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When Is a Male Turtle Not a Male?—Observations on Intersex Turtles

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ABSTRACT. – We report on an intersex green turtle, *Chelonia mydas*, observed at Raine Island, Australia that externally resembled an adult male but internally—based on laparoscopic examination—had both ovarian and testicular structures resembling those of an adult. This individual was the first intersex turtle that has been recorded on a nesting beach during the 39 years of extensive census studies in Queensland. Its observation confirms that the definitive identification of sex with marine turtles remains dependent on observations of their gonads or on observing eggs being laid.

One of us (CJL) had the pleasure of listening to the late Archie Carr and Joop Schultz at the 1978 IUCN SSC MTCG meeting debate the efficacy of using the presence of a long tail as a definitive criterion for determining the sex/maturity for adult male sea turtles. Joop's uncertainty with the use of "long-tailed" for identifying adult males was influenced by his observations of long-tailed *Chelonia mydas* coming ashore and digging deep nest cavities in Surinam during 1972–1973 breeding seasons (Pritchard 1979). Pritchard (1979) also observed a "male" *C. mydas* emerge from the sea and enter the nesting habitat before returning to the sea at James Island, Galapagos Islands in 1970. More recently, Troëng (2000) and Zárate Bustamante (2006) have described apparent male *C. mydas* ashore and behaving like nesting females on beaches in Caribbean Costa Rica and in the eastern Pacific, respectively.

We had difficulty in interpreting the presence of an apparent adult male *C. mydas* (tag number I22113) ashore among nesting females, 70 m inland of the high tide mark within nesting habitat at the western end of Raine Island on 29 November 1993. This turtle had an elongated tail (~30 cm beyond carapace) and large, curved front flipper claws. Raine Island, 11°36′S, 144°01′E, has been the primary index beach for monitoring the population dynamics of the northern Great Barrier Reef *C. mydas* genetic stock since 1976 (Limpus et al. 2003).

This same "male" was recorded ashore among nesting females at Raine Island on 3 December 2002 at 2034 hours: curved carapace length of 101.5 cm and tail length beyond carapace of 32.0 cm. It was removed from the beach to an adjacent vessel, its reproductive system visually examined using laparoscopy (Limpus et al. 2005), and released at 0520 hours on 4 December 2002. Only the right gonad and associated ducts were viewed in detail. It had ovaries with numerous large, ~32 mm diameter, well-vascularized, yellow ovarian follicles resembling normal mature follicles seen in breeding females. The abundance of large follicles was consistent with a turtle capable of laying several clutches within a breeding season. No corpora lutea and no large atretic follicles were observed on the ovary, in spite of an extended search. Also, no corpora albicantia (healed corpora lutea from previous breeding seasons) were found. Large numbers of small previtellogenic follicles and ~2 mm diameter "atretic disks" were present within the surface of the ovary. The atretic disks are the remnants of the resorption (atresia) of vitellogenic follicles in a previous season(s). The absence of corpora albicantia and presence of atretic disks indicated that this turtle had not ovulated in a previous breeding season but had resorbed large follicles. An enlarged and convoluted oviduct suspended within a mesentery lateral to the ovary and leading to the cloaca resembled a mature oviduct as seen in breeding females. It contained no oviducal eggs. A large testis lay dorsal and lateral to the ovary. The testis was distended with highly visible seminiferous tubules resembling that of a breeding male, but there was no pendulous, enlarged epididymis adjacent to the testis. A large penis comparable in size to that of normal breeding male *C. mydas* lay within the cloaca. Although externally this turtle morphologically resembled a breeding male *C. mydas*, it had none of the courtship damage bite marks usually seen on the rear edges of flippers or dorsal tail of breeding male *C. mydas* at this time of the year (Limpus 1993). Also, it had no bite marks to the neck and shoulders and no claw marks on the margin of the carapace adjacent to shoulders as are common for breeding females.

This turtle returned to the nesting habitat of Raine Island over 4 nights, 6–9 December 2002 (Fig. 1). Observations were made of its apparent nesting behavior (Table 1). Definition of nesting behavior follows Bustard and Greenham (1969). At no time did this turtle display behavior indicative of a male attempting to mate with the numerous females in its vicinity while it was on the nesting beach.

