CREATION AND MAINTENANCE PROCESSES OF MALACOLOGICAL COLLECTIONS OF CEPHALOPOD BEAKS

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ABSTRACT

Lopes, V. E. A. S. & Vaske Junior, T. (2022). Creation and Maintenance Processes of Malacological Collections of Cephalopod Beaks. Braz. J. Aquat. Sci. Technol. 26(1). ISSN 1983-9057. DOI: 17226/bjast.v26n1. The buccal apparatus of the Cephalopoda includes a chitinous two-part beak, which is used to triturate prey. Cephalopod beaks often present diagnostic features which, while difficult to analyze, can be used to both identify species and infer their ecological characteristics. The present study aimed to establish and maintain a didactic collection of cephalopod beaks. The curation process began with the preparation of the preserving liquid and the choice of jars, and ended with the selection and identification of the specimens. It was decided to maintain the collection in liquid medium, in a solution of 80% alcohol and 5% glycerin. The collection includes a total of 562 beaks, representing 50 taxa, which had been obtained from the stomach contents of predators, scientific collections, and commercial fisheries located along the whole of the Brazilian coast. Although the lack of published material on the curation of malacological collections, and in particular specimens of cephalopod beaks, hampered the curation process, it reinforced the need for the establishment of didactic collections and reference material to support research and teaching, with emphasis on the taxonomic identification of cephalopod beaks.

Key Words: Cephalopoda, Beak, Curation, Species Identification.

INTRODUCTION

The Cephalopoda is a class of marine invertebrates of the phylum Mollusca, the second-largest after the Arthropoda, with over 800 extant species catalogued to date, and an estimated 11,000-plus extinct taxa (Roper et al., 1984; Wilbur et al., 1985; Jereb & Roper, 2010; Jereb et al., 2014; CephBase, 2020; WoRMS, 2020). This metazoan group includes a variety of animals in two extant subclasses, the Nautiloidea (nautiluses) and the Coleoidea, which is divided into four orders, the Sepiida (cuttlefish), Octopoda (octopuses), Teuthida (squid), and the Vampyromorpha, the vampire squids (Jereb & Roper, 2010; Jereb et al., 2014).

The buccal apparatus of the cephalopods features a pair of chitinous jaws, known as beaks due to their resemblance to the beaks of parrots, which are secreted by a single layer of cells surrounding the buccal muscles (Dilly & Nixon, 1976; Lefkaditou & Bekas, 2004; Vaske, 2006). Given their predominantly carnivorous diet, which includes fish, crustaceans, and other mollusks, cephalopods use their beaks to seize their prey and crush rigid structures, such as bone and shell (Miserez et al. 2007; Miserez et al. 2008). Composed of water, glycine-histidine, and dopa-proteins interspersed between layers of melanin and chitin fibers, these beaks are rigid, odd-shaped structures with unique pigmentation, which can be used to diagnose cephalopod families, genera, and even species (Clarke, 1986; Broomell et al. 2007).

Akimushkim & Beteshava (1955) was the first study based on the identification of cephalopod beaks, which were used to determine which cephalopod taxa had been ingested by sperm whales. A number of subsequent studies adopted a similar approach to identify the cephalopods in the diets of a range of different marine species (Iverson & Pinkas, 1971; Furness et al., 1984; Lefkaditou & Bekas, 2004; Xavier et al., 2007; Chen et al., 2012). In Brazil, cephalopod beaks have been identified in the stomach contents of a number of different predators in studies such as those of Santos (2000), Santos and Haimovici (2001), Vaske (2006), and Leite et al (2010). The potential for the identification of taxa supports the establishment of reference collections of cephalopod beaks as a diagnostic tool for the analysis of trophic patterns in teuthophagous species, such as marine mammals, turtles, and fish.

Although a large number of studies has focused on the collection and preservation of specimens for malacological collections in Brazil (Matthews & Rios, 1987; Almeida & Oliveira, 2000; Sturm et al., 2006; Geier et al., 2007; D'Ávila, 2016; Martins et al., 2016; Nojoza et al., 2016; Silva & Barreira, 2016), relatively little attention has been given to the preservation of cephalopod beaks. The present study aimed to contribute to the rectification of this knowledge gap and identify the beaks in the malacological collection of the Laboratory of the Biology and Conservation of Pelagic Organisms (LABCOP) at ão Paulo State University (UNESP – IB/CLP) in São Vicente, São Paulo, Brazil.

