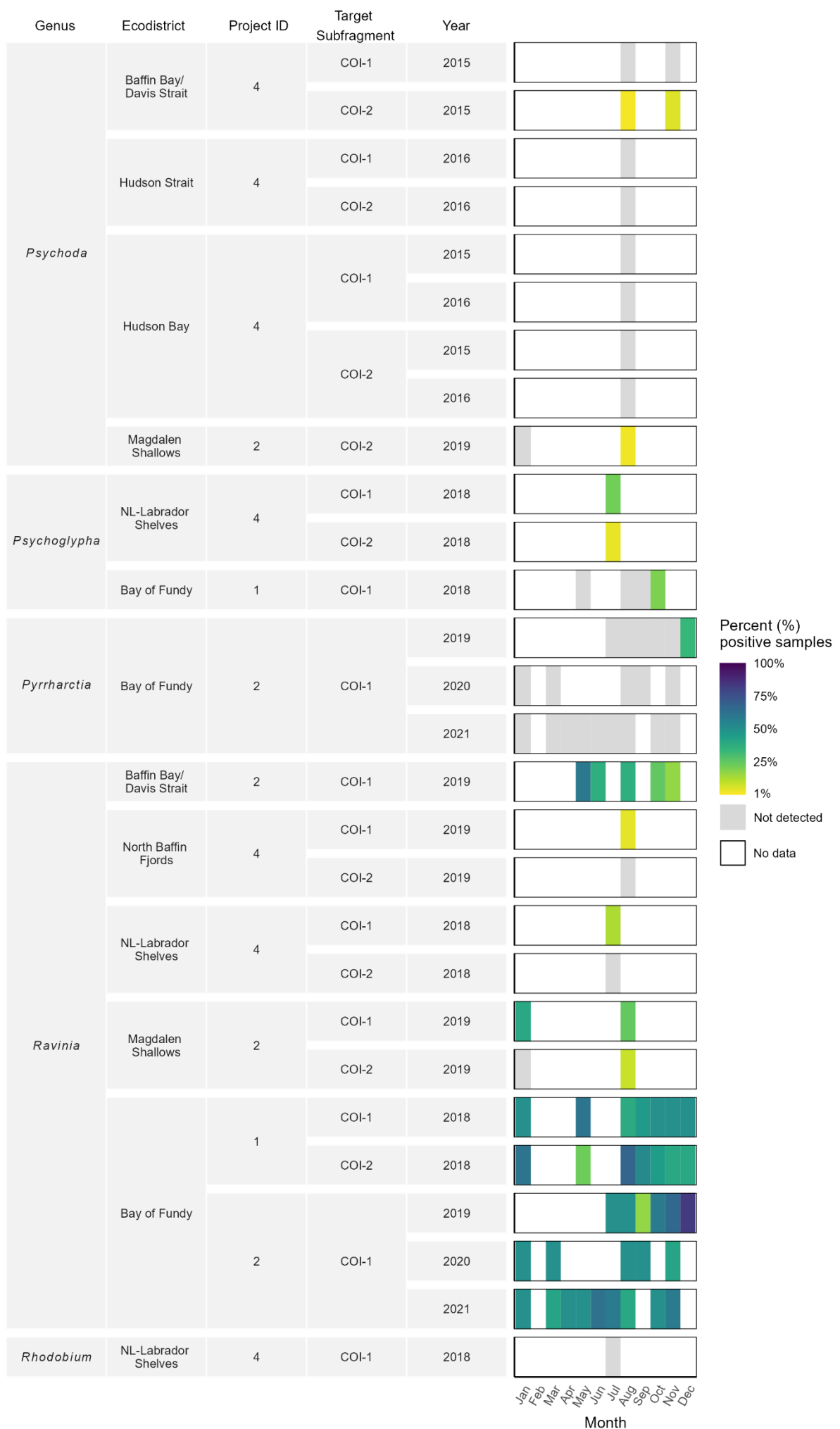


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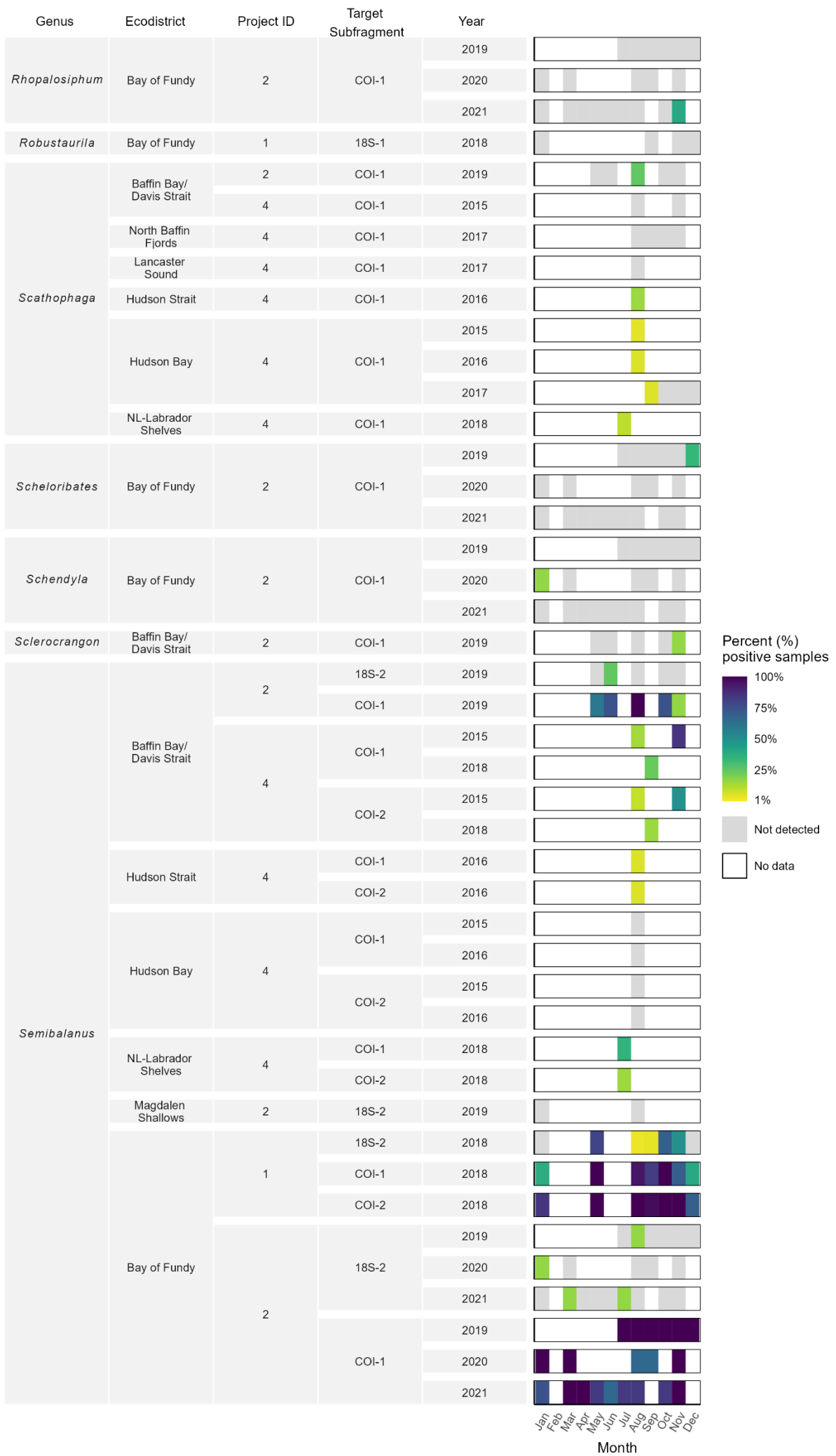


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Figure 13. (Continued)

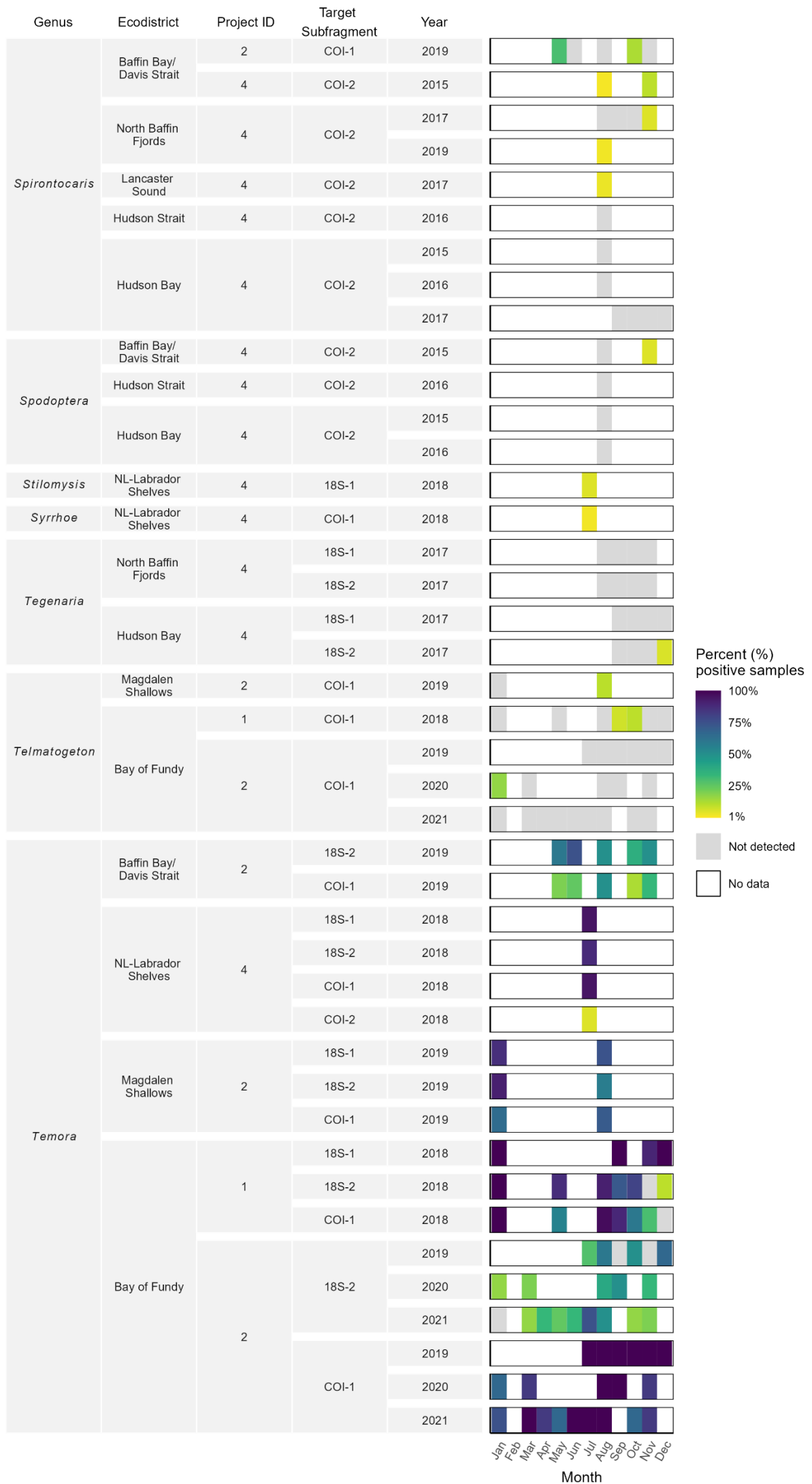


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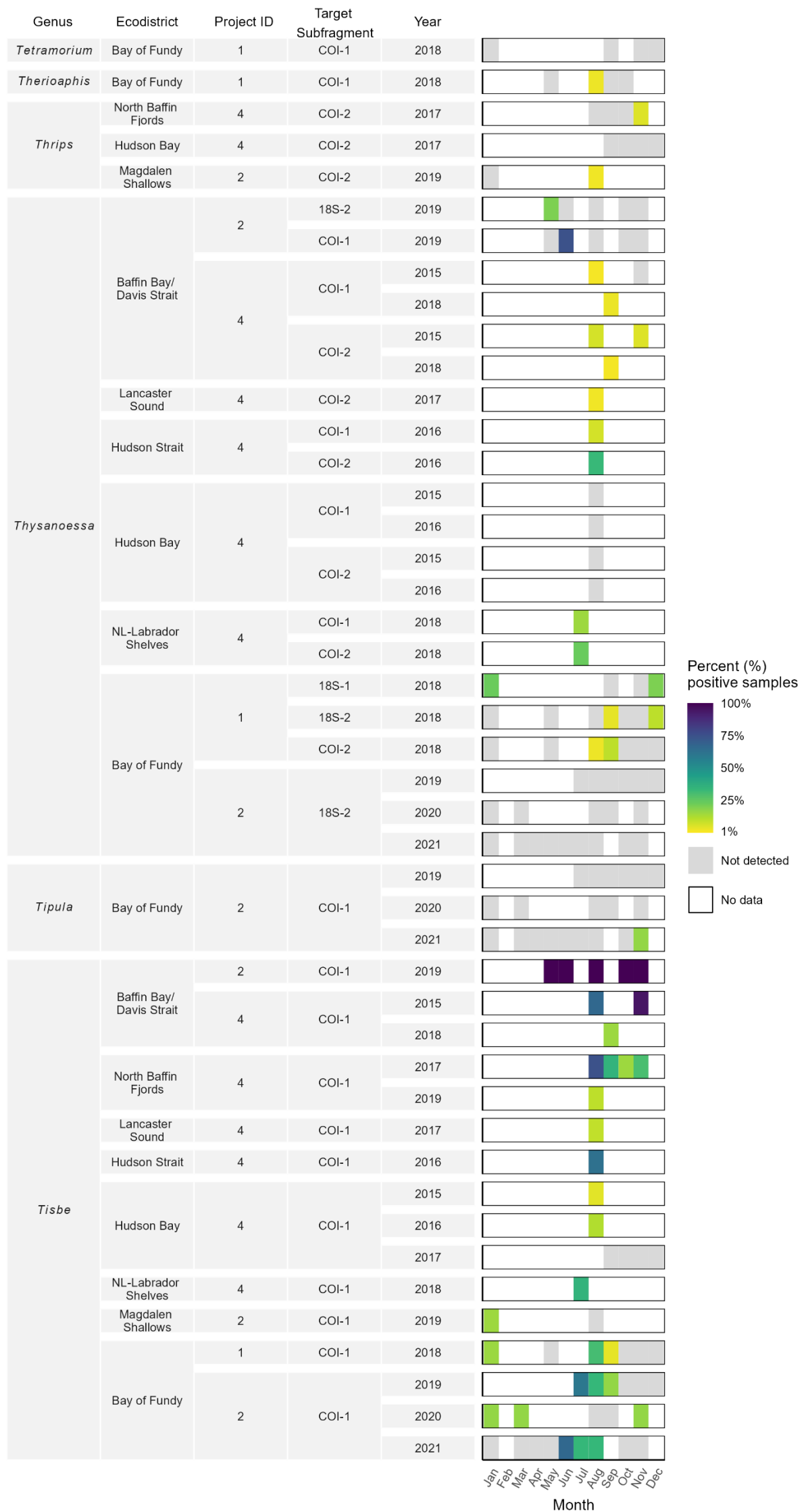


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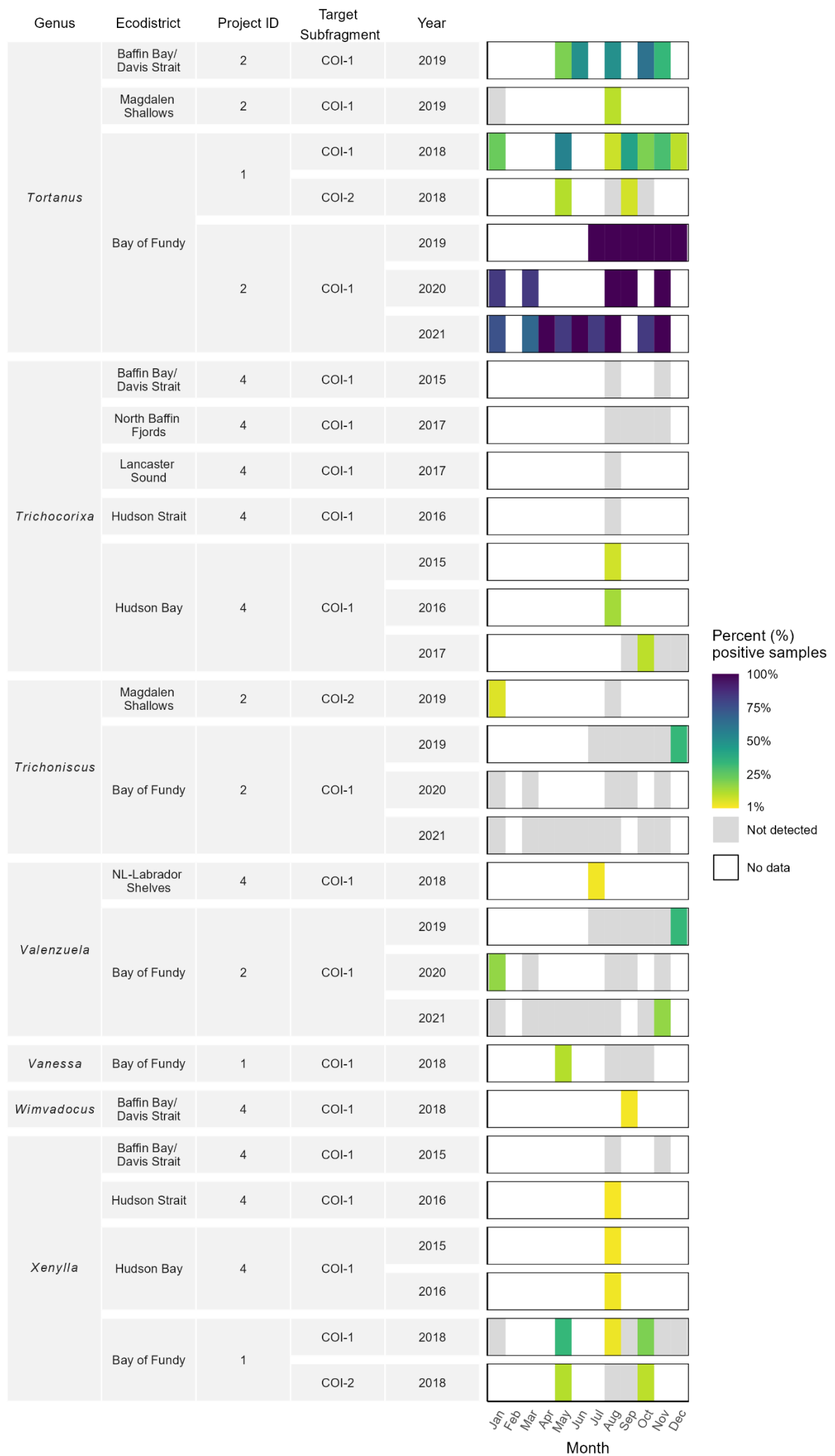


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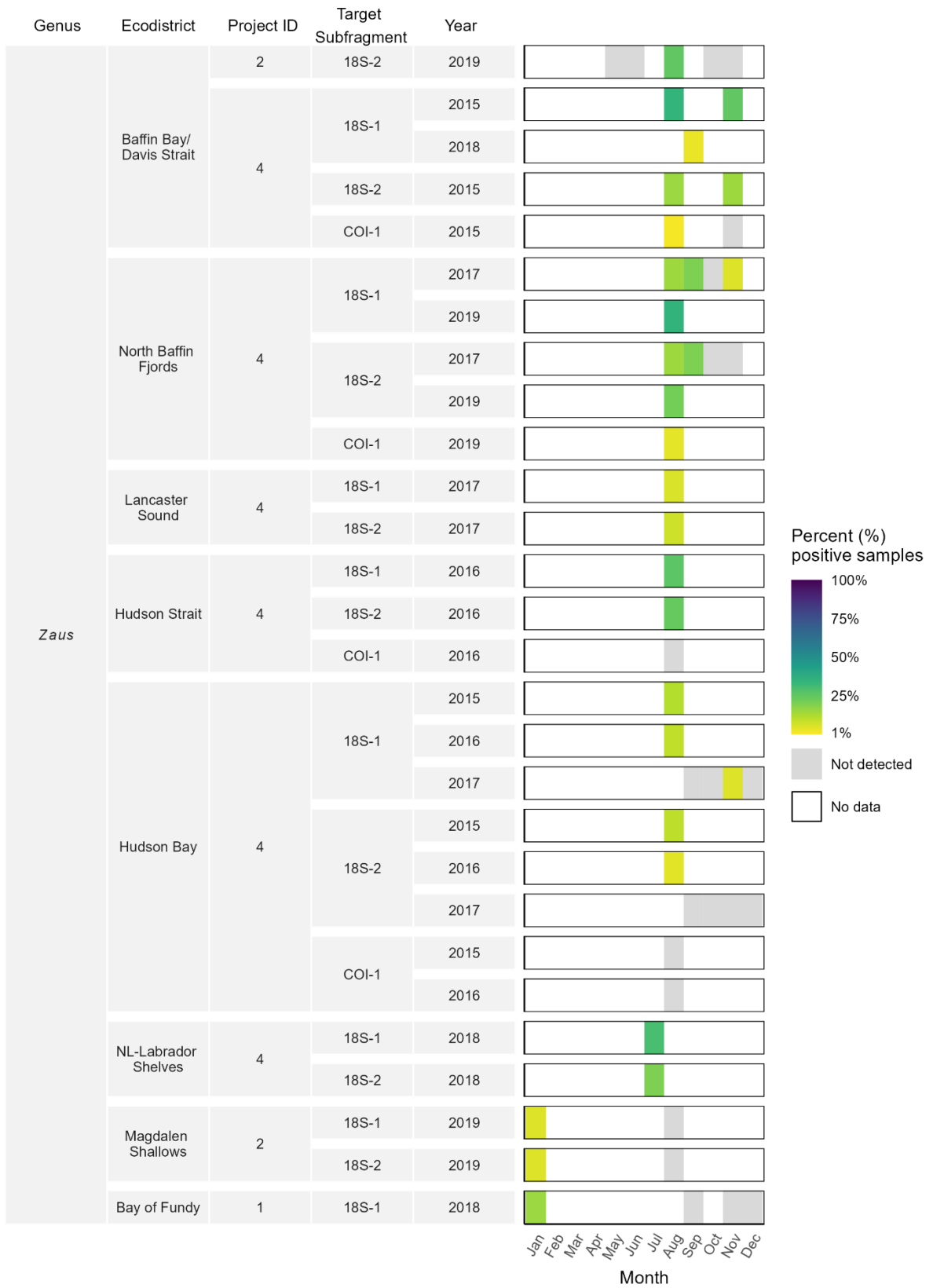


Figure 13. (Continued)

6.3 BRACHIOPODA



Figure 13. (Continued)

6.4 BRYOZOA



Figure 13. (Continued)

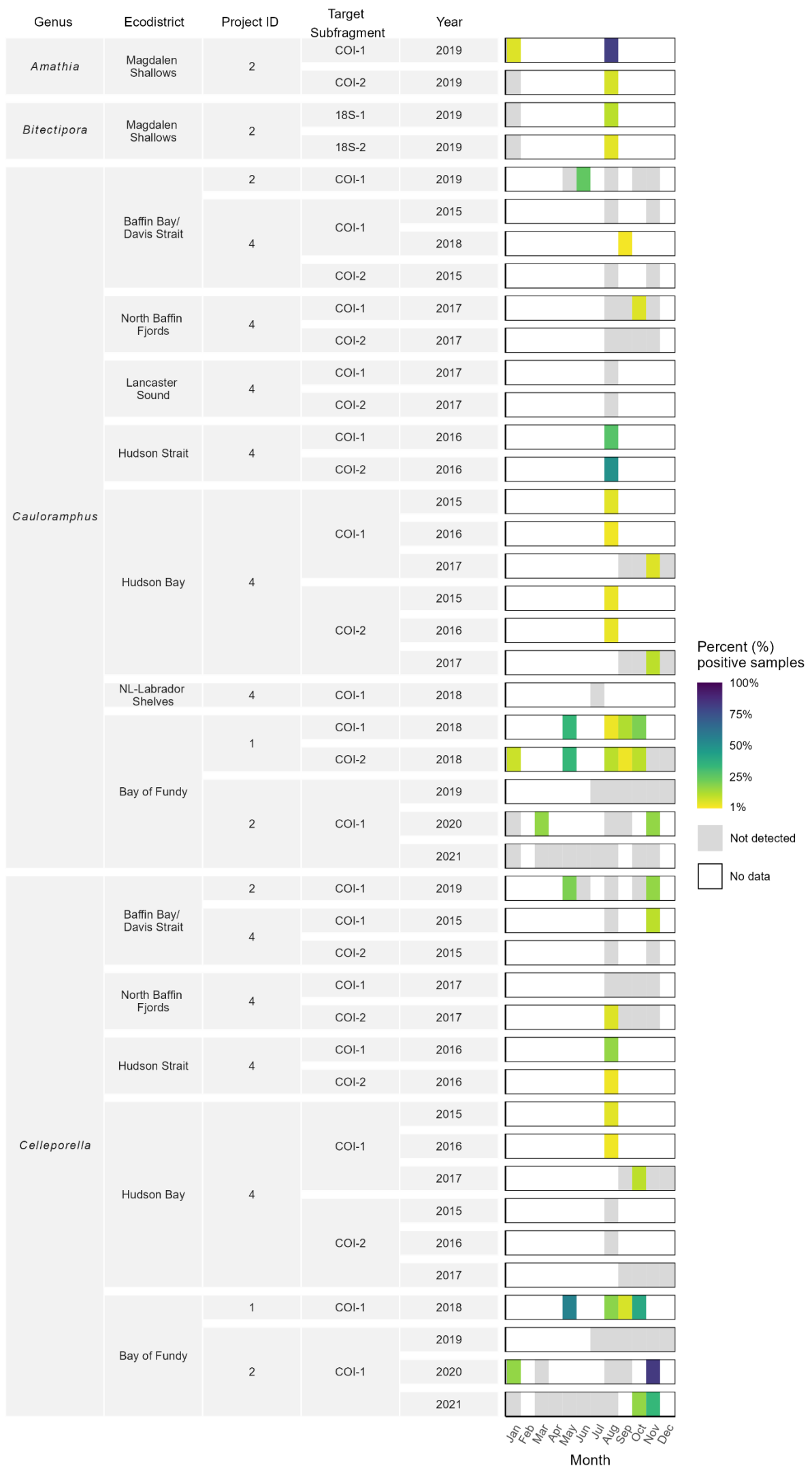


Figure 13. (Continued)

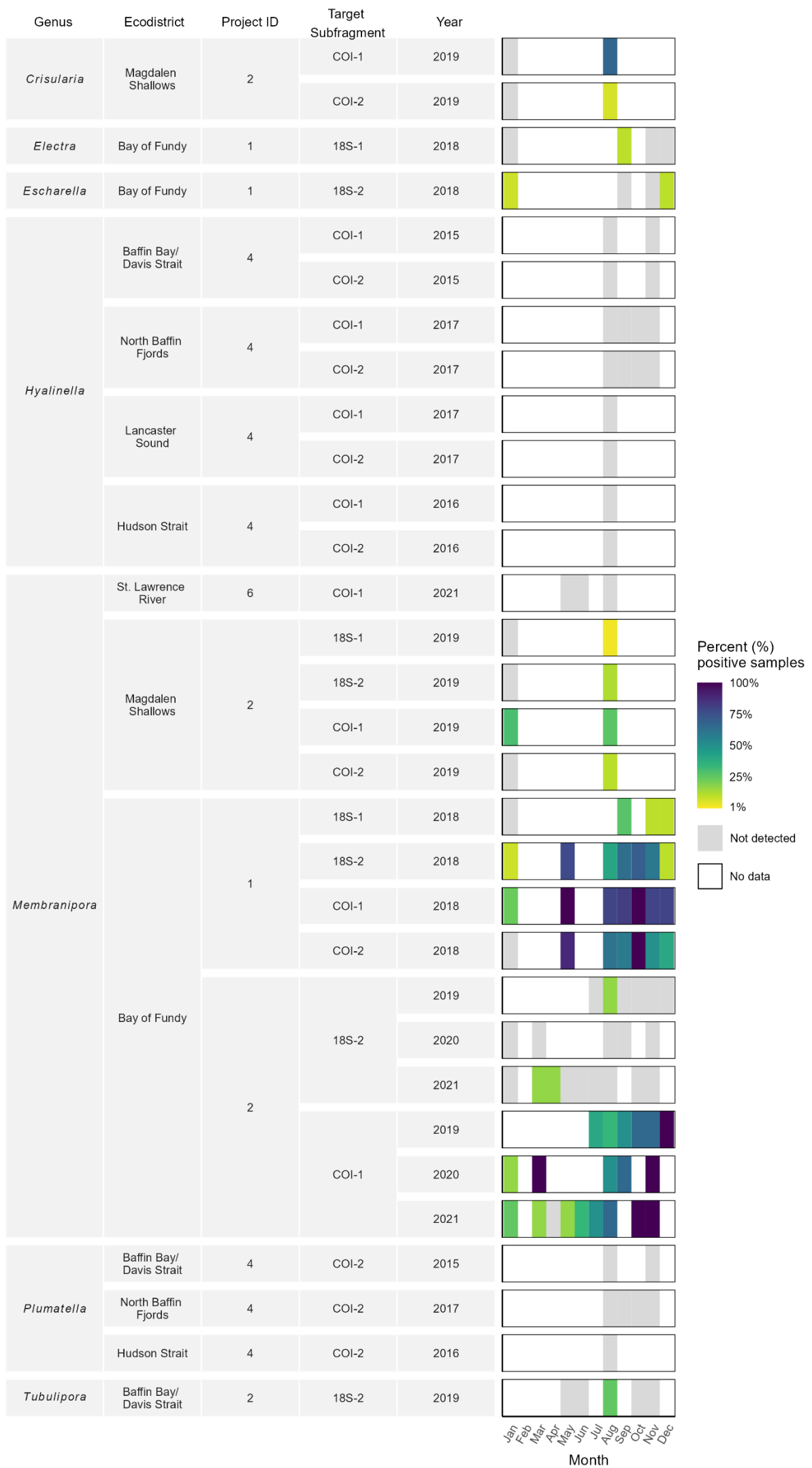


Figure 13. (Continued)

6.5 CHORDATA

6.5.1 Actinopterygii

Chondrostei

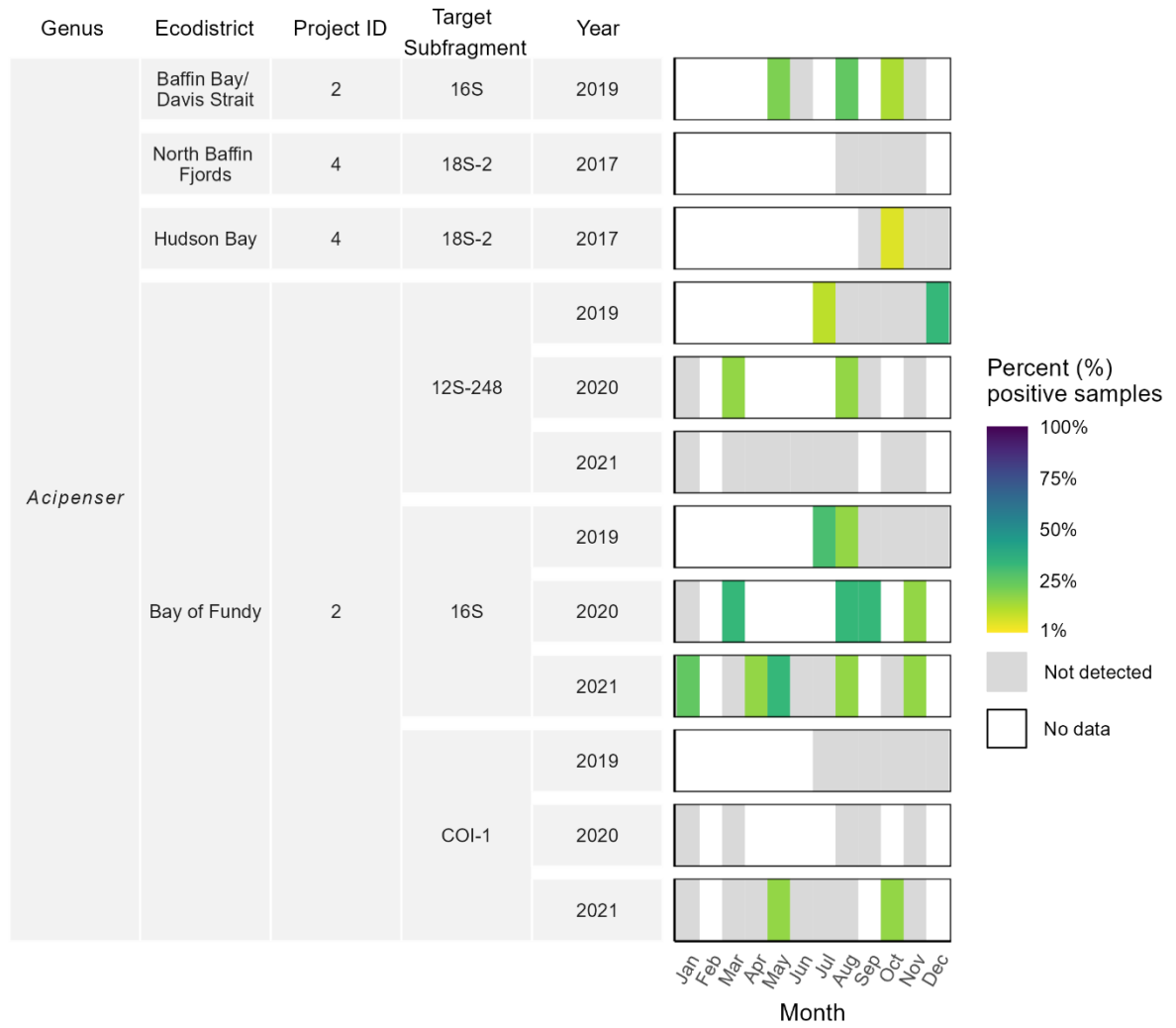


Figure 13. (Continued)

Holosteii



Figure 13. (Continued)

Teleostei

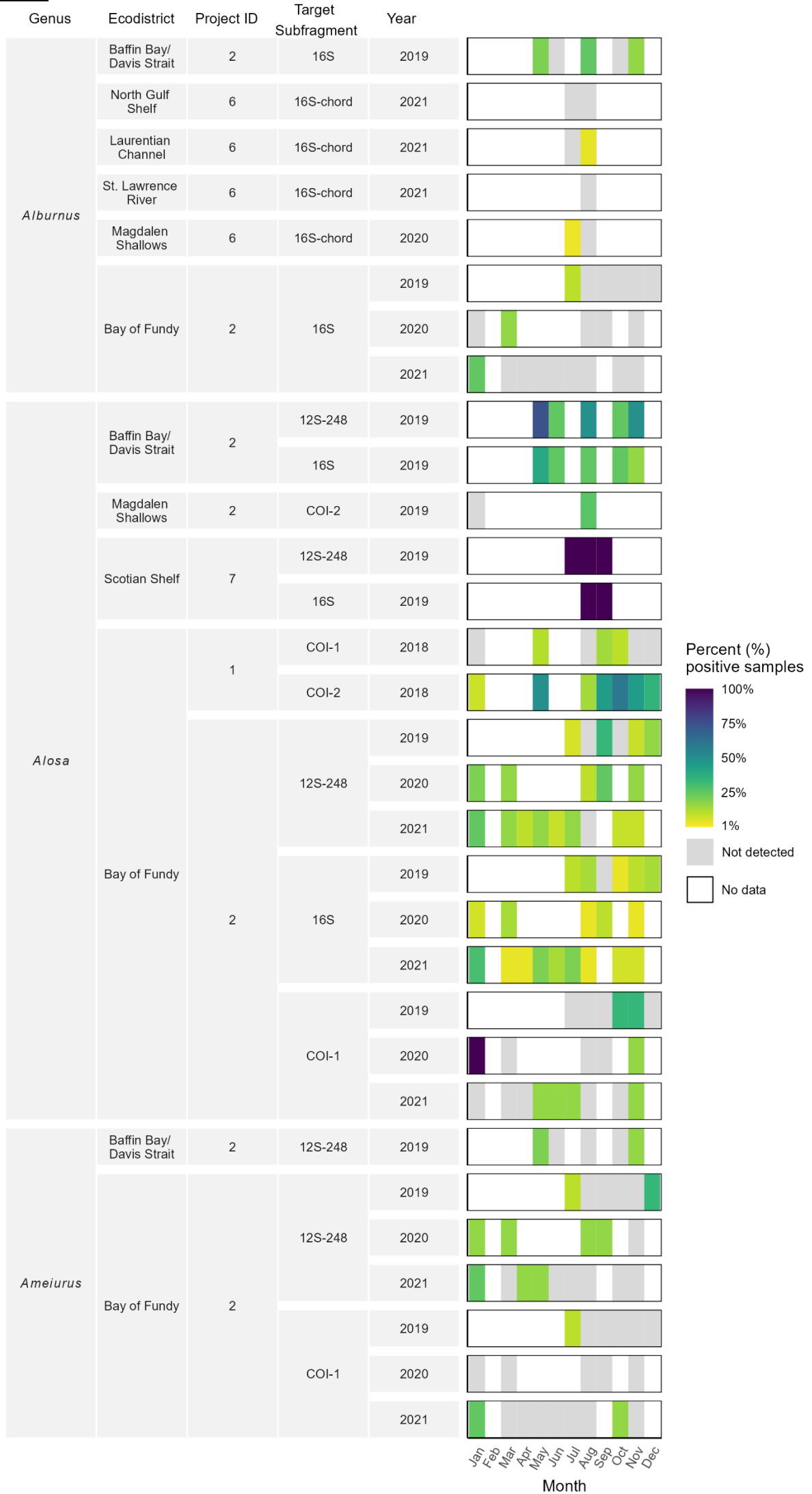


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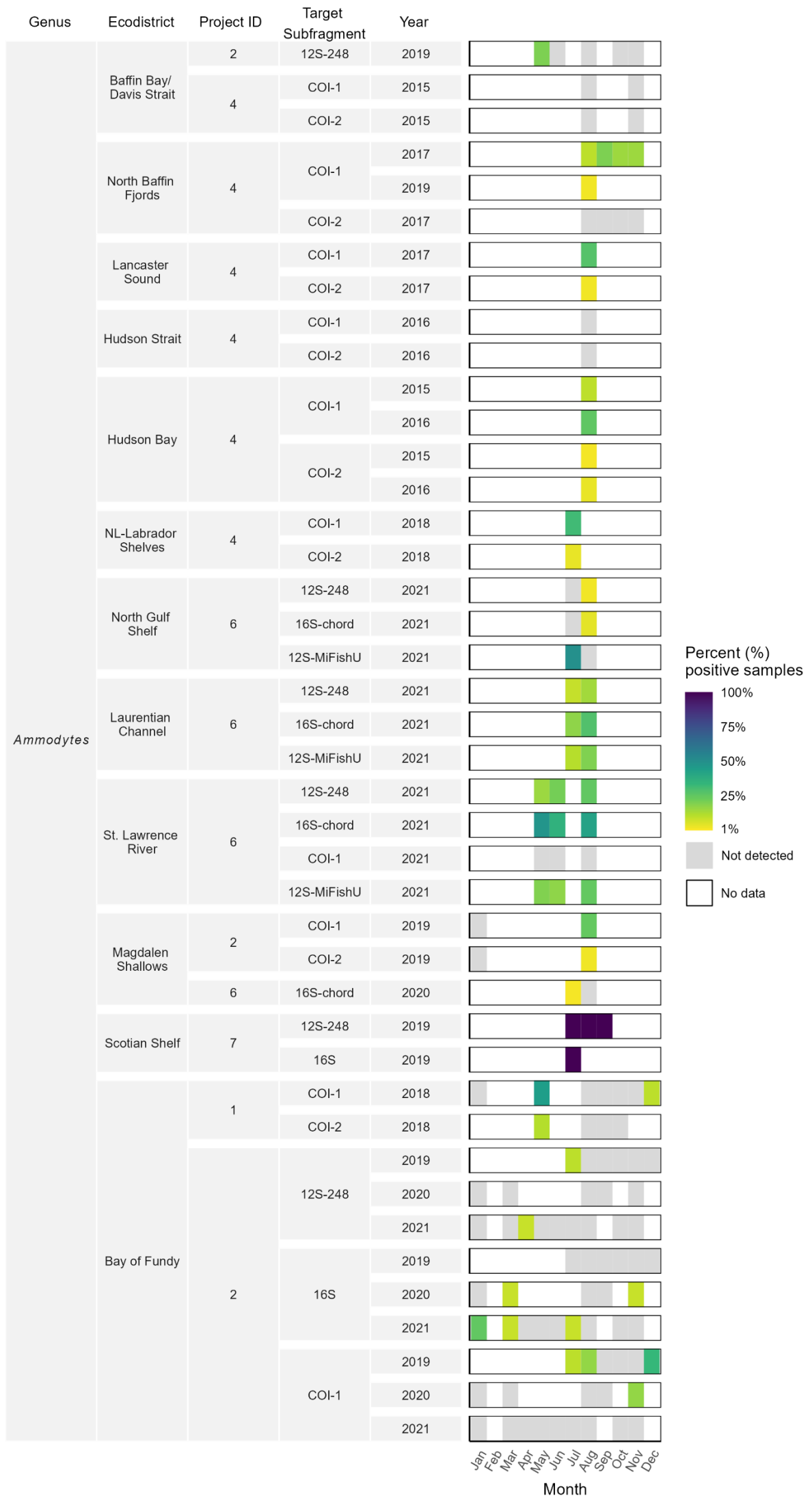


Figure 13. (Continued)

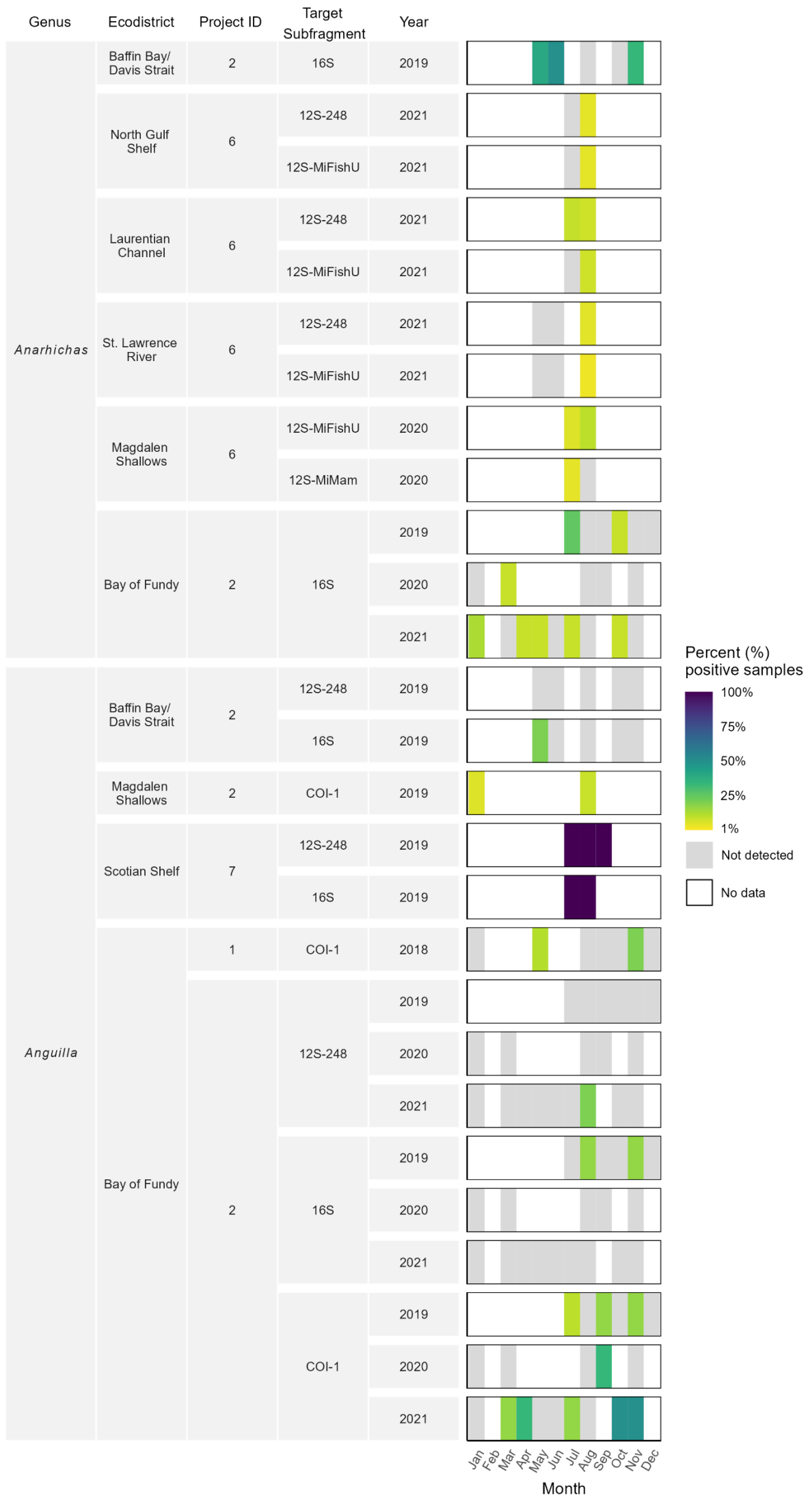


Figure 13. (Continued)

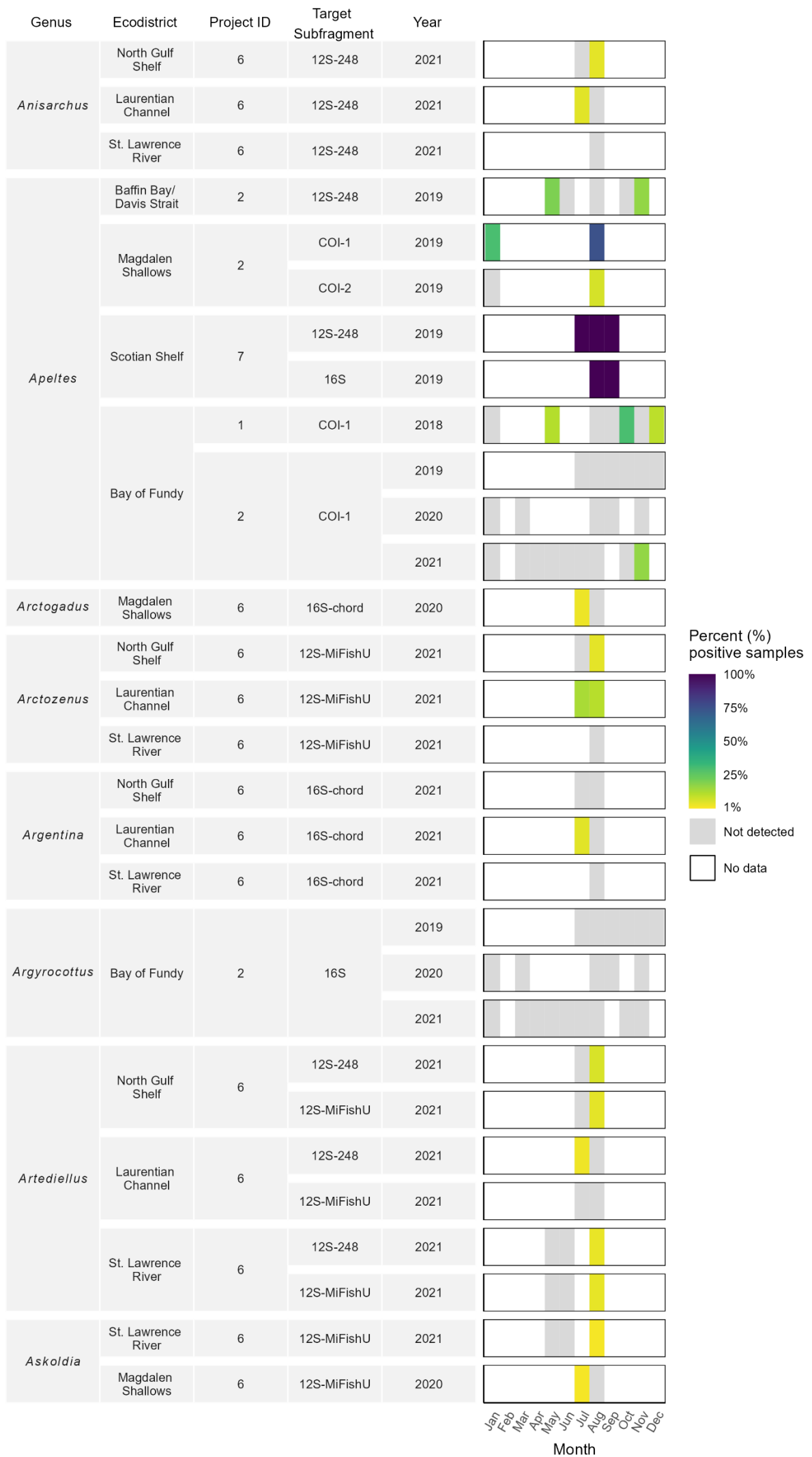


Figure 13. (Continued)



Figure 13. (Continued)

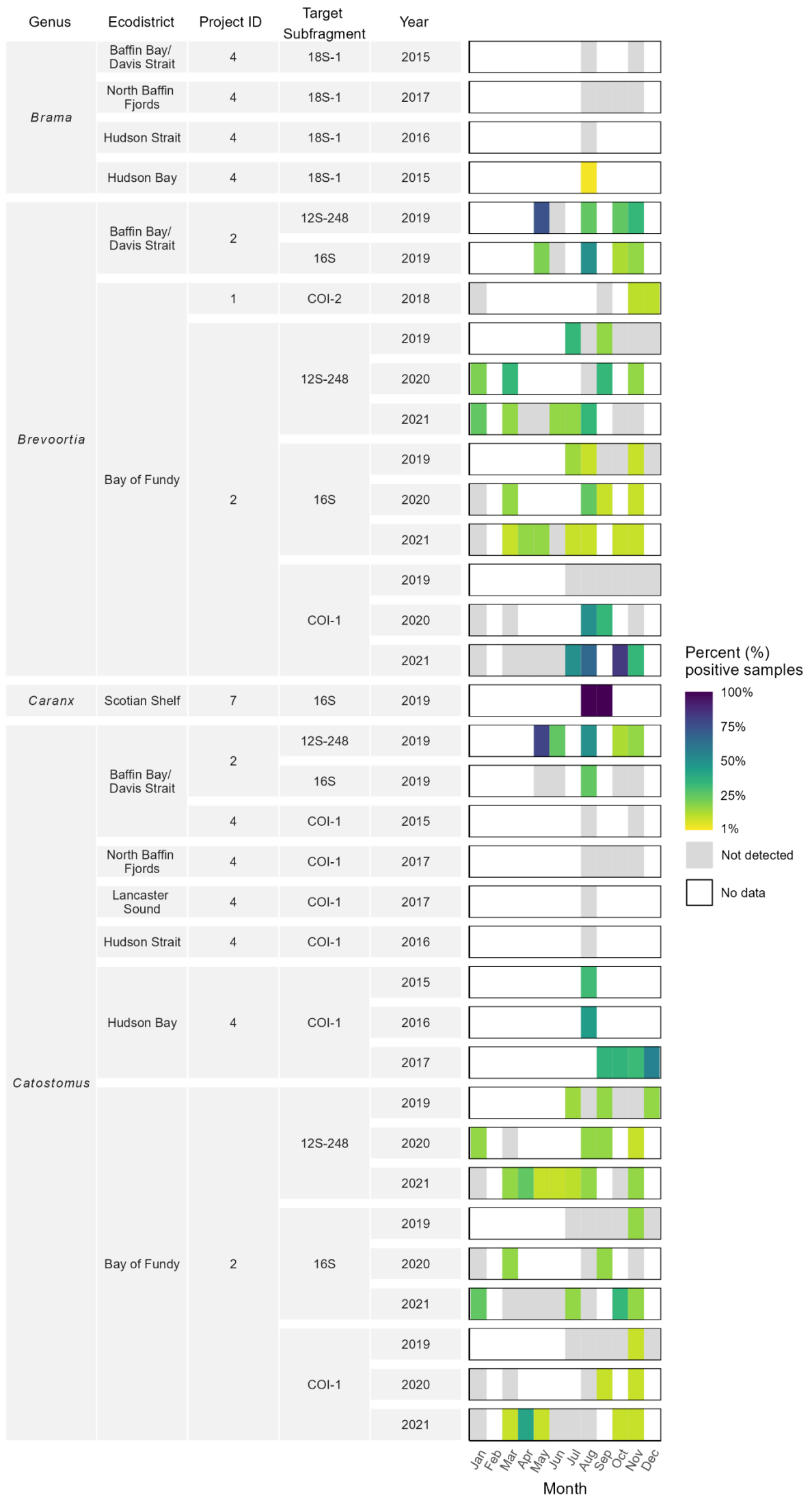


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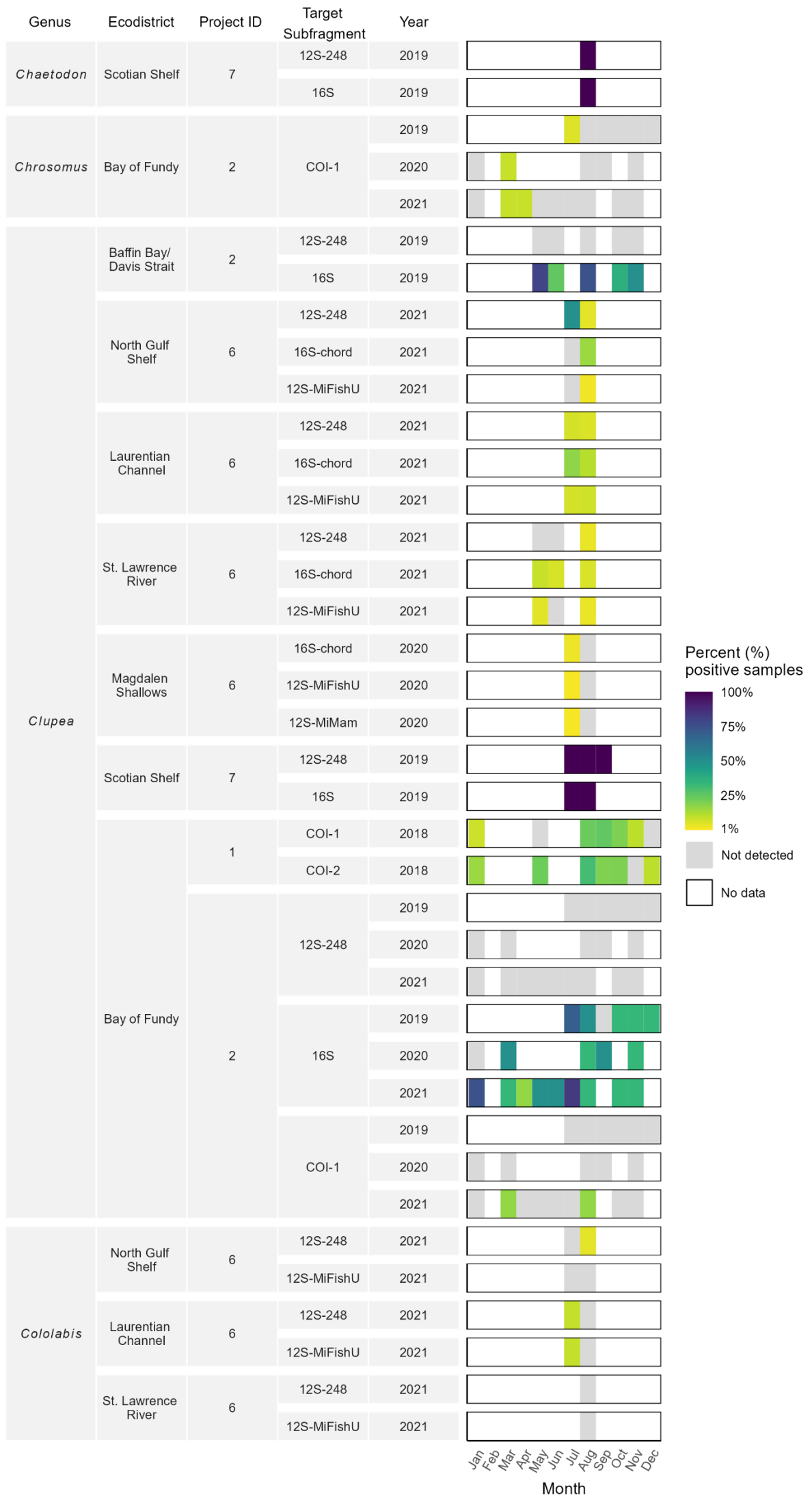


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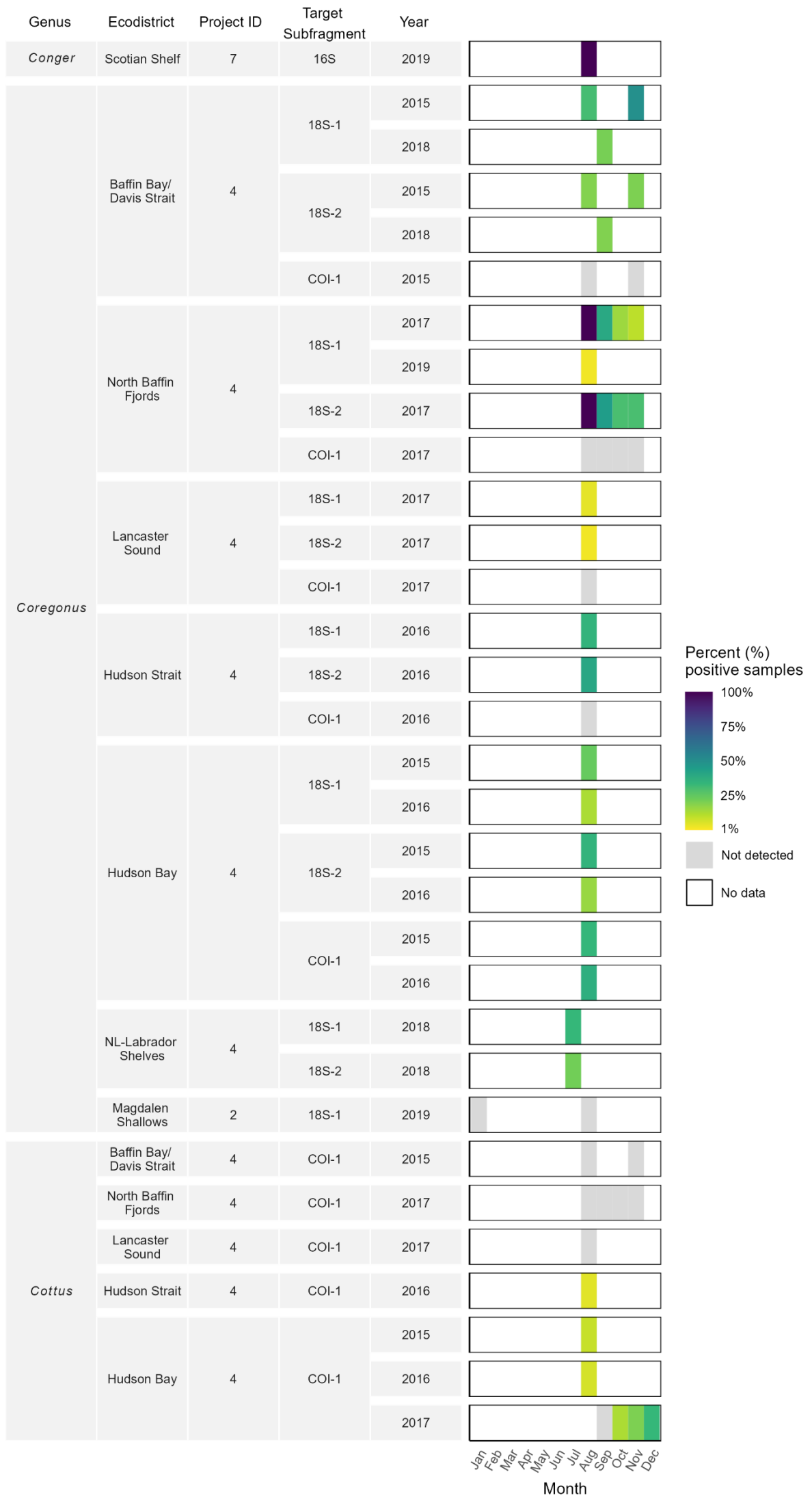


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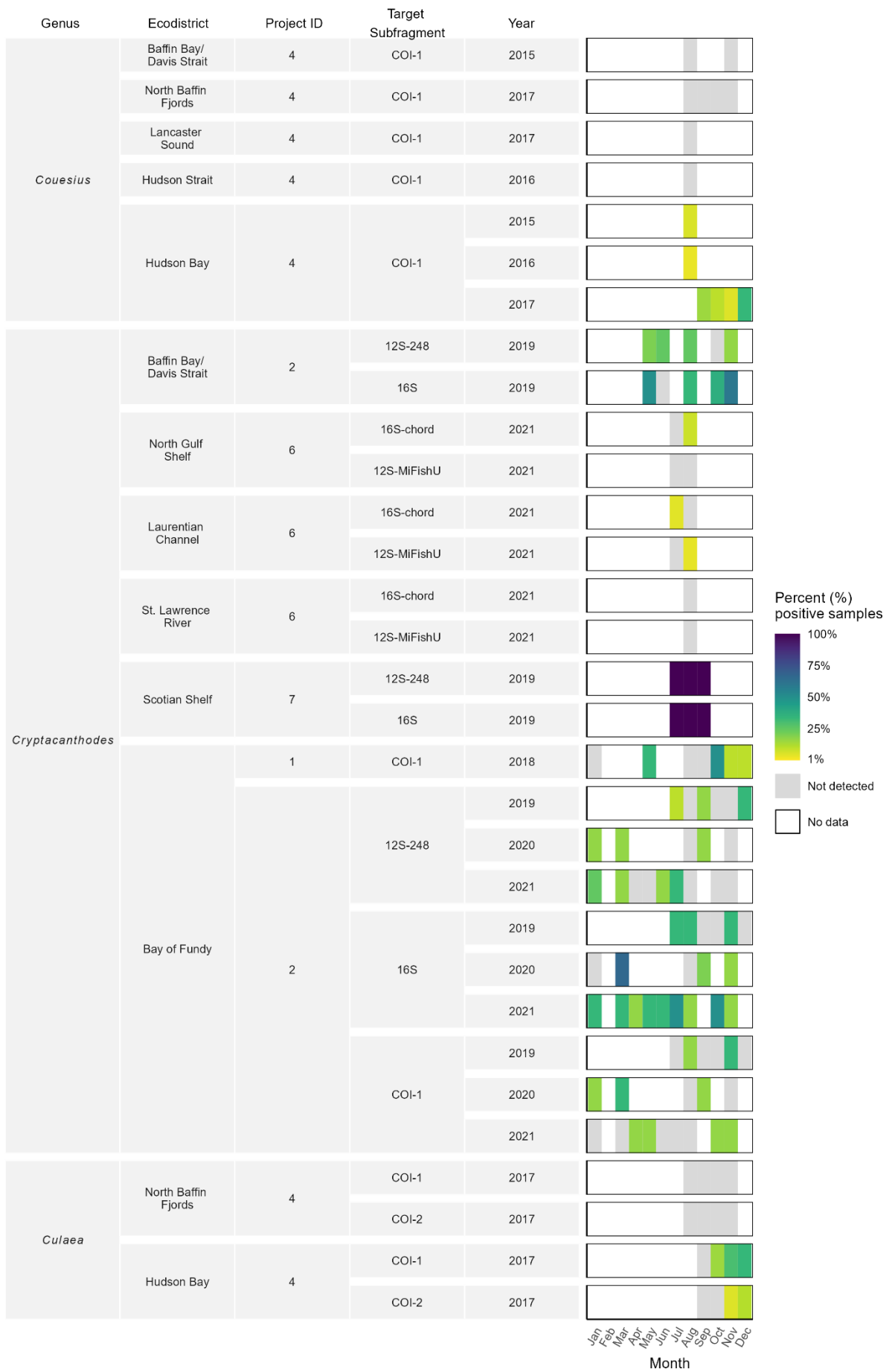


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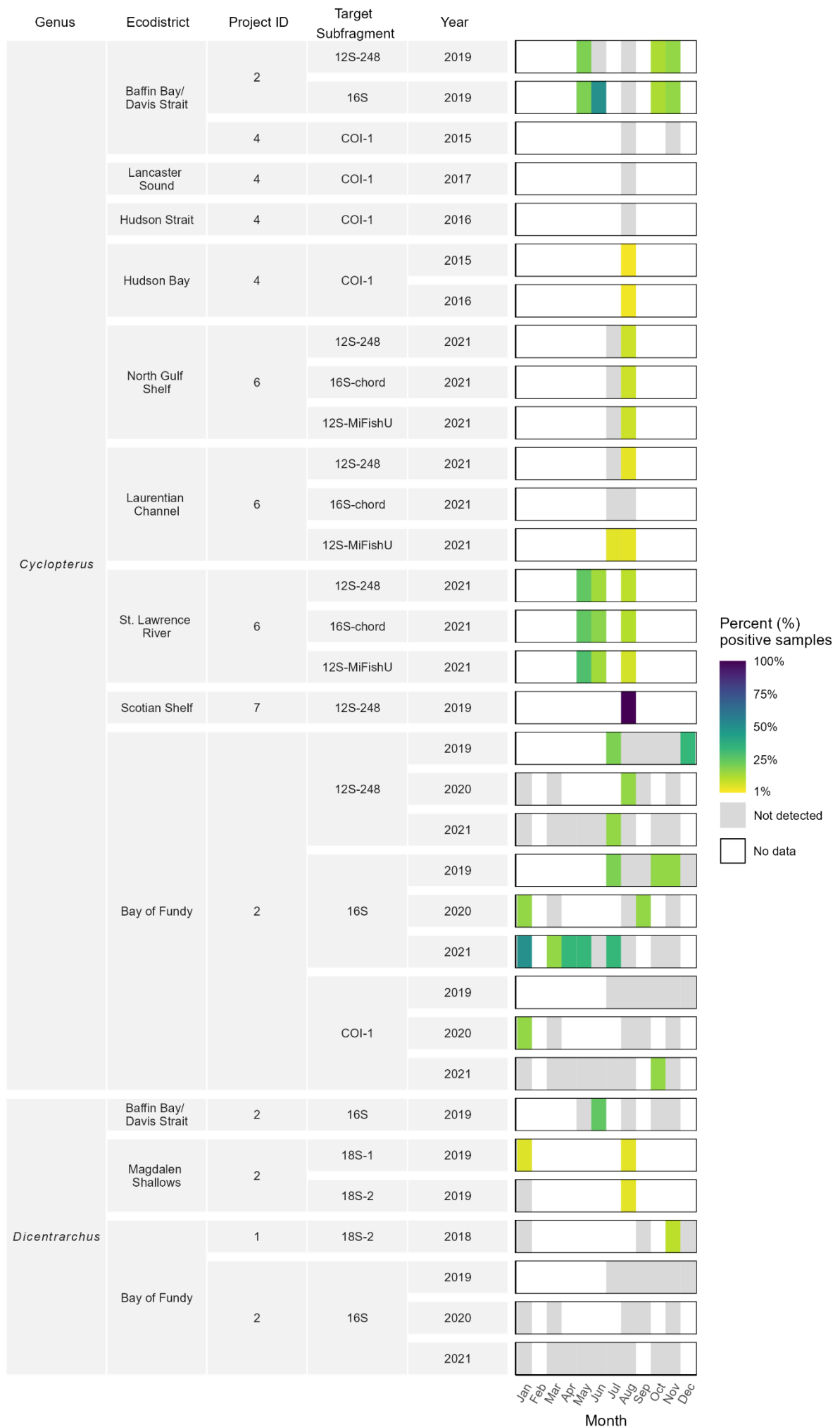


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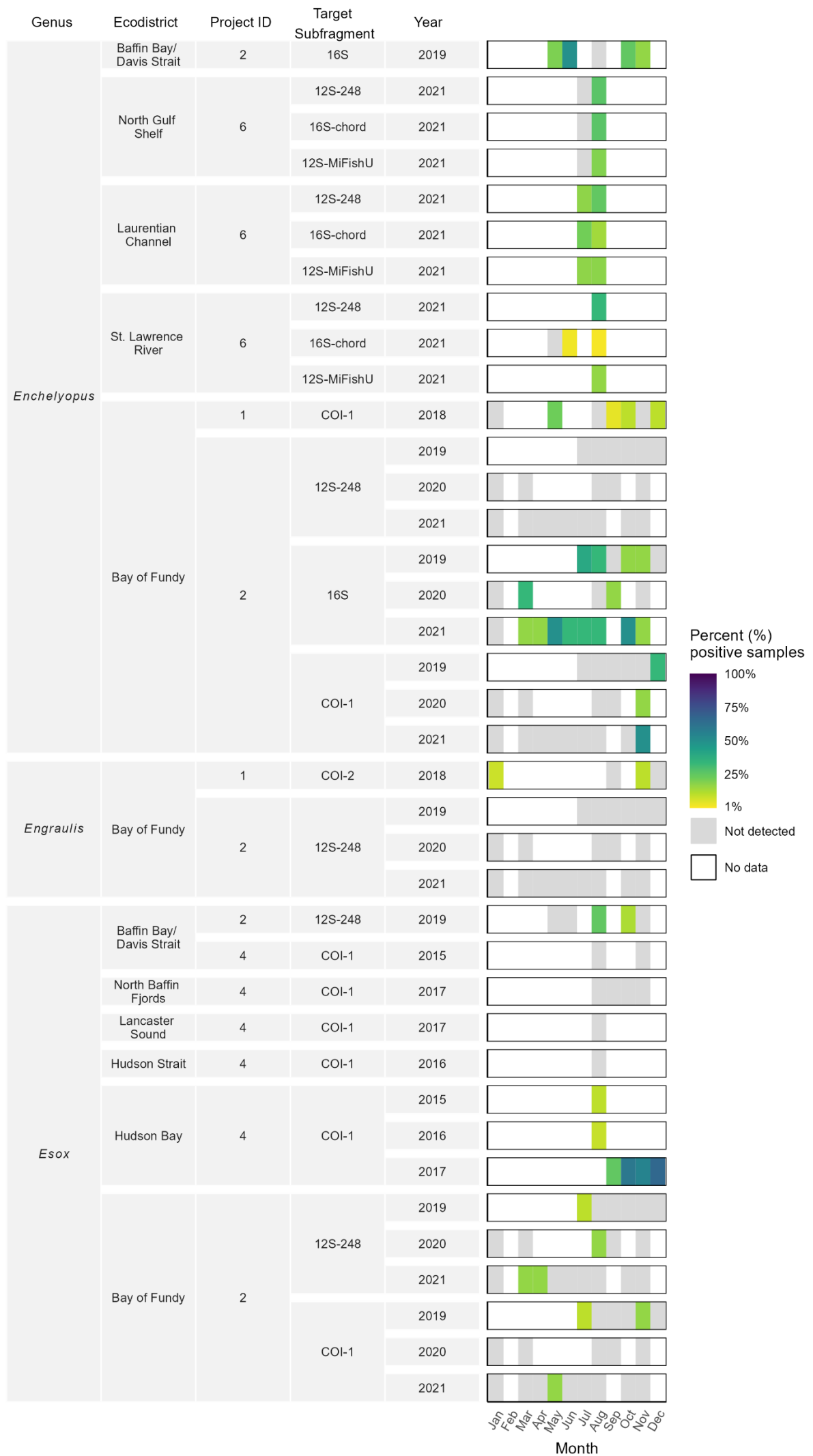


Figure 13. (Continued)



Figure 13. (Continued)