Basking turtles are regularly seen on some Australian beaches, by day and by night (Barrett 1919; Limpus et al. 1994a, 2005). This turtle did not remain on the beach during the day, and its continual movement and digging at night in no way resembled the resting behavior of basking

Figure 1. Intersex *Chelonia mydas* (I22113), on left, ashore within the nesting habitat at Raine Island, northern Great Barrier Reef, 6 December 2002. The *C. mydas* on the right was an adult female with a normal length tail.

turtles when on shore. The onshore activity of this turtle could not be interpreted as basking behavior.

We previously recorded an injured adult male *C. mydas* (T77695, 30 July 1994) digging what appeared to be an "egg chamber" while it was held ashore for rehabilitation at the Shoalwater Bay foraging area in central Queensland. It was allowed to wander on the ground in a shaded area over several days. This turtle dug a shallow hole beneath its tail using the characteristic egg chamber digging actions of a nesting female. Laparoscopic examination showed this male had normal gonad morphology for an adult that was not in a spermatogenic cycle: large testis with nondistended seminiferous tubules and pendulous epididymis with a translucent, nondistended duct. These observations suggest that stressed turtles may display displacement behavior that includes segments of nesting behavior. This does not appear to be the case with I22113, given its repetitive voluntary return to the nesting beach where it engaged in activities normally associated with nest digging (even though its was physiologically not prepared for oviposition).

During studies of loggerhead turtles, *Caretta caretta*, green turtles and hawksbill turtles, *Eretmochelys imbricata*, in eastern Australia feeding areas, large samples of turtles have been captured and their gonads examined visually by laparoscopy to determine their sex, maturity, and breeding status (Limpus 1992; Limpus et al. 1994b, 1994c, 2005; Limpus and Limpus 2003). It was unusual

Table 1. Summary of beach activity during the 2002 nesting season at Raine Island, Australia by a *Chelonia mydas* individual bearing tag number I22113.

Sighting no.	Date	Time (hours)	Beach activity
1	3 Dec	2034	This turtle was encountered on the beach and removed for laparoscopic analysis (see text; released at 0520 hours on 4 Dec).
2	6 Dec	2300	Attempting to dig a body pit using occasional front flipper sweeps, followed by an apparent attempt to dig an egg chamber with about 7 alternating digs by each hind flipper; the long tail obstructed this rear flipper digging.
2	6 Dec	2307	Moving and wandering through the nesting habitat, bumping into turtles attempting to nest nearby.
2	6 Dec	2320	Starting to dig another body pit with 3 sweeps of the front flippers.
2	6 Dec	2322	Moving off towards the waterline, attempting 2 further body pit digs with 2 sweeps of the front flippers at each site before leaving the beach.
2	9 Dec	0055	Attempting to dig a body pit using sweeps of the front flippers simultaneously.
2	9 Dec	0103	Commencing to dig an apparent egg chamber using alternate digs by the hind flippers with digging actions resembling that of nesting females; the tail did not obstruct this digging; because of dry sand, the egg chamber walls continually collapsed.
3	9 Dec	0117	Abandoned digging and left the nesting habitat.
4	9 Dec	at night	Made 3 successive attempts to dig body pits but moved off on each occasion after bumping or being bumped by other turtles; the turtle left the nesting habitat without attempting to dig an egg chamber.

Table 2. The occurrence of intersex turtles (turtles that are not distinctly male or female) identified by visual examination of their gonads via laparoscopy in feeding areas studies in eastern Australia.[a]

