REPRODUCTIVE CYCLE IN THE MALE AND FEMALE GREY MULLET, *LIZA KLUNZINGERI* IN THE KUWAITI WATERS OF THE ARABIAN GULF

by

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ABSTRACT. - The reproductive activities of the grey mullet, *Liza klunzingeri* (Day, 1888) in Kuwaiti waters were investigated from March 1998 to February 1999. Contrary to existing view that grey mullet in Kuwaiti waters spawn from December to February, evidence is presented that indicates a prolonged spawning period, beginning from November and ending in March. The maturation pattern is divided into seven stages in both male and female. Variations in GSI relative to fish length indicate that males reach maximum reproductive capacity at between 13.1-17.0 cm, and females at 14.1-18.0 cm. During the fishing campaign, females outnumbered males in the samples in a ratio of 2:1. The oocyte diameter-frequency distribution suggests a synchronous oocyte development with a single, total spawning. Fecundity ranged from 88,896-185,929 eggs. The species inhabits unstable habitats (estuaries and coastal areas) which, in the Kuwait Bay, are threatened. This, coupled with the fact that they generally grow to relatively small size, attain the most fecund status also at a small size, and have high fecundity, suggest that the species is r-selected, as opposed to the so-called K-selected species which inhabit stable environments, grow to relatively large sizes, with delayed reproduction, and low fecundity.

RÉSUMÉ. - Le cycle de reproduction mâle et femelle du mulet gris, *Liza klunzingeri* (Day, 1888) dans les eaux koweitiennes du golfe Arabique.

La reproduction du mulet gris, *Liza klunzingeri* (Day, 1888) a été étudiée dans les eaux du Koweït de mars 1998 à février 1999. Contrairement aux informations actuelles, selon lesquelles la reproduction aurait lieu de décembre à février, il s'avère que la saison de ponte est beaucoup plus longue : de novembre à fin mars. La maturation peut être décrite par 7 stades chez les mâles comme chez les femelles. Les variations du RGS avec la longueur indiquent que les mâles atteignent une capacité reproductrice maximale pour une taille comprise entre 13,1 et 17,0 cm et les femelles une fécondité maximale entre 17 et 18 cm. Durant la campagne de pêche, le sexe ratio observé est 2 femelles/mâle. Les distributions de fréquence du diamètre ovocytaire suggèrent un développement synchrone des ovocytes avec une ponte unique par saison de reproduction. La fécondité est comprise entre 88 896 et 185 929 œufs. Au Koweït, l'espèce colonise de façon instable divers habitats (estuaires et zones côtières) qui sont menacés par les pollutions. Cette observation, couplée au fait que les tailles maximales sont relativement faibles, que la maturité sexuelle est atteinte à une petite taille et que la fécondité est élevée, suggère que la démographie de l'espèce serait de type r sélection plutôt que de type K, pour laquelle les espèces ont une taille relativement élevée, une reproduction tardive et une faible fécondité.

Key words. - Mugilidae - *Liza klunzingeri* - Arabian Gulf - Kuwait - Maturation patterns - Maturity stages - Spawning rhythms - Fecundity.

The family Mugilidae plays an important role in commercial fisheries and aquaculture worldwide. Consequently, a body of information exists especially on various aspects of its reproductive biology and behavioral patterns, including those of Thomson (1957), Sarojini (1957), Anderson (1958), Abraham *et al*. (1966), Yashouv and Berner-Samsonov (1970), Chan and Chua (1980), Edwards *et al*. (1988), Hoda and Qureshi (1989), Abou-Seedo and Al-Khatib (1995) and Abou-Seedo *et al*. (2002). The abundance of grey mullets in estuarine and coastal areas of all tropical and sub-tropical regions of the world may be related to their food and feeding habits, as they occupy a relatively low position in the food web (Wright, 1988).

