

SERIAL SPAWNING AND BATCH FECUNDITY OF *MERLUCCIUS CAPENSIS* AND *M. PARADOXUS*

R. F. OSBORNE*, Y. C. MELO†, M. D. HOFMEYR‡ and D. W. JAPP§

Histological analysis of the ovaries and the presence of multiple modes in oocyte size frequency distribution of Cape hake *Merluccius capensis* and *M. paradoxus* indicates that they are serial spawners. Batch fecundity, calculated by means of the “hydrated oocyte method”, was positively correlated to ovary-free fish mass and total fish length. The mean relative fecundity (ovary-free mass) of *M. capensis* and *M. paradoxus* was 160 ± 12 and 306 ± 25 eggs·g⁻¹ respectively.

Two species of Cape hake, the shallow-water species *Merluccius capensis* and the deep-water species *M. paradoxus* are the main target species of the South African demersal fishery, contributing around 70% by mass of the catches (Stuttaford 1998). *M. capensis* is generally caught on the South Coast, and off the West Coast in waters shallower than 300 m. *M. paradoxus* form the bulk of the catches on the West Coast at depths of 200–600 m, but there is considerable overlap in the distribution of the two species at depths between approximately 150 and 450 m (Botha 1980).

Studies on the spawning behaviour of the Cape hake include the work of Hecht (1976) on the South African east coast, who used gonadosomatic index, frequency occurrence of ripe gonads, and condition factor to infer reproductive seasonality and spawning frequency in *M. capensis*. Hecht (1976) derived a seven-point maturity scale for *M. capensis*. From macroscopic examination of the gonads of hake from the West and South-East coasts, Botha (1980, 1986) and Payne (1986) determined the length and age at 50% and complete maturity, as well as the duration and time of the spawning season of *M. paradoxus* and *M. capensis*. However, there is, no understanding of the fecundity or the mode of spawning in Cape hake.

Histological techniques and oocyte size frequency analyses are used in the present study to investigate spawning type in *M. capensis* and *M. paradoxus*. Batch fecundity is estimated by means of the “hydrated oocyte method”.

MATERIAL AND METHODS

Spawning type

Spawning type was investigated by means of histological analysis of ovaries from hake collected on the south coast of South Africa in June 1996 (for *M. capensis*) and on the West Coast in July 1996 (for *M. paradoxus*). Cape hake are believed to spawn throughout the year (Botha 1980). For histological analysis, the ovaries were dehydrated through a series of ethanol immersions, cleared with toluene and embedded in paraplast. Sections (5–7 µm thick) were stained with Harris’s haematoxylin, followed by eosin counterstain. Each ovary was histologically classified according to the most advanced oocyte stage present in the ovary. Oocyte stages were analogous to the six ovarian maturity stages described by Herrera *et al.* (1988) for *M. gayi* and Balbontín and Bravo (1993) for *M. australis*: 1 – virginal, 2 – immature, 3 – maturing, 4 – mature/ripe, 3a – partially spent and 5 – regressing/resting. In the present study, the partially spent stage (3a) is referred to as Stage 5 and the regressing/resting stage (5) as Stage 6.

Oocyte diameter was measured, by means of an ocular micrometer, from five females per maturity stage. The diameter of 50 oocytes per female was measured. The measurements of oocytes found in hydrated (Stage 4) ovaries were taken from formalin-preserved

* Marine & Coastal Management (formerly Sea Fisheries), Private Bag X2, Rogge Bay 8012, South Africa. Department of Zoology, University of the Western Cape, Private Bag X17, Bellville 7535, South Africa. E-mail: rosborne@sfri.wcape.gov.za

† Marine and Coastal Management. E-mail: ymelo@sfri.wcape.gov.za

‡ Department of Zoology, University of the Western Cape. E-mail: retha@botany.uwc.ac.za

§ Formerly Marine & Coastal Management; now Fisheries and Oceanographic Support Services, 16 Orchard Avenue, Lakeside 7945, South Africa. E-mail: jappy@iafrica.com