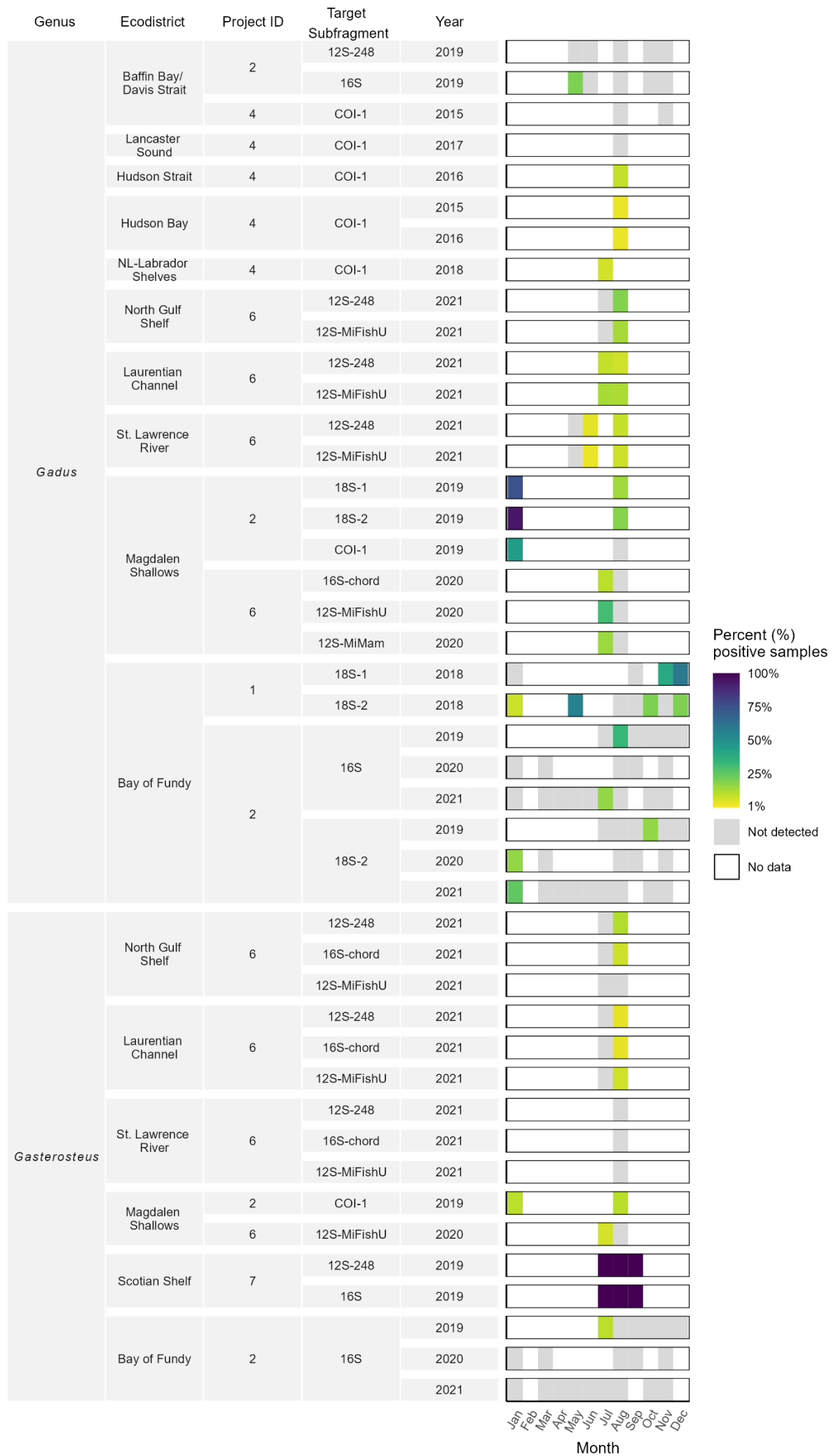


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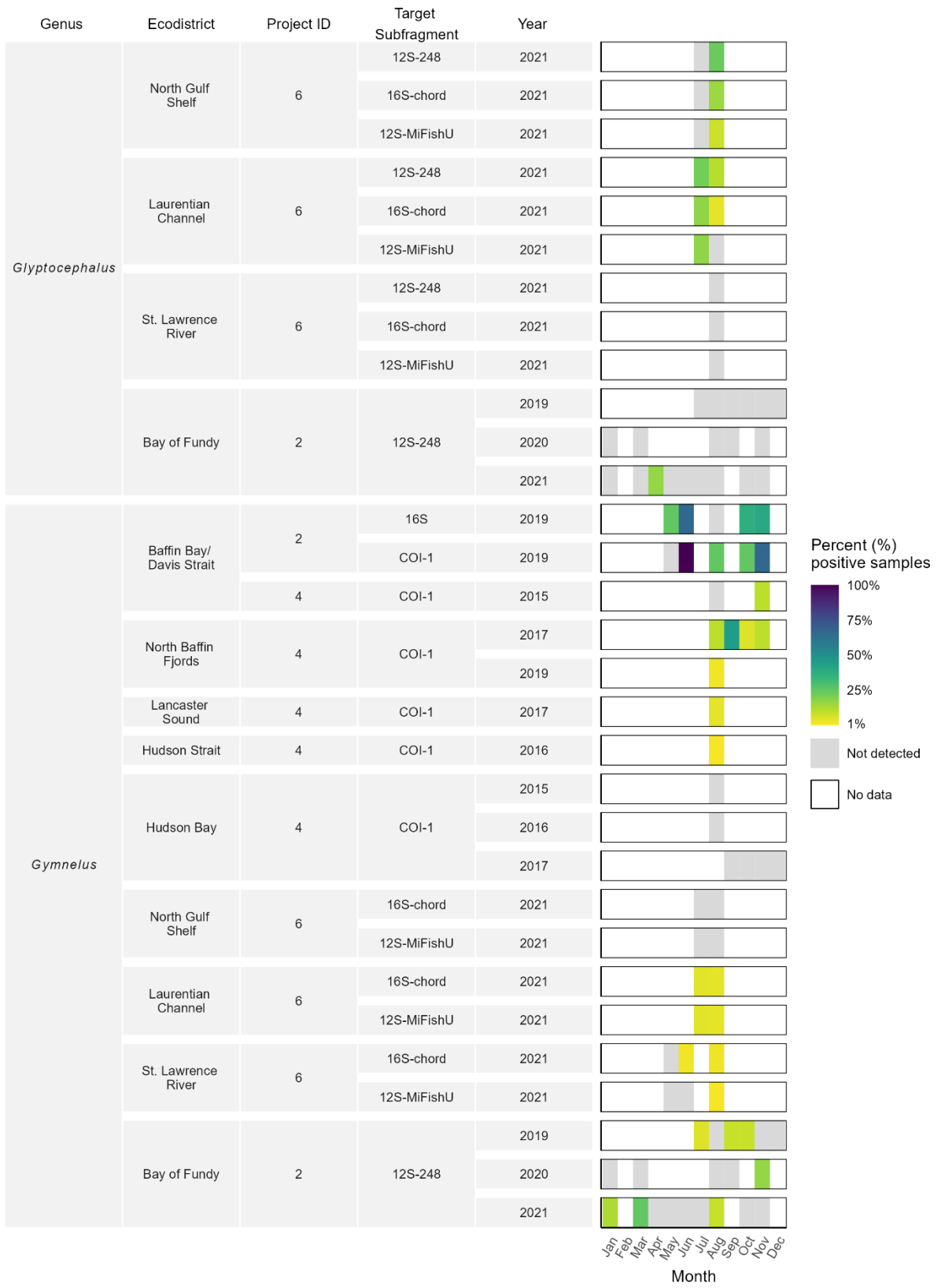


Figure 13. (Continued)



Figure 13. (Continued)

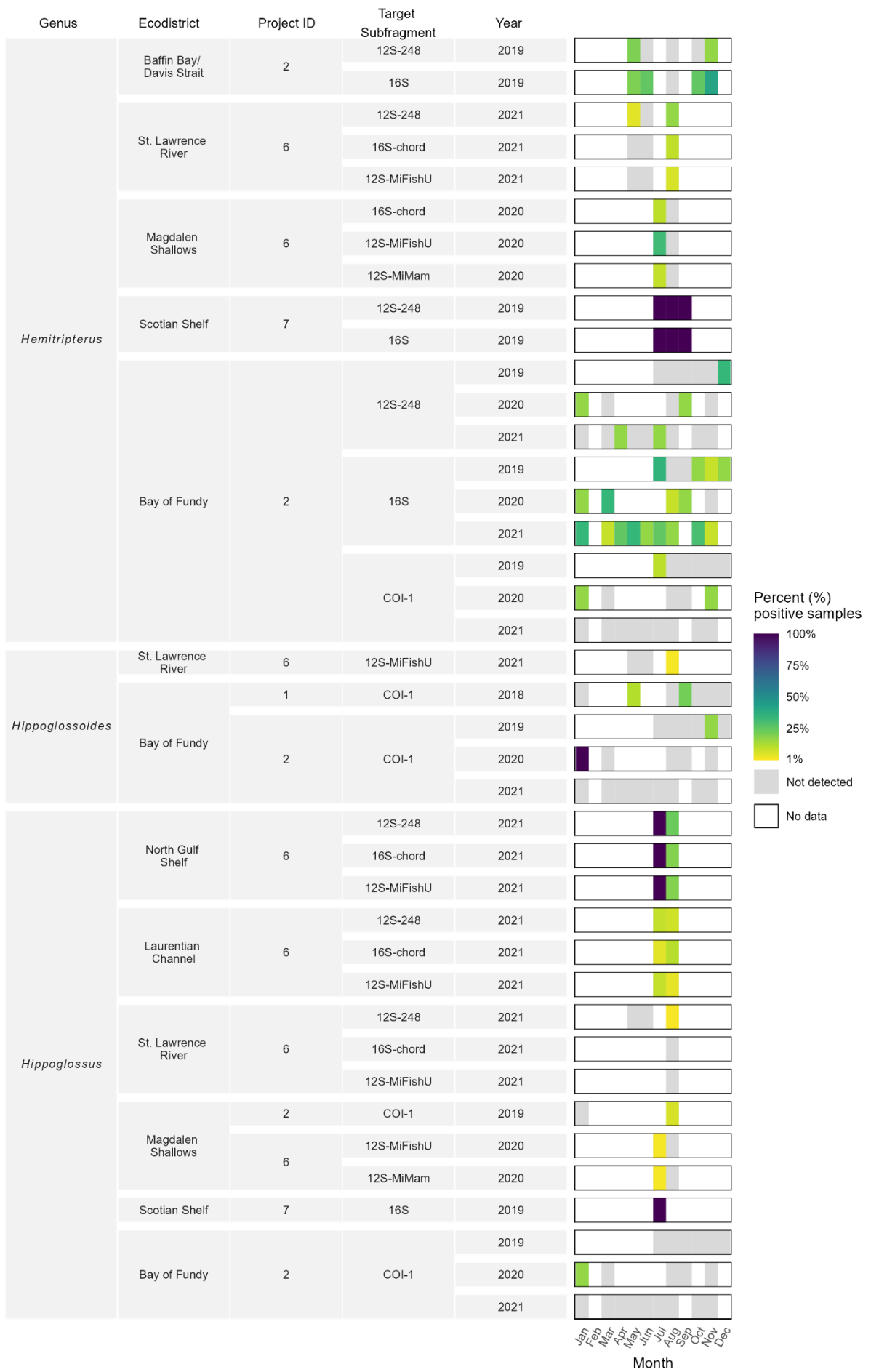


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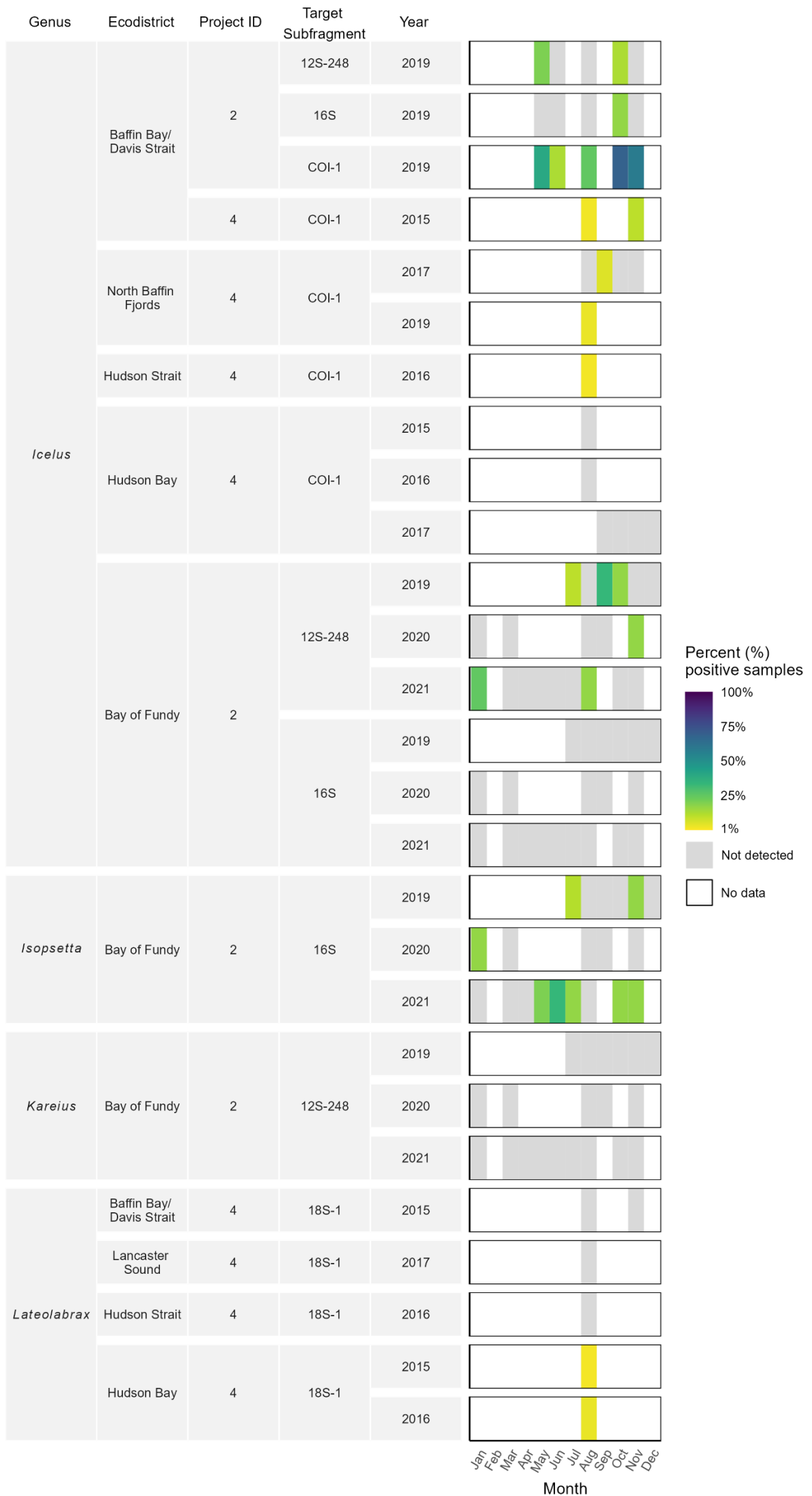


Figure 13. (Continued)



Figure 13. (Continued)

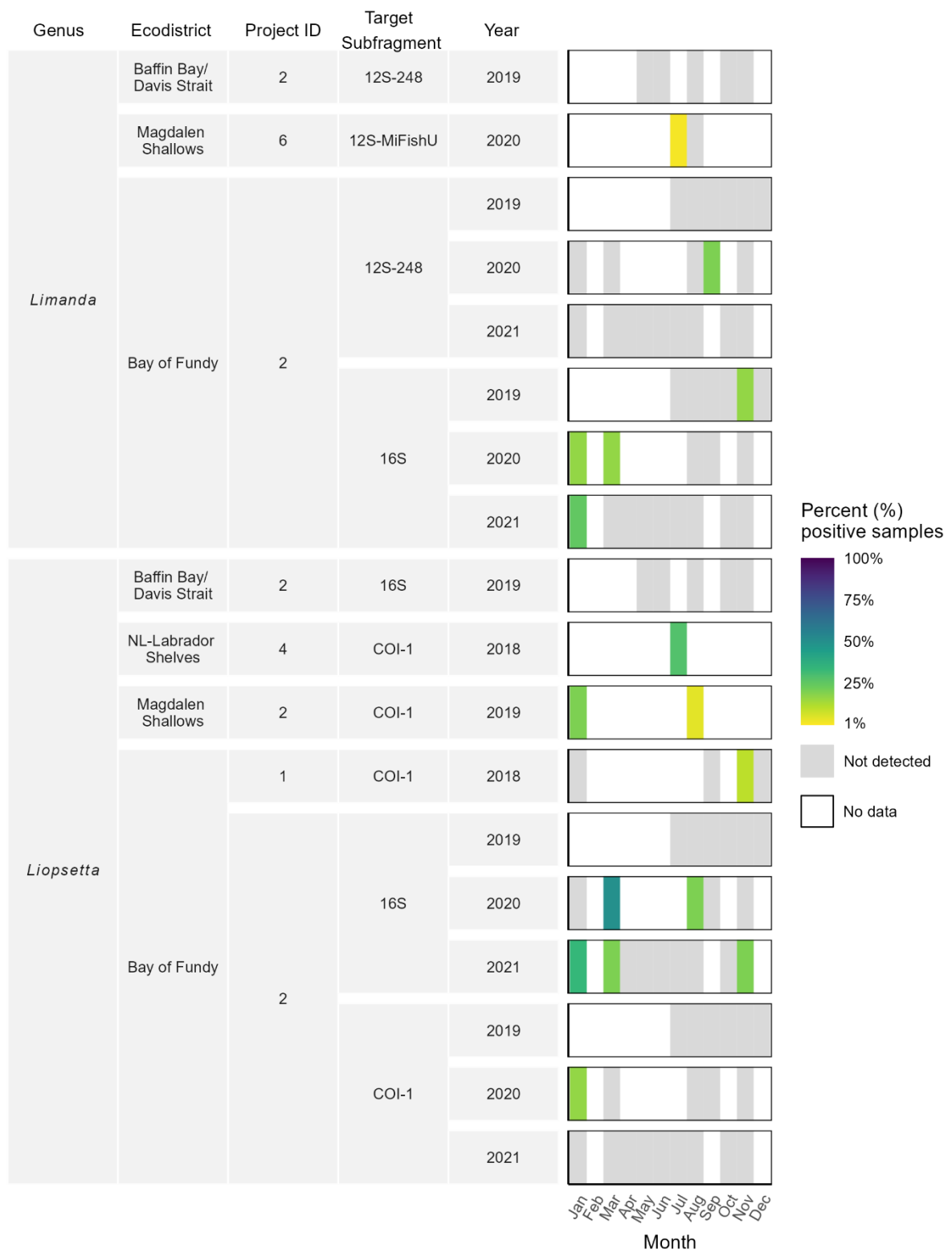


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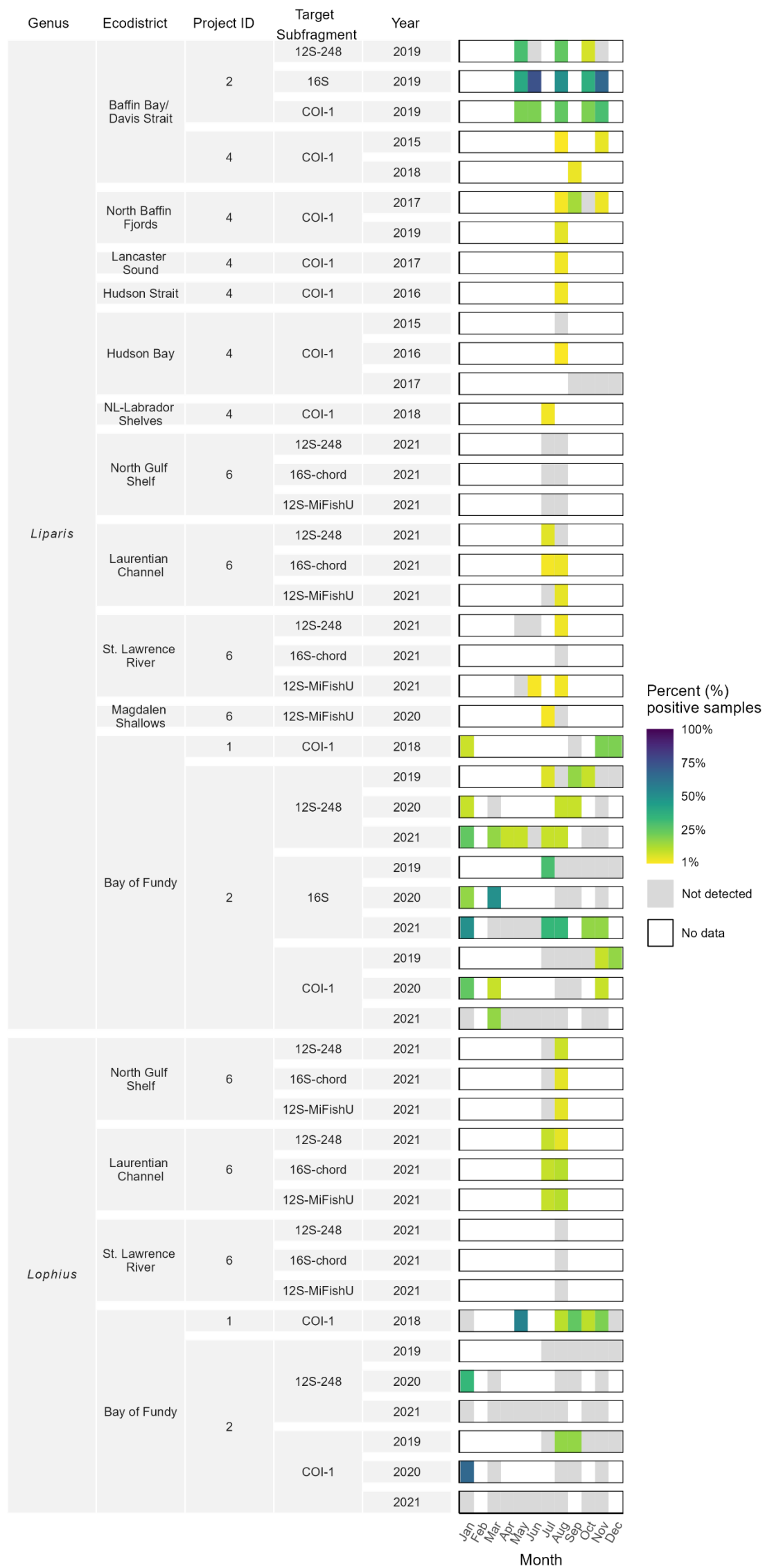


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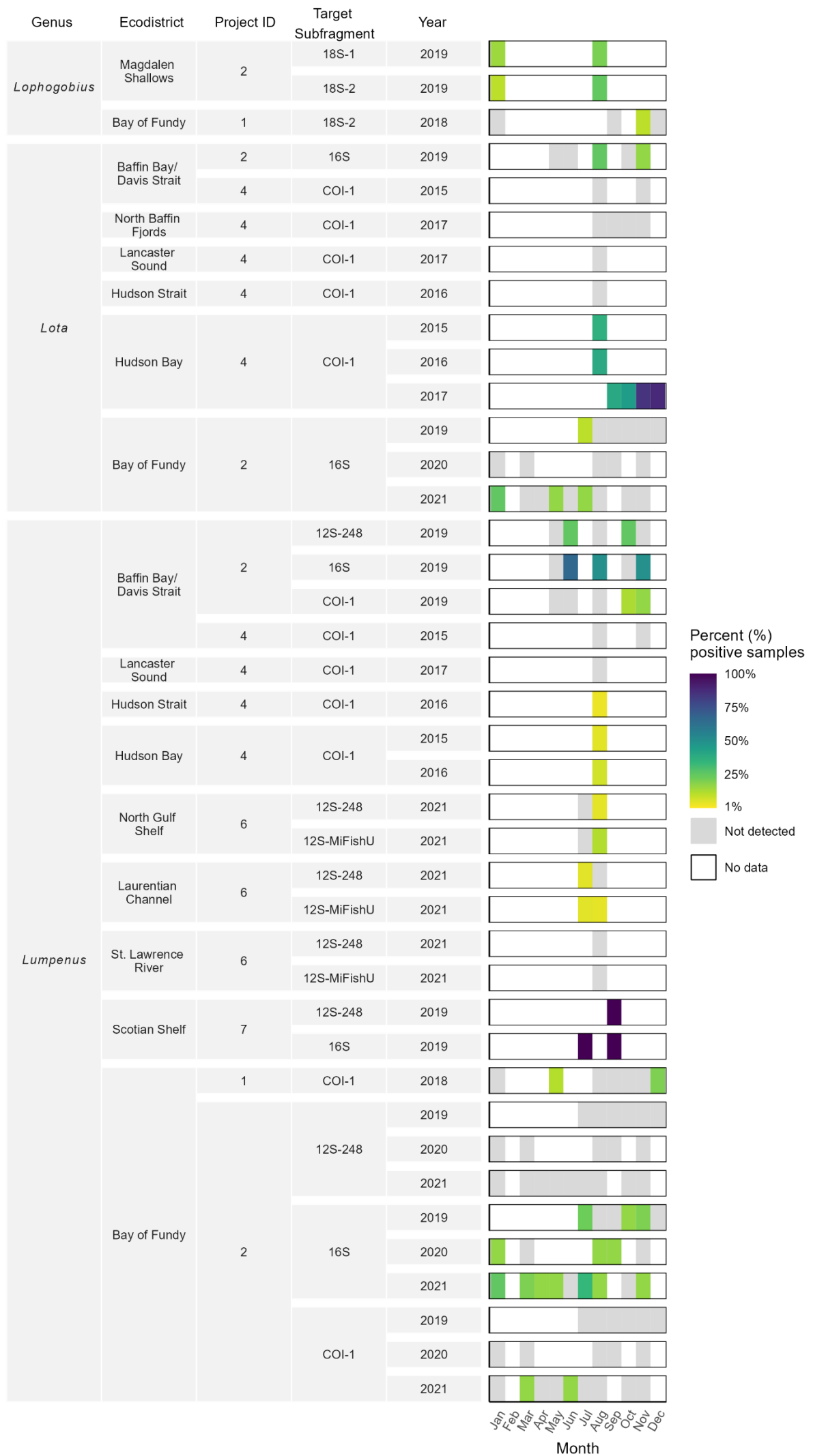


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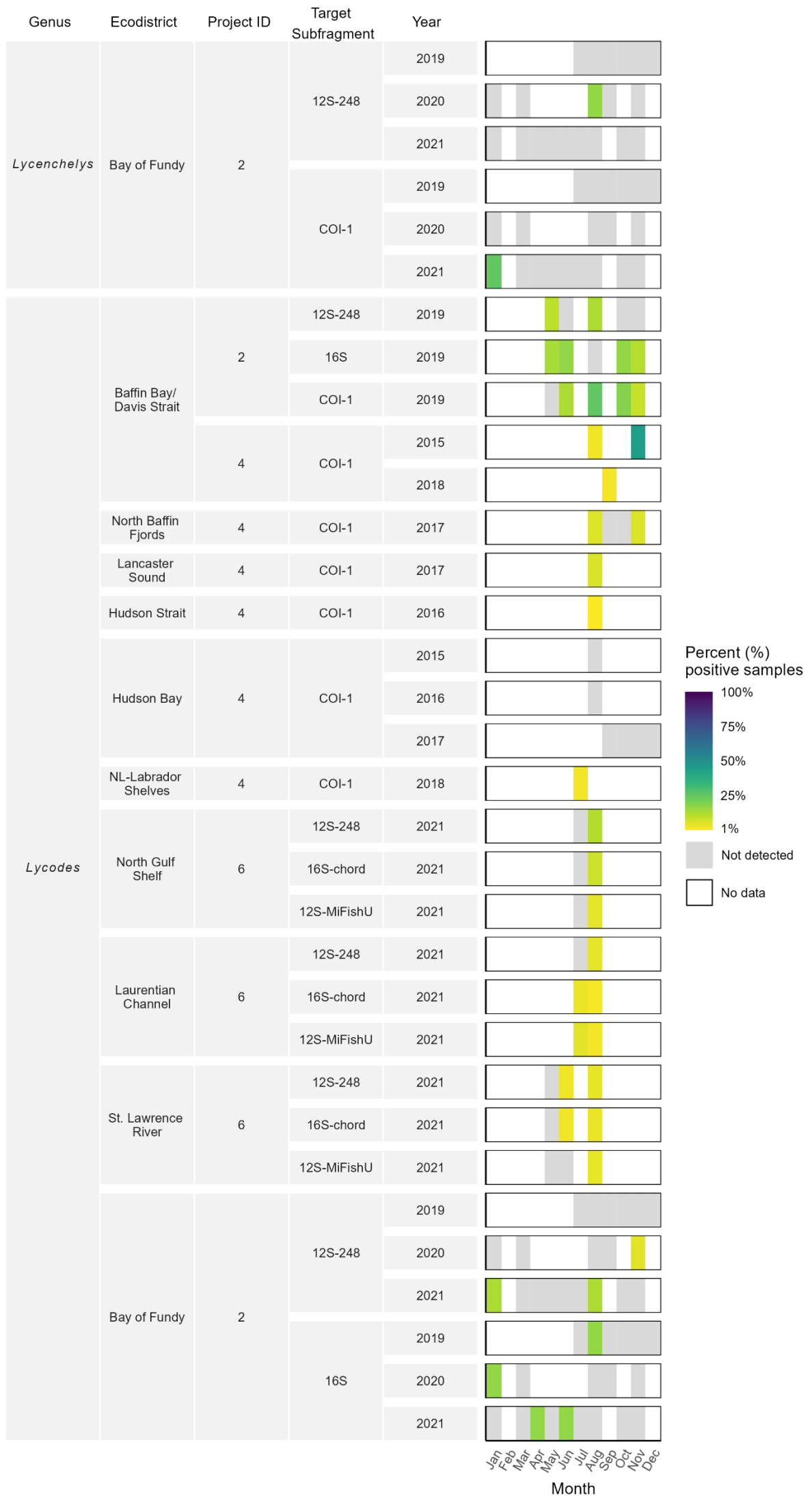


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Figure 13. (Continued)

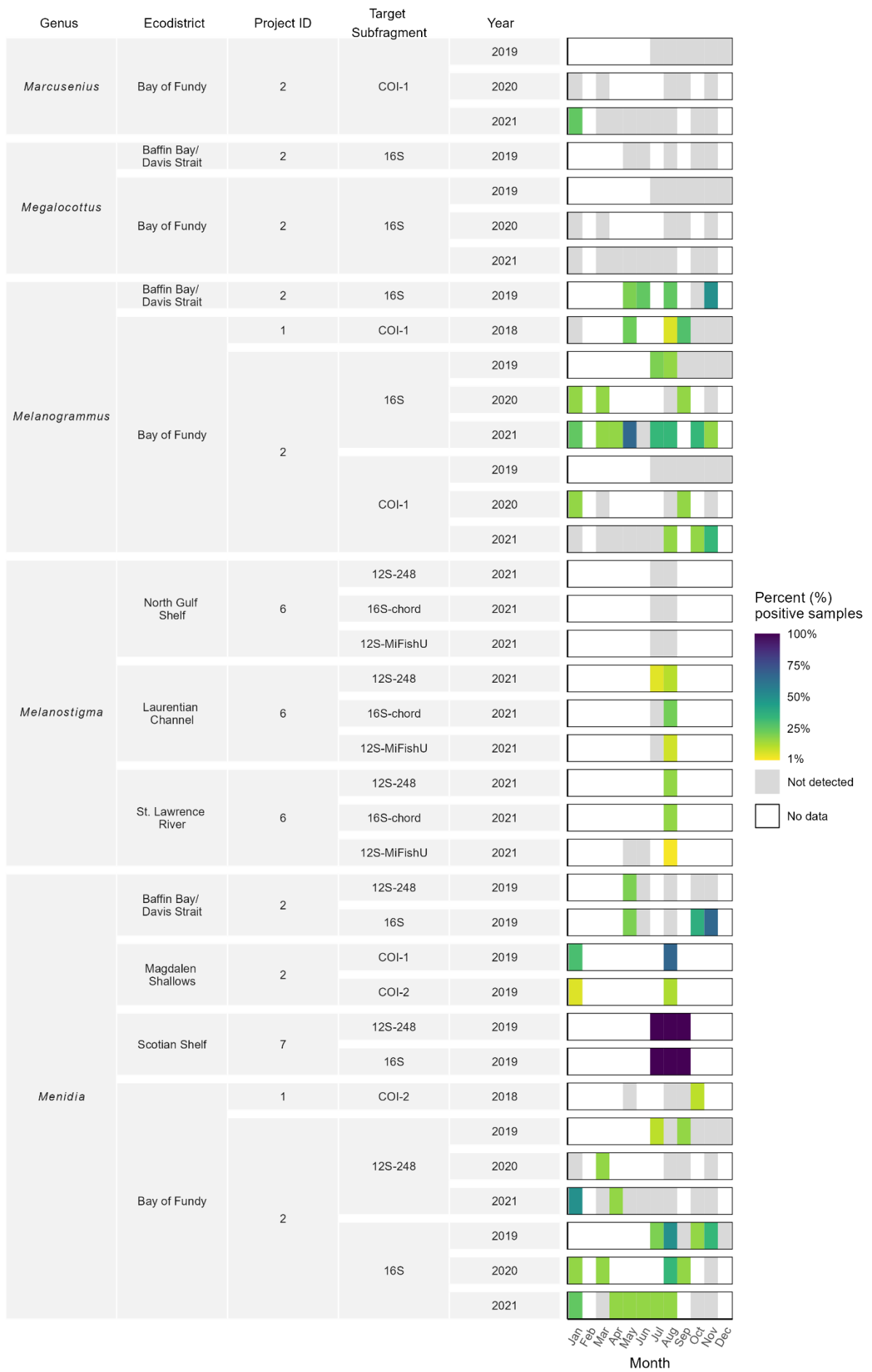


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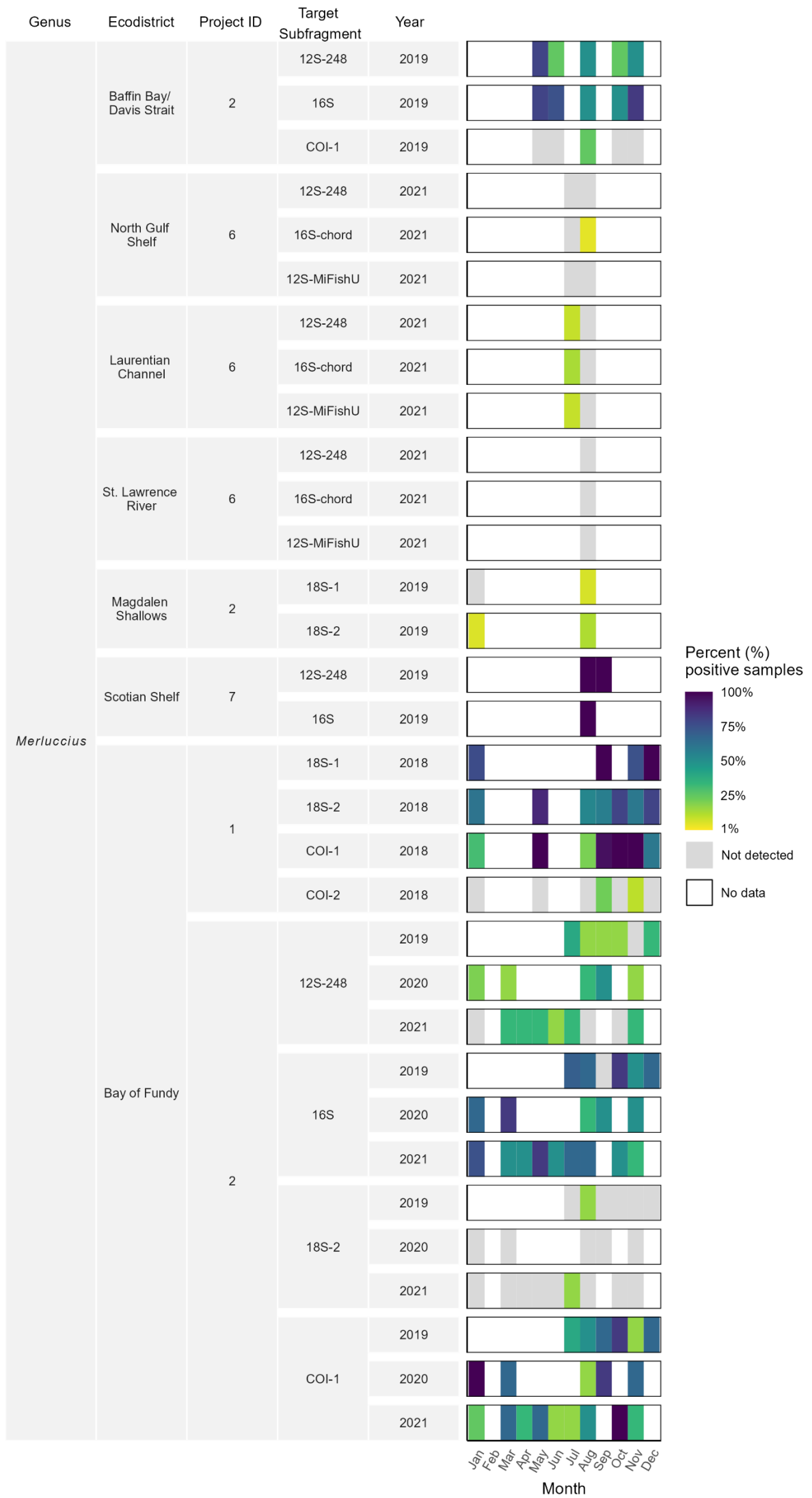


Figure 13. (Continued)



Figure 13. (Continued)

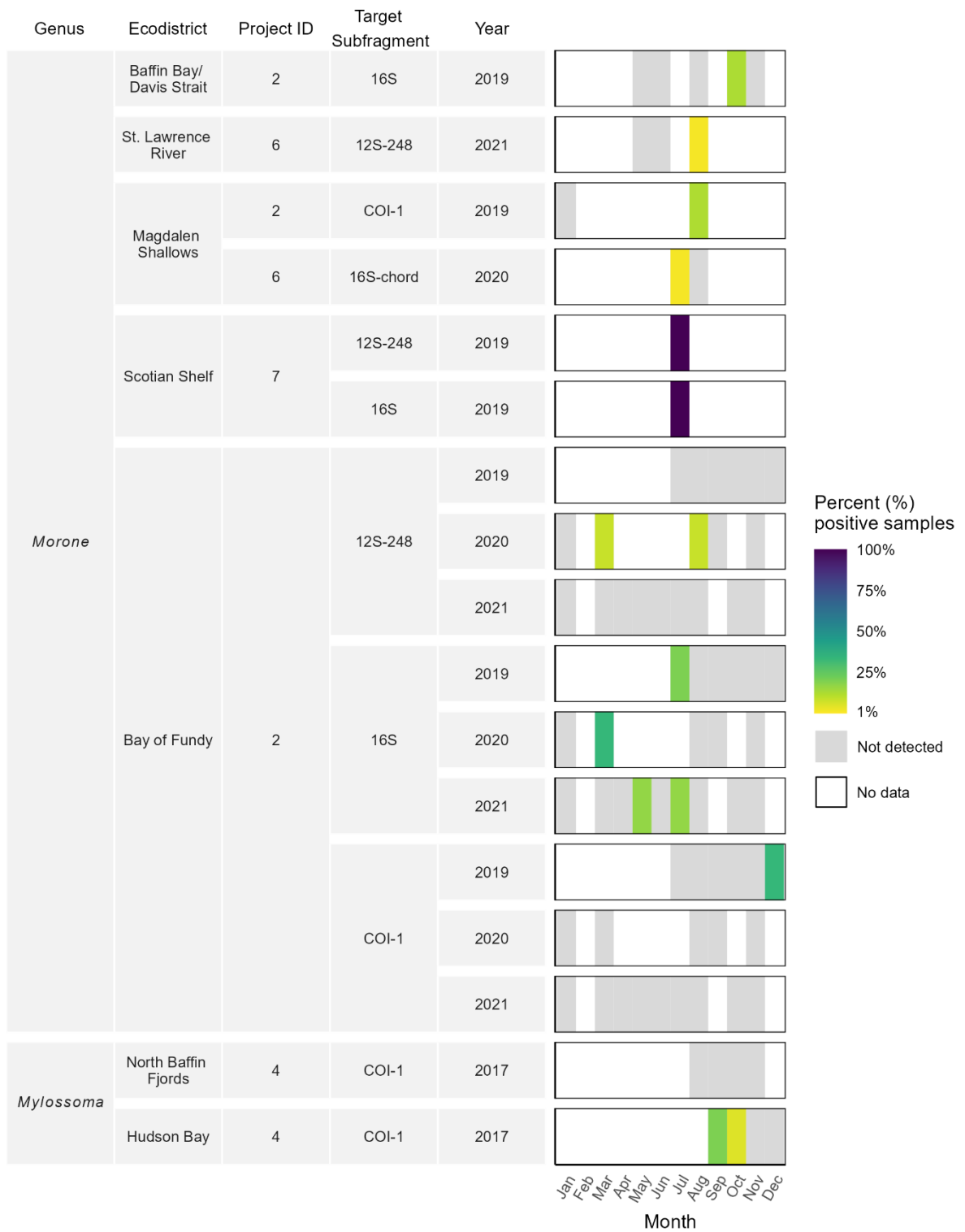


Figure 13. (Continued)



Figure 13. (Continued)

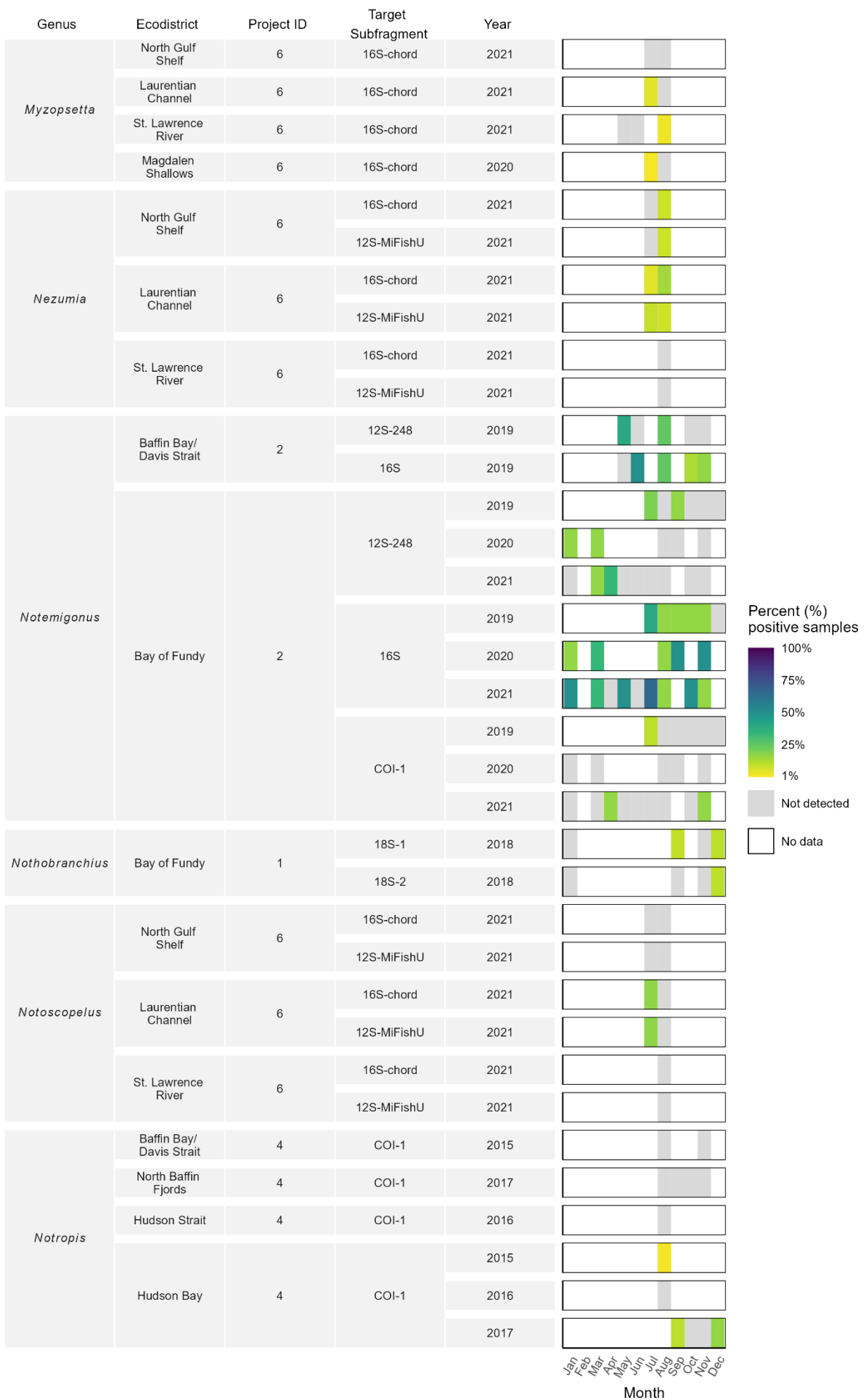


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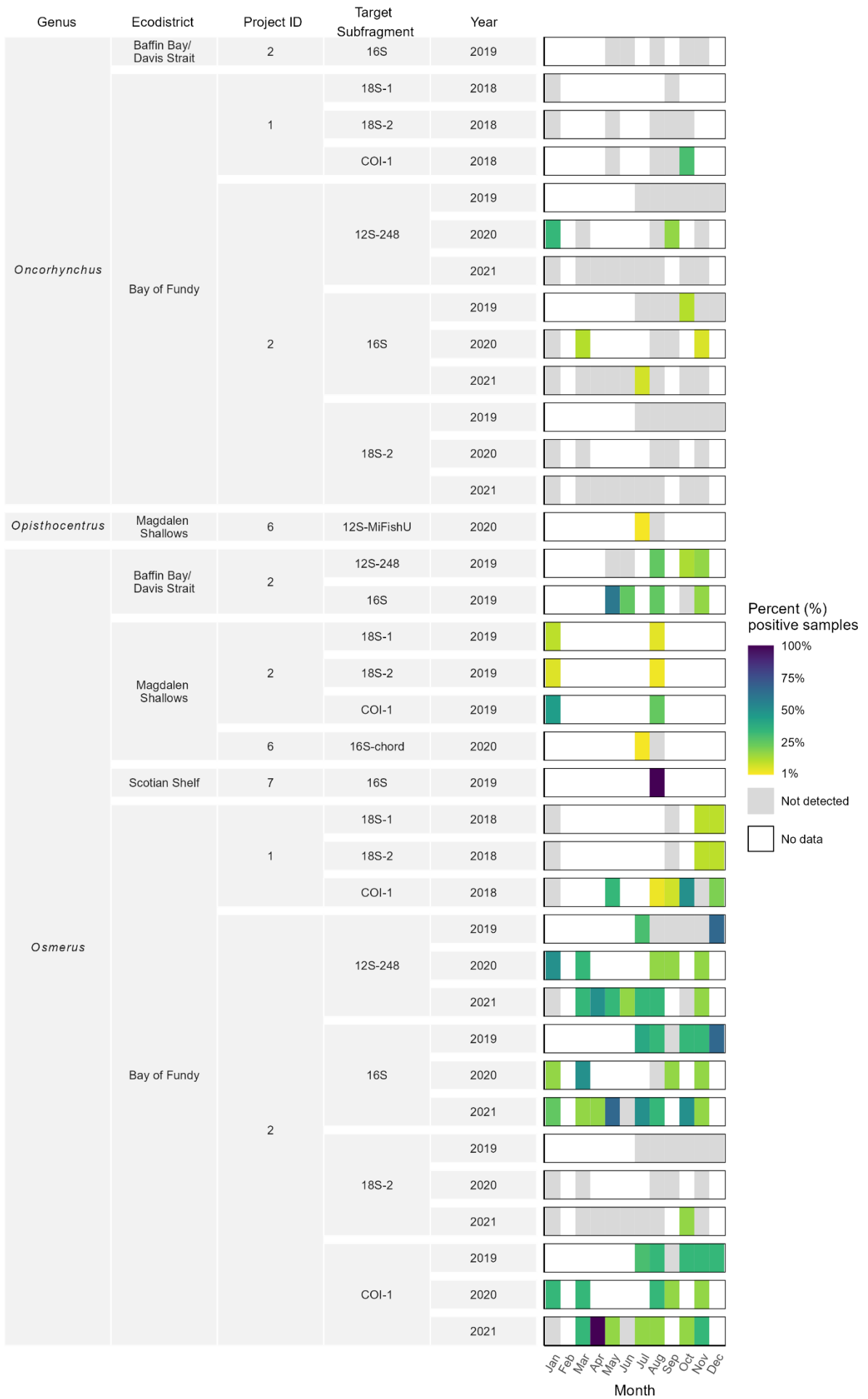


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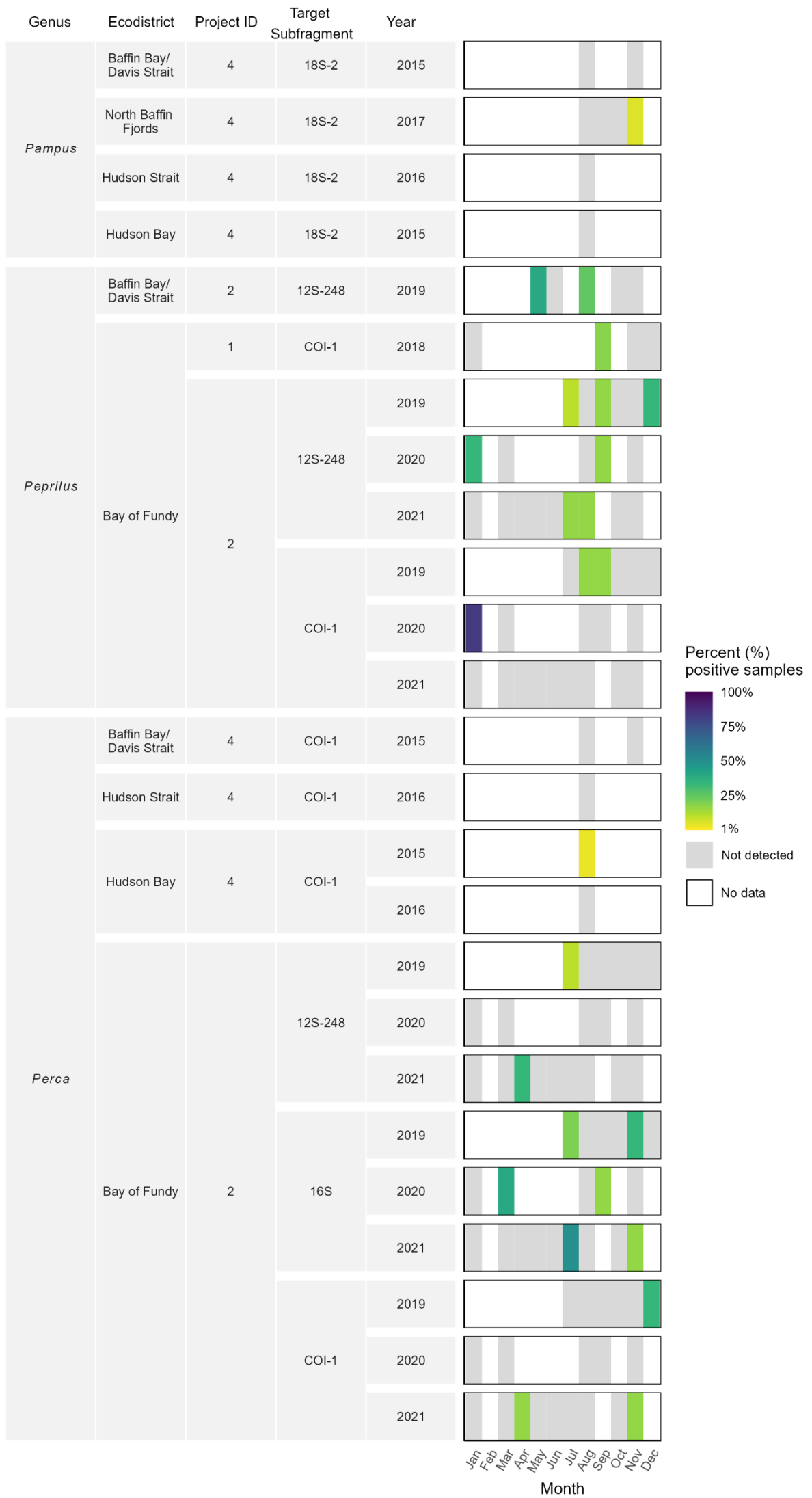


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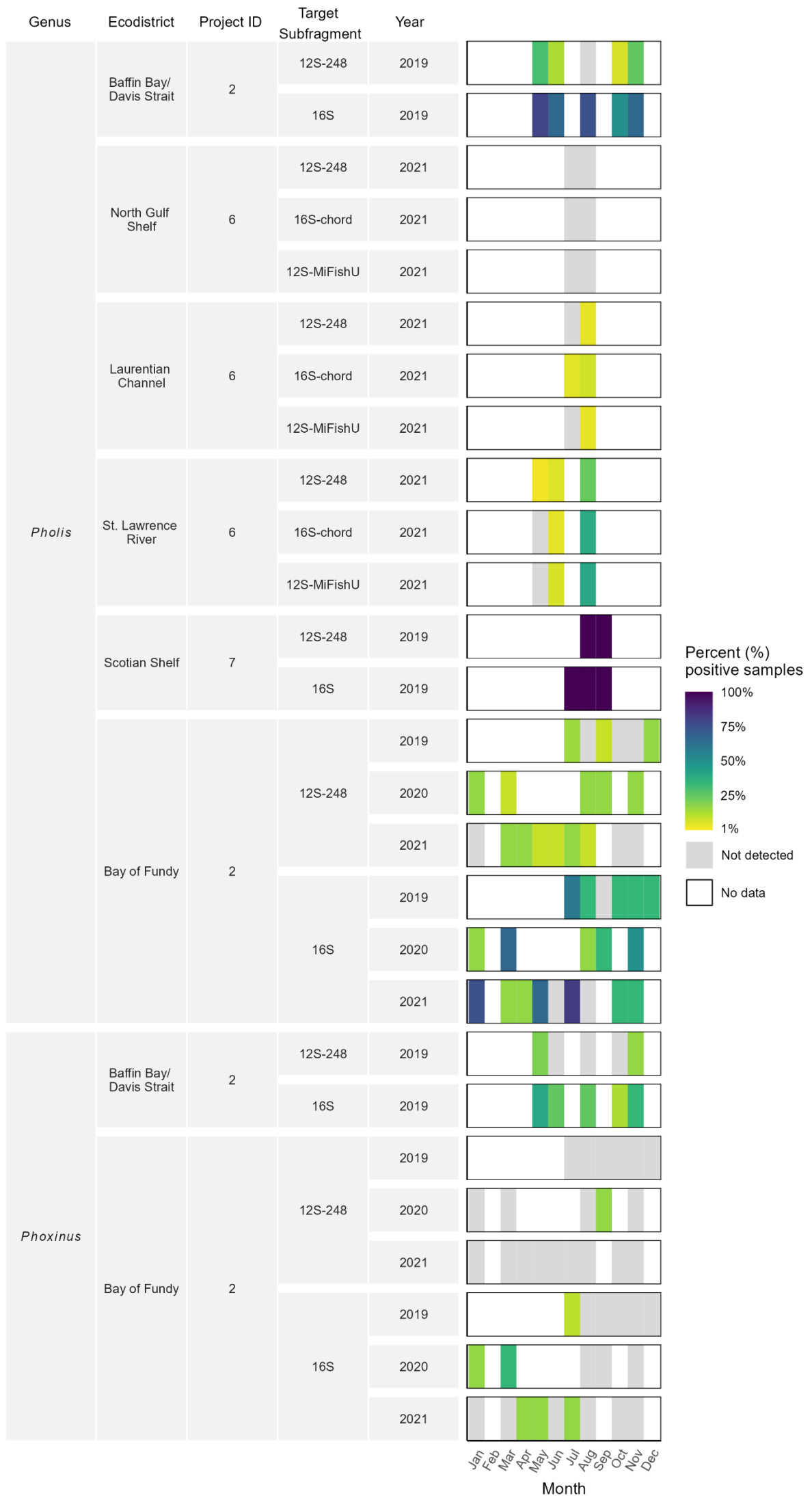


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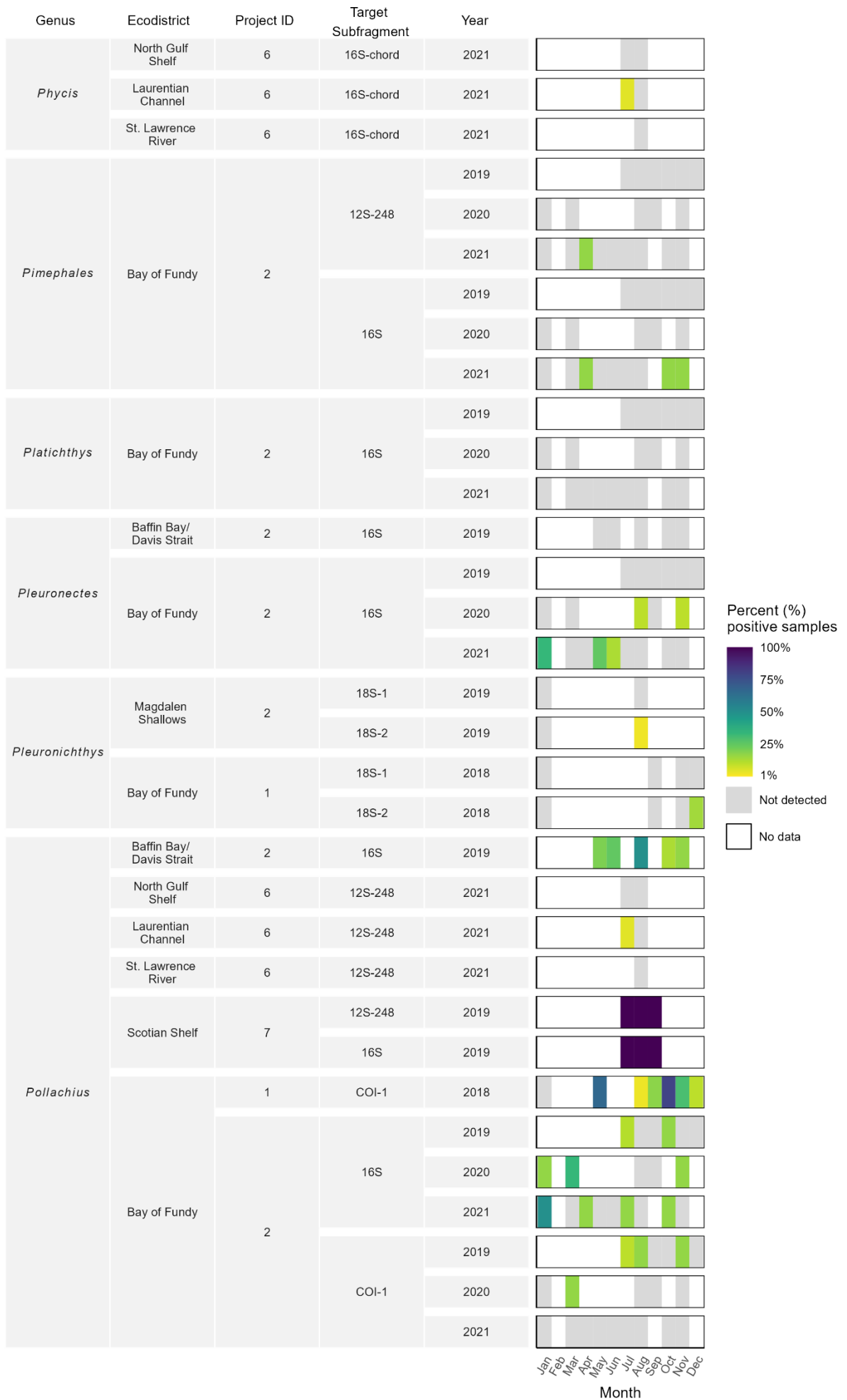


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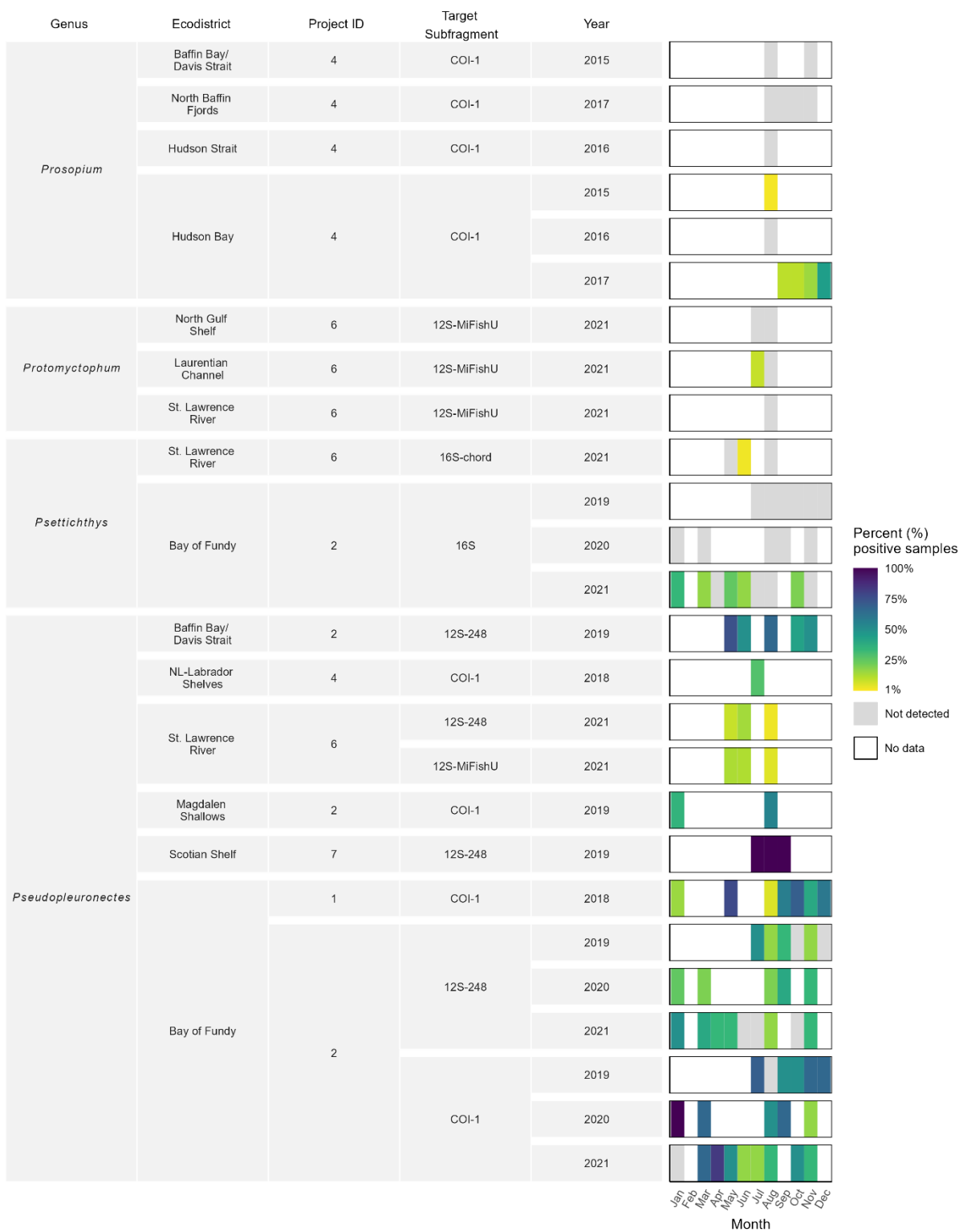


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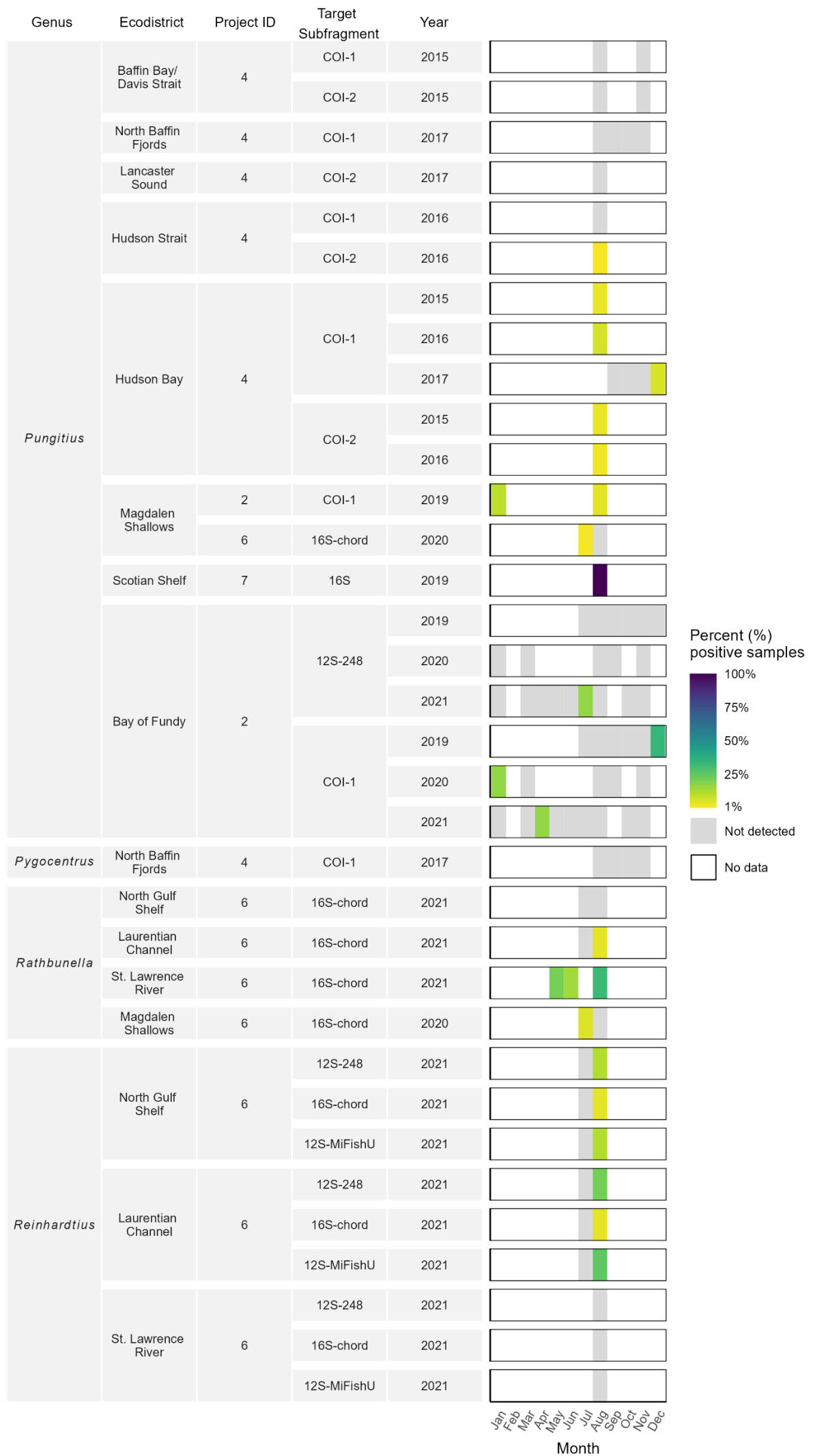


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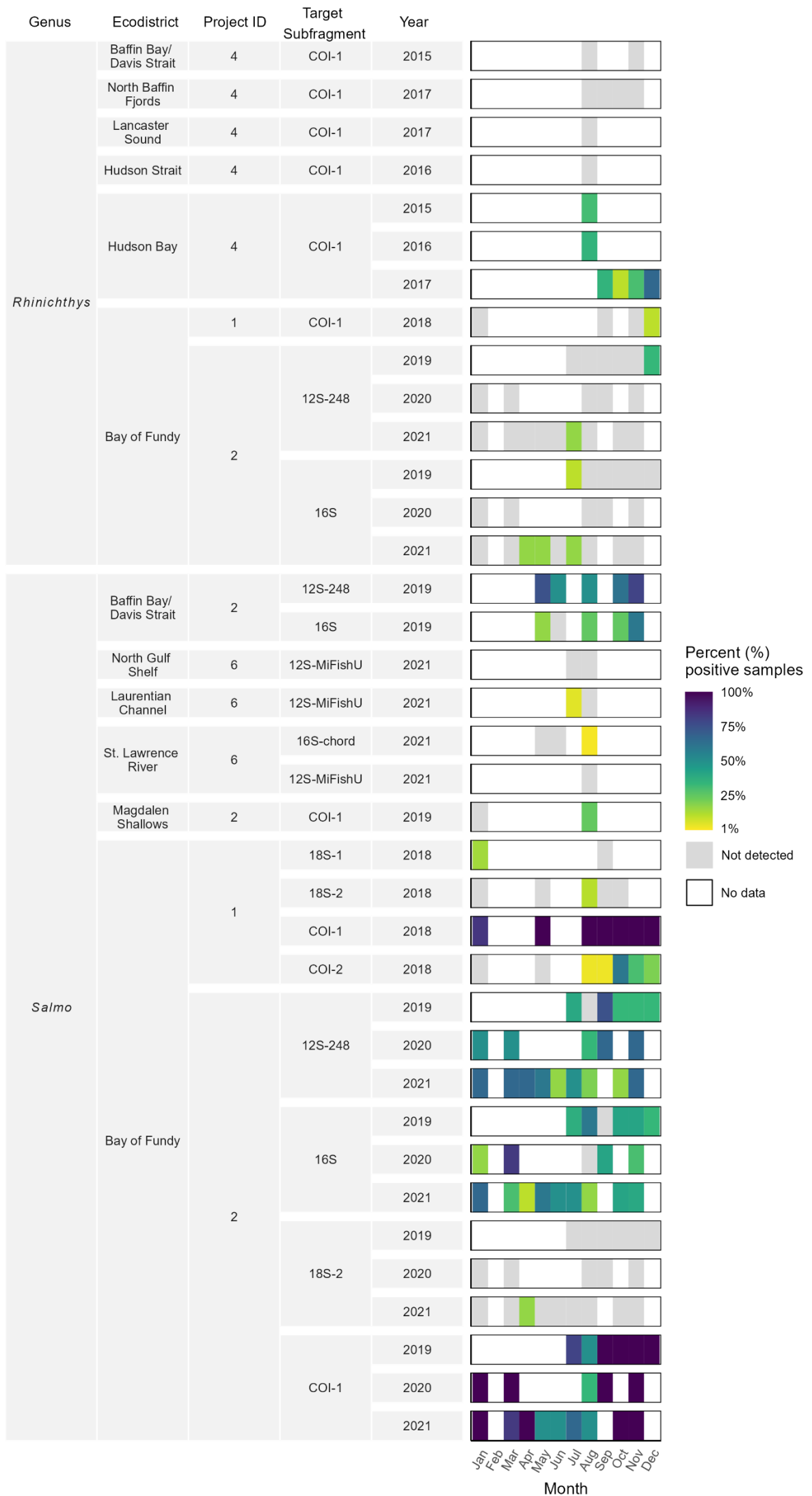


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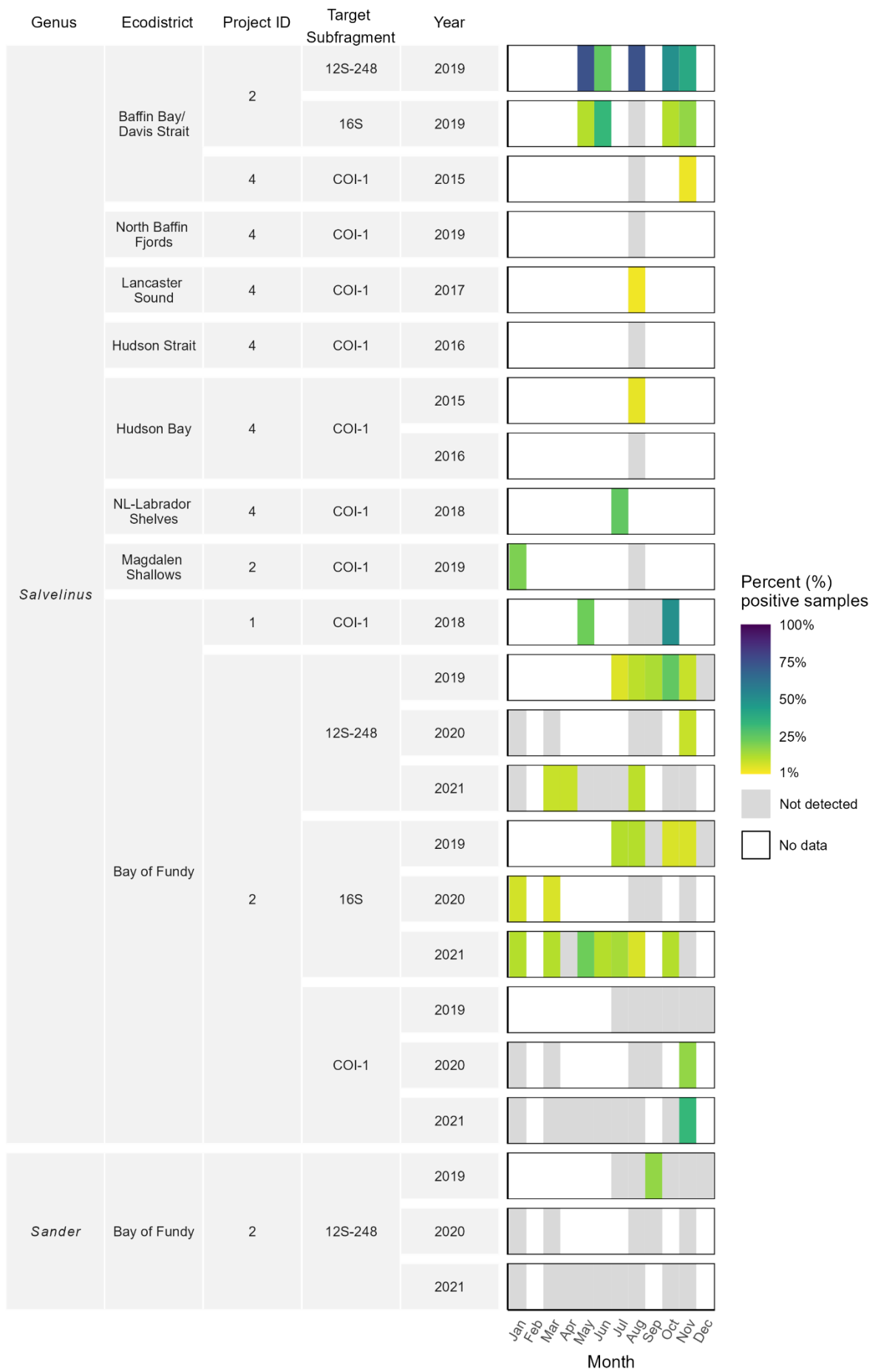


