

the potential to target delivery for selective destruction of certain types of cells, reducing the toxicity to nontargeted cells. It has also been observed that with decreasing pH, the loading of DOX reduces on SWCNTs. This is due to increased hydrophilicity and higher solubility of DOX at lower pH caused by increased protonation of $-NH_2$ groups on DOX. This results in reducing the hydrophobic interaction between DOX and SWCNT, and DOX is released. The pH dependence of binding/release of DOX could be profitably exploited for drug delivery applications. As the micro-environments of extracellular tissues of tumours and intracellular lysosomes and endosomes are acidic, the situation will potentially facilitate active drug release from SWCNT delivery vehicles. This method provides a novel, easy-to-make formulation of the SWCNT-DOX complex with extremely high drug-loading efficiency, which is remarkably higher than that reported for conventional liposomes and dendrimer drug carriers⁶.

Aqueous solution of functionalized SWCNTs on exposure to radiofrequency (RF) field experiences efficient heating and this property has been exploited by Gannon *et al.*⁷ for a noninvasive and selective thermal destruction of human cancer cells with minimal or no toxic

effects to normal cells. As 66% of the available SWCNTs are direct band-gap semiconductors, Gannon *et al.*⁷ assumed that exposure to RF field would lead to significant heat release by SWCNTs. Both *in vitro* and *in vivo* tests were carried out with cancer cells. In the *in vitro* test, three human cancer cell lines were incubated with various concentrations of SWCNTs and then treated in the RF field. After 48 h, tumours were harvested to assess viability. The authors observed CNT concentration-dependent cellular cytotoxicity *in vitro* in all three cancer cell lines after 2 min of exposure to the RF field. At a concentration of 500 mg/l of SWCNT, cytotoxicity essentially was 100% in all three cell lines. In another set of experiments, hepatic VX2 tumours in rabbits were injected with SWCNTs or with control solutions and were treated in the RF field. Rabbits bearing tumours tolerated the RF treatment⁷. After 48 h, all SWCNT-treated tumours demonstrated complete necrosis. However, tumours treated only with SWCNTs or RF field were viable. These observations indicate the thermal destruction of cancer cells⁷ in the presence of SWCNTs and exposure to RF field.

The two papers^{6,7}, one on supramolecular assembly of DOX on SWCNTs and the other employing SWCNTs and RF

field, demonstrate that carbon nanotubes are capable of leading to new exciting directions and approaches in therapeutic oncology.

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Adaptation of *Listeria monocytogenes* to low temperature and high salt

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Listeria monocytogenes, a Gram-positive bacterium and causative organism of listeriosis, a serious food-borne infection, is widely distributed in nature. Its ability to grow at low temperatures and high salt environment, makes it a potential risk factor in storage of food materials and also an attractive model for studies on bacterial adaptation to low temperature and high osmolarity.

A couple of years ago, using SCOTS (Selective Capture of Transcribed Sequences), a novel procedure for differential cloning of c-DNA, selective expression of RNAs in *L. monocytogenes* caused by a downshift of environmental temperature from 37°C to 10°C, was studied. The

RNAs, the steady-state level of which was elevated at low temperature, included those involved in synthesis of flagellar proteins, sigma factor Rpo N, histidine kinase (believed to be required for sensing environmental temperature), a transcriptional antiterminator, a repressor protein, some chaperone proteases, products related to the metabolism of some amino acids, enzymes required for some degradative metabolism and a novel fibronectin-binding protein. In addition to these, some genes unique to *L. monocytogenes* were also expressed at low temperature¹.

A recent study by Chassaing and Au-vray² reveals the role of another protein

in cold and salt tolerance of the bacterium. The investigators performed transposon mutagenesis of a strain of *L. monocytogenes* using the temperature-sensitive plasmid pTV32-OK containing the transposon Tn 917-*lac*. A total of 3720 random insertion mutants were screened for cold-sensitivity. A mutant that could grow at 37°C but not at 4°C in a microtiter plate was designated as cs 1. When cs 1 was checked for its ability to survive at 4°C over a period of 35 days with a culture of the wild-type strain maintained in the same condition as control, the mutant was found not only to survive but also to grow. However, its rate of growth was slower compared to

that of the wild-type strain. The mutant and the wild-type strain showed similar plating efficiencies on brain–heart infusion (BHI) agar at 8 and 37°C, but most of the colonies formed by *cs 1* at 8°C were smaller than those formed by the wild-type strain. The number of colonies formed by the mutant on BHI agar containing 3% sodium chloride at 37°C, was significantly lower than that formed by the wild-type strain. Hence it was evident that both cryotolerance and osmotolerance of the wild-type strain were adversely affected by transposon mutagenesis.

It was revealed by inverse PCR that Tn 917-*lac* was inserted between the putative promoter and ribosome-binding site of the gene *lmo 1078*. Sequence of the *lmo 1078* protein showed strong homology to bacterial UDP-glucose pyrophosphorylases. Following complementation of the mutant with a plasmid carrying a wild type copy of *lmo 1078*, the growth phenotype of the resultant strain was found to be similar to that of the wild type at low temperature, both in liquid

and solid medium. It was also able to grow in the presence of 3% sodium chloride, like the wild-type strain.

Hence expression of *lmo 1078* was adversely affected by transposon mutagenesis leading to cold-sensitivity and salt-sensitivity of the strain. The gene encodes a putative UDP-glucose pyrophosphorylase, an enzyme known to catalyse the formation of UDP-glucose. The cell-wall of Gram-positive bacteria contains a special structure made of teichoic acids, which helps in the attachment of peptidoglycan to lipids of the cytoplasmic membrane. The combined unit of lipid and teichoic acid is called lipoteichoic acid. A membrane glycolipid, diglucosyl-diacylglycerol, forms the predominant membrane anchor moiety of lipoteichoic acid. Biosynthesis of this glycolipid requires UDP-glucose. Thus, absence or shortage of UDP-glucose pyrophosphorylase may ultimately lead to destabilization of the architecture of bacterial cell wall and cell membrane. Further studies on complete membrane protein and membrane lipid

profile of *cs 1* vis-a-vis those of the wild-type strain are likely to reveal a more clear picture and also provide insight into the mechanism of bacterial adaptation to cold and salt stress. The fact that both cryotolerance and osmotolerance of the strain were adversely affected by a single mutation, gives credence to the postulation that mechanisms involved in bacterial tolerance to different types of stress conditions are interlinked³.

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