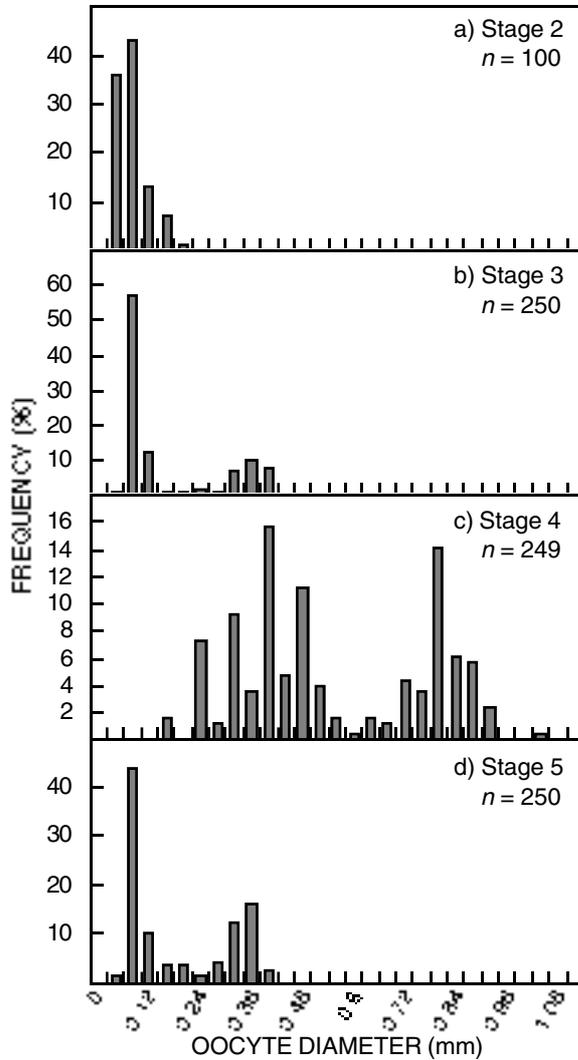


Fig. 1: Frequency distribution of oocyte size classes in the various ovarian stages of *M. capensis*

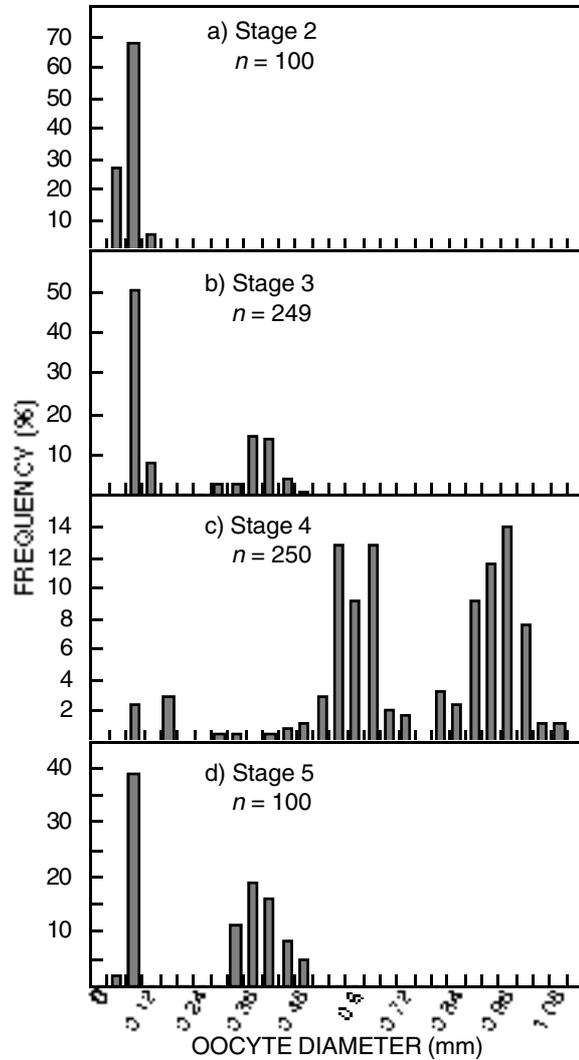


Fig. 2: Frequency distribution of oocyte size classes in the various ovarian stages of *M. paradoxus*

material, because the hydrated oocytes collapsed during histological processing.

