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Getting a G–RRP on regulated exocytosis in the heart

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A study by Rybkin et al. (see p. 527) substantially advances our understanding of regulated exocytois by specialized secretory cells, such as atrial myocytes. A second member of the Ras-related protein family, RRP17, was identified and shown to participate in regulating the secretion of the cardiac-derived peptide hormone, atrial natriuretic peptide. In addition to the heart, RRP17 was shown to be expressed in neuronal, pancreatic, and skeletal muscle cells, suggesting a widespread role in regulated secretion for this new protein.

Comment

Vesicle-bound cargo can be released from mammalian cells by constitutive or regulated exocytosis. All cells exhibit constitutive exocytosis; however, neuroendocrine cells also possess a specialized form of exocytosis that requires activation of the appropriate signal transduction pathways and is, therefore, referred to as regulated exocytosis (An and Zenisek, 2004; Silver and Pappas, 2005). Cardiac myocytes in the atrium are both contractile and endocrine cells capable of secreting several peptide hormones, including the blood pressure and volume lowering hormone, atrial natriuretic peptide (ANP) (Fig. 1). As in most peptide hormone-secreting cells, large dense-core vesicles (LDCVs) store cargo that is bound for regulated exocytosis, while constitutive exocytosis involves smaller vesicles (Michael et al., 2006). Although the regulation of ANP synthesis has been well studied, the packaging of ANP into LDCVs and its trafficking and exocytosis are less well understood. In this issue, Rybkin et al. (2007) make progress in understanding these processes by identifying a new Ras-related protein involved in regulated secretion of ANP.

In most cases, the signaling pathways required for regulated exocytosis culminate with an increase in cytosolic calcium (Oheim et al., 2006; Rutter et al., 2006), which is required for the priming, as well as the docking and the fusion of secretory vesicles with the plasma membrane (Stojilkovic, 2005). Numerous proteins are involved in the docking and fusion of LCDVs with the plasma membrane; some are located on the LDCV membrane, while others are found on the cytosolic face of the target membrane, or in the cytosol. SNAREs (soluble N-ethylmaleimide-sensitive factor attachment protein receptors) facilitate the docking of the LDCVs near the plasma membrane in preparation for the fusion event (Stojilkovic, 2005), while others, such as calcium activated protein for secretion 1, or CAPS1 (Walent et al., 1992) confer calcium dependence to the fusion event. The Ras superfamily of small GTPases, including all Rho family members, Rab, ARF, G_o-α, and Ral, are also involved in regulated exocytosis (Pfeffer, 2007).

Rybkin et al. (2007) identified a novel small GTPase in the heart, Ras-related protein on chromosome 17 (RRP17) which is also expressed in brain, pancreas, and skeletal muscle. They also demonstrated that RRP17 can interact with CAPS1, a protein known to mediate LDCV exocytosis, suggesting that RRP17 may also be involved in regulated exocytosis of LDCVs. Consistent with this concept are the similarities in the previously published expression patterns of CAPS1 and RRP17, although CAPS1 expression had not been previously examined in the heart. Rybkin et al. (2007) found that atrial myocytes expressed CAPS1, RRP17, and ANP; however, ventricular myocytes, which do not normally express ANP and are not known to have LDCVs under these conditions, expressed only RRP17 (Table I, Normal heart). This led to the hypothesis that RRP17 might require CAPS1 to participate in regulated exocytosis. The authors used two experimental approaches to examine this hypothesis. In the first, they took advantage of the fact that ANP expression in ventricular myocytes is induced during certain cardiac pathologies, such as pressure overload. In contrast to normal ventricular tissue, they found that CAPS1 and ANP were both expressed in the ventricles of mice subjected to maneuvers that mimic cardiac pathology (Table I, Pathologic heart). In the second approach, Rybkin et al. (2007) generated RRP17 knock-out mice and found that, compared with normal mice, the atrial myocytes in the $RRP17^{-/-}$ mice possessed smaller LDCVs. Moreover, the hearts of $RRP17^{-/-}$ mice contained less ANP, and the mice exhibited increased blood pressure, both of which are consistent with roles for RRP17 in the pathway leading to LDCV exocytosis of ANP from the heart.

The study by Rybkin et al. (2007) shows for the first time that a small GTPase, RRP17, can interact with CAPS1, and that RRP17 participates in ANP release from cultured cardiac

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Abbreviations used in this paper: ANP, atrial natriuretic peptide; CAPS1, calcium-activated protein for secretion 1; LDCV, large dense-core vesicles.