Liza klunzingeri (Day 1888), formerly known as *L. carinata* (Carpenter *et al*., 1997) is one of the commercially important fish species in Kuwait and the other Gulf countries. Caught usually by beach seines, gill-nets and stake traps (*hadra*), the species is the most abundant of the teleost fish caught in the Doha section of the Kuwait Bay (Abou-Seedo, 1992), and the second most abundant in the Sulaibikhat Bay (Wright *et al*., 1996).

Despite its worldwide commercial importance, only very limited and disparate information exists on any aspect of the biology of the species either locally (Abou-Seedo and Al-Khatib, 1995; Abou-Seedo *et al*., 2002) or regionally (Sorojini, 1957; Abraham *et al*., 1966; Chan and Chua, 1980; Hoda and Qureshi, 1989). It was against this background that the present research was undertaken to provide some of the much-needed data, on aspects of the reproductive biology of the species, for its management and rational

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exploitation in the Kuwaiti waters of the Arabian Gulf. The above aim was achieved by describing the: (1) seasonal fluctuations in the gonadosomatic index, (2) maturation pattern/seasonal distribution of maturity stages, (3) size at sexual maturation and spawning, (4) sex ratio, (5) spawning rhythm and (6) fecundity.

MATERIAL AND METHODS

Fresh samples of *Liza klunzingeri* collected from commercial catches from the Kuwaiti waters of the Arabian Gulf (Fig. 1) served as the material for this research. The sampling lasted 12 months, from March 1998 to February, 1999. The fish were caught using beach seines, gill nets or stake traps, and transferred immediately to the laboratory where total length (TL, cm), standard length (SL, cm) and body weight (g) of each fish were recorded. The fishes were then dissected, the gonads excised and weighed (mg) for the determination of the gonadosomatic index (GSI = weight of gonads/total weight of fish x100). Bouin's fixative was used to preserve the gonads which were then processed histologically. Sections of the ovaries (5-7 μm) and testes (5 μm) were stained in haematoxylin and counterstained in eosin to observe the histological changes in the gonads to confirm the maturity stages.

The maturity stages in the females were determined according to Abou-Seedo and Al-Khatib (1995) with minor modifications as summarized hereunder: Females - Stage I (Immature): Fish with ovaries containing early germ cells; sex differentiation of gonial cells difficult. Stage II (Immature and recovering): Ovary contains numerous mitotically dividing oogonia and few early, perinucleolar, pre-vitellogenic oocytes. Stage III (Developing): Ovary contains mostly early vitellogenic oocytes. Stage IV (Maturing): Presence of numerous advanced vitellogenic oocytes in ovary. Stage V (Mature): Ovary reveals numerous advanced oocytes which have completed vitellogenesis and nuclear polarization, a few of them exhibiting ovulation. Stage VI (Spawning). The majority of the ovarian cells are hyaline oocytes, some of them extruding to the exterior with a slight application of pressure on the belly. Stage VII (Spent): Ovary contains numerous ruptured follicles at different phases of resorption, some unexpelled oocytes undergoing atresia, as well as resting phase oogonia and perinucleolar oocytes.