Batch fecundity

Cape hake were collected on the South Coast (*M. capensis* – April 1996, 1997; *M. paradoxus* – May 1997). The hydrated ovaries of nine *M. capensis*

and 14 *M. paradoxus* were used to determine batch fecundity, according to the “hydrated oocyte method” (Hunter *et al.* 1985). Fish and gonad mass were measured to the nearest 1 g and total fish length to the nearest 1 cm. Fresh ovaries were preserved in 10% buffered formalin. For determination of batch fecundity, two samples of 0.5 g each, positioned about one-third of the distance from each end of the ovary, were removed from the right ovary and weighed to the

Table I: Estimated number of hydrated oocytes in two subsamples (a and b) taken from the right ovary of *M. capensis* and *M. paradoxus*

Fish number	<i>M. capensis</i>				<i>M. paradoxus</i>			
	Mean of three counts		Overall mean	SE	Mean of three counts		Overall mean	SE
	Subsample a	Subsample b			Subsample a	Subsample b		
1	646	*	646		629	590	610	19.2
2	665	700	682	17.5	535	498	517	18.7
3	739	678	709	30.8	633	642	637	4.3
4	610	606	608	1.7	642	617	630	12.2
5	804	619	712	92.5	590	594	592	2.0
6	696	665	681	15.7	595	688	642	46.3
7	774	755	765	9.7	632	709	671	38.7
8	*	918	918		685	703	694	8.8
9	715	633	674	41.0	489	511	500	10.8
10					653	632	643	10.5
11					629	693	661	32.2
12					860	726	793	66.8
13					658	617	638	20.8
14					473	550	512	38.2

* Eggs collapsed in one sample

nearest 0.01 g. The hydrated oocytes, identified by their translucent straw-coloured yolk and oil droplet, were counted under a dissecting microscope (Table I). Histological examination of the ovaries showed the absence of new post-ovulatory follicles. The presence of such follicles in the ovaries indicates that spawning has commenced, resulting in decreased numbers of hydrated oocytes. Such ovaries are not suitable for batch fecundity determination (Murua *et al.* 1996).

RESULTS

Description of maturity stages

Four of the above-mentioned six classes of ovaries were found in the hake under study, the virginal (Stage 1) and regressing (Stage 6) being absent. The oocyte stages were identical for each species, but the oocyte diameters differed slightly between species for each stage. The following lists the characteristics of each ovarian stage present. The measurements given in parenthesis refer to those of *M. capensis* and *M. paradoxus* respectively.

Stage 2 (immature ovaries, Figs 1a, 2a) – In both species, immature ovaries were characterized by a predominance of perinuclear and previtellogenic oocytes. The perinuclear oocytes were round to irregularly shaped, with a large nucleus and a thin surrounding layer of cytoplasm (diameter 0.08 and 0.02–0.09 mm). In the previtellogenic oocyte

(0.10–0.24 and 0.10–0.20 mm), the volume of cytoplasm had increased and yolk vesicles had developed as a single row of vacuoles around the periphery of the cytoplasm.

Stage 3 (maturing ovaries, Figs 1b, 2b) – Vitellogenic oocytes were present in both species (0.24–0.37 and 0.24–0.40 mm), characterized by the appearance of yolk granules in the cytoplasm. Early mature oocytes (0.40 and 0.44 mm) were present, in which the nucleus had started to migrate to the animal pole. A large oil droplet was evident in the mature oocyte.

Stage 4 (mature/ripe ovaries, Figs 1c, 2c) – Mature and hydrated oocytes predominated, with fewer vitellogenic oocytes present. In the mature oocyte, nuclear migration was almost complete and the yolk granules had started to fuse (0.60 and 0.84 mm). In the hydrated oocyte (diameter 0.70–1.10 and 0.88–1.08 mm), the nuclear membrane had disintegrated, and the yolk granules first fused to form yolk plates and then a homogenous mass that covered the entire oocyte.

Stage 5 (partially spent ovaries, Figs 1d, 2d) – Ovaries were similar to those of Stage 3. They contain vitellogenic oocytes (0.24–0.40 and 0.34–0.48 mm) in the process of yolk accumulation (indicative of another spawning) and developing perinuclear (0.08 and 0.09 mm) and previtellogenic oocytes (0.12–0.24 mm for *M. capensis*) for a subsequent spawning batch. As a result of poor histological preparation, no measurements were obtained for previtellogenic oocytes in the partially spent gonads of *M. paradoxus*. However, new post-ovulatory follicles were also present in both species, indicating that the fish had already spawned.