MATERIAL AND METHODS

Conservation of cephalopod beaks

Cephalopod beaks are chitinous structures that will shrink when dehydrated, and are thus best preserved in a liquid medium. This type of storage requires adequate containers, *e.g.* jars with sealing rings or diaphragms, to avoid the evaporation of the preserving liquid. Oscillations in temperature are also a major potential problem due to changes in pressure inside the containers, which may cause the contraction of the liquid and/or the seal, resulting in the exposure of the specimens to the elements, attack by mold or dehydration. Given this, periodic inspection of the collection is necessary to ensure the level and quality of the preserving liquid (Ingenito, 2014). Ideal storage conditions include relatively constant temperature of approximately 18ºC and low relative humidity (less than 65%).

At LABCOP, the cephalopod beaks were washed under running water and any incrustations were removed using tweezers. The beaks of specimens at an advanced stage of degradation were first kept in 92% alcohol for at least 24 hours. Fragments of beaks and specimens damaged by fungus were discarded.

Dry storage is not recommended as it results in extremely dry, wrinkled, and brittle beaks, which are difficult to handle and vulnerable to breakage. Given this, three different types of preserving liquid were tested to determine their suitability for the storage of cephalopod beaks, in particular, the maintenance of the coloration and malleability of the specimens (Table 1).

Table 1 - The three different types of preserving liquid tested in the LABCOP cephalopod beak collection.

Preserving Liquid	Preparation Method
Alcohol 70%	752.5 mL of alcohol 92.8% + 247.5 mL of distilled water
Alcohol 80%	860 mL of alcohol 92.8% + 140 mL of distilled water
Alcohol 80%/Glycerin	860 mL of alcohol 92.8% + 105 mL of distilled water + 35 mL of glycerin

Cephalopod beaks collected from the stomach contents of marine mammals, birds, and large oceanic fish were immersed for 30 days in each of the different preserving liquids (Table 1), with five beaks being tested in each liquid (N = 15). These tests indicated that the 80% alcohol/5% glycerin solution performed best to maintain the luster and coloration of the beak, as well as its malleability, so this solution was chosen as standard for the LABCOP Cephalopod Beak Collection (CMBC/LABCOP).

Identification of cephalopods based on their beaks

The beaks were identified using the available identification keys, scientific illustrations, and photographs (Clarke, 1986; Lu & Ickeringill, 2002; Vaske, 2006; Xavier & Cherel, 2009; Vaske, 2011). For this, each beak was examined under a stereomicroscope to visualize its diagnostic features, with some alcohol being applied to the specimen as it was focused, to avoid dehydrating the material. The principal characteristics of the upper and lower beaks of squids and octopuses are shown in Figure 1, including some of the morphometric parameters commonly used for the diagnosis of taxa.

