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RESEARCH ARTICLE
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Individual rearing of common octopus (*Octopus vulgaris* Cuvier, 1797) in tanks: Preliminary results

Ahtapot (*Octopus vulgaris* Cuvier, 1797)'un tanklarda bireysel **yetiştiriciliği: İlk** sonuçlar

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Abstract: The effects of individual rearing technique on the growth and survival of *Octopus vulgaris* (Cuvier, 1797) were investigated. Therefore, wild octopuses were reared in the transparent, perforated and caped polyethylene terephthalate (PET) pots (10 l). The assay was established as one free group rearing as a control (12.7 kg/m^3 of stoking density) and two individual rearing groups as I1 and I2 (13.8 kg/m^3 and 18.3 kg/m^3 of stoking density, respectively). The individuals were fed mainly fresh or thawed fish at 5% body weight per day. The experiments lasted for 120 days. The acclimatization period of the octopuses lasted within 3-5 days in the individual rearing and 7-10 days in the free group rearing. The final survival rates were 100% for the control and I1 or 85.7% for I2 (P>0.05). Statistically differences were not found among the growth or specific growth rates (between 0.5 and 0.6%/day for the trials) (P>0.05). The maximum density (30.7 kg/m^3) was obtained from the I2 trial.

Keywords: Octopus vulgaris, individual rearing, growth, survival

Öz: Bireysel yetiştiriciliğin *Octopus vulgaris* (Cuvier, 1797)'in gelişimi ve yaşama oranı üzerine etkileri araştırıldı. Bunun için doğadan yakalanan ahtapotlar, 10 litre hacmindeki şeffaf, delikli ve kapaklı polietilen tereflat (PET) kaplarda yetiştirildi. Denemeler, bir serbest grup yetiştiricilik (12,7 kg/m^3 stok yoğunluğunda, kontrol grubu) ve iki bireysel yetiştiricilik (I1, ve I2 olarak sırasıyla, 13,8 kg/m^3 ve I2, 18,3 kg/m^3 stok yoğunluğunda) grubu olarak kuruldu. Ahtapotlar, ağırlıklı olarak taze ve/veya taze donmuş çözülmüş balık ile günlük olarak vücut ağırlıklarının %5'i oranında beslendi. Denemeler 120 gün sürdü. Adaptasyon periyodu bireysel yetiştiricilik gruplarında 3-5 günde, serbest grup yetiştiricilik grubunda 7-10 günde tamamlandı. Hayatta kalma oranları kontrol ve I1 için %100, I2 için %85,7 olarak hesaplandı (P>0,05). Ahtapotların gelişimleri veya spesifik gelişim oranları (denemeler için 0,5 ve 0,6%/gün) arasında istatistiksel olarak önemli farklılıklar saptanmadı (P>0,05). Maksimum stok yoğunluğu (30,7 kg/m^3) stoklamanın en fazla olduğu I2 denemsinden elde edildi.

Anahtar kelimeler: Octopus vulgaris, bireysel yetiştiricilik, gelişim, yaşama oranı

INTRODUCTION

Octopus vulgaris (Cuvier, 1797), has become a potential species for the aquaculture industry, due to the high growth rate, high nutritional value, an easy adaptation to controlled conditions and high market demand (Vaz-Pires et al., 2004; Iglesias et al., 2004; Iglesias et al., 2007; García García et al., 2009). However, *O. vulgaris* culture has cannot be performed as commercially yet, due to the low survival rate of the paralarvae and lack of specific live and/or compound diets for its paralarvae and subadults (Vaz-Pires et al., 2004; Iglesias et al., 2007; Valverde et al., 2015). Despite these constraints, fattening of *O. vulgaris*, using floating cages in Spain (Galicia, NW) was achieved in 1998 and 1999 (FAO, 2002).