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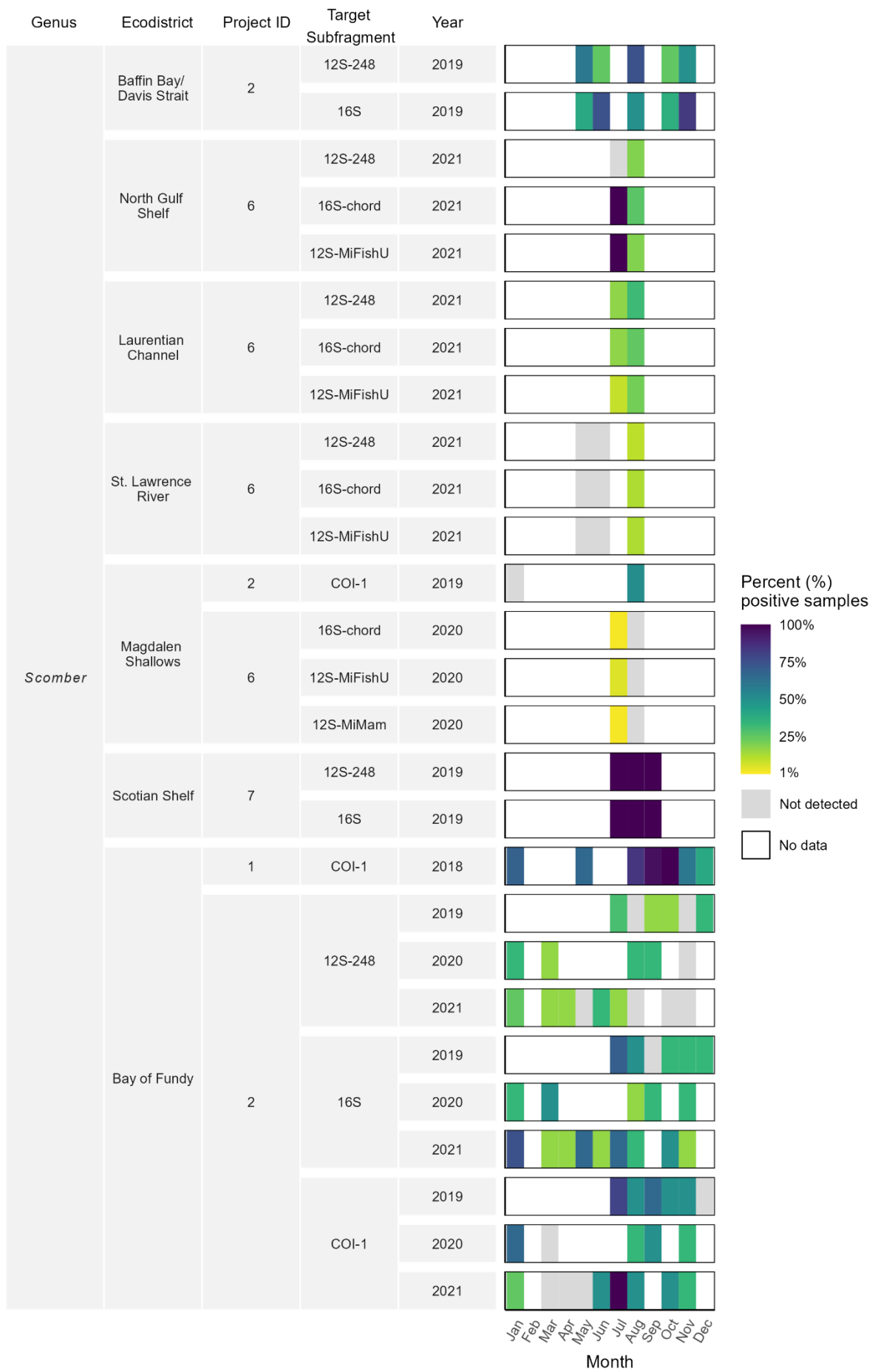


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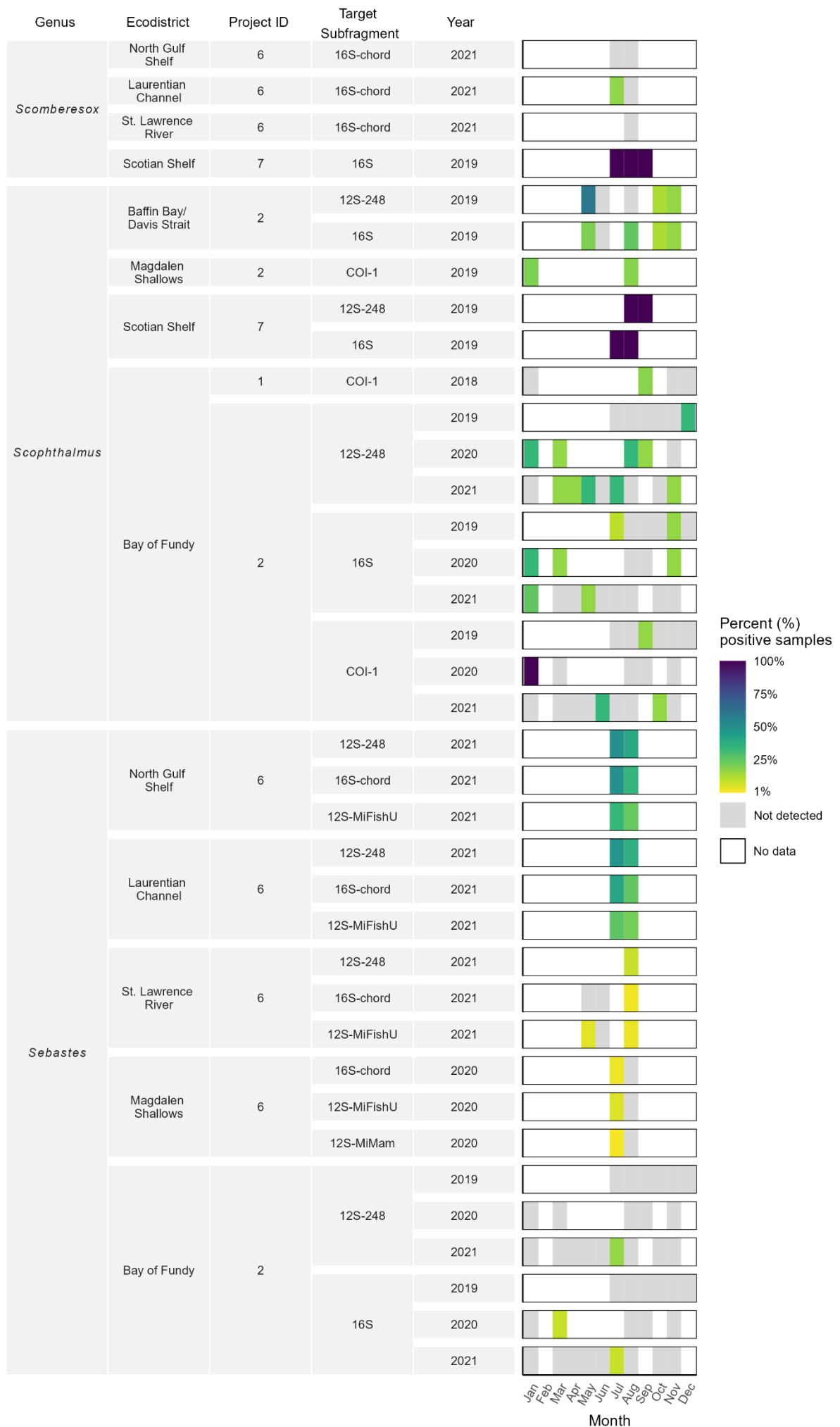


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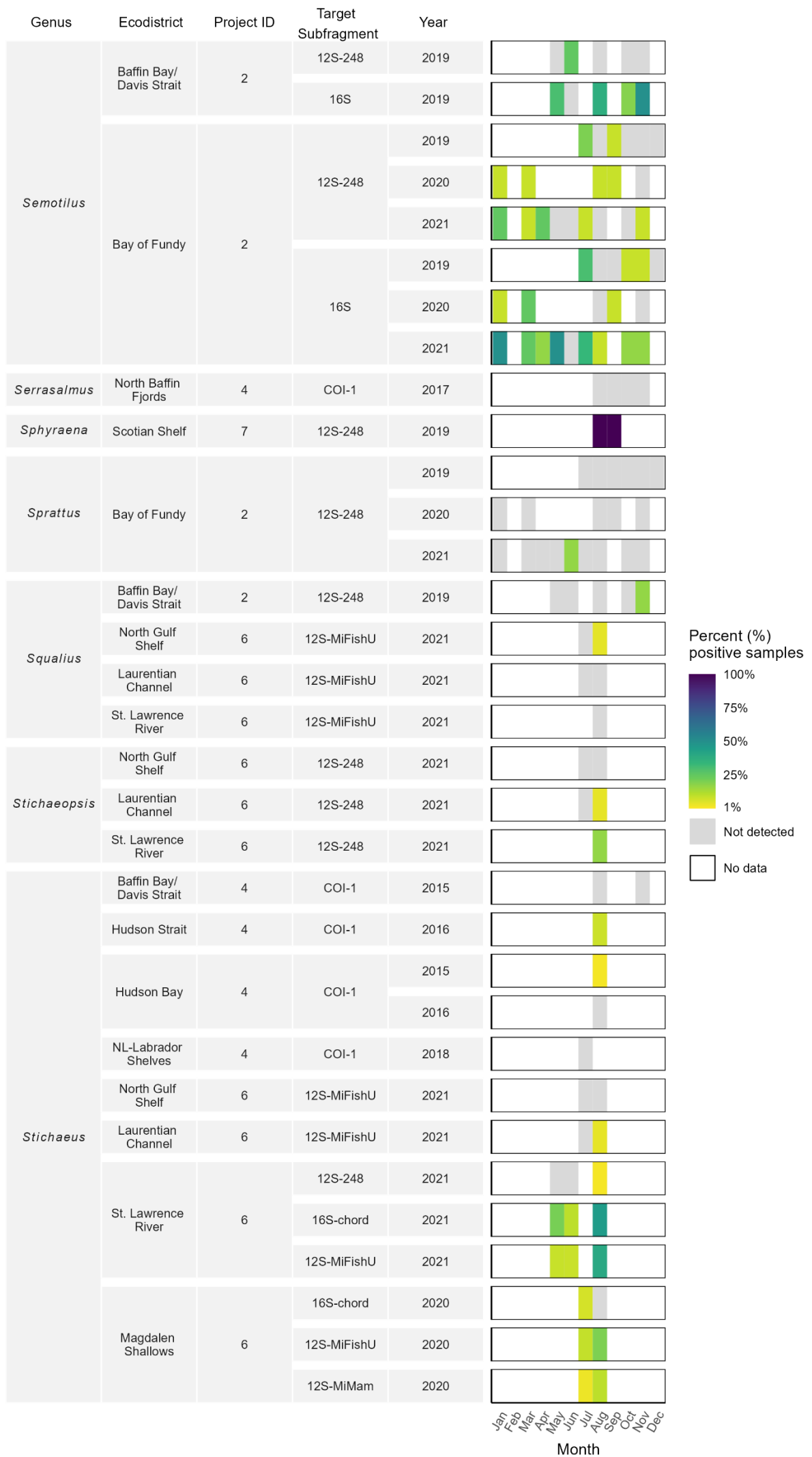


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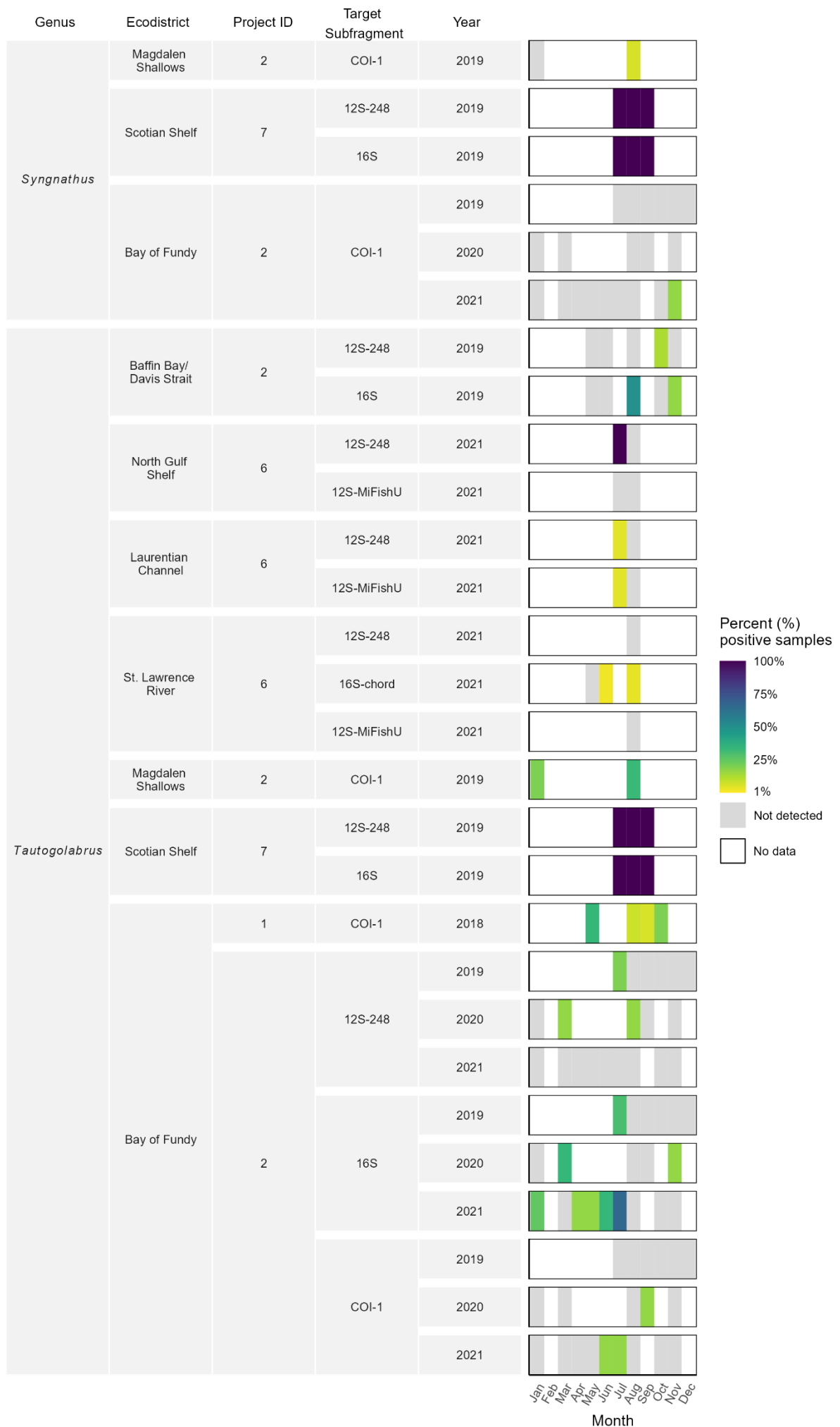


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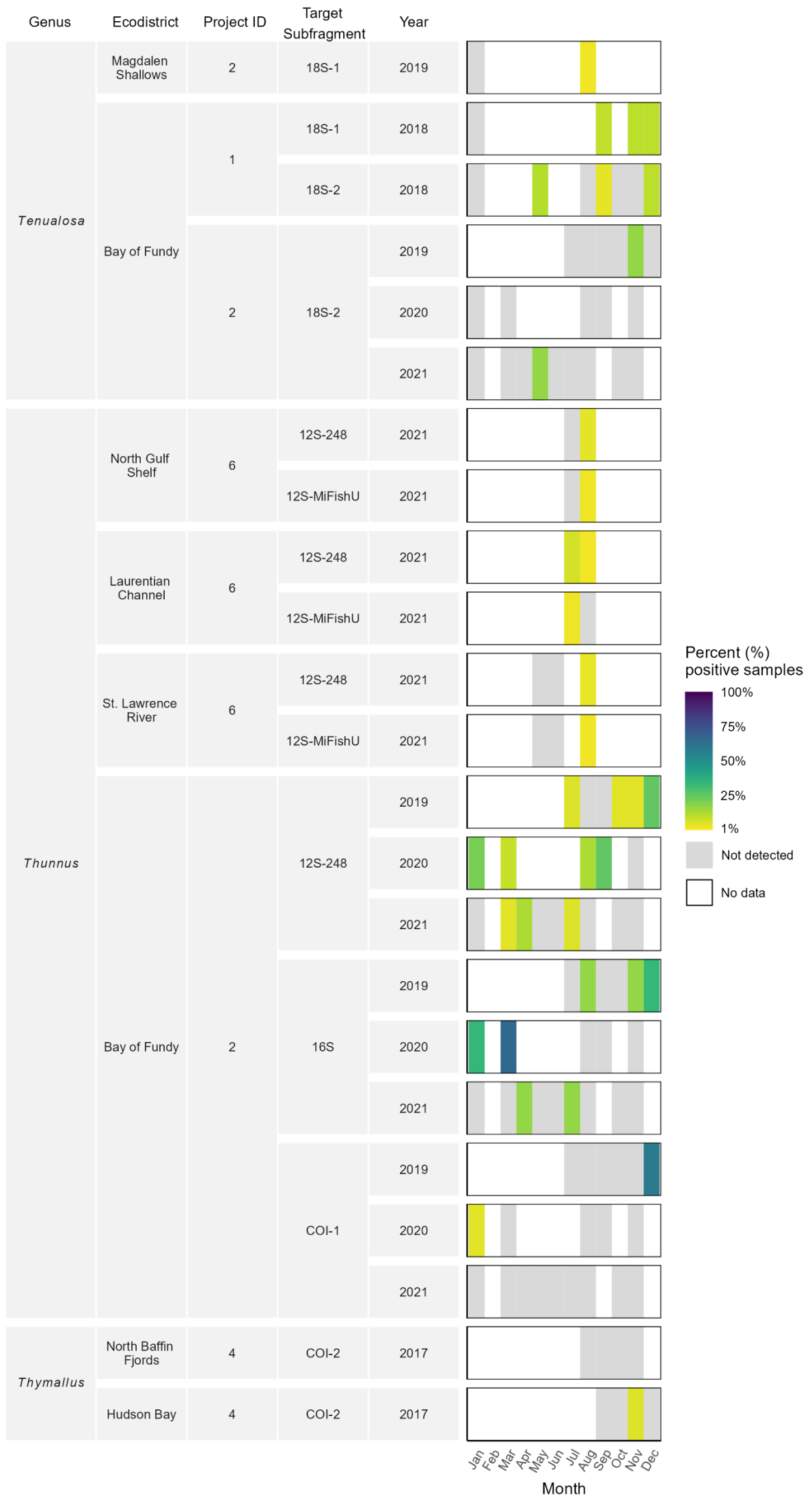


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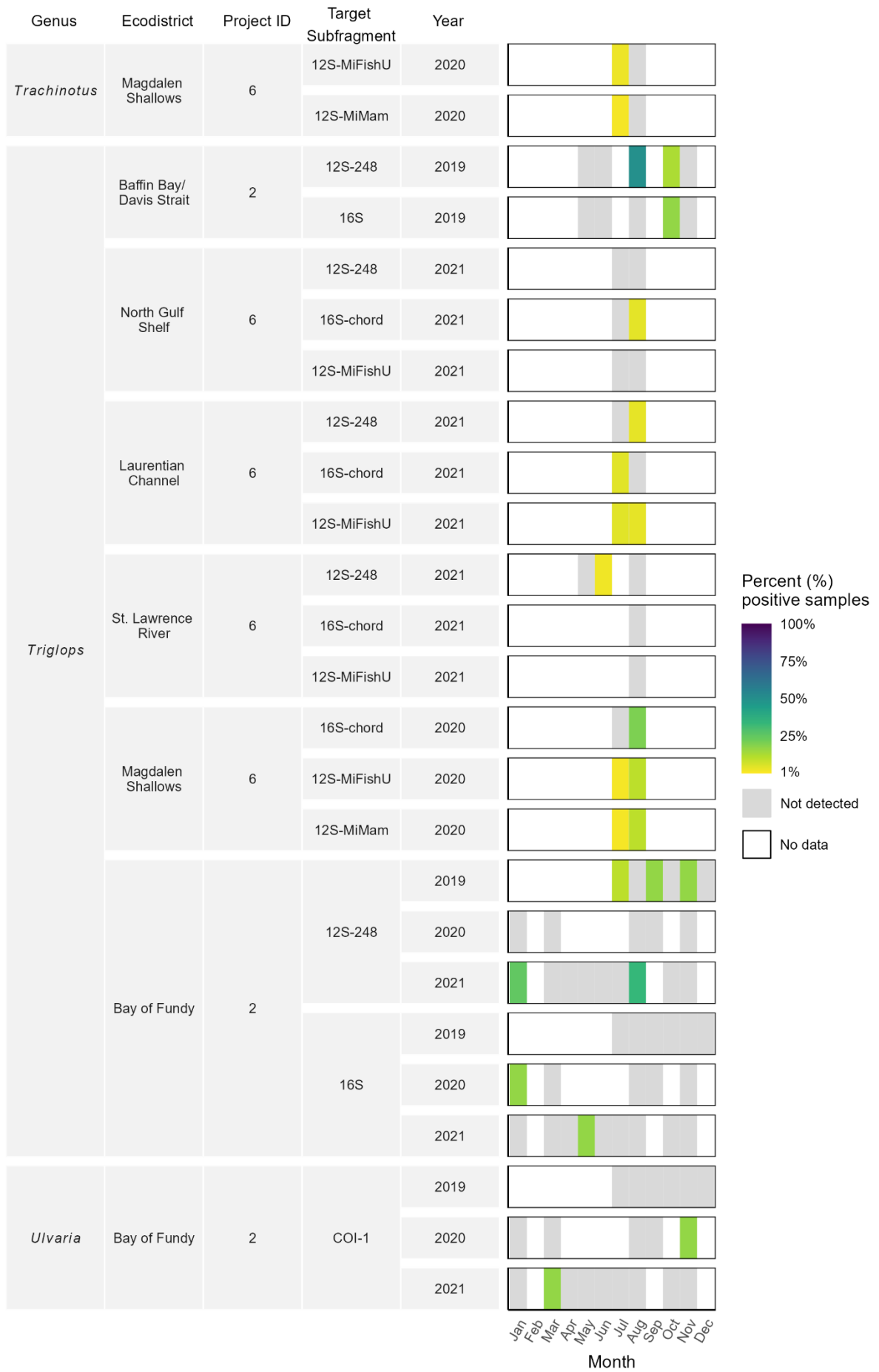


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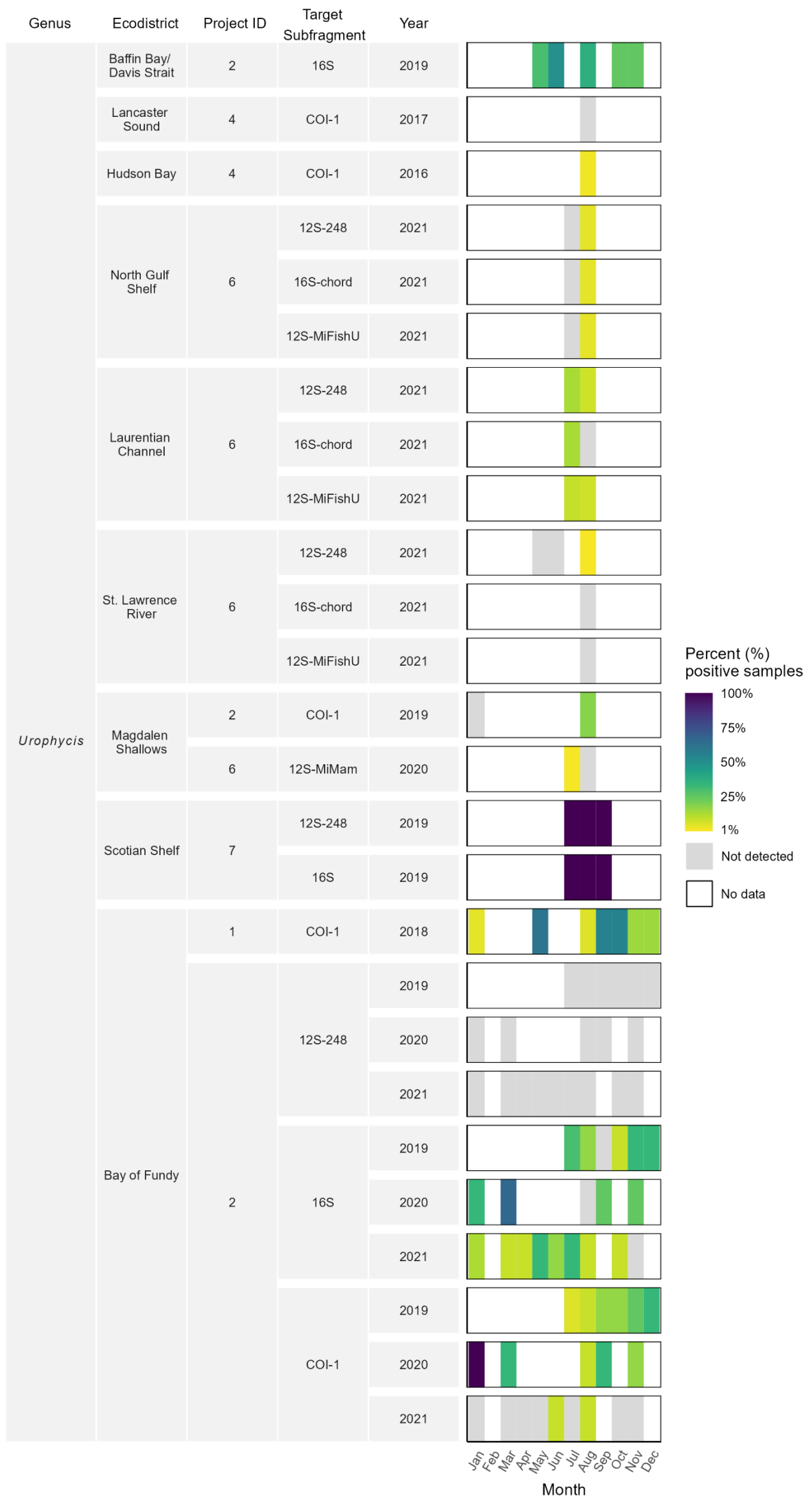


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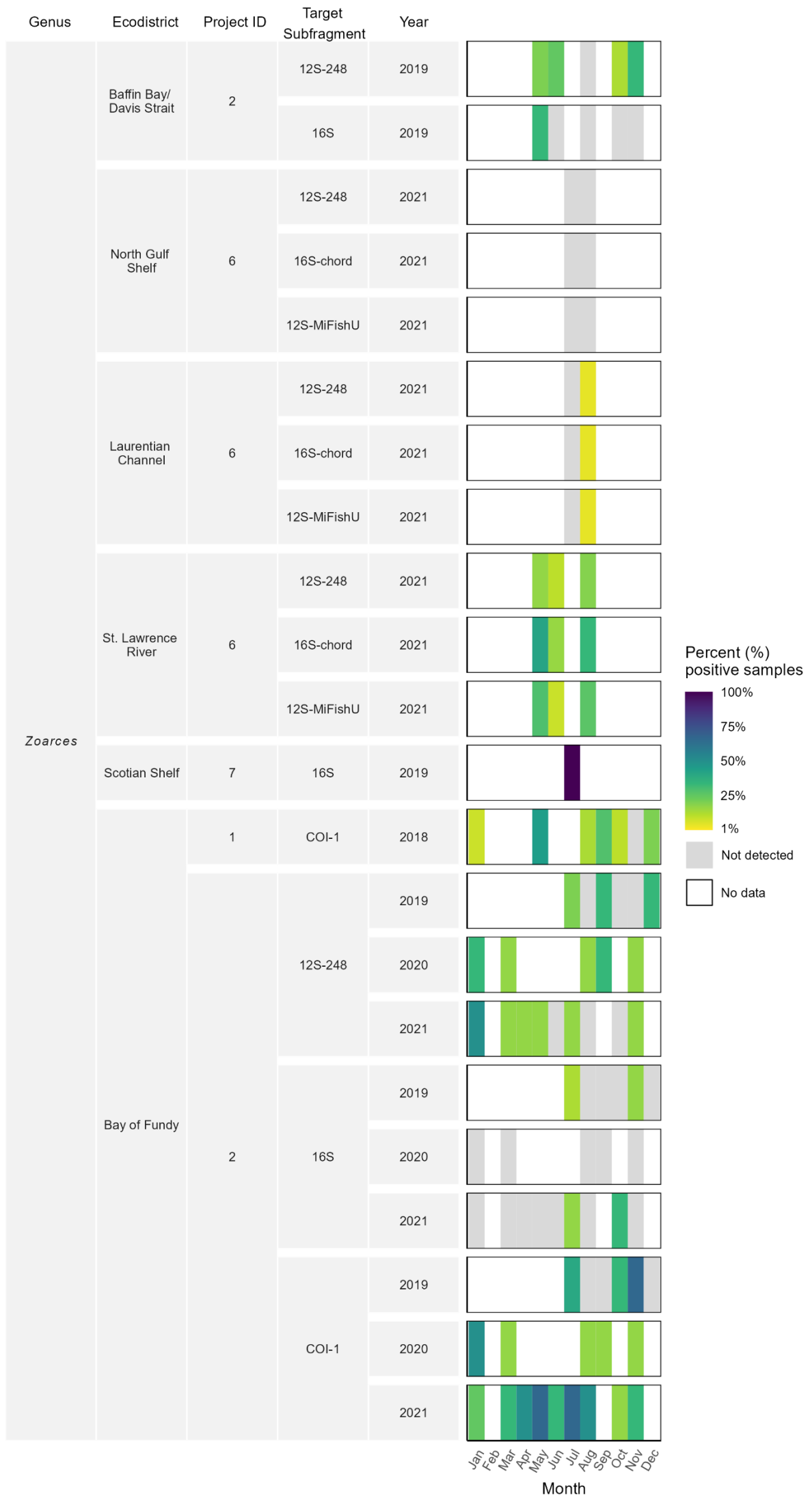


Figure 13. (Continued)

6.5.2 Amphibia



Figure 13. (Continued)

6.5.3 Ascidiacea

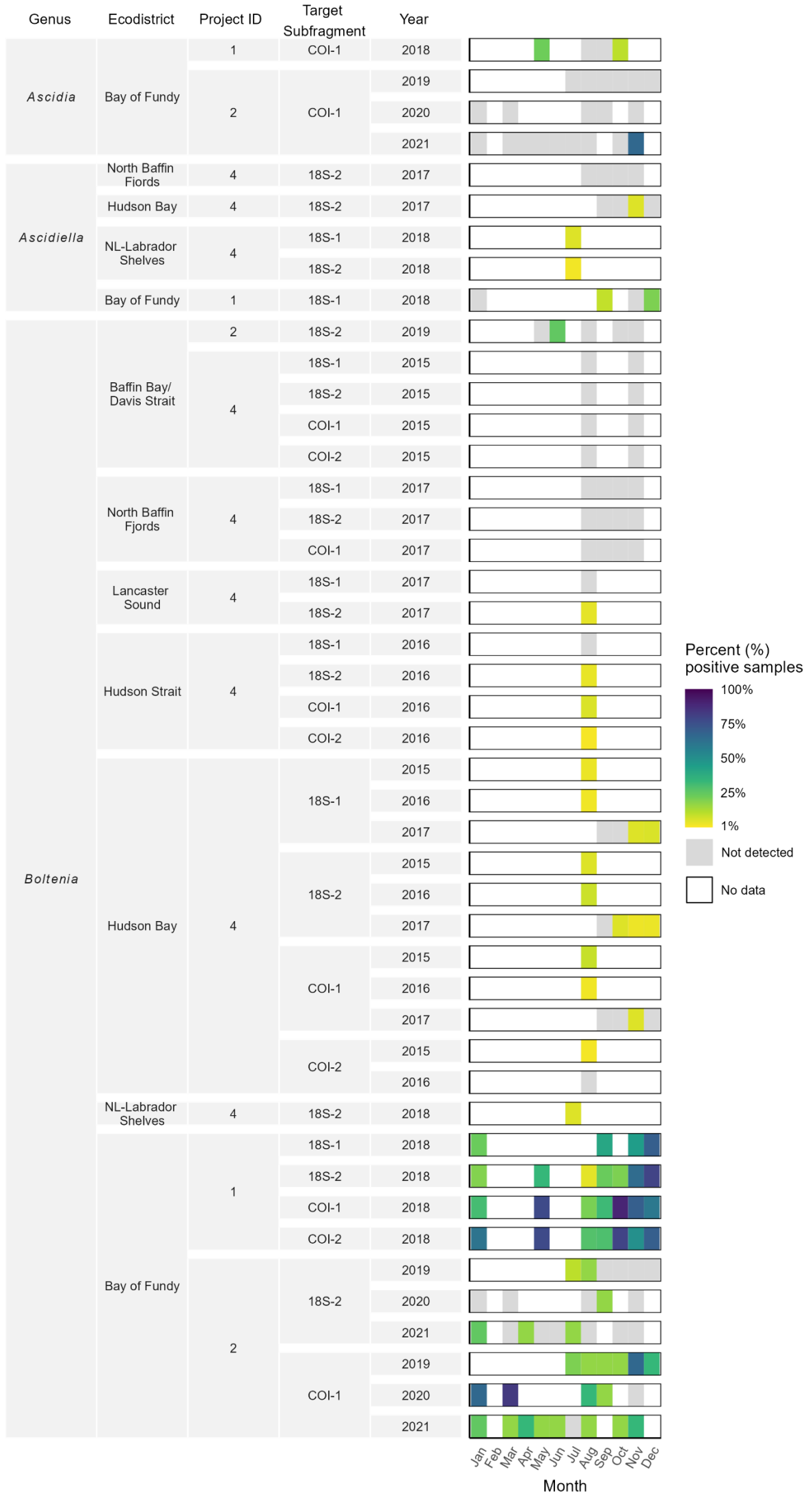


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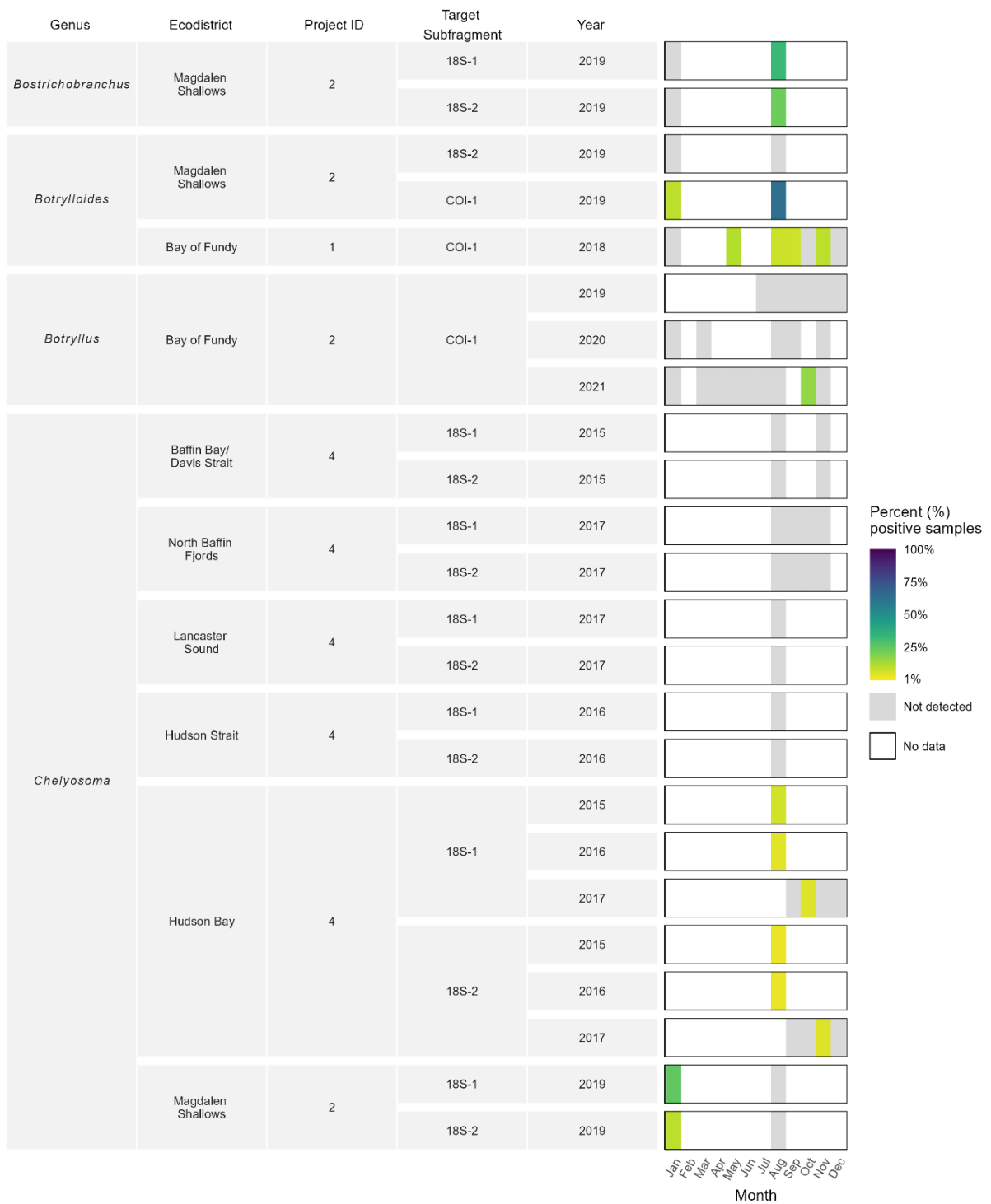


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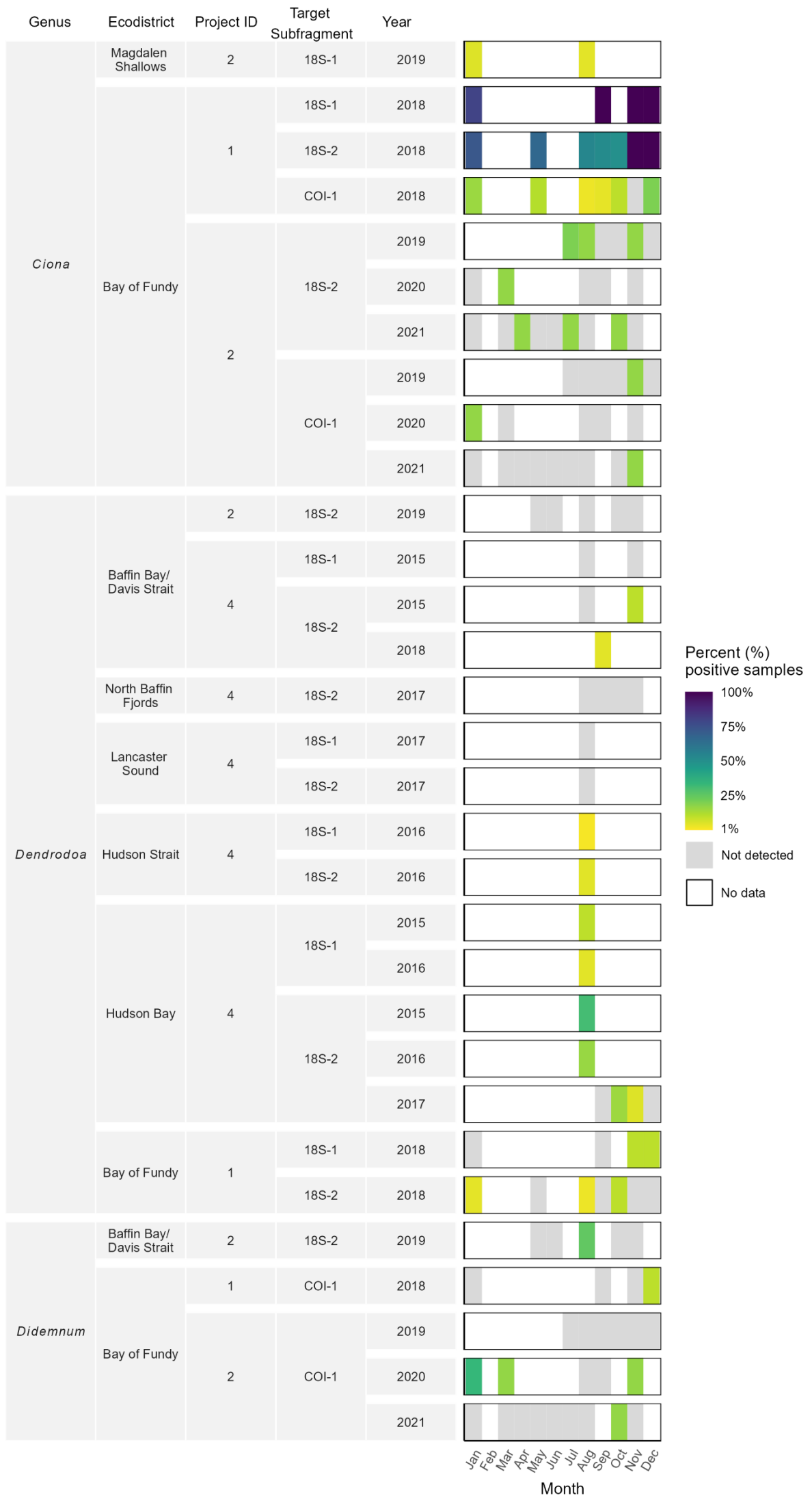


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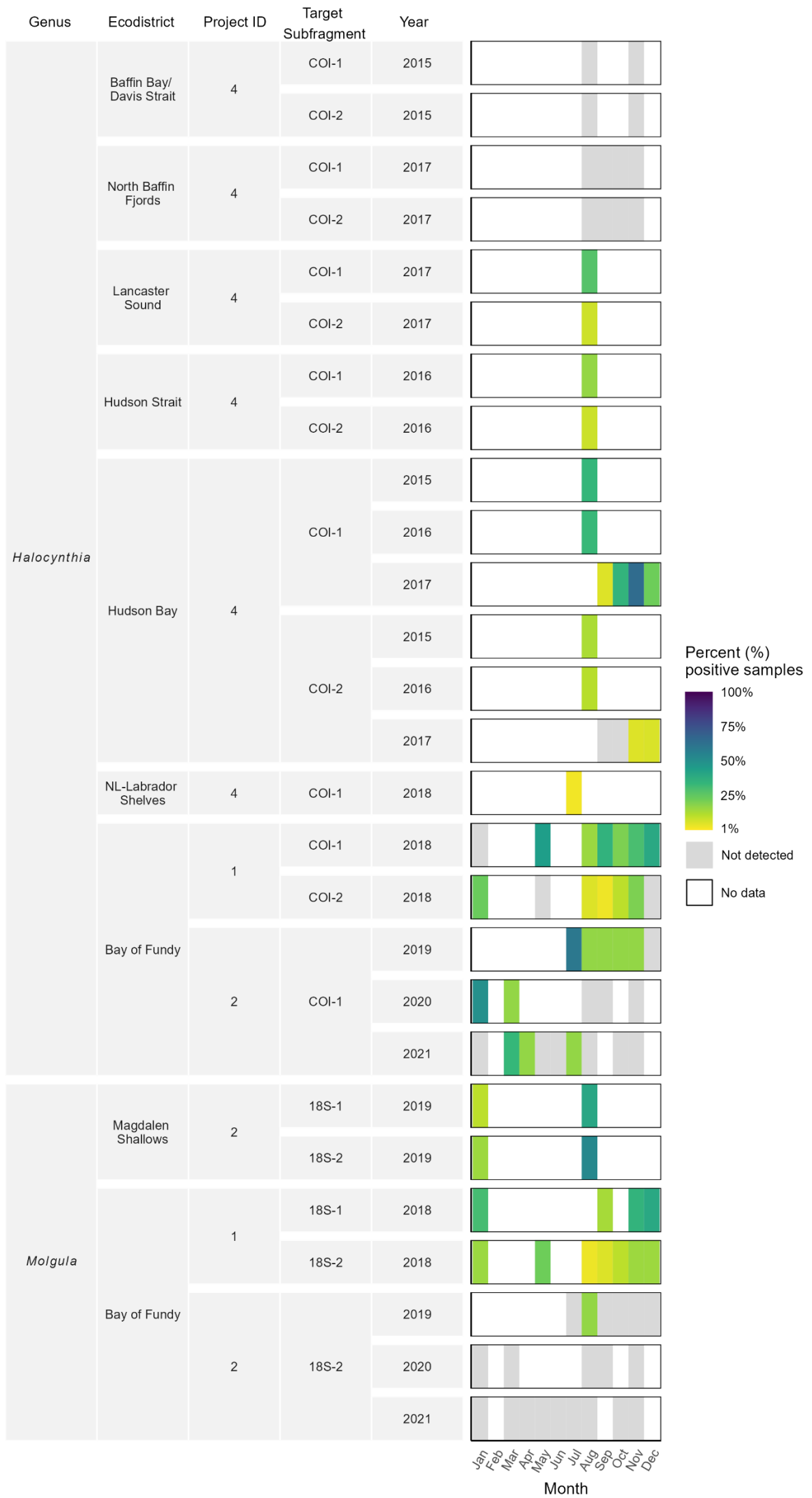


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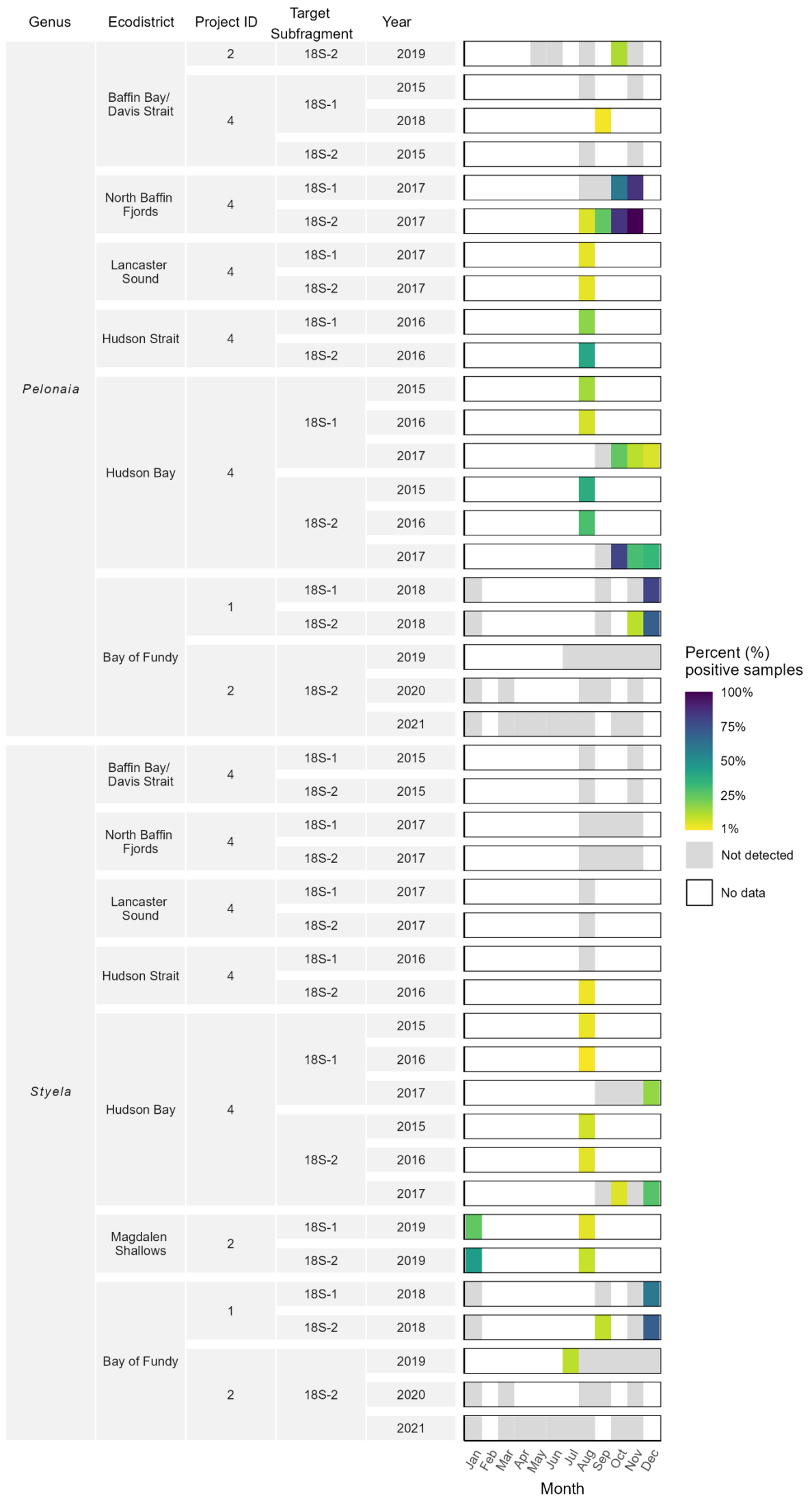


Figure 13. (Continued)

6.5.4 Aves

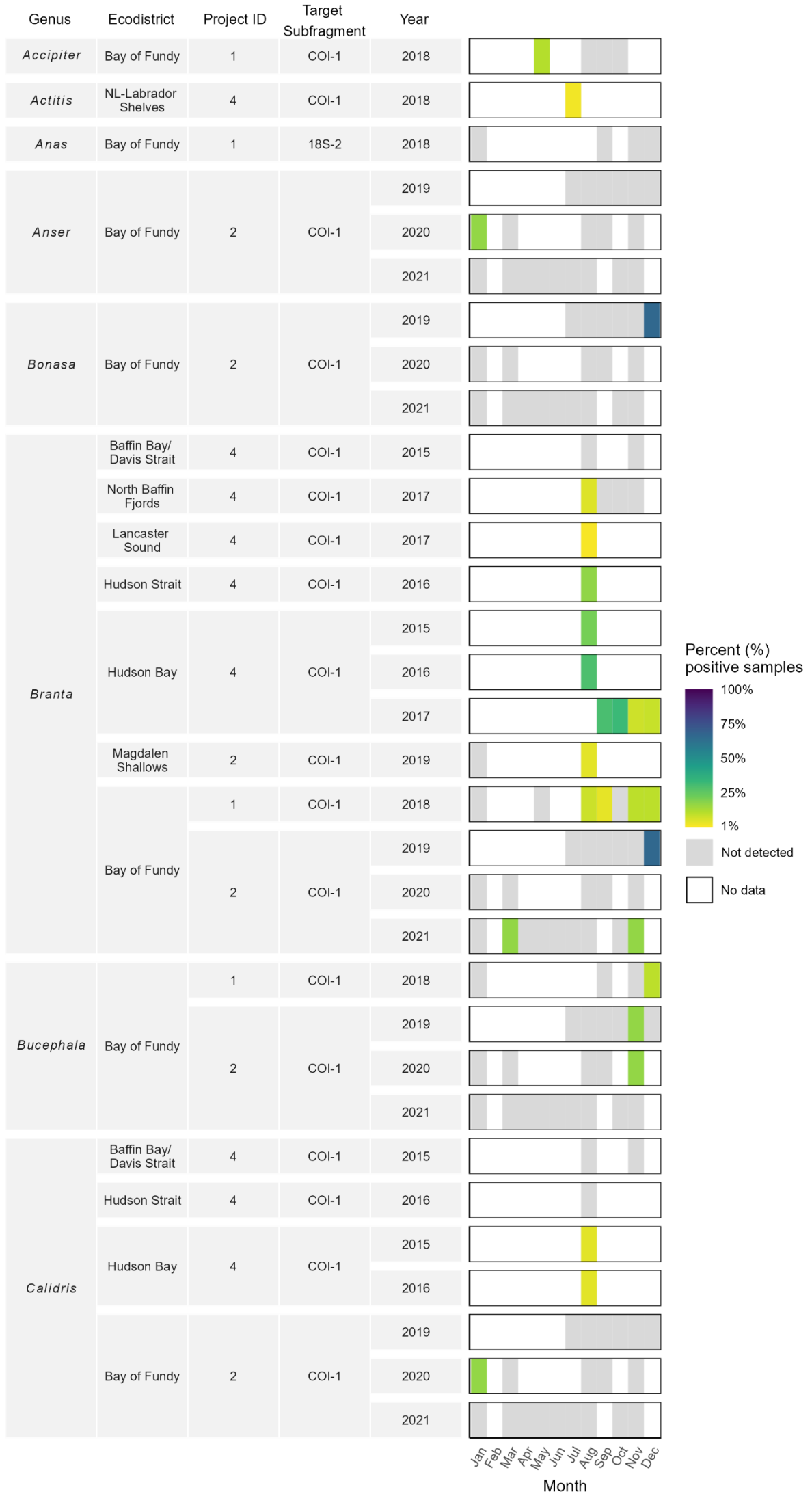


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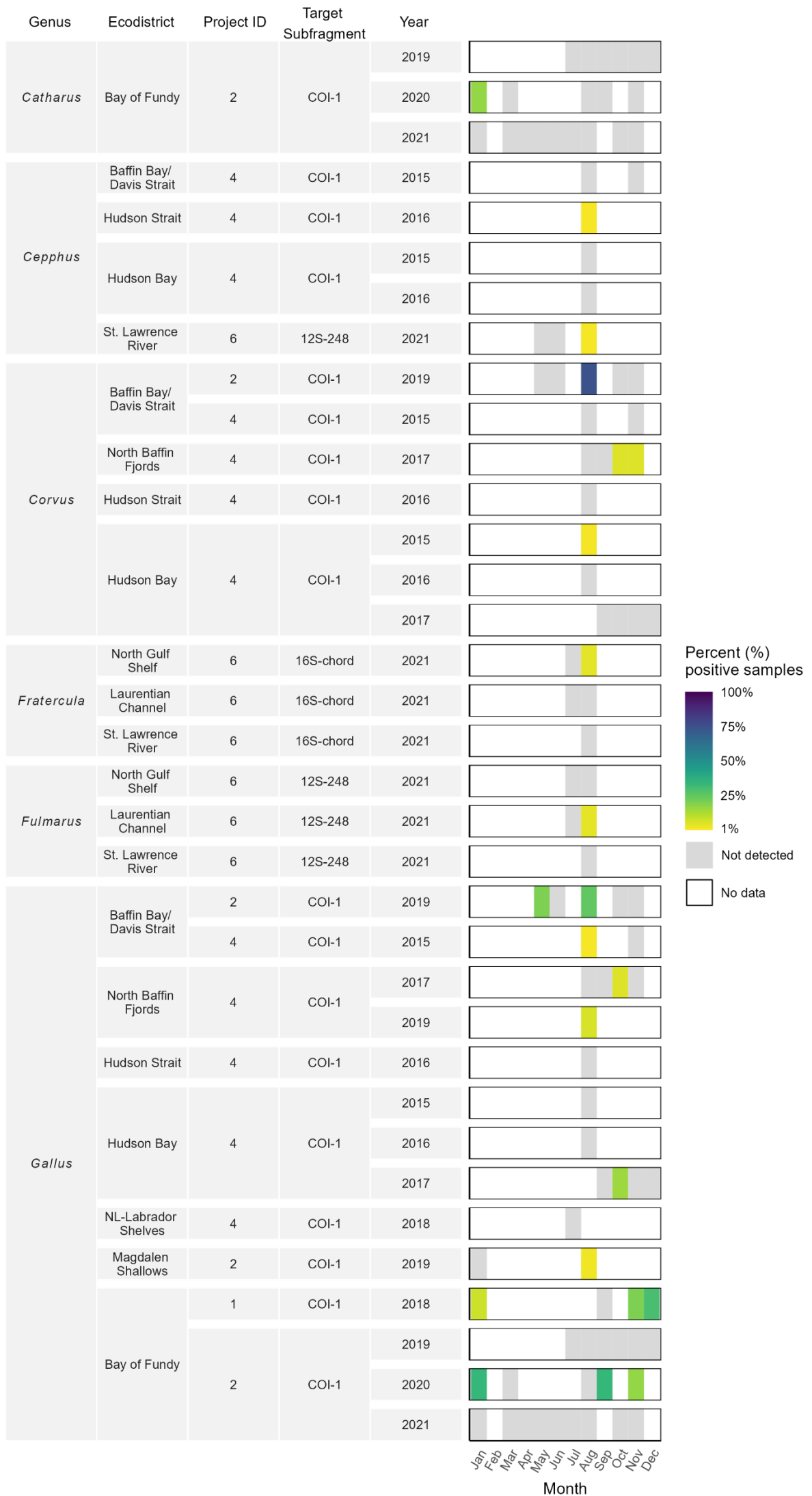


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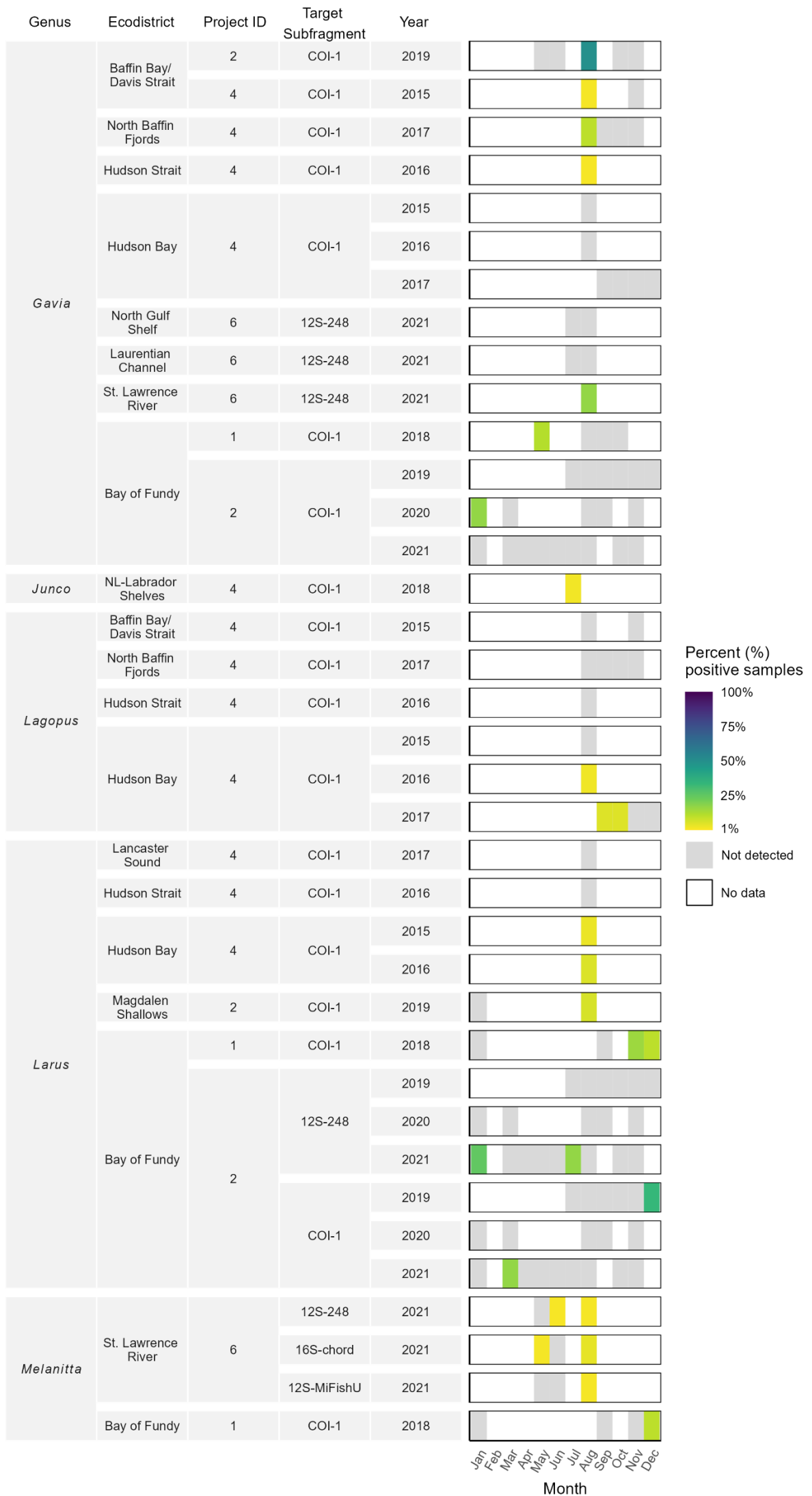


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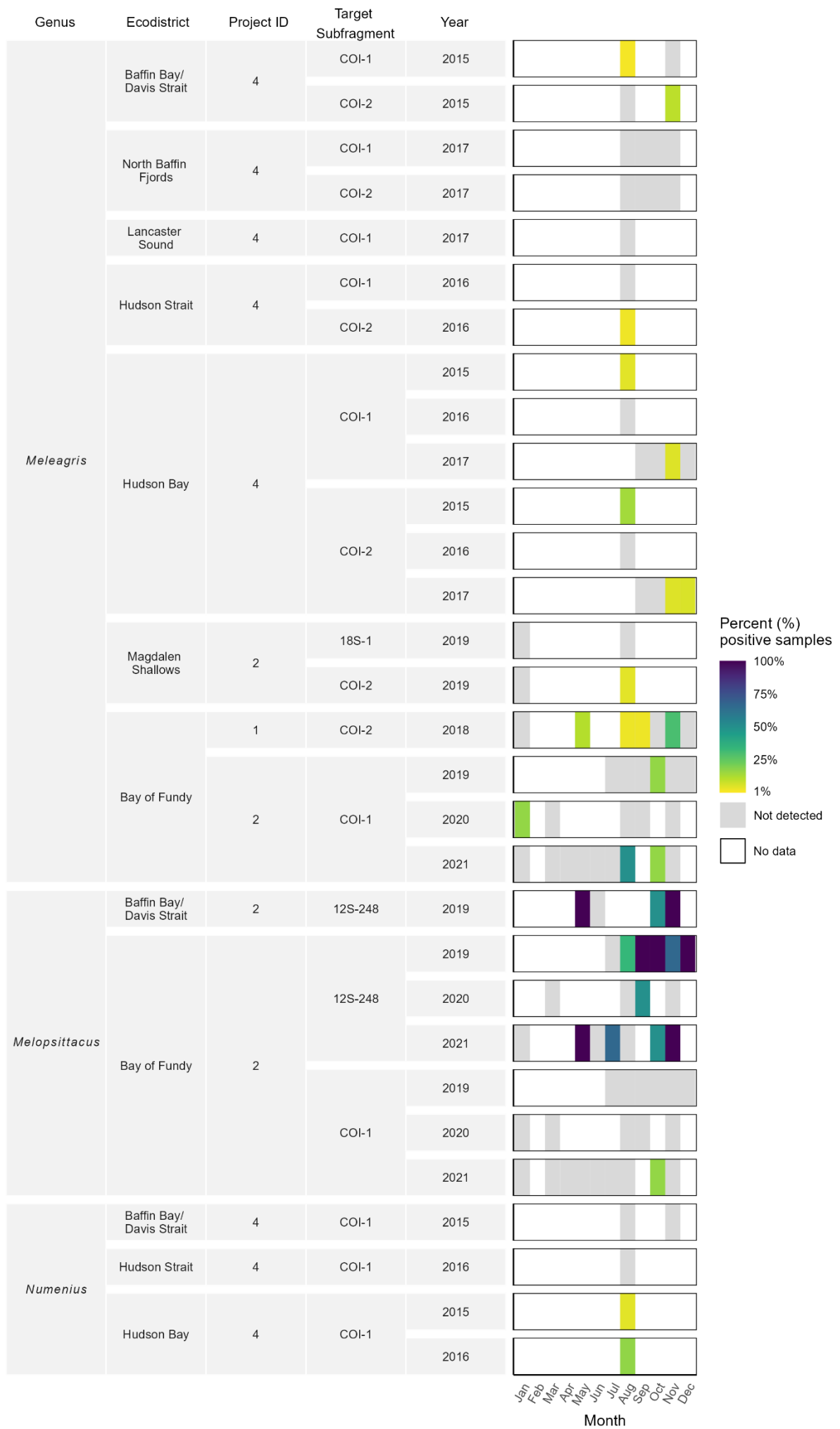


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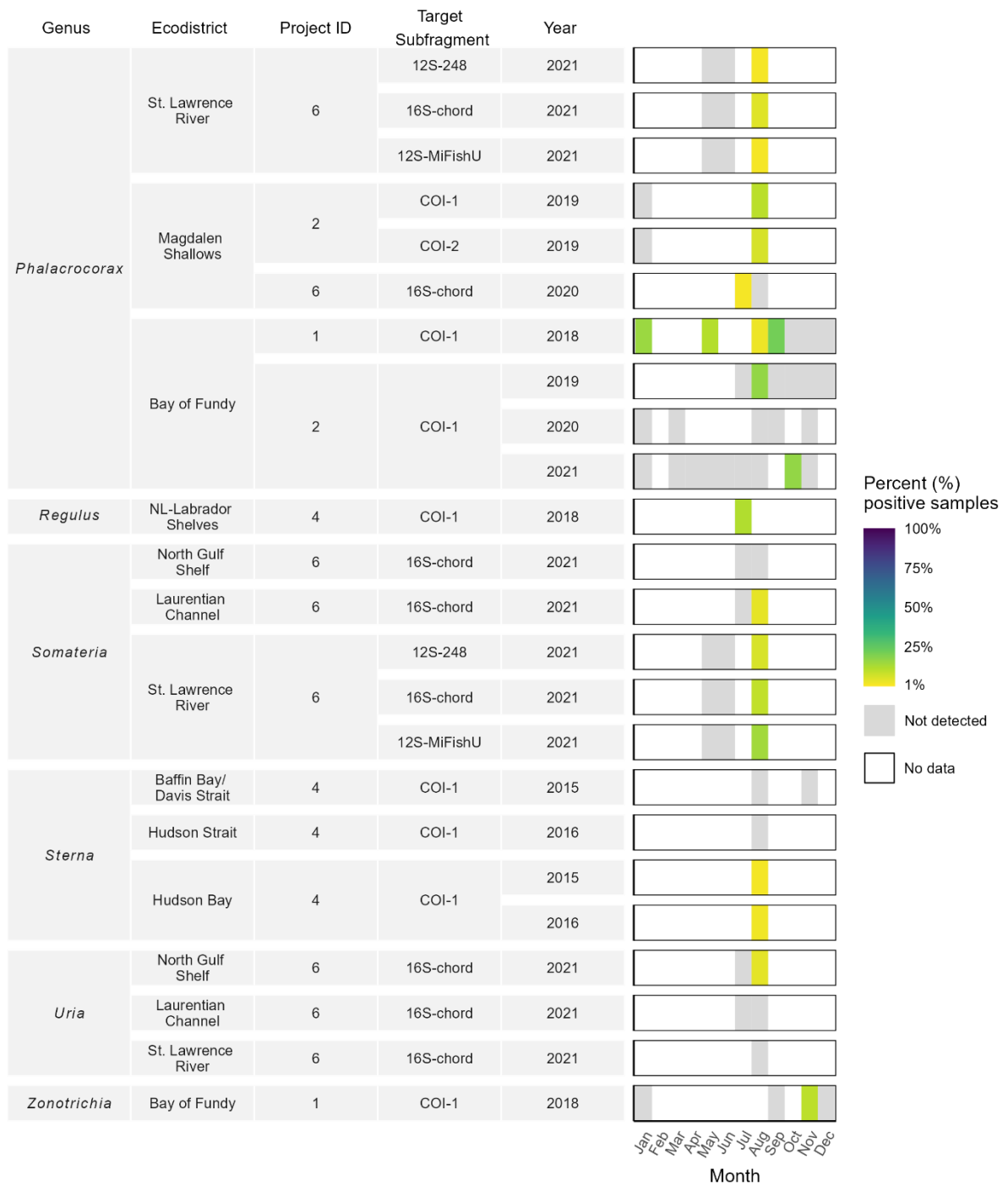


Figure 13. (Continued)

6.5.5 Chondrichthyes

Elasmobranchii



Figure 13. (Continued)

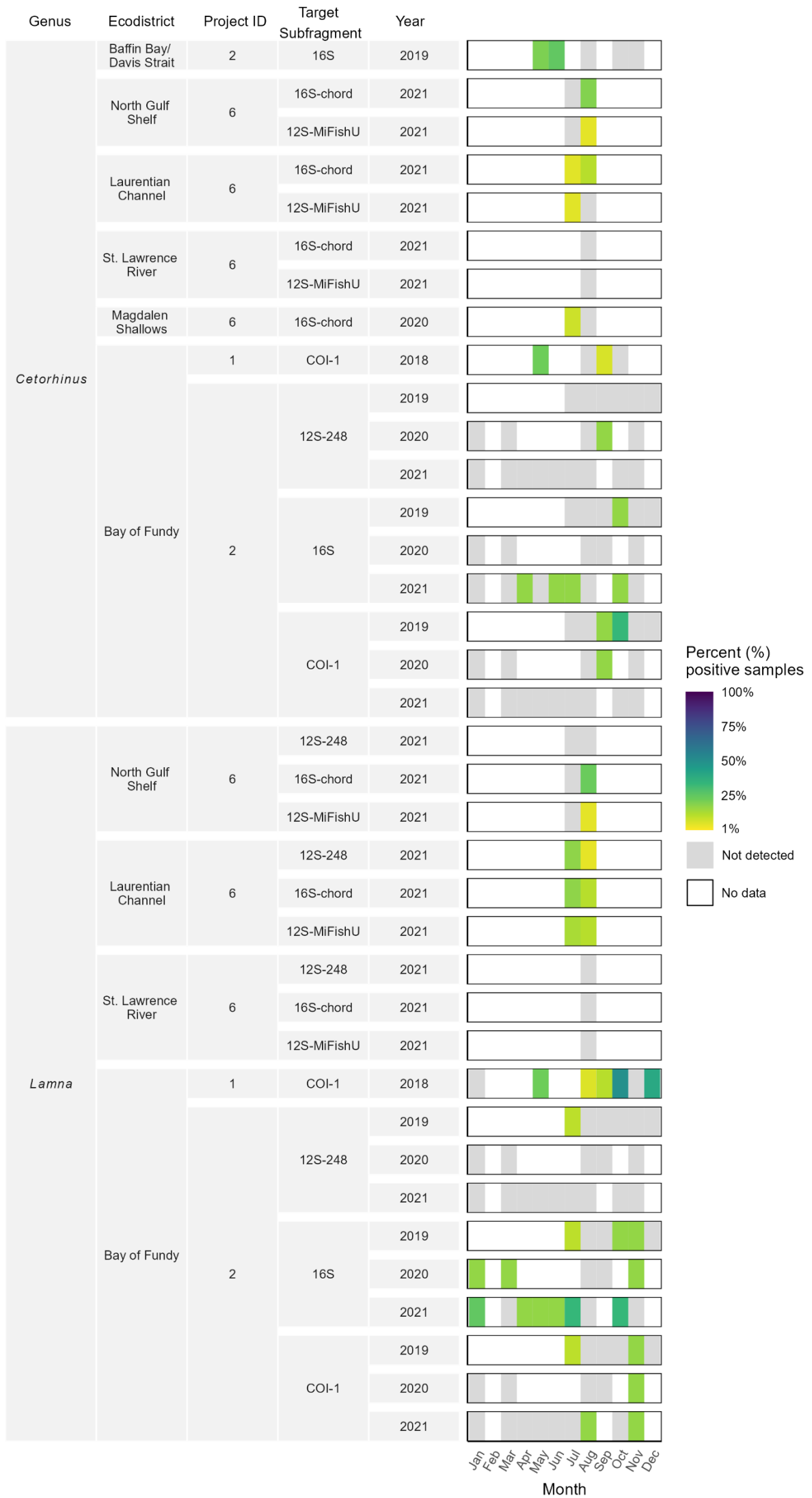


Figure 13. (Continued)