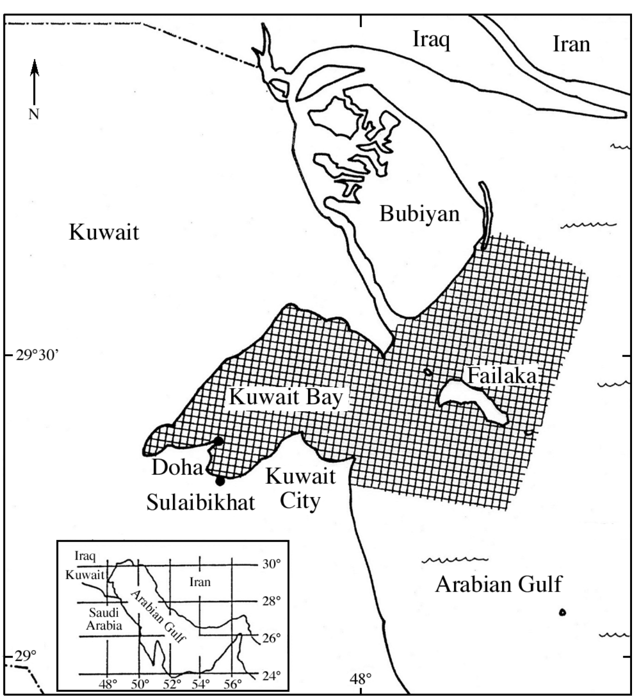

Figure 1. - Map of Kuwaiti waters of the Arabian Gulf, showing the fishing areas (hatched).

The maturity stages in the males were determined according to Al-Khatib (1986) with minor modifications: Stage I (Immature): Fish with testis containing early germ cells, and, as in the females, sex differentiation of gonial cells difficult. Stage II (Immature and recovering): Testis contains spermatogonia of different generations and cysts of spermatocytes. Stage III (Developing): Testis contains spermatogonia, numerous cysts of spermatocytes and spermatids. Stage IV (Maturing): Spermatids in cysts and in lobules dominate the testis, with some of the latter transforming into spermatozoa. The spermiogenetic process begins with transformation of spermatids first into sperm parachutes, with the heads pointing in one direction, while the tails adhere together. Stage V (Mature): Testis contains numerous spermatozoa released from the parachutes into the central parts of the seminiferous tubules. Stage VI (Spawning/running): The seminiferous tubules and main sperm duct of the testis are densely packed with ripe sperm, some of them oozing out. Stage VII (Spent): Empty tubules are observed in the testis, a number of them spotting residual sperm which become pycnotic and undergo degeneration. Many germ cells at different stages of spermatogenesis are also present.

The frequencies of the various maturity stages and the monthly variations in the GSI were used to study the maturation pattern and the extent of the breeding season. The GSI fluctuations in different size groups were also used to determine the size at maturation and spawning. The ratio of males to females in monthly samples was determined and the results tested statistically (χ^2 test of homogeneity).

For fecundity studies, ripe ovaries were fixed in Gilson's fluid and fecundity, defined as the number of ripe ova just prior to spawning (Bagenal and Braun, 1978), was estimated for 22 females by the gravimetric method (Kipling and Frost, 1969). Oocyte diameter-frequency distributions of 22 females sampled between December and February, the spawning period of *L. klunzingeri* in Kuwait waters (Abou-

Seedo and Al-Khatib, 1995), was determined by random selection of five sub-samples of oocytes from each ovary and measuring 100 oocytes from each sub-sample under the microscope at x40 magnification. The percentage frequencies of oocyte diameters in different size classes were then determined and used in plotting histograms depicting the rhythm of egg-laying.

RESULTS

Seasonal fluctuations in the gonadosomatic index (GSI)

Monthly changes in the GSI revealed an increase in reproductive activity detectable in the females in September, and in October in the males (Fig. 2). A further increase was observed, culminating in a peak in November, in both sexes, after which the GSI declined gradually until March/April. These results corroborate those reported below on the maturation patterns.

Maturation pattern/seasonal distribution of maturity stages

After a long, immature (Stage I)/recovering (Stage II) period lasting March-September in both males and females, the gametogenetic cycle in *L. klunzingeri* begins in October in both sexes as the gonads of the majority of the samples enter the developing stage (Stage III), while some males are found in the maturing stage (Stage IV) (Fig. 3). A resurgence in gametogenetic activity is observed as the ovaries and testes are filled with maturing (Stage IV) and mature (Stage V) germ cells, resulting in spawning from November when the first gonads in the spawning stage (Stage VI) were observed. Spawning lasted till March as only immature and mature/spawning stages were observed in this month, followed by the disappearance of spawning fish and the presence of only spent females (Stage VII) in the April 98 samples.

