DEDICATION

To my family

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Abstract

The growth rate and nutritional quality of the S-type rotifers *Brachionus rotundiformis* under four different salinity levels fed three different food types were tested and these resulted in differences in maximum density (production No. ml^{-1}), increment (%) and specific growth rate (SGR % day^{-1}) after 84 hours trial culture period. The maximum production rates were higher in the cultures fed on baker's yeast in all experimental groups. The production rates of rotifers (mean No±sd ml^{-1}) fed baker's yeast at 20, 30, 40, 50 ppt were (±SD)

320.7±3.80, 219.3±19.5, 211.3±3.2 and 204.0±5.3 individuals ml^{-1}, respectively. The lowest production rates were observed in the cultures fed instant algae® *Isochrysis sp*. under all salinity levels and the mean production rates recorded at 20, 30, 40, and 50 ppt were 218.3±15.6, 205.0±7.0, 198.0±10.0 and 177.3±27.9 individuals ml^{-1} respectively. The increments (%) and specific growth rate (SGR % day^{-1}) were significantly higher in the cultures fed on baker's yeast in all experimental groups and were lower in all treatments fed instant algae® *Isochrysis sp.* . Resting eggs production during the culture period was positively related to the production rate of rotifers**. The** highest number of resting eggs produced (no. ml^{-1}) was recorded for the rotifers cultures fed baker's yeast at all salinity levels at different intervals. On the other hand, the rotifers fed instant algae® *Isochrysis* sp. showed variations in the numbers of resting eggs produced during all intervals. The numbers were observed to decline with time and increasing salinity until it reached the minimum in the 50 ppt cultures.

The study suggested that there is a strong relationship between the food type and culture conditions. Accordingly, protein content and ether extract were found to be higher in all treatments fed baker's yeast under all salinity levels. The lowest protein and ether extract contents were recorded in the cultures fed instant algae® *Isochrysis sp*. Saturated fatty acids (myristic acid C14:0; palmitic acid C16:0; margaric acid C17:0 and stearic acid C18:0), a monounsaturated fatty acid (oleic acid C18:1 ω9) and one polyunsaturated fatty acid (linoleic acid C18:2 ω6) were detected.

The results clearly showed that the concentration of excretory products (nitrogenous compounds) was inversely correlated with the density of rotifers per ml under the affection of salinity and food type.

The cultures of rotifers fed backer's yeast at 20 ppt salinity level recorded the maximum ammonia concentration and the minimum concentration of ammonia were measured in the cultures of rotifers fed on *chaetoceros sp.* at 50 ppt salinity level. However, nitrite concentrations were maximum in the cultures of rotifers fed on *chaetoceros sp*. at 40 ppt salinity level and the minimum concentrations were measured in the cultures of rotifers fed on baker's yeast at salinity level of 50 ppt. Rotifers cultures fed *Chaetoceros* sp., instant algae® Isochrysis *sp·* and baker's yeast at 20 ppt salinity level showed relatively higher levels of dissolved oxygen concentrations which were 2.67±0.15, 2.63±0.06, and 2.53±0.12 $mg.l^{-1}$, respectively. The minimum dissolved oxygen concentrations were recorded in the cultures of rotifers fed on instant algae® *Isochrysis sp.* at 30 ppt salinity level followed by the cultures fed *Chaetoceros* sp. at 50 ppt salinity level and the cultures of rotifers fed instant algae® *Isochrysis sp.* at the same salinity level. The range levels of pH recorded during the experimental period was 7.22-7.78±0.01. This was considered to be within the optimum range for rotifers multiplication.

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الخلاصة

تمت دراسة تأثير الملوحة ونوع الغذاء على معدل النمو والقيمة الغذائية للروتفيرات ذات الحجم الصغير من نوع *Brachionus rotundiformis* تحت أربعة مستويات ملوحة (30,20، 40 و 50 جزء فى الألف) وثلاثة أنواع من الغذاء (خميرة الخبز Baker's yeast ، طحالب الكيتوسيروس *Chaetoceros sp.* و والطحالب الفورية المركزة من نوع