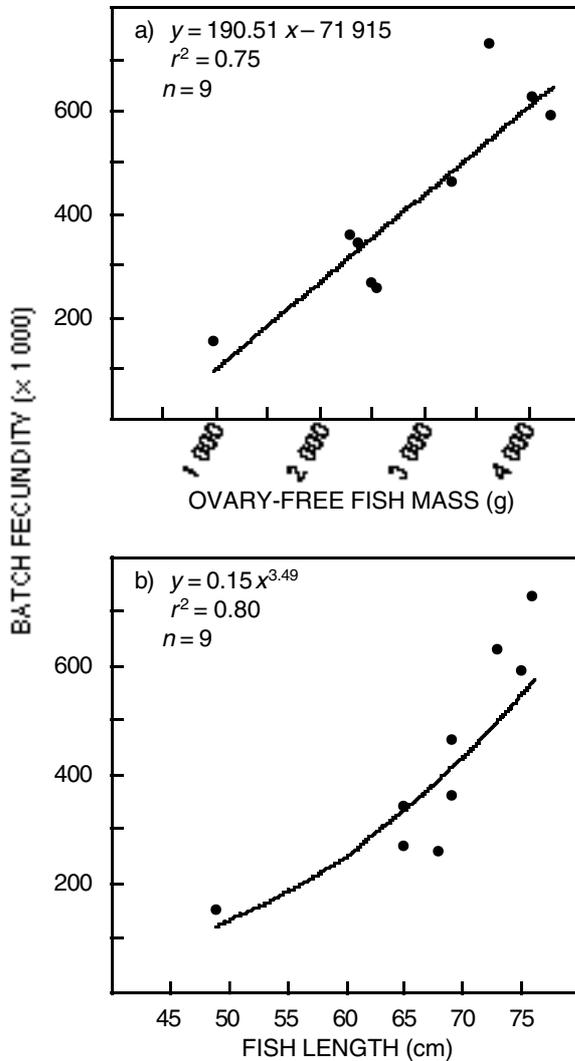


Fig. 3: Relationship between batch fecundity and (a) ovary-free fish mass (SE of slope = 41.7, mean mass of fish = 2 567 g) and (b) fish length for *M. capensis* (April 1996 and 1997)

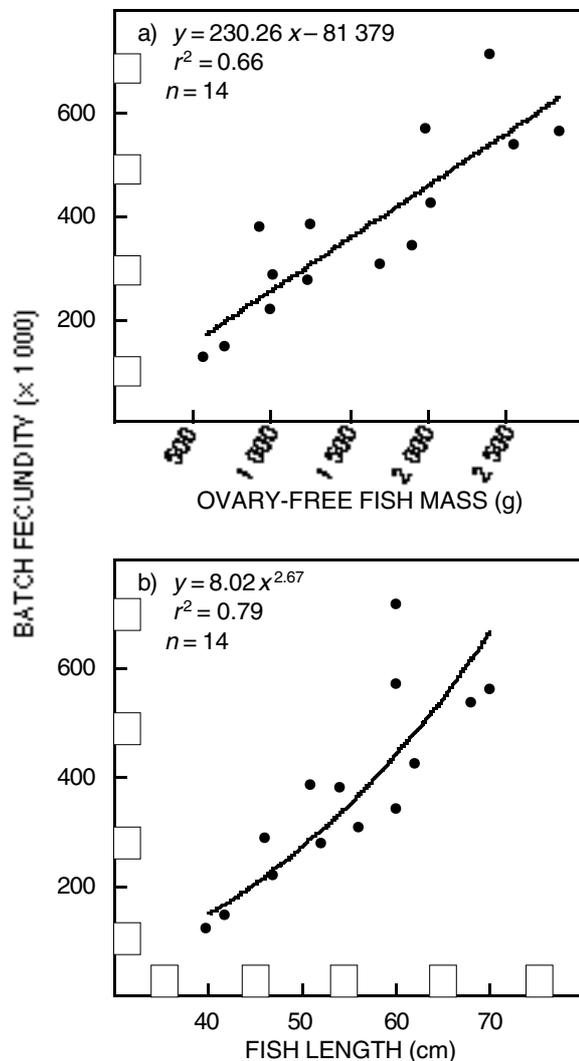


Fig. 4: Relationship between batch fecundity and (a) ovary-free fish mass (SE of slope = 48.1, mean mass of fish = 1 272g) and (b) fish length for *M. paradoxus* (May 1997)

Batch fecundity

Batch fecundity for both *M. capensis* and *M. paradoxus* was positively correlated with ovary-free fish mass and with total fish length (Figs 3, 4). The mean batch fecundity was $417\,205 \pm 64\,568$ (SE) eggs·female⁻¹ (range = 147 600–723 658) for *M. capensis* and $374\,375 \pm 45\,562$ eggs·female⁻¹ (range = 121 731–710 901) for *M. paradoxus*.