Size at sexual maturation and spawning

Developing males and females (Stage III) and above were considered mature for the determination of the minimum size at sexual maturation. As a mean, males mature at smaller sizes than females. The smallest males in the sampled population belonged to the 13.1-14.0 cm length class and 69.2% of them were already mature (Tab. I). Although mature and immature females were found in all length classes from 13 cm, only a minority of the mature females were less than 15 cm in length, and most maturing or spawning females measured 15 to 18 cm (Tab. II). The size of maximum reproductive capacity, *i.e.*, the mean size at which spawning (Stage VI) is observed, was recorded at a fish length of 13.1-17.0 cm with a mean of 15.1 ± 1.3 cm in males and 14.1-18.0 cm with a mean of 15.6 ± 1.6 cm in females.

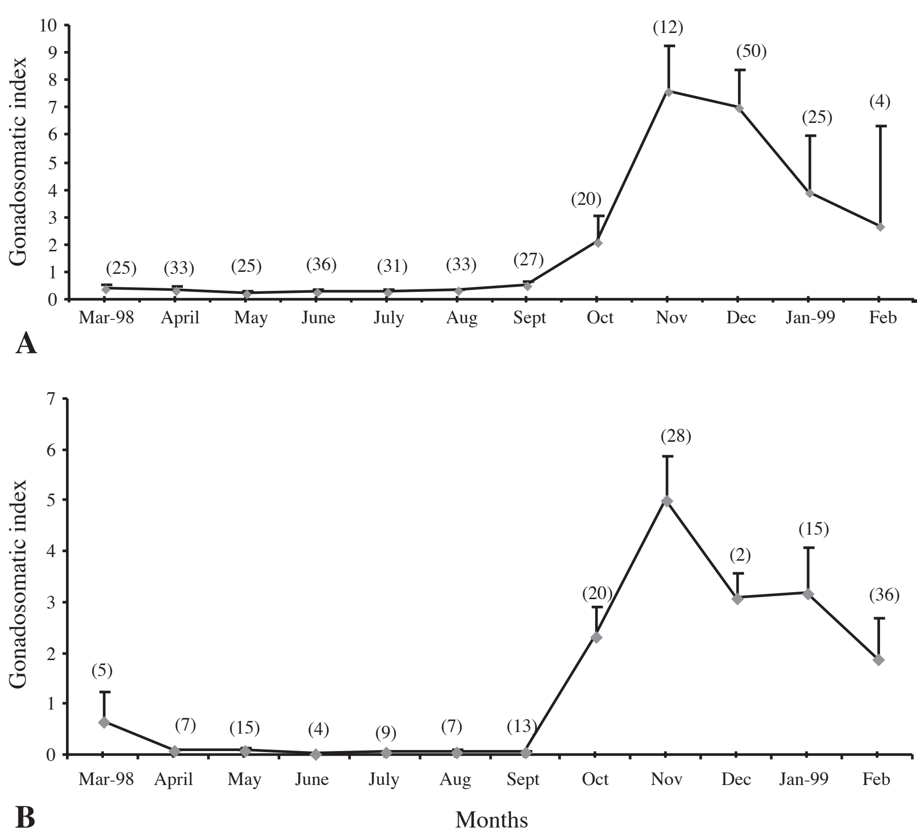

Figure 2. - Seasonal fluctuations in gonadosomatic indices in *Liza klunzingeri*. **A**: Females; **B**: Males. Vertical bars indicate standard deviations. Figures in parentheses indicate sample size.