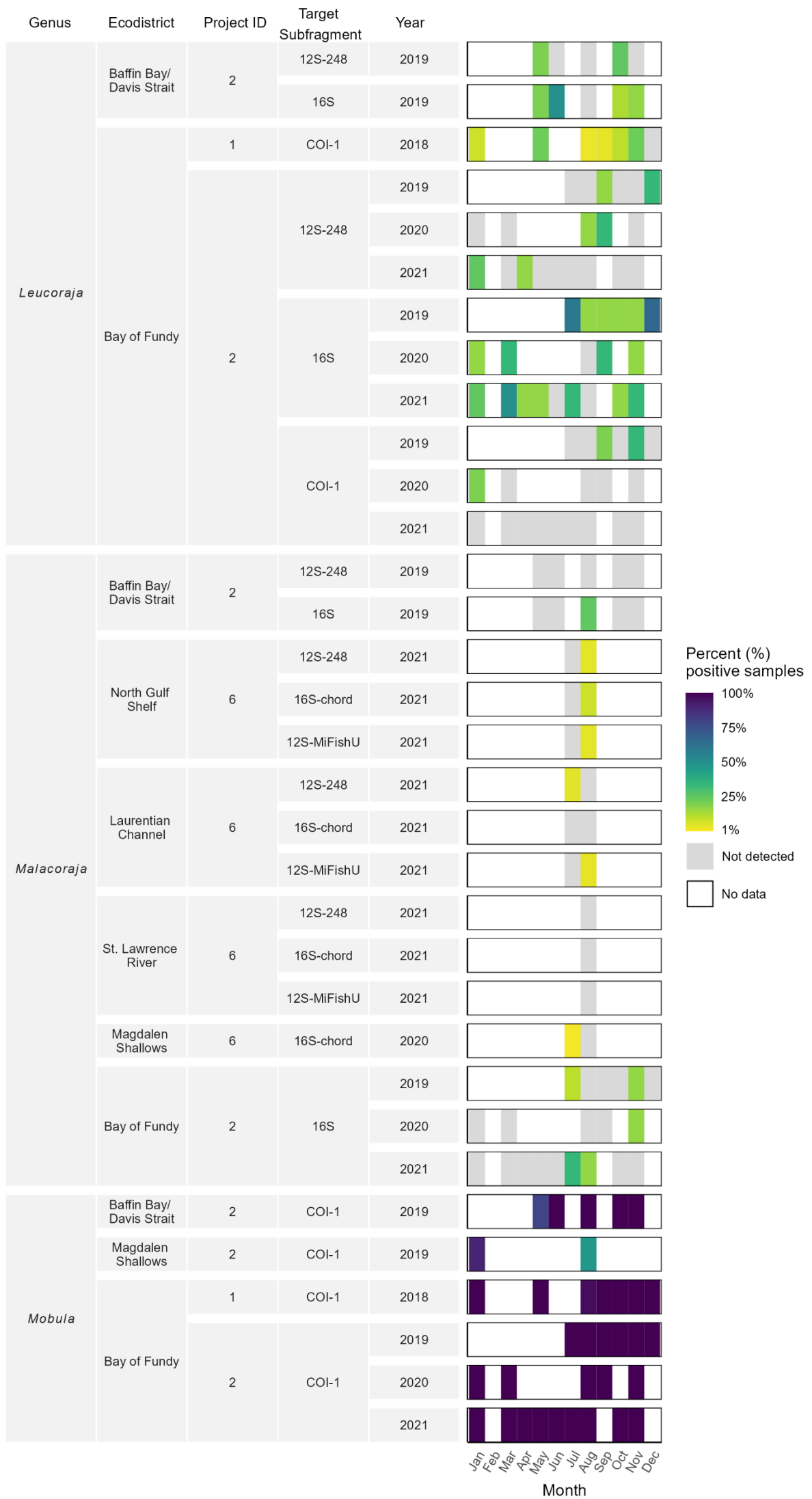


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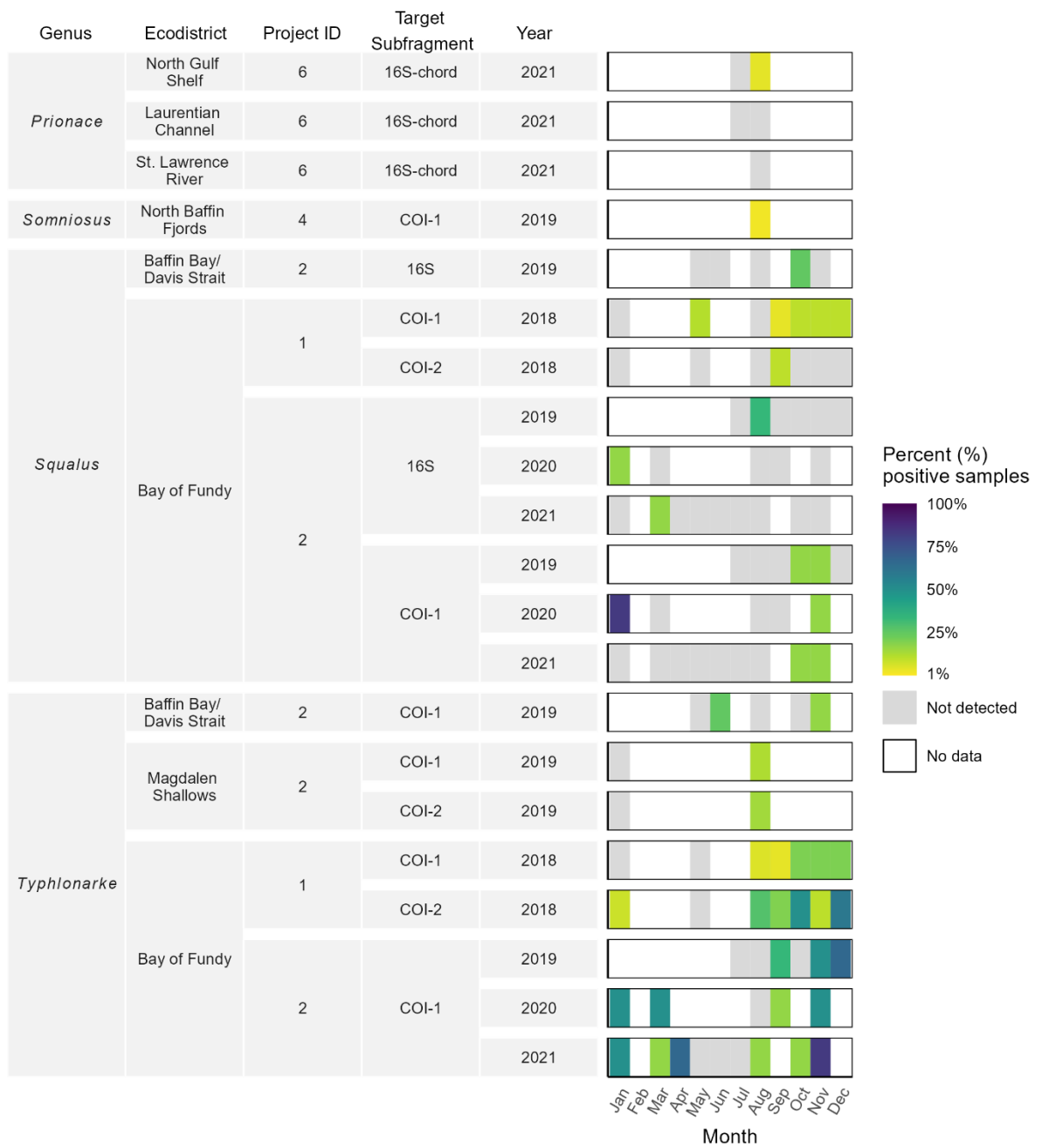


Figure 13. (Continued)

6.5.6 Mammalia

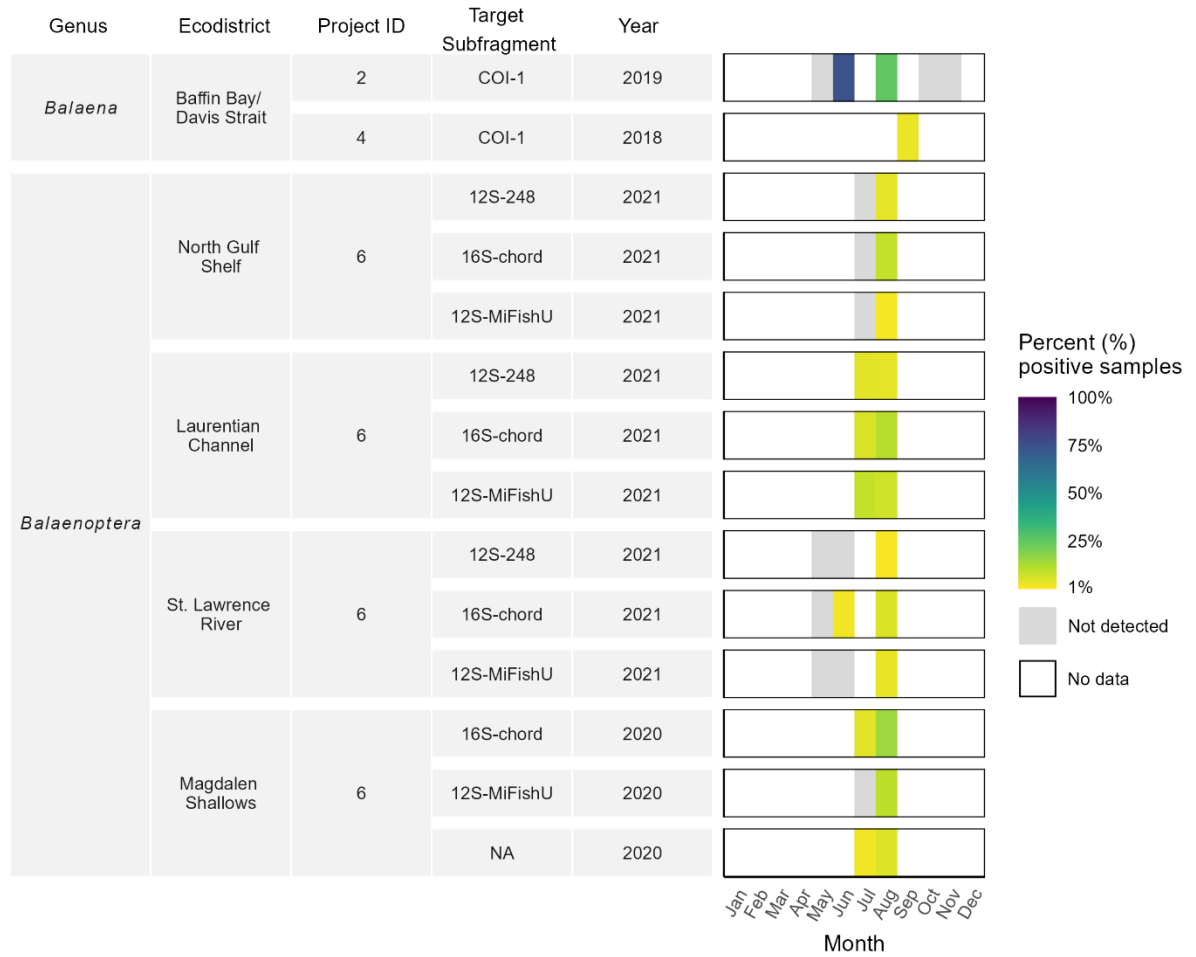


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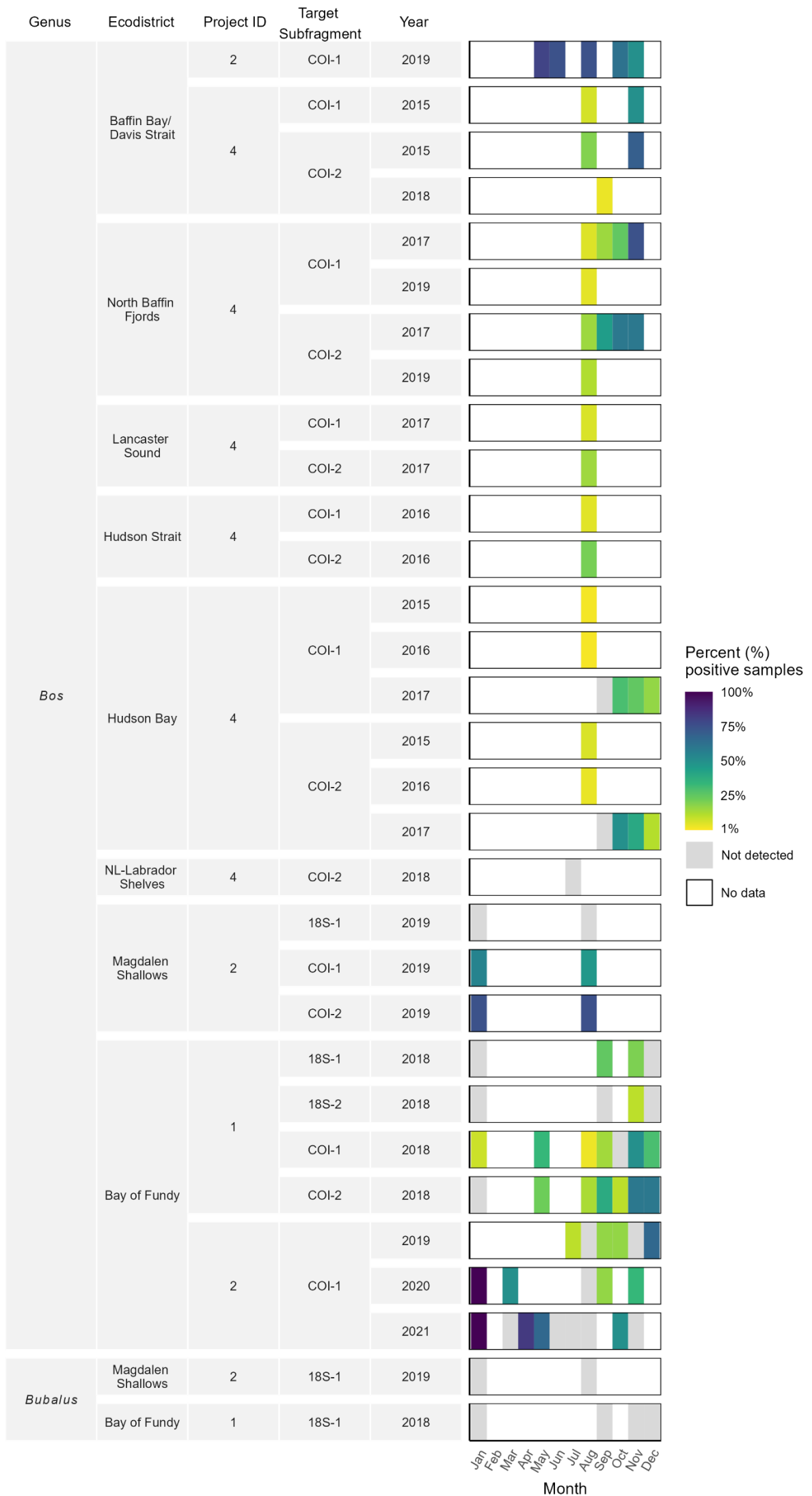


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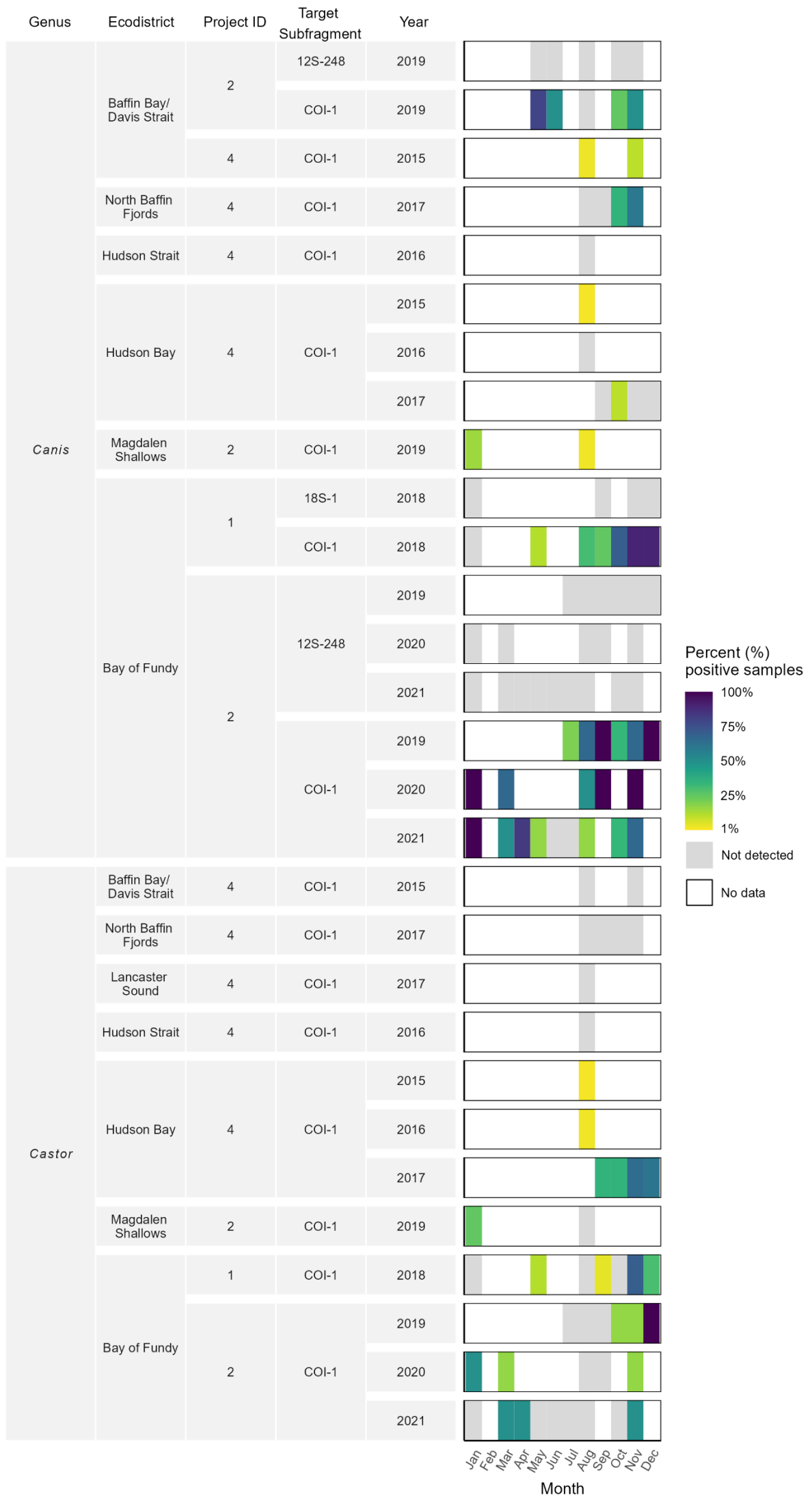


Figure 13. (Continued)



Figure 13. (Continued)

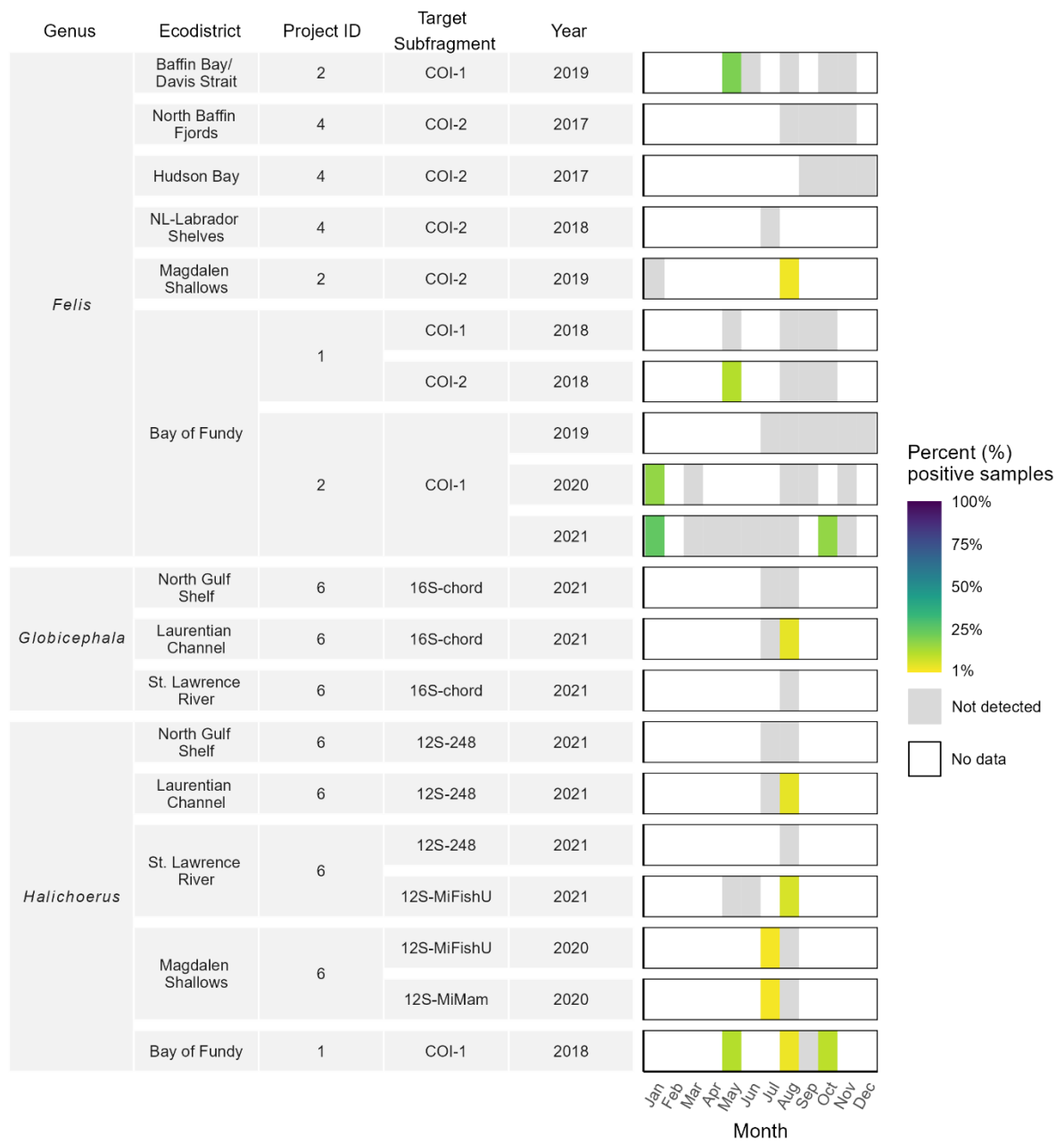


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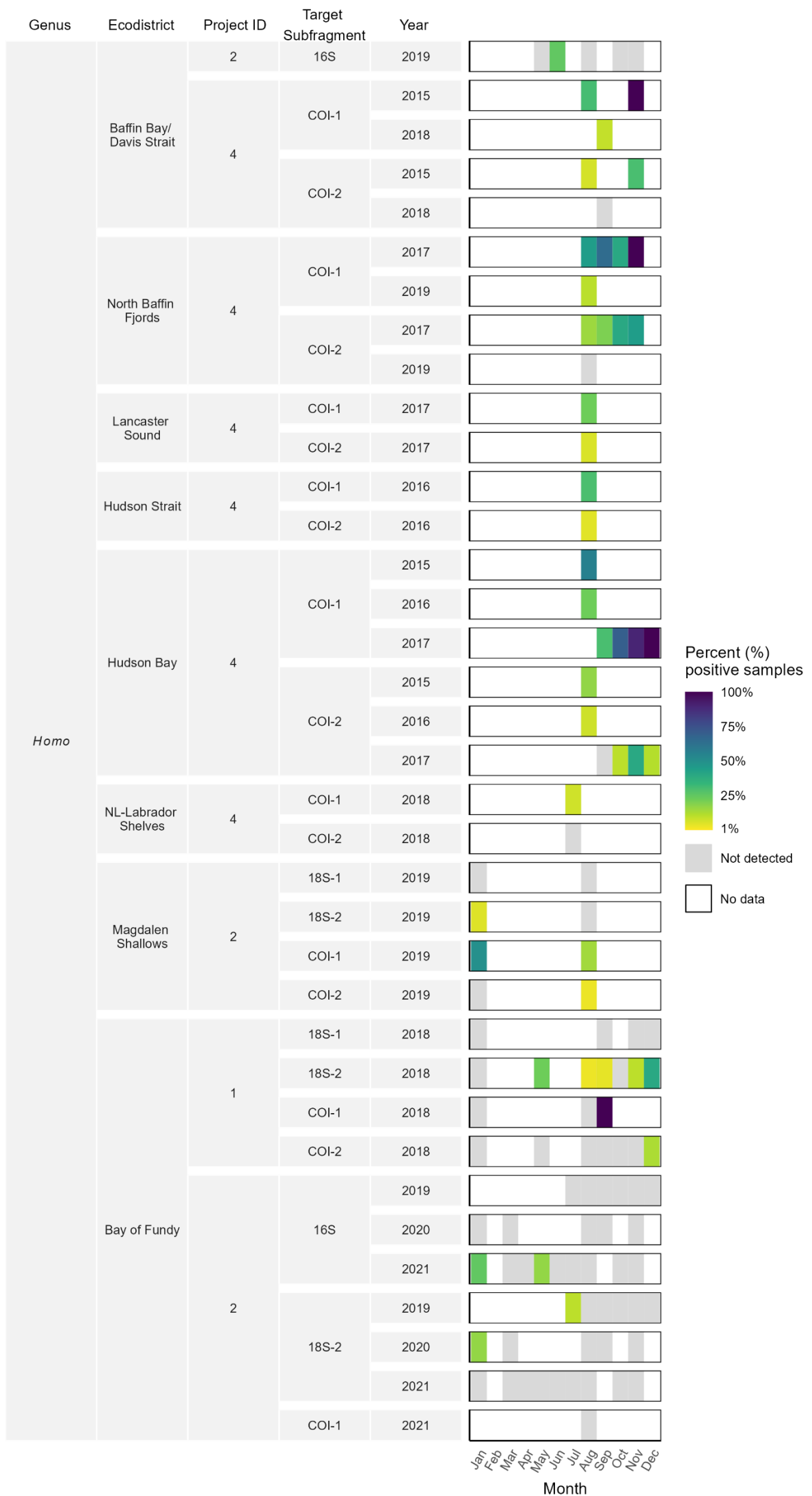


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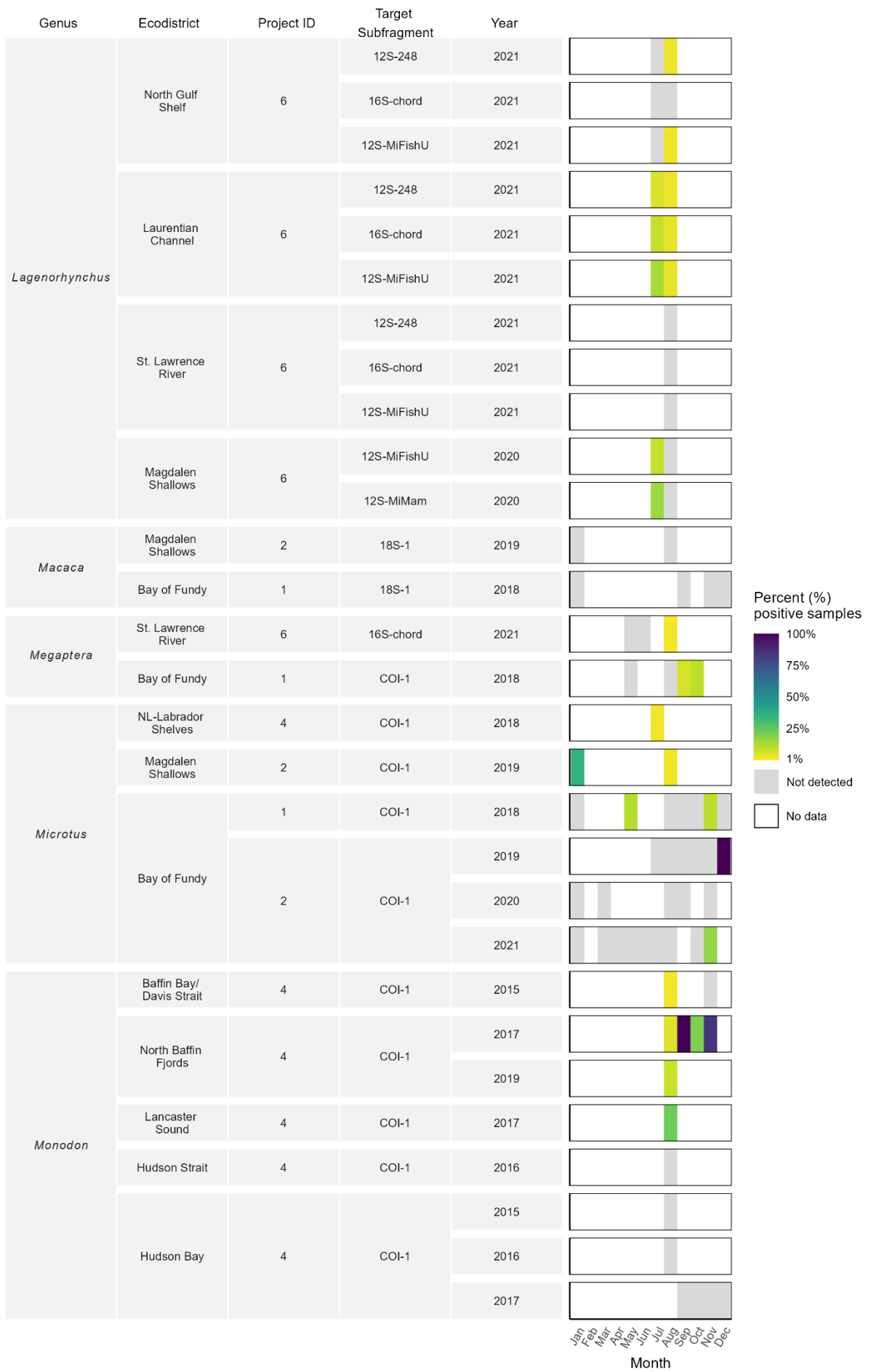


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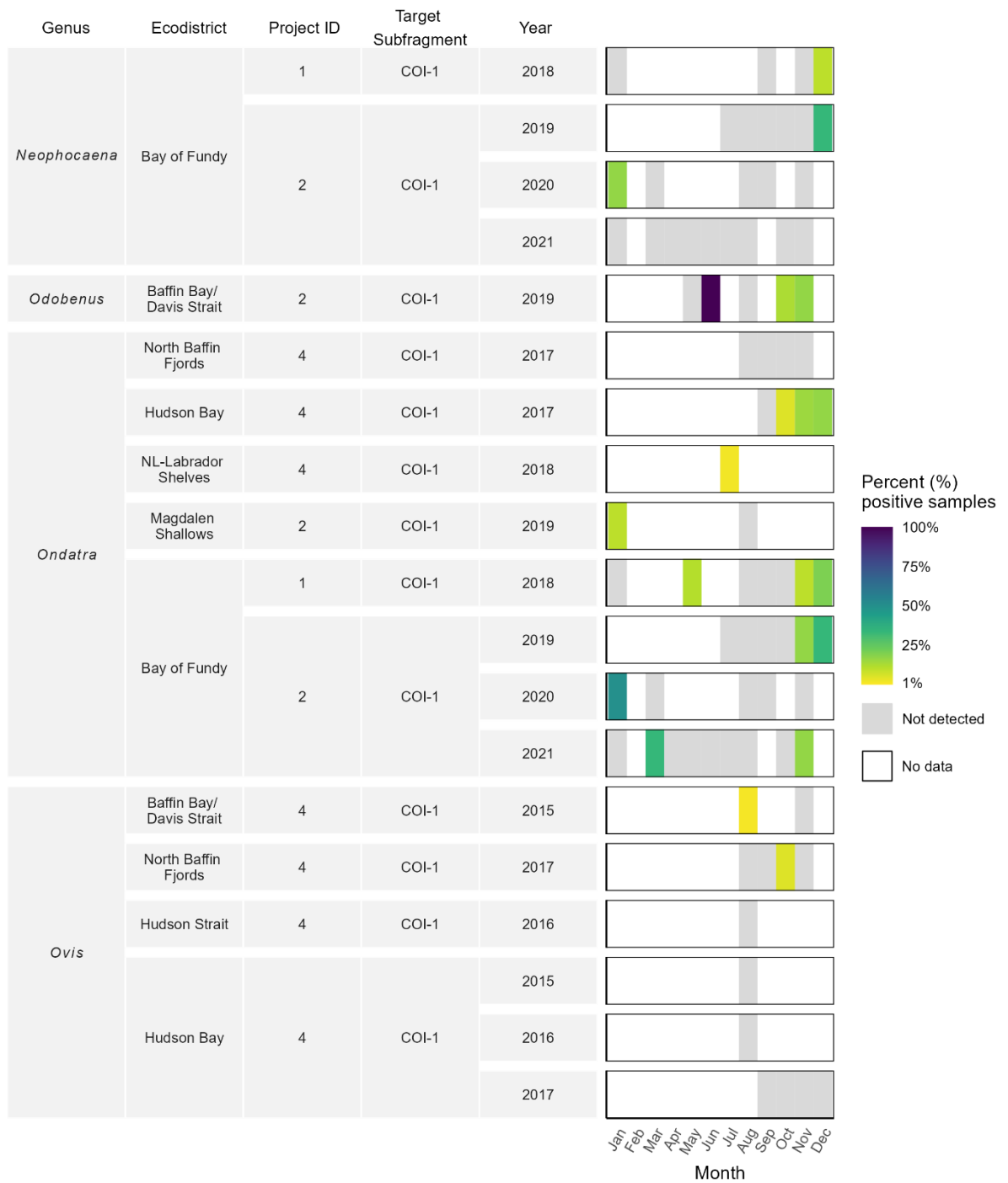


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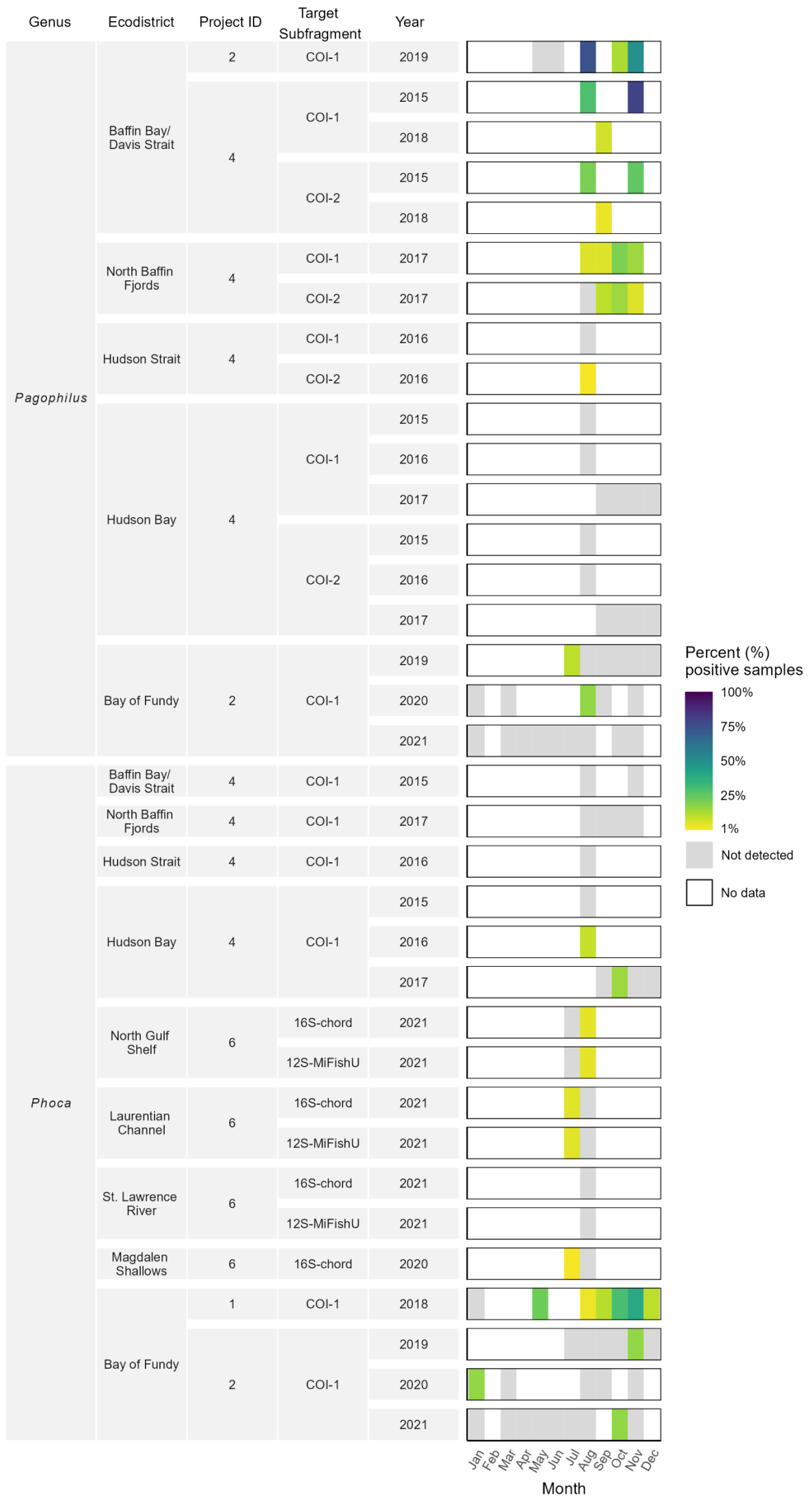


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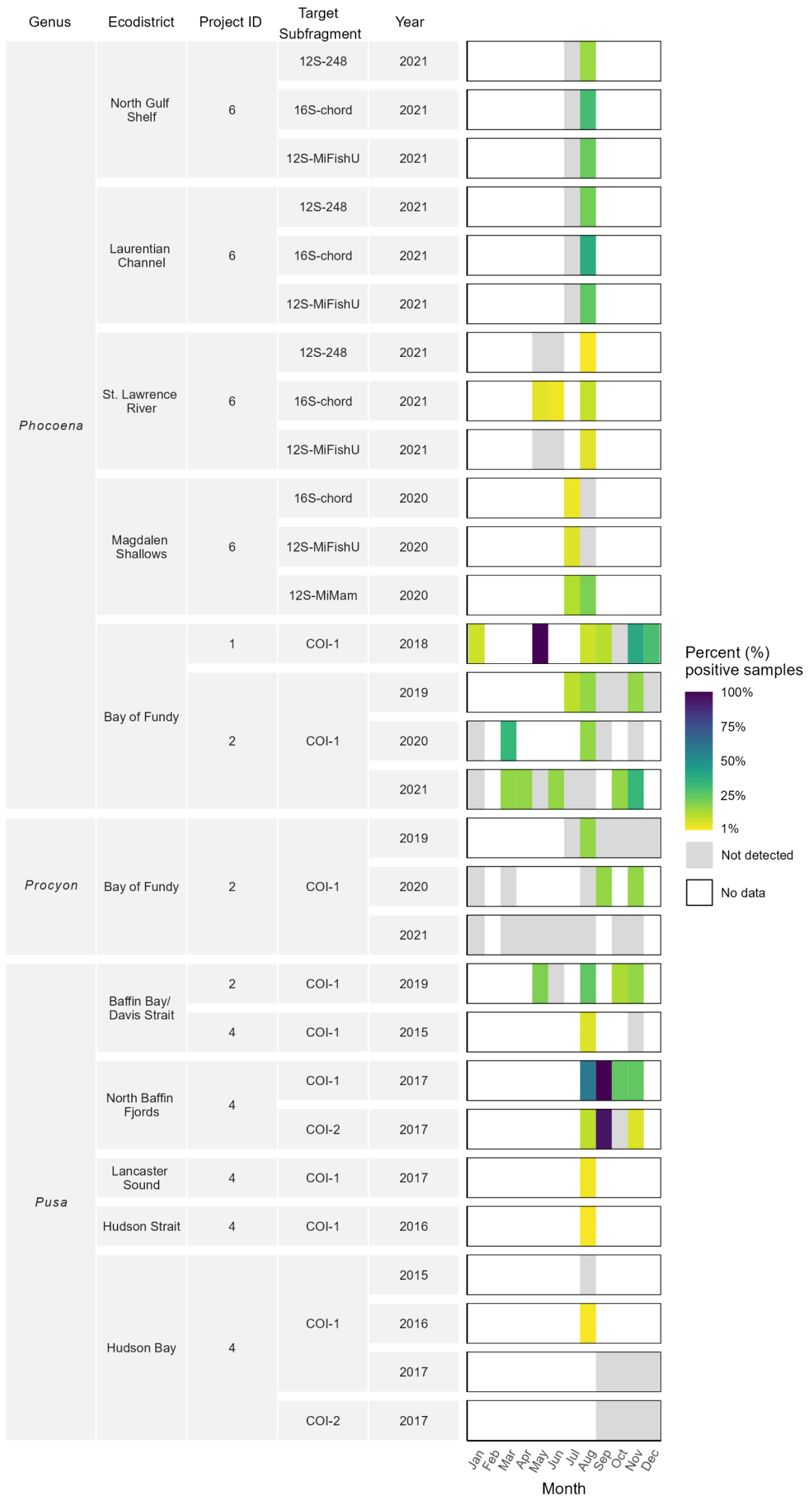


Figure 13. (Continued)



Figure 13. (Continued)

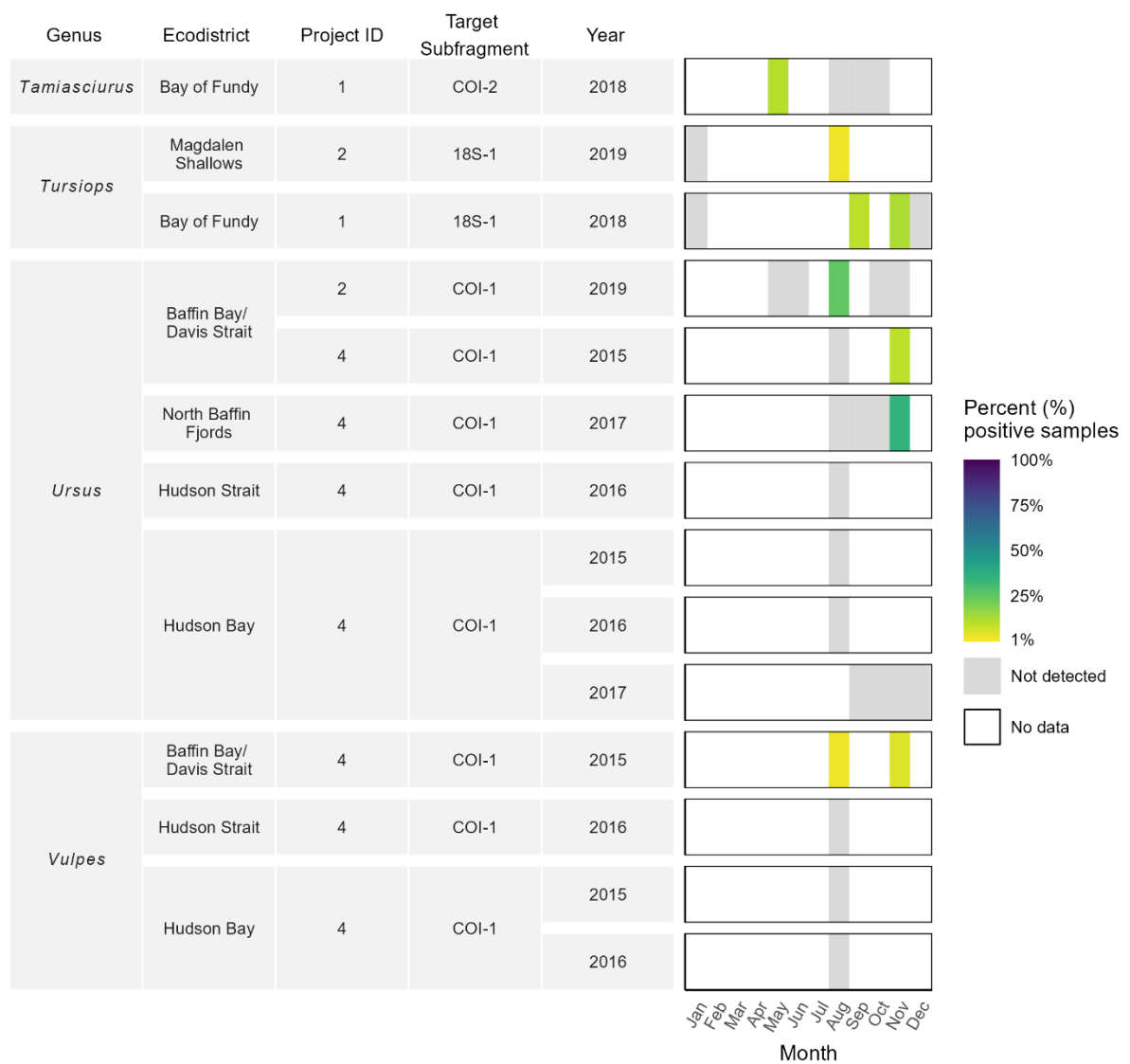


Figure 13. (Continued)

6.5.7 Myxini



6.5.8 Petromyzonti

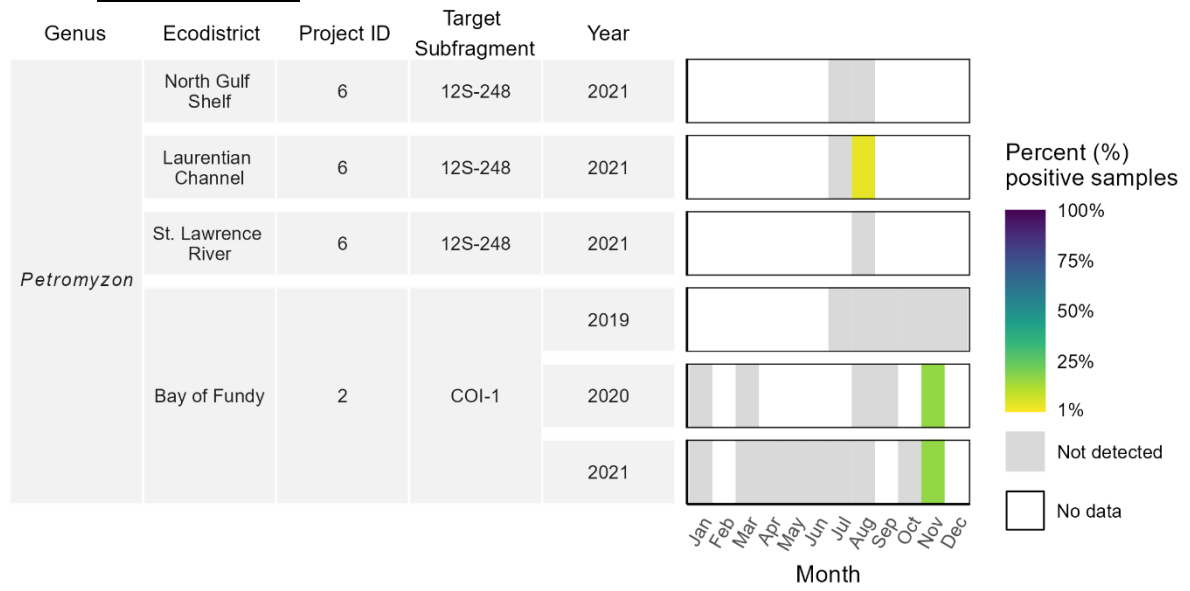


Figure 13. (Continued)

6.6 CNIDARIA

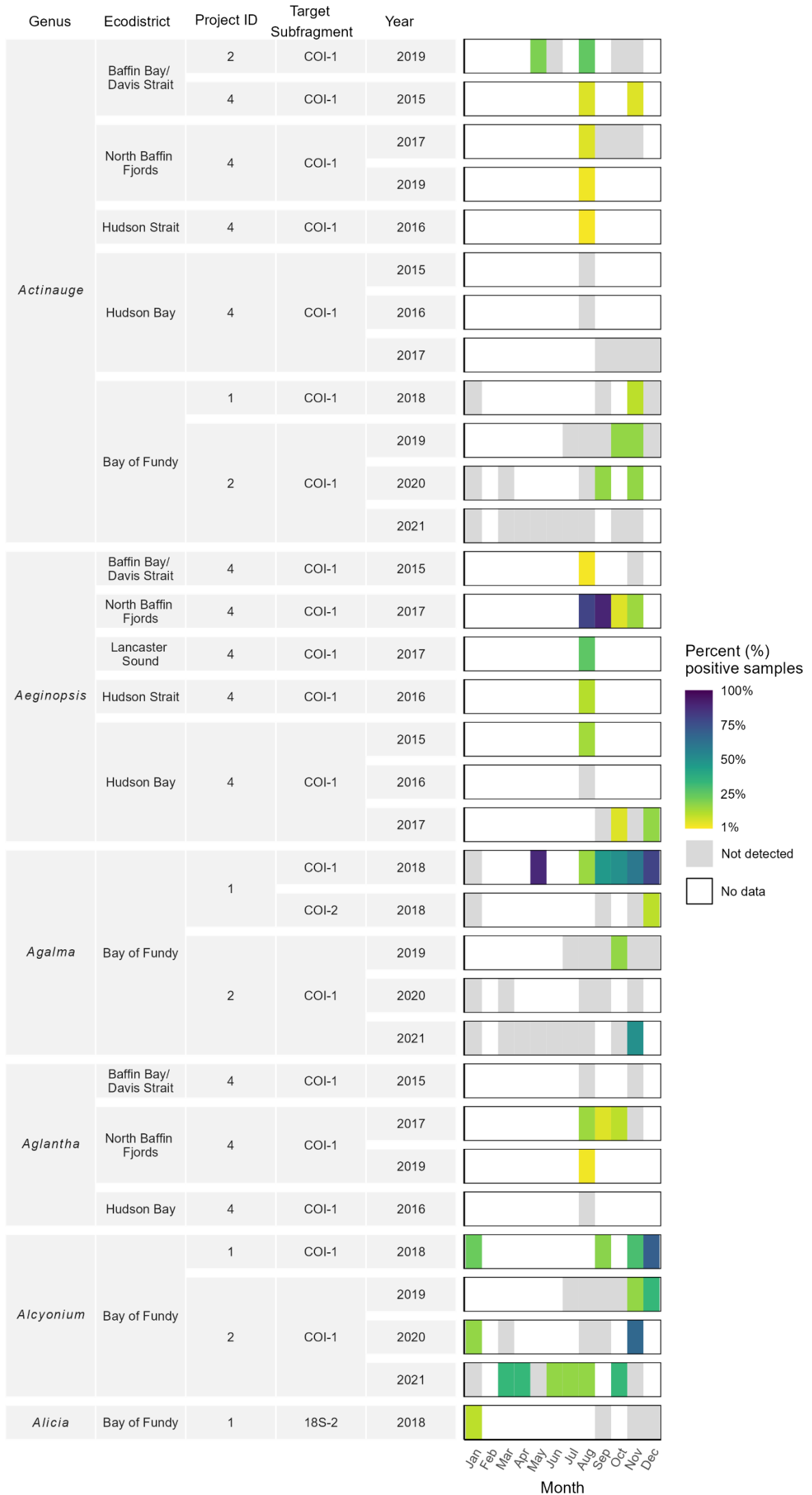


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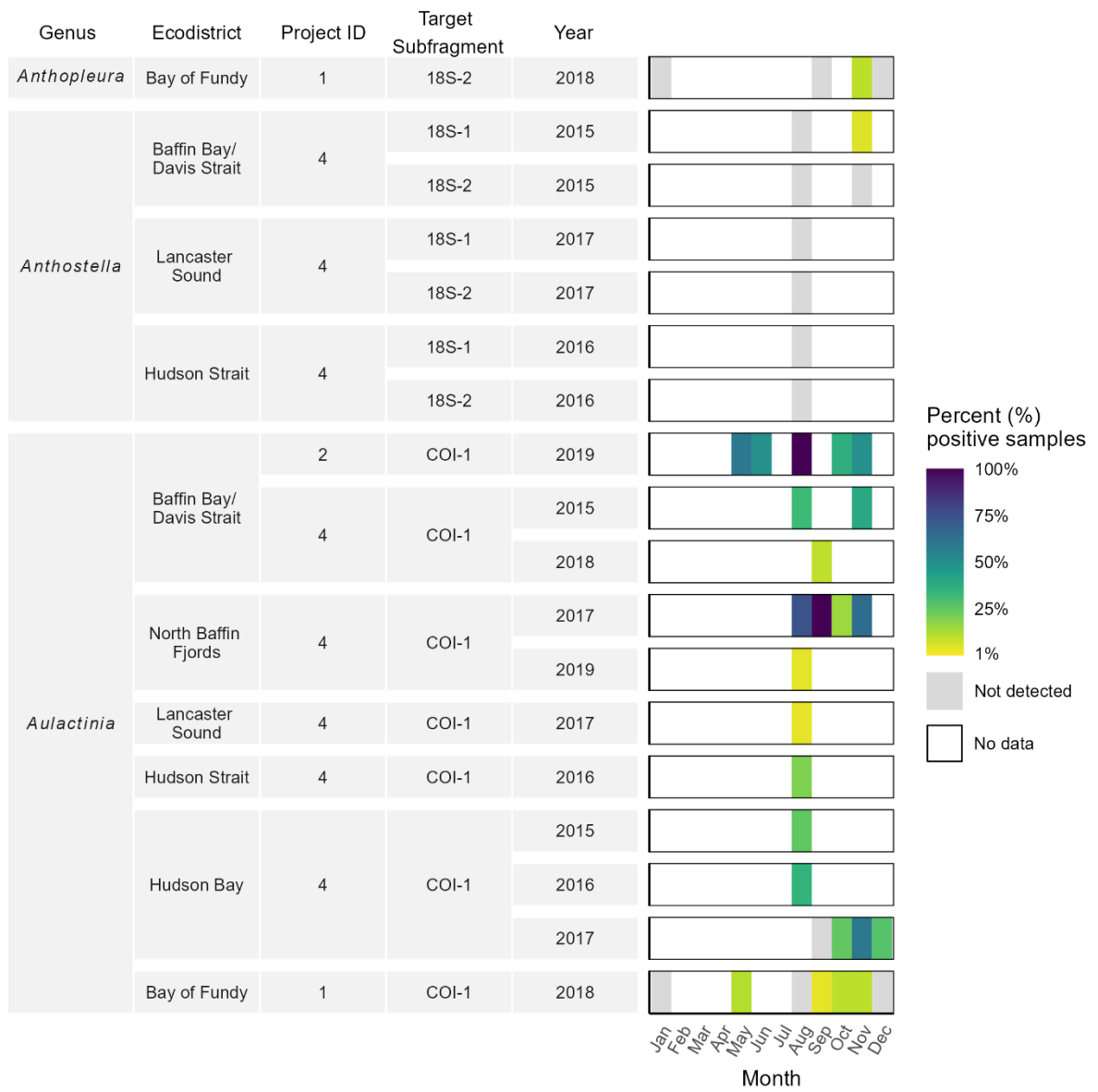


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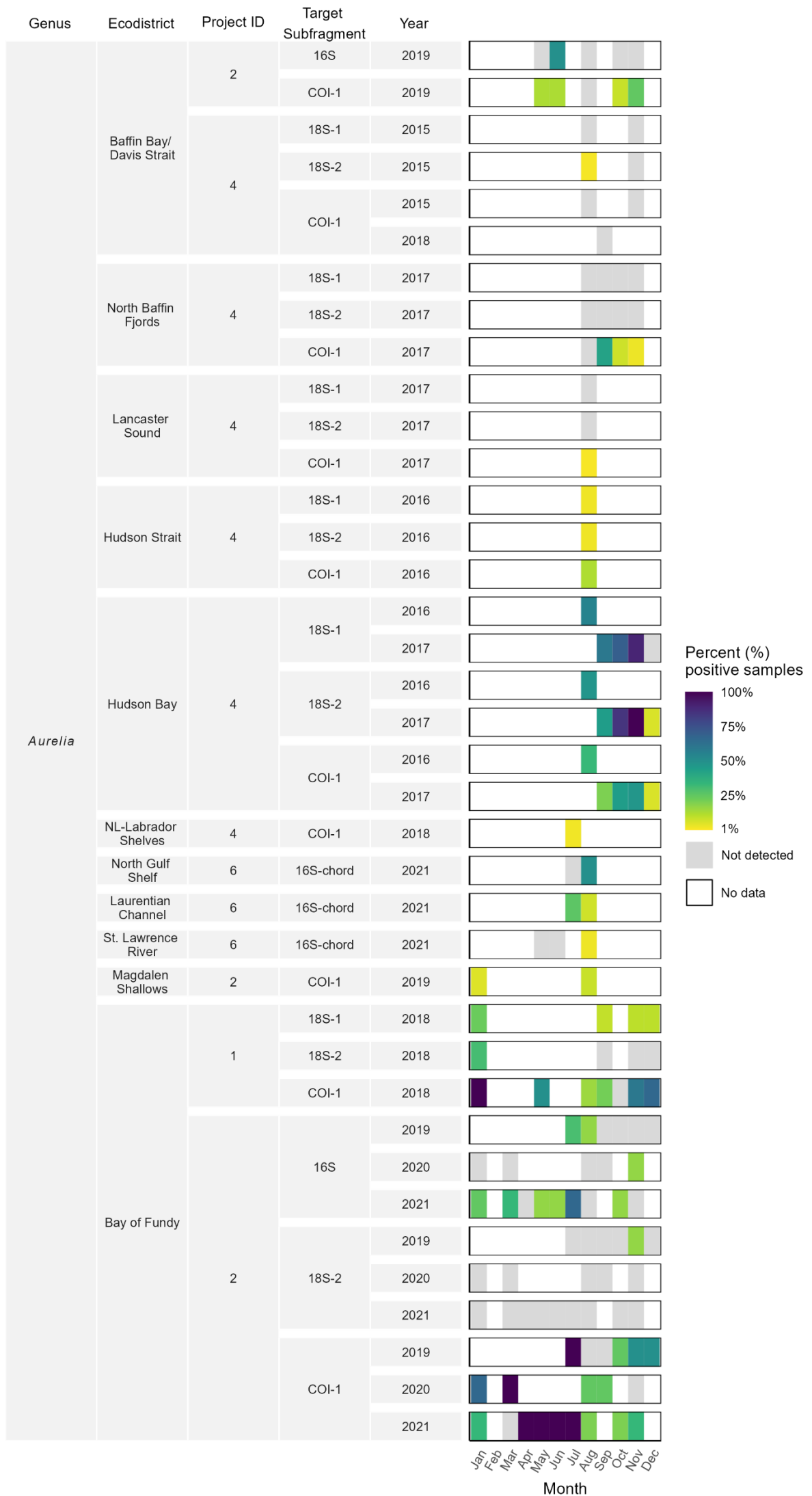


Figure 13. (Continued)



Figure 13. (Continued)



Figure 13. (Continued)

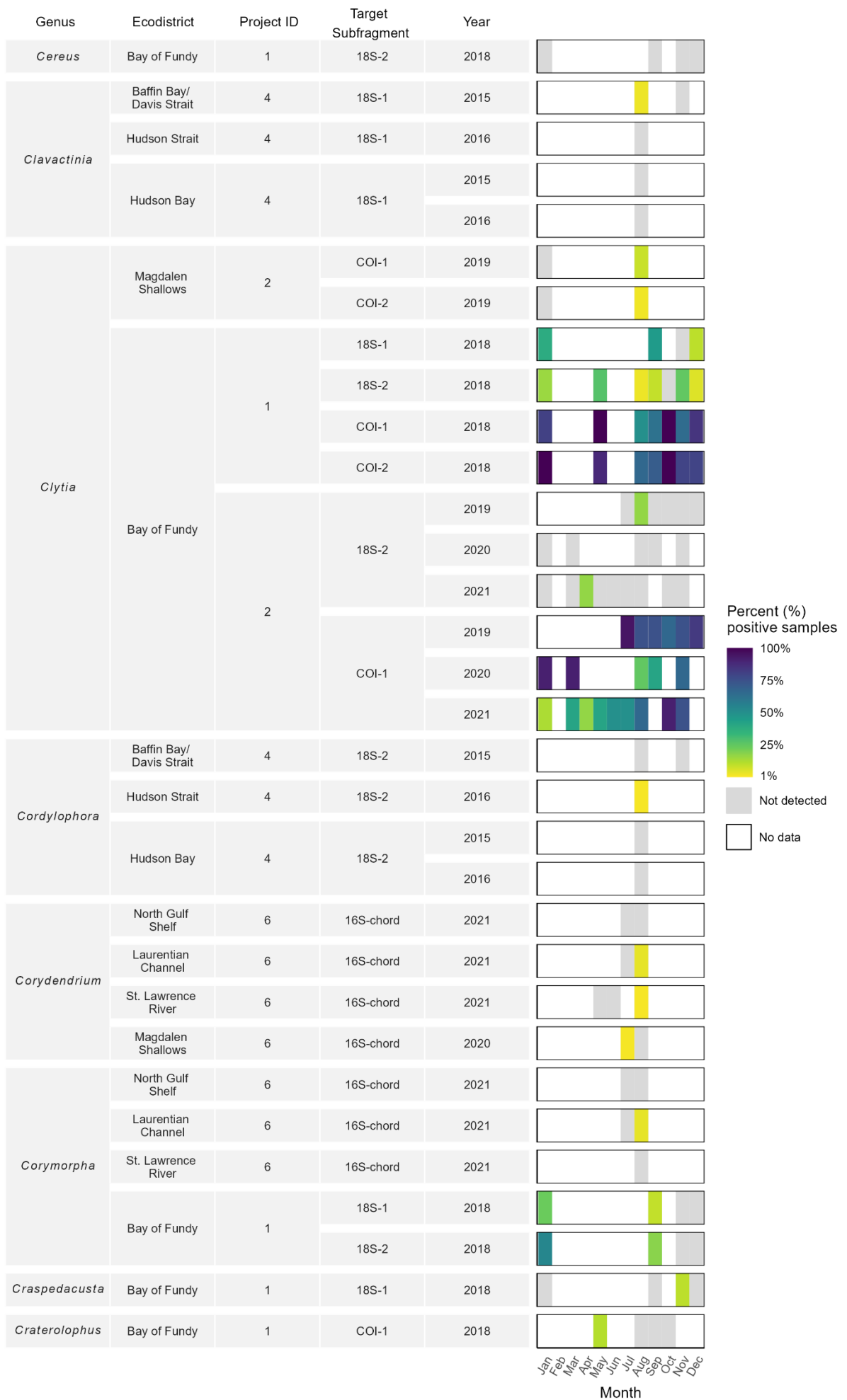


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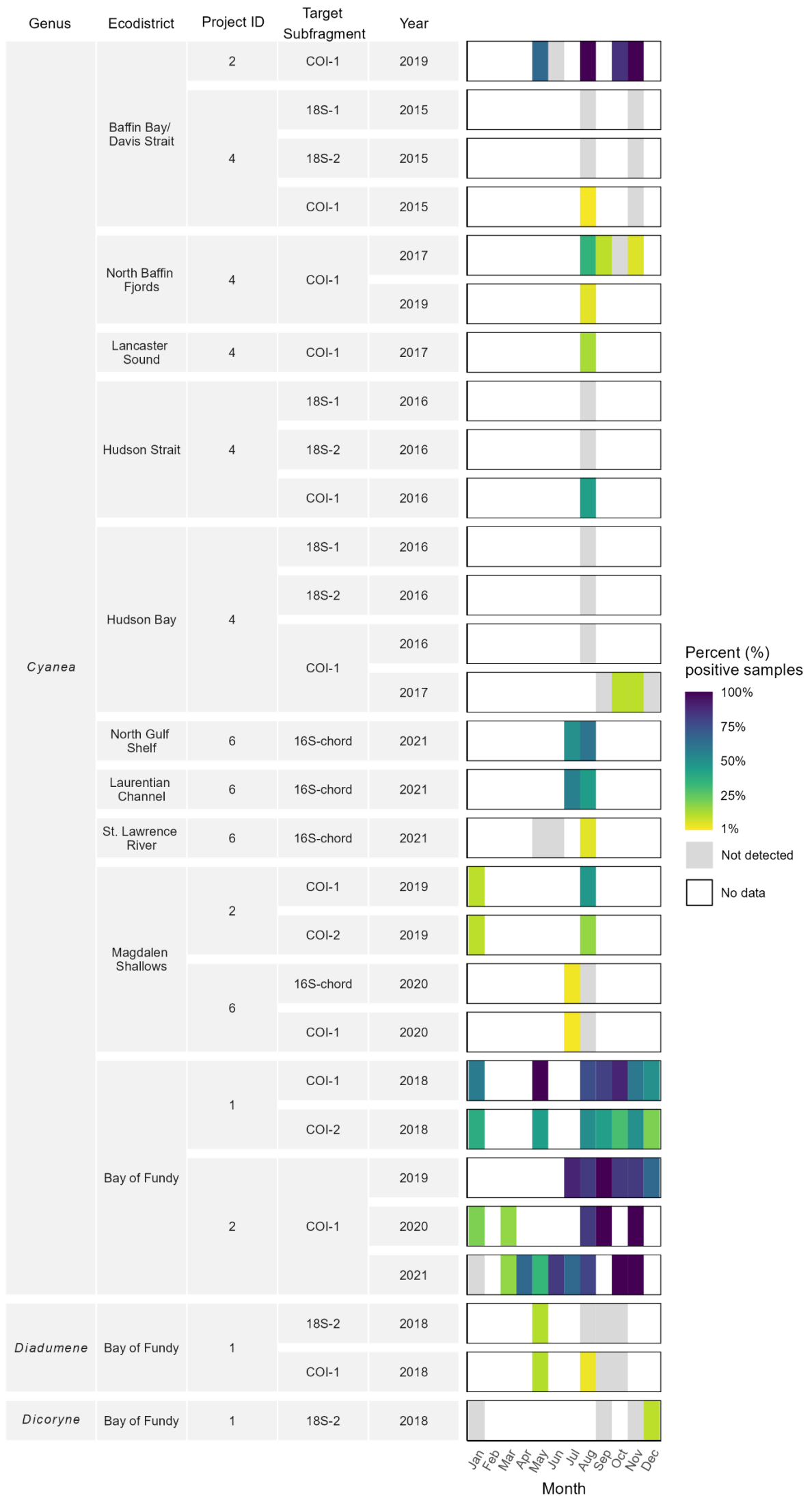


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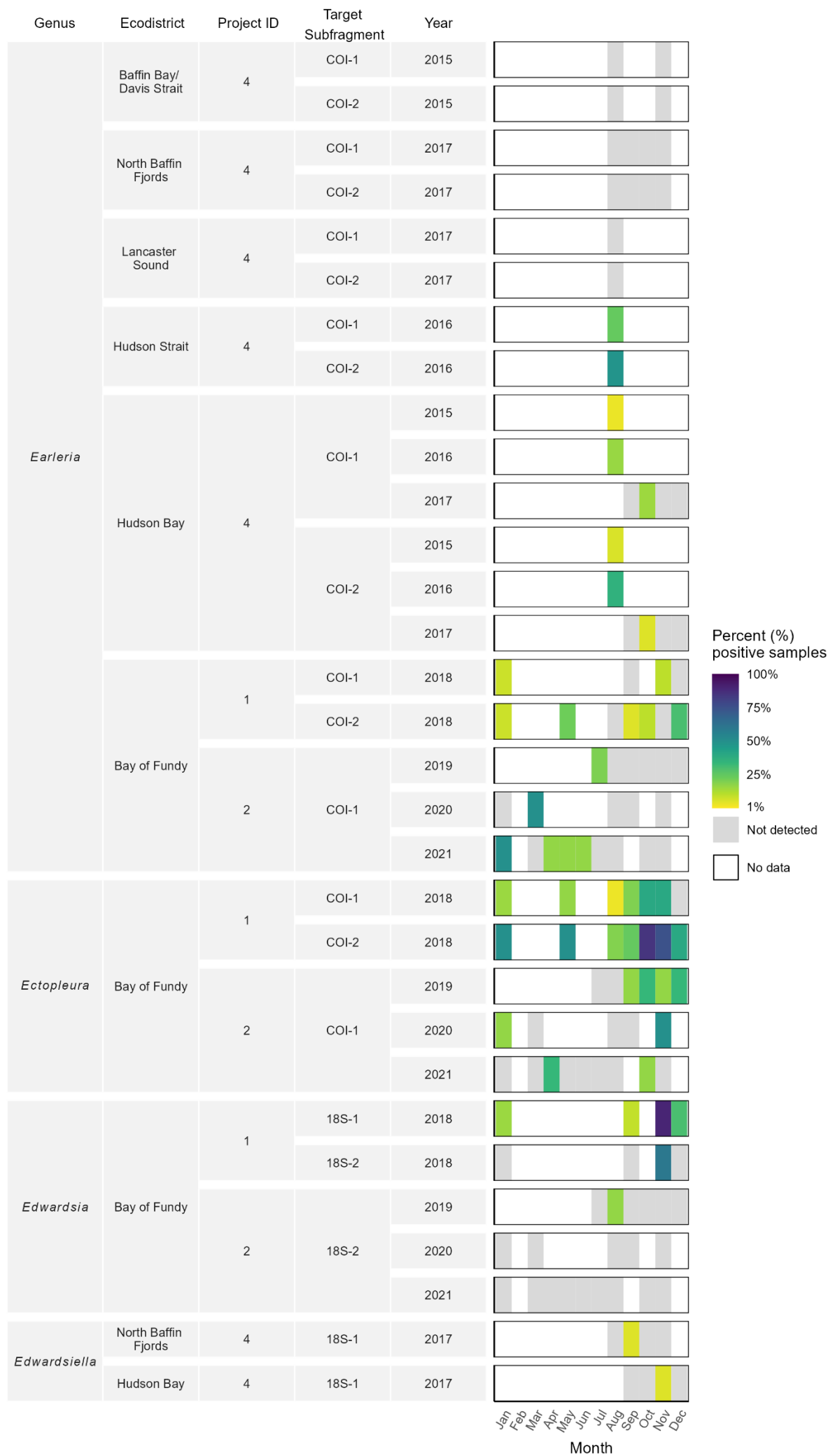


Figure 13. (Continued)



Figure 13. (Continued)

