



Waif or hybrid? Observation records of rare coloration grouper in Djibouti

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Abstract We observed groupers with an unusual color pattern in Ghoubet-Al-Kharab Bay, Djibouti, on two occasions in 2014 and 2020. It matches the field observation of a juvenile *Cephalopholis formosa* in India in 1980. If this is the case, our observation represents a major range extension for *C. formosa*. Alternatively, based on the intermediate coloration of the specimens and sightings, we propose that they may be hybrids of *C. formosa* and *C. oligosticta*. We present two possible scenarios that may have facilitated hybridization: rarity and overlapping ranges in Djibouti with self-recruitment or rarity and overlapping ranges in another location (e.g., Socotra or the Arabian Sea) with long-distance larval dispersal to Djibouti. This hybridization is possible given the genetic similarity between the two putative parent species and because similar hybridization cases have been recorded within this genus and family elsewhere. However, both of these scenarios would require a range extension for one or both parent species as

they are not previously known to overlap. Nevertheless, further field observations and genetic studies are required to verify the proposed identification of the putative hybrid and test the presented scenarios.

Keywords Grouper · Hybridization · Distribution range

The Gulf of Tadjoura is an extension of the Gulf of Aden that penetrates 20 km into the Horn of Africa via Ghoubet-Al-Kharab Bay in Djibouti (Fig. 1), which is a semi-enclosed bay separated from the rest of the Gulf of Tadjoura by a narrow pass (Moussa Omar et al. 2016). Located at the crossroads between the Red Sea and the Indian Ocean, the Gulf of Tadjoura harbors significant biodiversity (Boldrochi et al. 2018), including species endemic to the Red Sea, Gulf of Aden, and Arabian Sea (Lips et al. 2016). Since 1998, Djibouti has undertaken several surveys to update and refine the inventory of their ichthyological fauna.

In September 2014, M. A. Samoily observed a grouper with an unusual color pattern (Fig. 1) while surveying reef fishes at Vierge Rouge in Ghoubet-Al-Kharab Bay in Djibouti (Cowburn et al. 2019). On March 4, 2020, we observed a similar individual almost 20 km west of Veirge Rouge at a depth of less than 15 m on top of “The Crack” at Afar Triple Junction in Ghoubet-Al-Kharab Bay (latitude N11.58430, longitude E43.53760), which is the meeting point

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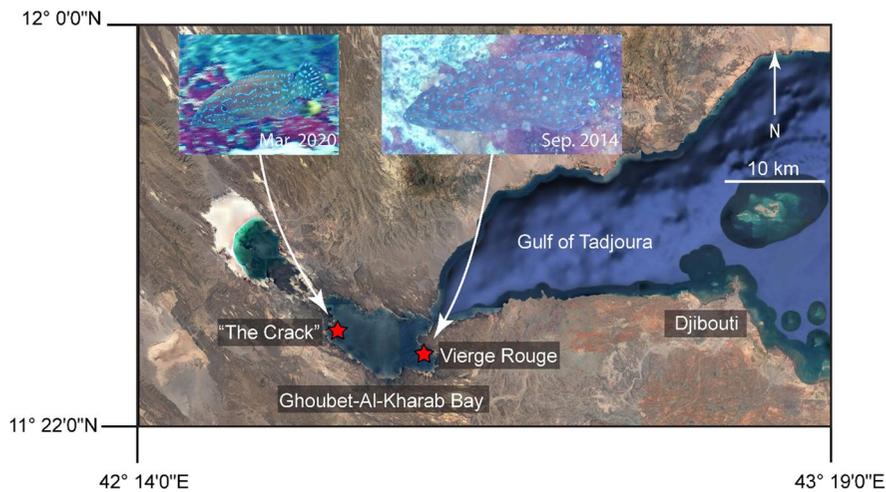


Fig. 1 We observed groupers with an unusual color pattern in Ghoubet-Al-Kharab Bay in Djibouti during underwater surveys conducted at Vierge Rouge in September 2014 and at “The Crack” in March 2020, which may be hybrids of *Cephalopholis formosa* and *C. oligosticta*. The putative hybrid images

have color balanced due to the poor quality of the original photos, taken using underwater cameras without independent light sources. Fish photo credits to M. A. Samoilys (2014 sighting) and A. L. Green (2020 sighting)

of the Arabian, Somali, and Nubian tectonic plates. Given slight differences in the color patterns (Figs. 1 and 2), these appear to be two different individuals, both of which appeared to be adults or large sub-adults (approximately 30 cm in length).