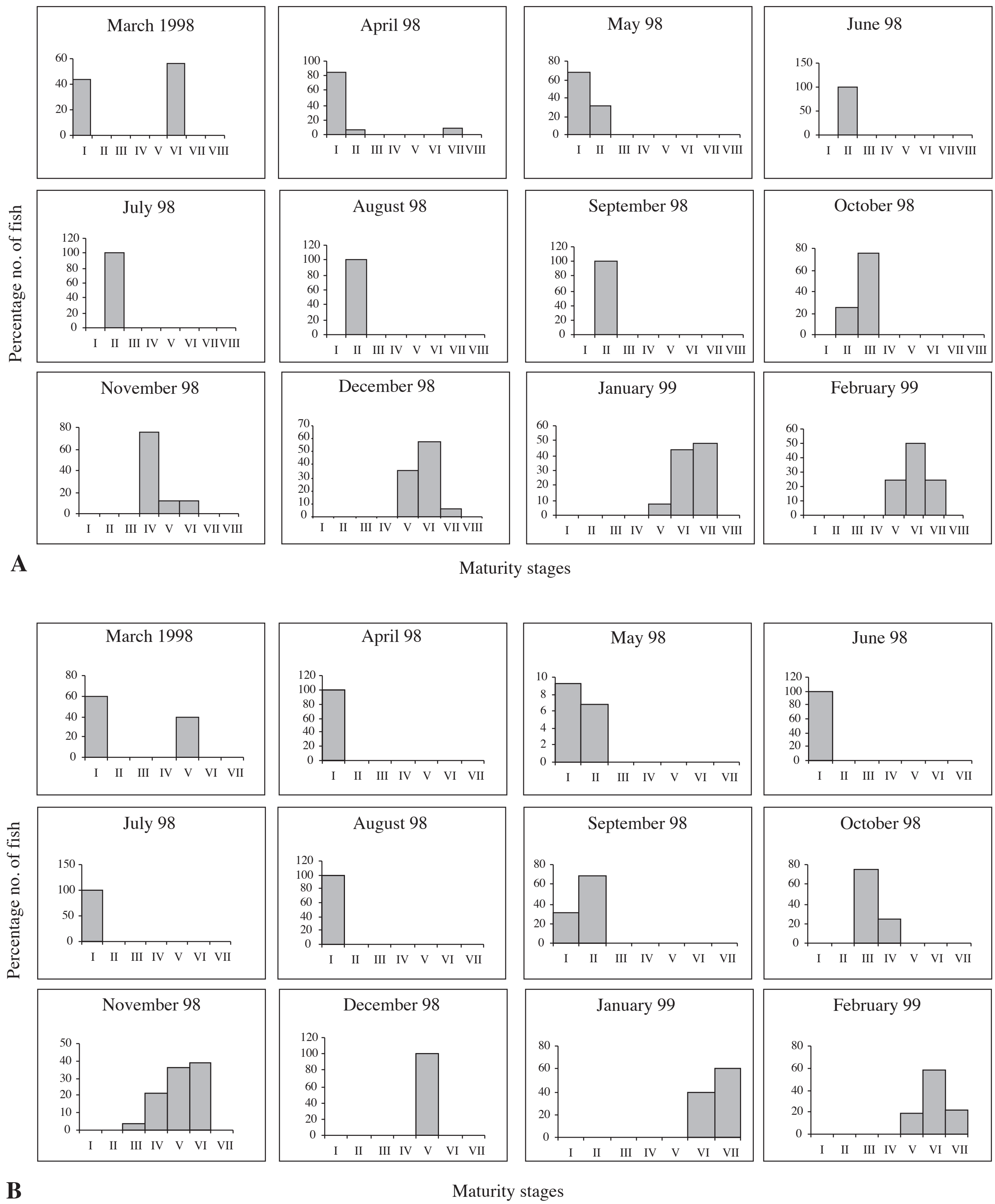

Figure 3. - Seasonal distribution of maturity stages in *Liza klunzingeri*. **A**: Females; **B:** Males.

Sex ratio

Out of 482 specimens sampled during the study, 321 were females and 161 were males, giving an overall sex ratio of 2: 1 which significantly deviated from the hypothetical distribution of 1:1 (χ^2 = 134; d.f. = 11; p < 0.001). A preponderance of females over the males was observed throughout the year except in October when the ratio was 1:1, and in November and the following February, when the males dominated the samples (Fig. 4).