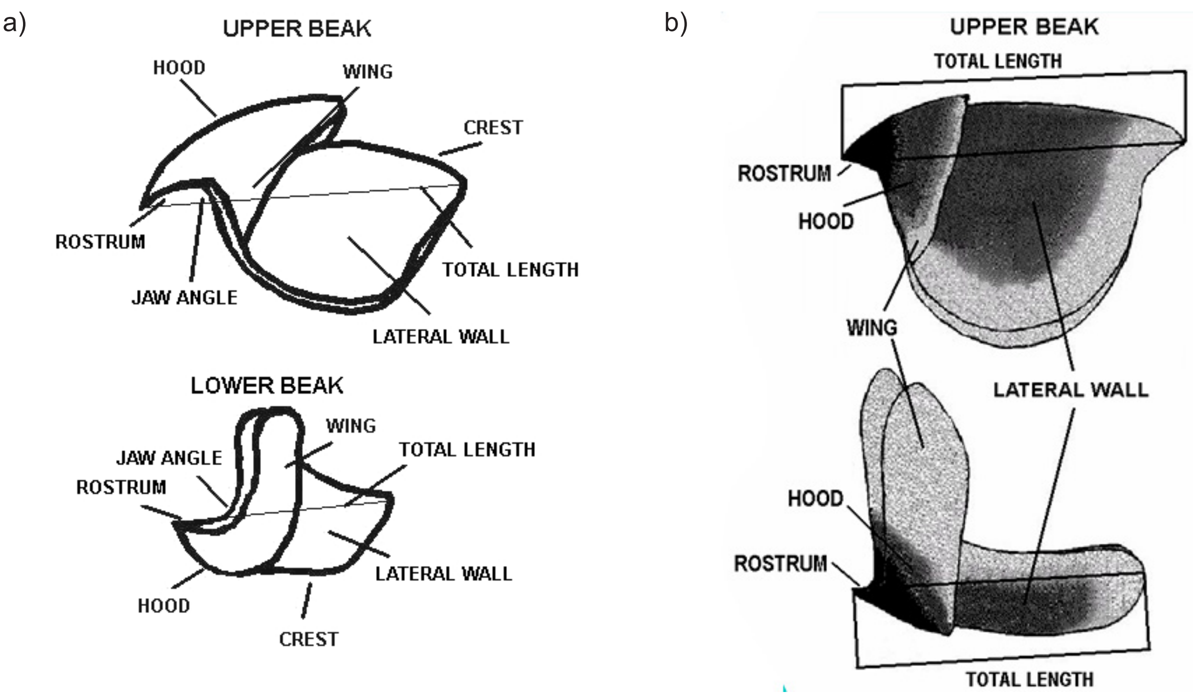

Figure 1 - Principal features and morphometric parameters of (a) squid and (b) octopus beaks. Adapted from Vaske (2006).

RESULTS

During the present study, a total of 562 beaks (270 lower and 292 upper) from the LABCOP Cephalopod Beak Collection (CMBC/LABCOP) were analyzed and 50 different cephalopod species were identified (Tables 2 and 3). It was necessary to update the nomenclature of some of the species identified in the reference material, based on the World Register of Marine Species (WORMS, 2020).

Table 2 - The species of the order Teuthida identified in the present study, with the number of upper and lower beaks assigned to each taxon.

Suborder	Family	Species	Collection Code	Lower Beak	Upper Beak
Myopsida	Loliginidae	Doryteuthis pleii	10	16	17
		Doryteuthis sanpaulensis	35	1	1
		Doryteuthis surinamensis	12	1	1
		Lolliguncula brevis	21	3	3
		Sepioteuthis sepioidea	3	2	2
Oegopsida	Ancistrocheiridae	Ancistrocheirus lesueurii	22	4	5
	Architeuthidae	Architeuthis dux	46	2	2
	Brachioteuthidae	Brachioteuthis riisei	38	1	2
	Chiroteuthidae	Chiroteuthis mega	42	1	1
		Chiroteuthis veranii	16	7	7
	Cranchiidae	Liguriella podophthalma	39	1	2
		Taonius pavo	2	7	9
	Cycloteuthidae	Discoteuthis discus	28	4	4
		Cycloteuthis akimushkini	31	1	1
	Enoploteuthidae	Abralia veranyi	50	0	4
		Enoploteuthis anapsis	5	3	2
		Enoploteuthis leptura	29	6	4
	Histioteuthidae	Histioteuthis corona	24	7	15
		Histioteuthis macrohista	26	9	11
	Lepidoteuthidae	Lepidoteuthis grimaldii	19	6	2
	Lycoteuthidae	Lycoteuthis lorigera	43	1	1
	Mastigoteuthidae	Magnoteuthis magna	48	2	3
	Pholidoteuthis	Pholidoteuthis adami	44	3	4
		Pholidoteuthis massyae	37	3	0
	Octopoteuthidae	Octopoteuthis megaptera	34	5	6
		Taningia danae	25	5	5
	Ommastrephidae	Eucleoteuthis luminosa	33	0	1
		Hyaloteuthis pelagica	11	13	17
		Illex argentinus	20	22	13
		Ommastrephes bartramii	18	1	1
		Ornithoteuthis antillarum	45	1	1
		Sthenoteuthis pteropus	8	46	56
		Todarodes filippovae	40	7	6
	Onychoteuthidae	Onychoteuthis banksii	30	2	2
		Onykia carriboea	32	8	9
		Onykia robsoni	41	3	3
	Thysanoteuthidae	Thysanoteuthis rhombus	1	9	11

Table 3 - The species of the orders Octopoda and Vampyromorpha identified in the present study, with the number of upper and lower beaks assigned to each taxon.

