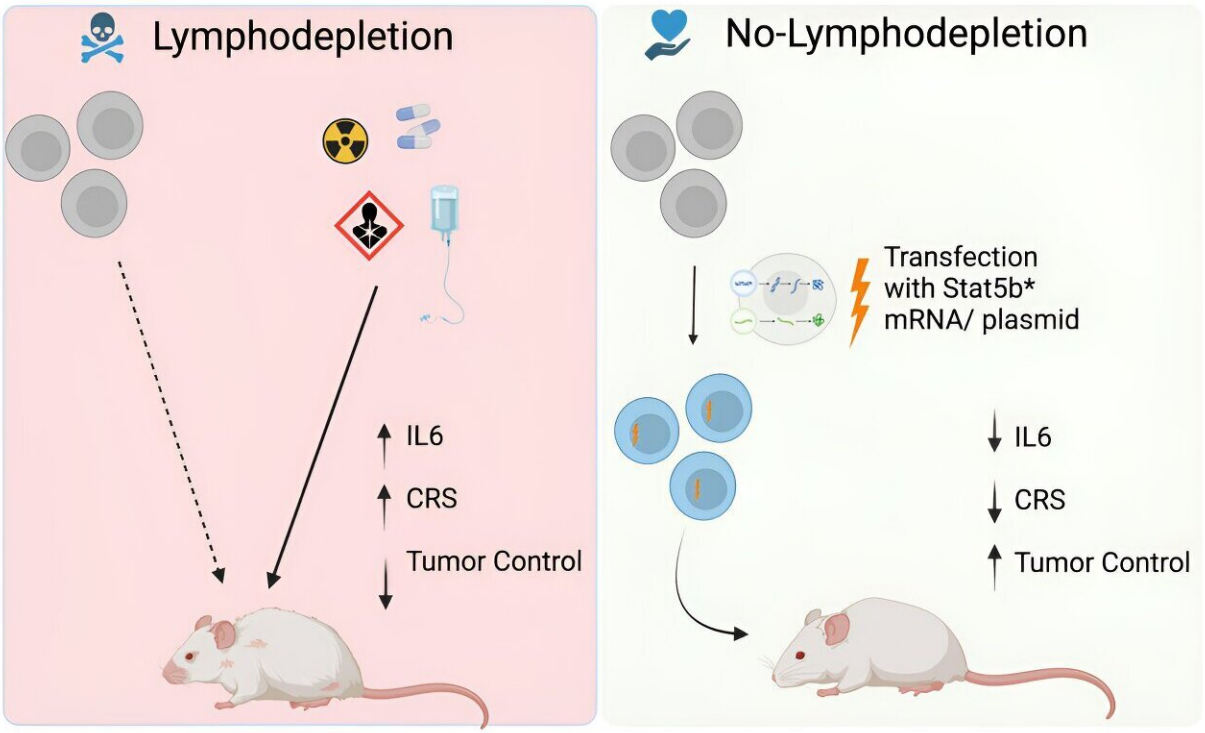
CAR T-cell therapy without side effects? Researchers show results in preclinical models

September 7 2023, by Leslie Cantu



Graphical abstract. Credit: *Molecular Therapy* (2023). DOI: 10.1016/j.ymthe.2023.07.015

When Richard O'Neil, Ph.D., joined MUSC Hollings Cancer Center two

years ago, he knew that he wanted to continue finding ways to make CAR T-cell therapy easier on patients.

What he didn't expect was that a side project—worked on by Megan Tennant, a graduate student in his lab, as a way to keep busy while a key piece of equipment was being serviced—would potentially open up this treatment beyond the world of cancer.

"I don't think that either of us expected that first initial experiment to work," Tennant said. "But when we saw how well it worked and really started to conceptualize where this could go and how important this could be, it was exciting."

O'Neil said they've begun conversations with biotechnology companies about how to push forward their findings.

"Right now, we're trying to out-license the technology," O'Neil added. "The most interest, actually, has come in the context of lupus."