Spawning rhythm

Histological observations made on the ovaries of *L. klunzingeri* during the mature/spawning period revealed the predominance of advanced mature oocytes of successive sizes in the ovaries. Oocyte diameter-frequency distribution revealed a unimodal pattern during the spawning period, from December till February (Fig. 5).

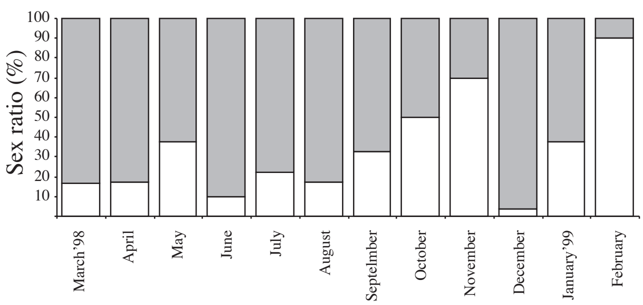

Figure 4. - Monthly sex ratio in captured *Liza klunzingeri*. Shaded: females; Unshaded: males.

Fecundity

The lowest fecundity estimated in *L. klunzingeri* in the Kuwaiti Bay was 88,896 eggs in a female measuring 15.9 cm SL and weighing 77.9 g, although the smallest female sampled (15 cm SL) had 125,955 eggs in the ovary, while the highest fecundity of 185,929 eggs was recorded in a 15.8 cm SL fish with a weight of 76.7 g. The mean fecundity from 22 females was 127,350 eggs.

DISCUSSION

Information on the spawning periodicity of *L. klunzingeri* is scarce and disparate both locally and regionally. In the only previous study conducted in 1984–85 in the Kuwaiti waters, Abou-Seedo and Al-Khatib (1995) reported a reproductive cycle beginning from May, after a long quiescent period, culminating in spawning from December to February. In the present study, recrudescence of gametogenesis was observed in October, and spawning started with a peak a month earlier than reported by Abou-Seedo and Al-Khatib (1995), in November, and ended also a month later, in March in both males and females. This prolonged spawning season is based on the presence of large numbers of males and females in Stages V-VII (Mature-spent) of maturity in the November-March samples, coupled with the very high GSI values recorded during this period. From this account, it may be concluded that nearly 24 years after the first study, a change has occurred both in the onset, and duration, of spawning in *L. klunzingeri* in Kuwaiti waters. In the Karachi-Sind waters of Pakistan, Hoda and Qureshi (1989)

Length-class (cm)	No. of fish	Number of fish in different maturity stages %							% mature fish
		I	II	III	IV	V	VI	VII	
13.1-14.0	26	26.9	3.9	3.9	0	7.7	46.2	11.4	69.2
14.1-15.0	82	22.0	9.7	14.6	11.0	13.4	22.0	7.3	68.3
15.1-16.0	49	38.8	6.1	6.1	4.2	16.3	12.2	16.3	55.1
16.1-17.0	4	25.0	0	0	0	0	25	50	75
17.1-18.0	0	0	0	0	0	0	0	0	0
18.1-19.0	0	0	0	0	0	0	0	0	0
19.1-20.0	0	0	0	0	0	0	0	0	0

Table I. - Distribution of males *Liza klunzingeri* at different size classes into maturity stages (March 1998 to February 1999).

Length-class (cm)	No. of fish	Number of fish in different maturity stages %							% mature fish
		I	II	III	IV	V	VI	VII	
13.1-14.0	13	15.4	76.9	0	0	0	0	7.7	7.7
14.1-15.0	104	19.2	48.1	8.7	1.9	6.7	10.6	4.8	32.0
15.1-16.0	137	17.5	34.3	4.4	2.2	8.0	16.1	17.5	48.2
16.1-17.0	55	18.2	36.4	1.8	5.5	7.3	20.0	10.8	45.4
17.1-18.0	9	0	55.6	11.1	0	22.2	11.1	0	44.4
18.1-19.0	2	0	100.0	0	0	0	0	0	0
19.1-20.0	1	0	100.0	0	0	0	0	0	0

Table II. - Distribution of females *Liza klunzingeri* at different size classes into maturity stages (March 1998 to February 1999).

