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How do sphingolipids and lipid rafts relate to pathology?

It had been realized for decades that glycosphingolipids with their complex carbohydrate structures must have dedicated functions [1]. This happened before it was realized that these functions may be intimately linked to the physical properties of sphingolipids and, more specifically their organization in microdomains or rafts. These lipid rafts [2] have been implicated as both signaling platforms, and also as a means to traffic glycolipids and certain proteins to specific membranes of polarized cells. They are now thought to be involved in the entry into the cell of toxins, viruses and bacteria, play a role in protein (mis)processing in prion disease and Alzheimer's, and in the cellular immune response.

Likewise, early notions on the involvement of sphingolipids in signaling have dramatically evolved with the finding that the balance between simple sphingolipid metabolites, the "sphingostat", regulates the cell cycle [3]. In terms of signaling, ceramide and sphingosine-1-phosphate (S1P) are the best-studied bioactive sphingolipids. They are produced for signaling pathways via the regulated activity of various enzymes. Ceramide and S1P often exert opposite effects, with ceramide usually inhibiting proliferation and promoting apoptosis, and S1P stimulating growth and suppressing apoptosis. Thus, many inducers of apoptosis or growth arrest activate one or more pathways of ceramide generation, and in many cases, there is evidence that endogenous ceramide mediates, at least in part, the apoptotic and growth arrest responses of these inducers. Ceramide has been implicated in the actions of, for instance, tumor necrosis factor-α, and in the cytotoxic responses to amyloid Aβ peptide, which suggests a role for this pathway in Alzheimer's disease and neurodegeneration. Conversely, many growth factors such as platelet-derived growth factor, as well as tumor necrosis factor-α, have been shown to induce S1P formation, which in turn has been implicated in mediating viability and inflammatory responses. Sphingolipids have also been implicated in disease etiology, notably cancer. Crucial roles for ceramide and S1P-mediated pathways have been identified in both cancer development and progression, with ceramide functioning as a tumor-suppressor lipid, inducing anti-proliferative and apoptotic responses in various cancer cells, and S1P acting as a tumor-promotor.

Not unexpectedly these two areas come together: (A) Signal transduction by ceramide apparently occurs in lipid rafts. In terms of signaling platforms, it is now believed that acid-sphingomyelinase becomes exposed on the cell surface in response to stimulation via certain receptors. The produced ceramide modifies pre-existing small rafts to fuse into larger signaling platforms and to trap activated receptor molecules in the platforms, thereby enhancing the efficacy of signaling, and providing a functional and conceptual link between ceramide signaling and sphingolipid-enriched microdomains. This kind of mechanism may be particularly important in signaling by immune T and B cells. (B) Complex glycosphingolipids in rafts are involved in signaling. Complex glycolipids might also be involved in cancer since they are shed from a number of cancer cells and ganglioside depletion has been shown to reduce tumorigenicity. Together, these discoveries are paving the way for the advancement of anti-cancer therapies involving manipulation of sphingolipid signaling pathways and the formation of complex glycolipids.

As an understanding of the roles of sphingolipids in disease etiology and treatment – most notably for cancer – has evolved, sphingolipid analogs such as safingol as well as anti-cancer chemotherapeutics that modify sphingolipid metabolism (such as fenretinide) have entered clinical trials, illustrating the strong partnership of basic and translational research in this field. Other challenges are to find small molecule inhibitors of ceramide clearance and S1P production, and to find ways to interfere with the role of sphingolipids in infectious diseases and as toxin receptors.

One major task in the field is to develop sensitive, holistic and high-throughput approaches to assess the sphingolipid status of cells and body fluids [4]. In addition, insight in the complex relationships between the various sphingolipids will require modeling of the metabolic pathways and the protein complexes involved. This does not only concern metabolomics, but also the subcellular localization of the lipids, of their metabolic enzymes and of their transporters. The localization of lipids will require improved methods of imaging: immunolocalization of lipids, and improved methods of cell fractionation in combination with refined analysis.

A second challenge is the analysis of the structure and function of lipid rafts. At this moment there is not a single general method that allows an unequivocal assessment of the structure and composition of lipid rafts. Existing methods like detergent-extraction, detergent-free isolation, single-molecule trapping, correlation microscopy, and atomic force microscopy must be optimized and standardized. Improvements in cell fractionation, like the use of viruses to sample the lipid composition of rafts during budding [5] must be applied to the problem, next to novel physical approaches, like STED microscopy [6]. Also in this case, the biochemical approaches must be supported by studies on the basic properties of the constituents, and molecular biophysical measurements on model systems, simulations and modeling will be needed to create the basis for understanding raft properties in vivo.

It is now clear that there is a sphingolipid component in many diseases: a defect in their hydrolysis results in a storage disease, they play a role in infection and toxin uptake, often via rafts, and the sphingolipid balance is involved in cell proliferation and death. The diagnosis and cure of these diseases will require contributions from the areas of basic biochemistry and biophysics, genetics, proteomics and metabolomics including lipidomics, developmental biology, and from the more applied areas of chemical biology and drug design. In line with postulates expressed in preceding editorials [7–9], only an integrated approach will provide the depth required to understand the chemistry, physics and human biology that underlie the diseased state, and to devise therapy.

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