ايزوكرايسيس *Isochrysis sp.* Instant algae®) ونتج عن ذلك اختلافات فى كثافة اعدادها ونسبة زيادتها ومعدل نموها النوعى بعد 84 ساعة فترة تربية. معدلات الإنتاج القصوى كانت أعلى فى جميع التجارب التى تمت تغذيتها على خميرة الخبز تحت مستويات الملوحة الأربعة ، 20، 30، 40 و 50 جزء في الألف ، حيث كانت قيمها (± الإنحراف المعيارى) 320.7±3.8 ، 219.3 ± 19.5، 211.3 ± 3.2 و 204 ± 5.3 خليه /المليمتر، على التوالىوأدنى معدلات تم تسجيلها لجميع التجارب التى تمت تغذيتها على الطحالب الفورية المركزة من نوع ايزوكرايسيس تحت مستويات الملوحة الأربعة حيث كانت قيمها : 218.3 ± 15.6، 208± 7،0، 198 ± 10 و 177.3 ± 27.9خلية/ المليمتر على التوالى. نسبة الزيادة (%)ومعدل النمو النوعى (%/ اليوم) كانت أعلى بشكل معنوي في جميع التجارب التى تمت تغذيتها على خميرة الخبز تحت مستويات الملوحة الأربعة. إنتاج البيض الساكن (خلية/ المليمتر) resting eggs فى جميع المجموعات التجريبية كان ذو علاقة بمعدل الإنتاج لكل مجموعة. وعليه فإن أعداده كانت اكبر فى جميع التجارب التى تمت تغذيتها على خميرة الخبز تحت مستويات الملوحة الأربعة. المجموعات التى تمت تغذيتها على الطحالب الفورية المركزة من نوع ايزوكرايسيس أظهرت إختلافات فى أعداد البيض المنتج تحت جميع مستويات الملوحة. ولوحظ ان أعداد البيض الساكن تتناقص بزيادة الملوحة و الفترة الزمنية للزراعة حتى وصل الى أقل أعداده فى جميع المجموعات تحت مستويات الملوحة 50 جزء في الألف.

الدراسة الحالية إقترحت وجود علاقة قوية بين الروتفيرات، نوع الغذاء وظروف التربية وتبعاً لذلك فإن نسبة محتوى البروتين الخام و المستخلص الإثيري كانت أعلى فى الروتفيرات التى تمت تغذيتها على خميرة الخبز تحت جميع مستويات الملوحة وأقل نسب للبروتين الخام و المستخلص الإثيري تم تسجيلها فى الروتفيرات التى تمت تغذيتها على الطحالب الفورية المركزة من نوع ايزوكرايسيس تحت جميع مستويات الملوحة. الأحماض الدهنية المشبعة (حمض المايريستك C14:0 ، حمض البالميتك C16:0، حمض المارقاريك C17:0 وحمض الستريك C18:0) والأحماض الدهنية الأحادية المشبعة (حمض الأوليك ω9 C18:1) وأحد الأحماض الدهنية المتعددة غير المشبعة (حمض اللينوليك ω6 C18:2) تم الكشف عنها فى جميع عينات الروتفيرات لكل المجموعات التجريبية.

أظهرت التجارب بوضوح أن تركيزات المواد الإخراجية (المواد النيتروجينية) الناتجة من الروتيفيرات داخل وسط التربية تحت تأثير الملوحة ونوع الغذاء المستخدم ذات تأثير عكسى على كثافة أعداد الروتفيرات فى المليمتر. المجموعات التجريبية التى تمت تغذيتها على خميرة الخبز تحت مستوى الملوحة 20 جزء في الألف سجلت أعلى قياس لتركيزات للأمونيا وأقل تركيزات له تم قياسها للمجموعات التى تمت تغذيتها على الطحالب من الكيتوسيروس تحت مستوى الملوحة 50 جزء في الألف. ومن ناحية أخرى فإن أعلى تركيزات النتريت تم قياسها للمجموعات التى تمت تغذيتها على طحالب الكيتوسيروس تحت مستوى الملوحة 40 جزء في الألف وأقل تركيزات له تم قياسها للمجموعات التى تمت تغذيتها على خميرة الخبز تحت مستوى الملوحة 50 جزء في الألف. المجموعات التى تمت تغذيتها على طحالب الكيتوسيروس ، الطحالب الفورية المركزة من نوع ايزوكرايسيس و خميرة الخبز تحت مستوى الملوحة 20 جزء في الألف أظهرت إرتفاعاً نسبياً في مستويات تركيزات الأكسجين الذائب والتى كانت: 2.67 ± 0.15، 2.63 ± 0.06 و 2.53 ± 0.12 ملجرام/الليتر على التوالى. و أقلتركيزات لمستويات الأكسجين الذائب تم تسجيلها للمجموعات التى تمت تغذيتها على الطحالب الفورية المركزة من نوع ايزوكرايسيس تحت مستوى الملوحة 30 جزء في الألف تليها المجموعات التى تمت تغذيتها على طحالب الكيتوسيروس تحت مستوى الملوحة 50 جزء في الألف والمجموعات التى تمت تغذيتها على الطحالب الفورية المركزة من نوع ايزوكرايسيس تحت نفس مستوى الملوحة. الأس الهيدروجينى pH خلال فترة التجارب تتراوح بين 7.22- 7.78±0.01والذي يعتبر ضمن المدى الأمثل لتكاثر الروتفيرات.

