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COMMENTARY

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RNA, a new member in the glycan-club that gets exposed at the cell surface

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When thinking about glycosylation, we mainly think of proteins or proteoglycans, which is not surprising as glycosylation is the second most frequent post-translational modification. However, glycans extend beyond proteins, and the glycosylation of lipids is now well established. To further document the nature of glycans and their associated roles in various biological functions, the Bertozzi laboratory has developed and applied original chemical approaches (biorthogonal chemistry).[1] As part of those, innovative metabolic labelling of glycans strategies were developed using azido-sugars to visualize and track glycoconjugate moieties.[2] According to a recent paper by Flynn et al, that relied on such an approach, RNAs can also be included into the world of glycans.[3] They coined these RNAs as "glycoRNAs" and found that they are small non-coding RNAs with an overrepresentation of Y RNAs, which were identified as constituents of Ro60 ribonucleoproteins[4] and small nucleolar RNA (snoRNAs), which are involved in a growing number of post-transcriptional processes.[5] Glycosylation of RNAs was sensitive to digestion with PNGaseF, an enzyme known to cleave the linkage between asparagine and the proximal GlcNAc of N linked-glycans in proteins. This implies that the glycosylation of RNAs involved an amide bond-containing linker. However, none of the nucleobases has such linkers. It is unlikely that a large linker acts as a carrier because the sedimentation experiments by Flynn et al suggest a linker with a small molecular weight. It is possible that precursors of nucleobases are modified in a manner that resembles an asparagine-like functionality. These open questions need further investigations to clarify the synthesis pathway of glycoRNAs.

Although the chemical synthesis pathway of glycoRNAs is unclear, it appears that the enzymes responsible for their synthesis are the same ones involved in N-glycan synthesis such as oligosaccharyltransferase or α-mannosidases I and II, proteins localized in the endoplasmic reticulum (ER) and Golgi apparatus.[6-8] Small non-coding RNAs were so far only reported to locate to the cytosol. However, the enzymes responsible for their modification localize in the lumen of the ER and in the Golgi complex. This implies that these small non-coding RNAs translocate to the lumen of the ER to become glycoRNAs. Strikingly, glycoRNAs were found to localize to the cell surface, thereby indicating they might use the secretory pathway to reach their final destination. This has however to be

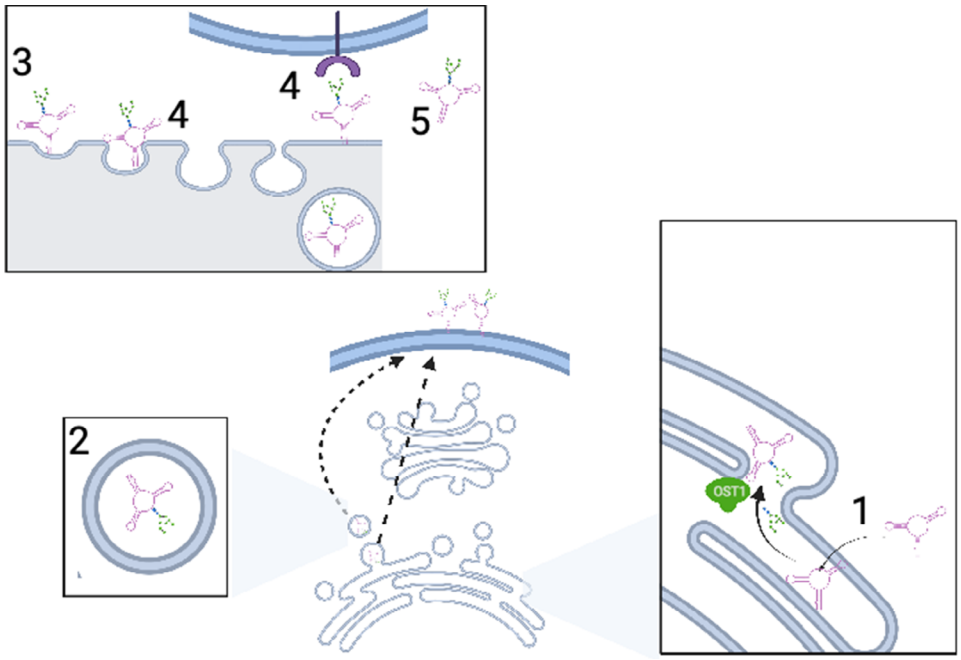

FIGURE 1 Schematic illustration of the main open question around glycoRNAs that are referred to in the main text. The numbers correspond to the numbering in the main text

formally demonstrated since small glycoRNAs were not detected at high levels in endomembrane compartments. At the cell surface, glycoRNAs might serve to regulate cell-cell communication by serving as ligands for receptors on other cells. In fact, Flynn et al propose that glycoRNAs are potential ligands for members of the Siglec receptor family. Besides this physiological role, glycoRNAs might potentially be involved in pathophysiological processes such as serving as triggering factors and/or targets for autoantibodies in autoimmune diseases such as systemic lupus erythematous where anti-RNA antibodies were found previously.

This study represents a major advance and opens several questions for future investigations. We have outlined five key questions that we represent graphically in Figure 1:

1. How does the small non-coding RNA translocate into the ER lumen? Is there any non-glycoRNA entering the secretory pathway?
2. How does the glycoRNA leave the ER to reach the cell surface? Is there a glycoRNA quality control system within the secretory pathway? The glycosylation pattern of glycoRNAs would suggest that they transit through the Golgi complex. If so, are there cargo receptors that mediate sorting of glycoRNAs into carriers, or do they leave non-selectively by bulk flow?
3. How are glycoRNAs anchored at the cell surface?
4. What is the fate of the glycoRNAs at the cell surface? Are they endocytosed, or do they bind to other molecules in other cells?
5. Are glycoRNAs released as soluble agents into the extracellular space? This could happen by shedding from the surface. In this case, they might serve as decoy ligands and compete with their membrane-associated counterparts or find new types of RNA-avid molecules.

In conclusion, Flynn and colleagues have reported the existence of a new biological species with unknown functions. This paves the way for many hypotheses that will require in-depth investigations in cellular models and in vivo.

PEER REVIEW

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