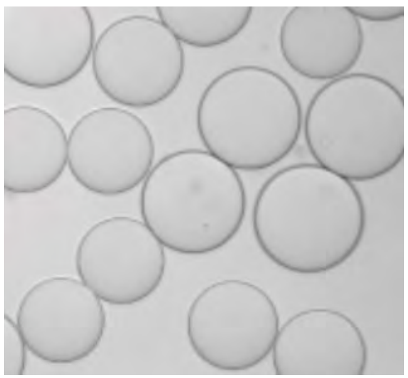
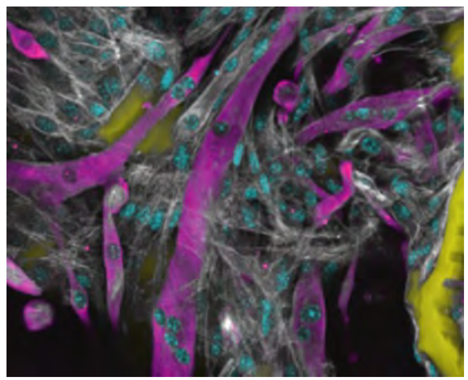
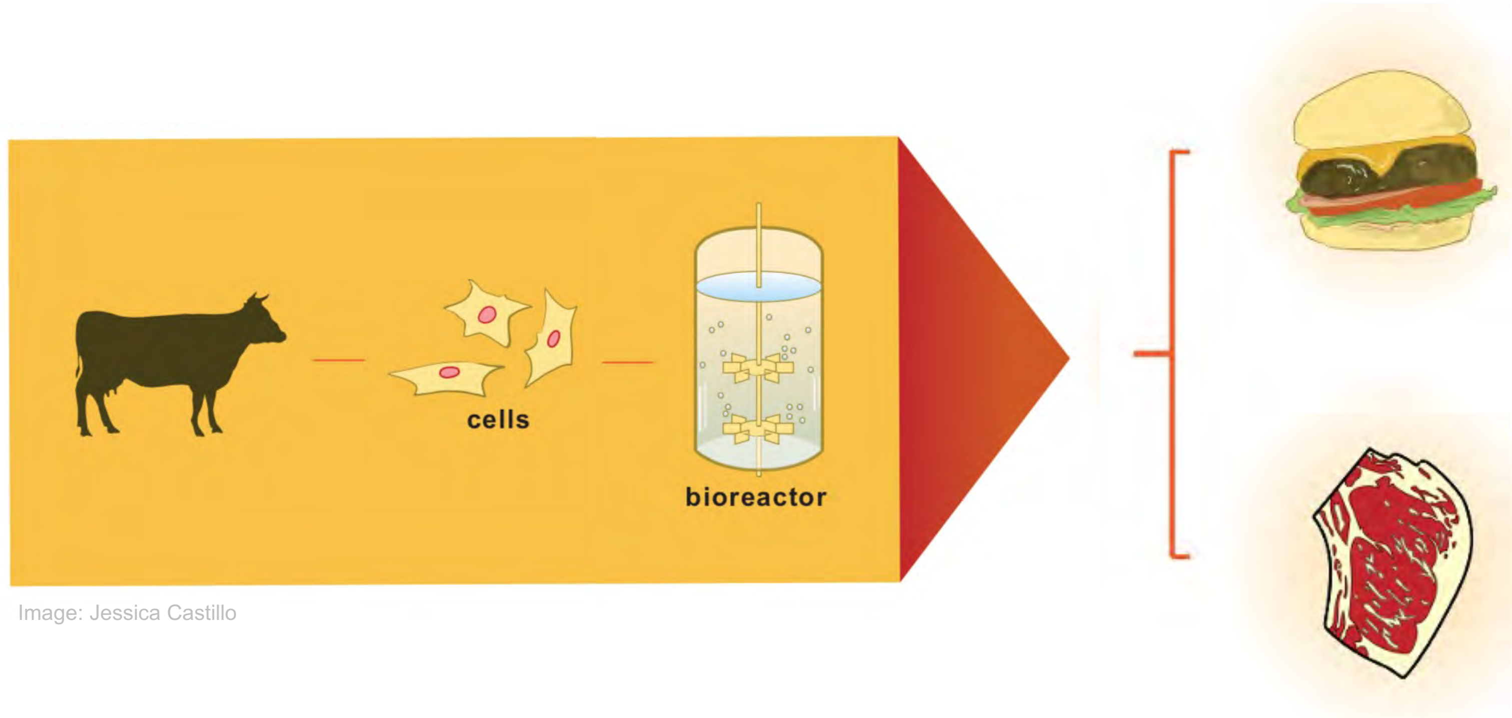
Towards marbled cultured meat that can be scaled for food production


Amy Rowat
Marcie H. Rothman Presidential Chair of Food Studies
Integrative Biology and Physiology, UCLA

SEMEL HCI Center at UCLA
UCLA Samueli Bioengineering
scienceandfood
UCLA Integrative Biology & Physiology
UCLA
CNSI at UCLA
Jonsson Cancer Center Foundation
UCLA Broad Stem Cell Research Center

What is cultured meat



Tomiyama…Rowat (2020) *Trends Food Sci Tech*

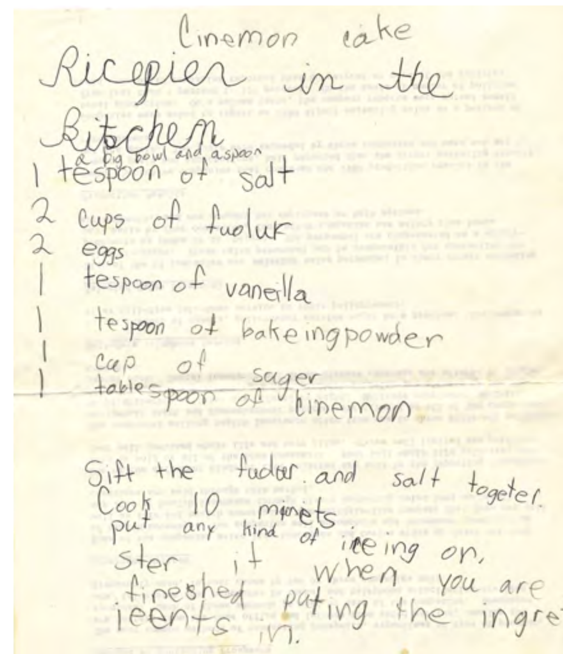
Roadmap for today's talk

CONTEXT	My academic background
	How my lab thinks (about cells and foods as materials)

CULTURED MEAT	The emergence of cultured meat
	Our research: 1. Marbled cultured meat 2. Culturing meat with a scalable process
	Future outlook

My first experiments in biophysics

My journey

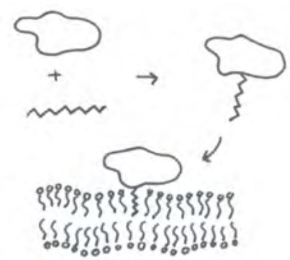


Cinemon cake
Ricepier in the
Kitchen
a big bowl and a spoon
1 tespoon of salt
2 cups of fuolur
2 eggs
1 tespoon of vaneilla
1 tespoon of bakeingpowder
1 cup of suger
1 tablespoon of cinemon

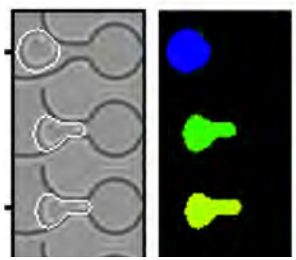
Sift the fudur and salt togeter.
Cook 10 minets.
put any kind of iceing on.
ster it when you are fineshed pating the ingret ieents in.

Photos: N. Rowat

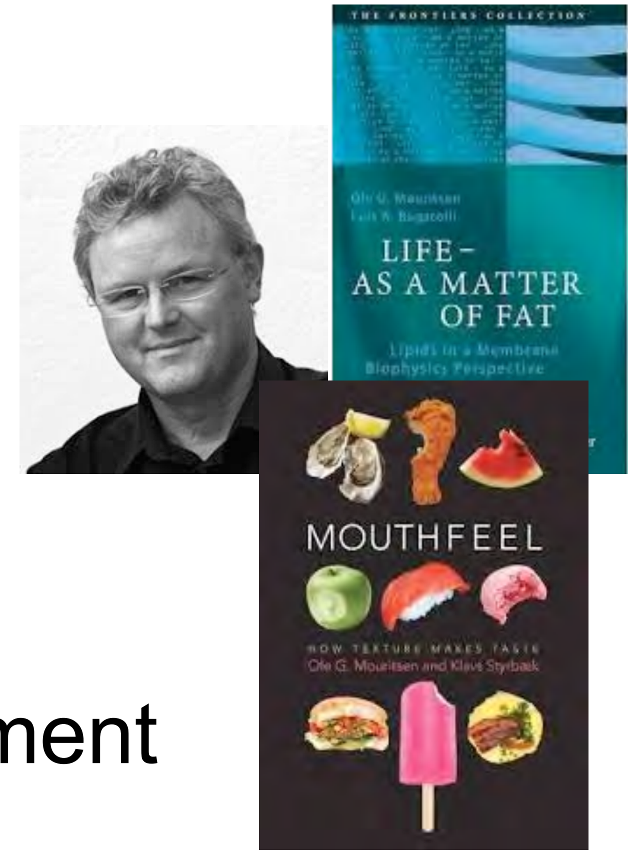
My academic background



- Graduate school:
Biomembrane physics



- Postdoctoral training:
Cell mechanics and biotechnology development



At Harvard, the Kitchen as Lab

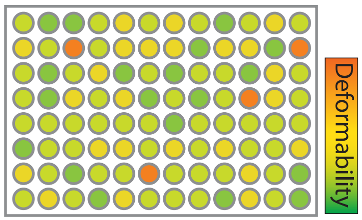
New York Times, Oct 2010. Photo: Michele McDonald for The New York Times.