The coloration of the observed fish in the current study matches a sample archived in the Bishop Museum (sample ID BPBM 27,656 – 5) (Fig. 2). This 45-mm-long (standard length) museum specimen is labeled as a juvenile *Cephalopholis formosa*. It was

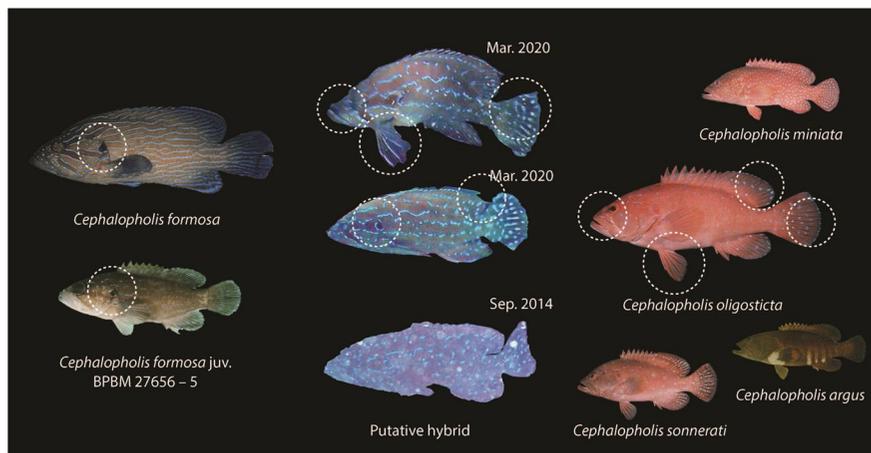


Fig. 2 Color pattern comparisons among the putative hybrid grouper *Cephalopholis oligosticta* X *C. formosa* observed in Djibouti (credits to M. A. Samoilys, 2014 sighting and A. L. Green, 2020 sighting) and potential purebred parent species (*C. formosa*, *C. oligosticta*, *C. miniata*, *C. sonnerati*, and *C. argus*; photo credits to J. E. Randall and R. F. Myers). A juvenile *C. formosa* held in the Bishop Museum (Hawaii,

USA, specimen ID BPBM 27,656–5) exhibited similar coloration patterns as the putative hybrids. The observation dates are labeled next to each putative hybrid photo (note that photos from March 2020 are of the same individual), and the white-dashed circles indicate the color patterns that are similar among the putative hybrids and parent species

collected in India, Kerala State, Kovalam, rocky point off Raja Hotel (about 300 m north of lighthouse), around 7 m depth with rotenone by J. E. Randall and W. F. Smith-Vaniz on February 11, 1980. Our sightings in Djibouti could represent a range extension of *Cephalopholis formosa* from the Arabian Gulf to the west side of the Gulf of Aden. The limited records of such an unusual color pattern in Djibouti indicate that such individuals may be waifs, a biogeographic term used to describe the occasional individual occurring outside of their species distribution (Backus 1986). Unfortunately, we cannot extract DNA to confirm the identification using genetic methods due to the formalin fixation performed on the Bishop Museum sample.

Individuals with intermediate color patterns can be hybrids (DiBattista et al. 2015; He et al. 2017; He et al. 2020; He et al. 2019; Hobbs and Allen 2014; Tea et al. 2020). Based on the intermediate color patterns of the individuals we observed in Djibouti, they may be hybrids of *Cephalopholis formosa* and *C. oligosticta* (Fig. 2). Key features that these individuals share with the putative parent species include a large black spot on the rear of the operculum, blue longitudinal lines and spots on a brown body, blue dots and

bars on the snout in front of the eyes, a blue dotted pattern on the rear of the second dorsal fin, and blue stripes on the pelvic fin rays (Fig. 2). Other alternative candidates for putative parents (e.g., *C. argus*, *C. miniata*, and *C. sonnerati*) seem less likely because they do not share this combination of unique traits with the putative hybrid (Fig. 2).

