

KARYOTYPE OF THE GIANT FRESHWATER PRAWN, *MACROBRACHIUM ROSENBERGII*

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ABSTRACT

Number and characteristics of chromosomes of the giant freshwater prawn, *Macrobrachium rosenbergii*, were studied from the second meiotic metaphase of the fertilized egg ($n = 59$), from mitotic metaphase of the actively dividing embryonic cell and from spermatogonial metaphase of the testicular tissue ($2n = 118$). The karyotype consists of 6 large, 26 medium, 15 small and 12 very small pairs. Most of these chromosomes are submetacentric and metacentric. Only 6 pairs appear to be either telocentric or acrocentric in configuration.

INTRODUCTION

Decapods possess the highest chromosome numbers in the animal kingdom.¹ Most chromosomes are small and are metacentric or submetacentric,¹⁻³ which makes them difficult to differentiate. Thus, discrepancy in chromosome numbers of the same species has been reported.⁴ Great variations in chromosome number even among related species of decapods have been found. For example, two related species of the freshwater crab, *Acanthotelphusa martensi* and *Paratelphusa jacquemonti* have 88 and 132 chromosomes, respectively,⁴ while the freshwater prawn, *Macrobrachium siwalikensis*⁵ and the American lobster, *Homarus americanus*³ have 106 and 138 chromosomes, respectively. The highest chromosome number ($2n = 376$) has been reported in the crayfish *Astacus trawbridgii*.⁶ Moreover, no distinguishable sex chromosomes have been found in most decapods studied^{4, 5} although their existence is claimed in some species.^{7, 8}

MATERIALS AND METHODS

Mature giant freshwater prawns, *Macrobrachium rosenbergii*, were obtained from prawn farms in Nakorn Prathom, Thailand. Haploid chromosomes were prepared from the fertilized eggs collected immediately after spawning from the mated females. Diploid chromosomes were prepared from testicular tissues and from one-day old embryos.

Fertilized eggs and embryos were fixed for several days in ethanol : acetic acid (3 : 1). The fixed specimens were washed briefly in absolute ethanol and stained with 2% acetoorcein for 20 min. They were placed in 45% acetic acid for 5 min and then squashed under cover slips.