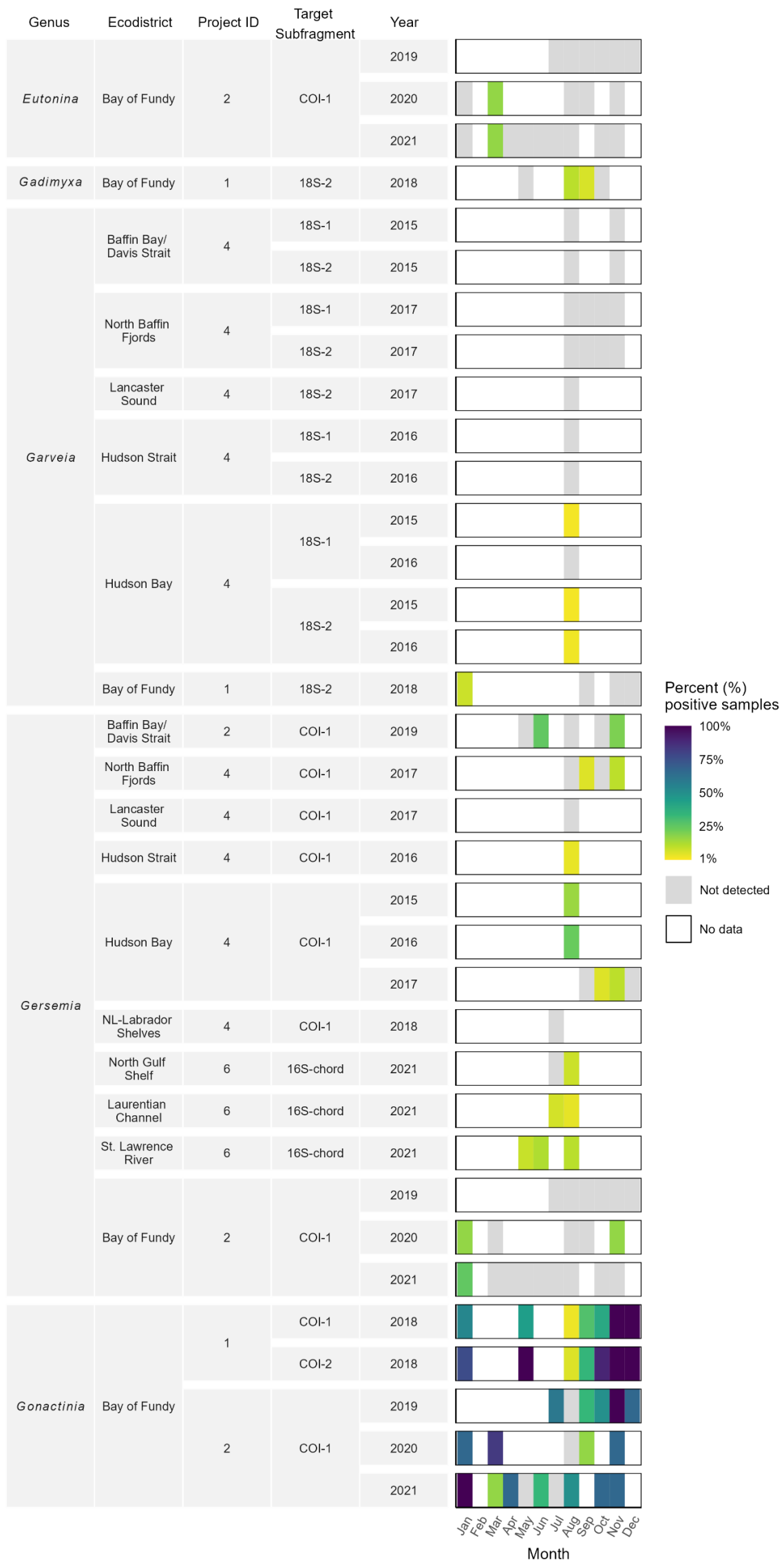


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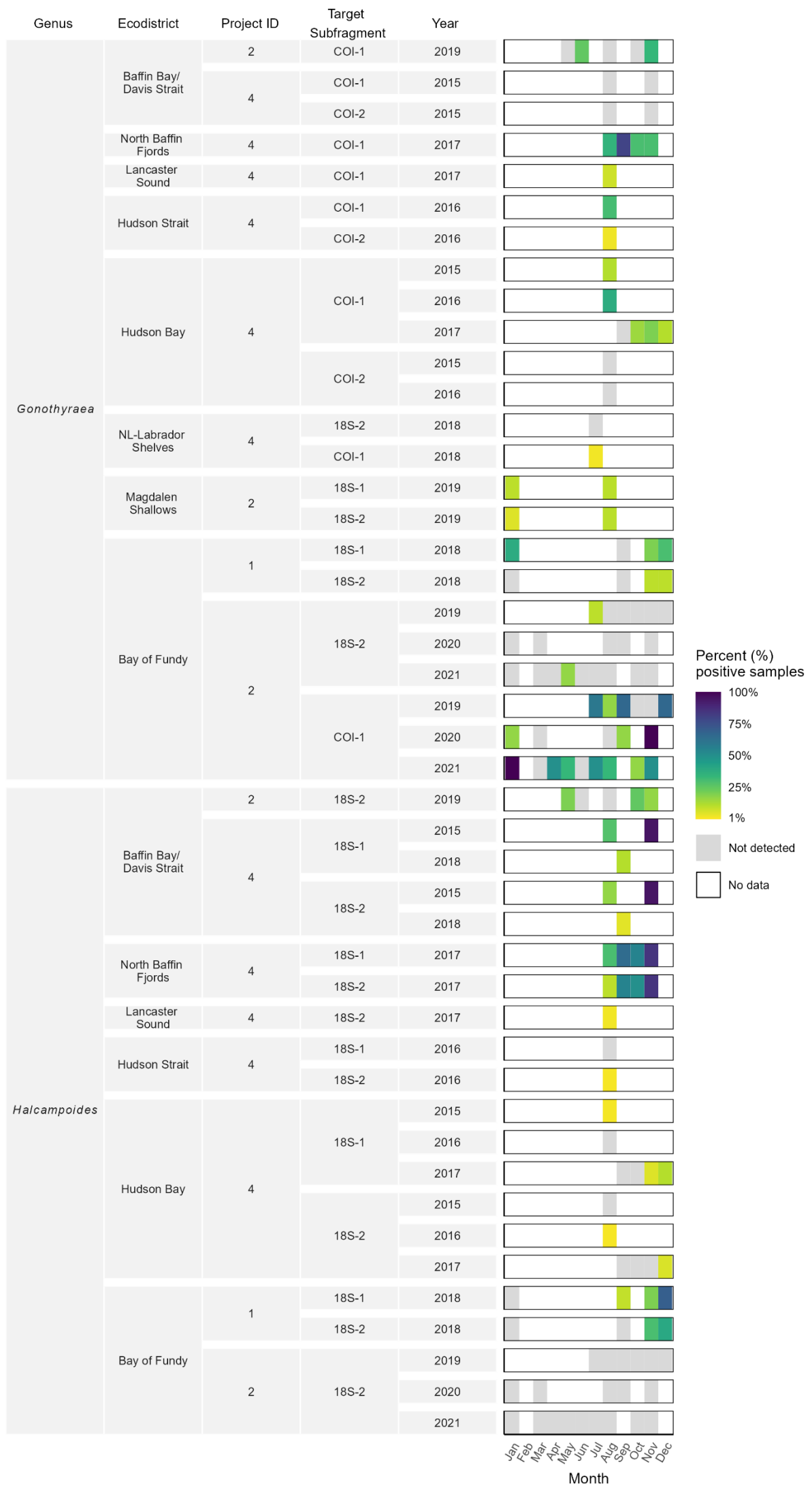


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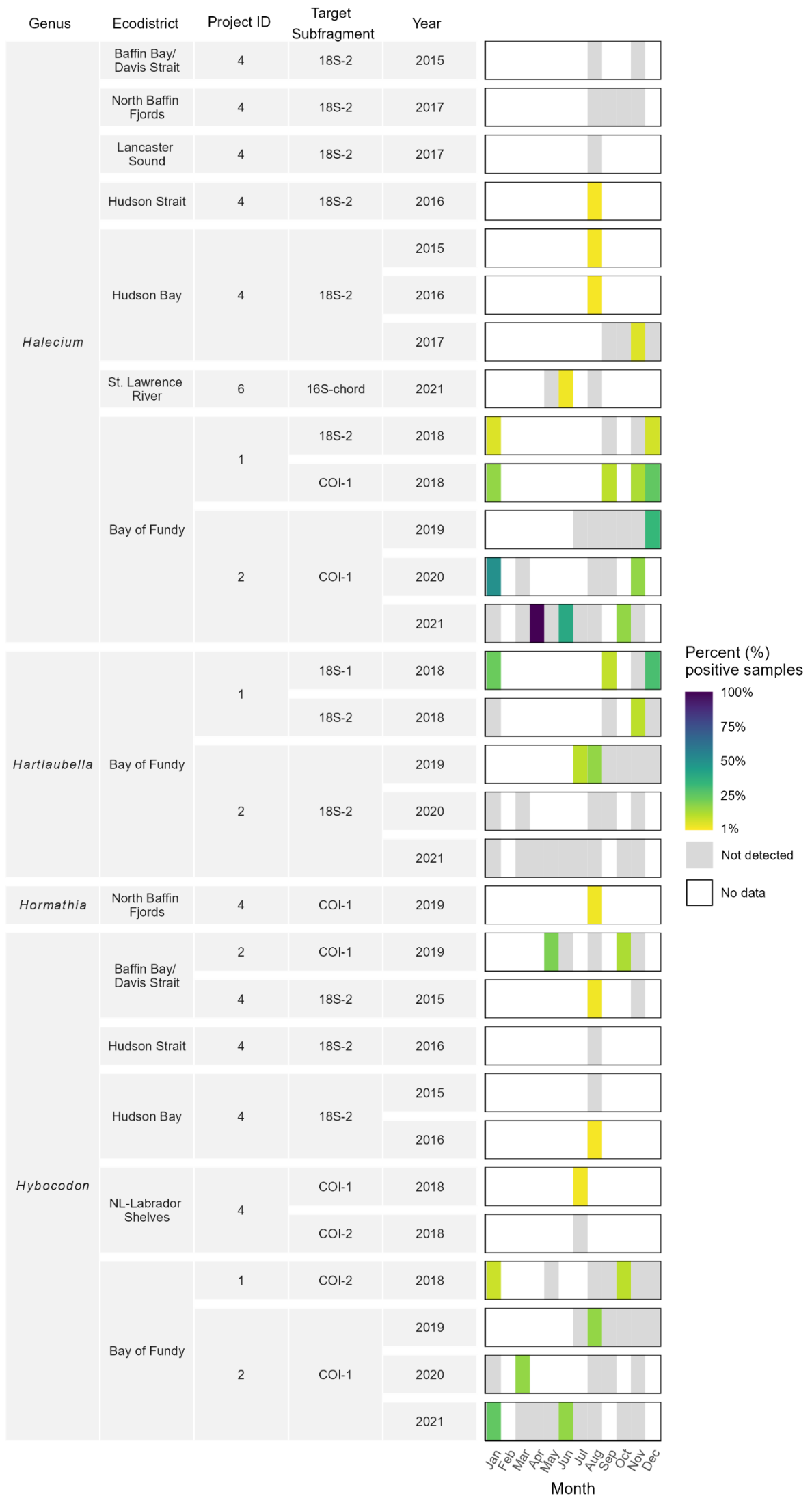


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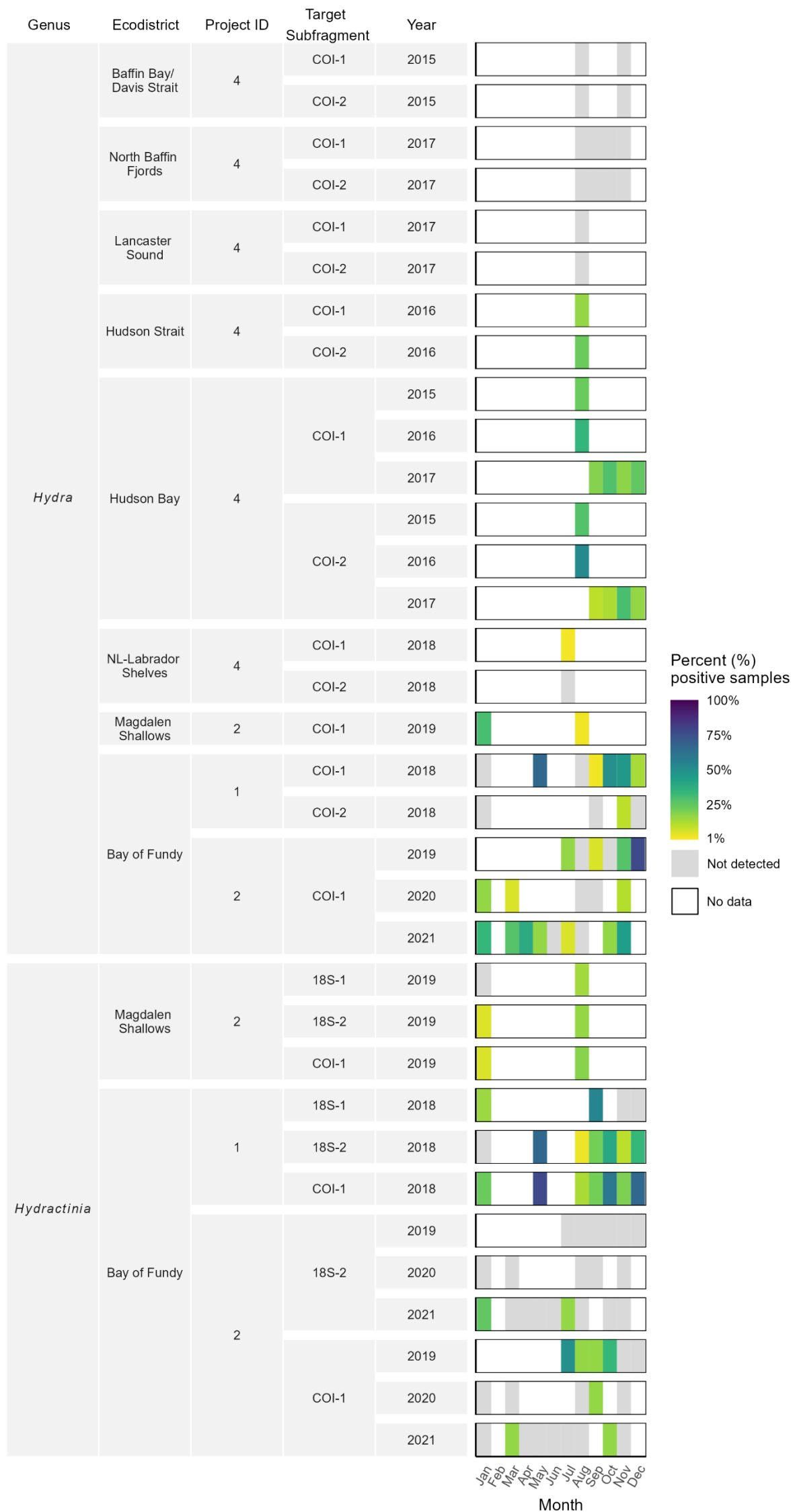


Figure 13. (Continued)



Figure 13. (Continued)



Figure 13. (Continued)

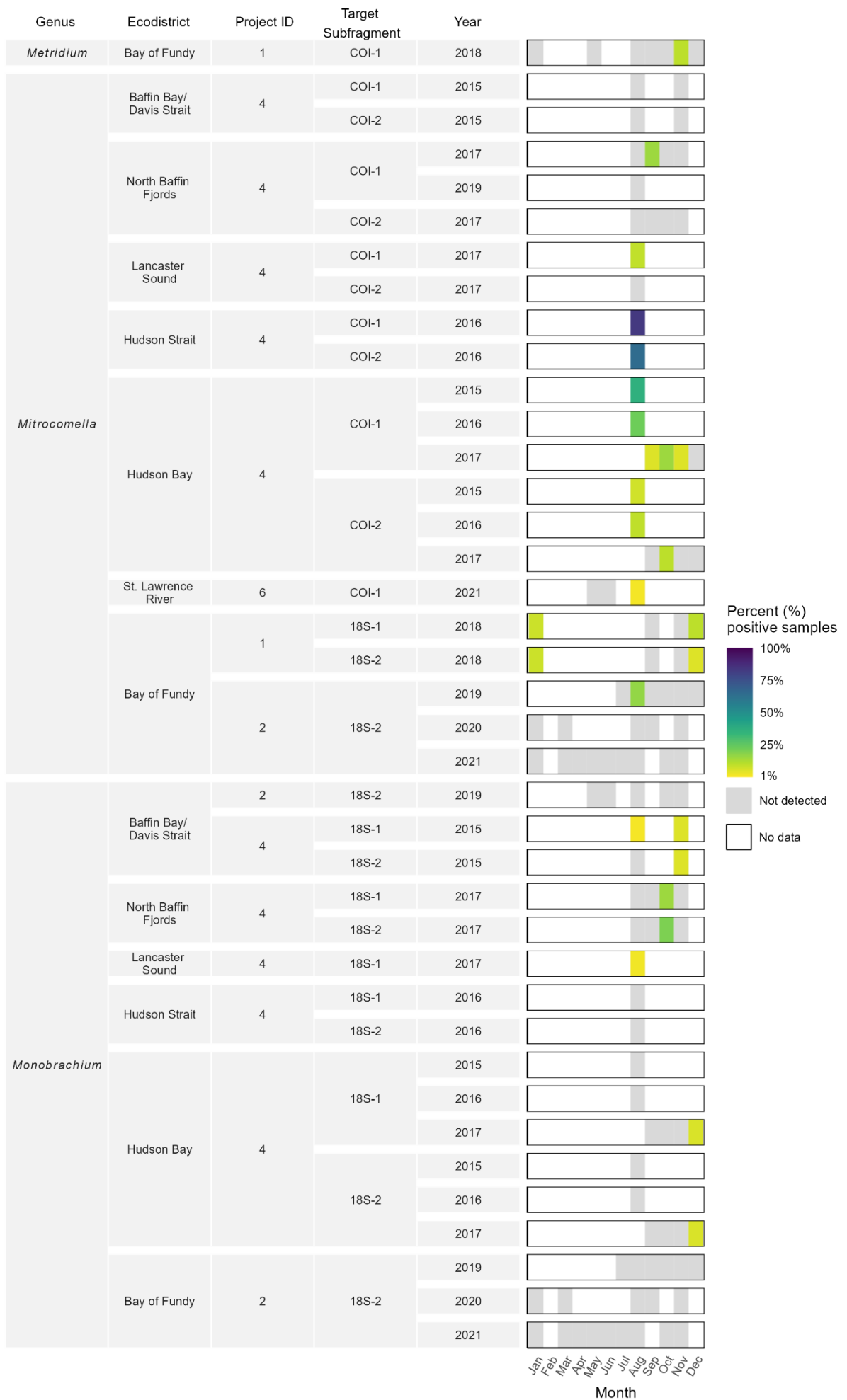


Figure 13. (Continued)



Figure 13. (Continued)

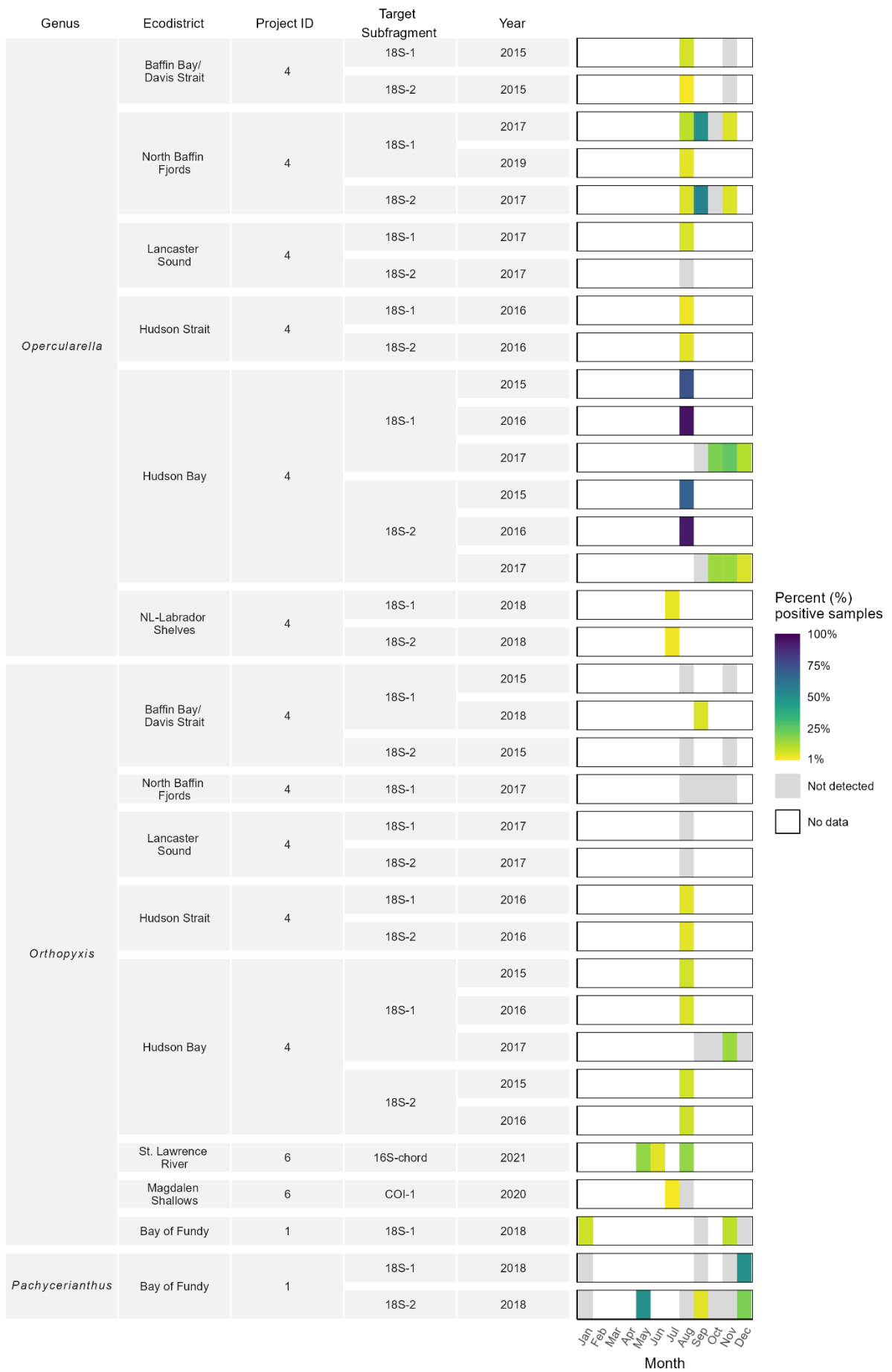


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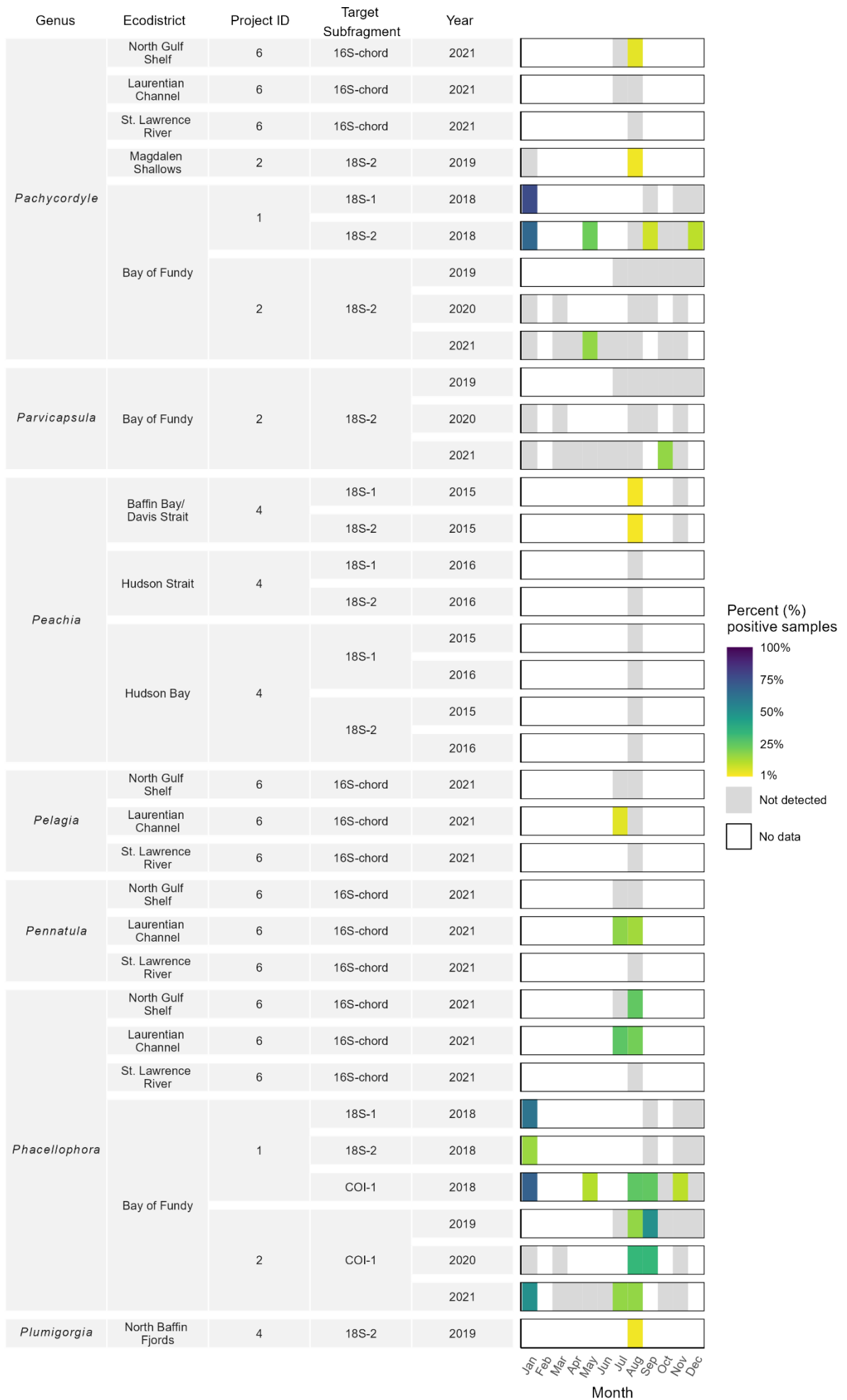


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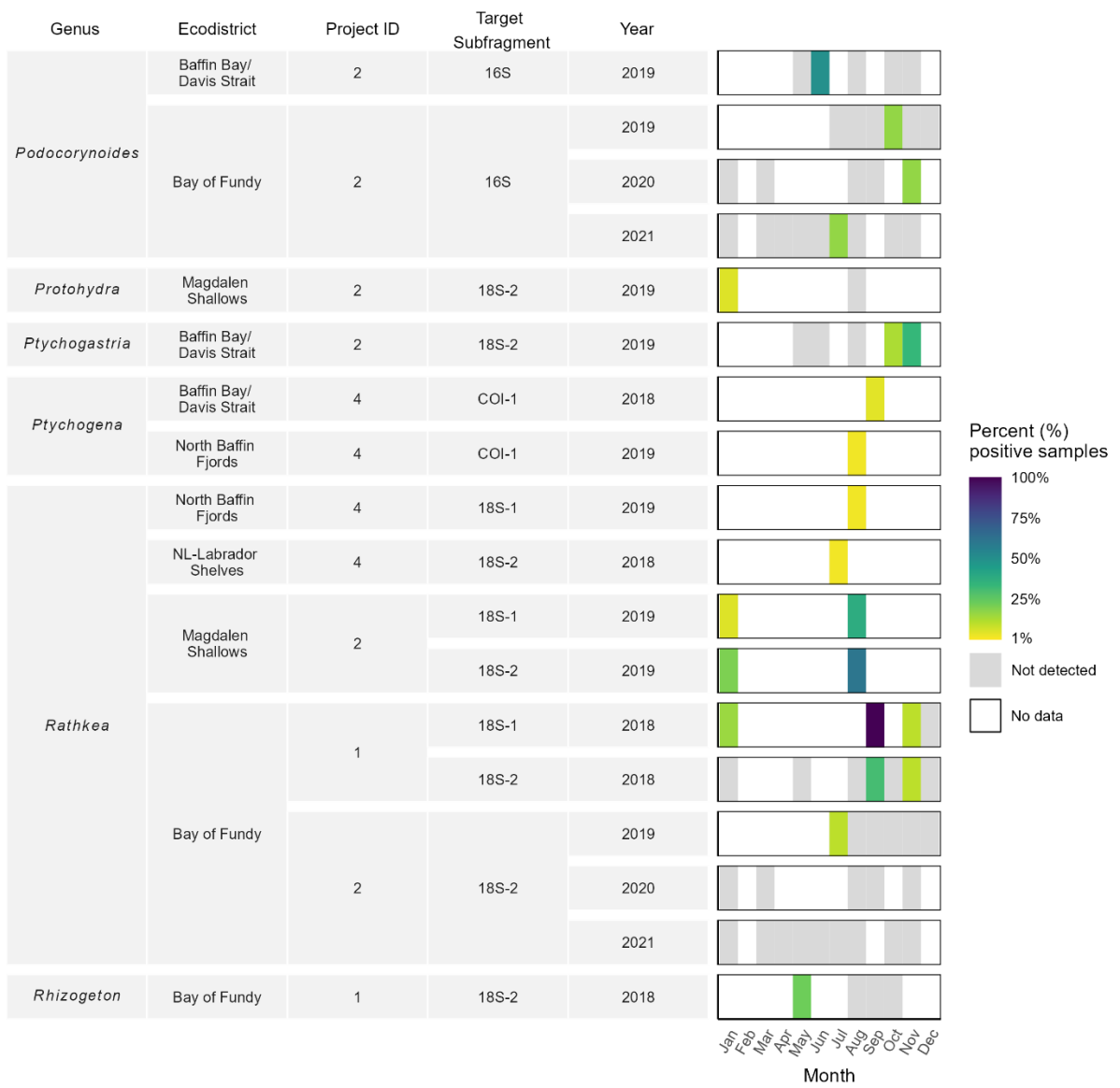


Figure 13. (Continued)



Figure 13. (Continued)

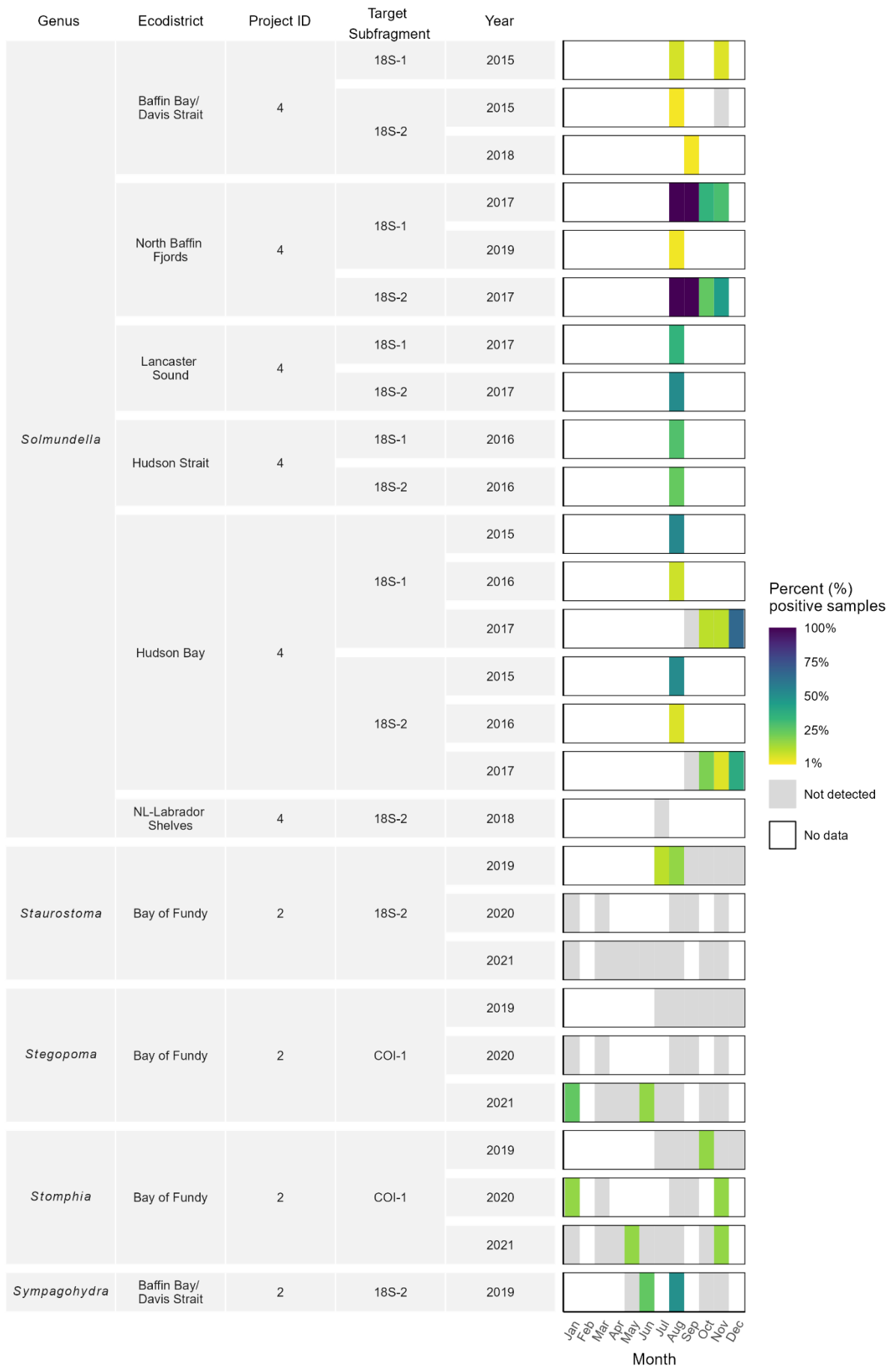


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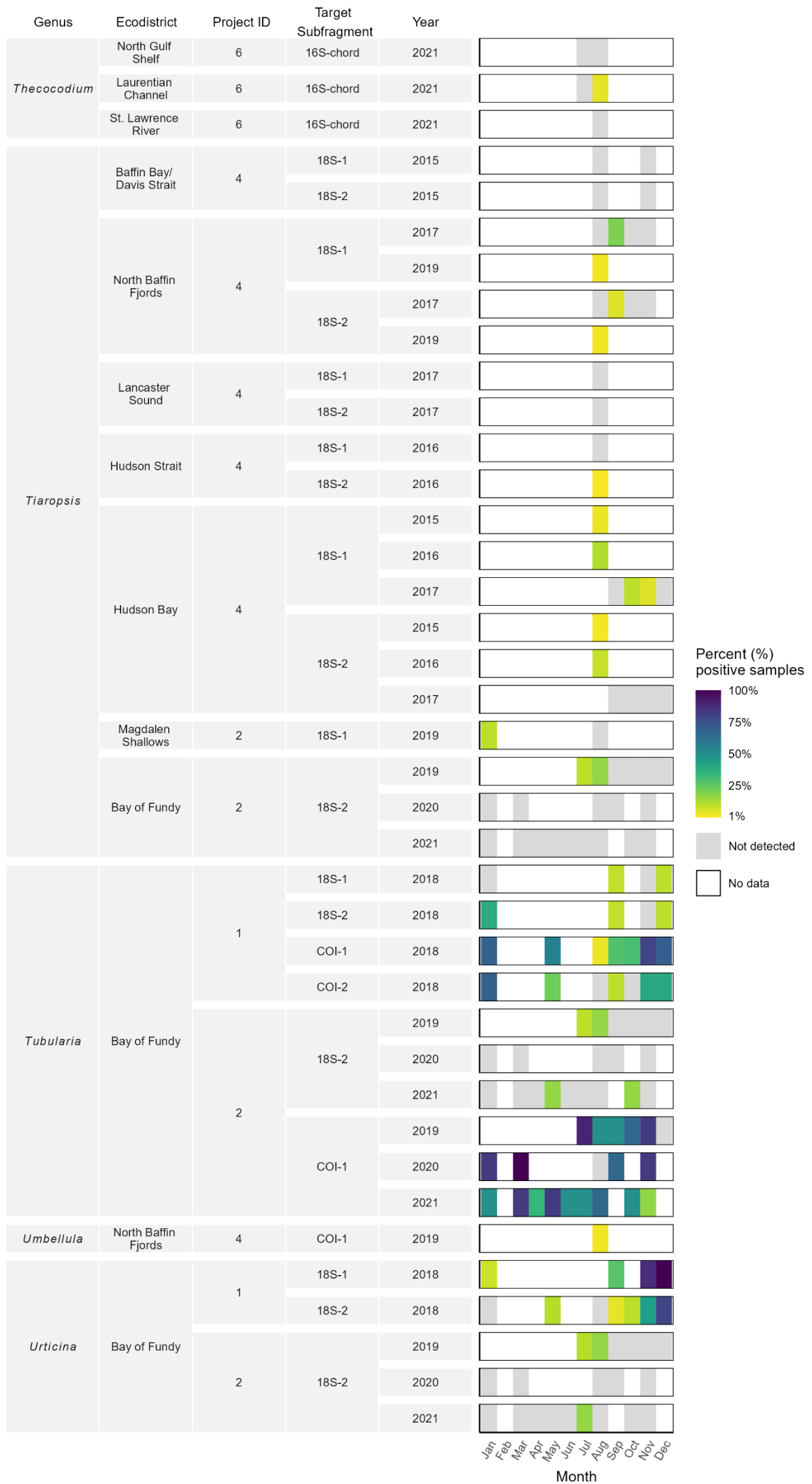


Figure 13. (Continued)

6.7 CTENOPHORA

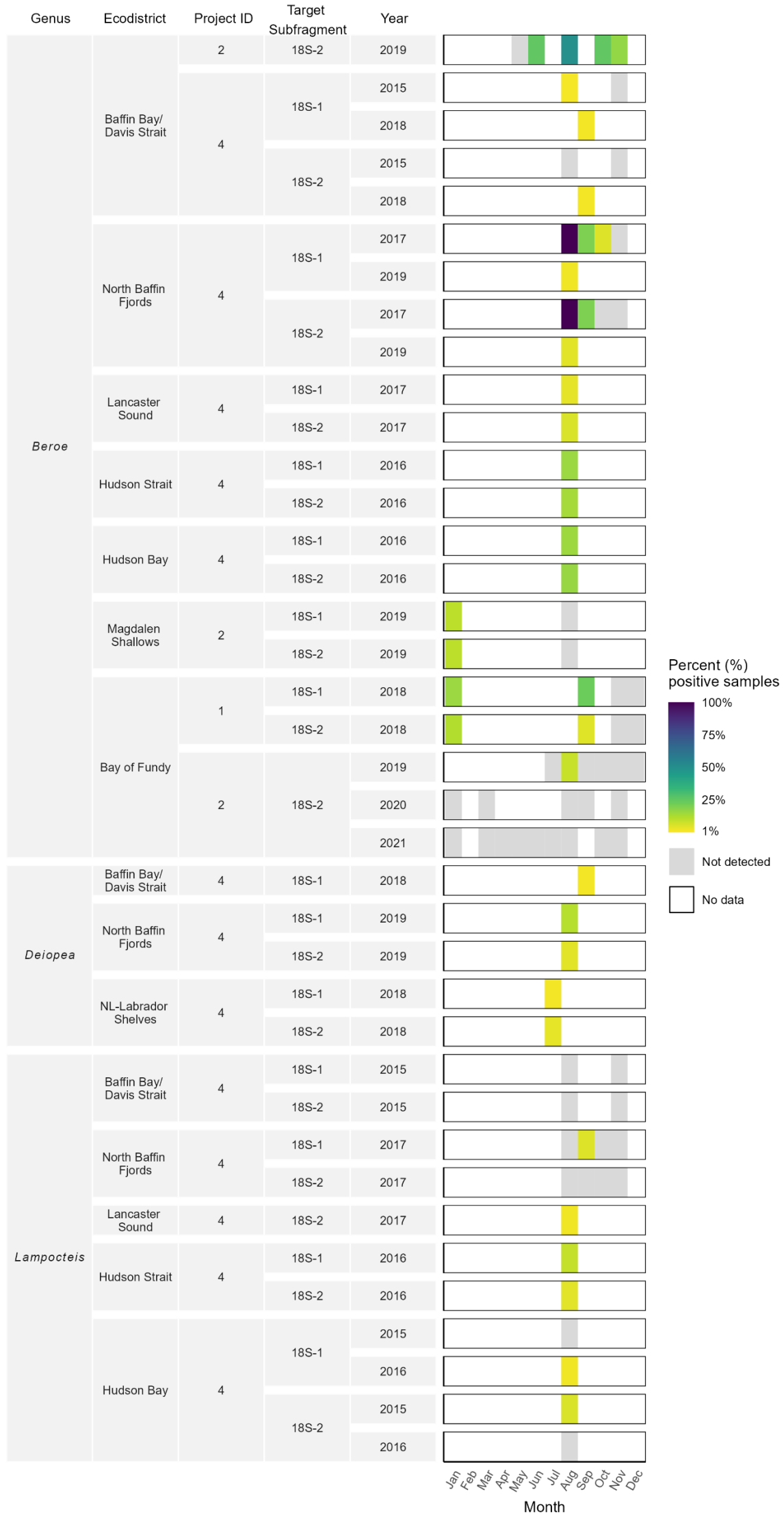


Figure 13. (Continued)

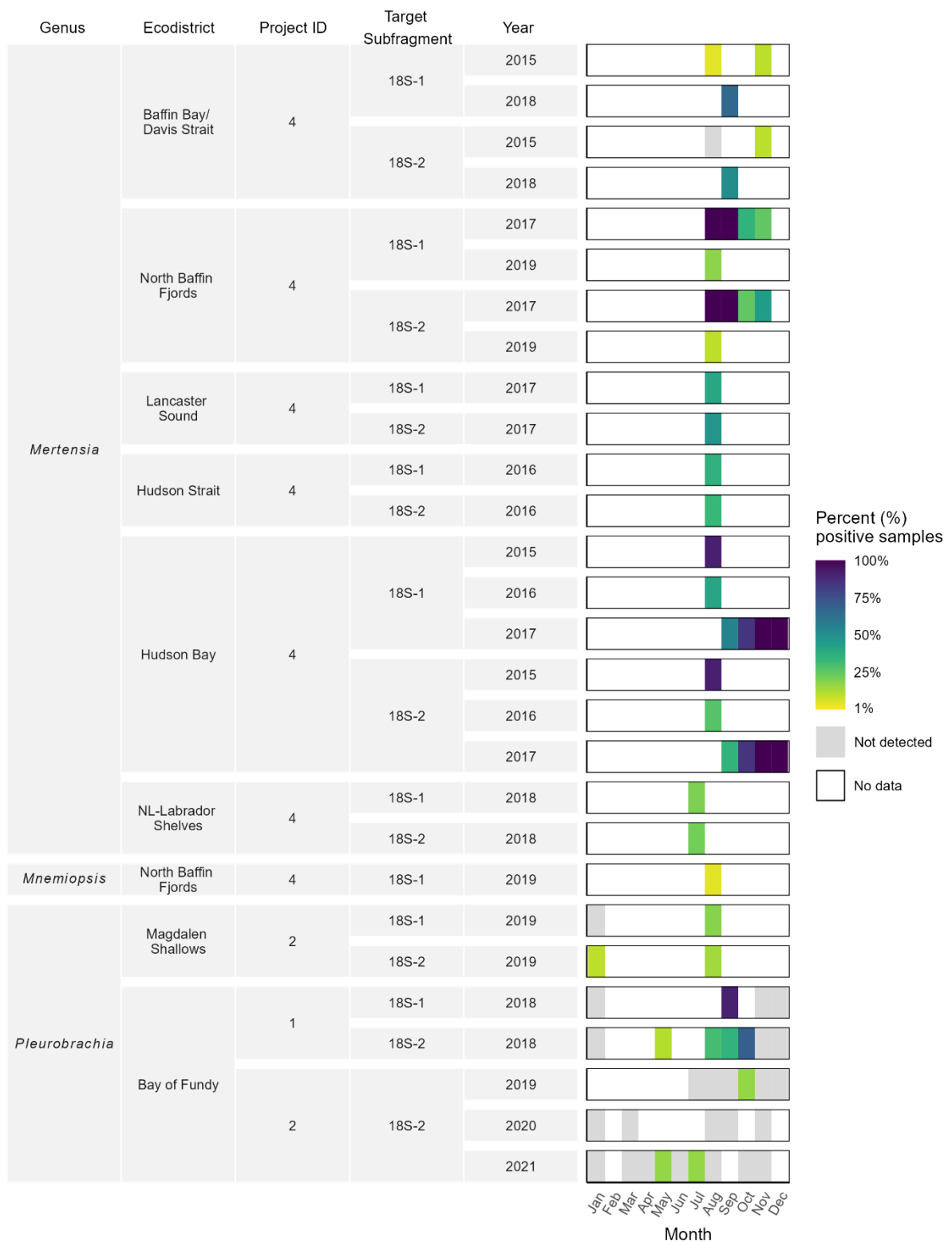


Figure 13. (Continued)

6.8 ECHINODERMATA



Figure 13. (Continued)

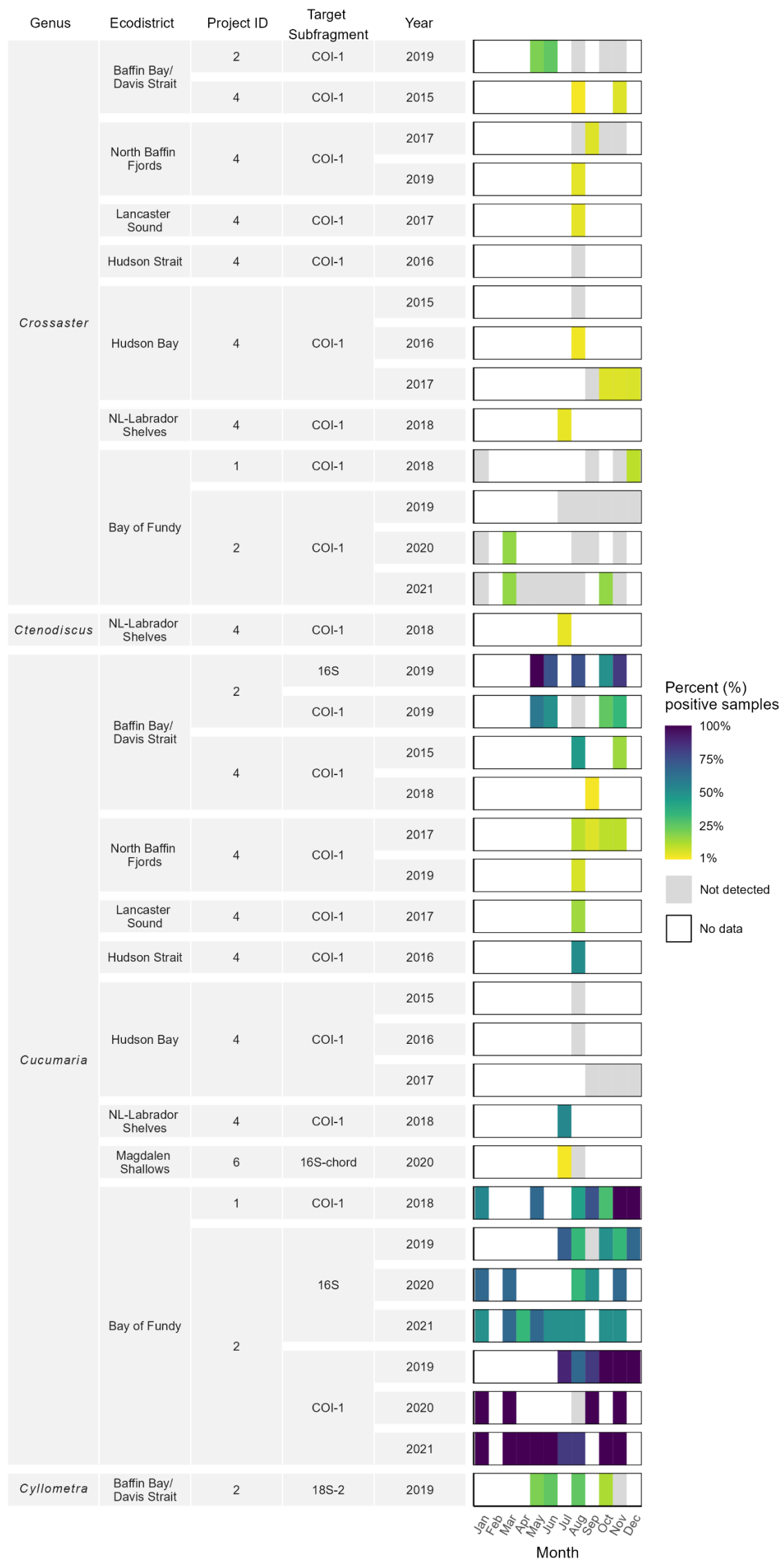


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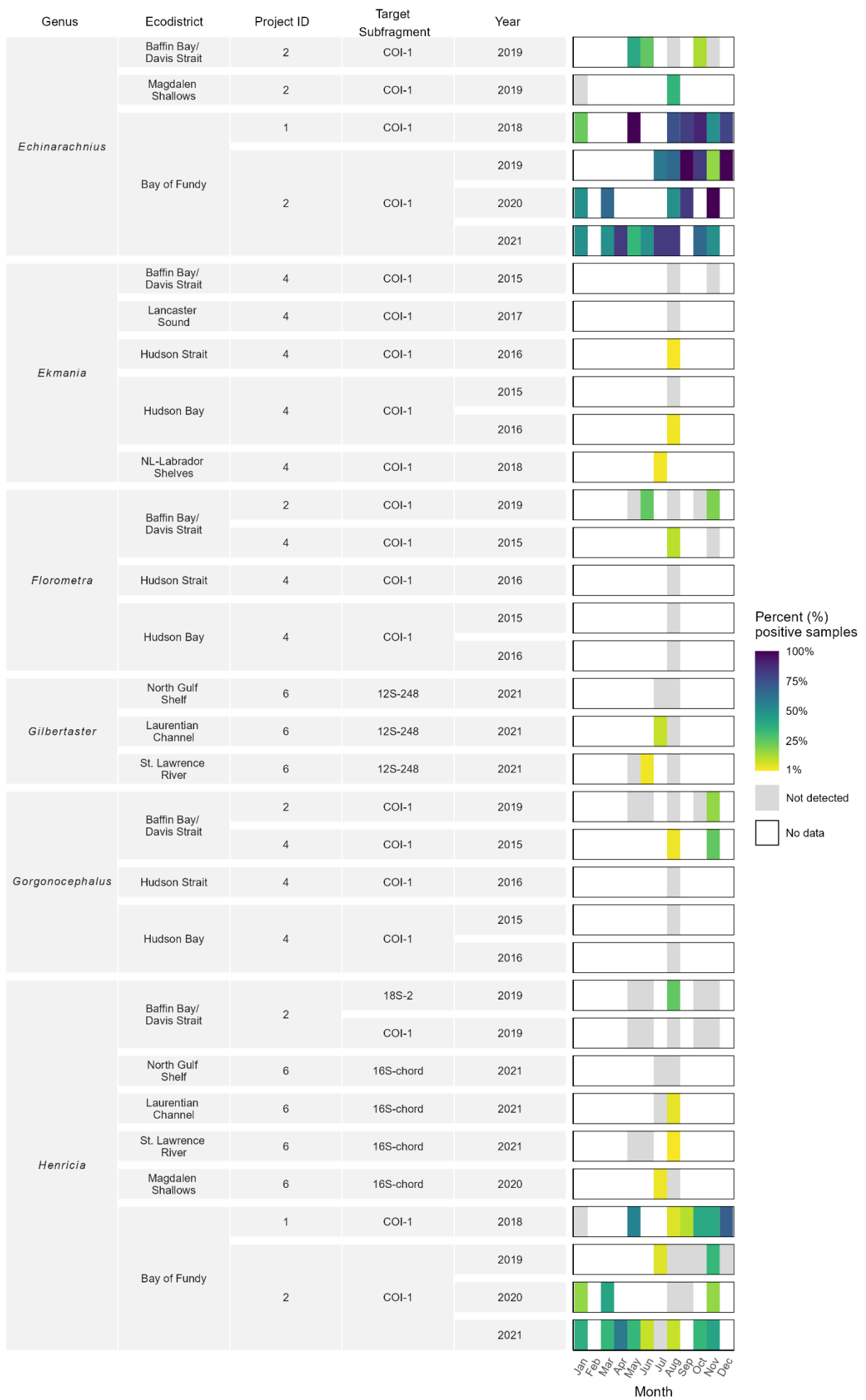


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Figure 13. (Continued)



Figure 13. (Continued)

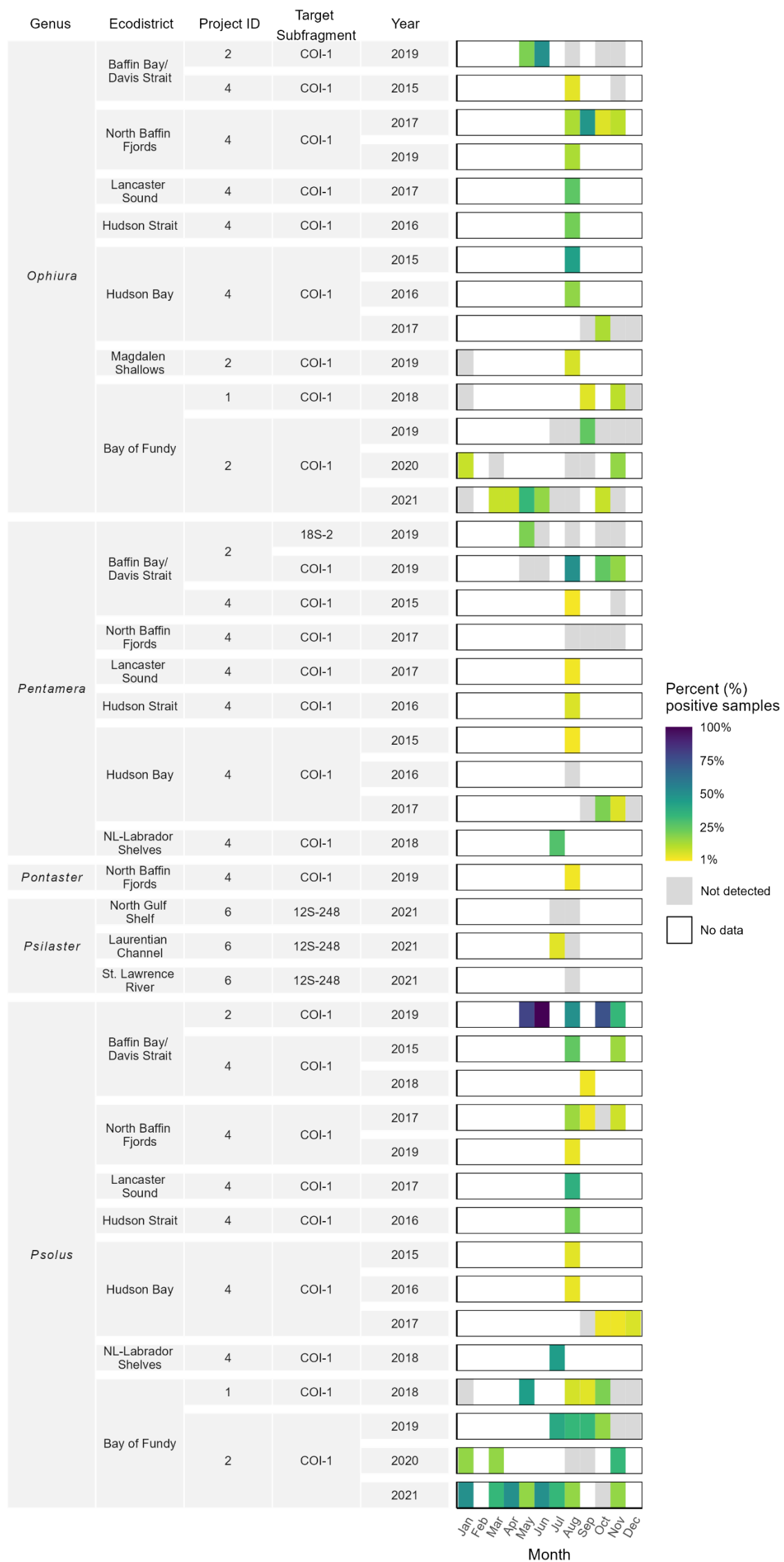


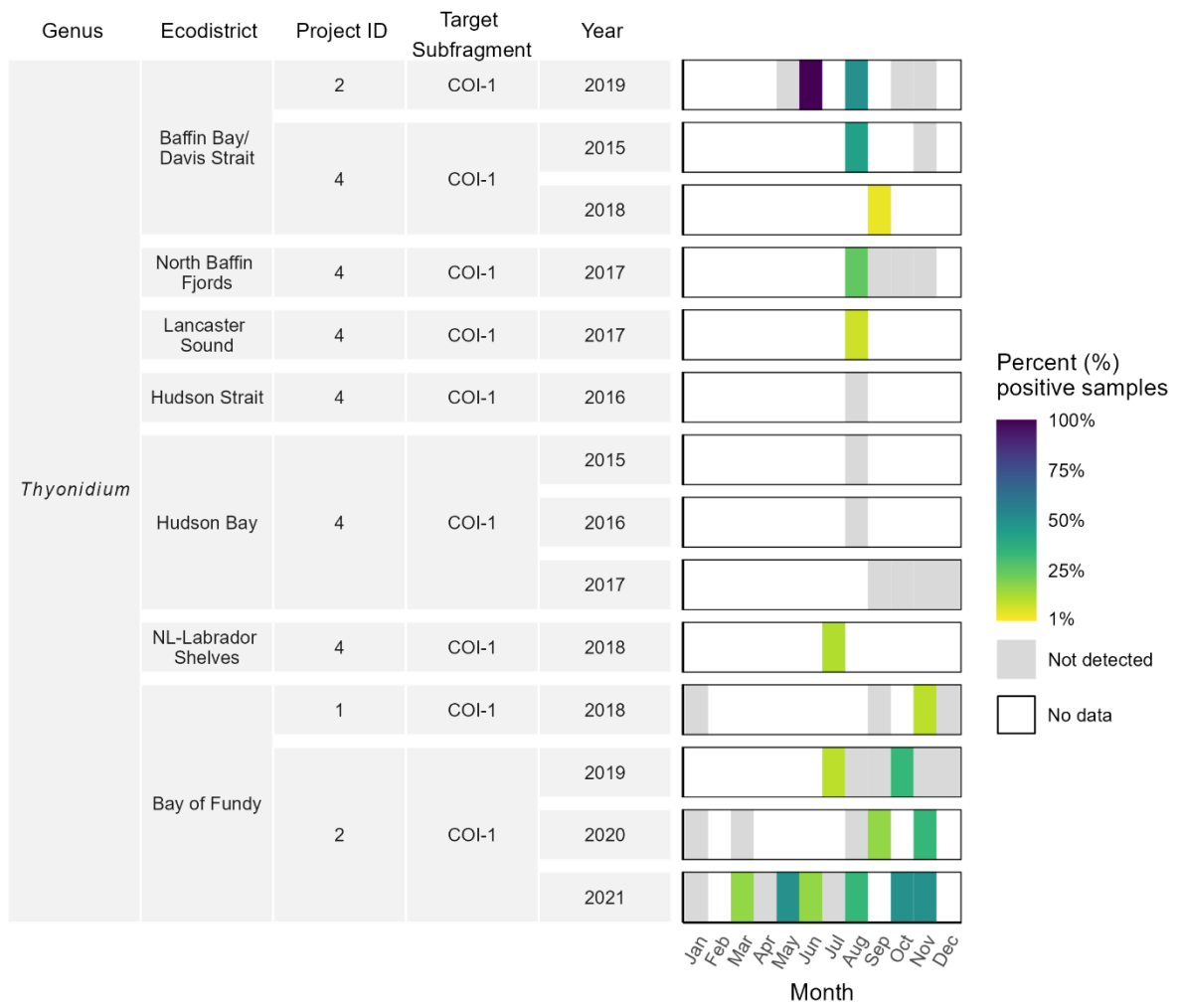
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Figure 13. (Continued)



Figure 13. (Continued)



6.9 ENTOPROCTA

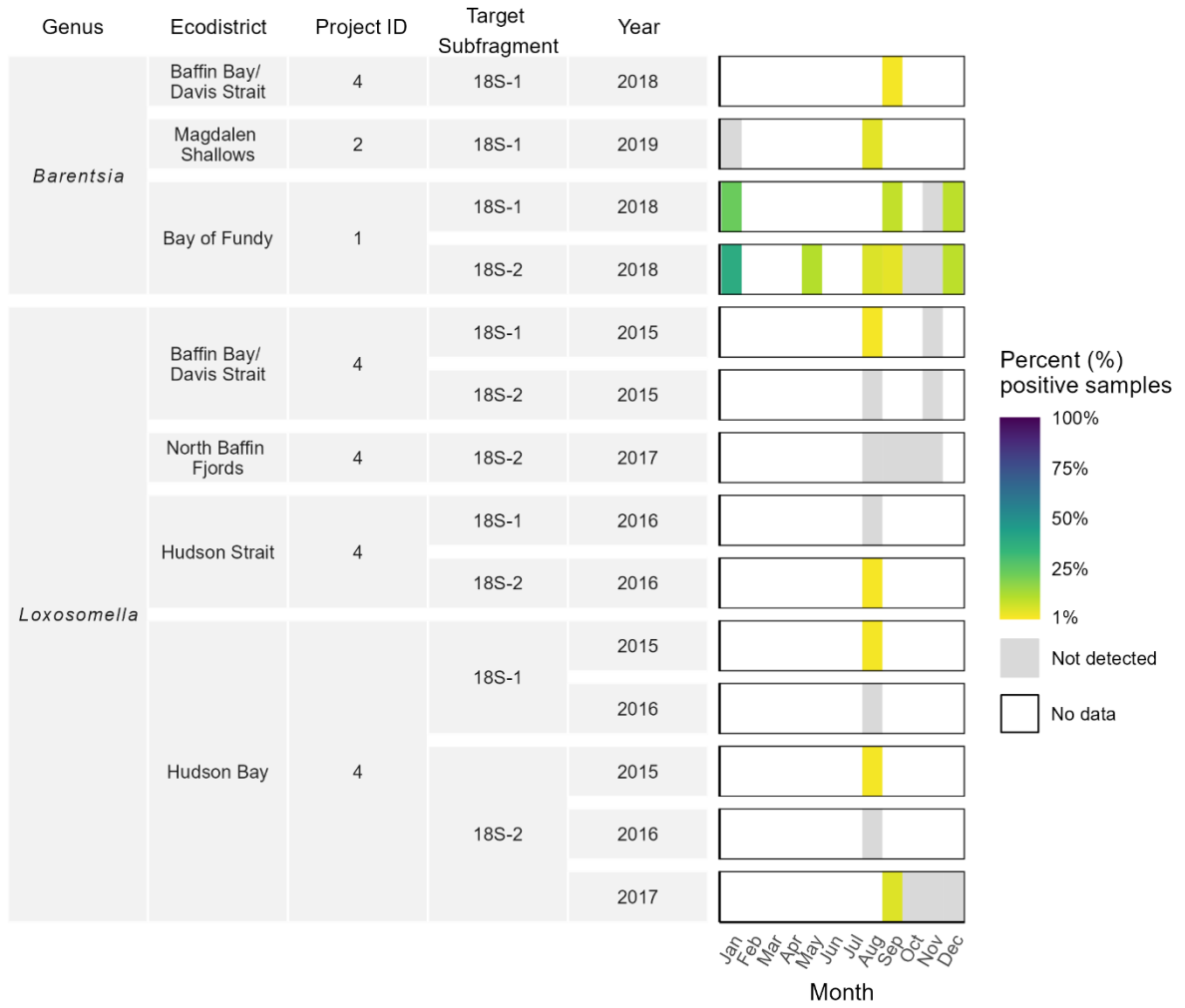


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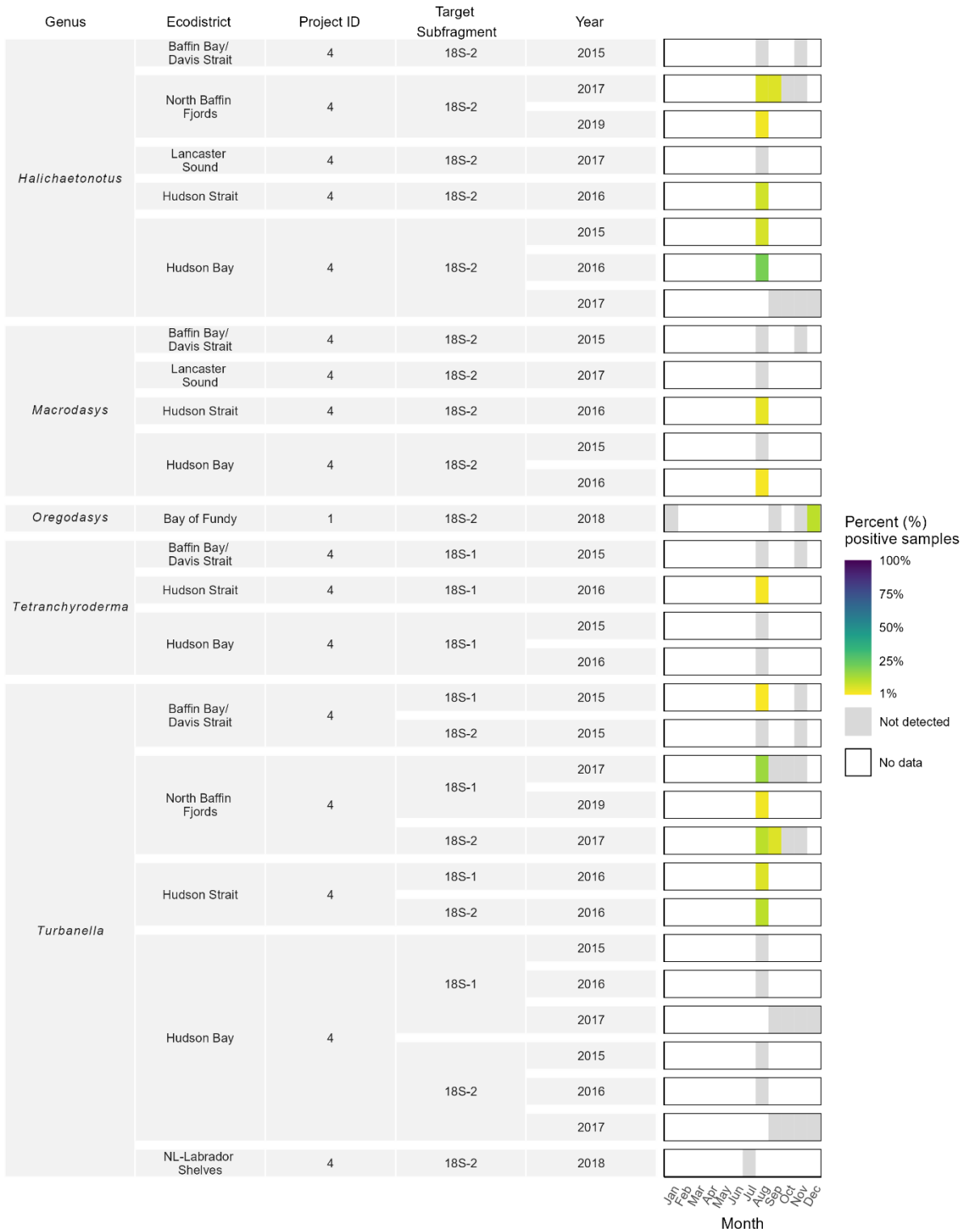


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6.11 GNATHIFERA

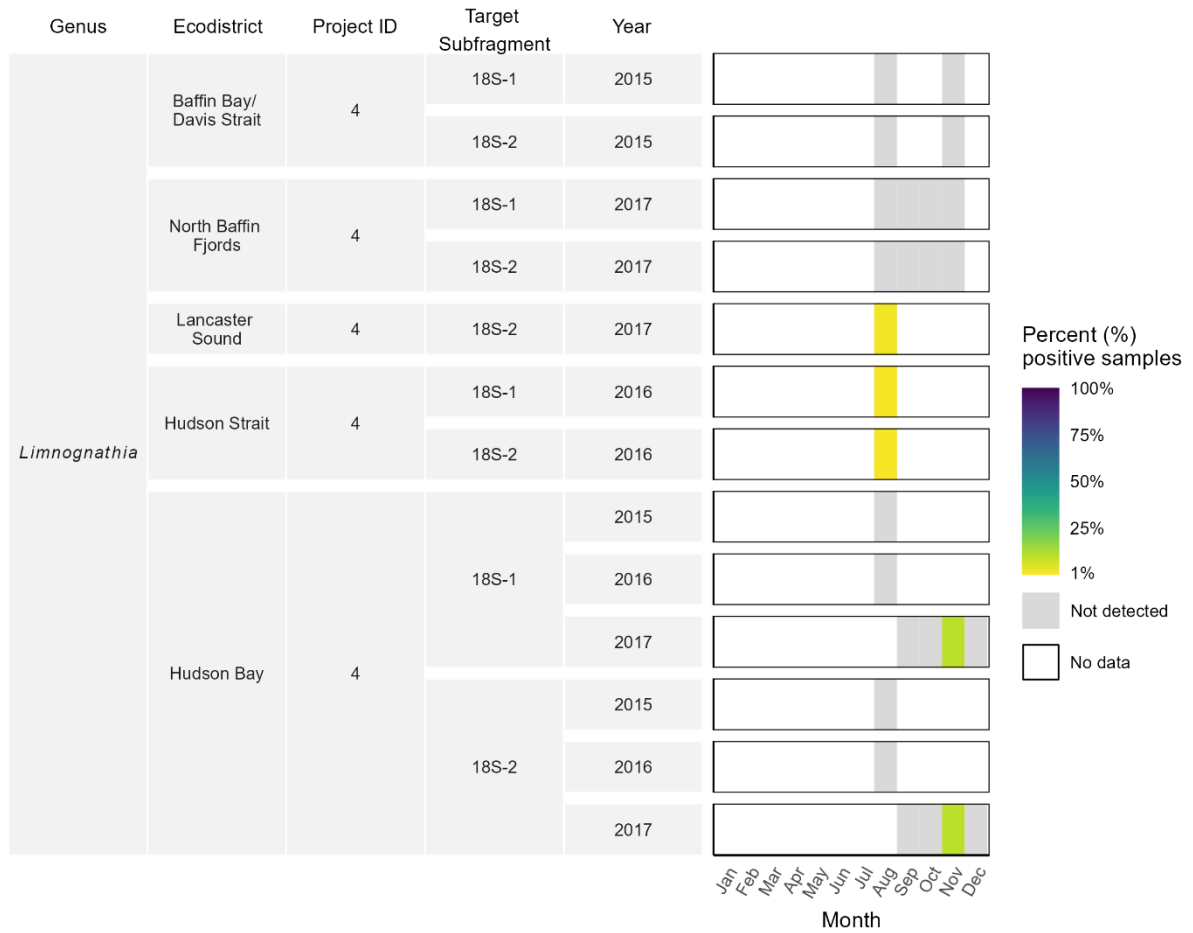
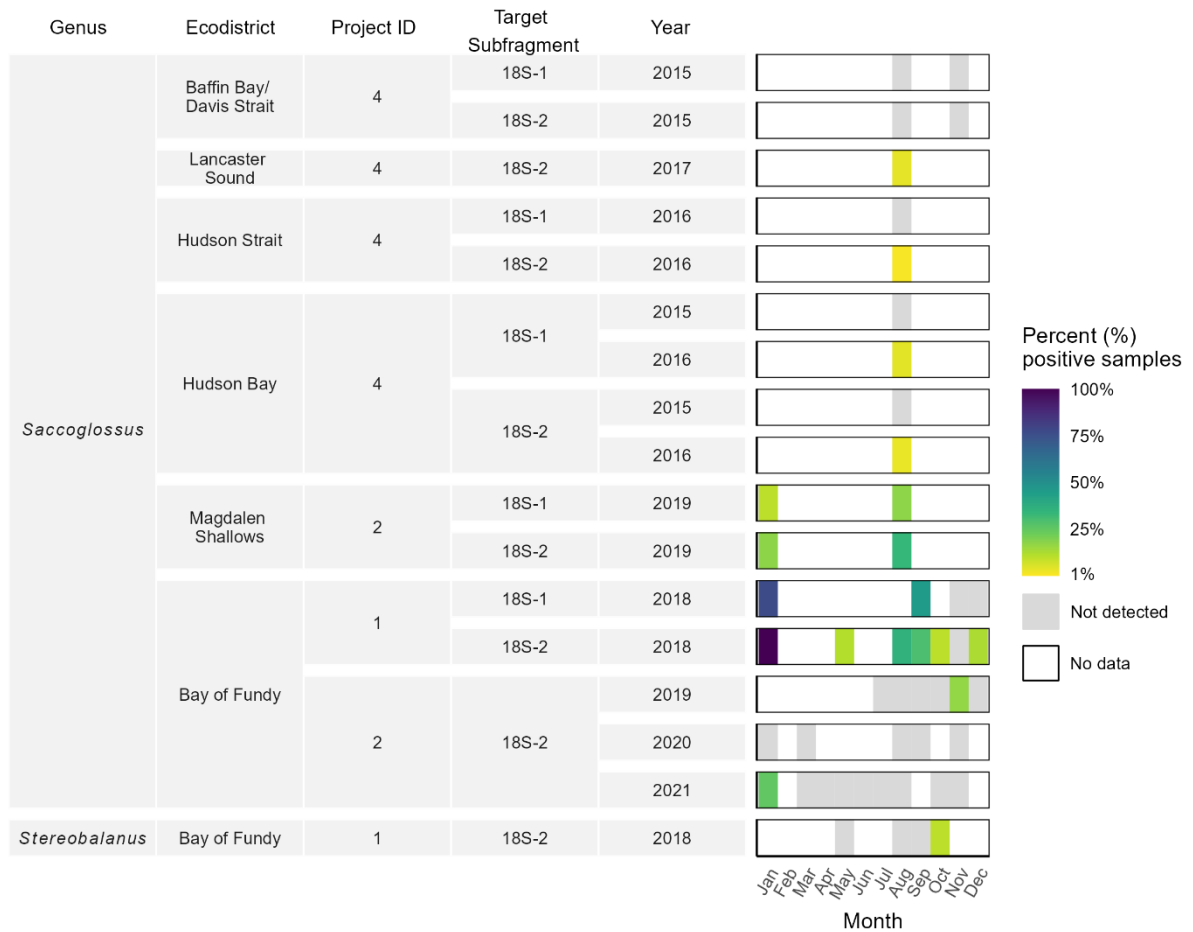
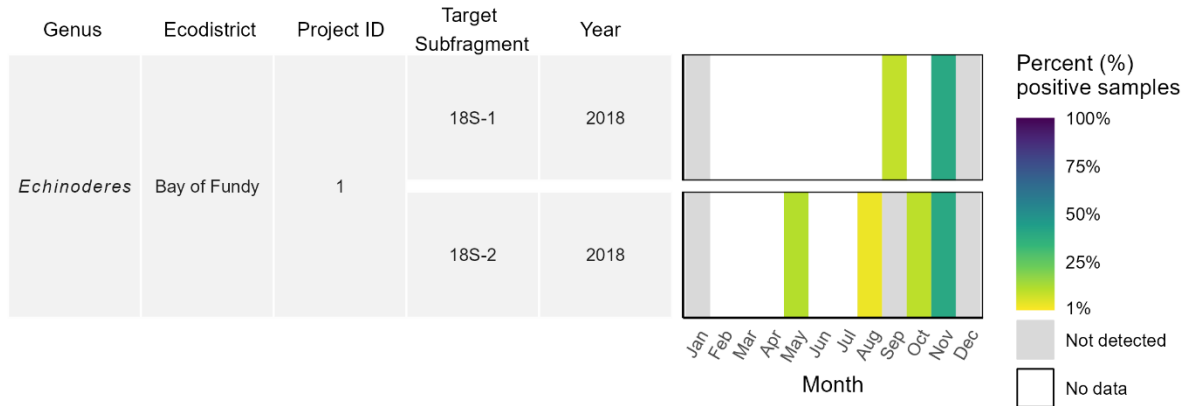


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6.12 HEMICHORDATA



6.13 KINORHYNCHA



6.14 MOLLUSCA



Figure 13. (Continued)

