In all animals, lipoproteins are used to transport lipids and other lipophilic compounds through the aqueous environment in the vascular system. The complex interconversion of different lipoproteins and their lipid and apolipoprotein compositions have been extensively characterized in mammals. For years, lipidologists have focused on lipoproteins as vehicles for the transfer of the major incorporated lipids, i.e. cholesterol, triglycerides and phospholipids. A novel concept in the field of lipoprotein research was recently introduced by Eaton, Thiele and coworkers [1]: by employing a combination of genetic and biochemical strategies, they showed a function for lipoprotein particles as vehicles for the movement of lipid-linked signaling molecules (morphogens) and glycosphatidylinositol-linked proteins.

It is well established that during development, secreted lipid-anchored signaling molecules are capable of acting over long distances. For instance, the cholesterol-anchored and palmitoylated Hedgehog protein [2,3] activates gene expression over 12 cell diameters from its site of production [4]. Another fatty acylated morphogen, Wingless, may signal through a range of over 30 cell diameters [5,6]. What has puzzled researchers for years is how these proteins with high affinity towards membranes travel over such long distances.

Panakova et al. approached the question by taking advantage of the model organism fruit fly, Drosophila melanogaster has been used in powerful genetic and developmental biological strategies but has remained less appreciated in lipoprotein research. Insects use only a single lipoprotein, lipophorin, a spherical particle slightly smaller than mammalian LDL and with a density similar to mammalian HDL (for a recent review, see [7]). It is composed of phospholipids, diacylglycerol, sterols and hydrocarbons, and one copy of each of the two nonexchangeable apolipoproteins, apolipoporphin I and II (apoLI and apoLII). Apolipoporphins are produced in the fat body of the animal, which combines the functions of the mammalian liver and adipose tissue: it stores and synthesizes lipids, lipophorin, glycogen, and other important metabolites. The apolipoporphins are generated by a posttranslational cleavage of the precursor proapolipophorin [8,9].

The apparent simplicity of the insect lipoprotein system turned out to be highly useful. By directing RNA interference (RNAi) against the apolipoporphin mRNA, a dramatic reduction of apoLI and apoLII (and hence, total lipoprotein) to 5% of wild-type levels could be achieved. This resulted in gross developmental effects – the animals prolonged the third larval instar and rarely pupariated – but experiments could be performed on third-instar larvae. The investigators found severe perturbations of lipid transport as evidenced by accumulation of neutral lipids in the midgut, reduced size of fat bodies and fewer lipid droplets in wing imaginal discs. When examining Hedgehog distribution and signaling in lipophorin-RNAi discs, they found that the range of Hedgehog signaling was decreased: transcript activation was narrowed from an 11-cell distance to a maximum six-cell distance. Furthermore, Hedgehog accumulated together with Patched to abnormally high levels in the first few rows of signal receiving cells. This suggests that lipophorin may be needed to help Hedgehog move to the more distal cells and decrease the probability of Patched endocytosing it.

In the case of Wingless, lipophorin-RNAi led to decreased levels of extracellular Wingless and an abnormally narrow gradient of target gene expression, suggesting that long-range Wingless signaling also requires lipophorin. Direct evidence for the association of lipid-linked proteins with lipophorin was provided by biochemical fractionation and immunoprecipitation experiments, upon generation of antibodies against fly apoLps. Although the vast majority of Hedgehog comigrated with cellular membranes, about 2% of the Hedgehog in discs was found to associate with low-density particles. Interestingly, the fraction was substantially higher (up to 22%) with several glycosphatidylinositol-anchored proteins, including, for example, acetylcholine esterase.

At present it is not known – although it seems likely – that a mechanism similar to that found for Hedgehog and Wingless signaling in the fly applies for long-range control of growth and patterning during mammalian
development. The complexity and apparent redundancy of lipoprotein trafficking systems in higher organisms may complicate the design of analogous approaches. The findings of Panakova et al., however, suggest that lipoproteins serve as long-distance carriers for functionally diverse lipid-modified proteins. In the proteomics era, it will now be tempting to look in more detail into the trace lipophilic components of mammalian lipoproteins. In addition to the vascular tree, paracrine mediators of signaling might be uncovered from the interstitial fluid.

References

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Boycott KM, Flavelle S, Bureau A, et al. Homozygous deletion of the very low density lipoprotein receptor gene causes autosomal recessive cerebellar hypoplasia with cerebral gyral simplification. Am J Hum Genet 2005; 77:477–483.