My journey

How we think about cells as materials

Our approach

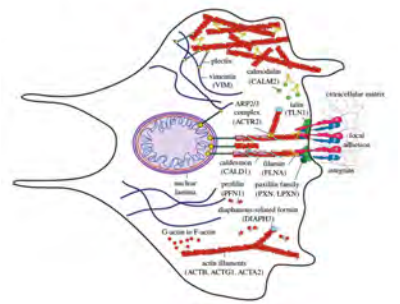
MEASURE: Build new technologies to study cells as materials



Qi...Rowat (2015) *Sci Reports;*
Gill…Rowat (2019) *Lab Chip*

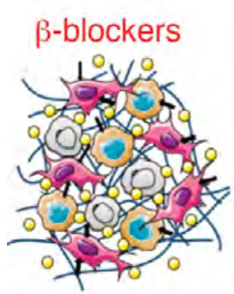
Nyberg…Rowat (2016) *Lab Chip*
Nyberg…Rowat (2017) *Biophys J*

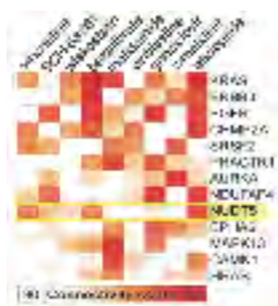
UNDERSTAND: Define mechanisms of how cells regulate mechanical behaviors



Chan…Rowat (2016) *Open Biology;* Kim et al (2016) *J Cell Sci;* Sobreiro et al (2018) *Cancer Res;* Lawrenson et al (2019) *Cell Reports;* Yokota et al (2020) *Cell;* Moose et al (2020) *Cell Reports*

TRANSLATE: Develop more effective cancer treatments





Chavez…Rowat, *Manuscript in preparation;*
Flores…Rowat, *Manuscript in preparation*

THE ROWAT LAB
UNIVERSITY OF CALIFORNIA LOS ANGELES

Engaging students and general audiences in science using food

1) Teaching students science & engineering through food

- Undergraduate class **PHYSCI7**: scienceandfood with hands-on experiences through food and cooking

2) Food-based resources and curricula for science

- Peer-reviewed resources for educators
- Food-based curricula for high school students that makes science & engineering more approachable

Zhou et al (2015) *Adv Physiol Edu*
Soh et al (2015) *J Coll Sci Teaching*
Rowat et al (2014) *Phys Edu*
Rowat et al (2011) *J Chem Edu*
Rowat et al (2010) *J Food Sci Edu*

3) Engaging general audiences in dialogue with scientists & chefs through food (scienceandfood.org)

- Public events on timely food issues (food waste, growing food in space)
- Educational blog managed by graduate and undergraduate students

Media Partners:

Discover MAGAZINE
Los Angeles Times FOOD BOWL

Featured in:

LA Times
ABC AUSTRALIA

Our approach

Why cultured meat as a complement

Our approach

Plant-based meat

Regenerative agriculture

Cellular agriculture

- Excitement for plant-based meat; but still demand for deliciousness

- Need meat production methods that:
  - support human and planetary health
  - build resiliency in food supply chains

> *Vision*: Delicious, nutrient-rich food that is accessible for all

How we think about cells as materials

Our approach

MEASURE: Build new technologies to study cells as materials

Qi...Rowat (2015) *Sci Reports;* Gill…Rowat (2019) *Lab Chip*

Nyberg…Rowat (2016) *Lab Chip*
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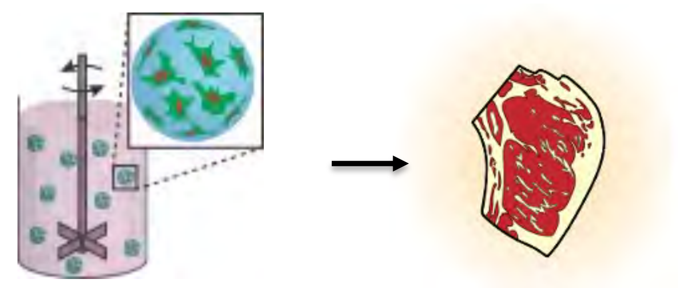
TRANSLATE: Develop more effective cancer treatments

Chavez…Rowat, *Manuscript in preparation;*
Flores…Rowat, *Manuscript in preparation*

TRANSLATE: Develop efficient and scalable processes for tissue culture



Tomiyama…Rowat (2020) *Trends Food Sci Tech*;
Norris…Rowat (2022) *Biomaterials*

THE ROWAT LAB
UNIVERSITY OF CALIFORNIA LOS ANGELES

Roadmap for today's talk

CONTEXT	My academic journey
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	Future outlook

Cultured meat technologies are rapidly evolving

Emergence of cultured meat

WIRED

Lab-Grown Meat Is Coming, Whether You Like It or Not

Soon enough, burgers will grow not just in fields but in vats. If the sound of that bothers you, know that you're not alone.

Feb 16, 2018

nature

NEWS | 06 February 2019

Sizzling interest in lab-grown meat belies lack of basic research

'Clean meat' firms have drawn tens of millions of dollars in investment in recent years, but technical hurdles remain.

Los Angeles Times

Prepare yourself for an avalanche of fake meat

Oct 21, 2021

The New York Times

Could This Be the Lab-Made Dinner Party of Our Future?

Feb 21, 2021

OUTLOOK | 09 December 2020

Will cell-based meat ever be a dinner staple?

Laboratory-grown meat has been stuck in the experimental stage. For it to become a commercially viable industry, tissue needs to be grown efficiently at scale.

Disclosures: I am a former Scientific Advisory Board of Beyond Meat and hold options in the company. I am currently a Scientific Advisory Board member of Orbillion Bio and exploring translational potential for a patent (WO2020219755A1) describing technologies developed in my laboratory.

Increasing support to develop cultured meat

- >100 companies dedicated to producing cultivated meat
- Jan 2021 — U.S. Government Invests in Lab-Grown Meat Research for The First Time
- Oct 2021 — Tufts Receives $10 Million Grant to Help Develop Cultivated Meat
- July 2022 — California just invested millions in lab-grown meat, becoming the first state to back the unproven industry
- Sept 2022 — Executive Order on Advancing Biotechnology and Biomanufacturing Innovation for a Sustainable, Safe, and Secure American Bioeconomy

Disclosures: I am a former Scientific Advisory Boa ... any.
I am currently a Scientific Advisory Board member of Orbillion Bio.

Why can't we just culture cells for food?

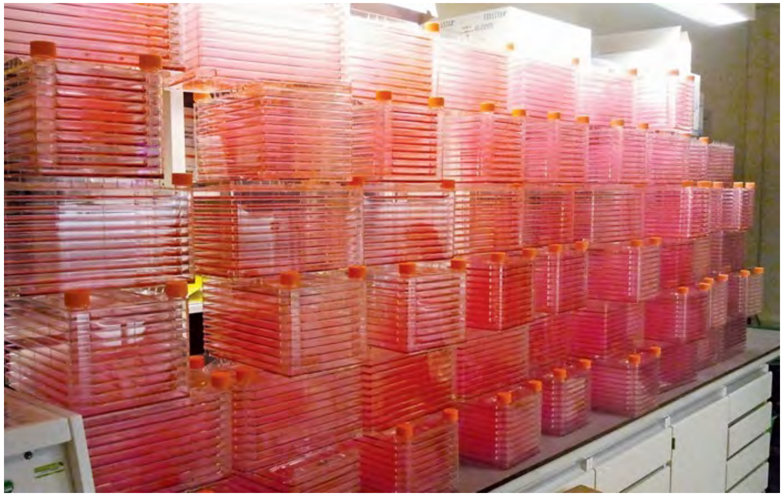
"With a greater knowledge of what are called hormones, i.e. the chemical messengers in our blood, it will be possible to control growth. We shall ***escape the absurdity of growing a whole chicken*** *in order to eat the breast or wing, by growing these parts separately under a suitable medium."*

- Winston Churchill: *'Fifty Years Hence'* in Strand Magazine, December 1931

Emergence of cultured meat

Image: Smithsonian Magazine

Why can't we just culture cells for food?