Species and sampled years	Locality	No. of intersex turtles (%)	Total no. of turtles examined for sex
Caretta caretta			
1990–2004	Moreton Bay	3 (0.36)	832
1982–1999	Southern Great Barrier Reef	2 (0.31)	646
1986–1999	Hervey Bay and Shoalwater Bay	0	32
1988–1997	Clack Reef, north Great Barrier Reef	0	4
Chelonia mydas			
1990–2004	Moreton Bay	1 (0.05)	2020
1882–1999	Southern Great Barrier Reef	1 (0.03)	3748
1986–1999	Hervey Bay and Shoalwater Bay	1 (0.03)	3960
1988–1991	Repulse Bay	0	350
1988–1997	Clack Reef, north Great Barrier Reef	1 (0.10)	1049
Eretmochelys imbricata			
1982–1999	Southern Great Barrier Reef	3 (0.81)	370
1988–1997	Clack Reef, north Great Barrier Reef	0	60

[a] Limpus (1992), Limpus et al. (1994a, 1994b), Limpus and Limpus (2003), J. Miller and CJL (unpublished data).

for a turtle not to be distinctly male or female: *C. mydas* < 1 per 1000 turtles examined; *C. caretta* $\sim$3 per 1000 turtles; *E. imbricata* $\sim$8 per 1000 turtles (Table 1). The term intersex was used in these studies to describe turtles that were morphologically neither male nor female. In most instances, the gonads of these intersex turtles were ovi-testes with seminiferous tubules in the medulla and scattered small previtellogenic follicles in the cortex. Several of these intersex turtles had their gonads re-examined over periods of up to 15 years, and none displayed evidence of changing sex. Only 1 turtle, *E. imbricata* T23066, had a gonad with no structures resembling either an ovary or testis (Limpus 1992). I22113 is the first intersex turtle we have observed with ovarian and/or testicular structures resembling that of an adult. It was also the first intersex turtle that has been recorded/recognized on a nesting beach during the 39 years of extensive census studies in Queensland during which over 100,000 nesting females were tagged. Long-tailed intersex turtles like I22113 are rare individuals.

While this turtle should display atypical endocrine profiles (Hamann et al. 2003), and hence engage in atypical behavior, it provides some insights into *C. mydas* reproductive biology. It had apparently completed vitellogenesis, as evidenced by the presence of many mature-sized ovarian follicles and was in advanced spermatogenesis as evidenced by a distended testis with enlarged seminiferous tubules. Apparently the physiological processes that underlie the maturation of female and male gonads are not mutually exclusive. In addition, one or both of these processes apparently was sufficient to set in train the drives that stimulated this turtle to make a breeding migration to this traditional nesting beach. Based on the absence of external injuries, there is no indication that it had been able to attract a mate into courtship behavior. Its presence at Raine Island in 2 breeding seasons, 9 years apart, is consistent with both male and female green turtle fidelity to their respective breeding sites (Limpus 1993; Limpus et al. 2003). The authors have examined the reproductive systems of more than 1000 marine turtles on nesting beaches, mostly at Raine Island (Limpus et al. 2003). This is the first individual recorded ashore for nesting that did not contain oviducal eggs.

With repetitive returns to the beach over successive nights, I22113 behaved like a gravid female that had failed to successfully lay and was returning for additional nesting attempt (Miller and Limpus 2003), yet it was not gravid. I22113 had mature-sized ovarian follicles and had not ovulated. No explanation is offered at this time as to what hormonal trigger(s) would stimulate it to repetitively haul-out on Raine Island to initiate nesting behavior over a series of nights in the absence of oviducal eggs or at least a prior ovulatory event.

Natural abnormalities in gonad development can be expected at low levels within wild populations. Data in Table 2 provide benchmark measurements from a series of foraging areas in eastern Australia against which the occurrence of such anomalies can be compared in the future. A "male" turtle is not a male when it is trying to make eggs. This turtle was obviously hormonally and behaviorally abnormal. It appears not to be able to contribute genes to the next generation and should not be considered as part of a breeding population.

The definitive identification of sex with marine turtles remains dependent on observations of their gonads or on observing eggs being laid. However, the error from using external characteristics to identify adult males, as with this turtle, should be trivial at a population level (Table 2). To assist in better defining the reproductive status of "male" turtles observed ashore and exhibiting nesting behavior (Troëng 2000; Zárate Bustamante 2006), it is recommended that additional data regarding their gonad development using laparoscopy or ultrasound scans and/or their hormonal status using blood sample analysis should be recorded where possible.

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