Order	Suborder	Family	Species	Colection Code	Lower Beak	Upper Beak
Octopoda	Cirrata	Opistoteuthidae	Opistoteuthis sp	47	2	3
	Incirrata	Alloposidae	Haliphron atlanticus	27	0	2
		Amphitretidae	Japetella diaphana	4	7	9
		Argonautidae	Argonauta nodosus	14	1	1
		Bathypolypodidae	Muusoctopus oregonae	49	1	1
		Eledonidae	Eledone massyae	13	13	11
		Octopodidae	Macrotritopus defilippi	9	1	1
			Octopus insularis	36	0	1
			Octopus tehuelchus	15	1	1
			Octopus vulgaris	7	8	8
		Ocythoidae	Ocythoe tuberculata	23	16	12
		Tremoctopodidae	Tremoctopus violaceus	6	3	3
Vampyromorpha	-	Vampyroteuthidae	Vampyroteuthis infernalis	17	4	5

DISCUSSION

The reliability of the identification of cephalopod species based on the morphology of their beaks depends on the adequate analysis of morphological features and morphometric parameters. While complex, this approach can provide a precise identification of taxa (Furness et al., 1984; Vaske, 2006; Chen et al., 2012). Lefkaditou & Bekas (2004) found that it is possible to identify *Eledone cirrhosa* of different ages based on the analysis of morphometric parameters and the pigmentation patterns of the beak, concluding that these features are essential diagnostic criteria for the evaluation of the specimens. Mercer et al. (1980) also showed that the morphological and morphometric characteristics of the beaks of *Illex illecebrosus* can be used to determine the sex in this species, given its sexual dimorphism.

The identification key for cephalopod beaks published by Clarke (1986) is the most traditional and comprehensive reference work for this task, given that it covers an enormous diversity of taxa from throughout the world's oceans. However, the author only included the lower beak in this publication, claiming that the greater number of folds, grooves, and protrusions found in this half of the beak made it more suitable for the reliable identification of taxa. While this may be true, it did complicate the process here, given that some specimens were represented only by the upper beak. Fortunately, Lu & Ickeringill (2002), Xavier and Cherel (2009) and Vaske (2006, 2011) provide photographs and illustrations of the upper beak, which facilitated the identification process. In this case, minor details, such as the pigmentation of the beak and the length of the rostrum, were sufficient for diagnosis, without using other types of morphometric measurement.

The CBMC/LABCOP collection includes a relatively large number of species in comparison with the known cephalopod diversity of the Brazilian coast (Vaske, 2006; Vaske, 2011). Based on a survey using the *SpeciesLink* information software (CRIA, 2020), specimens of 31 equivalent species were identified in Brazilian museums, although none of these institutions hold specimens of cephalopod beaks, but rather, only of the complete individual.

Although similar numbers of upper and lower beaks were found in the CBMC/LABCOP collection, many specimens lacked one half of the beak, which hampered species identification. This is reinforced by the fact that most published studies of cephalopod beak identification focus only on the lower beak to diagnose taxa. The present study also adopted new preservation techniques that are not mentioned in the available curation manuals.

As much of the material in the CBMC/LABCOP collection was of unknown provenance, and lacks collection dates and other standard information, it is of little value for scientific research, which means that the primary vocation of the material is as a didactic resource and/or reference collection for cephalopod research. As specimens in this type of collection are subject to constant handling (Ingenito, 2014), and thus more vulnerable to damage, they require extra care, ranging from constant checks on the liquid medium to adequate storage and handling. While museums in most countries tend to have only indirect links with universities, in Brazil, there is typically a strong relationship between research and teaching institutions, and zoological collections (Martins, 1988), which reinforces the need to maintain collections such as the CBMC/LABCOP within the academic sphere, where it may represent an invaluable resource for future research.

CONCLUSION

The LABCOP cephalopod beak collection (CBMC/LABCOP) contains 562 specimens representing 50 different cephalopod species. The lack of information on collection procedures and beak preservation for scientific collections limited the scope of the present study, and demanded the development of new methods and techniques. Given the lack of known provenance of many specimens and the difficulty of preserving these specimens in liquid medium, the collection will be most valuable as a didactic resource or reference collection for future cephalopod research. The next step in the consolidation of this collection will be the elaboration of a curation protocol in line with LABCOP standards.

The identification of cephalopod beaks is a demanding process, especially when the specimen is incomplete (i.e., only the upper or lower part is available). As the availability of both the lower and upper beaks may greatly enhance the reliability of the taxonomic diagnosis, priority should be given to the preservation of complete specimens, which will facilitate both teaching and research.

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