In recent years, most studies on the improvement of growth in *O. vulgaris* is concentrating on the development of specific enrichments and compound or formulated diets (Valverde et al., 2013; Valverde et al., 2015; Morillo Velarde et al., 2015; Querol et al., 2015a; Querol et al., 2015b). On the other hand, a few studies have been performed in order to test new rearing systems and/or techniques for *O. vulgaris* ongrowing (Chapela et al., 2006; Rodríguez et al., 2006; Estefanell et al., 2012a; Estefanell et al., 2012b).

Further, in the previous studies, it's clearly pointed out that better growth results in the common octopus have been obtained by free group rearing technique, but higher survival rates have been achieved by individual rearing (Chapela et al., 2006; García García and Valverde, 2006; Petza et al., 2006; Rodriguez et al., 2006; Valverde et al., 2008; Biandolino et al., 2010; Prato et al., 2010; García Garrido et al., 2011; Estefanell et al., 2012a). Therefore, the individual rearing seems to be a highly considerable method for *O. vulgaris* on growing when consideration of the handicaps of the free group rearing technique for *O. vulgaris* that mentioned above. The purpose of the current study was to test the growth and survival of *O. vulgaris* by individual rearing technique, and also whether it can solve the main problems in the free group rearing method.

MATERIALS AND METHODS

Wild live octopuses were obtained from local fishermen using by a jigging line, of Urla Port (Izmir Bay, Aegean Sea).

The octopuses were transferred with no mortality to the laboratory immediately after collection. They kept together till coming into the laboratory in a 40 l tank with the water being renewed every 20 minutes; the temperature was 16-18°C and oxygen above 6.5 mg/l. The specimens were weighed one by one after the sex determination. Then, the octopuses were randomly distributed according to the rearing technique (individual or free group). The adaptation period of the octopuses was performed in circular polyester 430 l tanks and open flow-through filtered seawater system (860 l/h). In the free group rearing trial, polyvinyl chloride (PVC; 110 mm in diameter and 350 mm in length) tubes as shelters, and a net cover to prevent octopuses from escaping were provided. In the individual rearing groups, transparent, perforated and with caps polyethylene terephthalate (PET) pots (10 l) were used (Figure 1). During the period, the octopuses were fed as ad libitum once a day at 09:00 hour, with a mixed diet including fresh or thawed bogue (*Boops boops*), annular seabream (*Diplodus annularis*), picarel (*Spicara smaris*) and mantis shrimp (*Squilla mantis*), provided on consecutive days.

Totally 17 octopuses (1132.9 ± 218.2 g) were weighed following controlled of their sexes and were stocked at different initial densities (Table 1). The experiment was established as one free group rearing as a control (12.7 kg/m^3 of stoking density) and two individual rearing groups as I1 and I2 (13.8 kg/m^3 and 18.3 kg/m^3 of stoking density, respectively). No significant differences were found in the weights of the individuals (P>0.05). The rearing experiments were carried out in the same tank conditions as mentioned above. In the control group, a net cover and the PVC tubes as the same number of the octopuses were used. In the individual rearing groups, the octopuses were placed one by one into the PET pots (Figure 1).

Table 1. Beginning data of the experiment (mean ± SD).

	C	I1	I2
N_i	5	5	7
Female:Male	3:2	1:4	5:2
Average W_i (g)	1093 ± 243^a	1187 ± 313^a	1122 ± 135^a
Min W_i (g)	784	809	937
Max W_i (g)	1447	1450	1324
D_i (kg/m^3)	12.7	13.8	18.3

N_i, initial number; D_i, initial density; Different superscript letters show the significant

During the trials, the specimens were fed a mixed diet as fresh and/or thawed fish (93%; 48% of *D. annularis*, 33% of *S. smaris*, 12% of *B. boops*) and mantis shrimp (*S. mantis* as 7%) once a day as 5% body weight per day (Domingues et al., 2010). The next morning, remaining foods were collected from the tanks by hand or net, and were weighed before feeding. This was used to determine food ingestion of the octopuses. The food ratio was arranged after each weighting period. The specimens were weighed individually every 15 days during the assay.

