Chapter 16
Cellular Nanotubes: Membrane Channels for Intercellular Communication

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Abstract Cells of living organism communicate in many different ways with their neighbor cells. This is accomplished by, for example, the secretion of signaling molecules or the formation of proteinaceous pores, referred to as gap junctions, between physically attached cells. In addition to these long-known communication routes, a novel mechanism was discovered recently based on de novo formation of membrane nanotubes, which facilitate the delivery of biological molecules and organelles between cells. Interestingly, chemists have been developing artificial carbon-based nanostructures with a similar architecture for communication with cells and delivery of clinically interesting drugs. Along with every new developed technology involving the use of foreign compounds in biomedical applications, concerns emerge on the biocompatibility and toxicity at the cellular level. This is particularly true for nano-sized materials, whose effects are yet to be thoroughly determined *in vivo*. Biocompatibilization of synthetic compounds may be done more efficiently if naturally occurring structures are taken as models.

Keywords Tunneling nanotube, TNT, cellular communication, intercellular transport

Abbreviations CNT, Carbon nanotube; F-actin, Filamentous actin; TNT, Tunneling nanotube

16.1 Structure and Formation of Tunneling Nanotubes

Tunneling nanotubes (TNTs) were first described in cultured rat pheochromocytoma PC12 cells as thin continuous membranous channels that span the shortest distance between connected cells (Fig. 16.1) (Rustom et al., 2004). They have a diameter between 25 and 200 nm, a length up to several tens of micrometers, and they are extended above the substratum and not in contact with it (reviewed in Gerdes et al.

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F. Cataldo, T. Da Ros (eds.) *Medicinal Chemistry and Pharmacological Potential of Fullerenes and Carbon Nanotubes,*

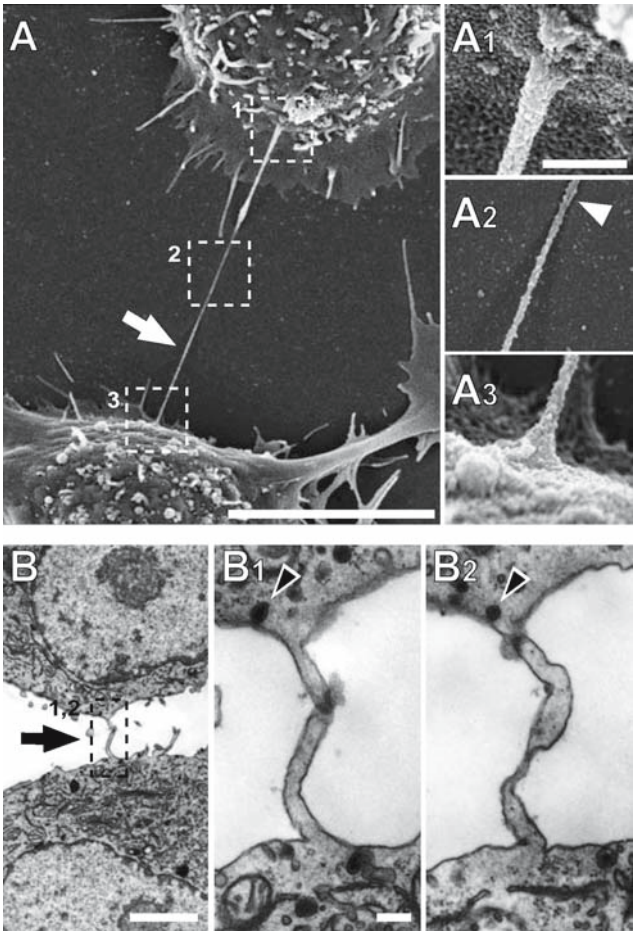

Fig. 16.1 Ultrastructure of tunneling nanotubes (TNTs). (A) Scanning electron microscopic (SEM) or (B) transmission electron microscopic (TEM) image of a tunneling nanotube connecting two cultured PC12 cells. The boxed areas in (A) and (B) are shown as higher magnification images (insets A1–A3, B1, B2). (B1) and (B2) represent consecutive 80 nm sections, which demonstrate membrane continuity between the connected cells. Bars: 10 µm (A); 2 µm (B); 200 nm (insets A1–A3, B1, B2) (Modified from Rustom et al., 2004)