Typical eukaryotic cell ~ 3.5×10^{-9} g

One 10 cm dish = 10^6 cells ~ 1 mg

↓ ?

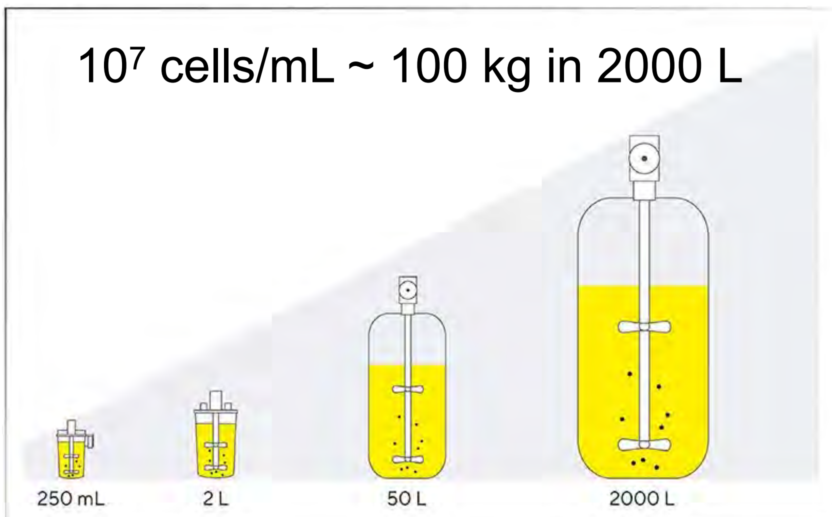
→ ?

Why can’t we just culture cells for food?

Typical eukaryotic cell ~ 3.5×10^{-9} g

One 10 cm dish = 10^6 cells ~ 1 mg

?



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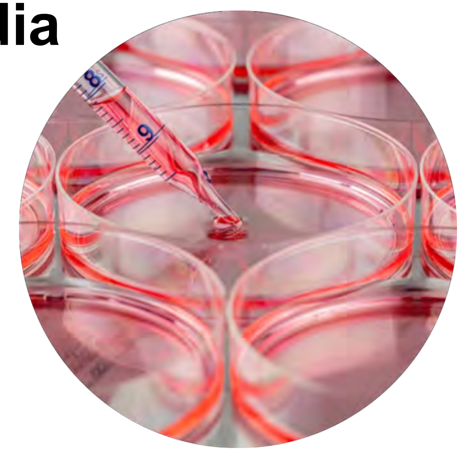
Emergence of cultured meat

Emergence of cultured meat

Challenges in culturing meat

Scale up

Media

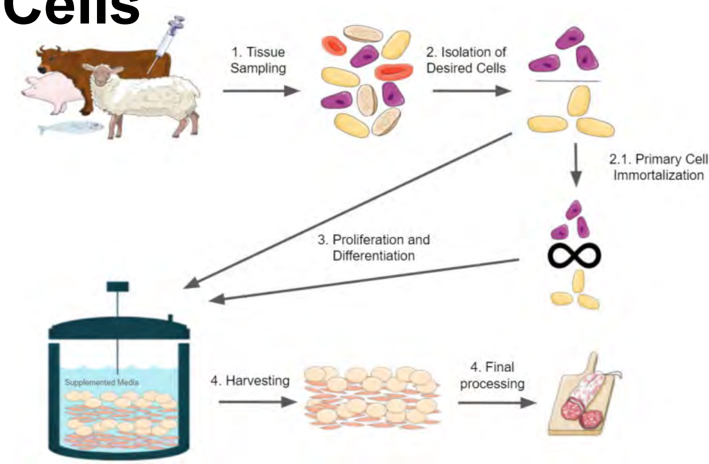


Messmer et al (2022) *Nature Food;* Stout et al (2022) *Comm Biol;* Cosenza et al (2022) *Biotech Bioeng*

Structure & Texture

Zagury et al (2022) *Comm Biol;* Kang et al (2021) *Nat Comm;* Furuhashi et al (2021) *NPJ Sci Food*

Cells



Soice & Johnston (2021) Int J Mol Sci; Dohmen et al (2022) *NPJ Sci Food*

Challenges in culturing meat

Scale up

Media

Messmer et al (2022) *Nature Food;* Stout et al (2022) *Comm Biol;* Cosenza et al (2022) *Biotech Bioeng*

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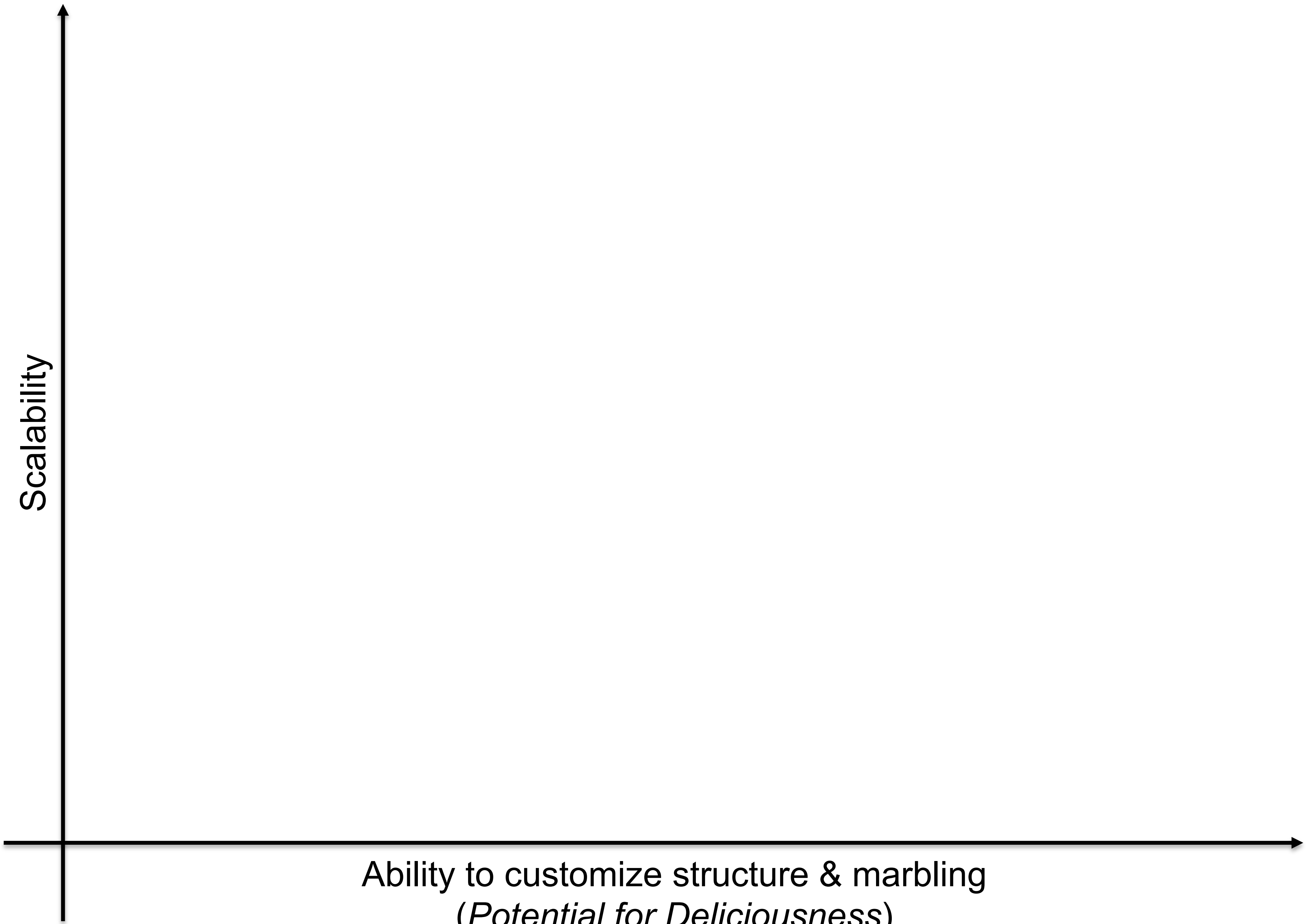
Cells