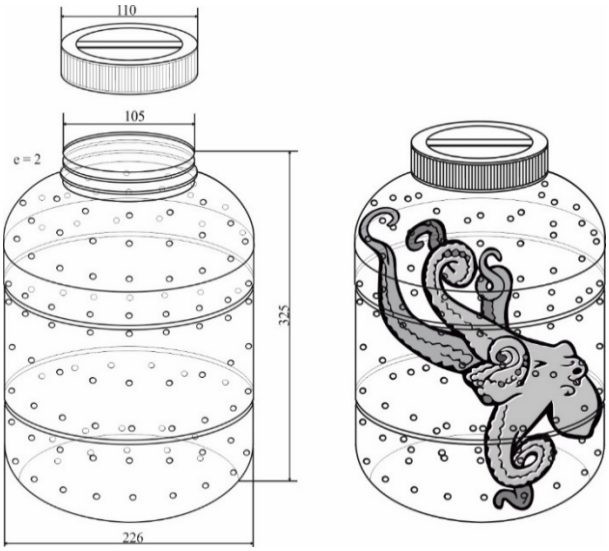

Figure 1. The polyethylene terephthalate (PET) pot (10 L) (dimensions in mm)

The experiments were finished in the 120th days of the trial since at this time mating behavior began among the octopuses in the control group.

Through the experimental period, each experimental tank illuminated by a 40W daylight fluorescent lamp. Photoperiodicity was natural (38˚21'K, 26°46'D; December to April). The water parameters were measured by YSI EcoSense DO 200A for oxygen and temperature, YSI EcoSense pH 100A for pH, and YSI EcoSense EC300A for salinity.

For every sampling period, the weight data were used to calculate: absolute growth rate (AGR)= (Wf - Wi) / t; specific growth rate (SGR) = (LnWf - LnWi) × 100 / t, where Wf and Wi are the final and initial weights of the octopuses, respectively, Ln the natural logarithm, and t the number of days of the experimental period; feeding rate (FR)= [IF / Average W(t)] × 100, where IF is the ingested food and Average W(t) is the average weight of the octopus during that period; food conversion (FC) = (Wf - Wi) / IF, where Wf - Wi is the weight gained by the octopus during that period; octopus density (D) per tank = total biomass of the octopuses (kg) × (1000 / 430); where 430 was available water volume (l) of the tank. Survival (%) was calculated by counting for each tank for each sampling period.

Statistical analyses of the data (means ± SD) were performed with SPSS 25.0 statistical software in 95% confidence interval (P < 0.05); following the ANOVA to test differences in weight, growth rate, feeding rates and feed

conversion rates, the significance of differences was tested using a Tukey multiple comparison test. The normal distribution analysis after the square root transformation of the data was carried out with the Kolmogorov-Smirnov test. Homogeneity of variances was tested with Levene's test (Zar, 1999). Differences in survival rates between were tested with the chi-square (χ^2) test.

RESULTS

The acclimatization periods of the octopuses in the individual and free group rearing completed within 3-5 days and 7-10 days, respectively. In addition, mortality related to cannibalism was not detected. During the period, the water temperature, salinity, oxygen, and pH were 15.2-17.3°C, 37.7-38.6‰, 8.5-9.6 mg/l and 8.1-8.2, respectively. During the assays, the experimental water parameters were given in Table 2.

Table 2. Experimental water parameters (Mean ± SD)

	C	I1	I2
Temperature (°C)	13.4 ± 1.3 a.	13.3 ± 1.2 a.	13.9 ± 0.9 a.
Salinity (‰)	39.4 ± 0.8 a.	39.4 ± 0.9 a.	39.4 ± 0.8 a.
Oxygen (mg/l)	9.4 ± 0.7 a.	9.4 ± 0.6 a.	9.2 ± 0.7 a.
pH	8.0 ± 0.07 a.	8.0 ± 0.07 a.	8.0 ± 0.07 a.

Different superscript letters show the significant differences (P < 0.05)

The final survival of 85.7 for I2 and 100% for I1 and control groups were recorded (P > 0.05). The mortality occurred in I2 with one octopus on the 90th day of the experiment.