(2007)). Branched TNTs occur occasionally between cultured cells with a typical angle of 120° between the different tubular junctions (Önfelt et al., 2004; Rustom et al., 2004). The interior of TNTs is filled with a bundle of filamentous actin (F-actin) over the entire length of the nanotube (Rustom et al., 2004), while for some cells an additional type of thicker nanotube connectors (with a diameter up to 1 µm), containing both F-actin and microtubules, was found (Önfelt et al., 2006). Live-cell imaging has provided the basis for the current proposed mechanisms for de novo formation of TNTs. Frequently, a filopodia-like protrusion from one cell may come into contact with the plasma membrane or a filopodium from another cell, and a

straight membrane tube is formed at the shortest distance between the connected cells. Alternatively, TNTs can emerge when two cells in close contact diverge (Önfelt et al., 2004; Rustom et al., 2004; Gerdes et al., 2007). Since their discovery, diverse intercellular membrane nanotubes have been described for different cell types, and the idea of broad structural and functional heterogeneity for this type of nanotube-mediated cell-to-cell communication is emerging (Gerdes and Carvalho, 2008; Gurke et al., 2008; Davis and Sowinski, 2008).

The discovery of TNTs may end up challenging the established cell theory concept of a cell as the primary unit of life in animals, if future studies can reveal a role of TNT-mediated communication in fundamental cellular processes. In the plant kingdom the multicellular organism is already considered the primary unit of life, driven by the role of plasmodesmata, cytoplasmic channels interconnecting plant cells, in processes such as cellular differentiation and development (reviewed in Baluška et al. (2004)).

16.2 Artificial Membrane Nanotubes

TNTs are fragile structures, prone to disruption by mechanical stress, chemical fixation, and prolonged light exposure during widefield microscopy (Rustom et al., 2004; Koyanagi et al., 2005; Watkins and Salter, 2005). These characteristics have made it difficult to analyze the molecular composition as well as the physical properties of such singular naturally occurring tubules. Despite these constraints, important information can be obtained from studies on model artificial membrane tubes such as those created by pulling tethers from synthetic lipid vesicles or cellular plasma membrane (Fig. 16.2). The morphology of such tubes resembles that of

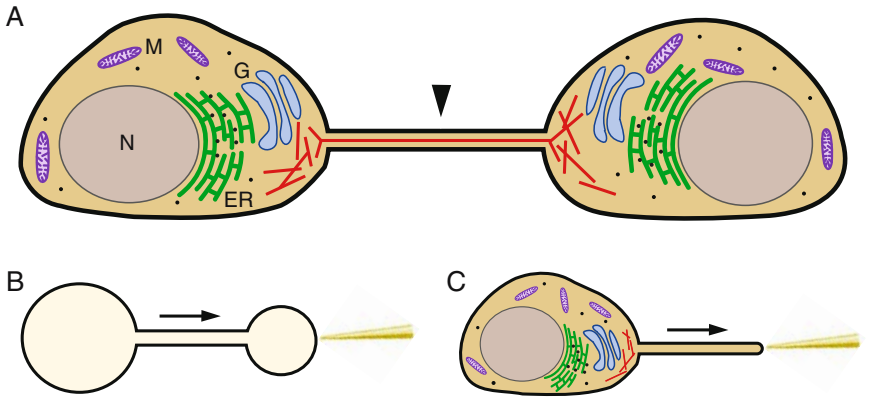

Fig. 16.2 Schematic representation of cellular and artificial membrane nanotubes. (A) Two cells are connected by a tunneling nanotube (arrowhead) containing a bundle of filamentous actin (red line). N (grey), nucleus; M (purple), mitochondrium; ER (green), endoplasmic reticulum; G (blue), Golgi apparatus. (B) Lipid nanotube connecting two lipid vesicles formed by pulling a membrane tether. (C) Membrane tether pulled from the plasma membrane of a cell (*see Color Plates*)

TNTs and it seems reasonable that the same physical laws govern the common shape in both structures. Both TNTs and artificial lipid tethers are straight tubes spanning the shortest distance between two connected points, which hints that this invariable geometry may be energetically favorable. The diameter of a nanotube extracted from membranes is determined by the balance between the bending rigidity and the tension of the membrane, and is typically in the range of 40–400 nm (Karlsson et al., 2001), comparable to TNTs. Similar Y-shaped junctions naturally found in TNTs can be produced artificially by pulling of tethers from lipid vesicles (Karlsson et al., 2002; Lobovkina et al., 2006) and even from straight TNTs (Pontes et al., 2007). Geometrical calculations estimate that this symmetric three-way nanotube junction has the lowest energy (Yin and Yin, 2006).