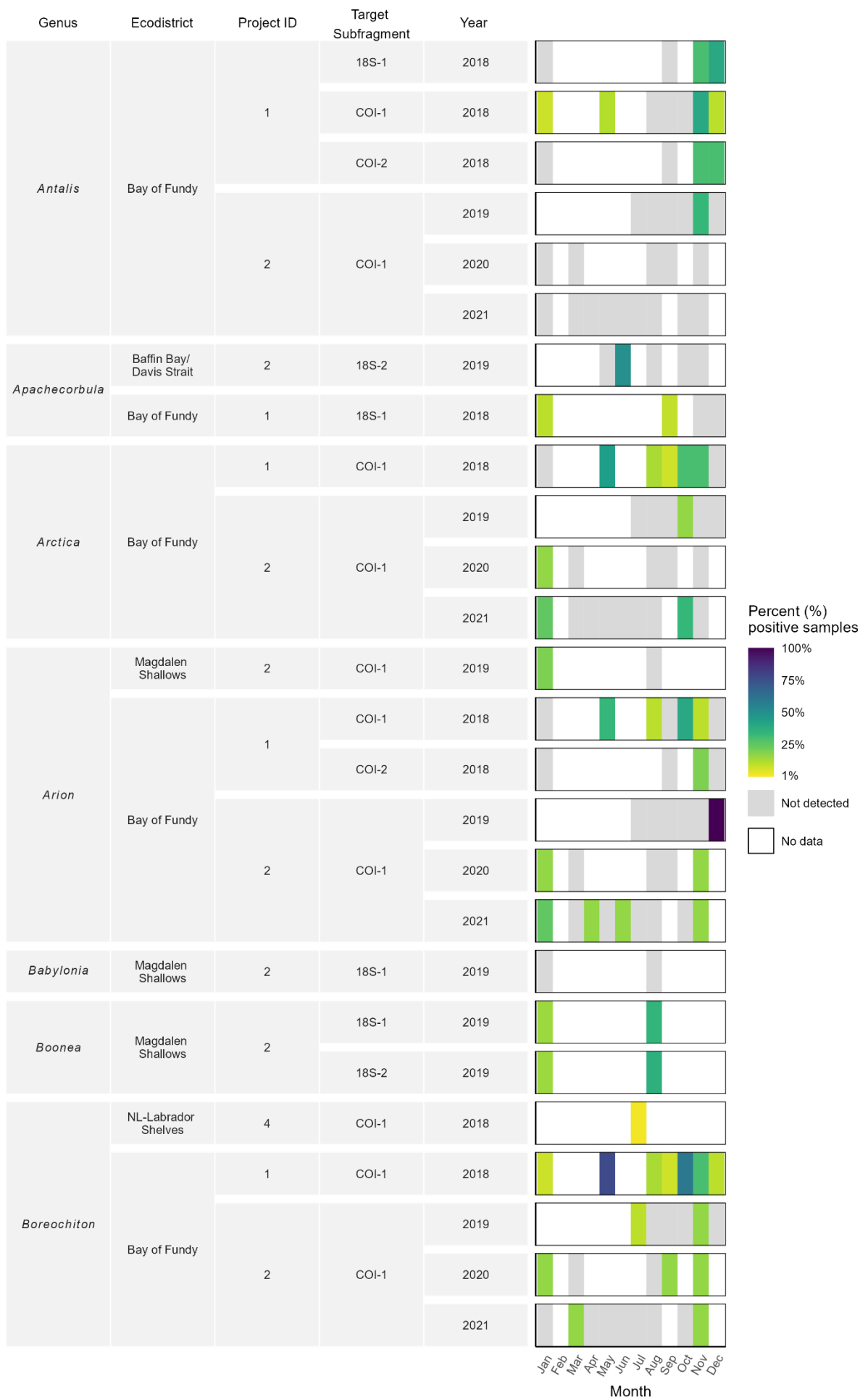


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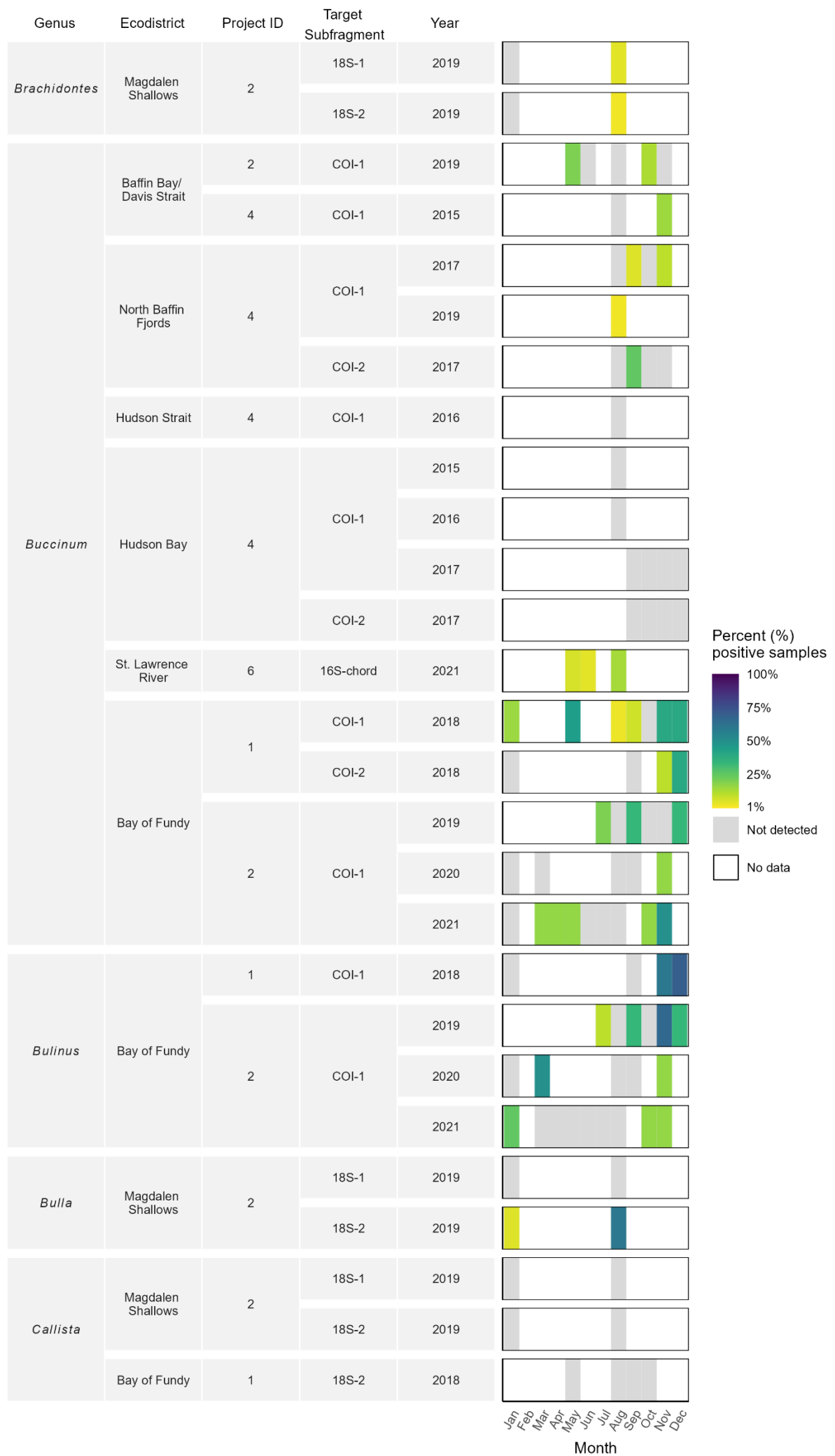


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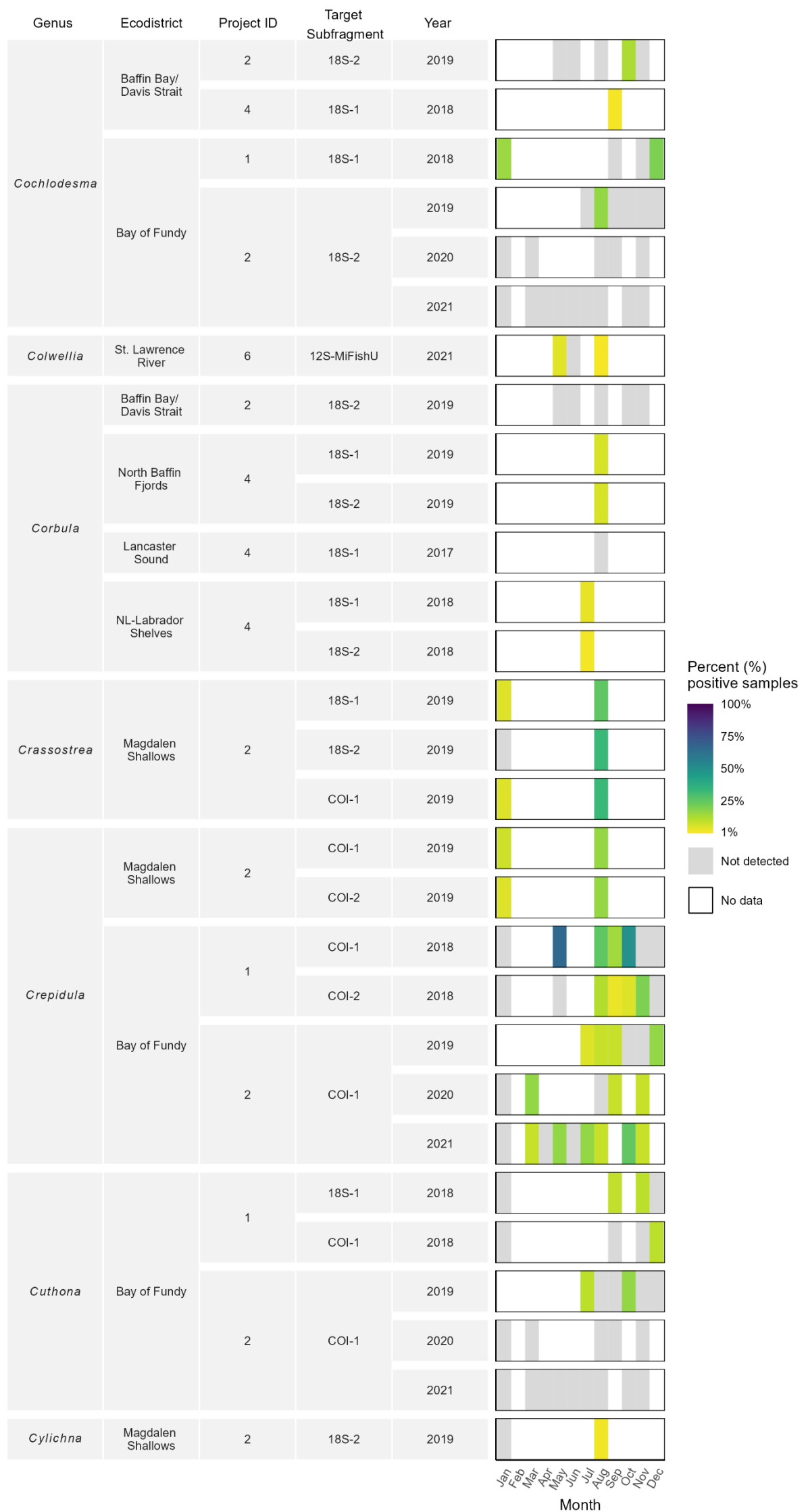


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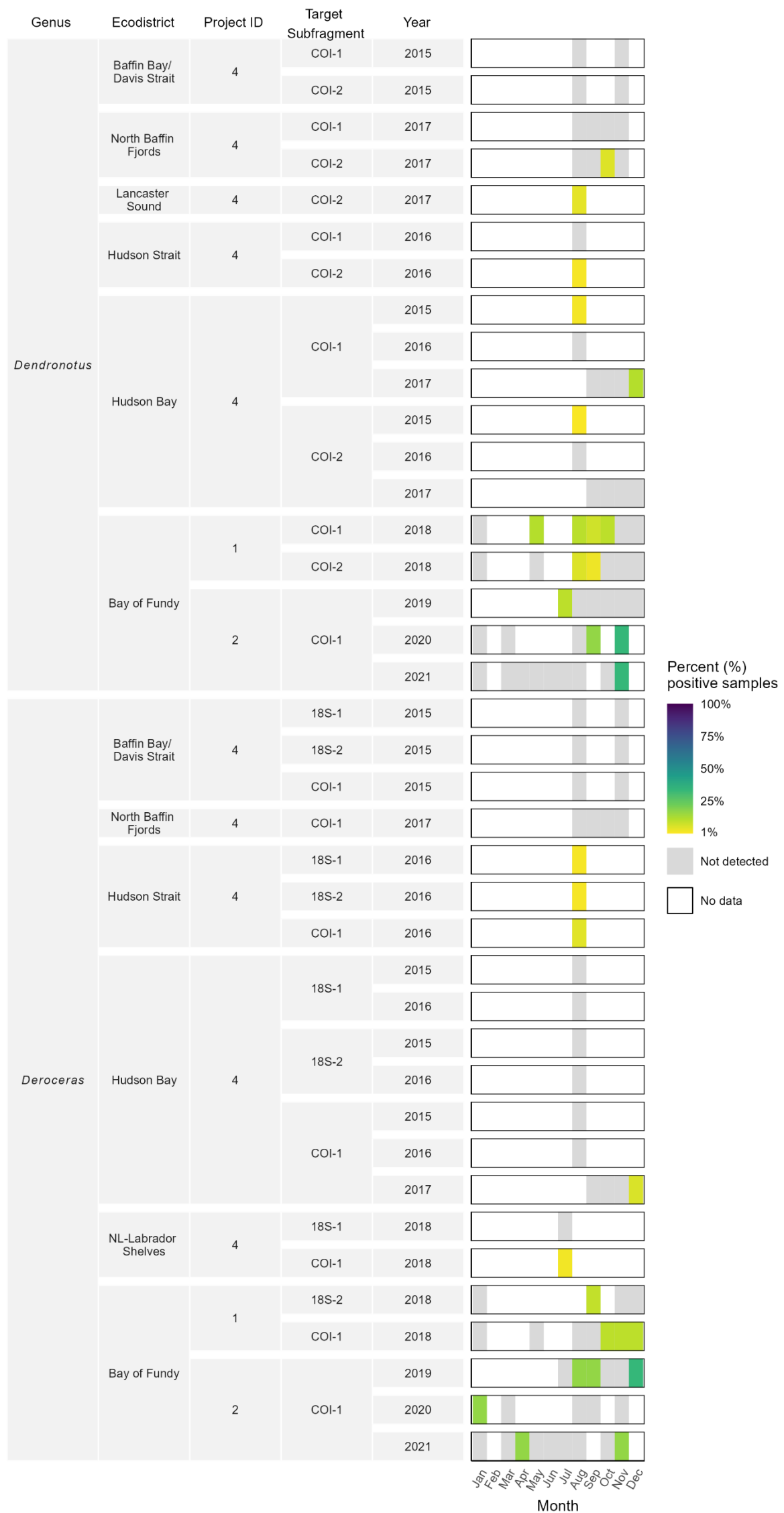


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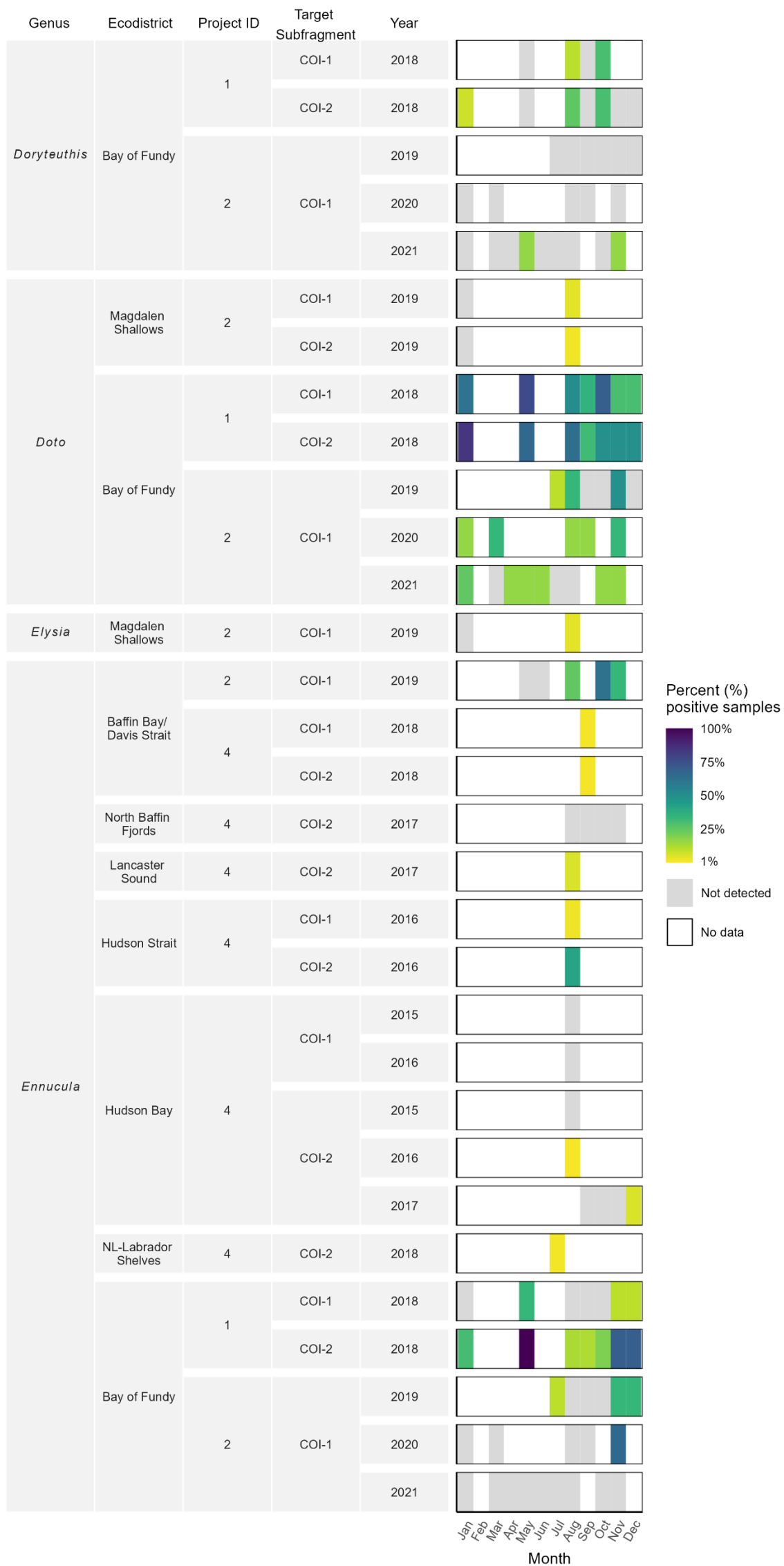


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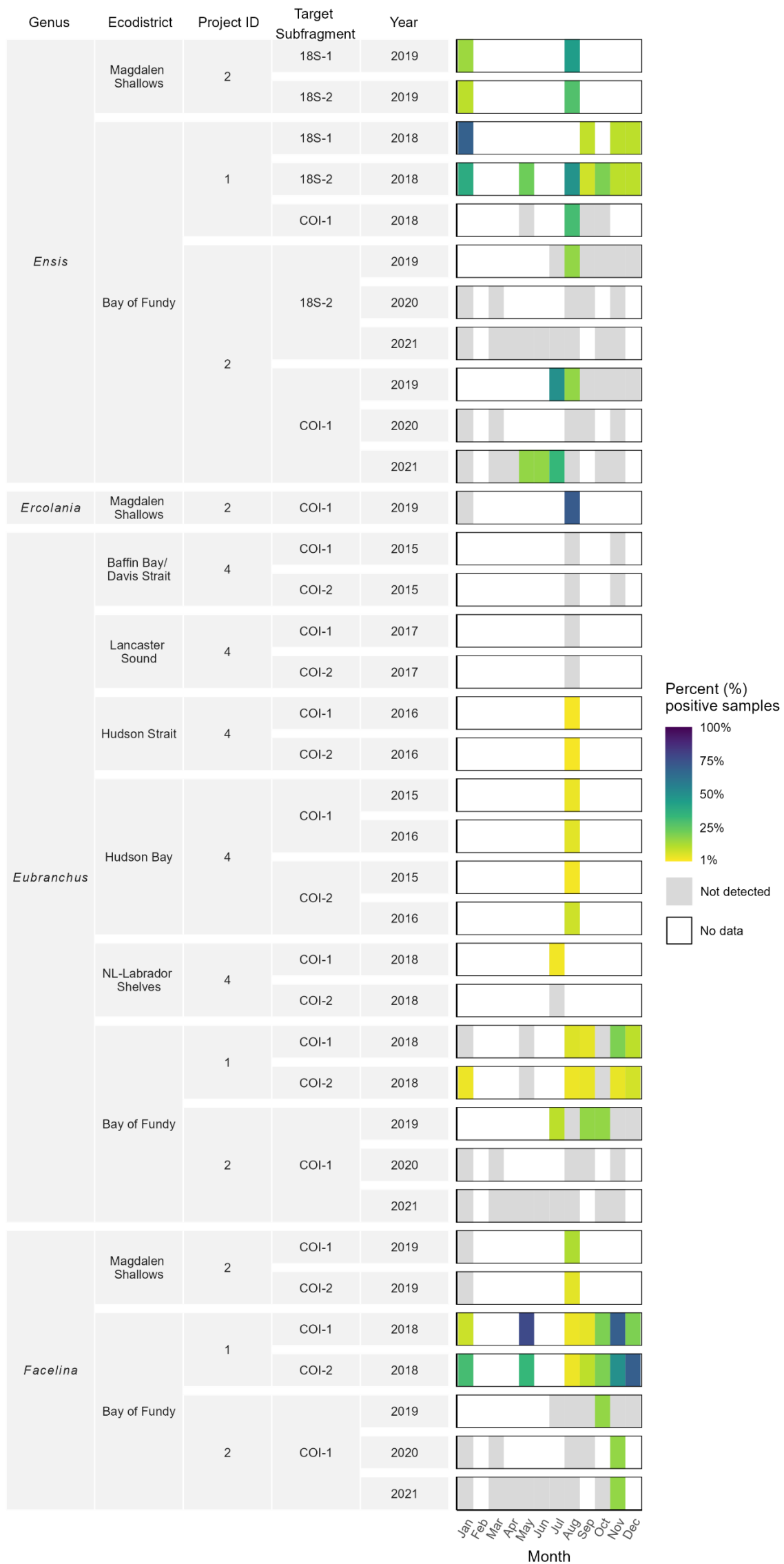


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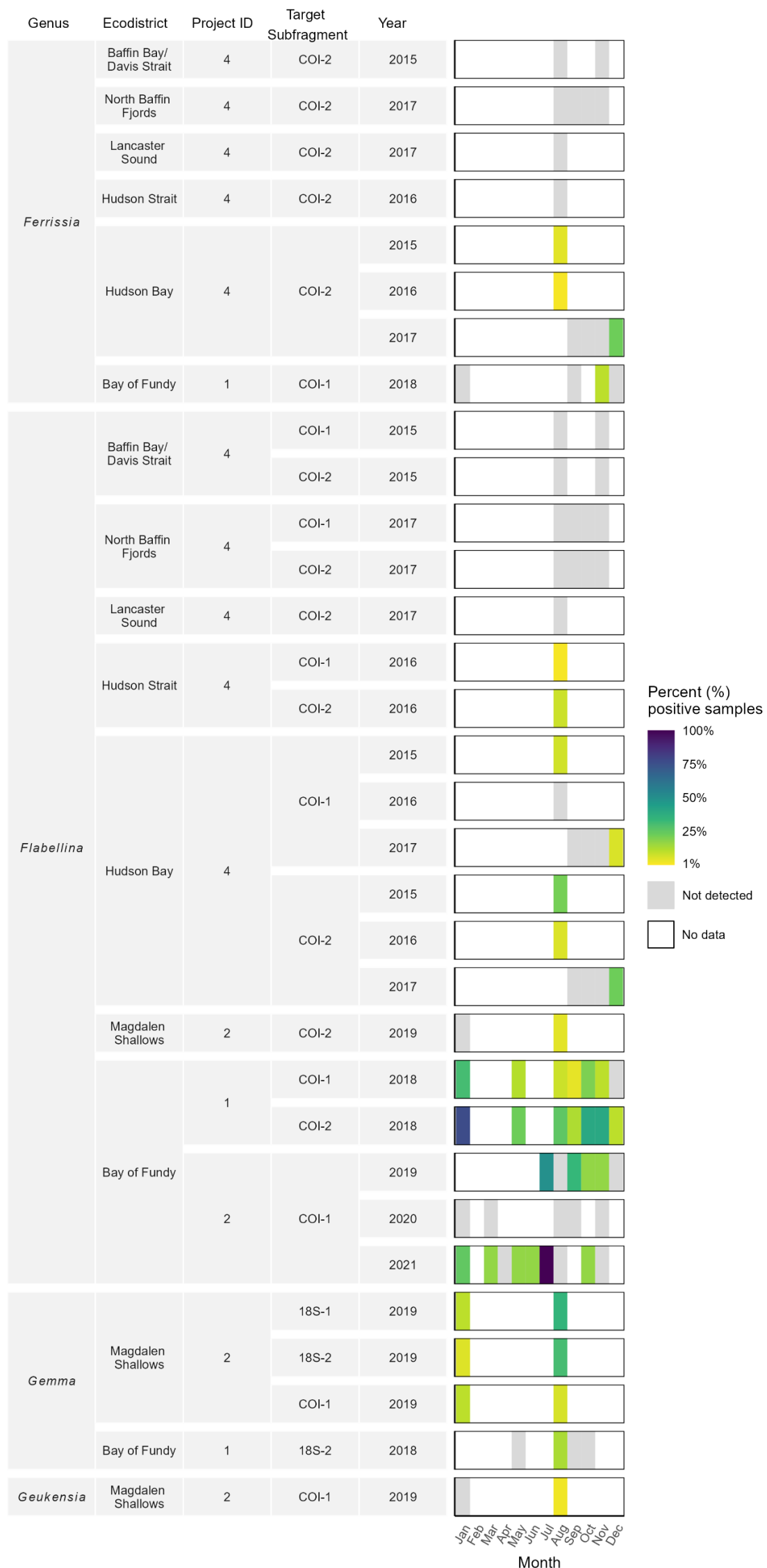


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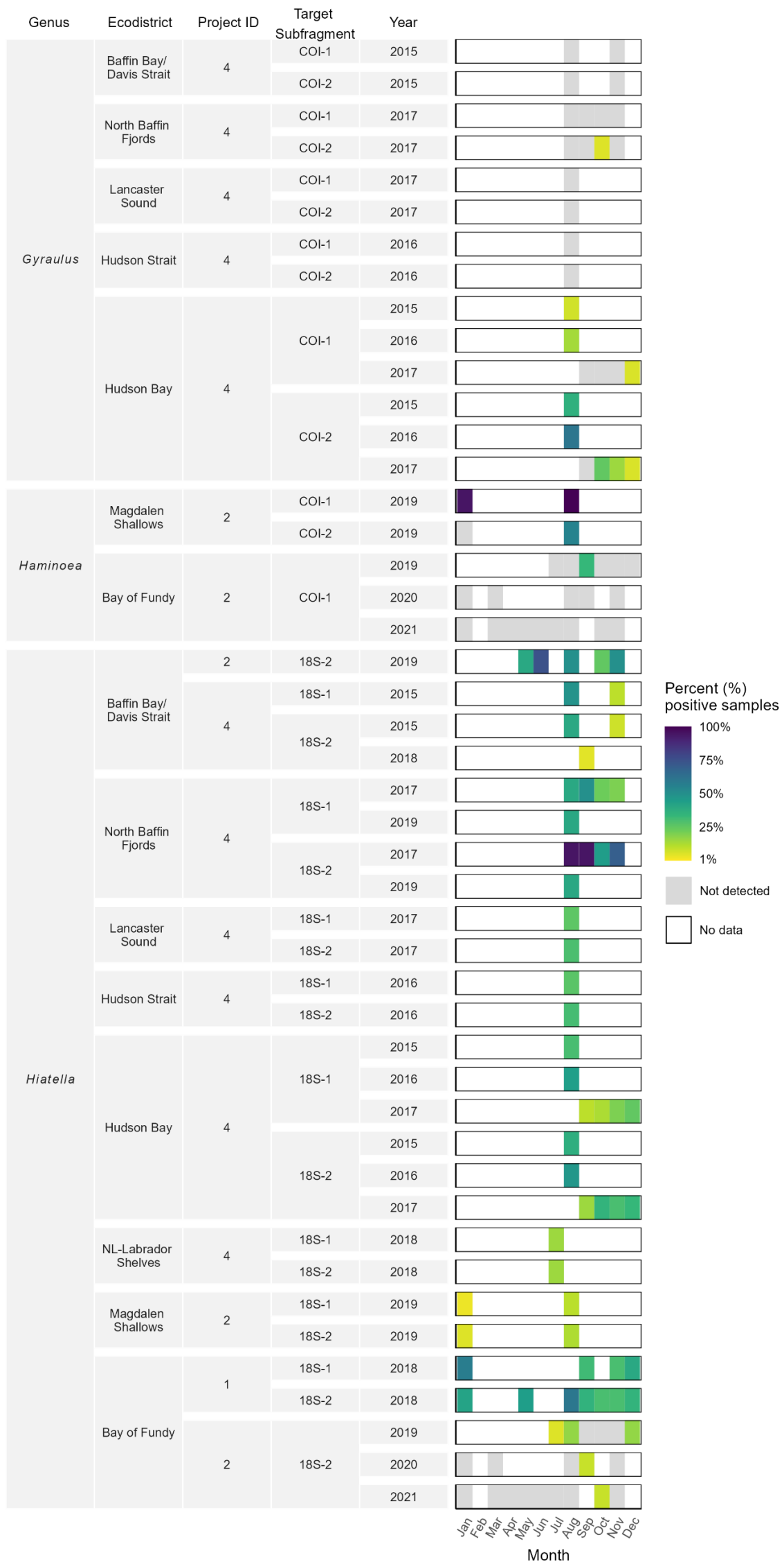


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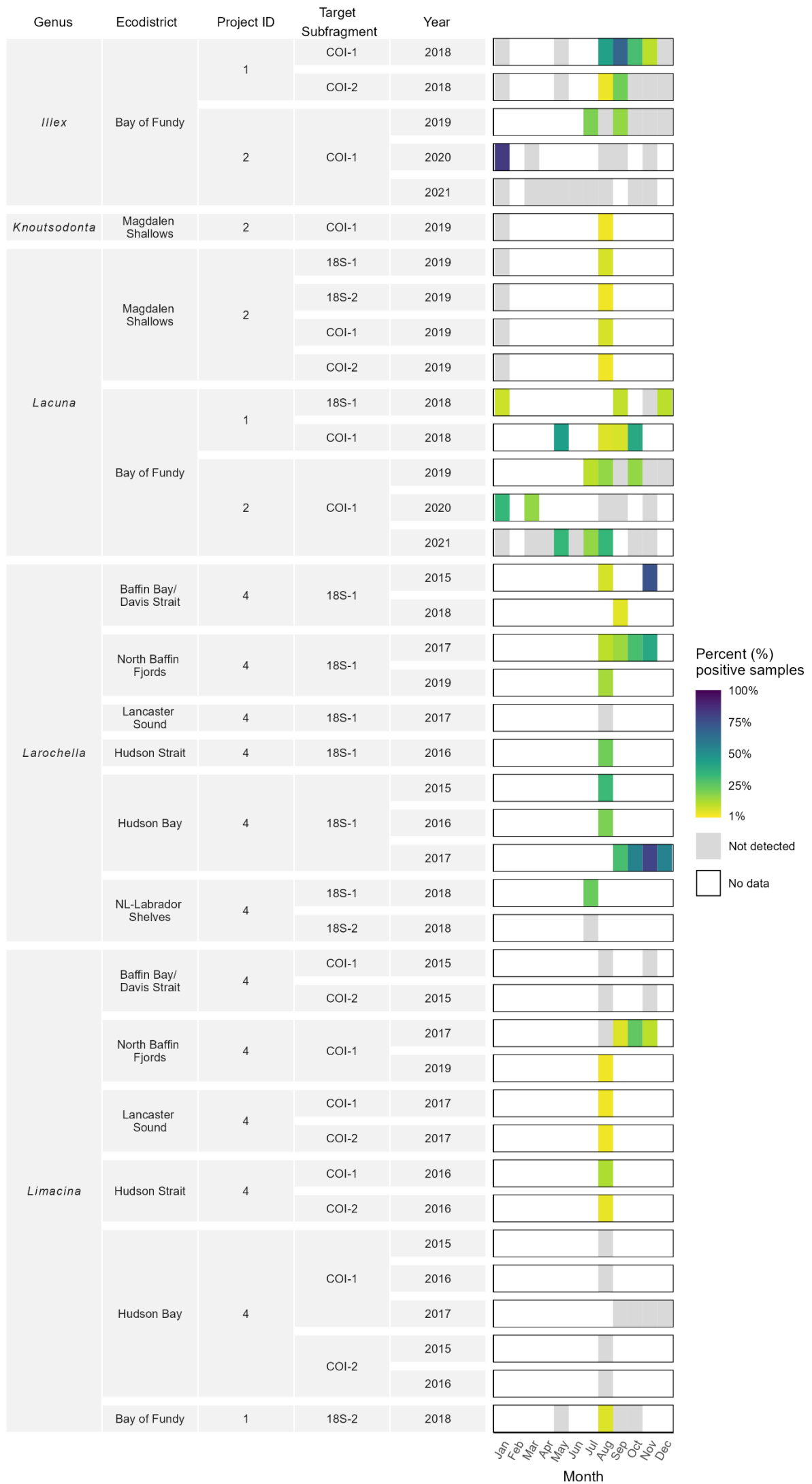


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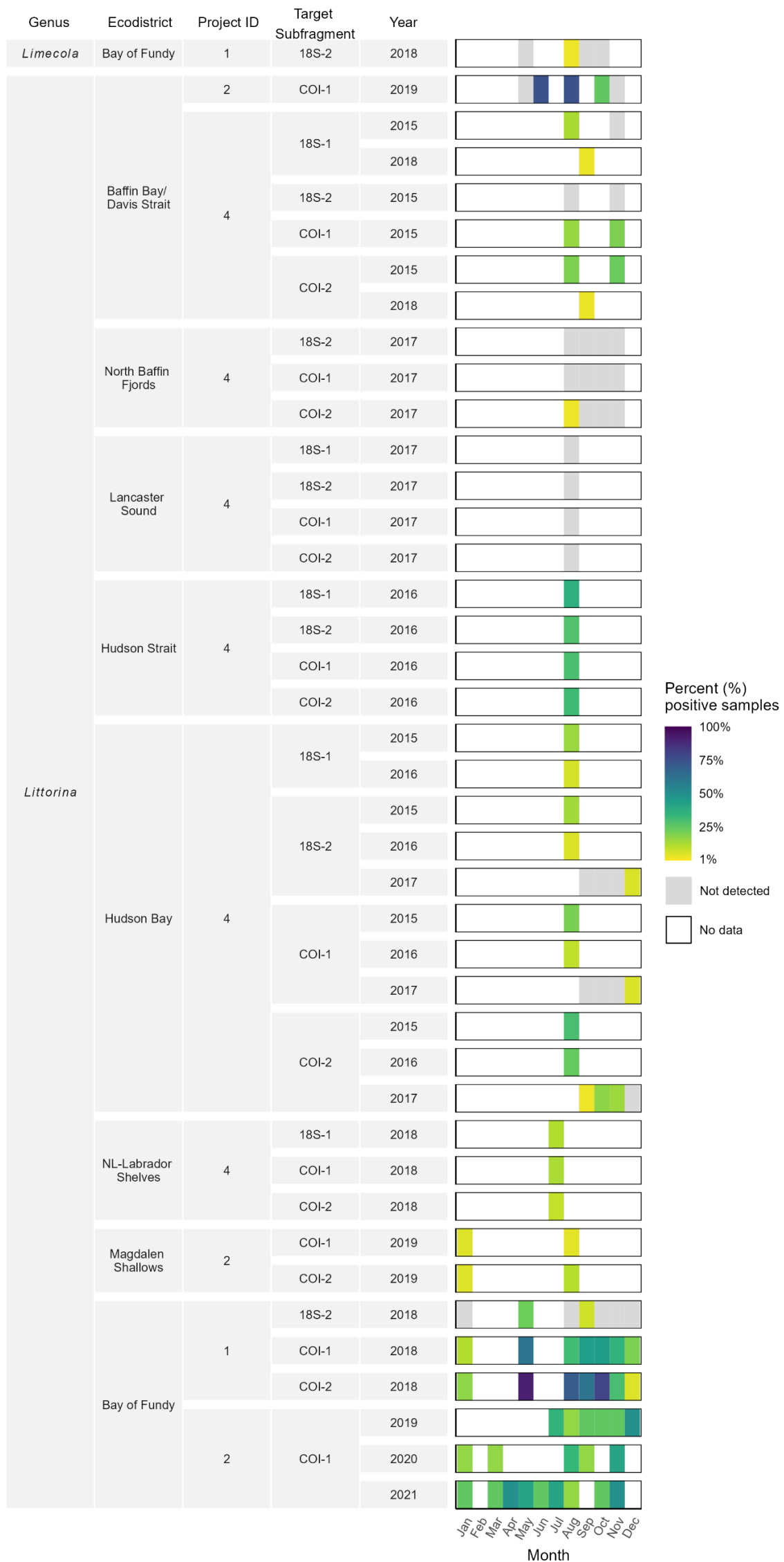


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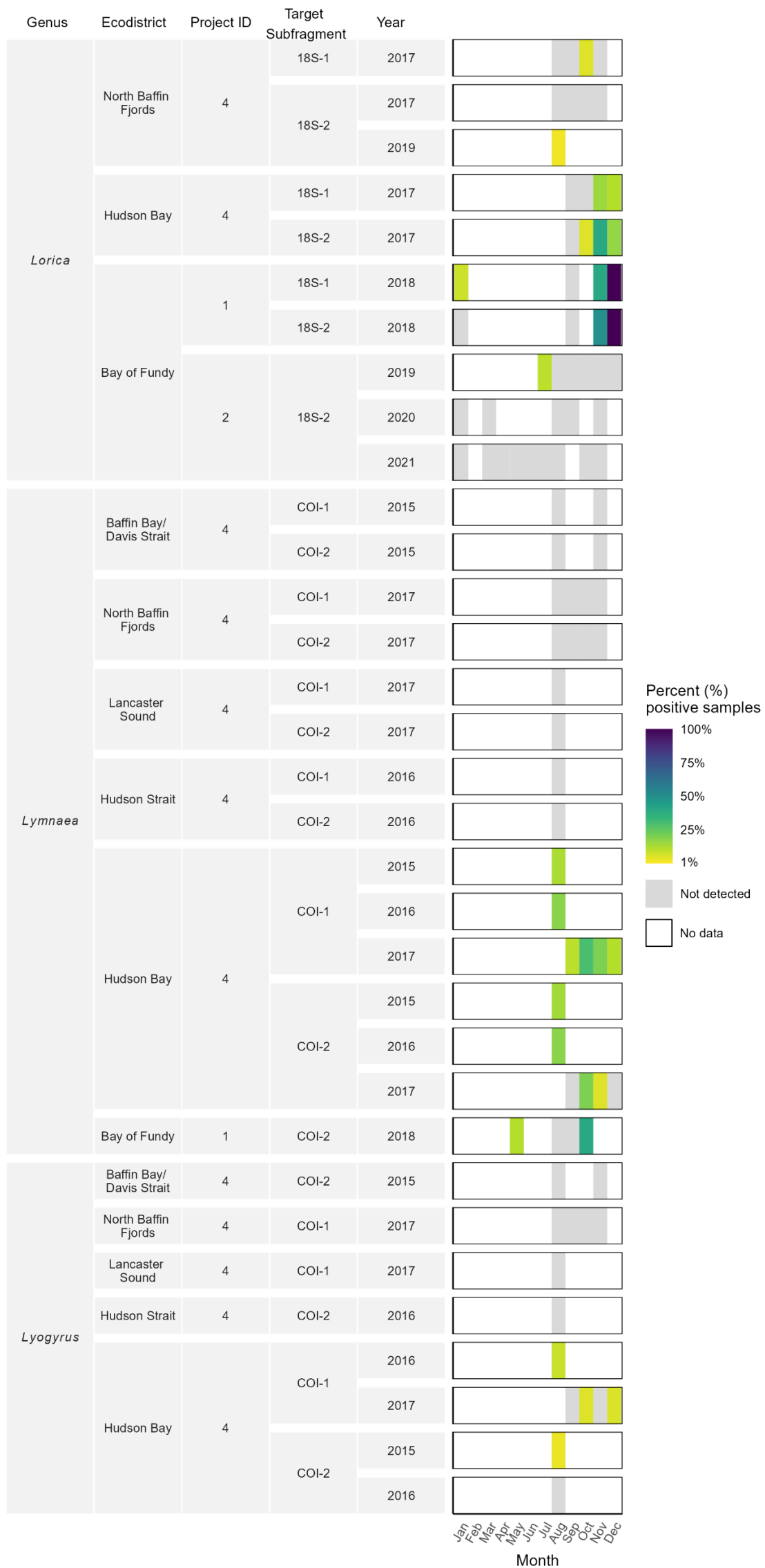


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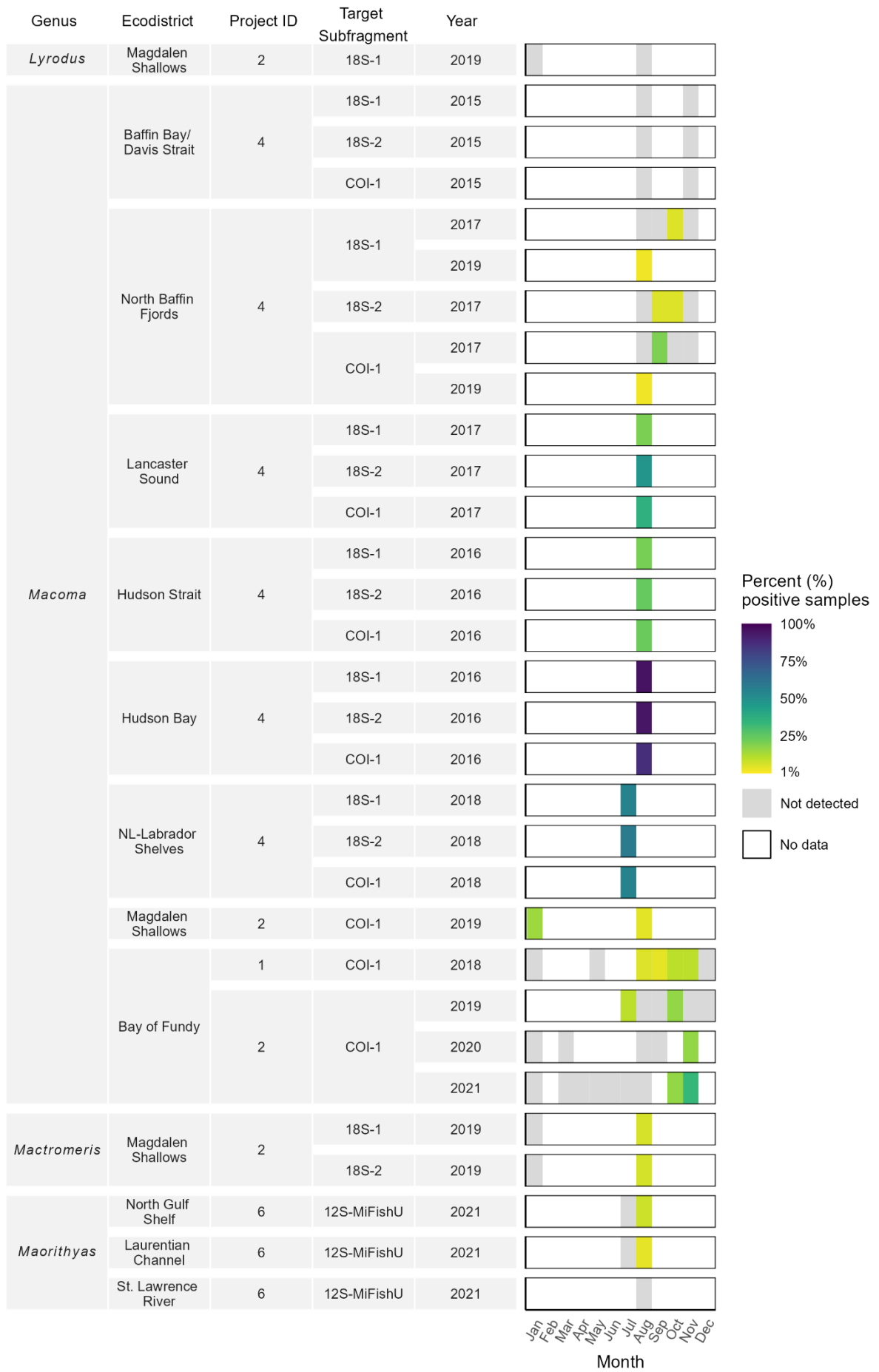


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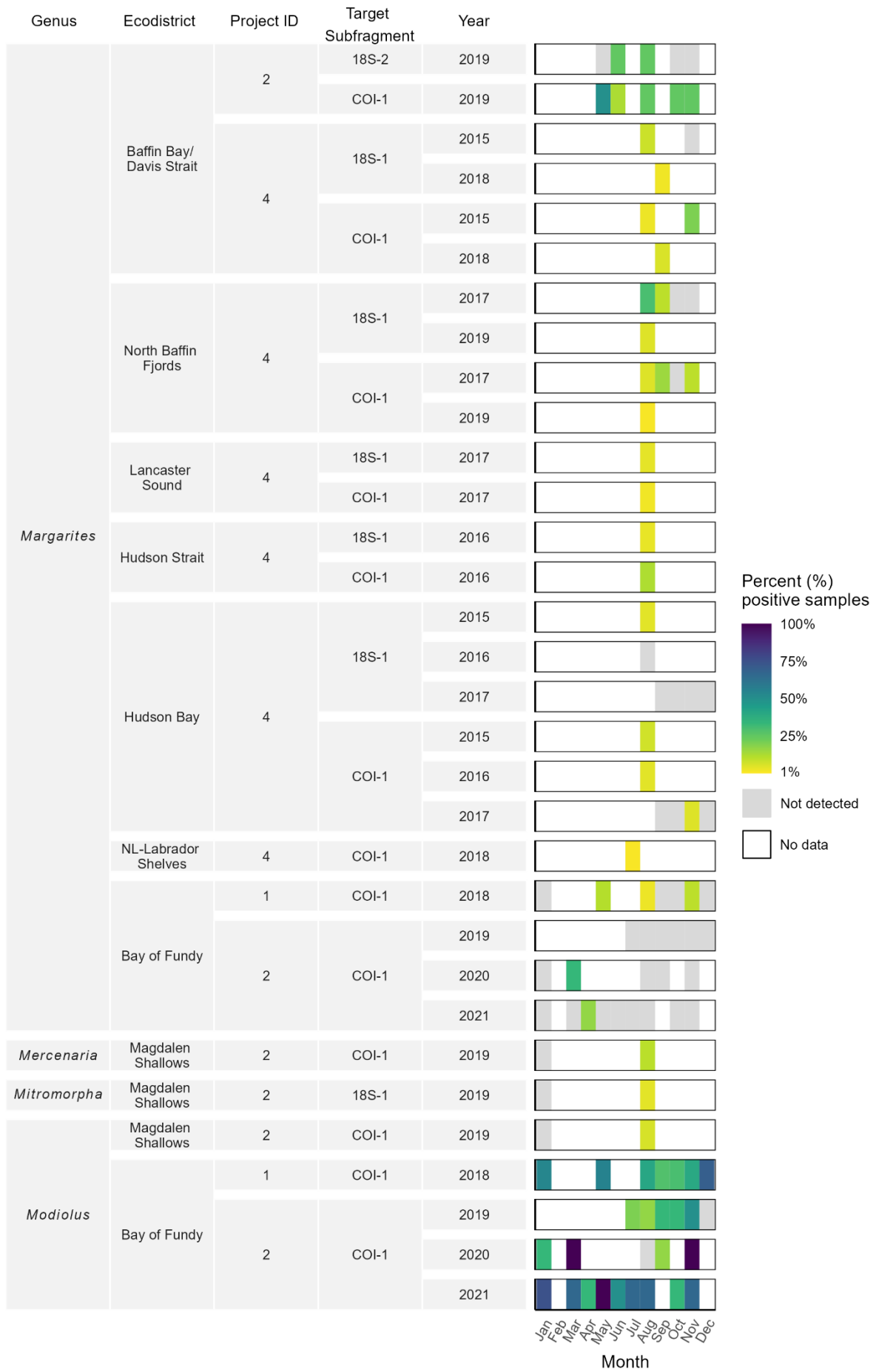


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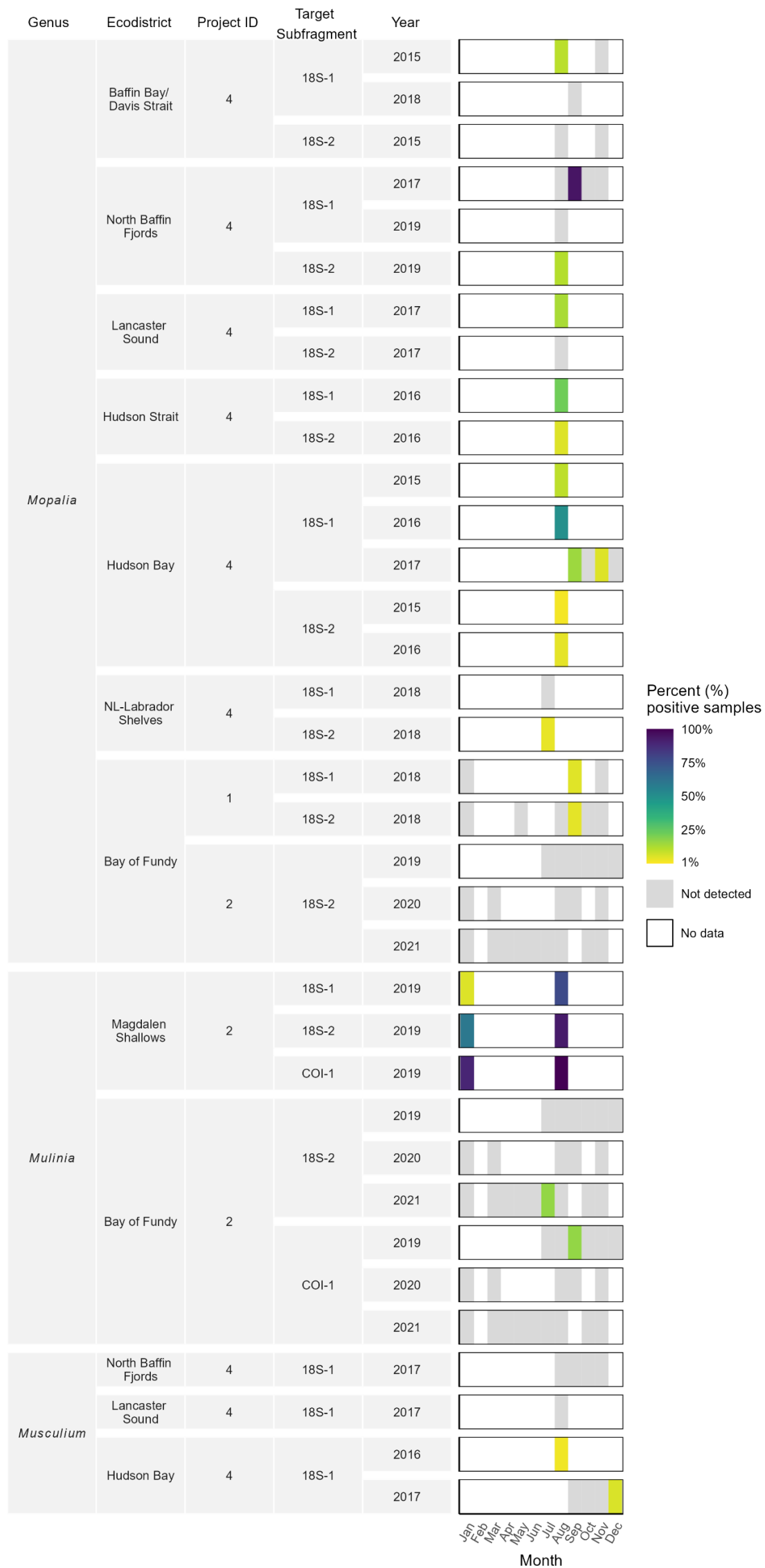


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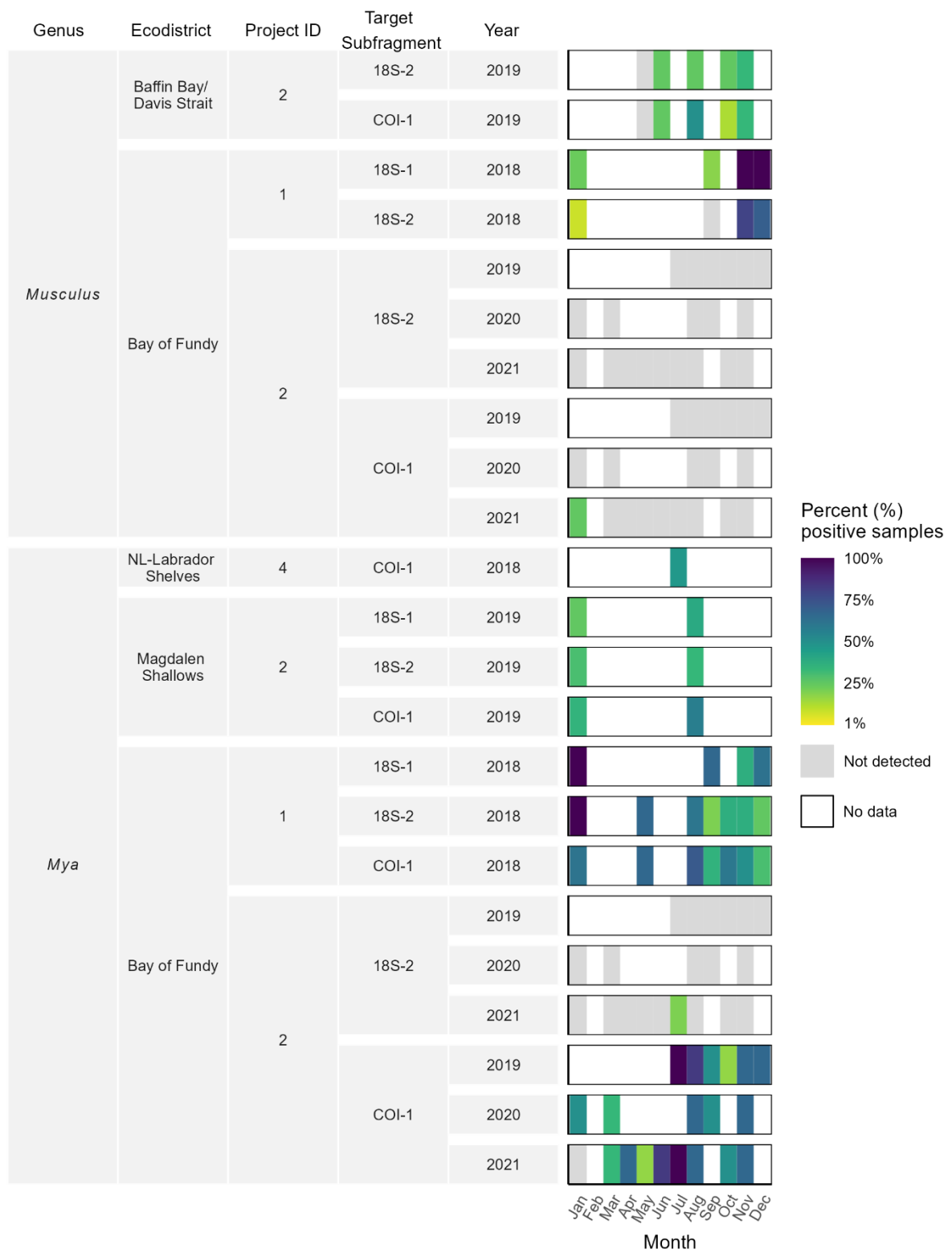
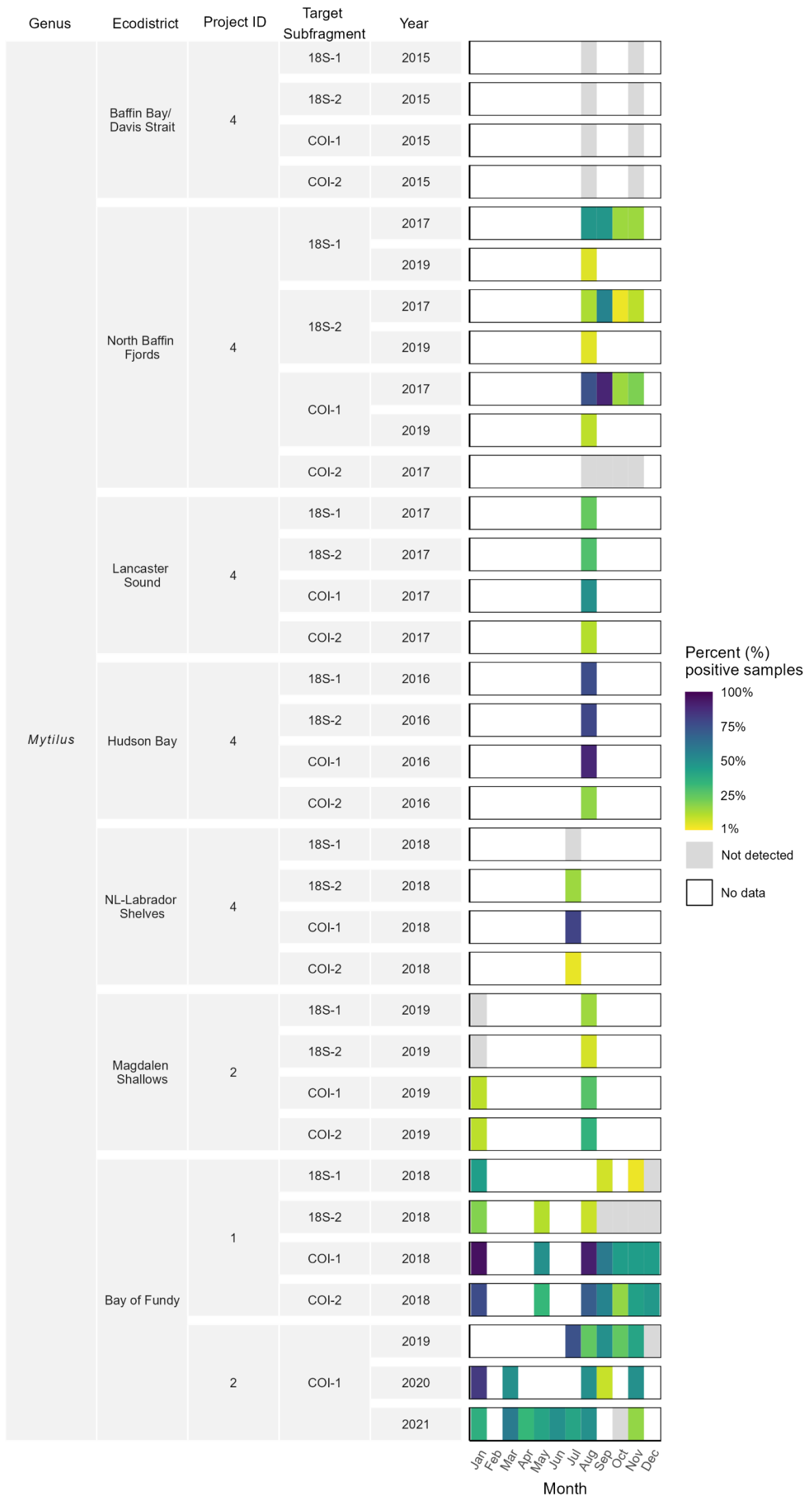


Figure 13. (Continued)



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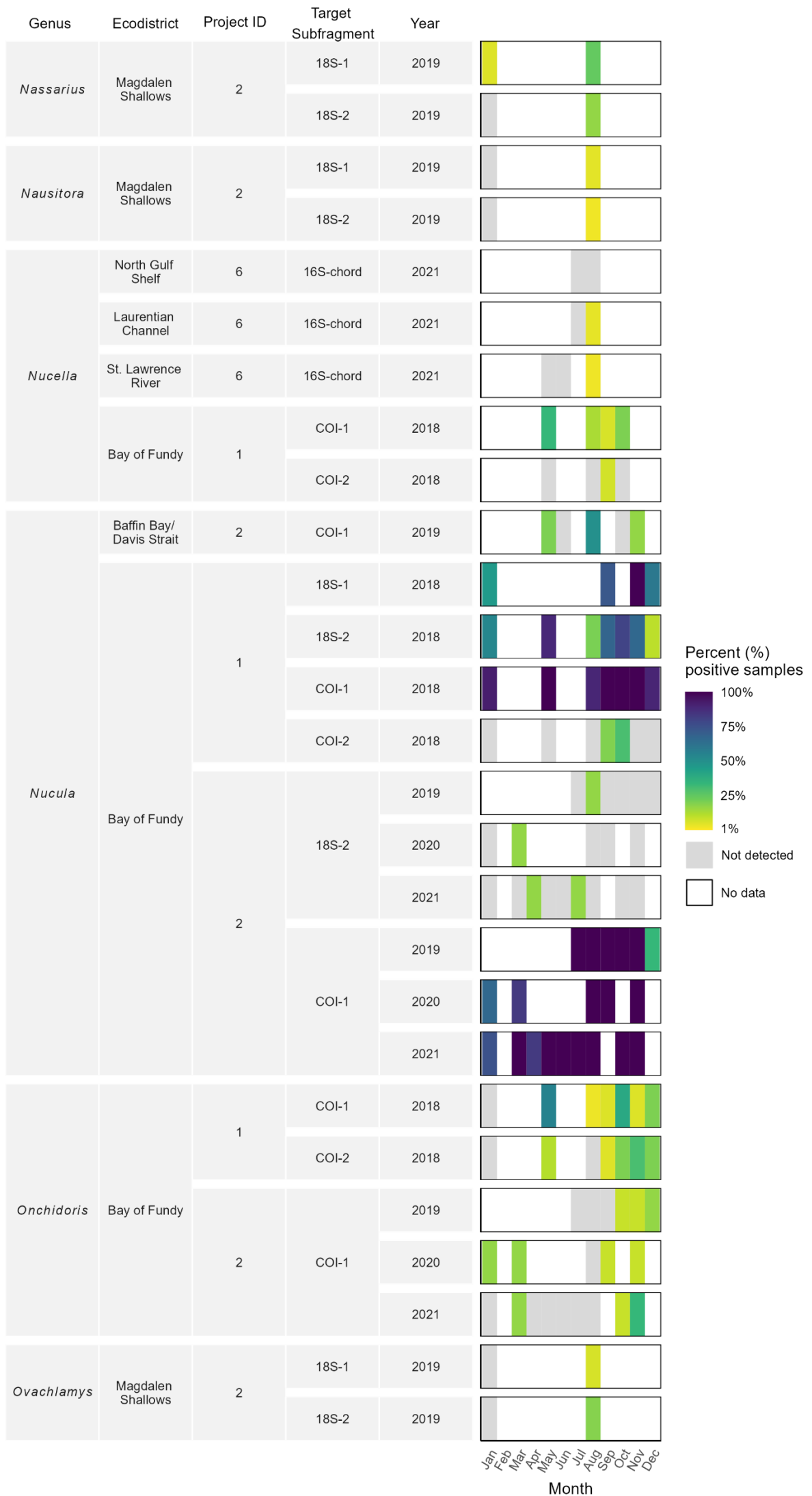


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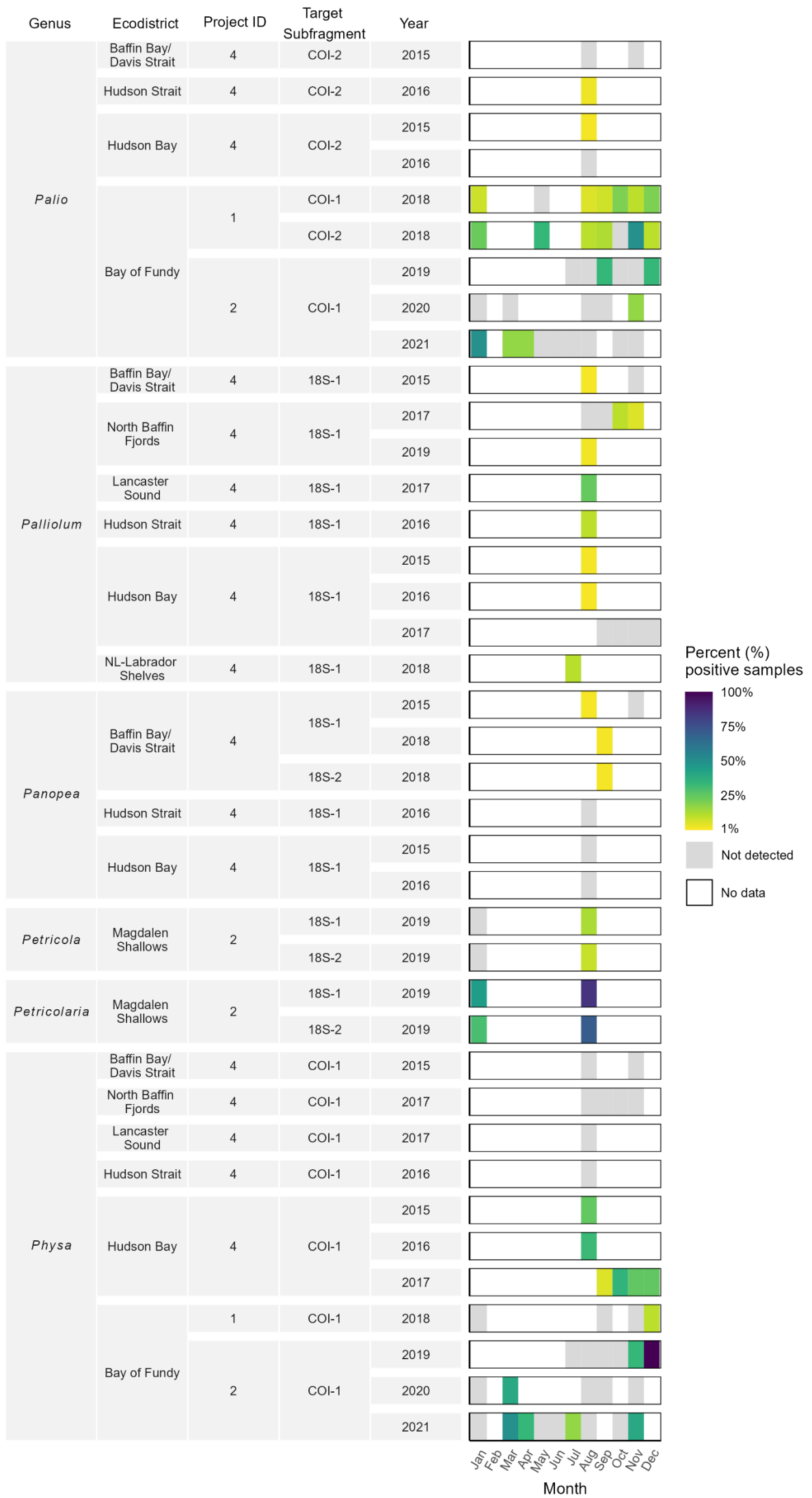


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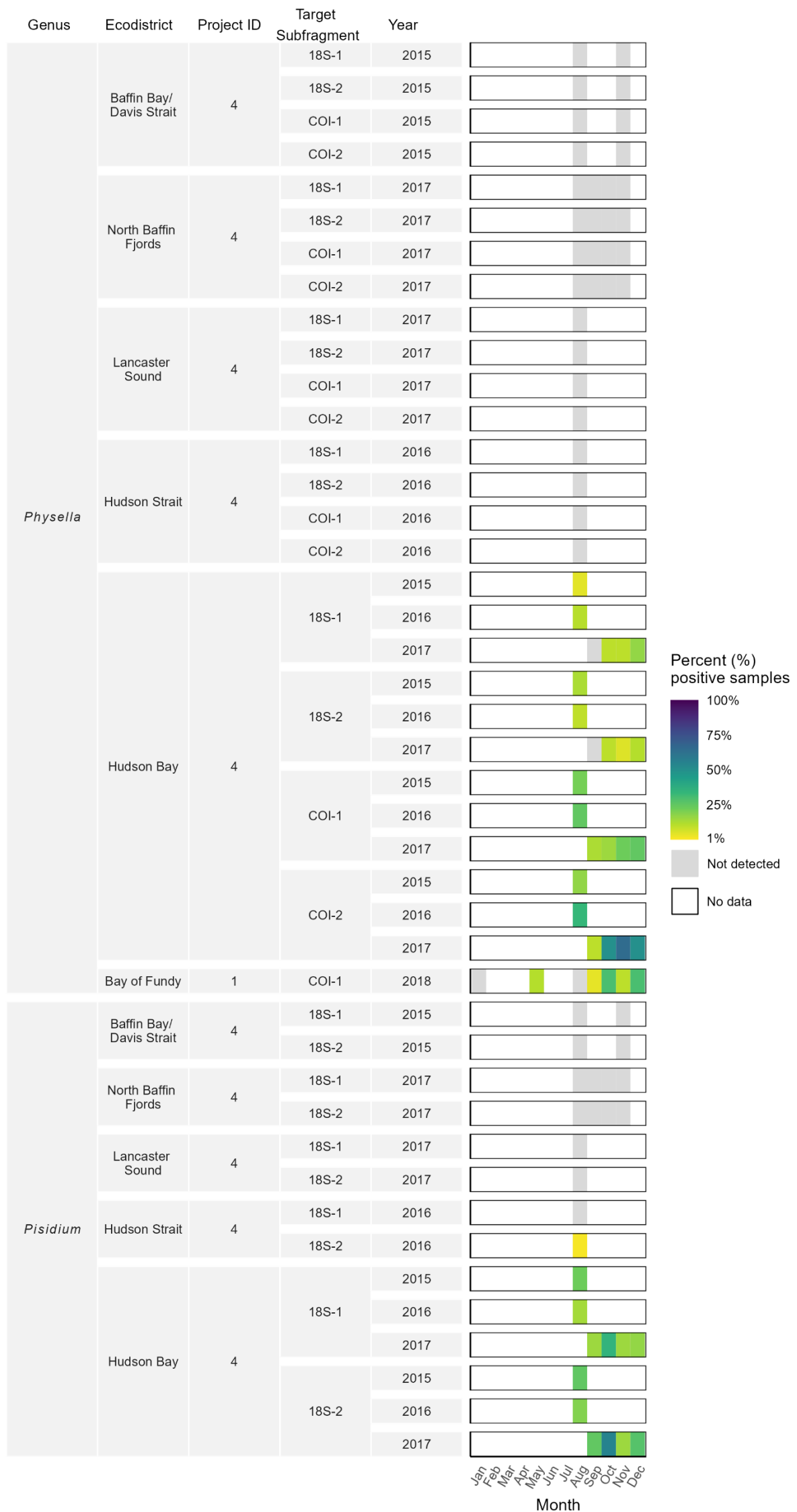


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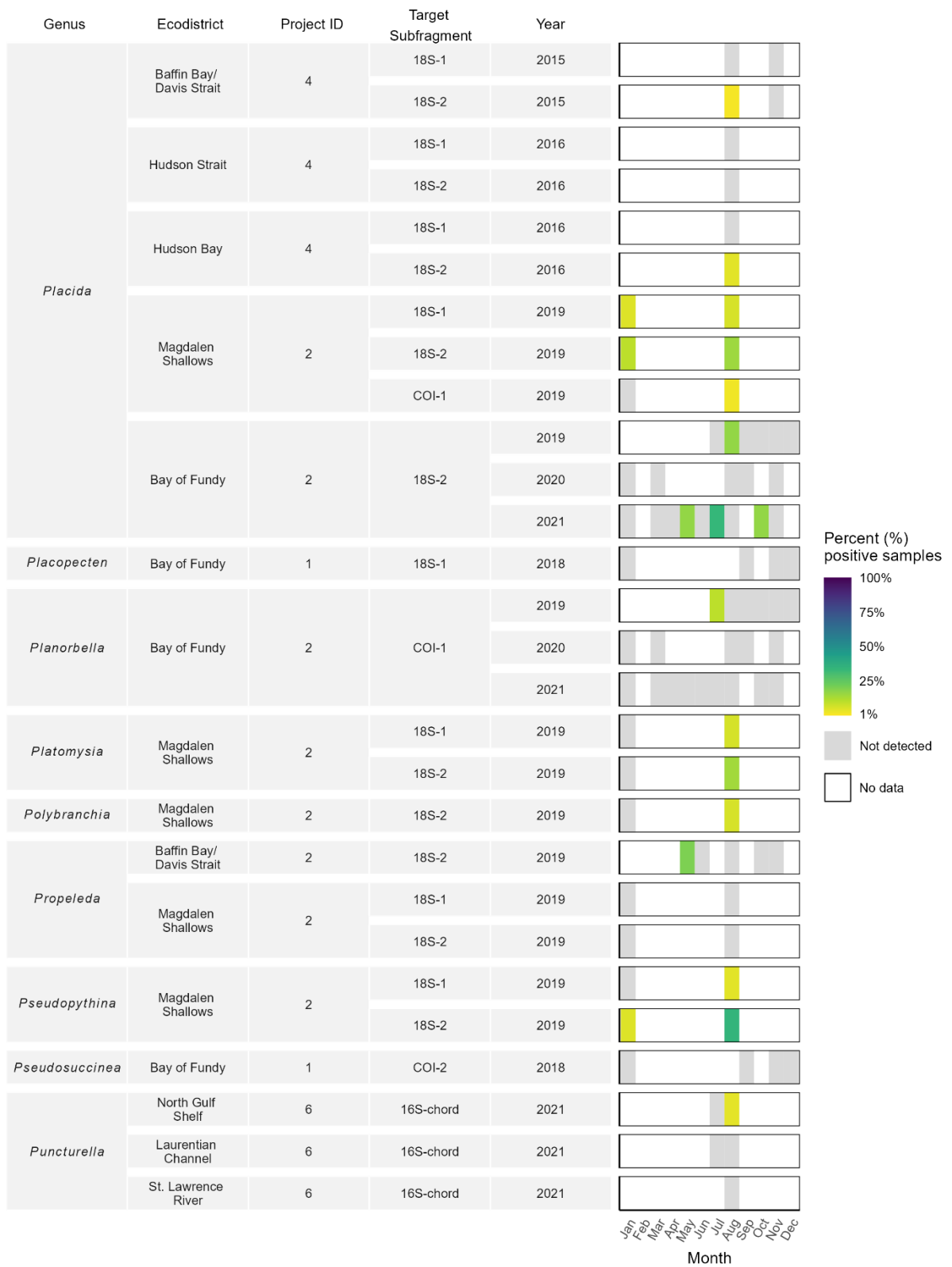