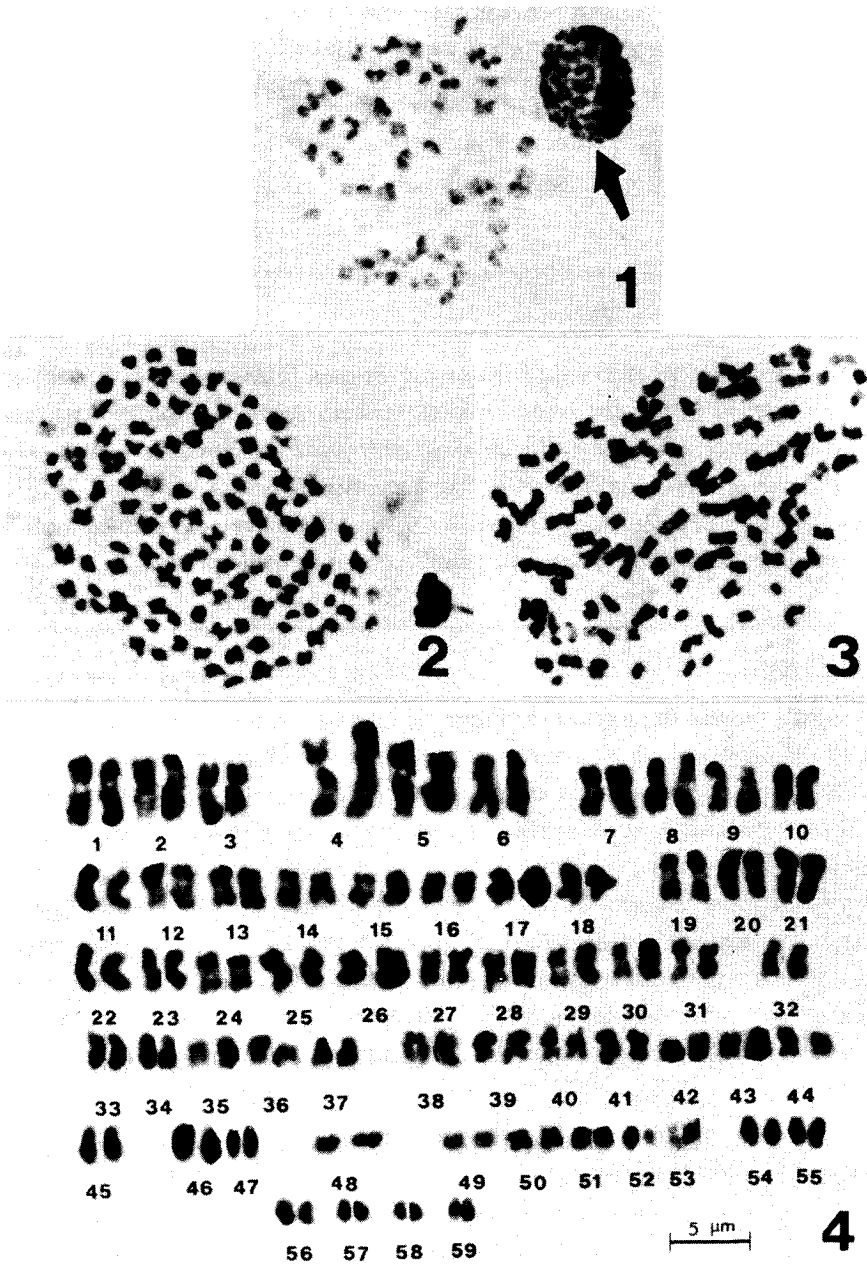


Fig. 1. Second meiotic metaphase prepared from the newly spawned fertilized egg. Arrow indicates chromosomes of the polar body.

Fig. 2. Spermatogonial metaphase prepared from the testicular tissue.

Fig. 3. Mitotic metaphase prepared from the actively dividing embryonic cells.

Fig. 4. Karyotype of *Macrobrachium rosenbergii*.