Formation of membrane tubes by directly pulling tethers from cell membranes requires an initial rise in the force elongation profile, to overcome the increased bending energy of the plasma membrane and the strength of the membrane–cytoskeleton links that need to be disrupted (Li et al., 2002). Alternatively, it was also shown that tubular budding in lipid vesicles can be induced by adding strong anisotropic amphiphilic molecules, which accumulate at the buds and cause a spontaneous curvature and tubular initiation. Under these conditions a direct pulling mechanical force was no longer required (Yamashita et al., 2002). Acquisition of specific proteins that drive membrane curvature has also been associated with tubule formation *in vitro* (Farsad et al., 2001) and *in vivo* (Razzaq et al., 2001). In contrast to artificial membrane tethers, TNTs are not hollow cylindrical membrane tubes, but contain bundles of F-actin attached to the cortical actin cytoskeleton. Thus, it is likely that actin polymerization plays a role in the outward pushing of the plasma membrane and nanotube formation *in vivo* as it is known for the extension of filopodia (Faix and Rottner, 2006). This is also consistent with the observation that the action of molecular motors bound to the bilayer of lipid giant unilamellar vesicles is sufficient to generate membrane tubes (Roux et al., 2002; Koster et al., 2003). In addition, elongation of plasma membrane nanotubes requires a membrane flow from the cell plasma membrane into the growing tube and it is suggested that cells maintain a membrane reservoir (e.g., ruffles, invaginations), controlled by the cytoskeleton, to provide a buffer against membrane tension over several micrometers of tube elongation (Raucher and Sheetz, 1999; Sun et al., 2005). This is also true when nanotubes are formed from lipid bilayer vesicles, where a slight rehydration of the vesicles by increasing the osmotic strength of the aqueous suspension is necessary to provide the bilayer reservoir (Evans et al., 1996).

16.3 Tunneling Nanotubes Are Conduits for the Delivery of Molecules

The current knowledge of TNTs points to their main function being in facilitating the unidirectional intercellular transport of biological molecules either by providing a channel for diffusion of small cytoplasmic molecules or by containing the neces-

sary motors for an active transport of membrane containers. Passive diffusion is likely to be the mechanism for the transfer of calcium fluxes observed between dendritic cells and THP-1 monocytes (Watkins and Salter, 2005), but on the other hand, the small molecule calcein with only 400 Da could not be seen passing through TNTs between PC12 cells (Rustom et al., 2004). This suggests that the structural heterogeneity of TNTs in different cells may determine the size exclusion limit of the channel or reflect the existence of a gating mechanism for the transfer of specific molecules. Different membrane containers can travel along TNTs and into the connected cell. TNTs were observed to traffic small organelles belonging to the endosomal/lysosomal system between PC12 cells (Rustom et al., 2004), NRK cells (Rustom et al., 2004), immune cells (Önfelt and Davis, 2004) and human prostate cancer cells (Vidulescu et al., 2004), and mitochondria between neonatal rat cardiac myocytes and human endothelial progenitor cells (Koyanagi et al., 2005). The mechanism by which such organelles are shipped along the interior of TNTs, tightly filled with bundles of F-actin, is still unknown, but is likely to be an active process. The presence of the actin-associated motor protein myosin Va inside TNTs (Rustom et al., 2004; Zhu et al., 2005), partially co-localizing with organelles (Rustom et al., 2004), further supports the involvement of an actin-/myosin-driven transport associated with the movement of organelles inside the TNTs. In addition, a slow transfer of several plasma membrane components including membrane proteins and lipid molecules has been described (Rustom et al., 2004; Watkins and Salter, 2005). Diffusion rates of membrane components in TNTs are likely to be lowered by the tight interactions between the membrane and the F-actin. Conversely, pulling tethers from cell membranes uncouples the lipid bilayer from the membrane-associated cytoskeleton and results in higher diffusion rates for integral membrane proteins in the tether than those observed in the plasma membrane (Berk et al., 1992).