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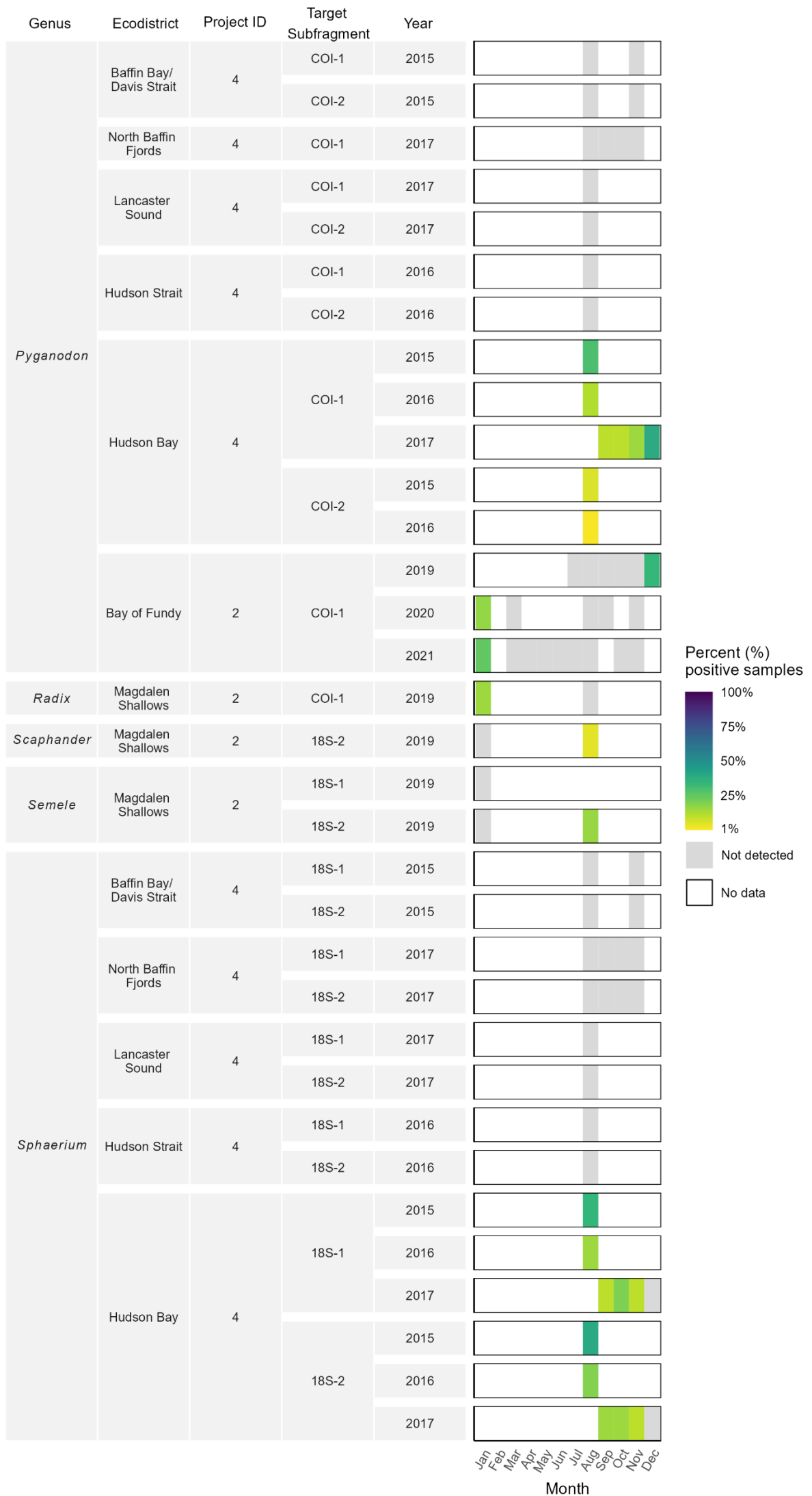


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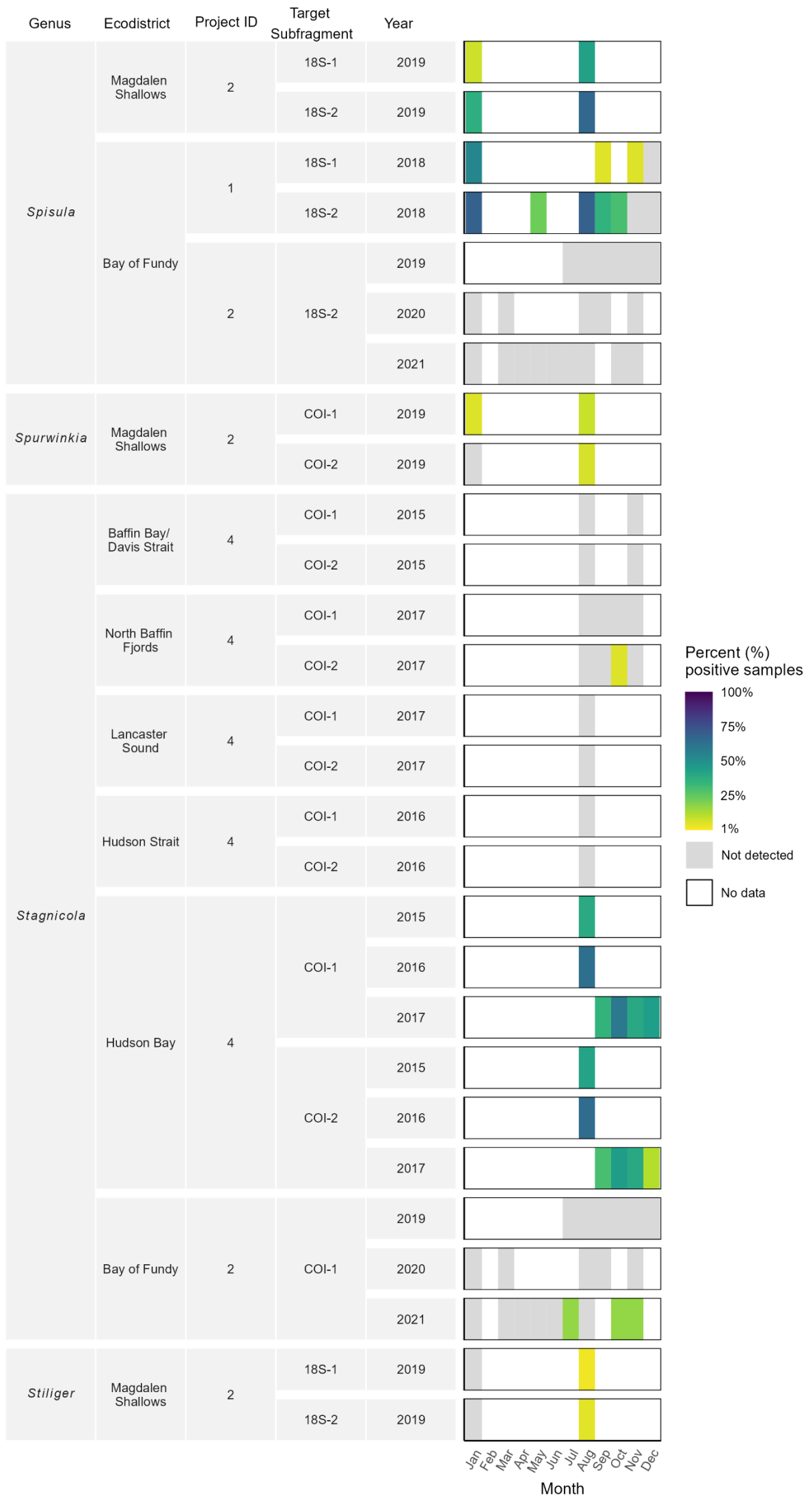


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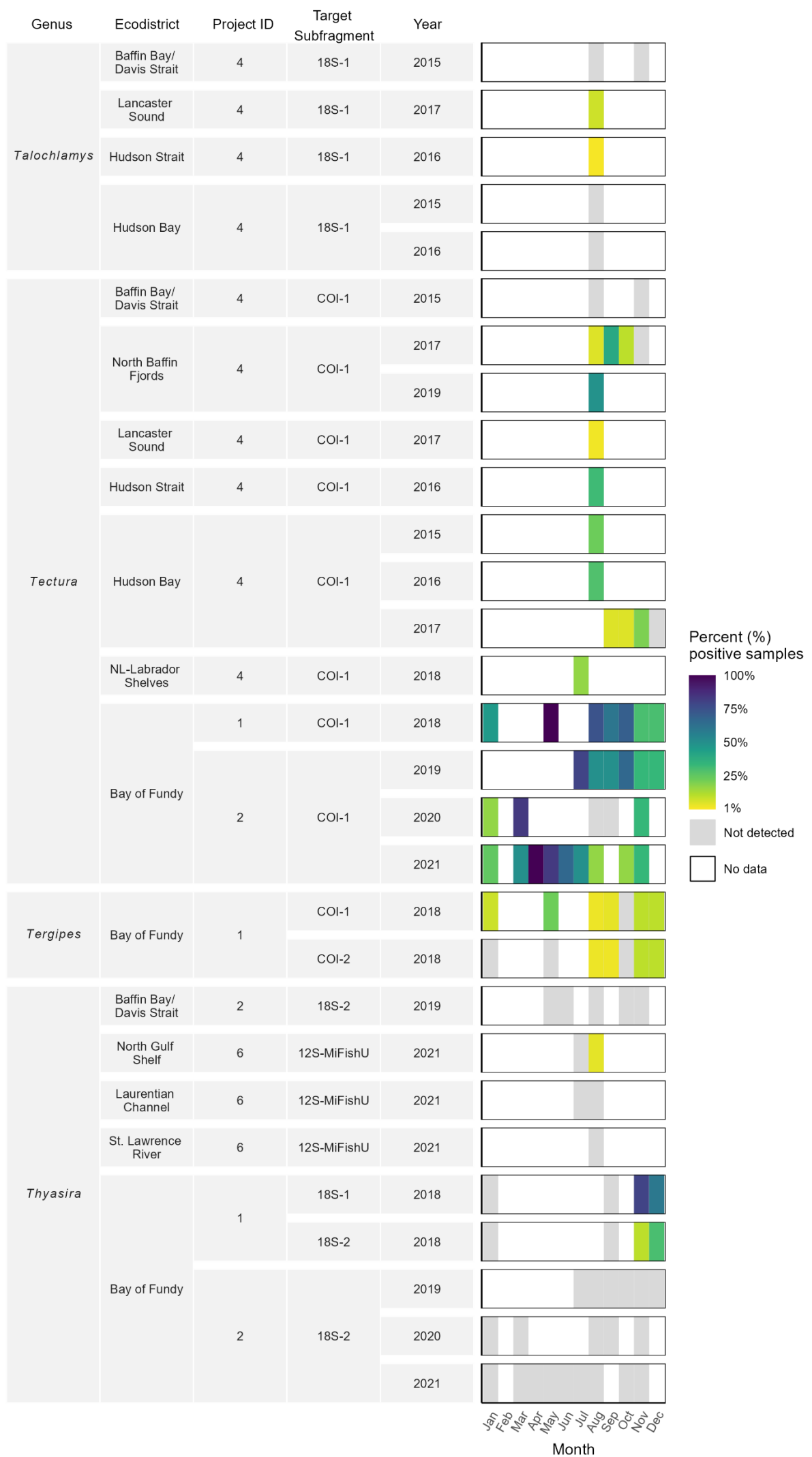


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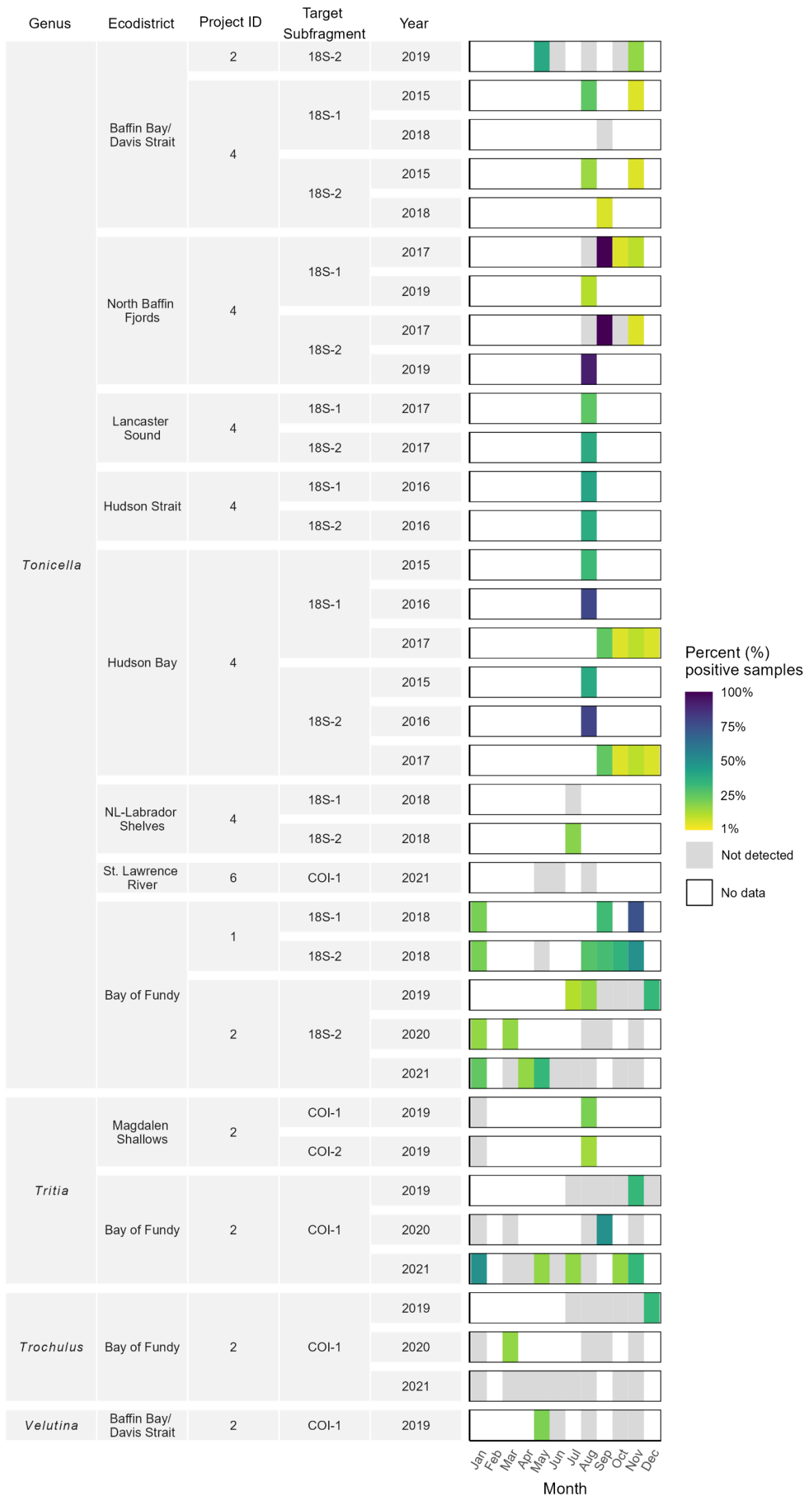


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6.15 MYZOOZOA

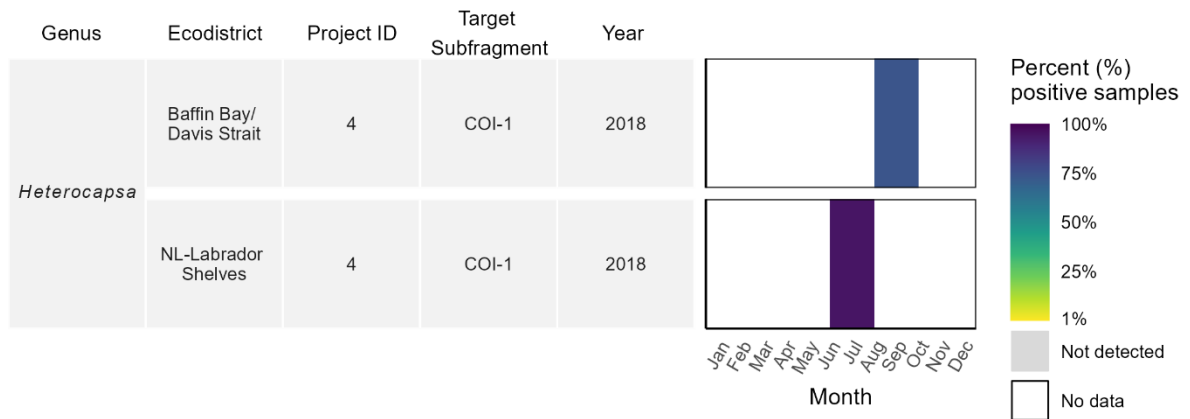


Figure 13. (Continued)

6.16 NEMATODA



Figure 13. (Continued)



Figure 13. (Continued)

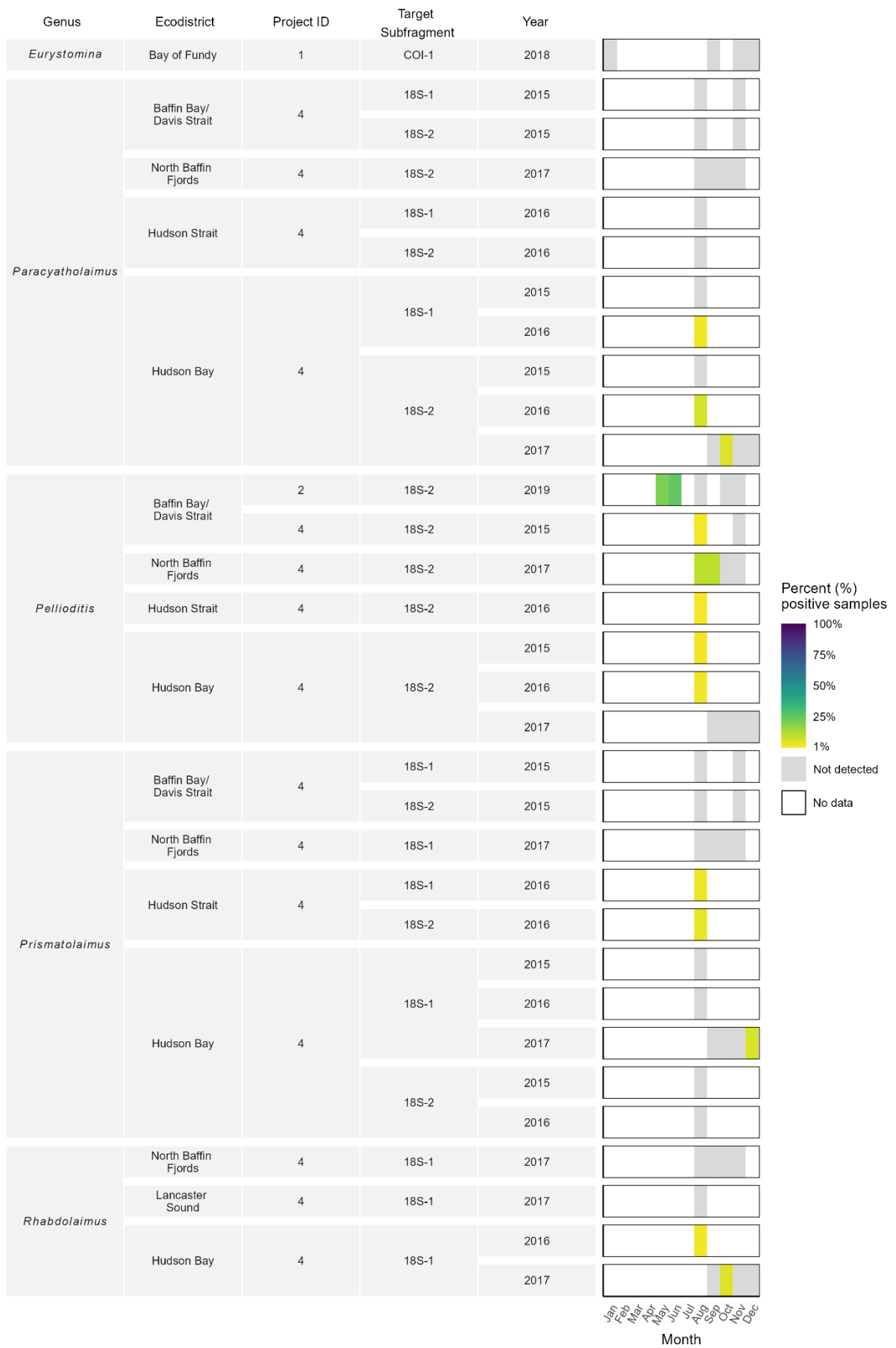


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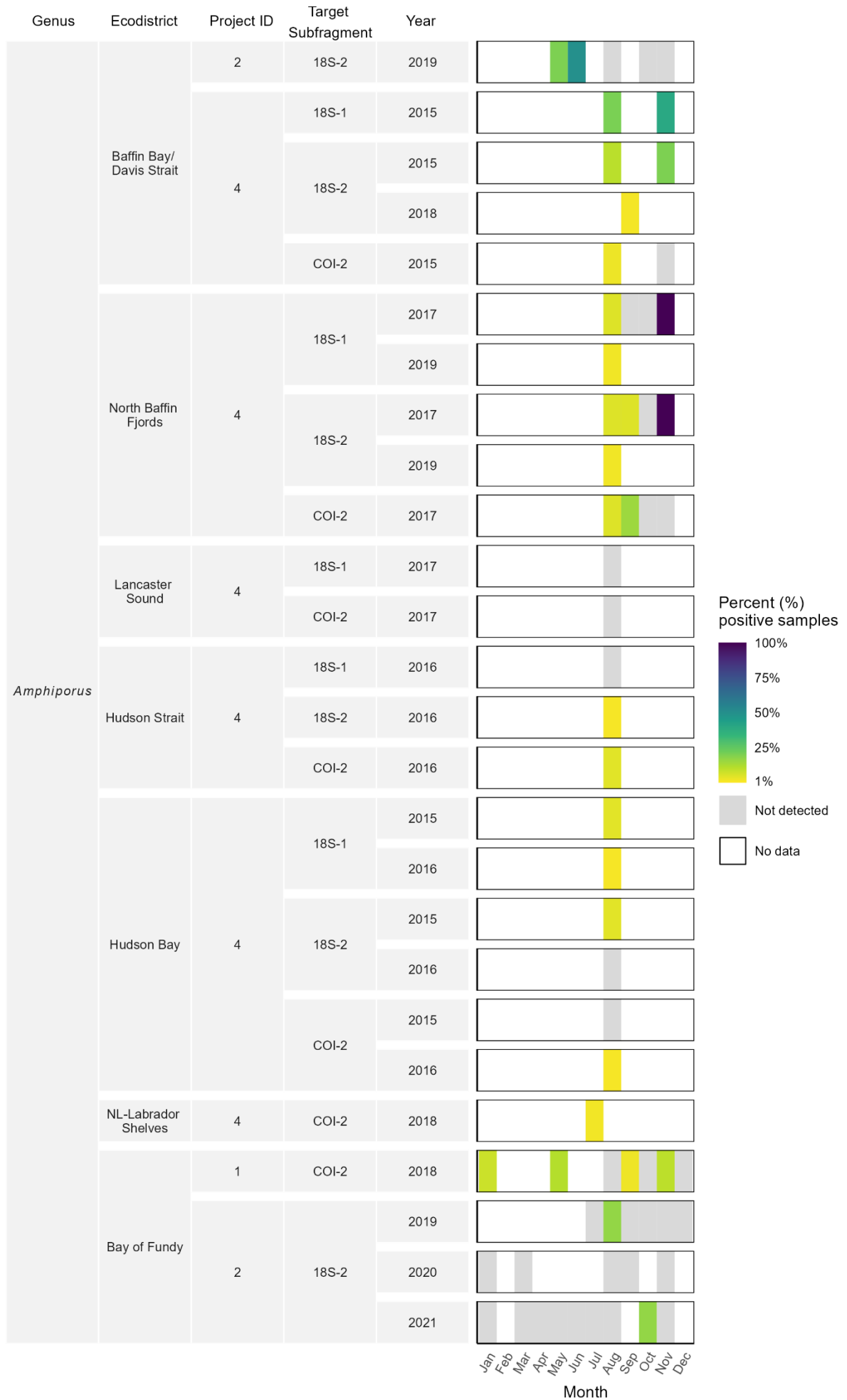


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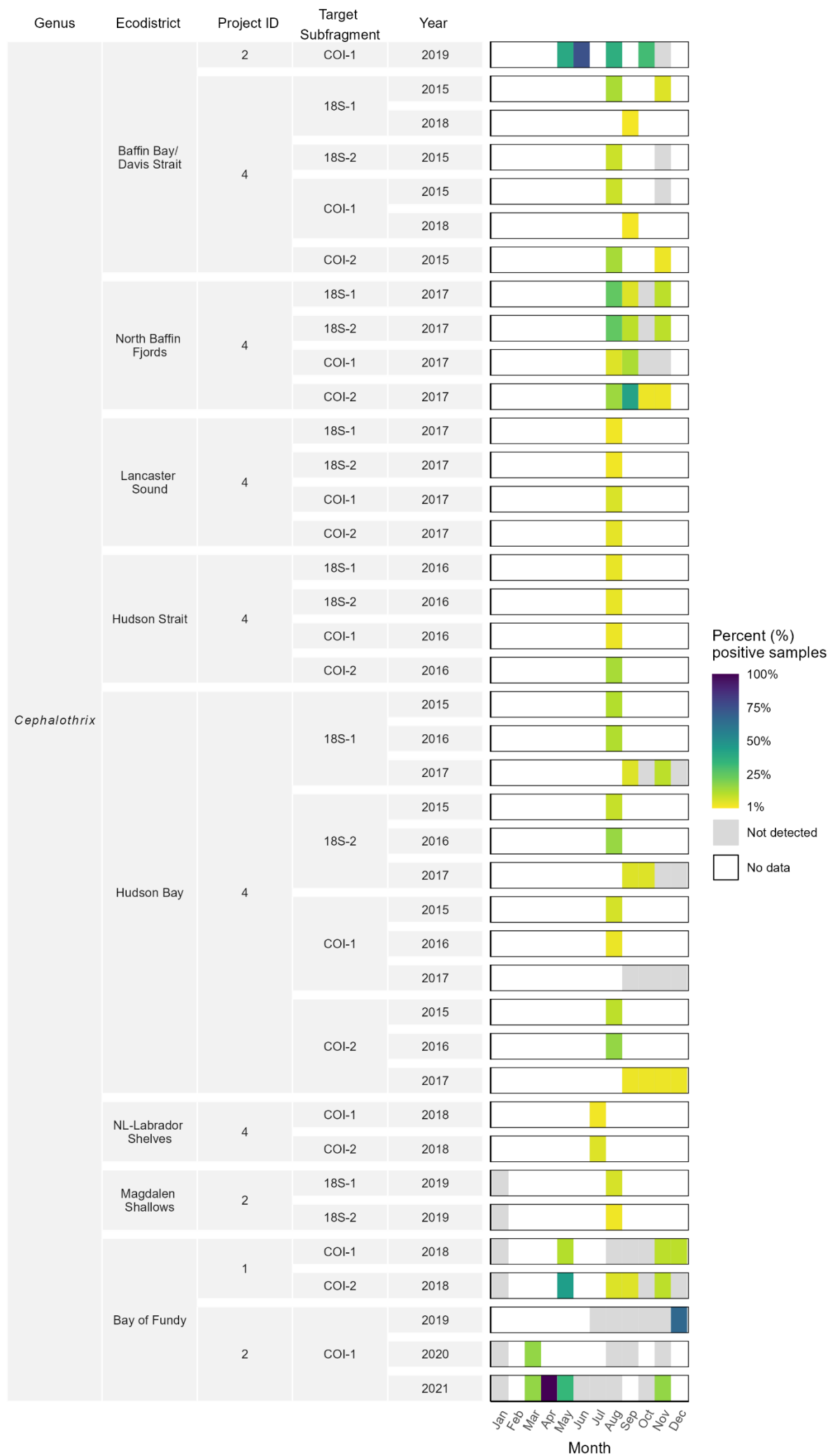


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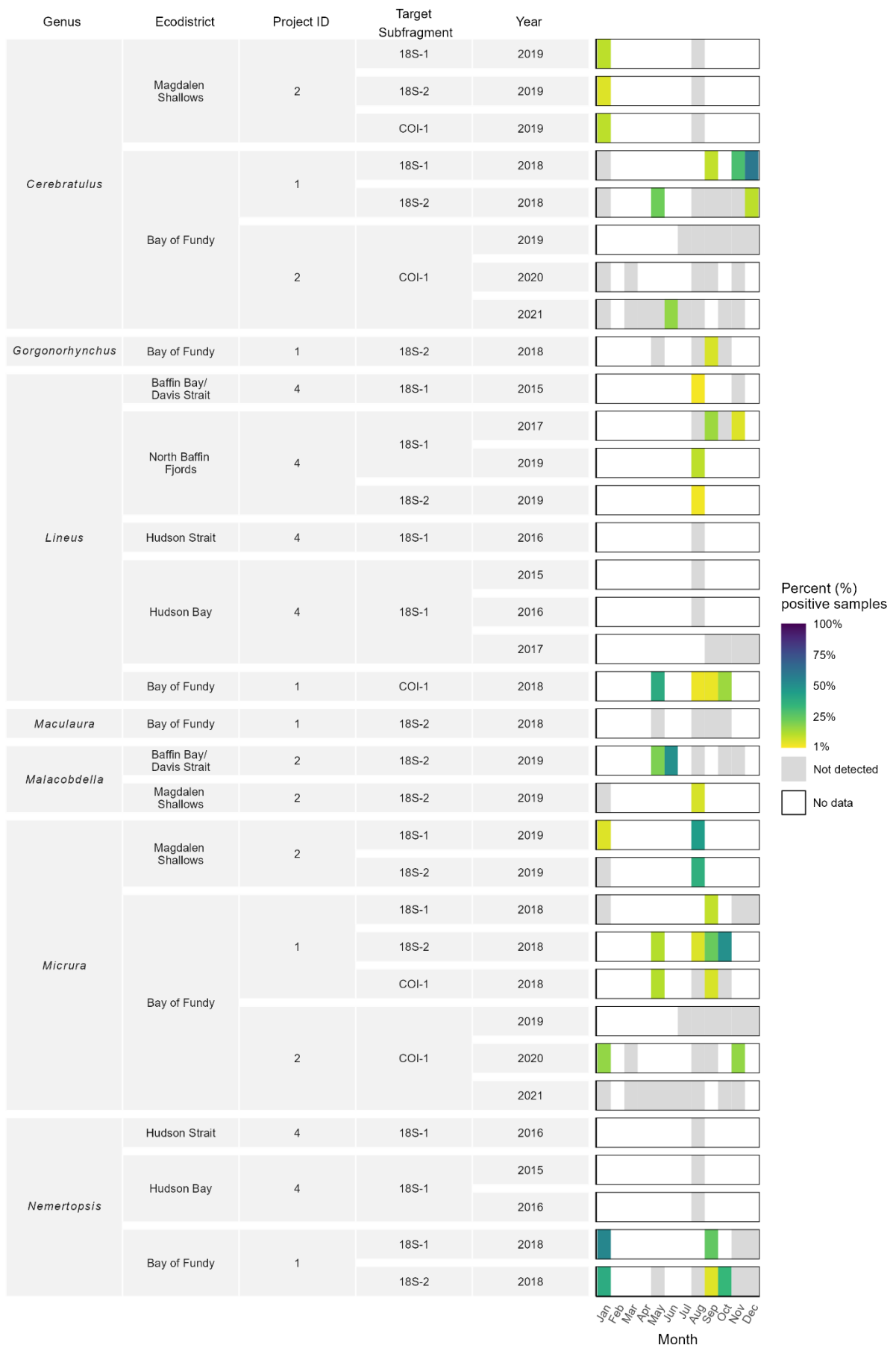


Figure 13. (Continued)



Figure 13. (Continued)

6.18 PLATYHELMINTHES



Figure 13. (Continued)



Figure 13. (Continued)

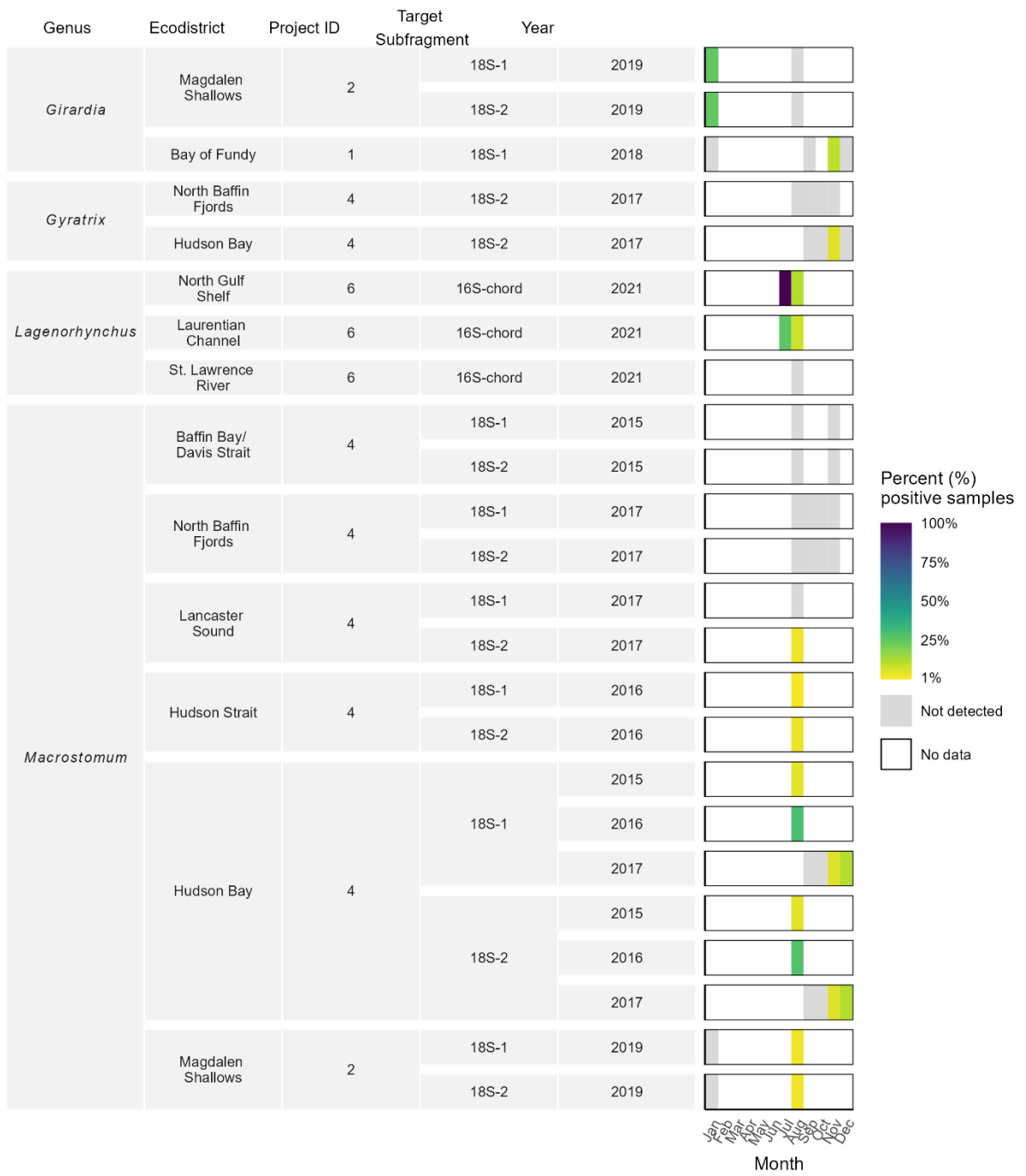


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Figure 13. (Continued)

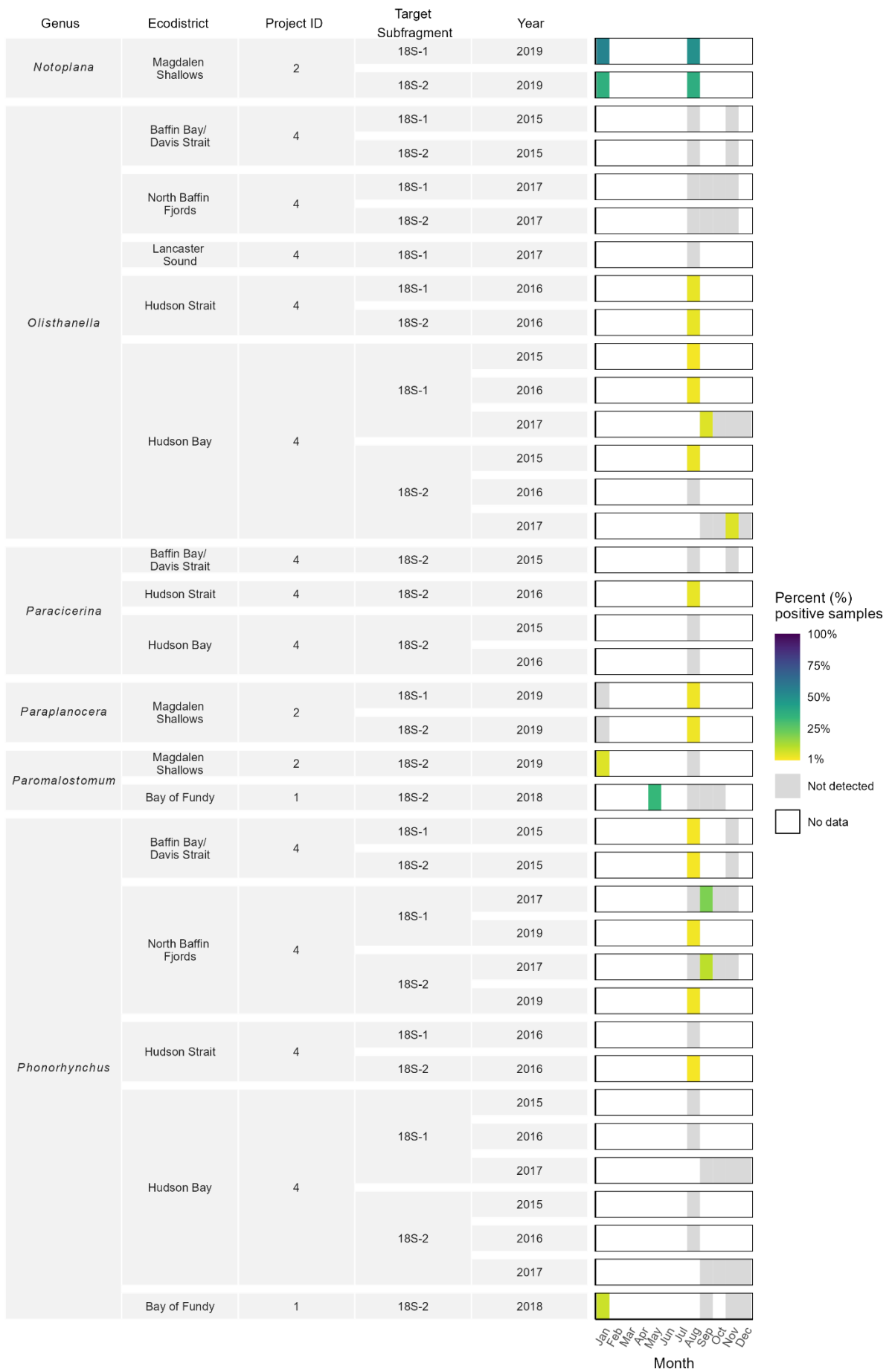


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Figure 13. (Continued)

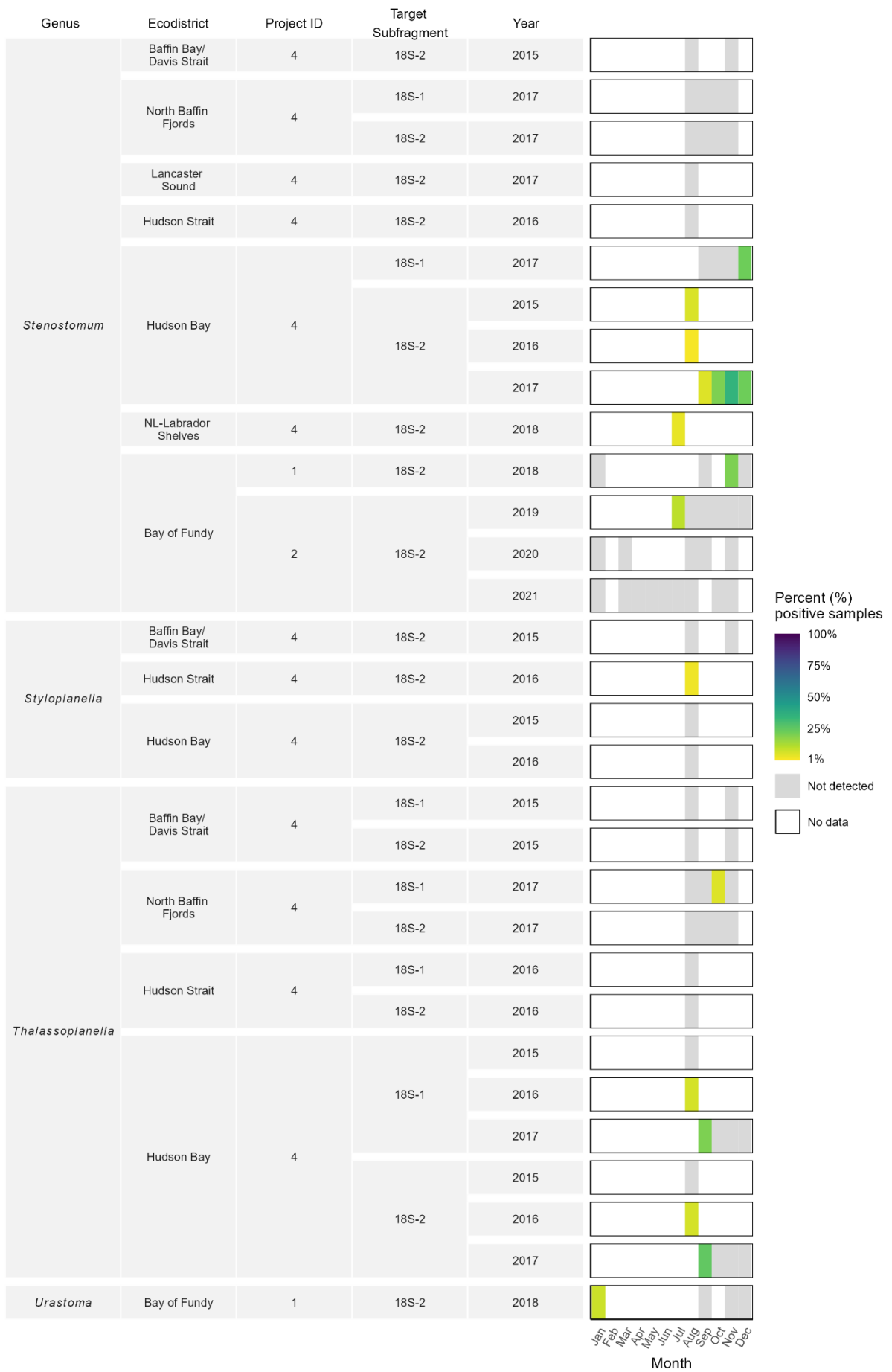


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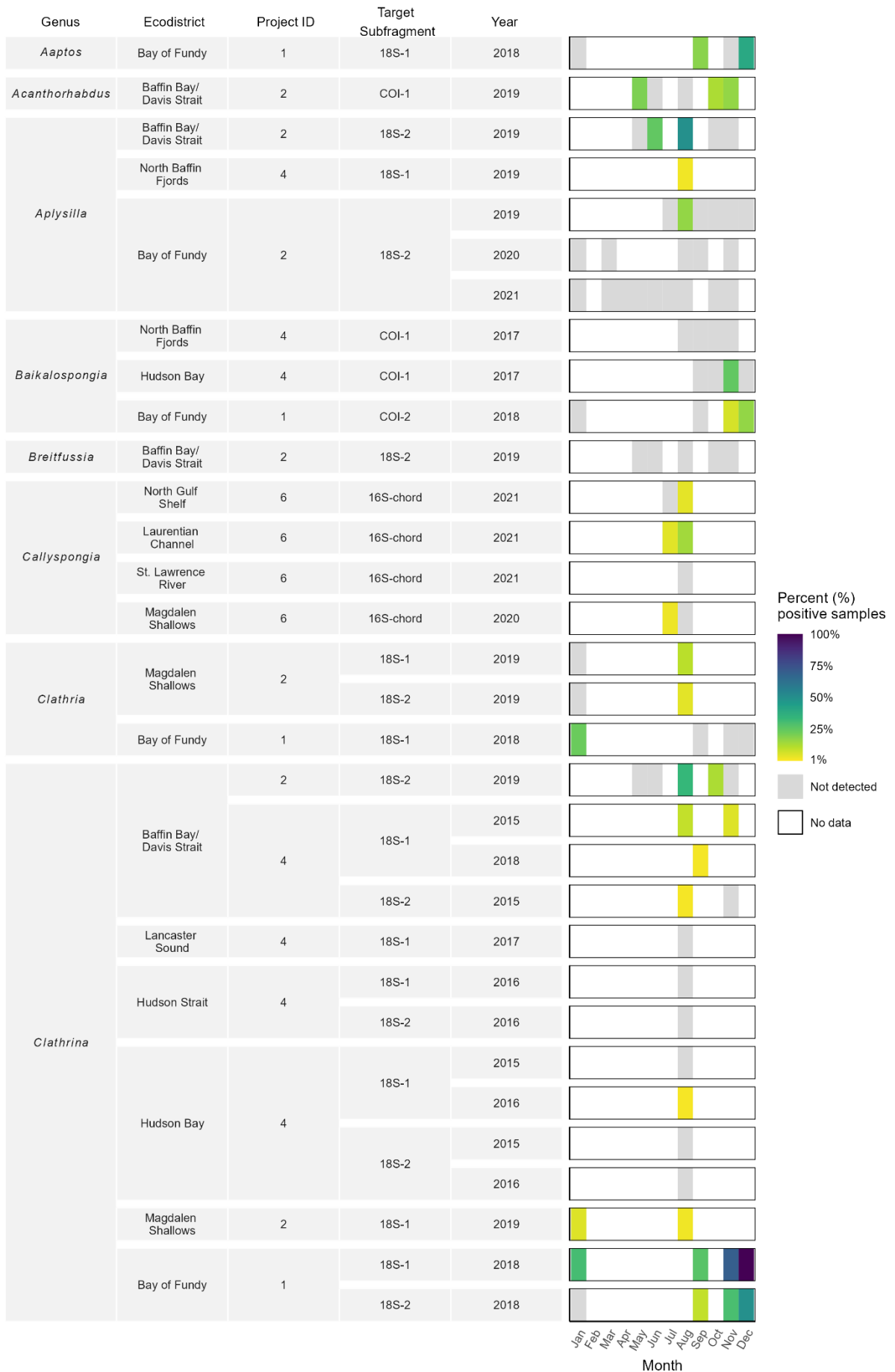


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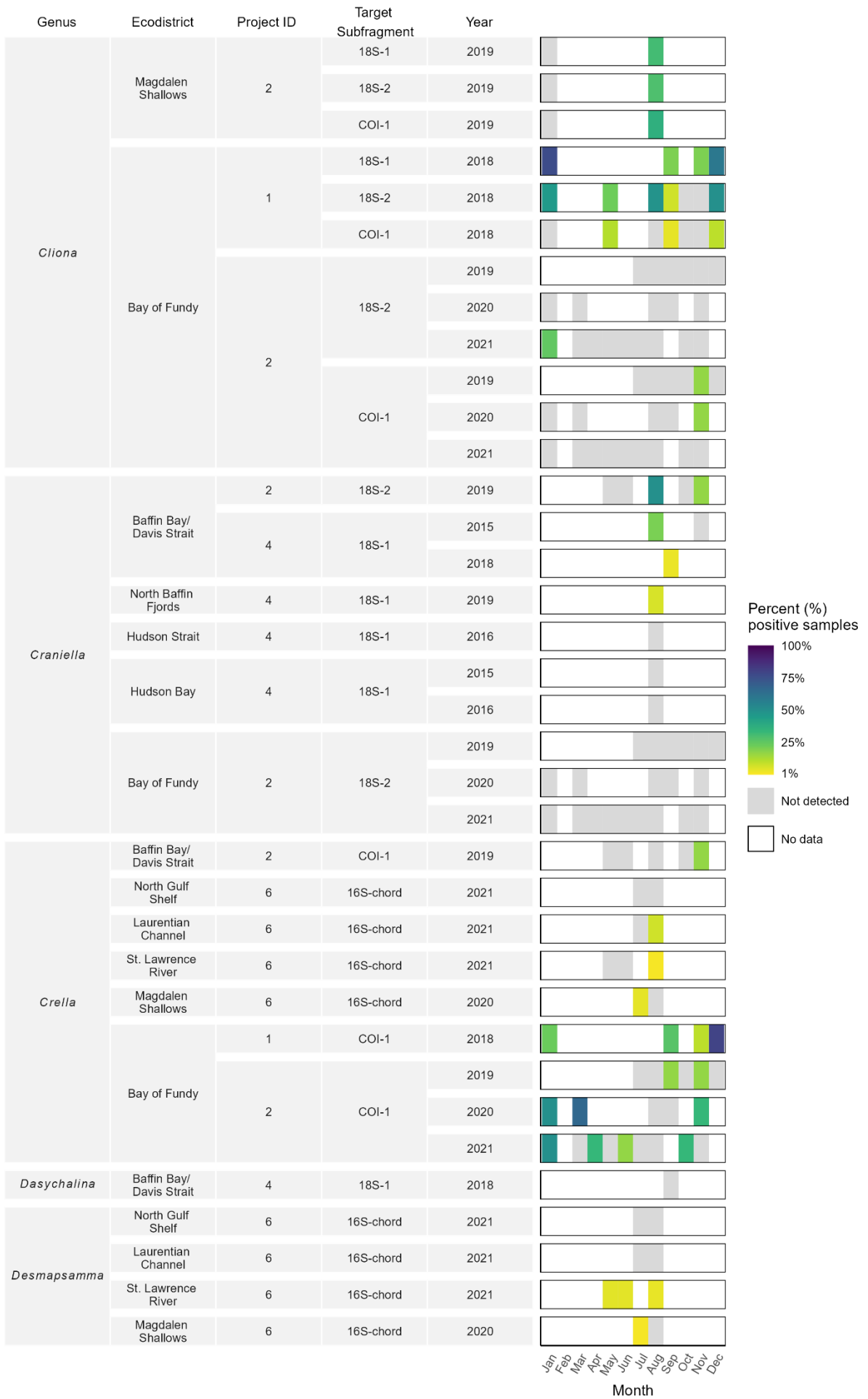


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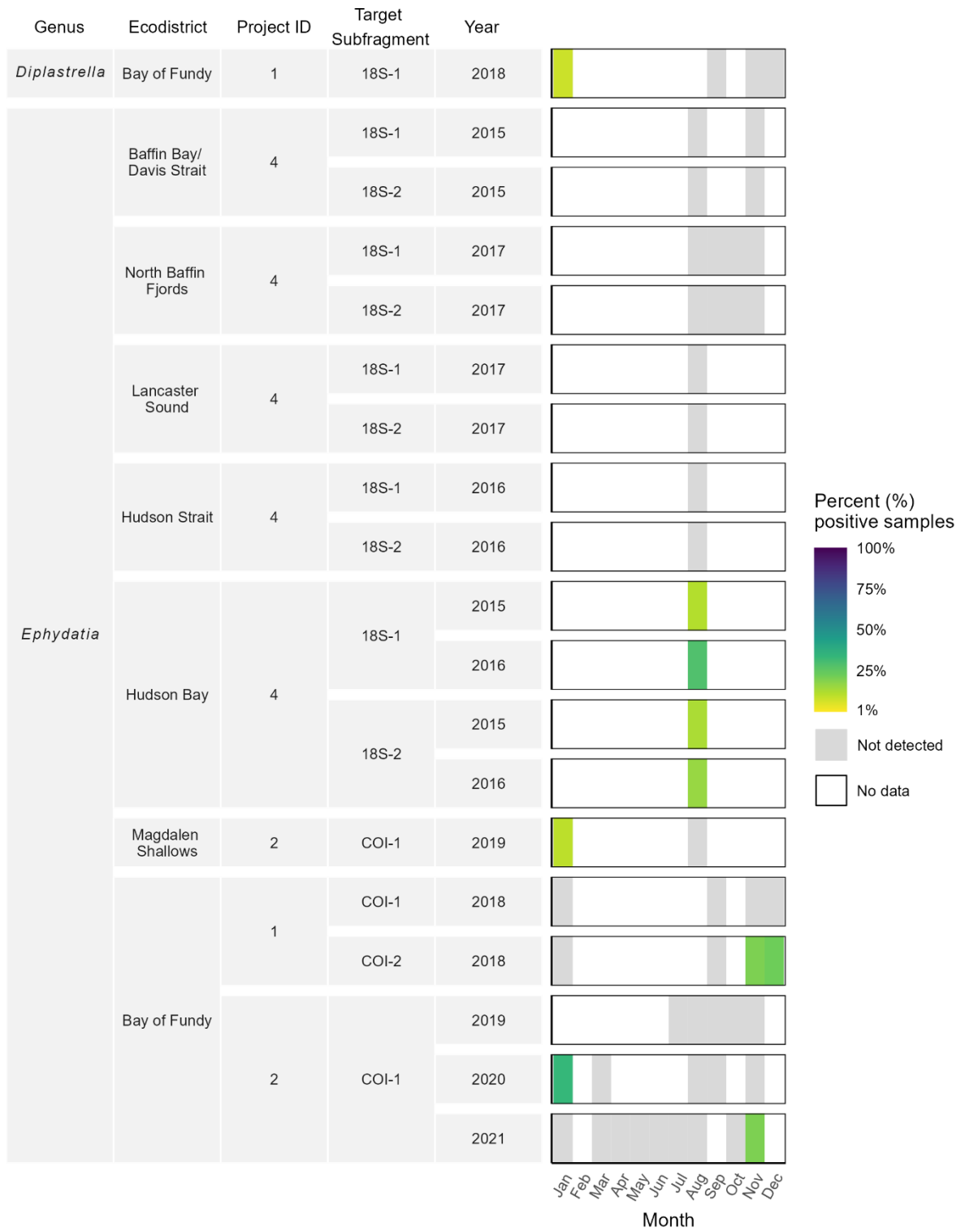


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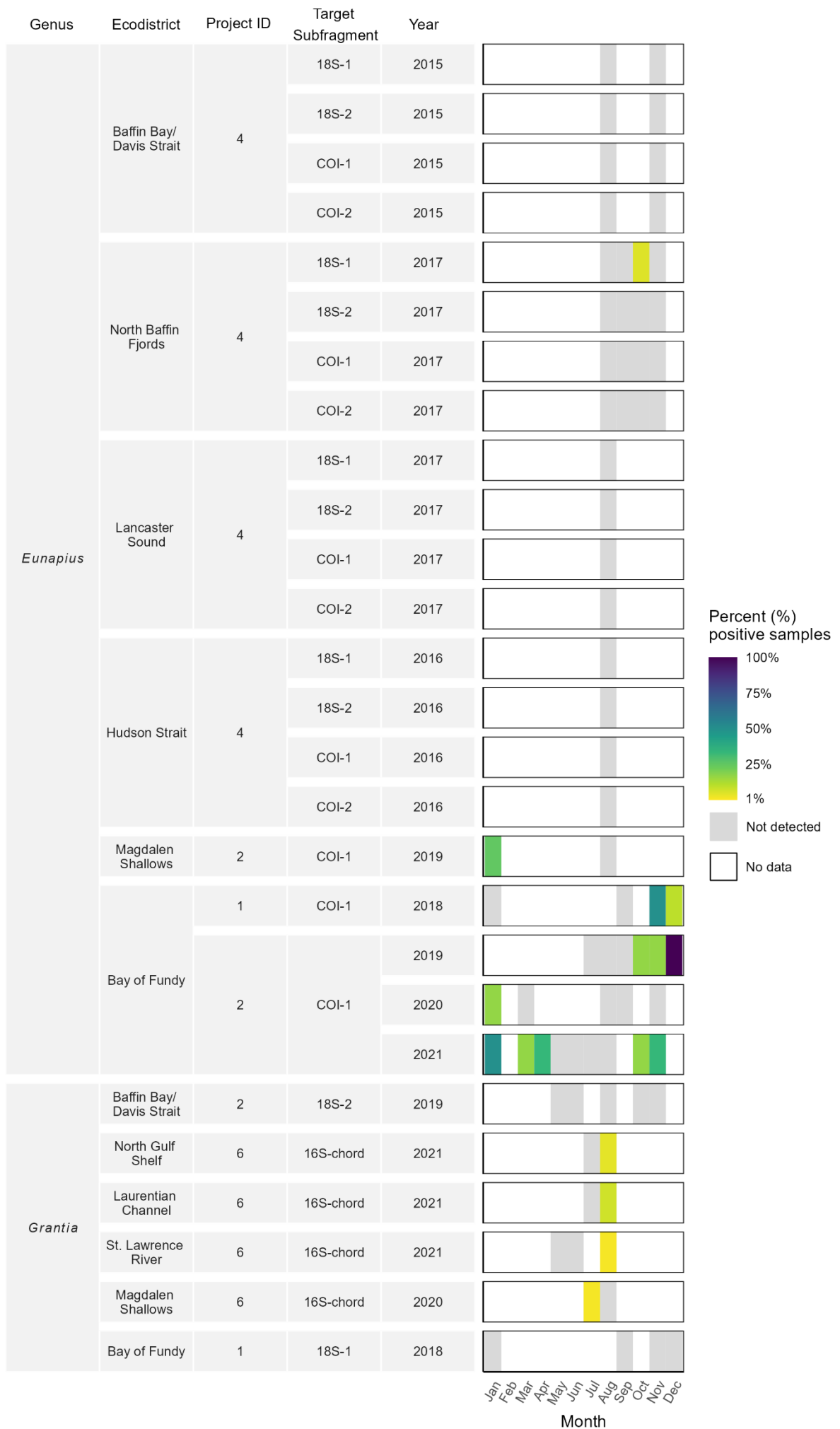


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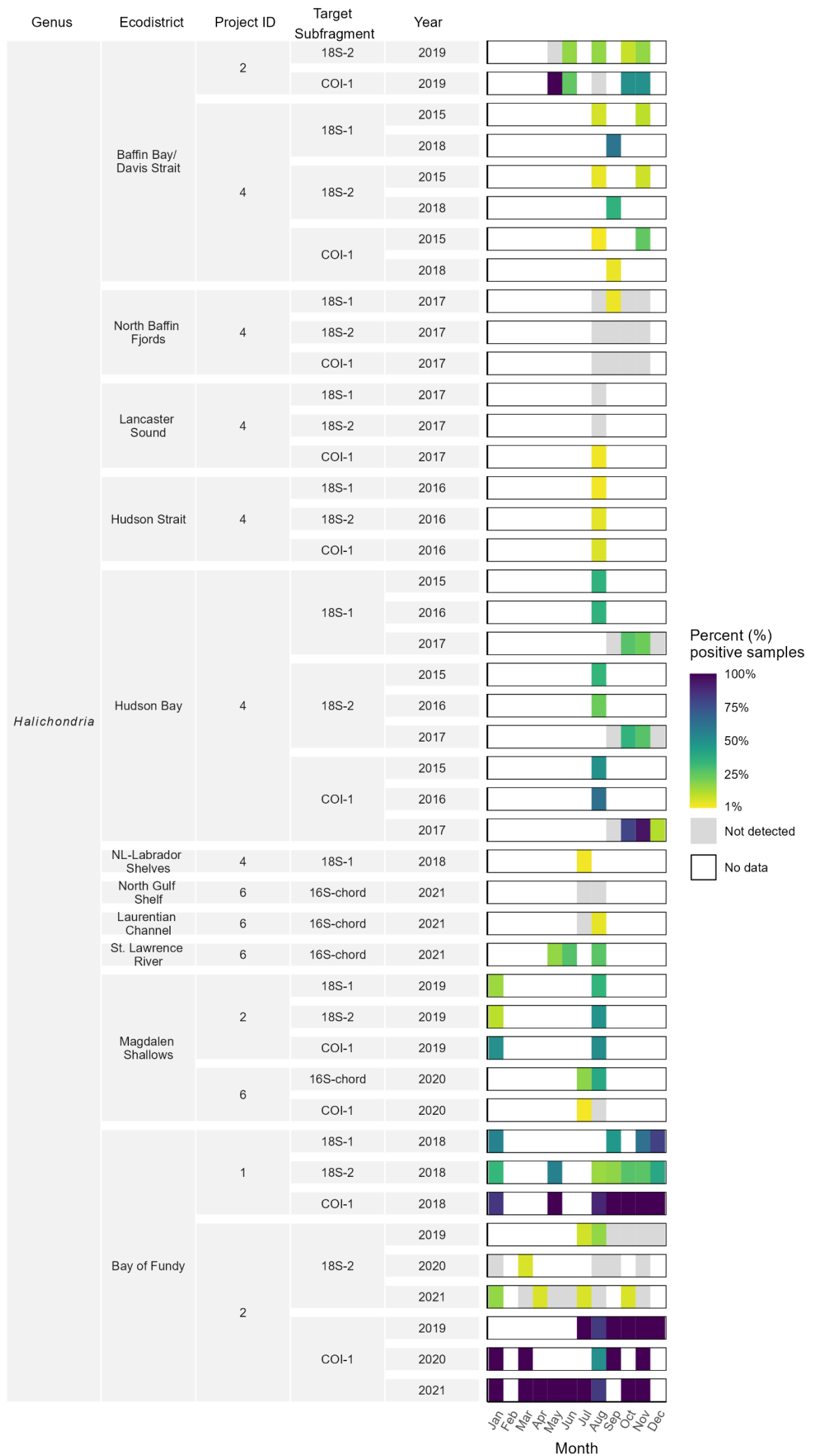


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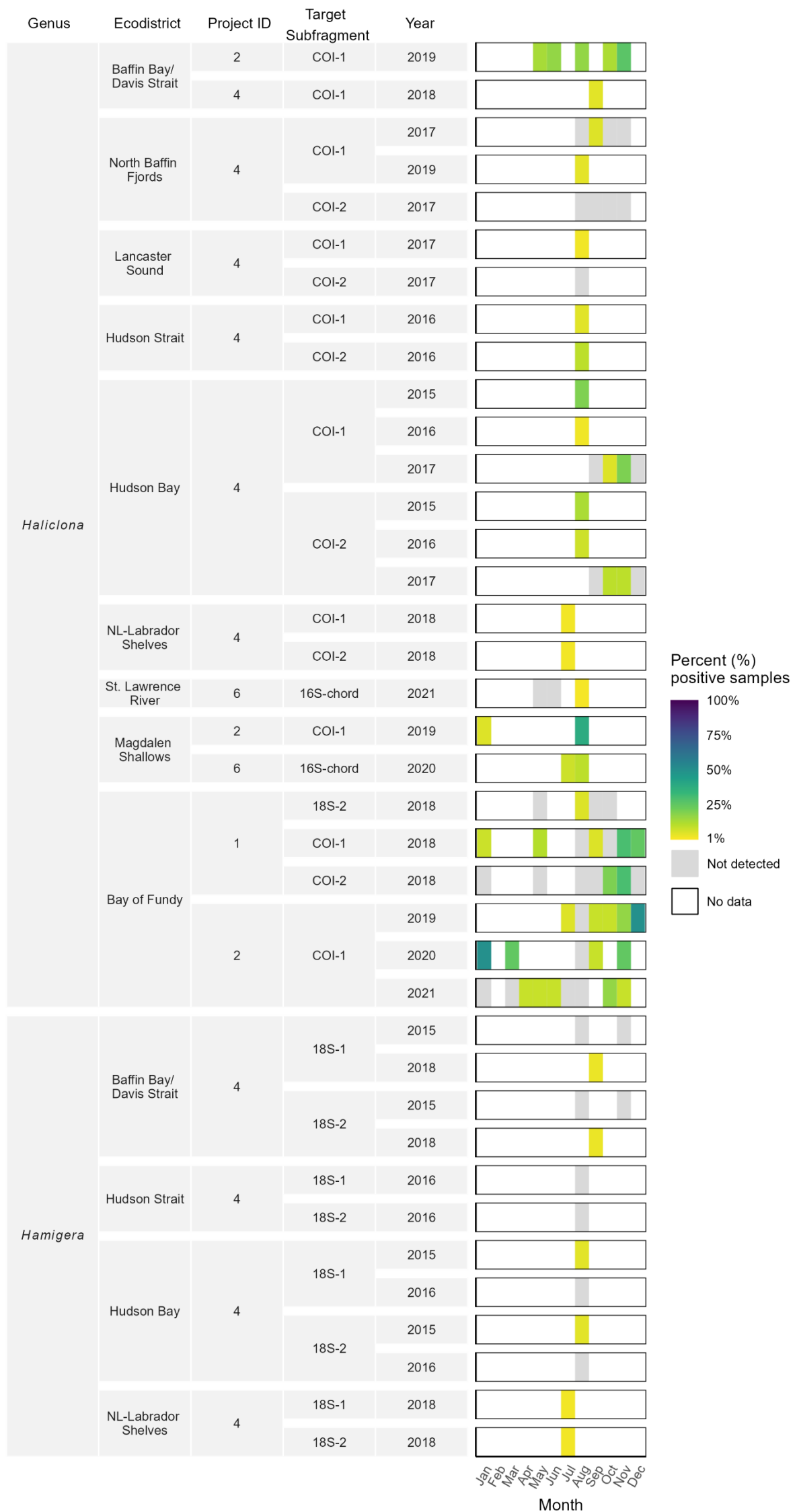


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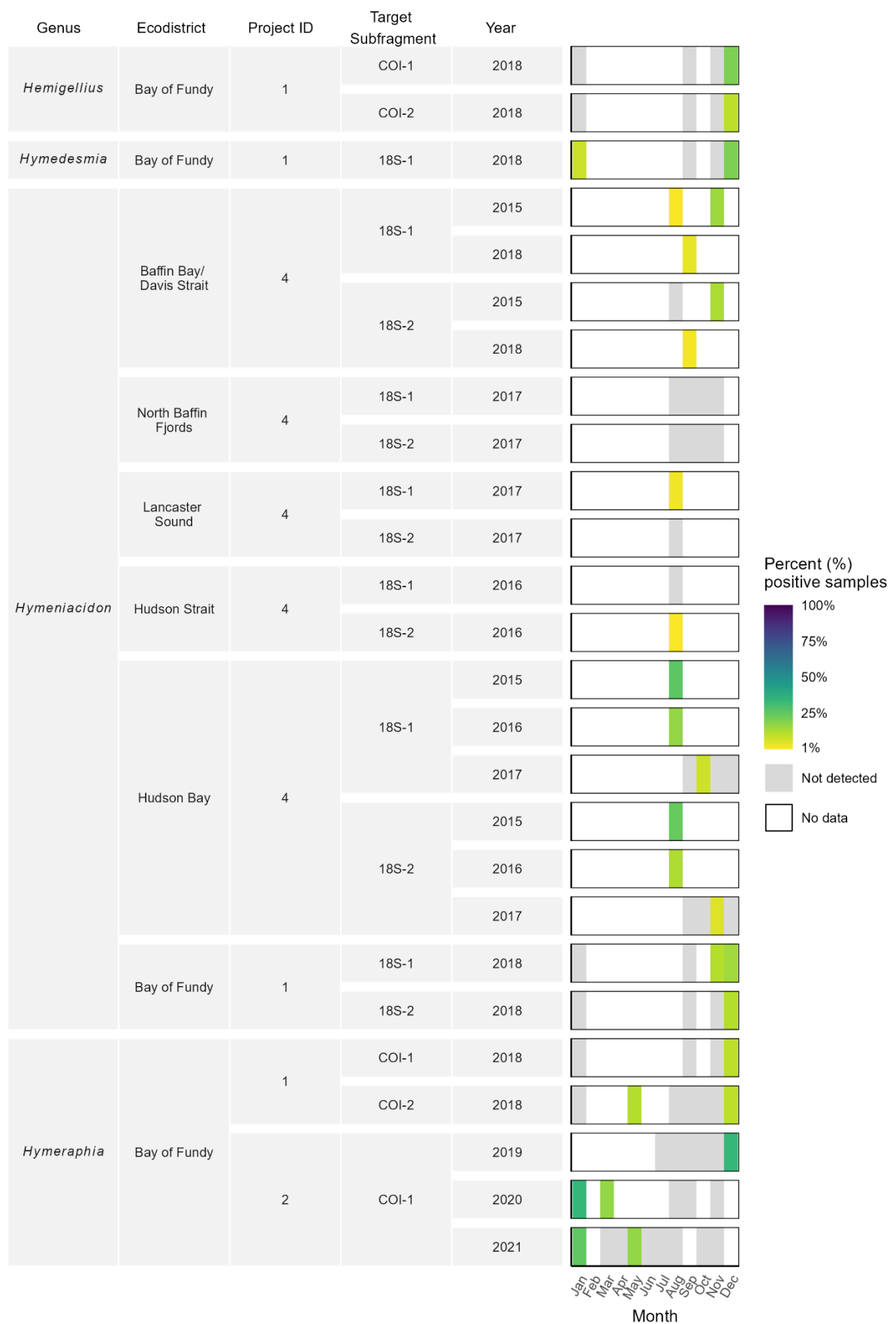


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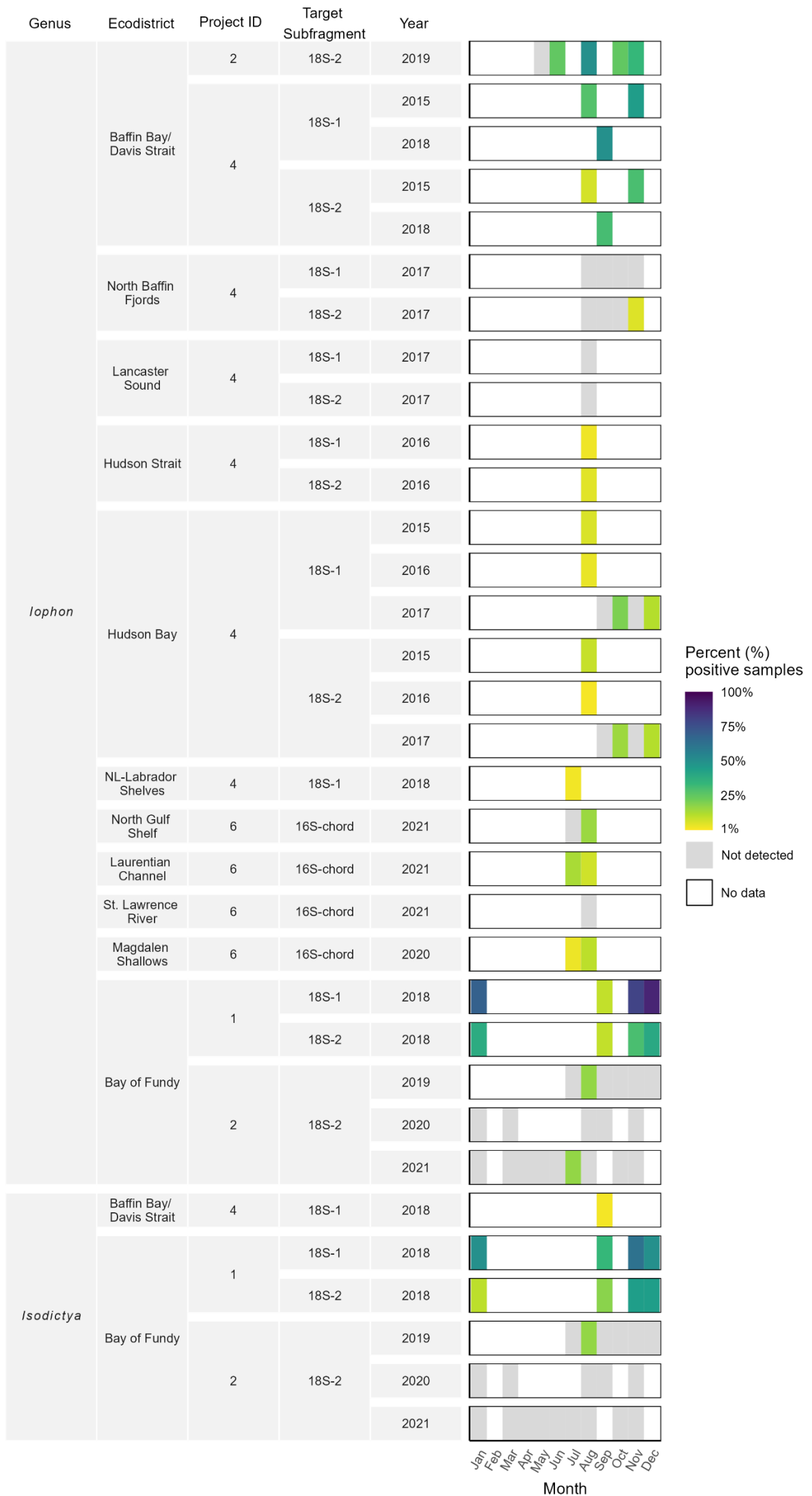