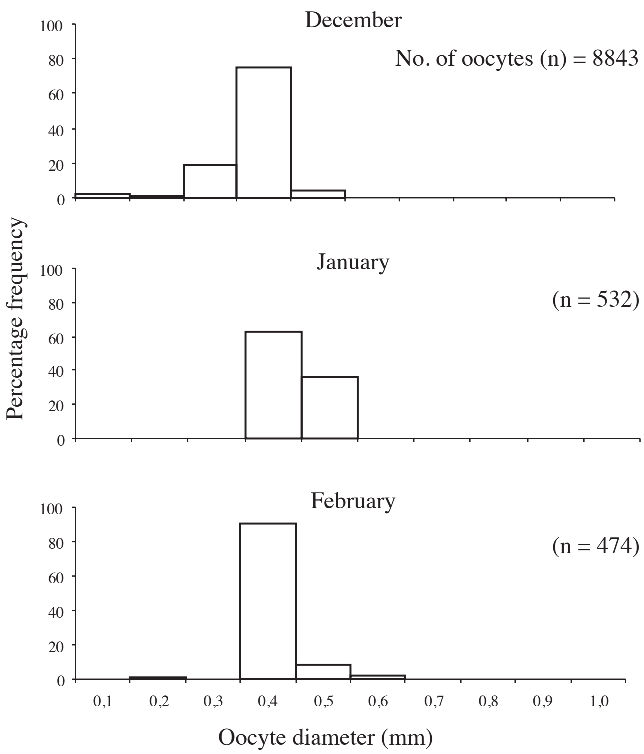

Figure 5. - Frequency distribution of oocyte diameter in *Liza klunzingeri*.

observed even a longer spawning period in the same species, from October to March.

The above conclusion has serious implications for the management of the mullet stocks in the Kuwaiti waters. Applying a fishing ban one month late, in December, especially after the month of peak spawning, and lifting the ban one month before the end of spawning can deplete spawning biomass, leading to recruitment overfishing. A similar change in the onset of spawning in *Pampus argenteus* in Kuwaiti waters has recently been observed (Dadzie *et al*., 1998), some twenty years after the last investigation, and the management protocols have been adjusted accordingly. This brings into sharp focus, the need to re-examine periodically, the reproductive patterns of fish, especially those of commercial importance, with the view to making the necessary management adjustments where necessary.

The present study has also revealed that *L. klunzingeri* in the Kuwaiti waters of the Arabian Gulf, spends six months (April-September) with gonads in the quiescent state, despite the prevalence of optimum water temperatures during the greater part of the year (Abou-Seedo *et al*., 2003), and spawns during the winter months (14-18°C). The other species spawning at equally low temperatures in the Kuwaiti waters is *Acanthopagrus latus* (Abou-Seedo *et al*., 2003). Apart from these, all the other local species studied take advantage of the increasing water temperatures after the winter spell for their gametokinetic process and begin to spawn with the warming of the water from April (Hussain and Abdullah, 1977; Abu-Hakima *et al*., 1983a, 1983b; Abu-Hakima, 1987; Dadzie *et al*., 1998, 2000a, 2000b; Al-Kandary, 2002). This timing in reproduction is common among many fishes of the same latitudinal distribution but inhabiting geographically different water bodies (Johnson *et al*., 1998; Taylor *et al*., 1998; Yoneda *et al*., 1998).