Soice & Johnston (2021) Int J Mol Sci; Dohmen et al (2022) *NPJ Sci Food*

Approaches in culturing meat

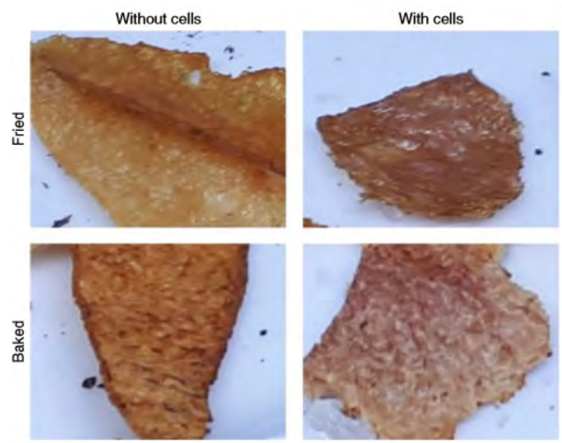


Our approach

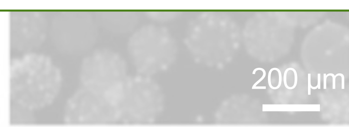
Approaches in culturing meat

Scalability

Natural material scaffolds



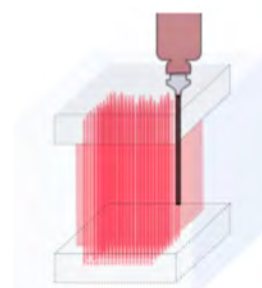
Ben-Arye et al (2020) *Nature Food*

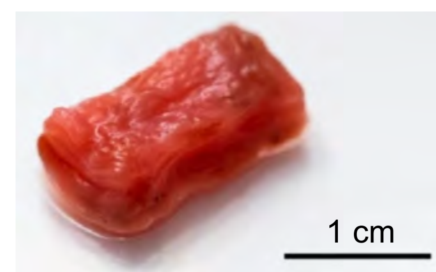


Verbruggen et al (2018) *Cytotechnology*

?

Bioprinting





Kang et al (2021) *Nature Commun;*
Furuhashi et al (2021) *NPJ Science Food*

Ability to customize structure & marbling
(*Potential for Deliciousness*)

Our approach

Our approach

How can we grow cultured meat that is delicious and cost-effective?

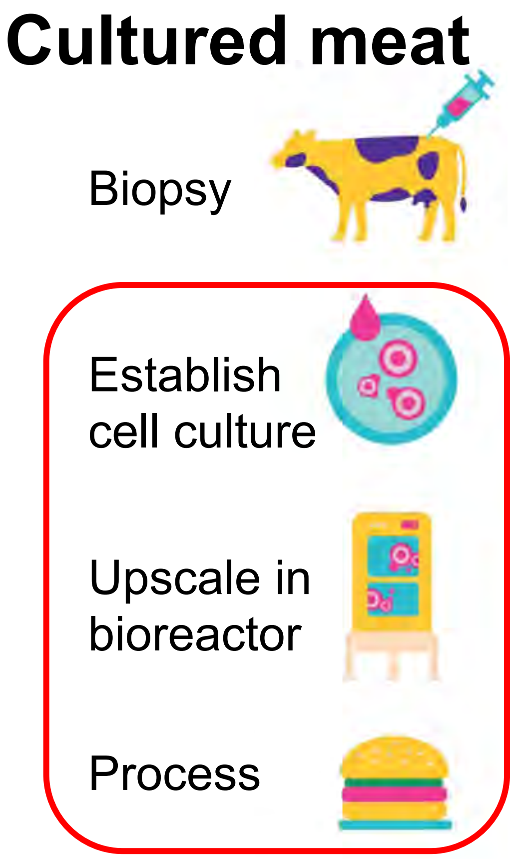
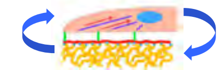
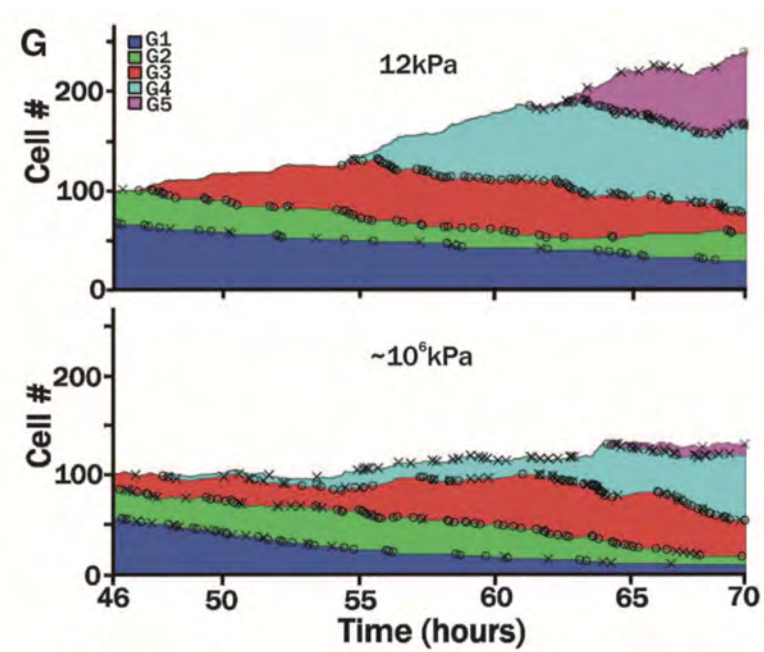