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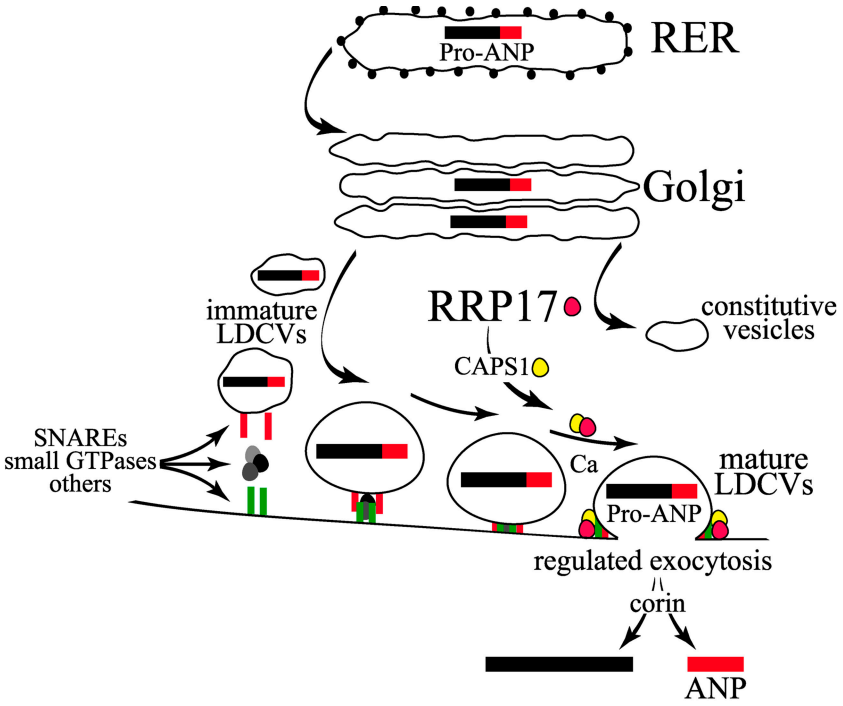

Figure 1. **Summary of regulated exocytosis of ANP from atrial myocytes.** Shown is the hypothetical subcellular route of ANP packaging into LDCVs, as well as the co-secretional processing of pro-ANP to ANP by corin (Chan et al., 2005). The hypothetical involvement of various SNAREs, Ras-related small GTPases, and many other proteins, including CAPS1 (yellow), is shown. The involvement of RRP17 (magenta) and CAPS1 in calcium-dependent regulated exocytosis of LDCVs in the heart is the topic of the study by Rybkin et al. (2007).

myocytes, and from the heart, in vivo. But other aspects of RRP17 function are yet to be addressed. For example, recent evidence suggests that CAPS1 plays roles in regulated, as well as constitutive exocytosis (Fujita et al., 2007); thus, as a CAPS1 binding partner, it is possible that RRP17 might participate in both forms of exocytosis. Also, because RRP17 is expressed in ventricular myocytes, even in the absence of CAPS1, perhaps it has CAPS1-independent roles in the release of membrane-bound cargo, or other membrane fusion events. Moreover, as described by Rybkin et al. (2007), the structure of RRP17 is such that it is likely to interact with novel regulators of its GTPase activity, which may reveal new molecular mechanisms of action of this new and growing family of small GTPases. Thus, while much remains to be discovered about RRP17, the study by Rybkin et al. (2007) adds to our understanding of the cell biology of LDCV exocytosis, as well as contributing to the mechanism of peptide hormone release by normal and diseased myocardium, in vivo.

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Table I. **Summary of atrial and ventricular myocyte characteristics**

	Normal heart		Pathologic heart	
	Atria	Ventricles	Atria	Ventricles
ANP	+	−	+	+
LDCV	+	−	+	?
Reg exocytosis	+	−	+	+
RRP17	+	+	+	+
CAPS1	+	−	+	+

This table summarizes the expression of ANP in the atria and ventricles of the normal and pathologic (hypertrophic) adult mouse heart, as well as our current state of knowledge concerning the presence of LDCVs and whether the myocytes in each tissue exhibit regulated exocytosis. Also shown is a summary of the expression of RRP17 and CAPS1 in the two tissues under normal and pathologic conditions, as demonstrated in the study by Rybkin et al. (2007).

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