The late events of fertilisation in the penaeoidean shrimp *Sicyonia ingentis*

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**Summary**

Antibodies to sea urchin β-tubulin and mammalian heavy neurofilaments were used to study the late events of fertilisation in the penaeoidean shrimp *Sicyonia ingentis*. The neurofilament antibody fortuitously stained centrosomes in eggs, as well as the subacrosomal region and acrosomal filament in sperm. The neurofilament antibody also stained a cortical site in eggs which was associated with the positioning of the mitotic spindle. During pronuclear migration, a large maternal microtubule aster formed in addition to the sperm aster. The activity of the maternal centrosome disappeared during synkary, while the sperm centrosomes formed the poles of the first mitotic spindle. Colcemid treatment modulated the size of the mitotic spindle and blocked pronuclear migration.

Keywords: Centrosomes, Fertilisation, Intermediate filaments, Microtubules, Penaeoidean shrimp

**Introduction**

Although previous studies have described the initial events of fertilisation and subsequent development in penaeoidean shrimp (Hudinaga, 1942; Kajishima, 1951; Zilch, 1979; Pillai & Clark, 1987; Lindsay et al., 1992; Hertzler & Clark, 1992), few studies have specifically addressed pronuclear migration and syngamy in these or other crustaceans. Pronuclear migration has been described in the caridian *Macrobrachium* (Damrongphol et al., 1991); however, the mechanism of pronuclear movement was not determined. Microtubules are likely to be involved in pronuclear migration in the brine shrimp *Artemia* (Anteunis et al., 1967; Criel, 1992) and the crab *Eriocheir sinensis* (Lee & Yamazaki, 1989), but neither study provided a description of the global microtubule patterns in crustacean eggs. Migration of the germinal vesicle is associated with microtubules in the caridan prawn *Palaemon serratus* (Cledon, 1986), but later events of fertilisation were not examined in this study. Therefore, it seemed appropriate to address this period of development in crustaceans, using whole-mount immunofluorescence techniques recently modified for the penaeoidean shrimp *Sicyonia ingentis* (Hertzler & Clark, 1992).

In most species, the sperm introduces a centrosome at fertilisation which becomes the primary microtubule-organising centre of the zygote and divides to form the poles of the first mitotic spindle (revived in Wilson, 1925; Mazia, 1987). The dynamics of microtubules and centrosomes during fertilisation have been studied in diverse phyla including nematodes (Albertson, 1984), echiurids (Luykx, 1991), molluscs (Kuriyama et al., 1986), insects (Callaini & Riparbelli, 1990), echinoderms (Harris et al., 1980; Bestor & Schatten, 1981), ascidians (Sawada & Schatten, 1988), and mammals (Schatten et al., 1985; Le Guen & Crozet, 1989; Sathaniathan et al., 1991). Ultrastructural and immunocytochemical studies have demonstrated the paternal inheritance of centrosomes in every case, with the single exception of the mouse (Maro et al., 1985; Schatten et al., 1985).

The migrations of both male and female pronuclei usually depend on the microtubules of the sperm aster, nucleated from the sperm centrosome. In addition, through the selective inactivation of the maternal centrosome and duplication of the paternal centrosome (Sluder et al., 1989a, 1993) the essential bipolarity of the ensuing cleavage division is established.