Image: New Scientist

Mechanical cues regulate cell behaviors that are important for culturing meat

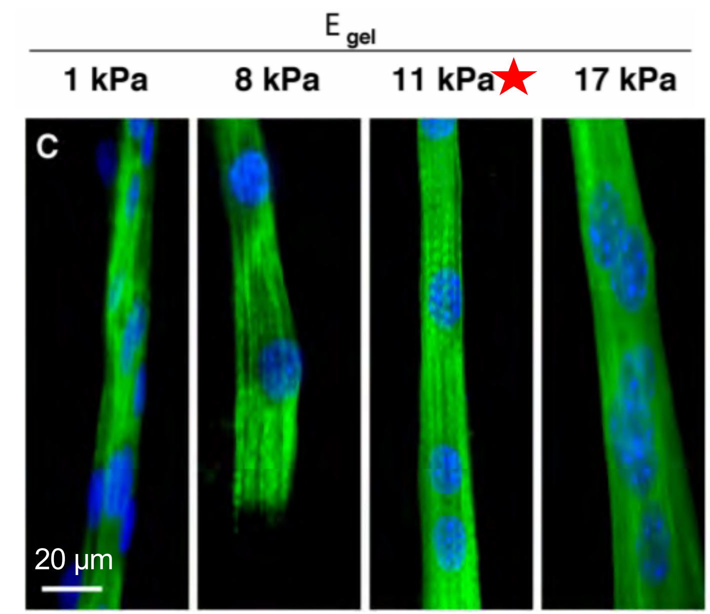


Muscle stem cell expansion



Gilbert et al (2010) *Science*

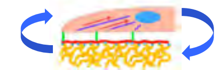
Myoblast differentiation



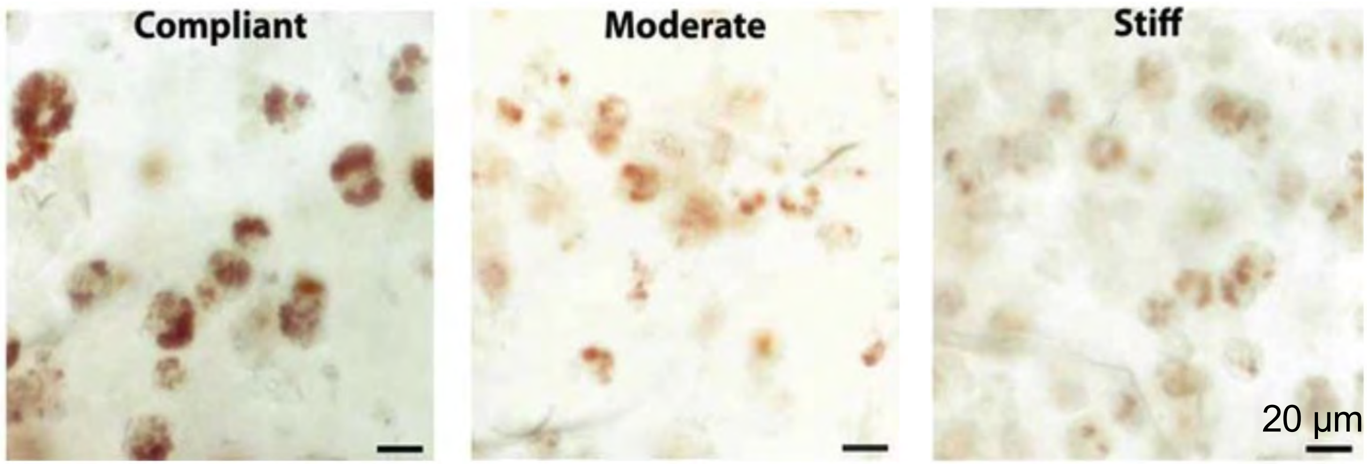
Engler et al (2004) *J Cell Biol*

Our approach

Mechanical cues regulate cell behaviors that are important for culturing meat



Adipogenesis



3.3 kPa | **7.9 kPa** | **12.4 kPa**

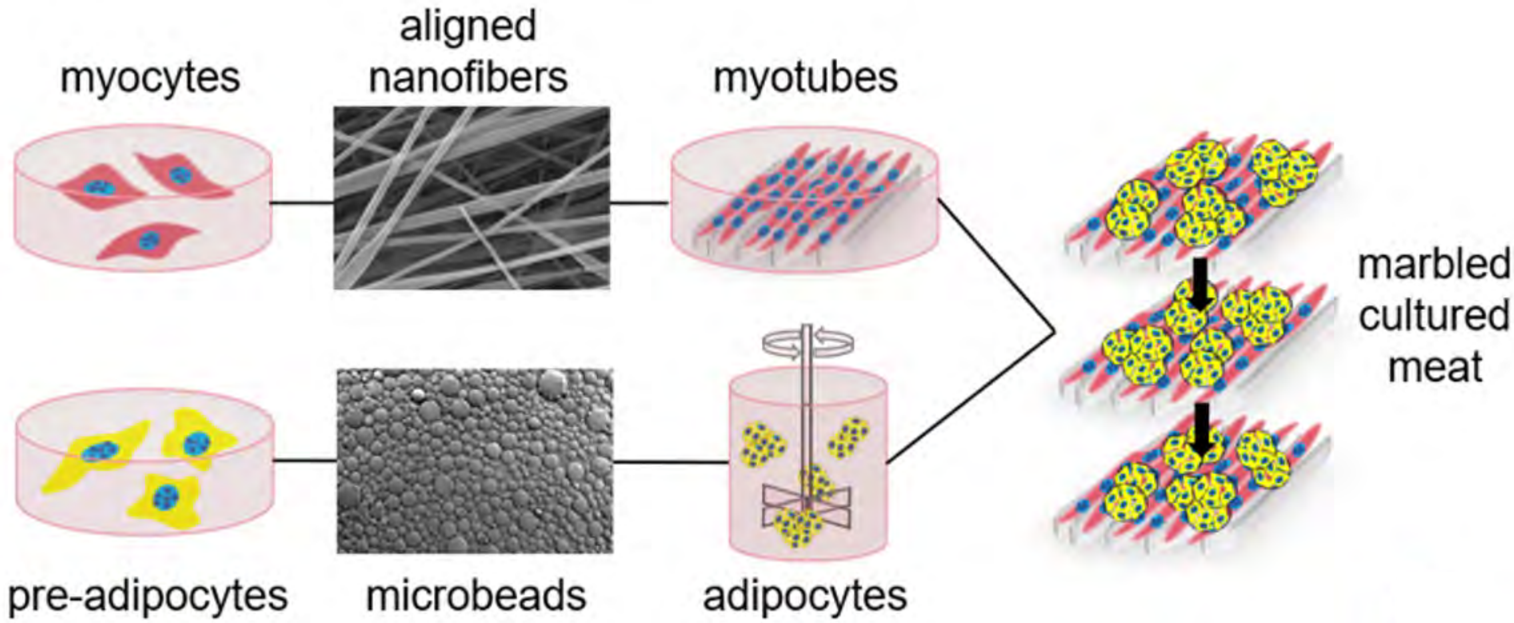
- 3T3-L1 cells with Oil Red O within alginate hydrogels

↑ Optimal stiffness for myotube differentiation (12.4 kPa)

Chandler et al (2011) *Biotech Bioeng*

Our approach

Building tissue constructs with muscle and fat



Stephanie
Kawecki

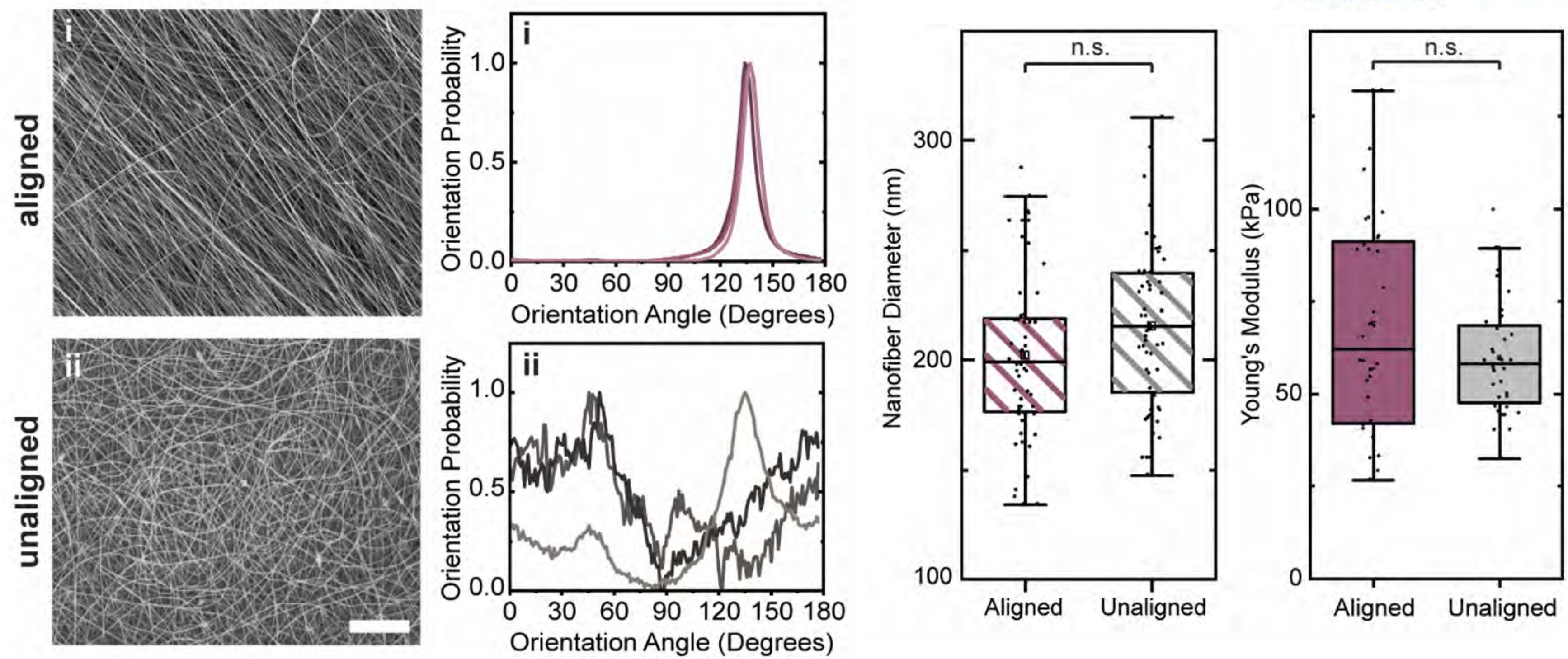
Rowat, Kawecki, et al, Patent Pending WO 2020/219755

Marbled cultured meat

Scaffolds to support muscle cell culture

- Generate scaffolds with aligned nanofibers using electrospinning

CNSI AT UCLA
Nano and Pico Characterization (NPC) Laboratory
Electron Imaging Center for Nanomachines