The average weight gains of the octopuses were shown in Figures 2 and 3, respectively. The absolute growth rates, the specific growth rates, the feeding rates, and the food conversion rates were not significantly affected by the method of rearing (P> 0.05). The experimental results were summarized in Table 3.

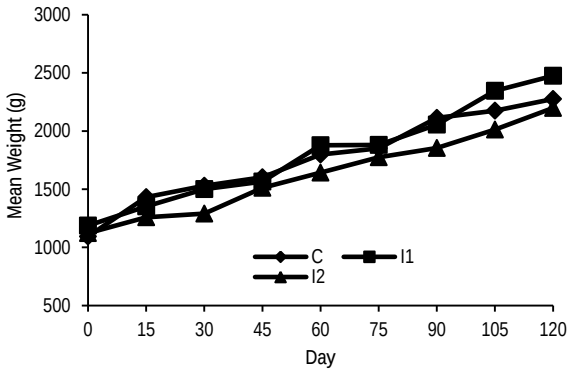

Figure 2. The mean weights (g) of *O. vulgaris*

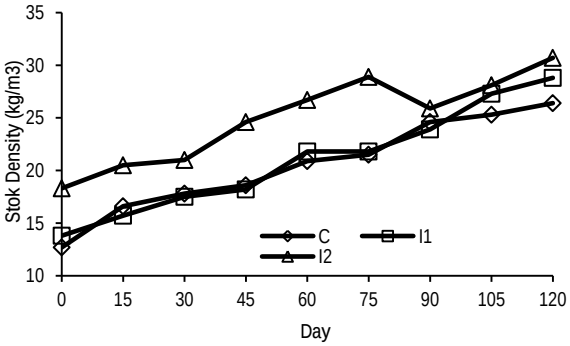

Figure 3. The stock densities of *O. vulgaris*

Table 3. The experimental results (mean ± SD).

	C	I1	I2
Average W_f (g)	2275 ± 235[a]	2475 ± 705[a]	2197 ± 370[a]
Min W_f (g)	1980	1749	1894
Max W_f (g)	2564	3491	2853
D_f (kg/m^3)	26.5[a]	28.8[a]	30.7[a]
S (%)	100[a]	100[a]	85.7[a]
AGR (g/day)	63.6 ± 21.2[a]	51.3 ± 5.3[a]	50.8 ± 11.7[a]
SGR (%/day)	0.6 ± 0.5[a]	0.6 ± 0.4[a]	0.5 ± 0.3[a]
FR (%/day)	21.9 ± 8.3[a]	20.8 ± 9.8[a]	20.1 ± 4.7[a]
FC (%)	41 ± 26[a]	39 ± 22[a]	50 ± 42[a]

Different superscript letters show the significant differences (P < 0.05); D_f, final density

In the control group, except cannibalism, food competition, aggressiveness, injuries, escape and mating behaviors were recorded. In the individual groups, these behaviors were not detected.

DISCUSSION

In this study, the individual rearing method for *Octopus vulgaris* ongrowing was applied for the first time. The present results showed that the individual rearing technique can be used for the octopus on growing due to the higher growth and survival rates.

It's pointed out that in the prior studies, the acclimatization period completes within 7-15 days for *O. vulgaris* in free group rearing method (Cagnetta, 2000; Iglesias et al., 2000a; García García and Valverde, 2006; Domingues et al., 2010; Delgado et al., 2011; Estefanell et al., 2012a; Estefanell, et al., 2012b). However, Şen (2012) notified that this period lasted 3-5 days for *O. vulgaris* by individual rearing technique. These results were found parallel to the present findings for both techniques. According to the current results, the individual rearing technique reduced the adaptation period of the octopus by almost one of the third when comparing to that of the free group rearing method.

The water parameters during the experimental period were optimal for the development of the octopuses, but especially the salinity value (41‰) was relatively higher than the previous data (García García et al., 2009; Delgado et al., 2011; Boletzky and Hanlon, 1983; Aguado Gimenez and García García, 2002). However, it's thought that the salinity data did not affect to the results.