Finally, it has been speculated that TNTs could represent a general mechanism for the intercellular spread of pathogens. In this respect, bacteria and retroviruses were seen to attach to the outer membrane and surf along the nanotubes towards connected cells, where they could be internalized (Önfelt et al., 2006; Sherer et al., 2007). Furthermore, the human immunodeficiency virus type 1 (HIV-1) was shown to move within nanotubes to infect connected cells (Sowinski et al., 2008).

16.4 Bio-Inspired Tuning of Carbon Nanotubes

It is interesting to know how the morphology of man-made carbon nanotubes (CNTs) resembles that of the nature-made TNTs with similar high length to diameter ratios. At the same time, it is evident that the material composite of both structures, lipid membranes in the case of TNTs versus graphitic backbone in the CNTs, is completely different and this determines their properties (Table 16.1) and applications. TNTs have a support of actin filaments enveloped within a tube of lipidic membrane that can easily be disrupted by mechanical and other stress conditions. TNTs are also very flexible and dynamic structures and their lifetime can range

Table 16.1 Comparison of major properties of tunneling nanotubes and carbon nanotubes

	TNT	CNT
Lifetime	Temporary	Stable
Elasticity	Flexible	Rigid
Strength	Fragile	High mechanical strength
Diameter	25–200 nm	0.4–2 nm (SWCNT) 1.4–100 nm (MWCNT)
Length	5–100 µm	20–1,000 nm (SWCNT) 1-several µm (MWCNT)
Conductivity	Ionic	Electrical, thermal
Transport mechanism	Diffusion or active transport through tunneling nanotubes	Diffusion or endocytotic uptake of carbon nanotubes

SWCNT, single-walled carbon nanotube; MWCNT, multiwalled carbon nanotube.

from minutes to hours. During the time of interconnecting a cell pair, TNTs can serve as a pathway for diffusion of ions and small molecules, while bigger molecules or organelles are likely to be transported by an active mechanism. By contrast, CNTs have rigid backbones made of graphite that accounts for their stability and high mechanical strength. In addition, CNTs have singularly high electrical and thermal conductivity properties that can be exploited in therapeutic applications. They are also easily internalized into cells, which is suggested to occur either by diffusion across the lipid bilayer of the cell membrane (Pantarotto et al., 2004) or by endocytosis (Kam et al., 2006). By attachment of interesting molecules to the surface of CNTs, these devices can be efficient transporters *in vivo*. Thus, CNTs are able to translocate nucleic acids and proteins (Kam and Dai, 2005; Kam et al., 2006) as well as drug molecules (Wu et al., 2005) into living cells.

Due to their unique nano-based properties, CNTs are considered as one of the most promising nanomaterials with applications in biomedicine and pharmacology. This includes their exploitation in diagnostics, tissue engineering, and drug delivery. With respect to the latter, the same idea of straight tubular structures interacting with living cells and delivering molecules of interest are behind both CNTs and TNTs. However, the central issue in applying CNTs to living organisms is their biocompatibility. Naked CNTs are hydrophobic and prone to nonspecific adhesion, properties that increase the chance of toxic effects. To overcome these obstacles, researchers have modified the surface of CNTs by introducing non-covalent modifications (Klumpp et al., 2006), to improve the solubility and reduce the associated toxicological responses. Furthermore, concerns on CNT uptake and toxicity have driven studies where biological molecules like proteins (Dutta et al., 2007; Zhang et al., 2007) were used to functionalize the nanomaterial surfaces. Despite the fact that these coatings certainly improved the trafficking and tolerance of CNTs in biological systems, it becomes evident that such modifications have their limits in real biological environments.