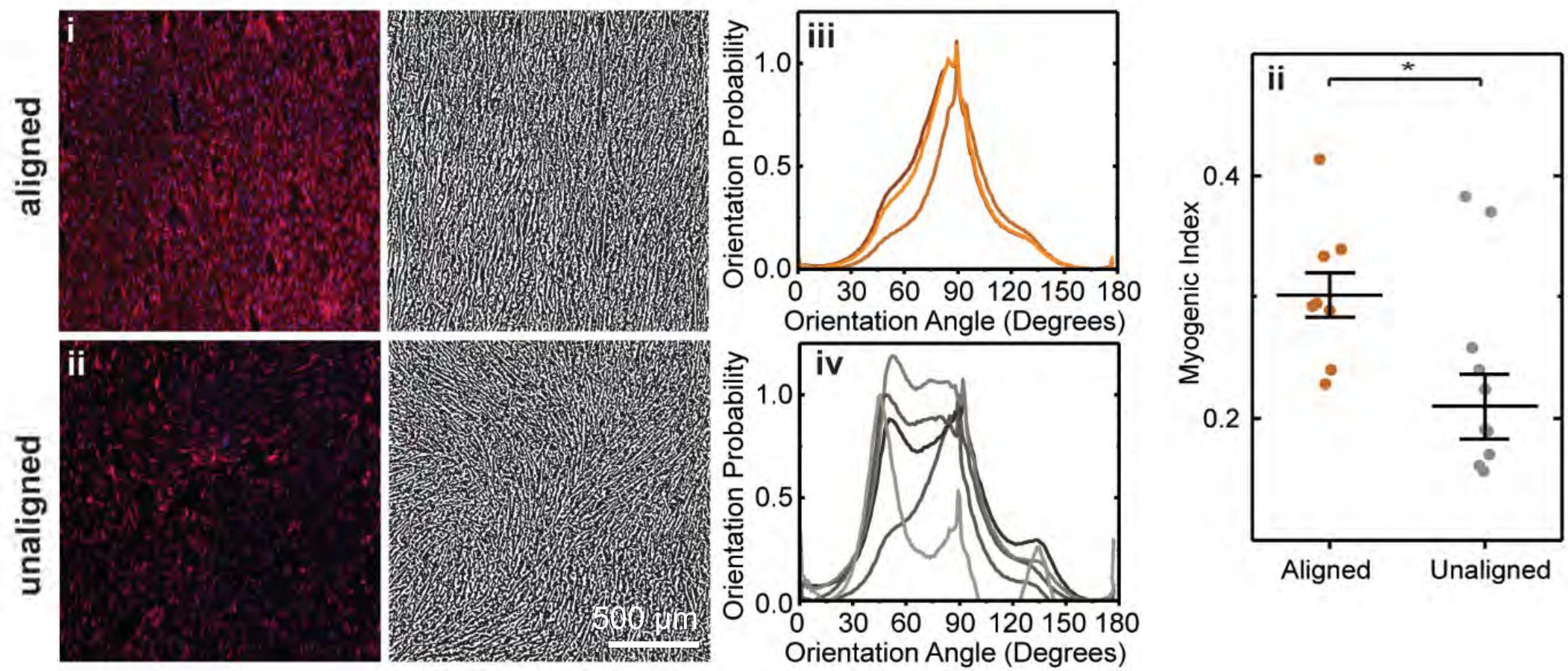


Kawecki et al, *Manuscript in preparation*

Scaffolds to support muscle cell culture

- Scaffolds with aligned nanofibers promote myotube formation in primary rabbit skeletal myocytes

Primary Rabbit Skeletal Myocytes



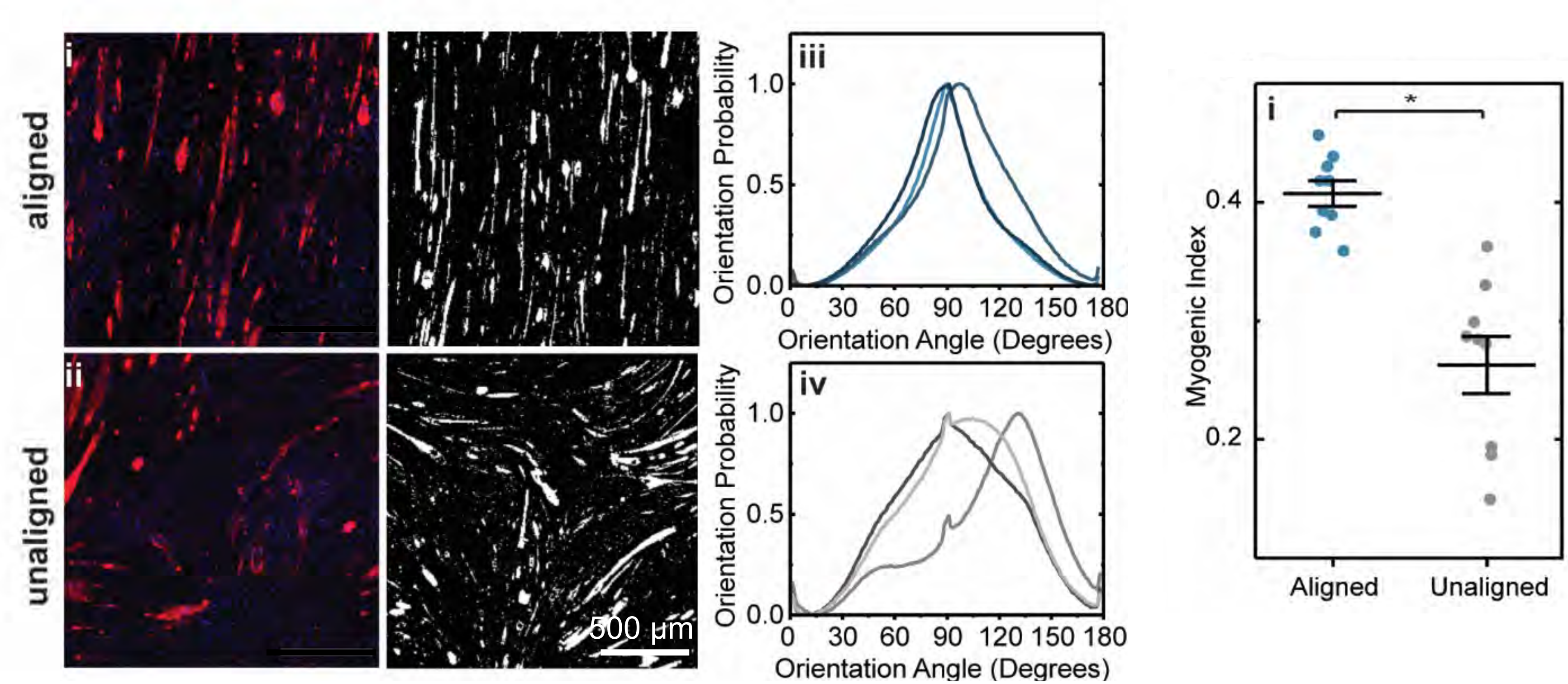
Red: Myosin Heavy Chain (Myh4)

Kawecki et al, *Manuscript in preparation*

Scaffolds to support muscle cell culture

- Scaffolds with aligned nanofibers promote myotube formation in mouse myocytes

Mouse Myocytes (C2C12)



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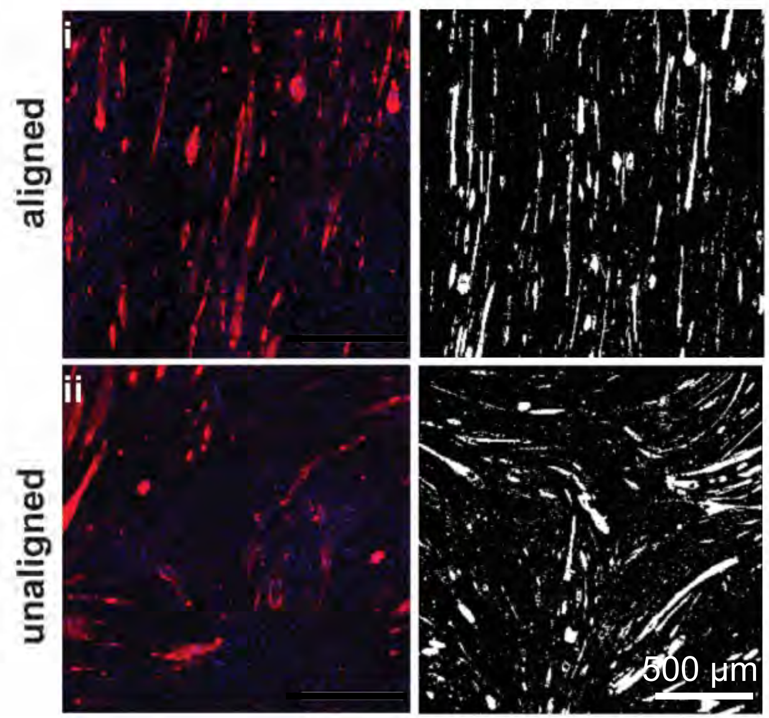
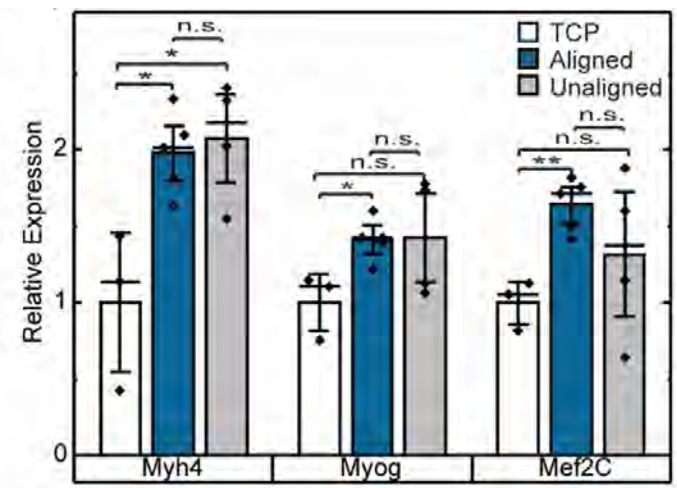


Red: Myosin Heavy Chain (Myh4)

mRNA levels of myogenic markers are higher than on tissue culture plastic (TCP)