CAR T-cell therapy is currently used to treat some types of blood cancers that have returned after treatment or that haven't responded to chemotherapy. It is both expensive and extensive—some of a patient's T-cells are removed and sent to a lab, where in a process that can take several weeks, they're engineered to add chimeric antigen receptors (CAR) that are tuned to home in on specific proteins on the surface of cancer cells. The newly formed CAR T-cells are then reinfused into the patient to attack the cancer.

Some patients have experienced incredible recoveries after CAR T-cell therapy. But they've also endured incredibly strong and scary side effects, in part because of the lymphodepleting chemotherapy that is performed before the CAR T-cells are reinfused.

Lymphodepleting chemotherapy kills off existing T-cells to create a blank slate for the CAR T-cells, and research has shown that the therapy is more effective after lymphodepleting chemotherapy.

But O'Neil and Tennant, along with colleagues Christina New, M.D., and Leonardo Ferreira, Ph.D., believe that lymphodepleting chemotherapy isn't necessary.

In a paper published in *Molecular Therapy*, they showed that encoding the CAR T-cells with instructions to create a hyperactive form of protein STAT5 prompted the CAR T-cells to engraft, or take root and begin multiplying, without requiring lymphodepletion. In their experiments, the engraftment process was cell autonomous, meaning it didn't depend on the surrounding environment but happened solely because of the instructions from within the cells.

"We present a lot of evidence in the paper to support that notion that it's a completely cell autonomous process, and that it's fundamentally driven by activation of STAT5," O'Neil said. "And so by transiently activating STAT5 during that phase of adoptive transfer, the initial engraftment phase, you're tricking the cells into essentially thinking they're going into a lymphodepleted environment.

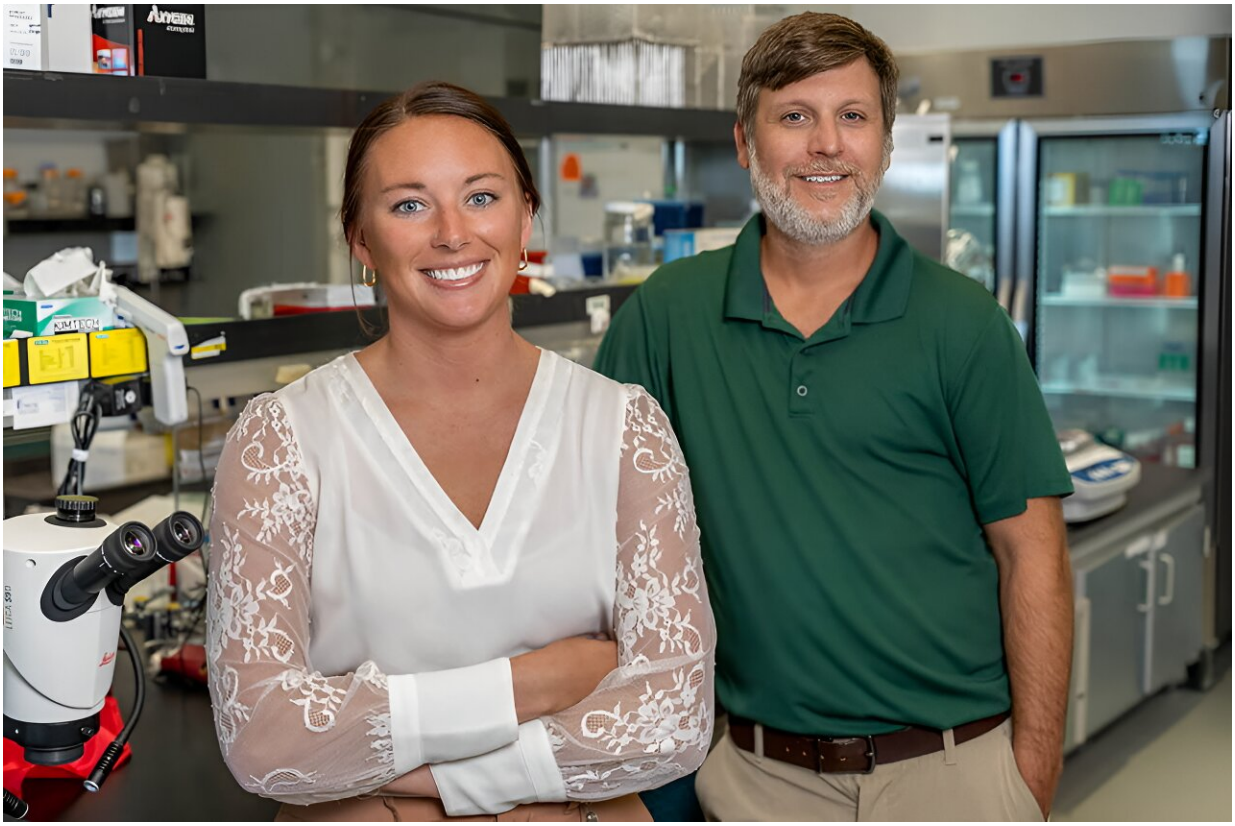
"And so they engraft and become very functional, and they do everything they're supposed to do," he continued. "Once we saw that they'd engraft, we just benchmarked how they functioned with as many different benchmarking assays as we could, comparing it to conventional adoptive transfer using lymphodepletion as our benchmark."