Both putative parent species are rare or not previously recorded in Djibouti, although they are known to occur elsewhere in the West Indo-Pacific (Fig. 3). *Cephalopholis oligosticta* is an endemic species of the Red Sea and Gulf of Aden (Bogorodsky and Randall 2019; Golani and Fricke 2018). Lips et al. (2016) noted that this species is rare in Djibouti, having seen it twice while diving in Ghoubet-Al-Kharab Bay and for sale in the fishing port. We observed one or two *C. oligosticta* while surveying reef fishes in Ghoubet-Al-Kharab Bay in September 2014 and March 2020. *Cephalopholis formosa* has a recorded distribution ranging from Arabian Gulf, India, and Sri Lanka to Western Australia (Allen et al. 2012; Barik et al. 2018; Humann et al. 2015; Taquet and Diringier 2012; Tavakoli-Kolour et al. 2015). *Cephalopholis formosa* has not been recorded in the Gulf of Aden (Lips et al.

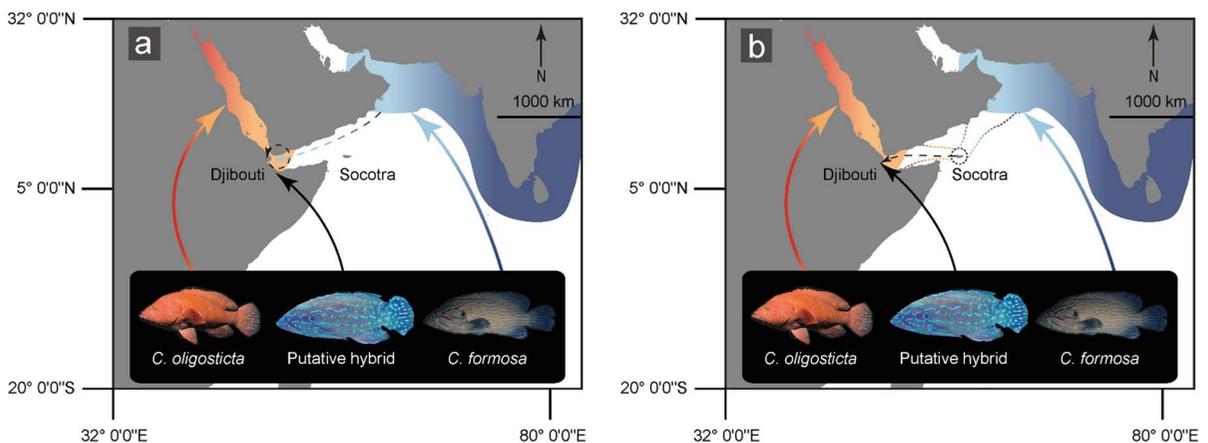


Fig. 3 Two scenarios that may explain the existence of the observed putative *Cephalopholis* hybrid (photo credit to A. L. Green) in Ghoubet-Al-Kharab Bay in Djibouti, where the recorded distribution ranges of *Cephalopholis oligosticta* (photo credit to R. F. Myers) and *Cephalopholis formosa* in the West Indian Ocean (photo credit to J. E. Randall) are represented by shaded red and blue colors, respectively. Please note that the putative hybrid images have not been color balanced (see Fig. 1). Scenarios include **a** hybridization may have occurred in Djibouti with local self-recruitment. This would require a major range extension for *Cephalopholis formosa*

(see blue-dashed line arrow). The dashed black circle indicates self-recruitment after hybridization in Djibouti. **b** Hybridization may have occurred in another location (e.g., Socotra) followed by long-distance larval dispersal to Djibouti. The dashed red/blue lines indicate the range extensions that would be required for both putative parent species and the dashed black circle indicates the possible hybridization zone. The black-dashed line with an arrow indicates the possible long-distance larval dispersal of the hybrid to Djibouti after hybridization via seasonal sea surface currents in the Gulf of Aden (described in Vitale et al. (2017)

2016 present five species of *Cephalopholis*, but not *C. formosa*). The most recent published version of a database of the region's reef fish distributions, maintained by R. F. Myers, does not report the presence of *C. formosa* in Djibouti (see Table S2 in DiBattista et al. 2016). Both *C. formosa* and *C. oligosticta* are known to occur in barren, shallow (< 15 m), silty, and sheltered reefs, which are conditions typical of Ghoubet-Al-Kharab Bay. Here we propose two potential scenarios to explain the existence of the observed putative *Cephalopholis* hybrid:

Scenario 1: Purebred *C. formosa* and *C. oligosticta* hybridized at Ghoubet-Al-Kharab Bay and the hybrid offspring were the result of local self-recruitment (Fig. 3a).

