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# CBD-1 organizes two independent complexes required for eggshell vitelline layer formation and egg activation in *C. elegans*

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## Abstract

Metazoan eggs have a specialized coat of extracellular matrix that aids in sperm-egg recognition. The coat is rapidly remodeled after fertilization to prevent polyspermy and establish a more permanent barrier to protect the developing embryo. In nematodes, this coat is called the vitelline layer, which is remodeled into the outermost layer of a rigid and impermeable eggshell. We have identified three key components of the vitelline layer structural scaffold – PERM-2, PERM-4 and CBD-1, the first such proteins to be described in the nematode *C. elegans*. CBD-1 tethered PERM-2 and PERM-4 to the nascent vitelline layer via two N-terminal chitin-binding domains. After fertilization, all three proteins redistributed from the zygote surface to the outer eggshell. Depletion of PERM-2 and PERM-4 from the scaffold led to a porous vitelline layer that permitted soluble factors to leak through the eggshell and resulted in embryonic death. In addition to its role in vitelline layer assembly, CBD-1 is also known to anchor a protein complex required for fertilization and egg activation (EGG-1-5/CHS-1/MBK-2). We found the PERM complex and EGG complex to be functionally independent, and structurally organized through distinct domains of CBD-1. CBD-1 is thus a multifaceted regulator that promotes distinct aspects of vitelline layer assembly and egg activation. In sum, our findings characterize the first vitelline layer components in nematodes, and provide a foundation through which to explore both conserved and species-specific strategies used by animals to build protective barriers following fertilization.

Graphical abstract