The sample size for the two species was too small to extrapolate the batch fecundity-mass relationship to the entire hake population. Therefore, batch fecundity was expressed at the individual level, relative to female mass, i.e. relative batch fecundity. Mean relative batch fecundity was estimated at 160 ± 12 eggs·g⁻¹ (ovary-free mass) and 143 ± 10 eggs·g⁻¹ (whole fish mass) for *M. capensis* and 306 ± 25 eggs·g⁻¹ (ovary-free mass) and 243 ± 17 eggs·g⁻¹ (whole fish mass) for *M. paradoxus*.

for *M. paradoxus*. Batch fecundity did not differ significantly (Mann-Whitney *U*-test, $p > 0.05$) between the species, but there were significant differences in the relative fecundity ($p < 0.05$). The batch fecundity, ovary-free fish mass and fish length regression relationships did not differ significantly between the two species ($p > 0.05$). There was also no significant relationship between relative fecundity and fish size ($p > 0.05$).

DISCUSSION

The presence of oocytes in different maturity stages in the ovaries of fish (asynchronous ovaries) is usually considered to be evidence for serial spawning (Hunter and Goldberg 1980, Hunter and Macewicz 1985, Melo and Armstrong 1991). Serial spawning has been shown for several species of *Merluccius*, e.g. *M. hubbsi* (Ciechomsky 1967, Christiansen and Cousseau (1971), *M. gayi gayi* (Balbontín and Fischer 1981) and *M. merluccius* (Murua et al. 1996).

Serial spawners usually possess indeterminate fecundity (Hunter et al. 1985), i.e. the annual fecundity (total number of eggs spawned by a female per year) is not fixed prior to the onset of spawning and un-yolked oocytes continue to mature and be spawned during the spawning season (Hunter et al. 1992). Therefore, estimation of the standing stock of advanced oocytes in the ovary is meaningless if, during the spawning season, oocytes are recruited to that stock (Murua et al. 1996). Given that identification of a predetermined annual spawning batch is probably impossible for hake, the only useful measure of their fecundity is the number of eggs produced in a single spawning batch, i.e. batch fecundity (Hunter et al. 1985).

Histological examination showed that *M. capensis* and *M. paradoxus* contained oocytes in different stages of development in all the ovarian classes examined. The presence of post-ovulatory follicles, together with vitellogenic oocytes, was also evident in the hake under study, a feature indicative of serial spawning (Goldberg 1985, Melo and Armstrong 1991). No hiatus in development was observed between the immature, previtellogenic and vitellogenic oocytes in the mature ovaries examined, which is suggestive of indeterminate fecundity (West 1990, Murua et al. 1996).

The positive correlations between batch fecundity and ovary-free fish mass and fish length for *M. capensis* and *M. paradoxus* have been shown for similar relationships in other species of hake, e.g. *M. merluccius* (Murua et al. 1996), *M. productus* (MacGregor 1966), *M. hubbsi* (Grunwaldt 1986), *M. australis* (Balbontín

and Bravo 1993) and *M. gayi gayi* (Balbontín and Fischer 1981). Also, similar relative batch fecundities (ovary-free mass) to those estimated for *M. capensis* and *M. paradoxus* (i.e. 160 and 306 eggs·g⁻¹ respectively) have been reported for *M. merluccius* (165 eggs·g⁻¹, Murua et al. 1996), *M. productus* (192 eggs·g⁻¹, MacGregor 1966) and *M. australis* (334 eggs·g⁻¹, Balbontín and Bravo 1993).

The finding that *M. paradoxus* had a significantly higher mean relative fecundity than *M. capensis* could be explained by the fact that the *M. capensis* examined had already spawned one or more batches of eggs, reducing the number of eggs available for the next spawning, whereas the *M. paradoxus* under study had not shed any eggs. Alternatively, *M. paradoxus* may be a more prolific spawner than *M. capensis*.

In conclusion, both *M. capensis* and *M. paradoxus* are serial spawners. The estimates of batch fecundity for Cape hake are preliminary and more detailed information, using larger sample sizes, is required to quantify their reproductive capacity. Appropriate research on the spawning fraction, the frequency of spawning and duration of the spawning season has been initiated in an attempt to apply the egg production method in addition to trawl data for future spawner biomass estimates of the Cape hakes.

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