Following spawning in winter, very young *L. klunzingeri* form shoals at the edge of the rising tide both day and night in clear water during spring in the Kuwait Bay, while fish two months older form shoals only on daytime rising tides (Abou-Seedo *et al*., 1990). Spawning in the winter coincides with the lowest green algae (*Ulva* and *Entermorpha*) productivity, while growth and development in the spring take place with rising water temperatures, of up to 31°C, and increasing algal productivity (Wright *et al*.,1996).

From the present study, it is also evident that females *L. klunzingeri* reach sexual maturity at a larger size than the males, similar to the report on the same species from Pakistan (Hoda and Qureshi, 1989). This could ensure that the fish have more accommodative capacity for increased egg production as suggested by Owiti and Dadzie (1989). Since *L. klunzingeri* commonly grows to 15.0 cm, with a maximum size of 20 cm (Carpenter *et al*., 1997), spawning in females at 14.1-18.0 cm is too close to the maximum size-expectancy, unless there is an advantage to be gained. The advantage here is the possibility of increasing the number of eggs produced in an unstable coastal and estuarine habitats of *L. klunzingeri*, which will tend to make the species r-selected (MacArthur and Wilson, 1967). It is implied, as it is also evident from the present study, that males mature at smaller sizes than females. This could suggest either that at some stage the males grow more slowly than the females, or that the males mature earlier than the females. Judging from our histological analysis, the testis of a large number of small males contained ripe spermatozoa, as compared to the ovaries of females of comparable size which did not contain mature oocytes. This suggests that the males mature earlier than females.

Females outnumbered the males by 2:1, which was a significant departure from the hypothetical 1:1. This could be due to the differential fishing factors related to seasons and schooling in feeding and spawning grounds (Sarojini, 1957; Silva and De Silva, 1981, 1982; Hoda and Qureshi, 1989). It could also result from selective fishing for the large fish preferred by the fishermen.

The predominance of mature oocytes of successive sizes in the ovaries of *L. klunzingeri* during the spawning period, coupled with the unimodal pattern of oocyte diameter-frequency distribution suggest that the species has a synchronous oocyte development with a single spawning, probably of short duration. This corroborates the report of Chan and Chua (1980) that spawning in a related species, *L. subviridis*, is restricted to a short and definite period, with all ripe ova being released within a single spawning act, and contradicts that of Hoda and Qureshi (1989) who suggested a prolonged period of spawning of individual fish. These reports are in contrast to the group-synchronous oocyte development and multiple spawning observed in many other teleosts (Wallace and Selman, 1981; Tucker and Campbell, 1988; Taylor *et al*., 1998). The phenomenon of synchronous oocyte development with a single spawning may also be related to the unstable nature of the habitat of the species, and hence the need to spend as little time as possible in the spawning grounds – a further confirmation that the species is r-selected.

Although the mean fecundity of 127,350 eggs for *L. klunzingeri* estimated in the present study may seem low as compared with that of other teleosts, it must be borne in mind that the species, as already established above, is naturally of small size. The fecundity data were based on mature females ranging in size from 15.0-17.5 cm which is very close to the maximum size to which the species generally grows (Carpenter *et al*., 1997). When compared with other teleost species which grow to much larger size, it is realized that the fecundity of *L. klunzingeri* in Kuwaiti waters is quite high. For example, Abu-Hakima *et al*. (1983b) reported the lowest fecundity of 53,676 eggs for *Otolithes argenteus* of 26.0 cm SL in the same area. This is much lower than the fecundity of 125,955 eggs recorded in the smallest *L. klunzingeri* (15.0 cm SL) encountered in this study. The high fecundity in relatively small-size *L. klunzingeri*, could be due, as mentioned earlier, to the ability of the species to mature at a larger size, thus having a more accommodative capacity for increased egg production, leading to high yield – an adaptation to ensure the conservation of inheritance in an otherwise unstable environment. It must be stated, though, that at the inter-specific level, a number of grey mullet species grow to much larger sizes, with correspondingly much higher fecundities (Kesteven, 1942; Jacob and Krishnamurthy, 1948).

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