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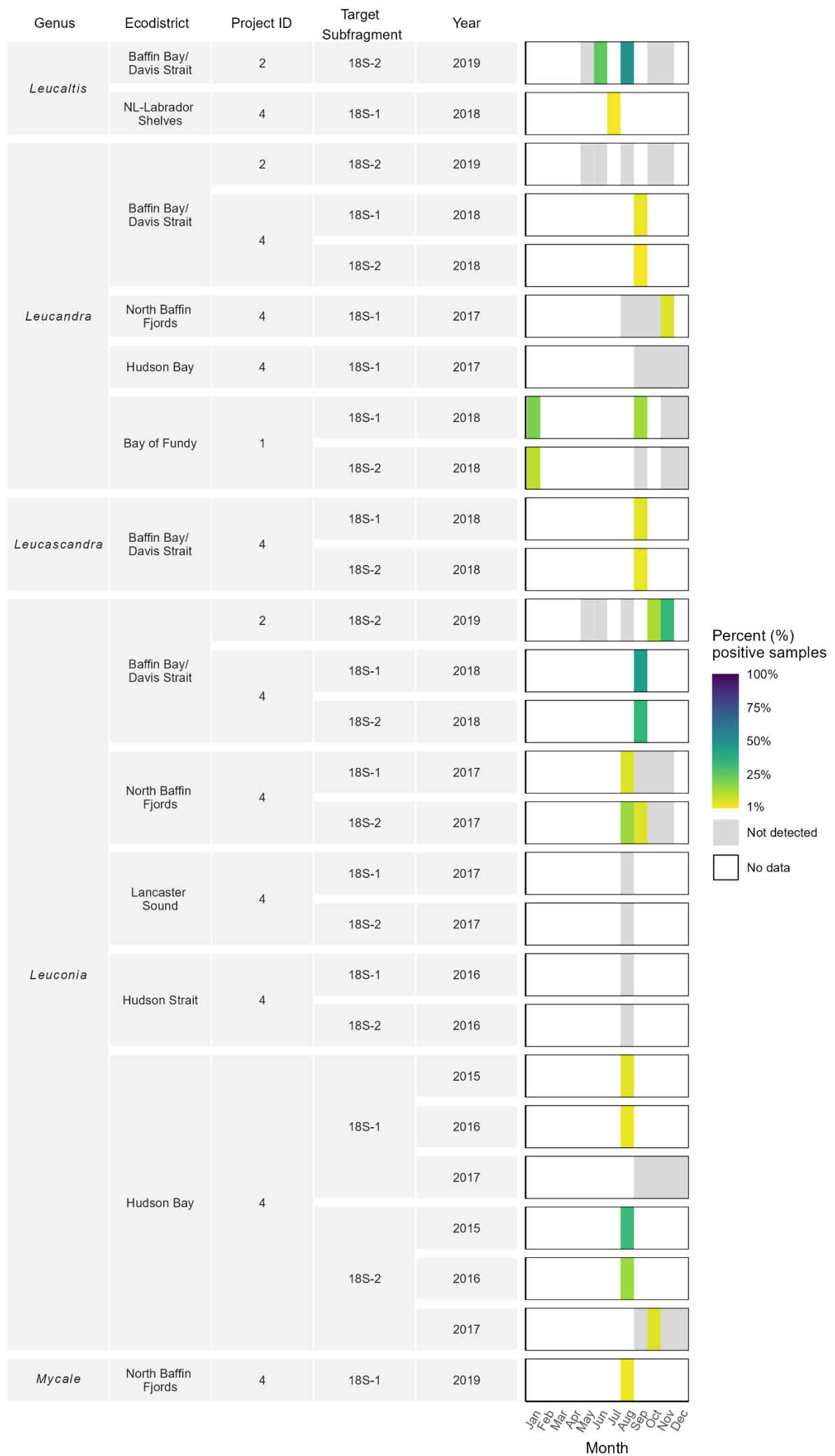


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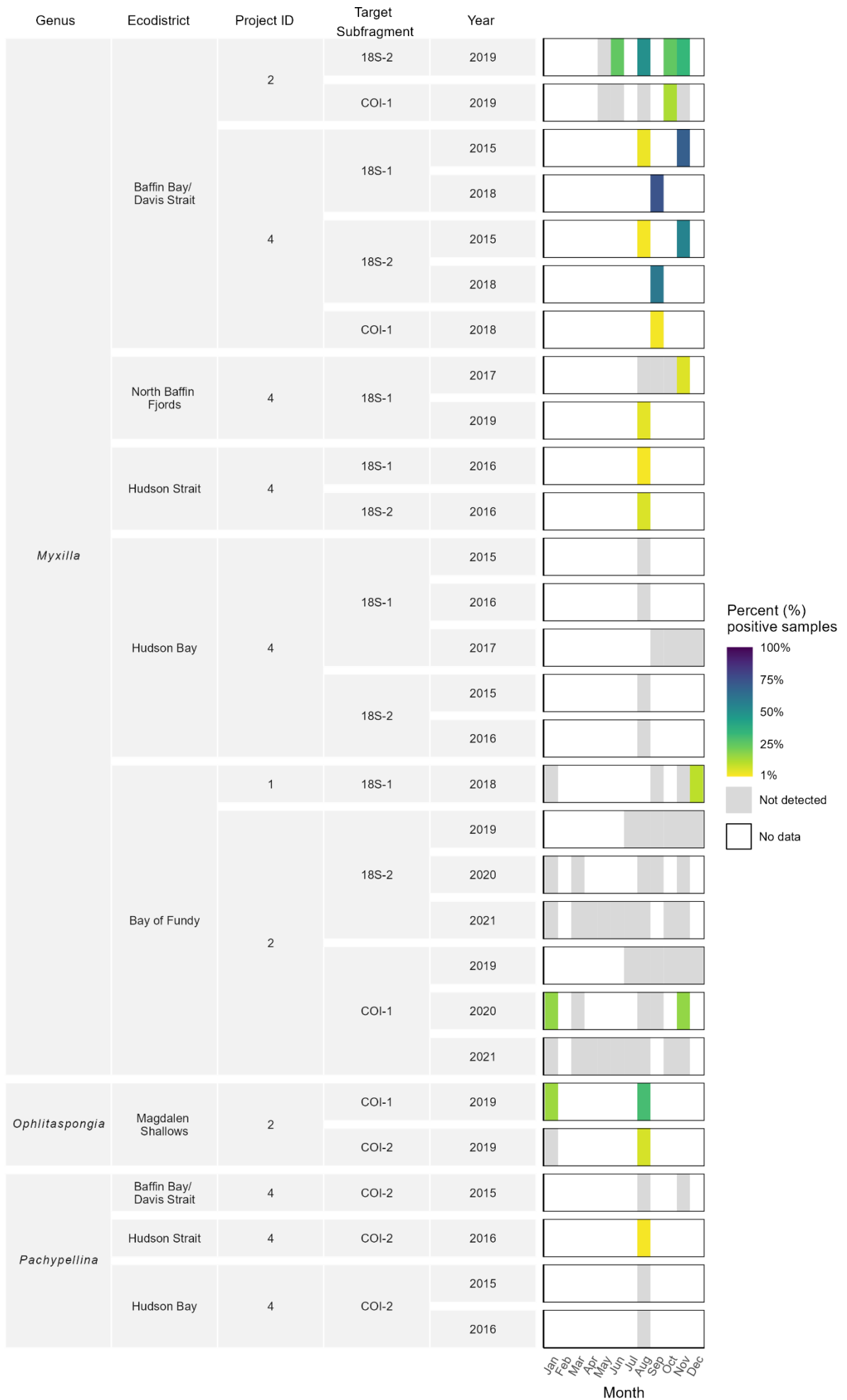


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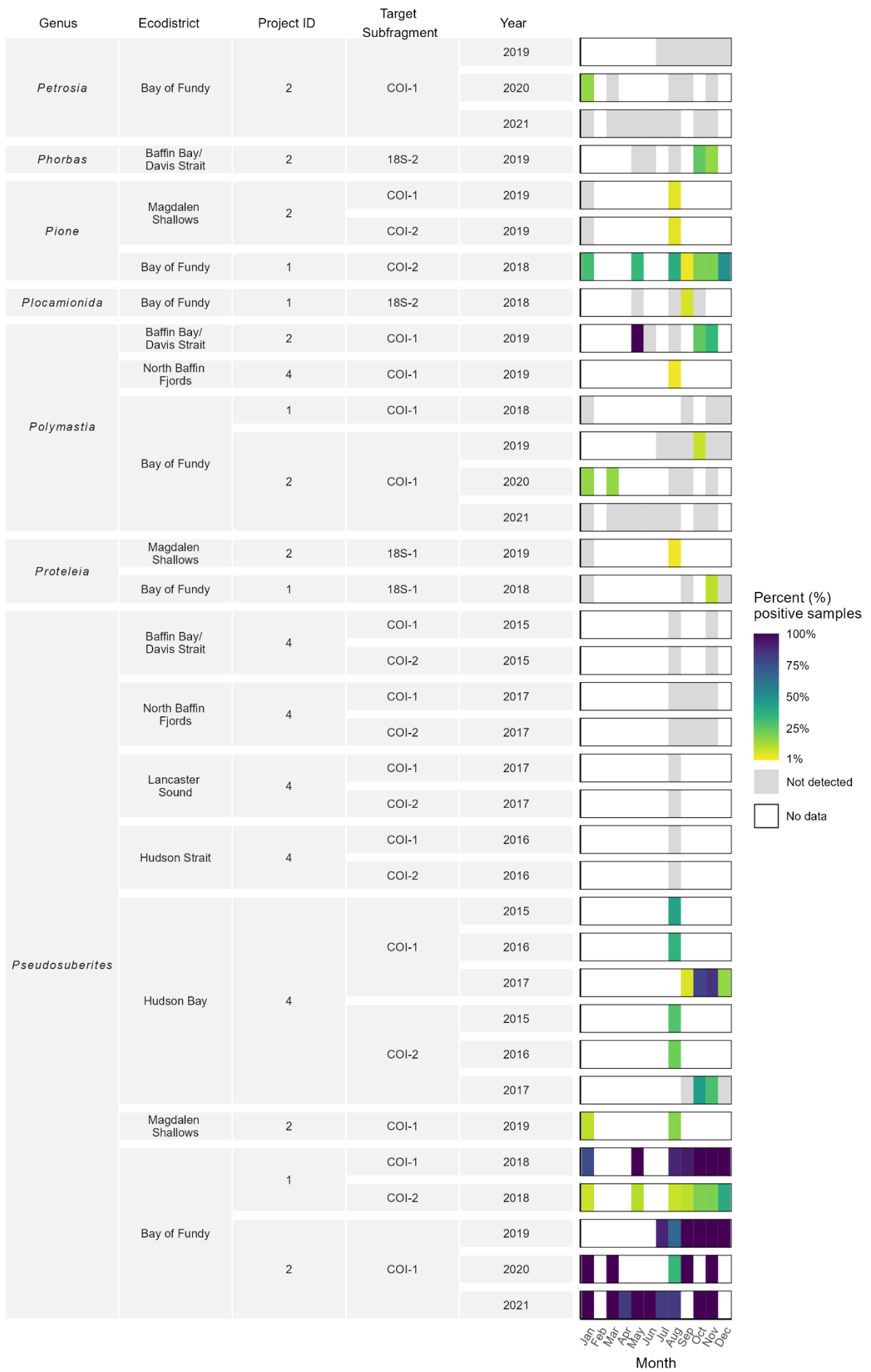


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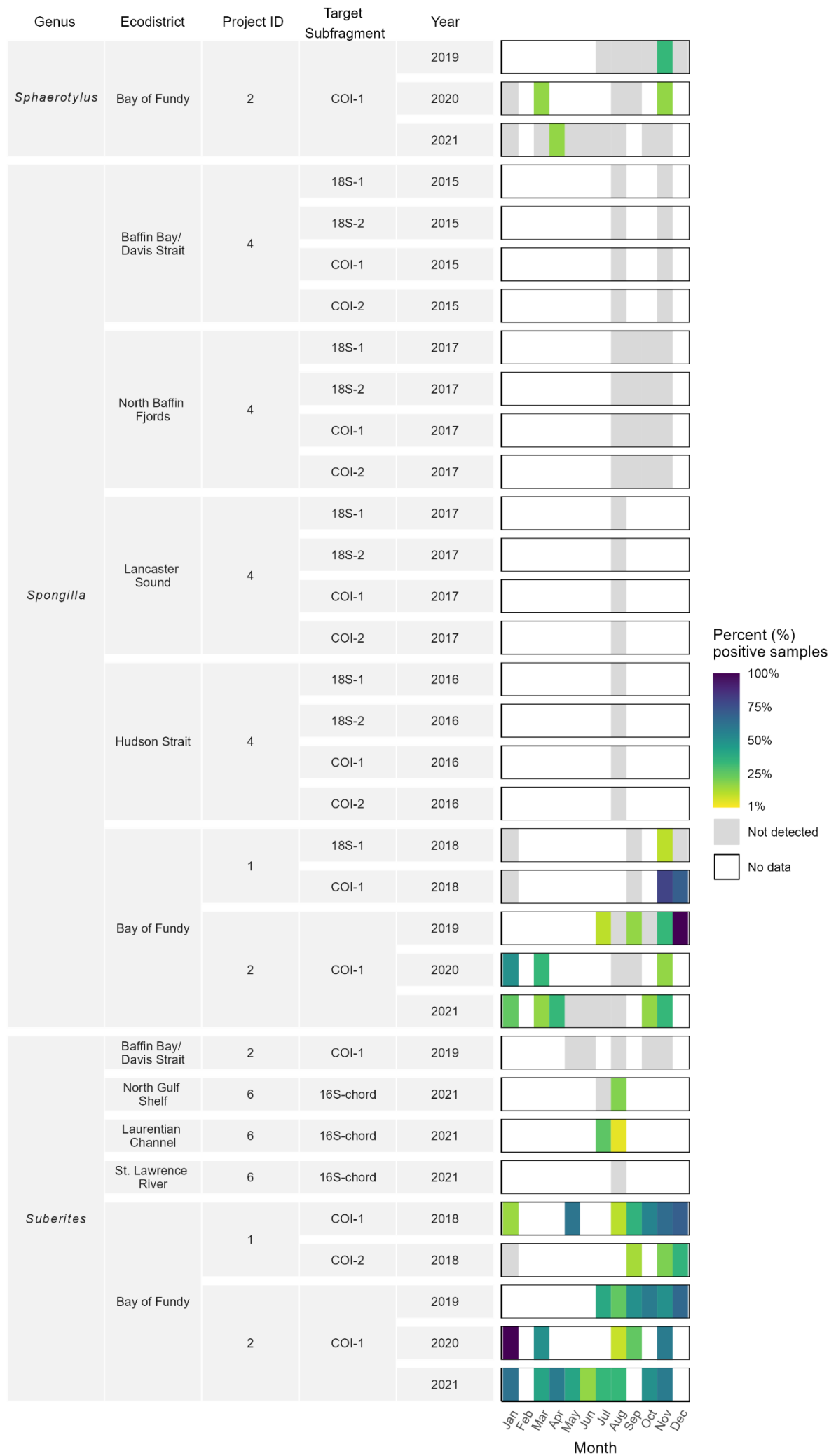


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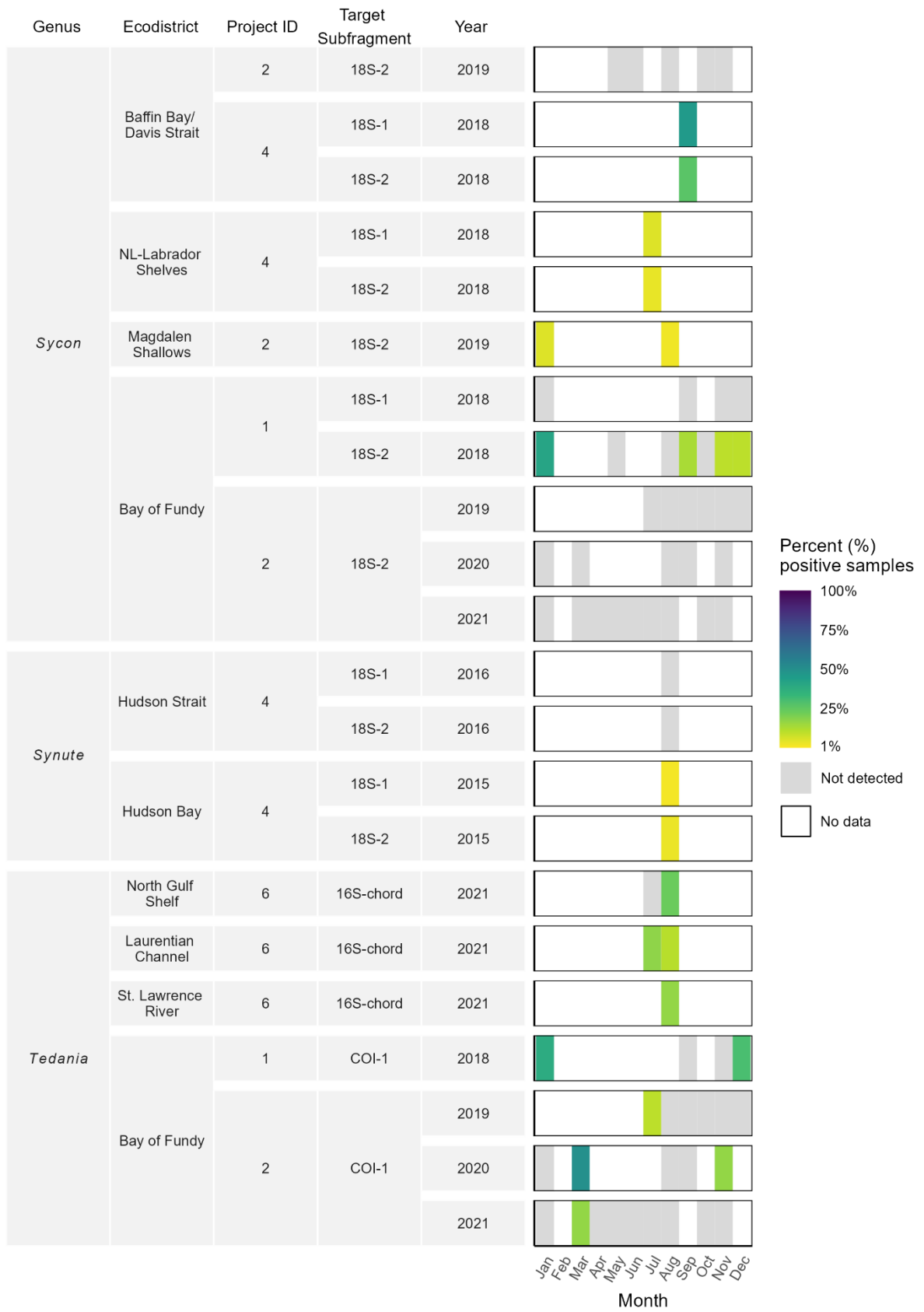


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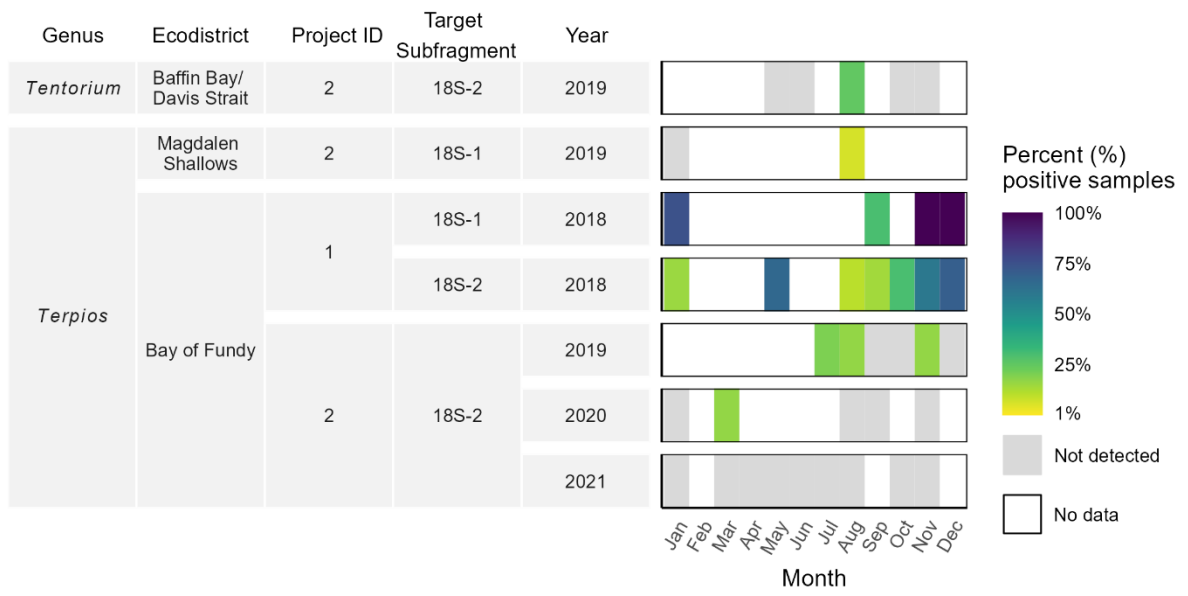


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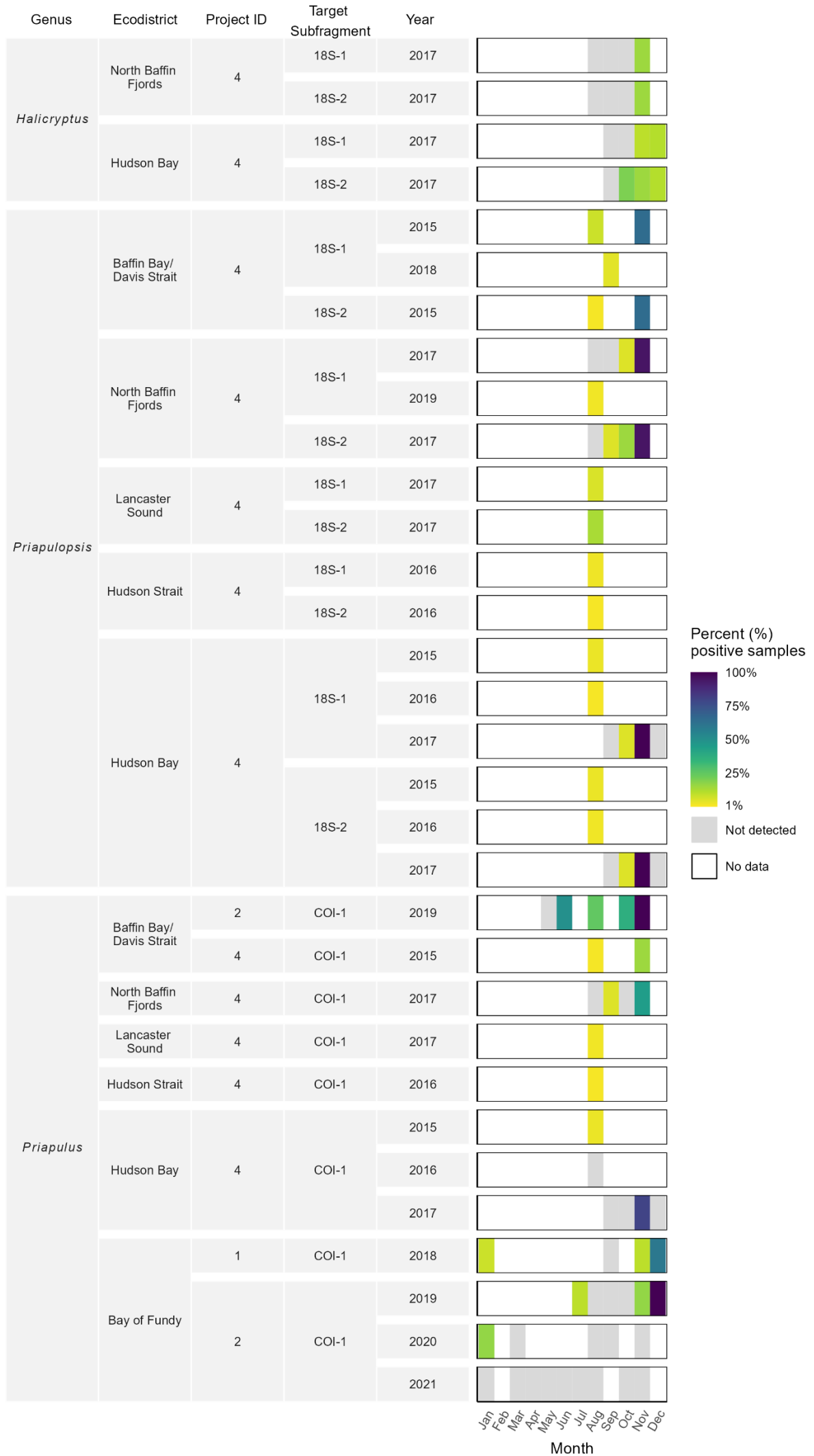


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6.21 ROTIFERA

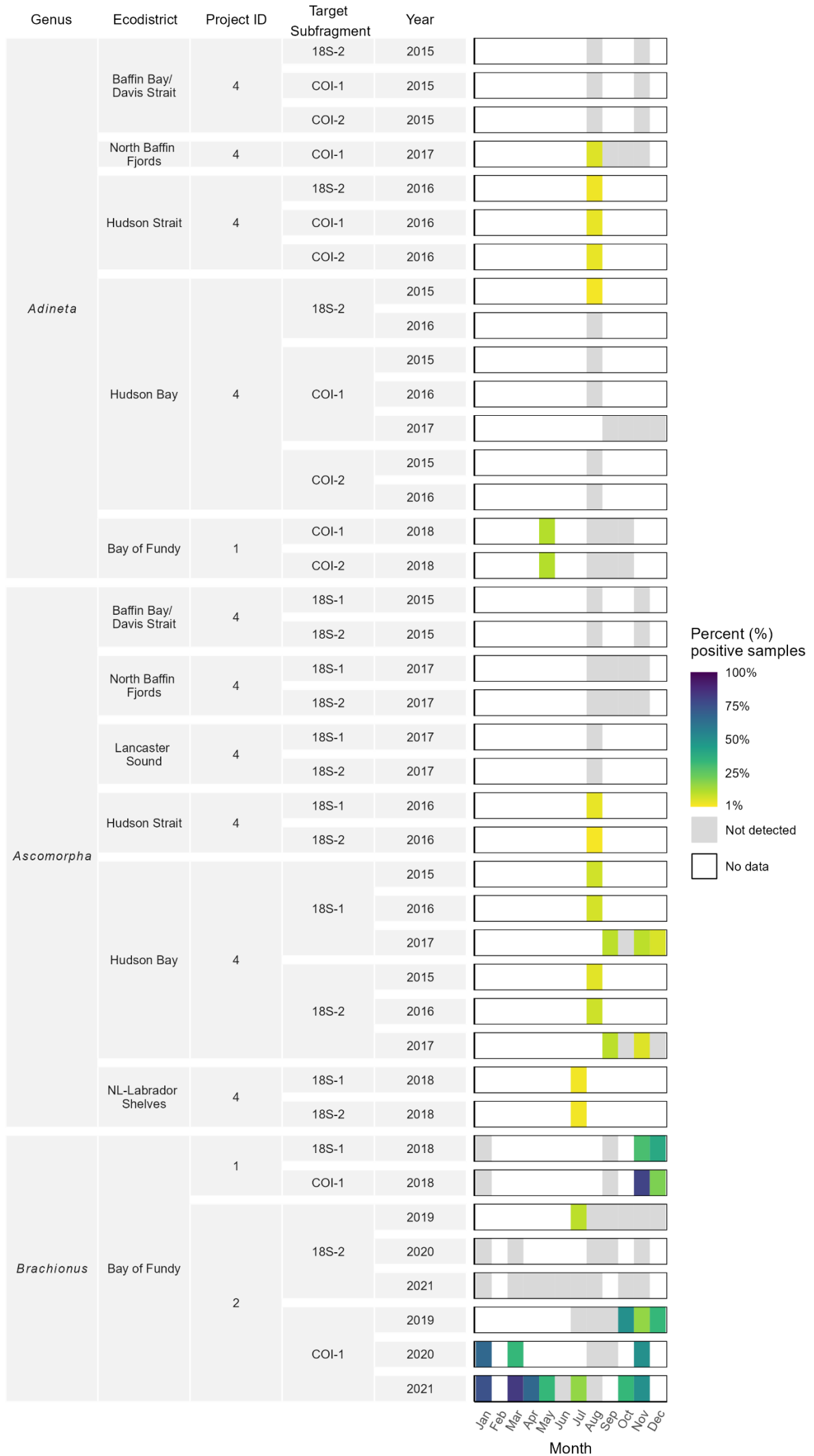


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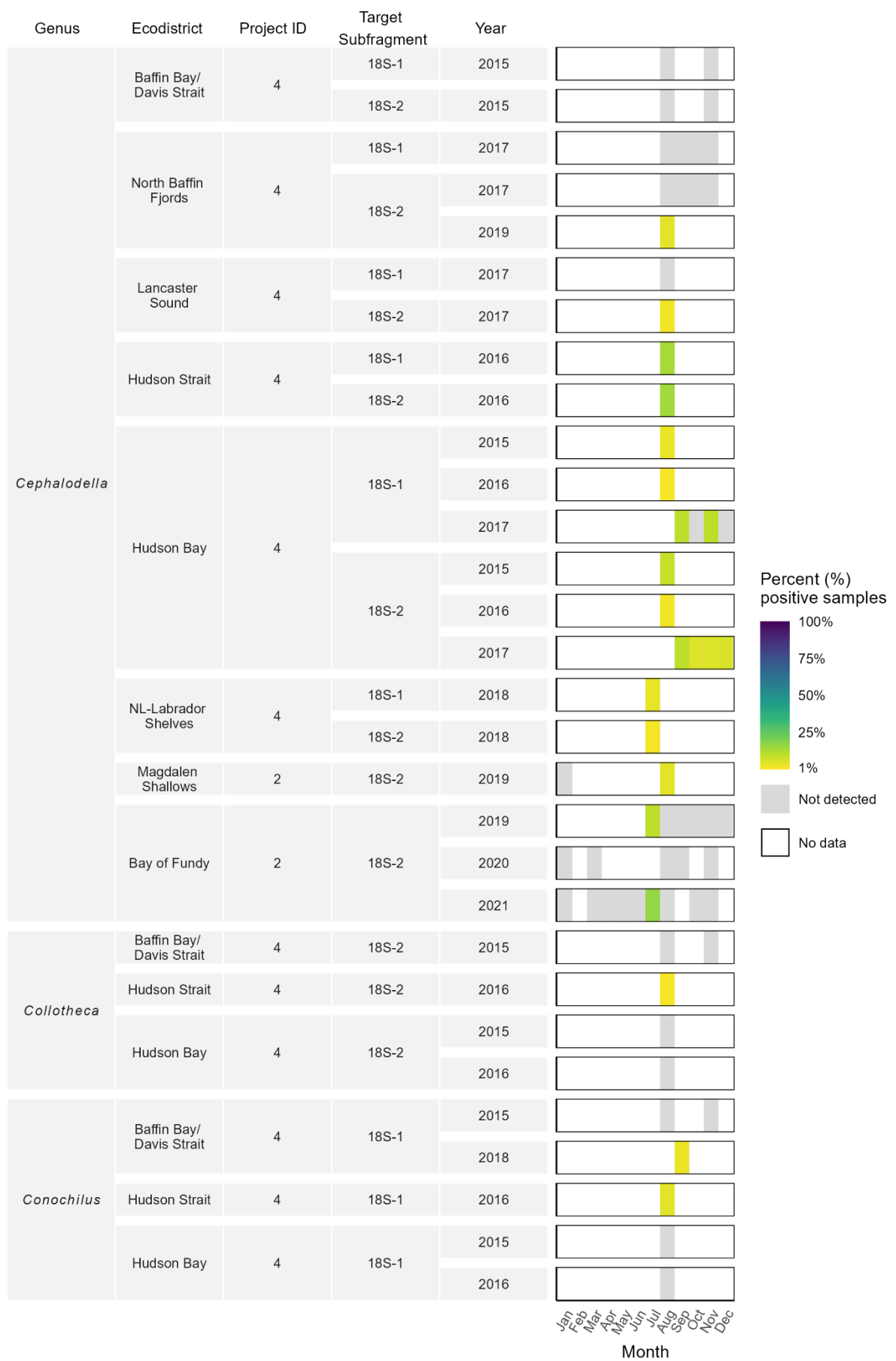


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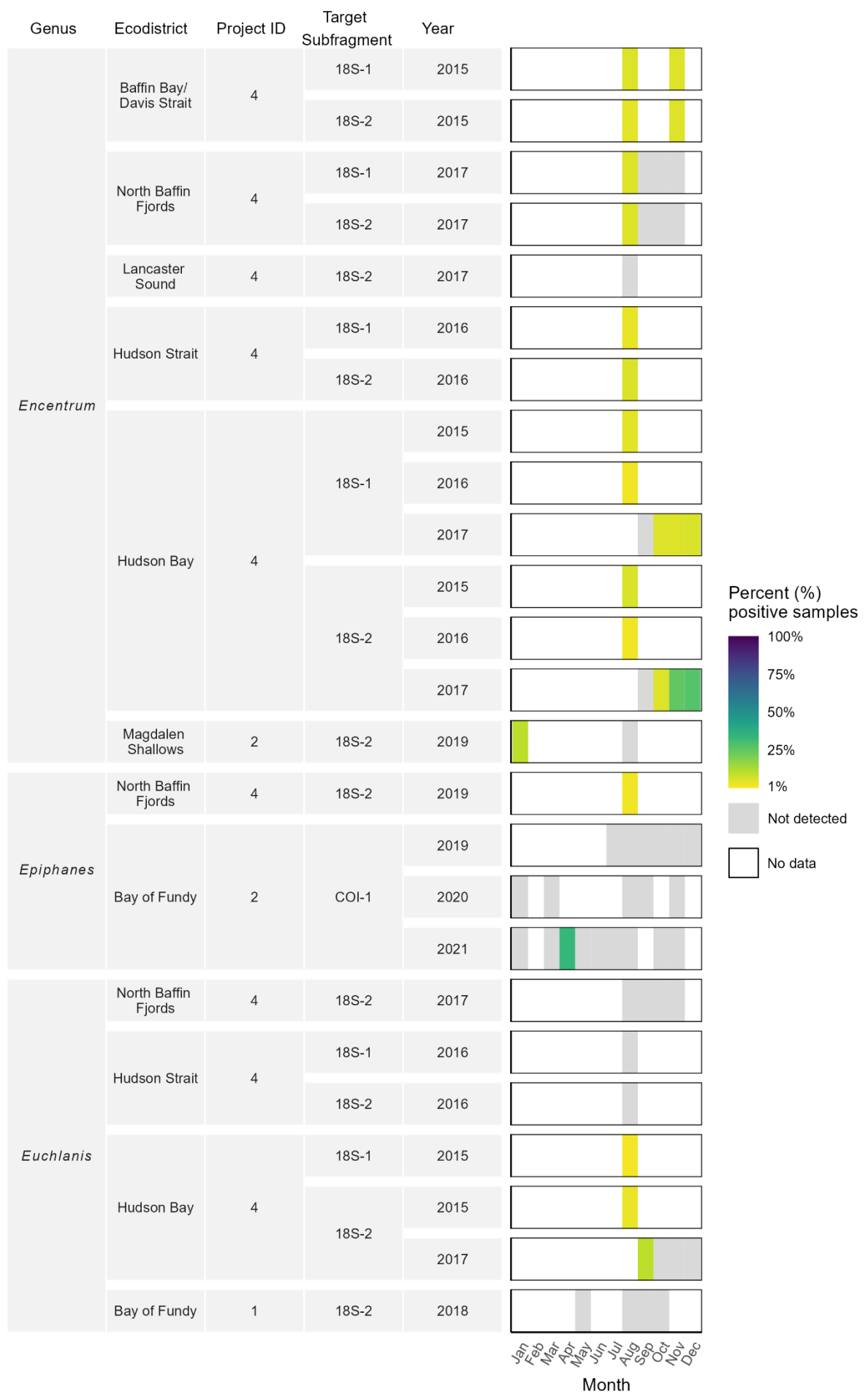


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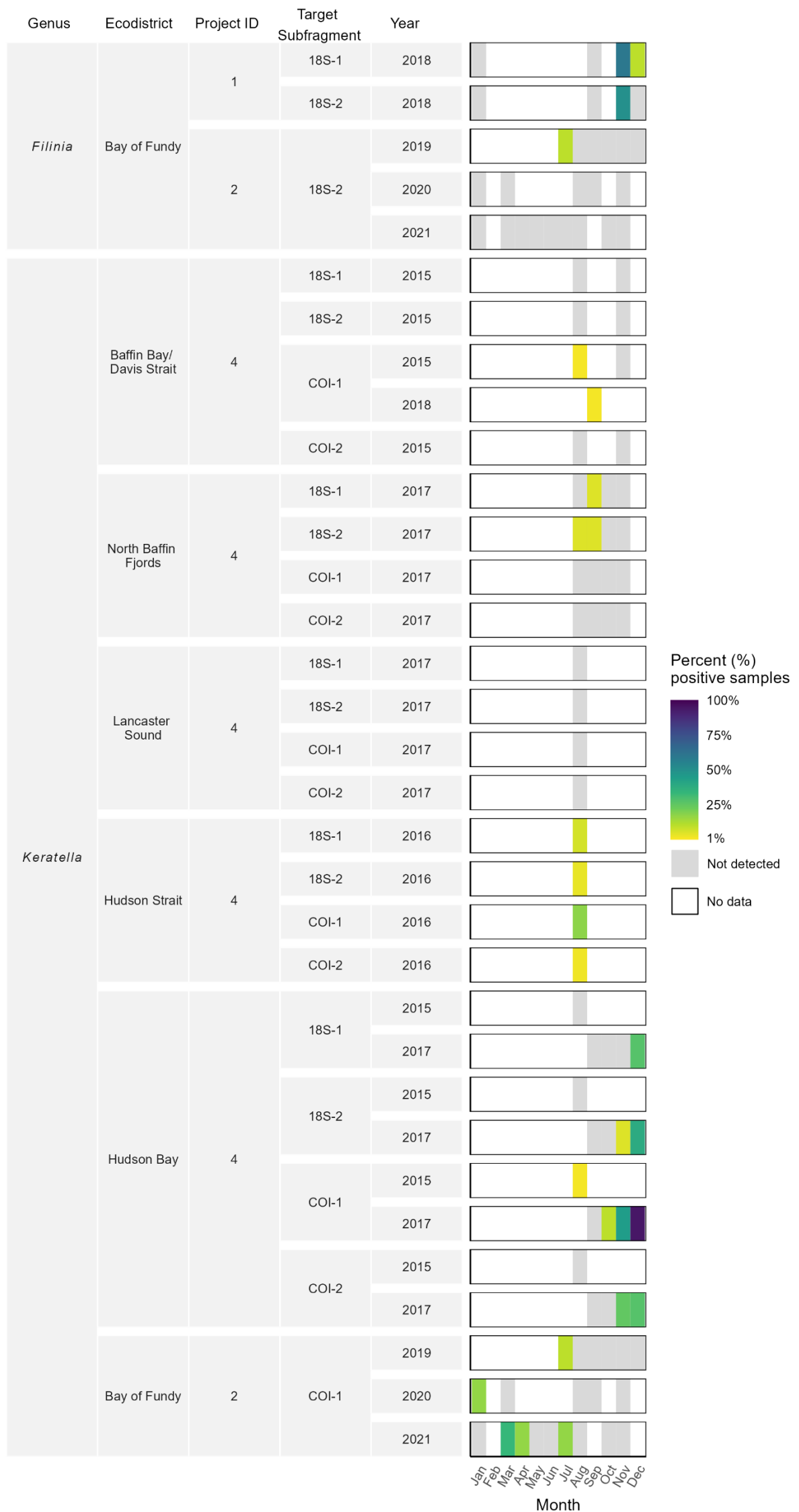


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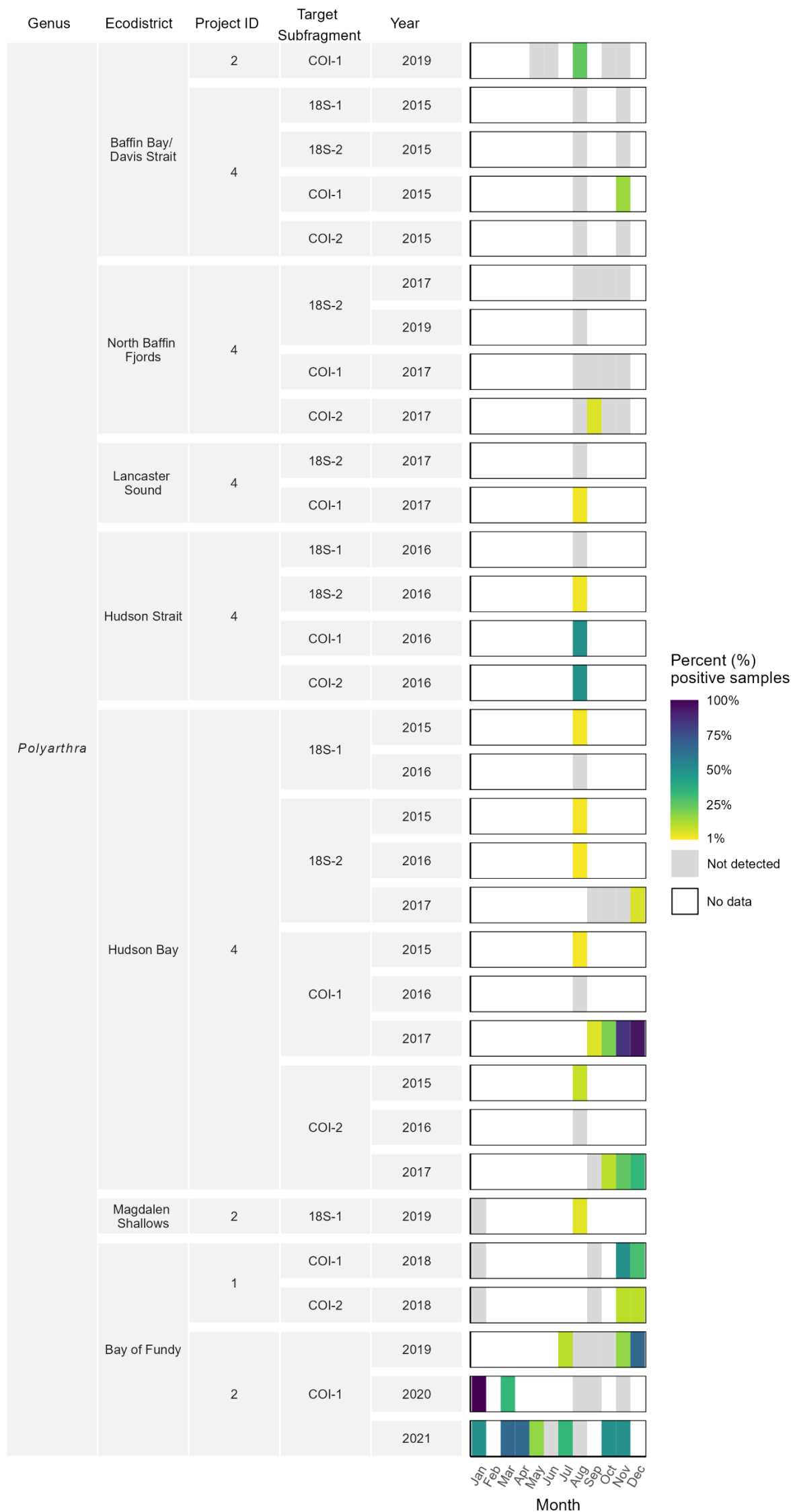


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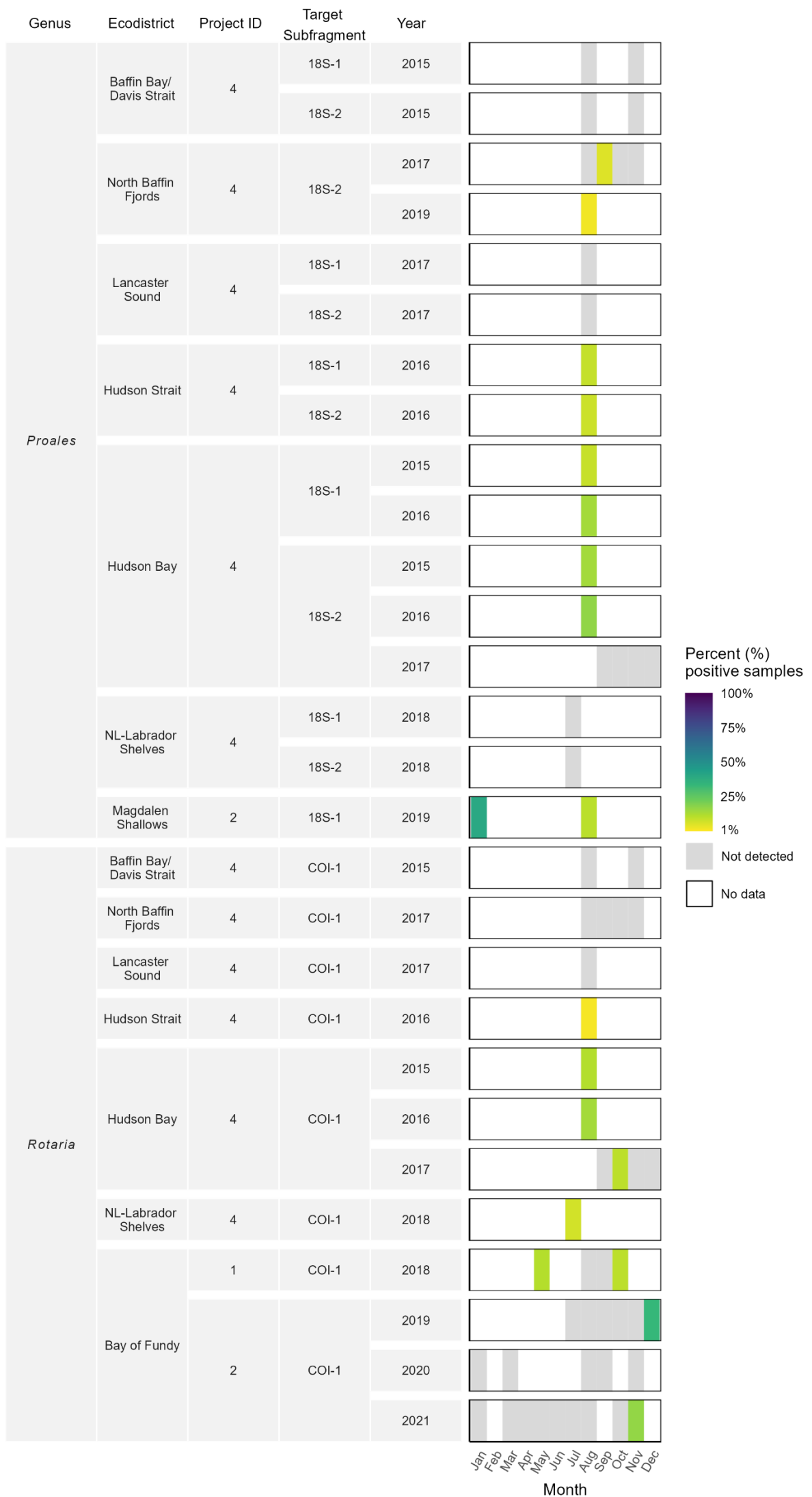


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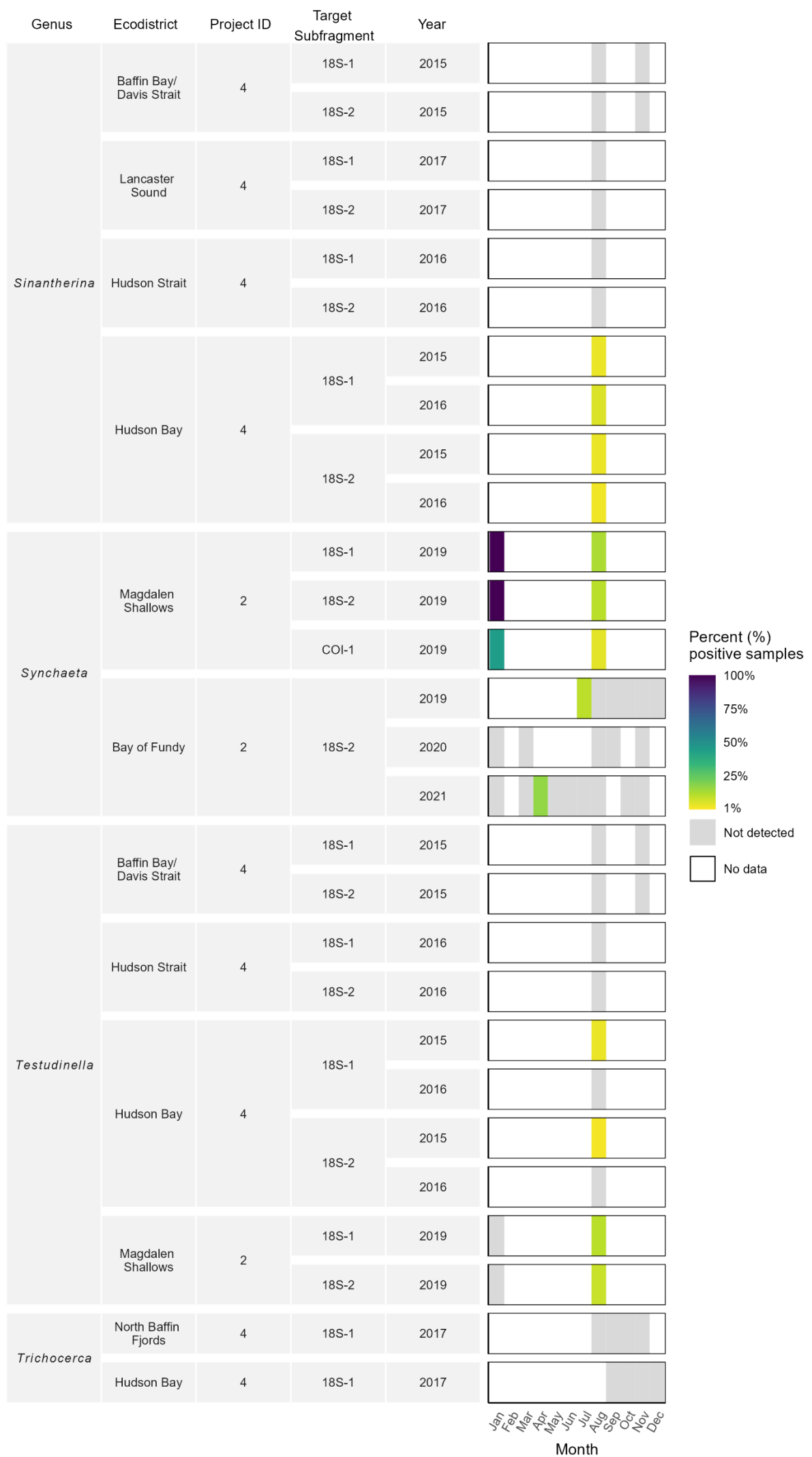


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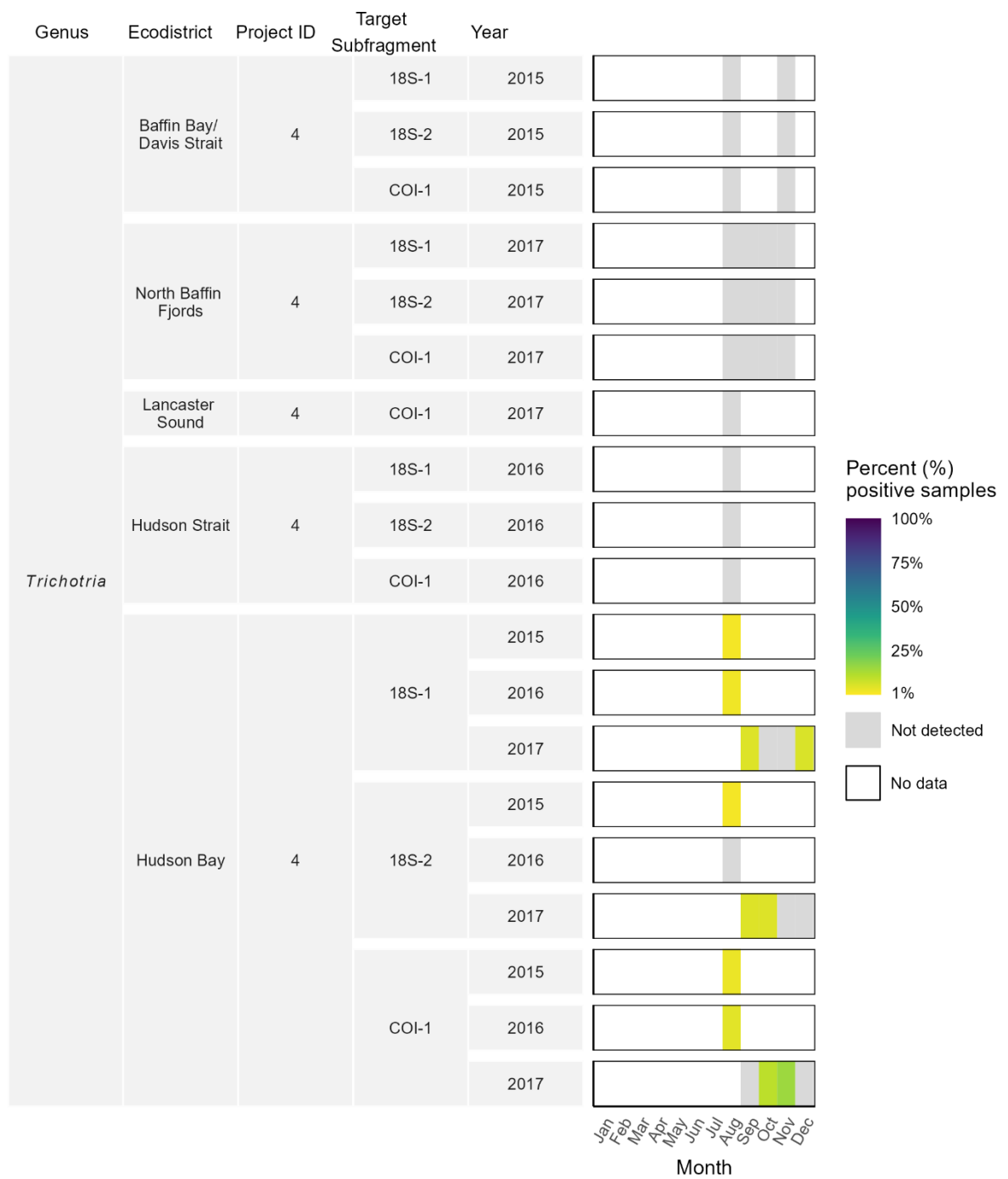
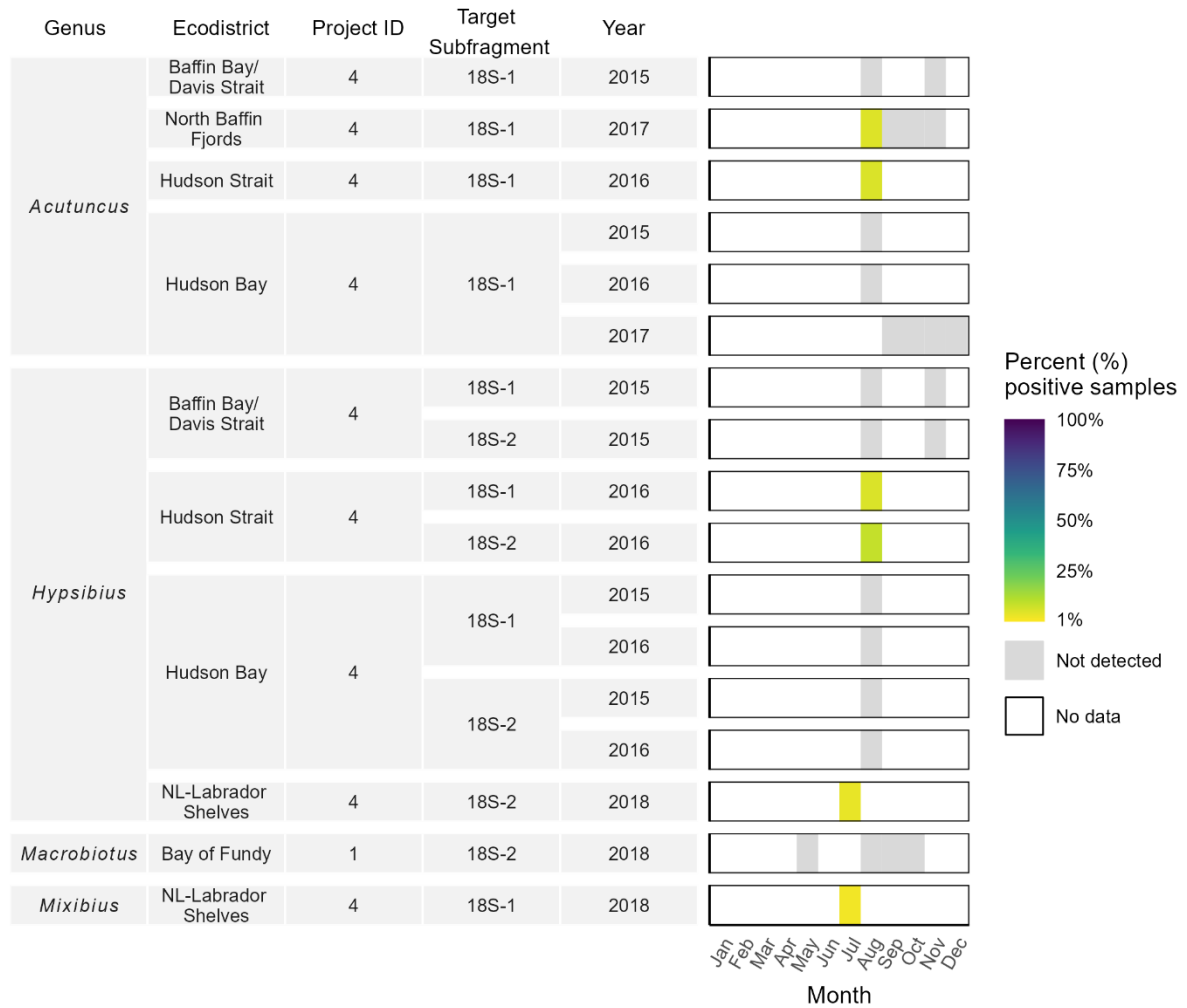


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6.22 TARDIGRADA



6.23 XENACOELOMORPHA

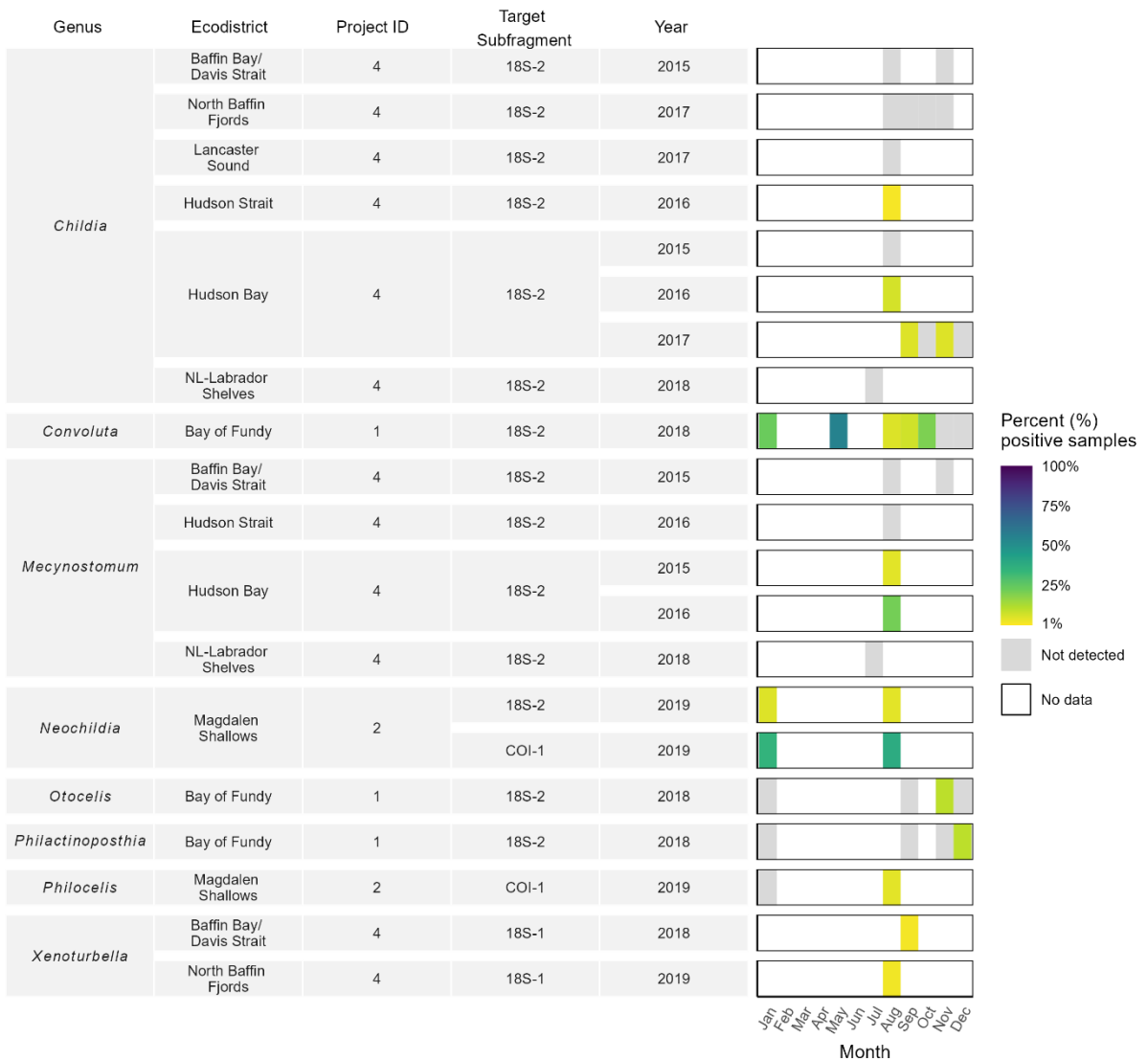


Figure 13. (Continued)

7. CONCLUDING REMARKS AND NEXT STEPS

The data visualizations presented herein can allow managers to optimize field work plans and develop AIS, SAR, and biodiversity monitoring programs based on observed detection periods for target taxa and regions. Direct interpretation of these results as species abundance measurements is not recommended as eDNA concentrations and metabarcoding sequence reads may not directly relate to species abundance without further analyses. More guidance will come in subsequent publications as we are testing the statistical variance of the detection window within and among taxa and will explore how biological and physical factors affect spatiotemporal variation in eDNA detection windows for different taxonomic groups. Finally, using both metabarcoding and qPCR data may allow managers to cross-reference taxa detection periods and allow for the decision of which method to use to optimize resources. Nevertheless, it is highly recommended that non-eDNA experts consult eDNA experts to interpret the spatiotemporal trends of any taxa using this report.

An online interactive tool (Guidance on Optimal Timing for eDNA [GOTeDNA]) is currently in development to report/visualize the detection window (*i.e.*, detection probability, length, and period[s]) for all taxonomic levels and locations. GOTeDNA is a machine learning application for ocean ecology and sustainable natural resource management, with the potential to radically improve the motivation of organizations to coordinate, share, and utilize their data. By updating the tool as time-series data/information is accrued, the degree of confidence in predicted optimal sampling periods will increase and be spatially-refined. This comprehensive approach has the potential to improve confidence by streamlining operational requests and contributing to address inconsistencies in reporting among eDNA studies.

ACKNOWLEDGEMENTS

We acknowledge that this research was conducted on the unceded and unsurrendered traditional territories of the Mi'kmaq, Passamaquoddy, Wabanaki (Bay of Fundy ecodistrict); Mi'kmaq, Wabanaki (Magdalen Shallows, Scotian Shelf); Métis peoples (Churchill Estuary-Hudson Bay); and the Inuit homelands of Inuit Nunangat including: Nunatsiavut (NL-Labrador Shelves), Nunavik (Southern Hudson Strait), and Nunavut (Baffin Bay/Davis Strait, North Baffin Fjords). We thank Louis Bernatchez for his significant contribution in developing these datasets and all of his support in eDNA research and development. This research was primarily supported by Fisheries and Oceans Canada (DFO), including the Genomics Research and Development Initiative (GRDI) and the Competitive Science Research Fund (CSRF) to ALR, KH, and FL. Arctic data collections were funded by ArcticNet, Polar Knowledge Canada, DFO (Aquatic Invasive Species Monitoring Program, Strategic Program for Ecosystem-Based Research and Advice [SPERA], Ocean Protection Plan [OPP] Coastal Environmental Baseline Program, Arctic Science Funds, GRDI), Nunavut Wildlife Management Board, Nunavik Marine Region Wildlife Board, and World Wildlife Funds to KH and ALR. Field accommodations and/or logistic support during Arctic field campaigns were provided by Churchill Northern Studies Centre, Glencore-Raglan, Baffinland Iron and Vale Mines, NRCan (Polar Continental Shelf program), Nunatsiavut Government, Environment and Climate Change Canada, and Government of Nunavut MV Nuliajuk. We are especially grateful for help with field coordination and/or assistance from the following individuals: Andrew Arreak, Antoine Dispas, Alex Flaherty (Polar Outfitting), Adamie Keatanik, Austin MacLeod, Chris Idlaut, Chris Lewis, Chris McKindsey, Colin Gallagher, Daniel Gibson, Dick Hunter, Emmanuelle Porter, Eric Solomon, Erin Keenan, Fatma Dhifallah, Frédéric Hartog, Frederic Lemire, Jari Aariak, Jennifer Amagoalik, Kathleen MacGregor, Heather Clark, LeeAnn Fishback, Lillian Angnatok, Markusie Jaaka, Melissa Nacke, Nathalie Simard, Noemie Leduc, Ken Cameron, Rachel Smale, Rory McDonald, Shelley Elverum, Sylvia Pewatoalook, Tapisa Kasarnak, Thomas Whittle, Trevor Arreak, Valérie Cypihot, Willie Keatanik, Willie Alaku, and Zoya Martin. We are also grateful to the large number of other colleagues who contributed to field sampling and laboratory work; please see the details in the various publications cited in Tables 2 and 3.

AUTHOR CONTRIBUTIONS

ALR and KH conceived the study. ALR, KH and FL contributed resources. ALR, KH, FLB, MC, NG, LH, NJ, GJP and NS provided data of which compilation was led by MKM. MKM and ALR analyzed the data and wrote the manuscript. All authors helped draft and improve the manuscript and approved the final version.

ACRONYMS

AIS: Aquatic invasive species

COI: Cytochrome *c* oxidase subunit I

CSAS: Canadian Science Advisory Secretariat

cytb: Cytochrome *b*

DNA: Deoxyribonucleic acid

eDNA: Environmental DNA

LOD: Limit of Detection

NAD5: NADH dehydrogenase subunit 5

PCR: Polymerase chain reaction

qPCR: Quantitative PCR or real-time PCR

RNA: Ribonucleic acid

rRNA: Ribosomal RNA

SAR: Species at risk

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APPENDIX 1

Annelida

<i>Abarenicola</i>	28	<i>Dendrobaena</i> ..	39	<i>Maldane</i>	51
<i>Achaeta</i>	28	<i>Dendrodrilus</i> ...	39	<i>Marenzelleria</i> ..	51
<i>Alitta</i>	28	<i>Dero</i>	39	<i>Marionina</i>	51
<i>Amblyosyllis</i>	28	<i>Dinophilus</i>	40	<i>Melinna</i>	51
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<i>Amphicorina</i>	29	<i>Echiurus</i>	40	<i>Micronephthys</i>	52
<i>Amphitrite</i>	29	<i>Eiseniella</i>	40	<i>Myrianida</i>	52
<i>Anobothrus</i>	29	<i>Enchytraeus</i>	41	<i>Myxicola</i>	52
<i>Aphelochaeta</i> ..	30	<i>Enipo</i>	41	<i>Naineris</i>	52
<i>Apistobranchus</i>		<i>Epigamia</i>	41	<i>Nais</i>	52
.....	30	<i>Erpobdella</i>	41	<i>Nephasoma</i>	53
<i>Aporrectodea</i> ..	30	<i>Eteone</i>	42	<i>Nephtys</i>	53
<i>Arcteonais</i>	31	<i>Euclymene</i>	42	<i>Nereimyra</i>	53
<i>Arenicola</i>	31	<i>Eulalia</i>	42	<i>Nereis</i>	54
<i>Aricidea</i>	32	<i>Eumida</i>	43	<i>Nicolea</i>	54
<i>Artacama</i>	32	<i>Eunoe</i>	43	<i>Nicomache</i>	54
<i>Aulodrilus</i>	32	<i>Flabelligera</i>	43	<i>Ninoe</i>	55
<i>Aurospio</i>	33	<i>Fridericia</i>	43	<i>Nothria</i>	55
<i>Axionice</i>	33	<i>Galathowenia</i> ..	44	<i>Oconnorella</i>	55
<i>Barantolla</i>	33	<i>Gattyana</i>	44	<i>Octolasion</i>	55
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<i>Bispira</i>	34	<i>Glycera</i>	45	<i>Ophelia</i>	56
<i>Bothrioneurum</i>	34	<i>Golfingia</i>	45	<i>Ophelina</i>	57
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<i>Cautleriella</i>	35	<i>Helobdella</i>	46	<i>Paranaitis</i>	58
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<i>Circeis</i>	38	<i>Lepidonotus</i>	48	<i>Piscicola</i>	61
<i>Cirratulus</i>	38	<i>Limnodriloides</i>	48	<i>Pista</i>	62
<i>Cirrophorus</i>	38	<i>Limnodrilus</i>	48	<i>Polycirrus</i>	63
<i>Clymenella</i>	38	<i>Lumbricillus</i>	49	<i>Polydora</i>	64
<i>Clymenura</i>	39	<i>Lumbriculus</i>	49	<i>Polygordius</i>	64
<i>Cognettia</i>	39	<i>Lumbricus</i>	50	<i>Polyphysia</i>	64
<i>Ctenodrilus</i>	39	<i>Lumbrineris</i>	50	<i>Praxillella</i>	64
		<i>Lysippe</i>	50	<i>Prionospio</i>	65

<i>Pristina</i>	65	<i>Ameronothrus</i> .	75	<i>Cypris</i>	83
<i>Proceraea</i>	65	<i>Ampelisca</i>	75	<i>Cythere</i>	83
<i>Pseudopotamilla</i>	65	<i>Amphibalanus</i> .	75	<i>Cytheromorpha</i>	83
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<i>Pygospio</i>	67	<i>Anonyx</i>	76	<i>Dermestes</i>	84
<i>Rhodine</i>	67	<i>Anoplodactylus</i>	76	<i>Dexamine</i>	84
<i>Rhycodrilus</i>	67	<i>Apherusa</i>	76	<i>Dienerella</i>	84
<i>Ripistes</i>	67	<i>Apis</i>	76	<i>Diporeia</i>	84
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<i>Schizobranchia</i>	68	<i>Aurila</i>	77	<i>Epitheca</i>	85
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