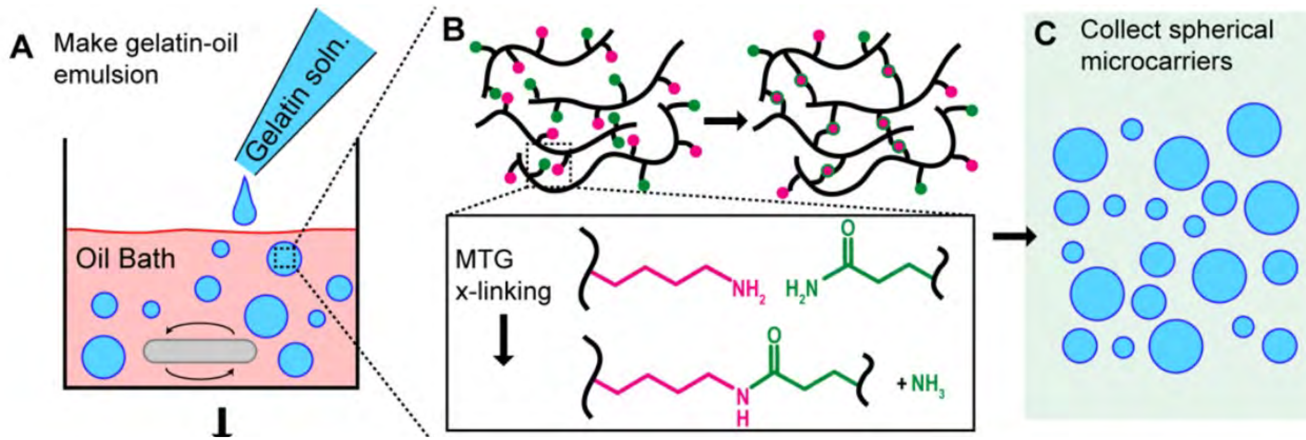


Myosin heavy chain (Myh4), Myogenin (Myog), and Myocyte Enhancer Factor 2C (Mef2C)

Kawecki et al, *Manuscript in preparation*

Microcarrier scaffolds to support adipocyte culture

- Generate compliant microcarrier scaffolds using emulsion droplets

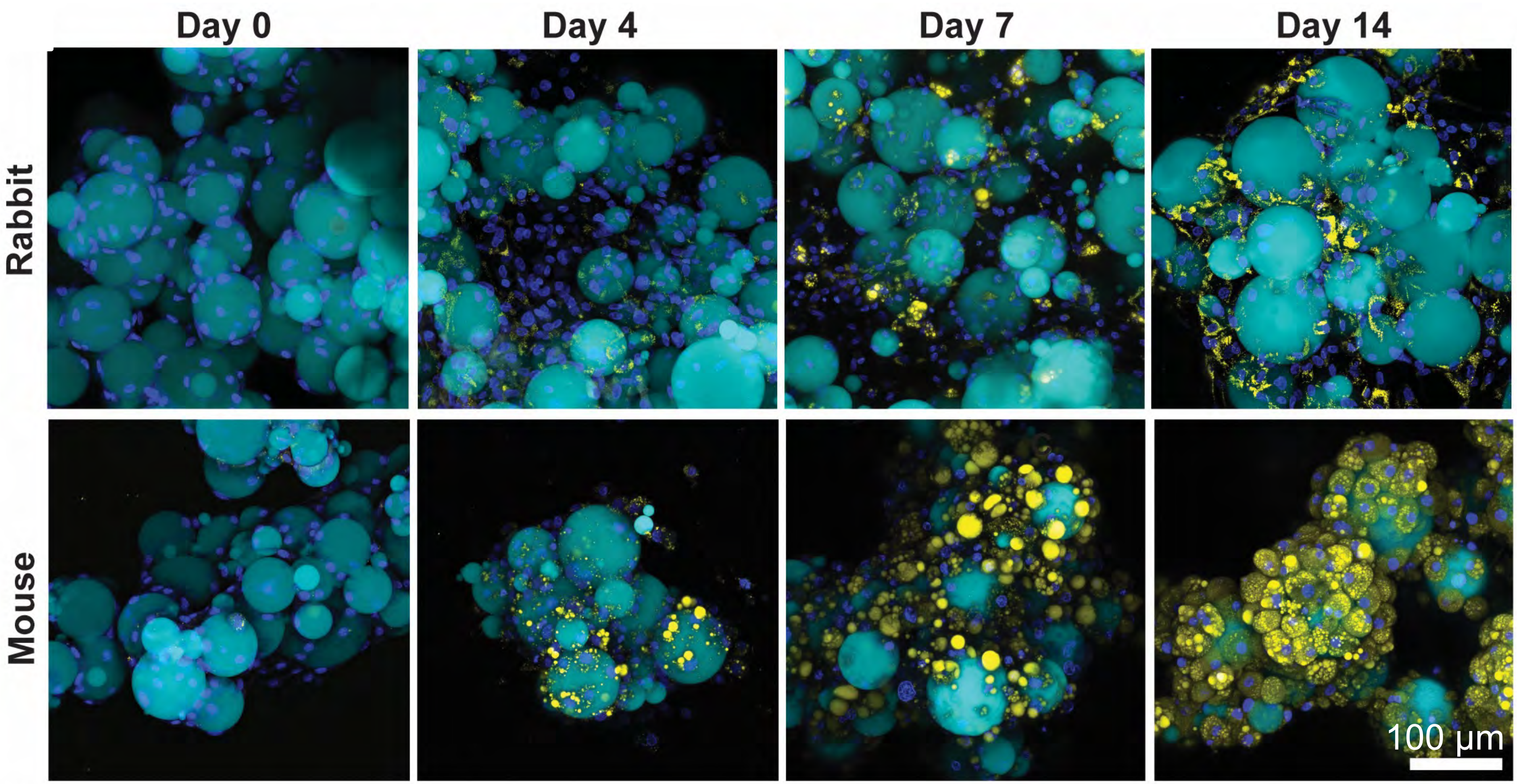


Marbled cultured meat

Kawecki et al, *Manuscript in preparation*

Microcarrier scaffolds support adipogenesis

- Lipid accumulation in primary rabbit and mouse 3T3-L1 adipocytes



Yellow: Intracellular lipids (Nile Red); Cyan: Microcarriers; Blue: DNA (Hoechst)

Marbled cultured meat

Kawecki et al, *Manuscript in preparation*

Microcarrier scaffolds support adipogenesis

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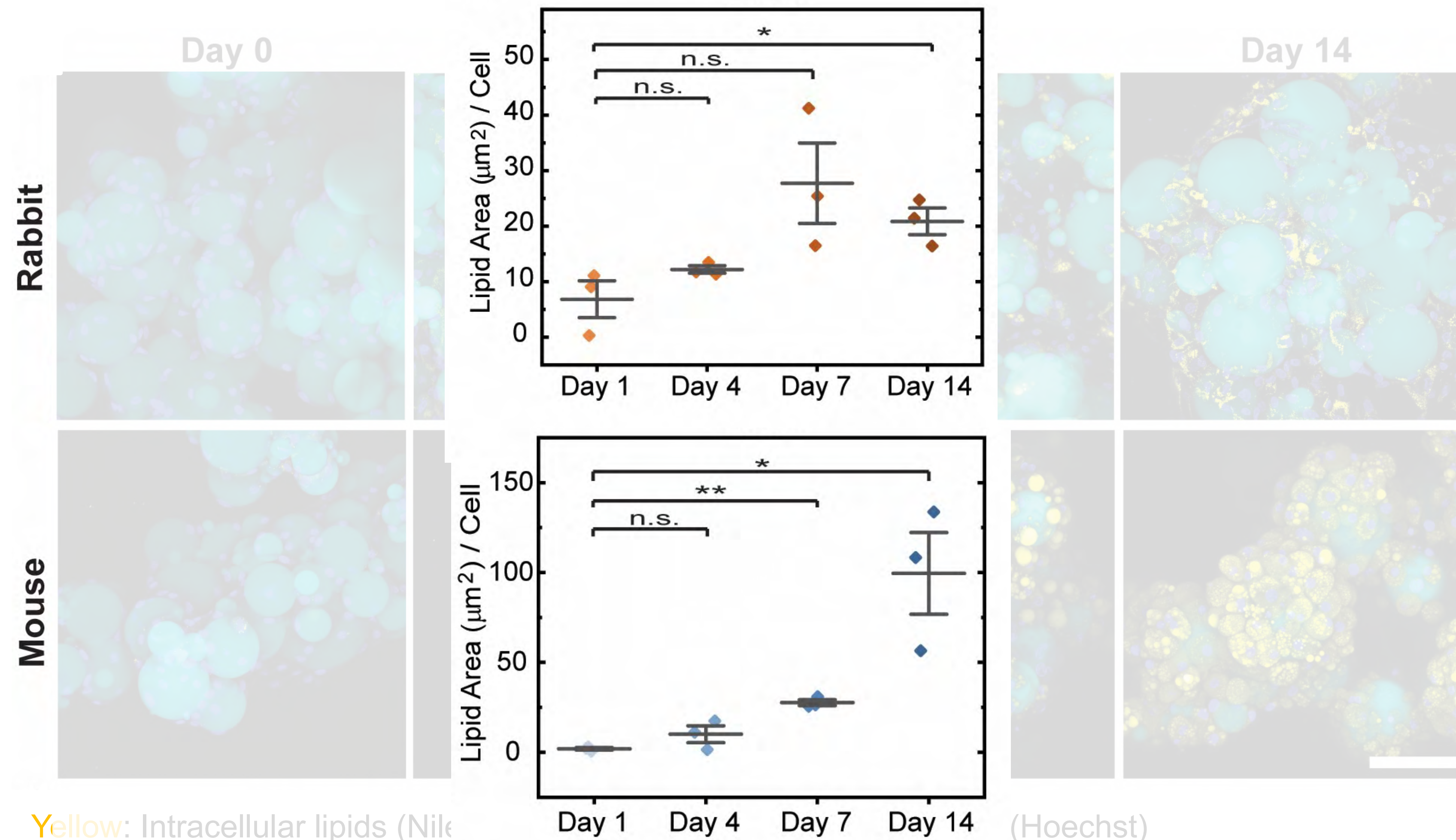