For the free group rearing method, it's suggested that initial stocking density should be up 10 kg/m^3 for *O. vulgaris* ongrowing (Iglesias et al., 2000a; García García et al., 2004; Rodriguez et al., 2006). However, the present survival rates showed that culture density up to 18 kg/m^3 could be used for on growing of the octopus by the individual technique. On the other hand, reason of the mortality in I2 could not have been understood, clearly.

Common octopus reaches 2.5-3 kg in 3-5 months in the optimal conditions (García García et al., 2009). However, the rearing period should not exceed 3.5 months in order to commercial farming of the octopus, otherwise, the rearing practice would not be economically successful (García García et al., 2004; García García et al., 2009). In the present study, according to the current growth results, the octopuses reached almost to the commercial market size within 120 days (4 months). The major factor related to the delay in the growth of the specimens was the water temperature, which was under the suggested values (18-21°C) (García García et al., 2009). Likewise, the feeding with mainly fish might have been caused by the delay of the growth. Because, Aguado and García García (2002) reported that a diet mainly of fish, leads to comparatively poor growth.

The current growth rates (below to 1%/day) were nearly close to the ones reported for *O. vulgaris* at lower temperatures (Otero et al., 1999; Iglesias et al., 2000a; Chapela et al., 2006). Also, these data were lower than those reported for the ones rearing at a higher temperature (García García and Giménez, 2002). Temperature is the major factor that effects to cephalopod growth, food conversions and ingestion (Domingues et al., 2010).

Reported feeding rates for *O. vulgaris* range 2-8% in the previous studies (Sanchez et al., 1998; Iglesias et al., 2000a,b; García García and Valverde, 2006; Biandolino et al., 2010; Domingues et al., 2010; Estefanell et al., 2012a). However, given the average feeding rates, the current values were below the reported rates. Further, it's thought that the low FRs in this study might stem from the low water temperature and/or mainly fish diet (Aguado Giménez and García García, 2002).

Food conversions between 30 and 60% are reported for *O. vulgaris* (Aguado Giménez and García García, 2002; Vaz-Pires et al., 2004). In the present study, the food conversions got from the trials stayed within this range. Besides, food conversions in the trials indicated that the rearing methods did not affect normal growth, and the conditions were convenient for the on-growing of octopuses. More, since feeding rates in the assays were similar for the three groups, the octopuses in I2 did not consume more energy in order to metabolic purposes and biomass increment. This might indicate that the initial density of the octopuses could be raised up to 18 kg/m^3 by the individual rearing technique.

In the free group rearing trial, it was observed that the female and male octopuses kept with together demonstrates mating behavior ending with spawning. These events were also emphasized in previously (Otero et al., 1999; Iglesias et al., 2000a,b; Vaz-Pires et al., 2004; García García and Valverde, 2006; Domingues et al.,2010). Although there were no contact possibilities between the octopuses in the individual rearing method, only the one female octopus laid eggs in I2 group, interestingly. It is thought to be that the female octopus was mating in its natural environment before being caught, probably. Because, in *O. vulgaris*, 580 g females and 250 g males can reach sexual maturity and reproduce (Silva et al., 2002).

In conclusion, using the individual rearing method is a common application for high priced and highly cannibalistic farmed species (Nicholson et al., 2008; Perez et al., 2010). So, it's thought that the present results supported the authors' findings. Because it's obvious that the individual rearing technique for *O. vulgaris* on growing can be used due to the positive effects on the survival and growth of the octopuses. Furthermore, according to the present results, the individual rearing method presented important advantages, when compared to the free group rearing; no mortality related to cannibalism, no shelter, no aggressiveness, no food competition, a high survival rate, high growth rate, high initial stocking density, co-rearing of female and male octopuses, co-rearing of large and small octopuses, a short adaptation period, and any marketing size harvesting possibility. As a result, the preliminary findings showed that the individual rearing technique could solve the main problems in the free group. Finally, the preliminary results showed that for *O. vulgaris* on growing, the individual rearing technique could solve the main problems in the free group. However, there are needed more detailed studies should be carried out to the improvement of this technique.

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