In light of these constraints, perhaps the best strategy to further improve the properties of CNTs for medical applications could be to exploit the cellular principles as blueprints. Natural systems use lipid membranes as a universal host matrix, which

possess a large number of membrane proteins, receptors, and channels to fulfill the manifold host functions ranging from signal transduction to transport of molecules across the membranes. In this respect, the recently discovered TNTs with their striking similarity to the nano-sized morphology of CNTs could be most instructive in understanding the basic principles of delivery of substances into cells.

Given the fact that the very first encounter of a CNT with a cell is through the cellular plasma membrane and because membrane translocation is such an important step in any drug delivery strategy, one has to consider how and why nature has designed membranes. Cellular membranes are natural barriers that allow the coexistence of several compartments with specific molecules and specific functions inside. They also provide the matrices for a wide number of integral or attached proteins to associate with their substrates or binding partners. In addition, nature has developed versatile environments at the cellular surface that allow molecular recognition events to guide selective internalization of molecules. Most of these membrane features are expected to be present in cellular TNTs. De novo formation of TNTs is accomplished by an outgrowing of cellular extensions in the direction of neighboring cells presumably following a chemo-tactical guidance. Direct contact of the protrusion with the plasma membrane of the neighboring cell is likely to involve recognition of special plasma membrane molecules that precede the final fusion event. Molecular sensing and recognition events by the CNTs are often required if these nanomachines are to be used in biomedical applications. This can be achieved by attachment of molecules that modify and functionalize the CNT surface.

Thus, from a cell biological point of view, it would be most promising to use molecules present in the natural membrane environments of, for example, TNTs for the biocompatibilization of CNTs and their selective recognition at the cellular level. A major breakthrough in this direction has been the demonstration that coating of CNTs with lipid bilayers results in an efficient and biocompatible barrier between the nanotube surface and the surrounding solution (Artyukhin et al., 2005). In addition, lipid molecules in such a structure were able to diffuse along the bilayer plane much like in a normal cell membrane (Artyukhin et al., 2005). As a second major step, it would be interesting to extend this bio-inspired approach by including selected membrane protein receptors on such an artificial system. These should include membrane proteins, which recognize specific cell surface receptors and thus enable CNTs to selectively interact with certain cell types. This could be of great importance for cancer treatment where a selective targeting mechanism for tumor cells is essential to avoid harmful side effects of the therapy on healthy tissue. In this respect, receptor ligand molecules attached to CNTs have already been proven to recognize selectively the respective binding partners of viruses (Zhang et al., 2007) or tumor cells (Kam et al., 2005; McDevitt et al., 2007).

In addition to molecules providing a cell type-specific targeting, extra signaling molecules could be included to direct the CNTs to cellular compartments such as the nucleus or the mitochondrium. The way nature accomplishes the selective intracellular distribution of molecules is by using targeting signals that are recognized at the surface of specific cellular organelles. The use of signal sequences could be extended to intentionally determine the fate of molecules delivered by CNTs.

We envision that researchers developing strategies for carbon nanotube-based *in vivo* delivery of nanomedicines can benefit from taking into consideration the physiology and function of existing cellular mechanisms for the selective delivery of molecules. It can be expected that further characterization of the molecular composition and transport machinery inside TNTs will help to elucidate the structure and function of these curious ways of intercellular transport up to the tissue level. Understanding how nature functions will provide the ideas and tools to design smart nanoscaled devices for medical and pharmacological applications.

Acknowledgments We would like to thank Steffen Gurke and João Barroso for critical comments on the text and the Norwegian Research Council for financial support (project number 164959/V40).

References

Artyukhin AB, Shestakov A, Harper J, Bakajin O, Stroeve P, Noy A (2005) Functional one-dimensional lipid bilayers on carbon nanotube templates. J. Am. Chem. Soc. 127: 7538–7542.

Baluška F, Volkmann D, Barlow PW (2004) Eukaryotic cells and their cell bodies: Cell theory revised. Ann. Bot. (Lond) 94: 9–32.

Berk DA, Clark A, Jr., Hochmuth RM (1992) Analysis of lateral diffusion from a spherical cell surface to a tubular projection. Biophys. J. 61: 1–8.

Davis DM, Sowinski S (2008) Membrane nanotubes: dynamic long-distance connections betweein animal cells. Nat. Rev. Mol. Cell Biol. 9: 431–436.