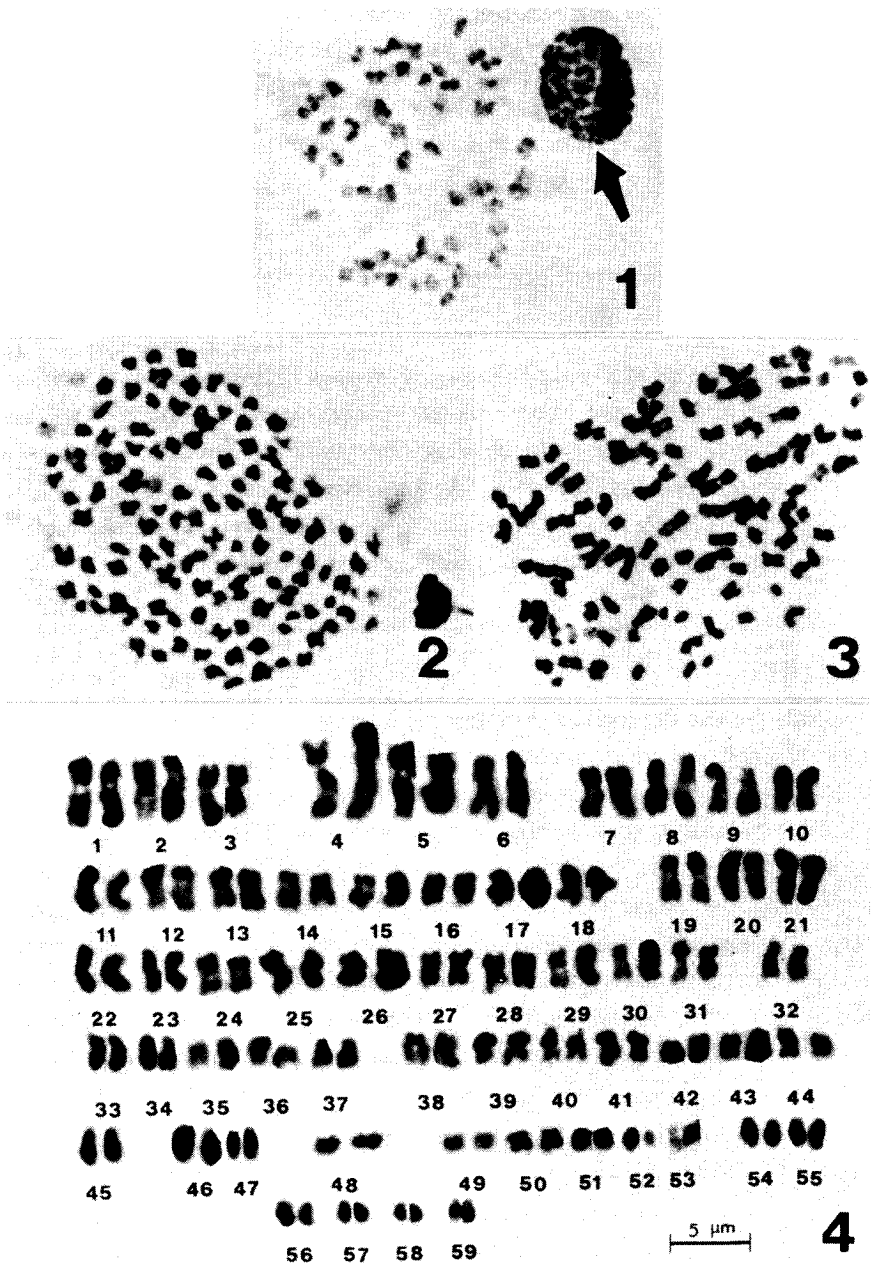


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Mature males were injected with 0.2 ml of 0.3% colchicine about 24 hours prior to sacrifice. The testes were dissected and placed in 2% sodium citrate for 20 min. They were fixed in ethanol : acetic acid (3 : 1) for 15 min and then minced in 60% acetic acid for 15 min. The samples were then dropped on the warm slides (45°C) and air dried. The slides were stained with 2% Giemsa in 0.1 M phosphate buffer for 20 min.

The haploid chromosome number was scored from the second meiotic metaphase of the newly spawned fertilized eggs. And the diploid chromosome number was observed from spermatogonial metaphase of the testicular tissues and from mitotic metaphase of the actively dividing embryonic cells.

RESULTS AND DISCUSSION

Examination of at least 28 well-spread chromosomes from at least 7 individuals in each type of chromosome preparations have revealed that haploid chromosome number of *Macrobrachium rosenbergii* is 59 (Fig. 1) and the diploid chromosome number is 118 (Figs. 2, 3). The karyotype can be arranged into 4 groups (Fig. 4). These consist of 6 pairs of large (Nos. 1-6), 26 pairs of medium (Nos. 7-32), 15 pairs of small (Nos. 33-47) and 12 pairs of very small chromosomes (Nos. 48-59).

All large chromosomes are submetacentric (Nos. 1-6) but half of the medium chromosomes are submetacentric (Nos. 7-18 and 32) and half are metacentric (Nos. 19-31). Among the small chromosomes, 6 pairs are submetacentric (Nos. 33-37 and 45), 7 pairs are metacentric (Nos. 38-44) and 2 pairs appear to be telocentric or acrocentric (Nos. 46-47). And of the very small chromosomes, 2 pairs are submetacentric (Nos. 54-55), 6 pairs are metacentric (Nos. 48-53) and 4 pairs appear to be telocentric or acrocentric (Nos. 56-59). No marked distinction of the sex chromosomes was observed in this study.

Macrobrachium rosenbergii shows typical chromosome characteristics of the decapods, which have high chromosome numbers with variation in size and shape. These present a technical problem in cytotaxonomy. Moreover, like other cases of *Macrobrachium*,⁵ sex chromosomes could not be indentified in this study.

An advantage of chromosome preparation from eggs over that from testicular tissues in Crustaceans has been suggested¹ Utilization of eggs or embryos of *Macrobrachium rosenbergii* which deposit externally at the brood chamber of the female is economically safe and technically simple. No tedious dissection or sacrifice of animal is required. However, precise timing of chromosomal stages of the eggs or developing embryos is prerequisite. Chromosomal stages of the eggs or developing embryos have been reported previously.⁹ The present study selects stages which give the second meiotic metaphase during oocyte maturation and mitotic metaphase of the developing embryo to represent the haploid and diploid chromosomes respectively. Furthermore, the squash technique is necessary for chromosome preparation from the eggs due to large cytoplasmic masses.

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บทคัดย่อ

จำนวนแฮพลอยด์โครโมโซมของกุ้งก้ามกรามที่ศึกษาจากระยะไมโอติคเมทาเฟสที่สองของไข่ที่ได้รับการปฏิสนธิใหม่ ๆ แสดงว่ามี 59 แห่ง และจำนวนดิพลอยด์โครโมโซมที่ศึกษาจากระยะไมโทติคเมทาเฟสของเซลล์เอมบริโอ และจากระยะไมโอติคเมทาเฟสที่หนึ่งของเซลล์ในอวัยวะแสดงว่ามี 118 แห่ง โครโมโซมเหล่านี้ประกอบด้วยโครโมโซมขนาดใหญ่ 6 คู่ ขนาดกลาง 26 คู่ ขนาดค่อนข้างเล็ก 15 คู่ และขนาดเล็ก 12 คู่ ส่วนใหญ่จัดว่าเป็นพวกสับเมทาเซนทริกและเมทาเซนทริก