INTRODUCTION

Most of the marine finfish larvae with aquaculture potential have very limited yolk reserves and at start of exogenous feeding most of the larvae were observed to die in the past. It was later recognized that the problem was due to

the fact that newly hatched larvae are usually very small, extremely fragile, and generally not physiologically fully developed. For example, their small size (i.e. small mouth size), the uncompleted development of their perception organs (i.e. eyes, chemoreceptor) and digestive system, are limiting factors in proper feed selection and use during the early first-feeding or start-feeding period (Fountaine and Revera, 1980; Reitan et al., 1994; Bromage and Roberts, 1995; Sanoamuang et al., 2001; Arimoro 2006). To overcome this problem, various types of food, including a number of zooplankton, protozoa, larvae of marine finfish, minced fish…etc., were tried without success (Fountaine and Revera, 1980). This situation continued until the rotifers were recognized as the most suitable live food starter (Lubzens, 1987). The success of rotifers as a culture organisms are due to their planktonic nature, nutritional quality, tolerance to wide range of environmental conditions, high reproduction rate (0.7-1.4 offspring/female/day). Moreover, their small size (130-320μm), slow swimming velocity and tendency to stay suspended in the water column make them a suitable prey for fish larvae that have just reabsorbed their yolk sac (Hirata, 1979, 1980; James et al., 1983; Lubzens, 1987; Dhert et al., 1995; Dhert, 1996; Fengqi, 1996). On the other hand, the filter-feeding nature of rotifers allows inclusion in body tissues of specific nutrients (bioencapsulation) essential for fish larvae such as essential fatty acids, vitamins and antibiotics (Watanabe et al., 1983; Watanabe, 1991; Theilacker and Kimball, 1984; Lubzens, 1987). The most widely used species is *Brachionus plicatilis*, a cosmopolitan inhabitant of inland saline and coastal brackish waters (Fukusho, 1989). However, for use in aquaculture, a simple classification is used which is based on two different morphotypes, namely *B. rotundiformis* or small rotifers (S-type) with a lorica length ranging from 100 to 210 μm (average 160 μm) and *B. plicatilis* or large rotifers (L-type) with a lorica

length ranging from 130 to 340 μm (average 239 μm) (Fielder et al., 2000; Hagiwara et al., 2007).

Basically, several algal species have been used to feed seawater rotifers species. Other alternative food types such as yeast, waste water from food industry and live stock, minced fish and a number of species of zooplankton, protozoa and larvae of marine finfish have been used for rotifers mass culture (Klekot and Klimowicz, 1981; Takano, 1995; Samra et al., 2001). Baker's yeast was first used by Hirata and Mori (1967) as food for the saline water species *B. plicatilis*. Since then a number of investigators have used bakers' yeast as food for this species and rotifers grown in this way have also been nutritionally enriched (Fernandez-Reiriz and Labarta 1996, Lie et al., 1997).

Beside the effect of the food type, the nutritional quality of rotifers is also very much affected by the biotic factors of the seawater. The rotifer *B. plicatilis (rotundiformis*) is a euryhaline species and can withstand a wide salinity range from 1 to 97 ppt. (Fengqi, 1996). However, optimal reproduction can only take place at salinities below 35 ppt (Lubzens, 1987).

Objectives of the study

Despite the abundant information on the culture of rotifers, the combined effect of food type and salinity still remain an area of interest for further investigation. Therefore, this study was designed to study the production rates and nutritional quality of S-type rotifers, *B. rotundiformis*, fed two different species of microalgae (*Chaetoceros* sp. and concentrated instant algae® *Isochrysis* sp.), and baker's yeast under four salinity levels.