This is evidently the first report of a human lipoprotein receptor mutation leading to a malformation syndrome. A large deletion encompassing the entire LDL-receptor gene was identified as the cause of a recessively inherited syndrome of moderate-to-severe mental retardation and predominantly truncal ataxia in the genetically isolated Hutterite population. Neuropathologically, the disease is associated with cerebellar hypoplasia and cerebral gyral simplification. Interestingly, the Hdl–/– mouse also manifests with cerebellar hypoplasia. In the mouse, the effects on lipoprotein metabolism are largely masked by the LDL-receptor. The animals, however, have elevated triglycerides after a high-fat diet and are protected from diet-induced obesity. Notably, half of the human patients had an abnormally low body mass index, suggesting that there may be some protection from obesity.

Boucard AO, Flach J, Kupatt E, et al. Enhanced transmembrane cholesterol flux in oxysterol-sensitive cells. J Biol Chem 2005; 280:284–293.

The authors demonstrated that atrial electrical function could be specifically modified without affecting ventricular electrophysiology.

Panakova D, Sproson H, Marius E, et al. Lipoprotein particles are required for Hedgehog and Wingless signalling. Nature 2005; 435:58–65.

The authors provide strong evidence for a novel function for lipoprotein particles in which they act as vehicles for the movement of lipid-linked secreted signaling molecules and glycosphingolipid/inositol-linked proteins. The findings help to explain why some secreted proteins are lipid anchored and how proteins with a highly lipophilic nature can spread long distances in the aqueous intercellular milieu. They also raise the possibility that lipoprotein particles or selective lipid uptake is linked to processing of the signal in the receiving cells.

Pelkmans L, Zerial M. Kinase-regulated quantal assemblies and kiss-and-run recycling of caveolae. Nature 2005; 436:128–133.

This elegant study, together with that of Tagawa et al. (see below), provides important new insights into caveolar dynamics. The results show that the majority of caveolae on the plasma membrane are static. Those that are mobile undergo local fission–fusion cycles with the plasma membrane, without disassembling the caveolar coat. A molecular switch mechanism, for example phosphorylation, may shift the mode of cycling from short to long-range transport to intracellular locations, such as caveosomes. A set number (estimated at 144 ± 39) of caveolin-1 molecules is incorporated in one caveolar coat. Interestingly, caveolar lipids may also be reduced in mobility as suggested by the findings that the GM1-bound cholera toxin B in caveolae remained sequestered after caveolar fusion with the plasma membrane.

Tagawa A, Mezzacasa A, Hayer A, et al. Assembly and trafficking of caveolar domains in the cell: caveolae as stable, cargo-triggered vesicular transporters. J Cell Biol 2005; 170:769–779.

The authors reported that caveolae constitute cholesterol-dependent membrane domains, which first assembled in the Golgi complex, were transported as vesicles to the plasma membrane and remained together as stable units. The fraction of mobile caveolae both in the plasma membrane and in the caveosome could be doubled, for example by a virus using caveolae for cell entry. Even under these conditions, however, individual caveolar domains underwent little exchange of caveolin-1. Remarkably, transient cholesterol depletion and repletion allowed the exchange of caveolin molecules and stimulated the reformation of caveolae with mixed caveolin populations, reinforcing the idea that cholesterol stabilizes the caveolar structure. The mobility of cholesterol per se as part of the containers remains to be elucidated.

Wolfrum C, Poy MN, Stoffel M. Apolipoprotein M is required for preβ-HDL formation and cholesterol efflux to HDL and protects against atherosclerosis. Nat Med 2005; 11:418–422.

ApoM is a recently identified component of HDL with an unknown function. This important study shows that apoM is a major apolipoprotein determining HDL and preβ-HDL concentrations, and thereby modulates macrophage cholesterol efflux and susceptibility to atherosclerosis. Overexpression of apoM doubled plasma HDL and preβ-HDL levels. In contrast, apoM deficiency decreased plasma HDL concentrations and led to an unfavorable remodeling of the particles, with an absence of preβ-HDL, decrease in HDLβ levels and formation of large HDL particles (HDLc). Moreover, cholesterol efflux from macrophages to apoM-deficient HDL was impaired. Finally, atherosclerotic hepatic overexpression of apoM protected LDL–/– mice against atherosclerosis. This compelling data set warrants several lines of investigation: to study the role of low apoM levels in atherosclerosis susceptibility at the population level, to investigate the molecular mechanisms of apoM-induced HDL remodeling, and to exploit this pathway for antithrombotic strategies.