O'Neil said they came to STAT5 because of its role in the cytokine signaling pathway. It's been known for some time that the interleukin cytokines IL2, IL7 and IL15 are instrumental to the engraftment process. Some have suggested injecting patients with IL2 or IL15 in place of

lymphodepleting chemotherapy, he said. But each of those cytokine pathways requires STAT5.

"We reasoned that we could recapitulate that entire signaling process at the node of STAT5, rather than trying to tickle it up at IL 15, or IL2 receptors. And by doing that, we also have more of a cell autonomous effect where we don't have to expose the patient to a bunch of IL7, IL15 and IL2, which can be dangerous," O'Neil said.

The team used messenger RNA transfection to implant the instructions within the CAR T-cells for activating STAT5.



Graduate student Megan Tennant and Richard O'Neil, Ph.D., have promising preclinical results of a new way to administer CAR-T-cell therapy that could mean fewer side effects and the possibility of treating more disease types.

Credit: Clif Rhodes

In preclinical models, they found that their method reduced cytokine release syndrome, sometimes called cytokine storm, one of the most serious side effects. The CAR T-cells carrying super STAT5 also got the cancer under control and appeared to create memory cells trained on that cancer.

O'Neil said that eliminating the need for lymphodepleting chemotherapy could mean that CAR T-cell therapy would become viable for more types of diseases. The balance of harms and benefits of lymphodepleting chemotherapy is different for someone facing a recurrent lymphoma that no longer responds to chemotherapy than for someone with a chronic condition like lupus.

"You might be able to treat some of these more serious lupus cases," O'Neil explained. "Doctors would never want to lymphodeplete somebody with lupus with the combined chemotherapies fludarabine and cyclophosphamide, but if we don't have to lymphodeplete them anymore, then you can imagine trying out this therapy with them."

Eliminating lymphodepletion could also change dosing schedules.

"Now you can imagine re-administrating dose after dose to the patient instead of 'one and done.' If you don't have to lymphodeplete them, you might be able to just give them an injection every month," he said.

O'Neil is working with the MUSC lupus erythematosus research group on potentially developing clinical trials. He's also talking to biotechnology companies that work in the CAR T-cell realm and have the ability to launch new clinical trials.

He praised Tennant, in her last year of graduate school, for her diligence in pursuing the project.

"This has been Megan's labor of love," he said. "It's actually been pretty remarkable. She just joined my lab when I came here two years ago, so this entire project came from conception to fruition in two years, which is pretty amazing and definitely shows what a talented and hard-working scientist Megan is."

More information: Megan D. Tennant et al, Efficient T cell adoptive transfer in lymphoreplete hosts mediated by transient activation of Stat5 signaling, *Molecular Therapy* (2023). DOI: 10.1016/j.ymthe.2023.07.015

Provided by Medical University of South Carolina