Dutta D, Sundaram SK, Teeguarden JG, Riley BJ, Fifield LS, Jacobs JM, Addleman SR, Kaysen GA, Moudgil BM, Weber TJ (2007) Adsorbed proteins influence the biological activity and molecular targeting of nanomaterials. Toxicol. Sci. 100: 303–315.

Evans E, Bowman H, Leung A, Needham D, Tirrell D (1996) Biomembrane templates for nanoscale conduits and networks. Science 273: 933–935.

Farsad K, Ringstad N, Takei K, Floyd SR, Rose K, De Camilli P (2001) Generation of high curvature membranes mediated by direct endophilin bilayer interactions. J. Cell Biol. 155: 193–200.

Faix J, Rottner K (2006) The making of filopodia. Curr. Opin. Cell Biol. 18: 18–25.

Gerdes H-H, Bukoreshtliev NV, Barroso JF (2007) Tunneling nanotubes: A new route for the exchange of components between animal cells. FEBS Lett. 581: 2194–2201.

Gerdes H-H, Carvalho RN (2008) Intercellular transfer mediated by tunneling nanotubes. Curr. Opin. Cell Biol. doi. 10.1016/j.ceb.2008.03.005.

Gurke S, Barroso JF, Gerdes H-H (2008) The art of cellular communication: tunneling nanotubes bridge the divide. Histochem. Cell Biol. 129: 539–550.

Kam NW, Dai H (2005) Carbon nanotubes as intracellular protein transporters: Generality and biological functionality. J. Am. Chem. Soc. 127: 6021–6026.

Kam NW, Liu Z, Dai H (2006) Carbon nanotubes as intracellular transporters for proteins and DNA: An investigation of the uptake mechanism and pathway. Angew Chem. Int. Ed. Engl. 45: 577–581.

Kam NW, O'Connell M, Wisdom JA, Dai H (2005) Carbon nanotubes as multifunctional biological transporters and near-infrared agents for selective cancer cell destruction. Proc. Natl. Acad. Sci. USA 102: 11600–11605.

Karlsson A, Karlsson R, Karlsson M, Cans AS, Strömberg A, Ryttsén F, Orwar O (2001) Networks of nanotubes and containers. Nature 409: 150–152.

Karlsson M, Sott K, Davidson M, Cans AS, Linderholm P, Chiu D, Orwar O (2002) Formation of geometrically complex lipid nanotube-vesicle networks of higher-order topologies. Proc. Natl. Acad. Sci. USA 99: 11573–11578.

Klumpp C, Kostarelos K, Prato M, Bianco A (2006) Functionalized carbon nanotubes as emerging nanovectors for the delivery of therapeutics. Biochim. Biophys. Acta 1758: 404–412.

Koster G, VanDuijn M, Hofs B, Dogterom M (2003) Membrane tube formation from giant vesicles by dynamic association of motor proteins. Proc. Natl. Acad. Sci. USA 100: 15583–15588.

Koyanagi M, Brandes RP, Haendeler J, Zeiher AM, Dimmeler S (2005) Cell-to-cell connection of endothelial progenitor cells with cardiac myocytes by nanotubes: A novel mechanism for cell fate changes? Circ. Res. 96: 1039–1041.

Li Z, Anvari B, Takashima M, Brecht P, Torres JH, Brownell WE (2002) Membrane tether formation from outer hair cells with optical tweezers. Biophys. J. 82: 1386–1395.

Lobovkina T, Dommersnes P, Joanny JF, Hurtig J, Orwar O (2006) Zipper dynamics of surfactant nanotube Y junctions. Phys. Rev. Lett. 97: 188105.

McDevitt MR, Chattopadhyay D, Kappel BJ, Jaggi JS, Schiffman SR, Antczak C, Njardarson JT, Brentjens R, Scheinberg DA (2007) Tumor targeting with antibody-functionalized, radiolabeled carbon nanotubes. J. Nucl. Med. 48: 1180–1189.

Önfelt B, Davis DM (2004) Can membrane nanotubes facilitate communication between immune cells? Biochem. Soc. Trans. 32: 676–678.

Önfelt B, Nedvetzki S, Yanagi K, Davis DM (2004) Cutting edge: Membrane nanotubes connect immune cells. J. Immunol. 173: 1511–1513.

