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In vitro model system for the study of milk protein secretion

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To study intracellular mechanisms of milk protein traffic to the cell surface and secretory vesicles biogenesis in mammary epithelial cells (MEC), we are establishing a *in vitro* system using the mouse MEC line HC11. HC11 cells grown on plastic, although heterogeneous, can easily be induced to synthesize β casein (β cn) in the presence of insulin, cortisol and PRL. When HC11 cells are grown on porous filters, they expressed ≥ 10 times more β cn. Milk proteins secreted by these cells were essentially found in the apical medium and identified by western blotting as lactoferrin and γ cn. Only a minor proportion of the cellular β cn was found in the apical medium. Analysis by SDS-PAGE showed that β cn from cells and medium migrated as a doublet of lower Mr. than the mature mouse milk β cn. This increase in electrophoretic mobility suggested that this form had not undergone all post-translational modifications and was blocked in an early compartment of the secretory pathway. Consistent with this hypothesis, we observed by electron microscopy that the rough endoplasmic reticulum of these cells was highly dilated. In some experiments, HC11 cells also expressed α cns and, interestingly enough, the mature form of β cn was observed in addition to the β cn doublet. The amount of mature β cn was found to be proportional to the level of expression of α cns.