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the recognition motif in fibronectin. Pierschbacher and Ruoslahti used this finding to isolate the fibronectin receptor using a modified affinity–column approach (Pytela et al., 1985).

Much work would come later, defining the roles of all of the ECM components, cytoskeletal players (Otey et al., 1990), and integrin subunits (e.g., Wayner and Carter, 1987), including those acting in lymphocyte adhesion (Jalakanen et al., 1987; Dustin and Springer, 1988; Diamond et al., 1990). But Buck notes that out of the 1982 CSAT paper "came definitive identification of the integrins as a complex linking the cytoskeleton to the ECM. It also led to the sequencing of the β1 subunit, and the field developed rapidly after that."

The search also integrated the Buck, Horwitz, Hynes, and Yamada laboratories into lasting friendships and collaborations. "We believed in what we did," Horwitz recalls. "We first had a handle on adhesion, and then neuronal outgrowth and cancer came into view. At one point, we thought we had the whole world." The field would go on to show that this receptor was part of a larger cell biology story—initiating cell signaling events central to many cell activities. JCB

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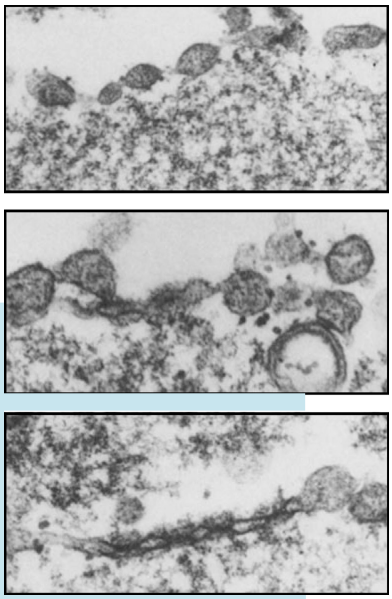

LOHKA

Frog egg extracts allow the study of nuclear envelope formation.

Frog egg extracts can do a cell's work

For his graduate project at the University of Toronto, Manfred Lohka wanted to answer one simple question: what enzyme was controlling the decondensation of a sperm nucleus when it entered the egg? At the time, an egg protease was thought to be involved, so he figured he should start by making egg extracts in which to test sperm. He got much more than he bargained for, including the birth of a potent cell-free biochemical assay.

After harvesting a test tube of eggs from *Rana pipiens* frogs, Lohka could spin the eggs at low speed and pop the cytoplasmic contents out of their plasma membranes "just like taking the skin off a grape." If he added *Xenopus laevis* sperm heads to the activated cytoplasm, he observed that the sperm heads transformed into pronuclei and then mitotic chromosomes (Lohka and Masui, 1983).

But it wasn't until Lohka and advisor Yoshio Masui looked at their extract-plus-sperm preparations by electron microscopy that they realized that not only did the sperm nucleus decondense, but a nuclear envelope was assembling around it as well (Lohka and Masui, 1984). Lohka provided one of the first descriptions of envelope assembly: membrane vesicles flattened and fused into a double-membraned structure, complete with nuclear pores. In addition, if Lohka fractionated the egg extracts with a higher spin and separated them into soluble and particulate fractions, he could show that envelope assembly required both.

Lohka credits Masui's love of unorthodox approaches for the discovery that egg cytoplasm can support cellular activities at least for short periods. "That was back when how you did experiments was an expression of your personality," says Lohka, now at the University of Calgary in Canada.

He notes that the study outcomes were not as important in the long run "as the notion that you could actually get quite complex cell processes to occur in cell-free extracts." The system was used to purify metaphase-promoting factor (Lohka et al., 1988), which led to the identification of cdc2 and cyclin as its components (Gautier et al., 1990) and the extracts became a valuable tool for further investigating the cell cycle (Murray, 1991). The system has also been used to study nuclear transport (Newmeyer et al., 1986), DNA replication (Mills et al., 1989), and spindle microtubule dynamics (Heald et al., 1996). JCB

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