Önfelt B, Nedvetzki S, Benninger RK, Purbhoo MA, Sowinski S, Hume AN, Seabra MC, Neil MA, French PM, Davis DM (2006) Structurally distinct membrane nanotubes between human macrophages support long-distance vesicular traffic or surfing of bacteria. J. Immunol. 177: 8476–8483.

Pantarotto D, Briand JP, Prato M, Bianco A (2004) Translocation of bioactive peptides across cell membranes by carbon nanotubes. Chem. Commun. (Camb): 16–17.

Pontes B, Viana NB, Campanati L, Farina M, Neto VM, Nussenzveig HM (2007) Structure and elastic properties of tunneling nanotubes. Eur. Biophys. J. [Epub ahead of print]

Raucher D, Sheetz MP (1999) Characteristics of a membrane reservoir buffering membrane tension. Biophys. J. 77: 1992–2002.

Razzaq A, Robinson IM, McMahon HT, Skepper JN, Su Y, Zelhof AC, Jackson AP, Gay NJ, O'Kane CJ (2001) Amphiphysin is necessary for organization of the excitation-contraction coupling machinery of muscles, but not for synaptic vesicle endocytosis in Drosophila. Genes Dev. 15: 2967–2979.

Roux A, Cappello G, Cartaud J, Prost J, Goud B, Bassereau P (2002) A minimal system allowing tubulation with molecular motors pulling on giant liposomes. Proc. Natl. Acad. Sci. USA 99: 5394–5399.

Rustom A, Saffrich R, Markovic I, Walther P, Gerdes H-H (2004) Nanotubular highways for intercellular organelle transport. Science 303: 1007–1010.

Sherer NM, Lehmann MJ, Jimenez-Soto LF, Horensavitz C, Pypaert M, Mothes W (2007) Retroviruses can establish filopodial bridges for efficient cell-to-cell transmission. Nat. Cell. Biol. 9: 310–315.

Sun M, Graham JS, Hegedüs B, Marga F, Zhang Y, Forgacs G, Grandbois M (2005) Multiple membrane tethers probed by atomic force microscopy. Biophys. J. 89: 4320–4329.

Sowinski S, Jolly C, Berninghausen O, Purbhoo MA, Chauveau A, Köhler K, Oddos S, Eissmann P, Brodsky FM, Hopkins C, Önfelt B, Sattentau Q, Davis DM (2008) Membrane nanotubes physically connect T cells over long distances presenting a novel route for HIV-1 transmission. Nat. Cell Biol. 10: 211–219.

Vidulescu C, Clejan S, O'Connor KC (2004) Vesicle traffic through intercellular bridges in DU 145 human prostate cancer cells. J. Cell. Mol. Med. 8: 388–396.

Watkins SC, Salter RD (2005) Functional connectivity between immune cells mediated by tunneling nanotubules. Immunity 23: 309–318.

Wu W, Wieckowski S, Pastorin G, Benincasa M, Klumpp C, Briand JP, Gennaro R, Prato M, Bianco A (2005) Targeted delivery of amphotericin B to cells by using functionalized carbon nanotubes. Angew Chem. Int. Ed. Engl. 44: 6358–6362.

Yamashita Y, Masum SM, Tanaka T, Tamba Y, Yamazaki M (2002) Shape changes of giant unilamellar vesicles of phosphatidylcholine induced by a de novo designed peptide interacting with their membrane interface. Langmuir 18: 9638–9641.

Yin Y, Yin J (2006) Geometric conservation laws for cells or vesicles with membrane nanotubes or singular points. J. Nanobiotechnol. 4: 6.

Zhang YB, Kanungo M, Ho AJ, Freimuth P, van der Lelie D, Chen M, Khamis SM, Datta SS, Johnson AT, Misewich JA, Wong SS (2007) Functionalized carbon nanotubes for detecting viral proteins. Nano Lett. 7: 3086–3091.

Zhu D, Tan KS, Zhang X, Sun AY, Sun GY, Lee JC (2005) Hydrogen peroxide alters membrane and cytoskeleton properties and increases intercellular connections in astrocytes. J. Cell Sci. 118: 3695–3703.