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Comparative studies on fatty acid composition of the ovaries and hepatopancreas at different physiological stages of the Chinese mitten crab

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Abstract

The fatty acid composition of the ovary at different physiological stages (immature, mature, spawning, egg loss and abortion) of the Chinese mitten crab *Eriocheir sinensis* was investigated with capillary gas chromatograph. A total of 18 types of fatty acids were found in the ovary of *E. sinensis.* Three of them were major fatty acids: oleic acid (C18:1) (31.96–37.31%), palmitic acid (C16:0) (16.42–23.03%) and palmitoleic acid (C16:1) (16.46–18.43%). Among the total fatty acids, the content of monounsaturated fatty acids (MUFA) were the highest (50.71–55.65%), saturated fatty acids (SFA) were the second (20.23–29.22%), and polyunsaturated and high unsaturated fatty acids (PUFA) were the lowest (16.58–27.87%). Comparative studies of the ovary and hepatopancreas at different physiological stages found significant differences in the content of fatty acids, SFA, MUFA, PUFA and ω_3/ω_6. Some fatty acids were not detectable at certain stages. It is noteworthy that arachidonic acid (C20:4) was only found in the egg-losing crabs. The fatty acid composition and the content of fatty acids in the ovaries have a direct relationship with *E. sinensis* abortion or egg loss.

Keywords: Chinese mitten crab; *Eriocheir sinensis*; Ovary; Fatty acid; Abortion; Embryo attachment

1. Introduction

Lipids play an important role during the development of decapod crustaceans, not only as energy sources, but also as essential nutrients (Kanazawa et al., 1985). Lipids are believed to be one of the key nutritional factors affecting egg hatching rates and larval survival (Xu et al., 1994). Some essential fatty acids (EFA) have also been shown to be of special significance for gonad maturation and brood quality (Soudant et al., 1996). In crustaceans, the hepatopancreas is generally regarded as a major lipid storage organ. In the case of female crustaceans, ovaries also contain higher levels of lipid than other organs, and this suggests that lipids are important for maturation of crustacean ovaries (Ando et al., 1977; Teshima and Kanazawa, 1983). The fatty acid content and composition of the crustacean ovaries have a direct influence on reproduction, egg survival and embryonic development (Teshima and Kanazawa, 1983; Lautier and Lagarrigue, 1998; Alava et al., 1993). The purpose of studies on variation and characteristics of fatty acids in crustacean ovarian development at

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different physiological stages is to understand reproduction at the biochemical level.

In the past decades, studies on crustacean fatty acids have focused on the nutritional requirements of brooders during ovarian development (Gonzalez-Baro and Pollero, 1988; Lytle et al., 1990; Mourente, 1996; Teshima and Kanazawa, 1983; Wen et al., 2002) or juvenile growth (Wen et al., 2003; Xu et al., 1994). Large quantities of fatty acids were found to be necessary for the development of ovaries (Teshima and Kanazawa, 1983). However, little information is available for fatty acid composition and variation in ovaries of spawning and aborting crabs.

The Chinese mitten crab, *Eriocheir sinensis* (H. Milne Edwards 1854), is one of the most important cultivated species and an important food source for South–East Asia. During the brooding period, individuals of *E. sinensis* often lose their embryos ahead of hatching time. This causes economic loss in Southern China. Many studies have been done on the abortion mechanism of shrimps and crabs (Fisher and Clark, 1983; Goudeau et al., 1987; Harper and Talbot, 1984; Cheung, 1966; Saigusa et al., 2002), and the relationship between egg loss and tegumental gland development has been reported (Yang et al., 1996; Yang and Zhou, 2000). The fatty acid composition of the hepatopancreas of *E. sinensis* at different physiological stages as well as the morphological features of normally attached embryos and aborted embryos were studied (Ying et al., 2004a, b,c). We hypothesize that the fatty acid composition and the content of fatty acid in the ovary had a close relationship with *E. sinensis* abortion or egg loss. To our knowledge, there are no reports about the relationship between fatty acid composition in the ovary and abortion in *E. sinensis*. In the present paper, we report variation in the fatty acid composition during *E. sinensis* ovarian development, in order to elucidate the relationship between fatty acid composition and abortion in the Chinese mitten crab.

2. Materials and methods

2.1. Animals

E. sinensis were collected from Qingjiang Breeding Farm of Yueqing City, Wenzhou, Zhejiang Province, China. Sixty immature, mature and spawning crabs were collected in July, October and December of 2002; 150 Wenzhou native-produced female crabs were collected in January of 2003. Animals were placed into glass tanks (55 × 50 × 40 cm) filled with fresh seawater (18 °C), fed with fresh clam meat and aerated with oxygen. After 6 d, the female crabs spawned. Abortion was artificially induced by increasing temperature abruptly as well as by changing the light cycle. With the temperature rising from 18 to 20 °C and the illuminating time elongating from 12 to 16 h, embryos of partial spawning crabs shed from their abdomen in 12 h after treatment. By continually rising the temperature to 22 °C, the number of aborting females increased by 30%. Sixty percent of the embryos shed after 3 d of fluctuating water temperatures and culture conditions. Sixty aborted crabs were sampled, and 60 immature, mature, spawning and aborted female crabs were divided into 6 groups of 10 individuals. Fresh ovaries were removed for moisture and fatty acid determination, and length and breadth of the carapace were measured (Table 1).

2.2. Moisture determination

Fresh ovaries were dried in an oven at 60 °C for 24 h, and the moisture in ovary was determined by weighing the ovaries before and after drying.

2.3. Fatty acid analysis

Fatty acid analysis was determined by gas chromatography (Shimadzu GC-9A) equipped with an autosampler, which was also linked to a spectral data recording microcomputer (C-R2AX). Analysis was conducted under the following: the carrier gas was nitrogen, the revolving rate was 20 ml/min. The analysis column was a Stabilwax capillary column (25 m in length, 0.25 mm in diameter), and the detecting temperature was 260 °C. The oven was programmed to rise from initial temperature to 230 °C, and the detection mode was the flame ionization detector (FID).

We used the potassium hydroxide–methanol high-temperature esterification method. The benzene to petroleum aether ratio was 1:1 to dissolve the lipids, and we used potassium hydroxide–methanol solution to esterify. Each fatty acid was determined according to the characteristics of standard fatty acid chromatograms under the same chromatogram conditions. Mixed standard fatty acids were purchased from Sigma Chemical Company (St. Louis, MO).

2.4. Statistical analysis

Data were presented as means ± standard deviation. The Statistic software package (Version 6.0, Stat Tech) was used to analyse the data of fatty acids of ovaries collected from different physiological stages (immature,