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# NTU team develops 'trojan horse' drug delivery system

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**Scientists from NTU (Nanyang Technological University) Singapore have developed a novel drug delivery method using protein-based microdroplets.**

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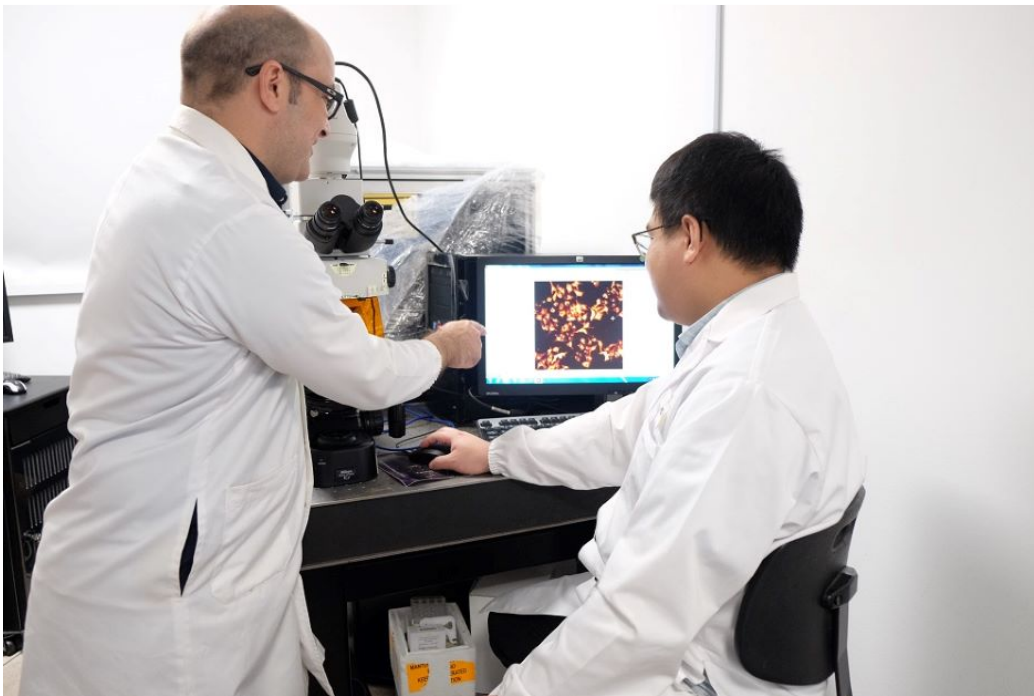
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*Image credit: NTU Singapore*

The discovery promises to be faster, safer, more effective, and better suited for gene therapy, cancer treatment and vaccine delivery – including mRNA-based vaccines such as those used for Covid-19 vaccinations.

Made up of small proteins named peptides, the microdroplets can encase large biomacromolecules that carry drugs inside them. In doing so, they allow these biological molecules to enter cells, something the molecules can't do alone.

Biomacromolecules are large biological molecules such as nucleic acids (DNA, mRNA), proteins and carbohydrates. They can carry large amounts of drugs, are non-toxic, can target specific sites and do not trigger the body's immune response, making them preferable to synthetic carriers currently used in drug delivery.

However, their large size and inability to pass through cell membrane have held them back from widespread clinical use.

Led by professor Ali Miserez from NTU's School of Materials Science & Engineering and School of Biological Sciences, the team's method of first encasing biomacromolecules in protein-based microdroplets was found to let them enter cells reliably and effectively.

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Researchers said they synthesised a peptide derived from squid beak to form the microdroplet due to its biological origin, high efficiency in storing molecules, and low toxicity. They were then able to entrap the biomacromolecules inside it through

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Similar to how oil and water can mix yet easily separate into two liquids, the LLPS process forms what is known as a coacervate that can merge into the cell membrane.

“Presumably, the liquid-like properties of coacervates achieved via the liquid-liquid phase separation process is critical in their ability to cross the cell membrane, although the precise entry mechanism is still unclear and currently under investigation,” said the paper’s first author, NTU PhD student Yue Sun.

The discovery allows biomacromolecules to avoid endocytosis – the process where cells allow foreign substances to enter by surrounding it with a protective membrane. Traditional drug delivery methods cannot cross into the cell membrane without first being caught by the cell and wrapped within a ‘bubble’ of cell membrane, or endosome.

Therefore, these types of drug packages must also be encoded with further instructions to ‘escape’ the endosome in order to efficiently release the drugs.

According to researchers, their coacervates can smoothly cross the cell membrane without triggering endocytosis. Once inside the cell, carrier droplets disintegrate and release the biomacromolecules.

“You can think about these droplets as molecular ‘Trojan Horses’: they trick the cells into letting them enter, and once inside, they deliver the biomacromolecular ‘soldiers’ that target the disease,” said Prof Miserez.

The team said they successfully delivered fluorescent proteins, commonly used to demonstrate efficiency of drug carriers, as well as the protein drug saporin through this method. They discovered that the cell entry process had a 99 per cent success rate compared to the 50-70 per cent of current commercially available carriers.

“Our peptide droplets can work as a universal delivery system without the need for individual adjustment. One delivery system for a whole range of proteins of various sizes, from big to small, and that can carry both positively- and negatively-charged proteins, is very appealing,” added Miserez.

Findings were published in *Nature Chemistry*. The team has filed two patents based on its study and is working to commercialise the drug delivery platform.

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