This scenario would require both parent species occurring in Ghoubet-Al-Kharab Bay, which includes suitable habitats for these species. *Cephalopholis oligosticta* are rare but known to occur in Ghoubet-Al-Kharab Bay. However, the presence of *C. formosa* in Ghoubet-Al-Kharab Bay may require a major extension to its known distribution since the closest published occurrence records of this species is in the Arabian Gulf. Therefore, this scenario would require one or a few *C. formosa* waifs, traveling via ocean currents (Franklin et al. 2019) as pelagic larvae, westwards across the Arabian Sea and settling in Ghoubet-Al-Kharab Bay.

Scenario 2: Purebred *C. formosa* and *C. oligosticta* hybridized at another location (e.g., Socotra) and the hybrid offspring in Ghoubet-Al-Kharab Bay result from long-distance larval dispersal (Fig. 3b).

Ghoubet-Al-Kharab Bay is approximately 1000 km west of the well-known hybridization hotspot at Socotra, which is considered a suture zone of multiple biogeographic providences (DiBattista et al. 2015). However, despite extensive surveys, neither of these two species has been recorded in Socotra (Zajonz et al. 2019). The known distribution range of *C. oligosticta* in the Gulf of Aden is limited to Djibouti, while the closest published distribution record of *C. formosa* to Socotra is in the Arabian Gulf. However, it is possible that a few waifs of these two species may have scattered beyond the edge of their known distribution to Socotra by larval transport via seasonal sea surface currents in the adjacent area (Vitale et al. 2017).

Given the extensive nature of surveys conducted in Socotra (Zajonz et al. 2019), if the putative parents are there, they must be very rare or in habitats not well surveyed to date (e.g., in deep water). Like other marine fish hybridization cases in Socotra, the rarity of at least one of the putative parent species may promote the formation of interspecific breeding pairs (Hobbs and Allen 2014), such as *C. oligosticta* and *C. formosa*, if they co-occur.

After parental spawning, the putative hybrid offspring could have traveled westwards via long-distance larval dispersal across deep water in the Gulf of Aden by the seasonal sea surface currents (Vitale et al. 2017) and settled in suitable habitat in Ghoubet-Al-Kharab Bay. Similar long-distance larval dispersal has been inferred to have occurred in coastal areas of Oman (Simpson et al. 2014).

Another possibility is that one or more *C. oligosticta* waifs may occur in another location within *C. formosa*'s range (e.g., in the Arabian Sea), being transported there by long-distance larval dispersal from the Gulf of Aden. Once the parent species hybridized, the hybrid offspring may have dispersed all the way back to Djibouti. However, this seems less likely given that this scenario would require rare long-distance dispersal of larvae in two directions, although it may be possible given variations in seasonal currents in the region (Vitale et al. 2017). Furthermore, if this were the case, hybrids would likely be more widespread than in a remote corner of Djibouti. This could even be a possible explanation for the aforementioned sample in the Bishop Museum; it may be an unidentified hybrid individual.

Since we believe that we observed two individuals with similar color patterns in Djibouti, scenario 1 seems more likely than scenario 2 because scenario 1 would require only a single waif *C. formosa* to produce multiple hybrid offspring that are retained within Djibouti, while scenario 2 would require multiple hybrid offspring to be produced in another location and undergo long-distance dispersal all the way to the same far-flung corner of Djibouti.

The putative hybridization we observed between *C. formosa* and *C. oligosticta* is possible since similar natural hybridization cases have been observed within the genus *Cephalopholis* in the Christmas-Cocos Islands hybridization hotspot (Payet et al. 2016). Also, at the family level, both natural hybridization and artificial hybridization attempts for aquaculture

have been recorded for other genera in the Serranidae (Ching et al. 2018; He et al. 2018; Qu et al. 2018; van Herwerden et al. 2002). Furthermore, from a genetic point of view, the similarity (p -distance, based on GenBank COI sequences) between *C. formosa* and *C. oligosticta* is ~0.14, which falls within the range of p -distances for documented grouper hybridization cases in aquaculture and in the wild (0.00~0.17). Although *C. formosa* and *C. oligosticta* are not genetically similar enough to be considered sister species (Ma and Craig 2018), the p -distance value suggests that crossbreeding is possible.

Further studies are now required to determine the validity of the putative hybrid and the potential scenarios to explain their existence. Genetic investigations should be performed using non-lethal tissue samples from putative hybrids to confirm the parent species. Field studies are also required to determine if the ranges of *C. oligosticta* and *C. formosa* overlap in the Gulf of Aden, Socotra, or other locations (e.g., the Arabian Sea). Additional field surveys should also be conducted in Djibouti to determine the rarity or otherwise of the putative hybrids with similar intermediate coloration patterns.

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