Yellow: Intracellular lipids (Nile … (Hoechst)

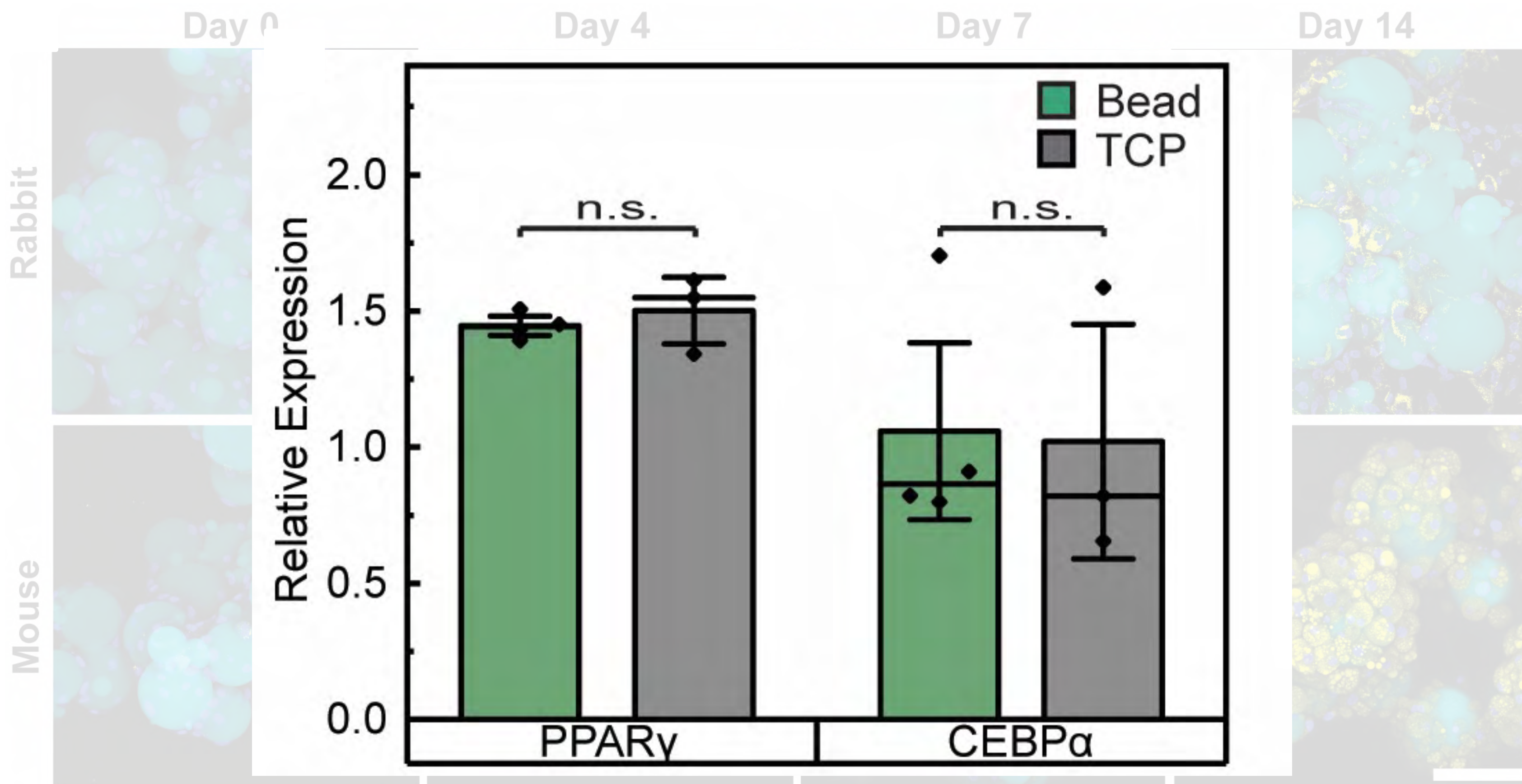
Kawecki et al, *Manuscript in preparation*

Marbled cultured meat

Marbled cultured meat

Microcarrier scaffolds support adipogenesis

- Increased expression of adipogenic markers in 3T3-L1 cells similar to tissue culture plastic (TCP)

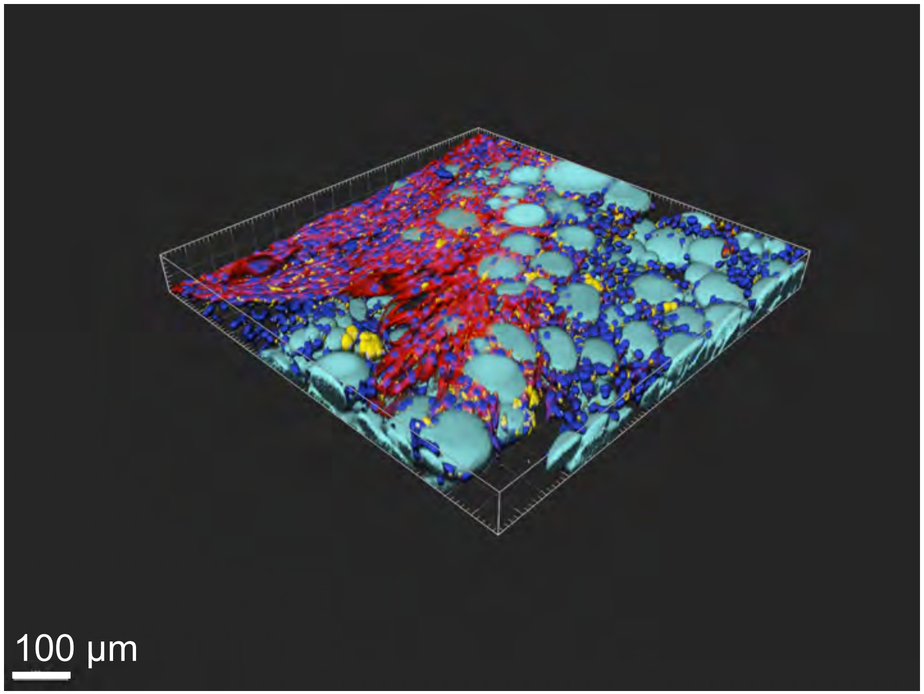


Yellow: Intracellular lipids (Nile Red); Cyan: Microcarriers; Blue: DNA (Hoechst)

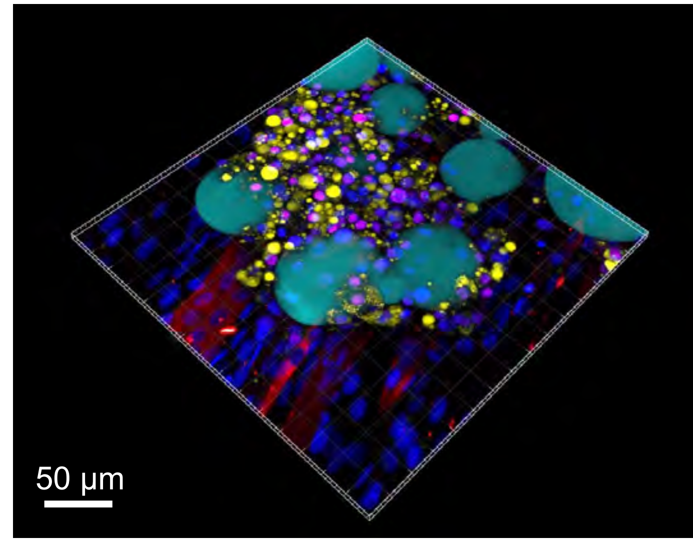
Kawecki et al, *Manuscript in preparation*

Building multicomponent 3D tissue constructs with muscle and fat microtissue

Rabbit



Mouse



Blue: DNA (Hoechst)
Red: Myosin Heavy Chain
Magenta: PPARγ
Yellow: LipidTox
Cyan: Gelatin Beads

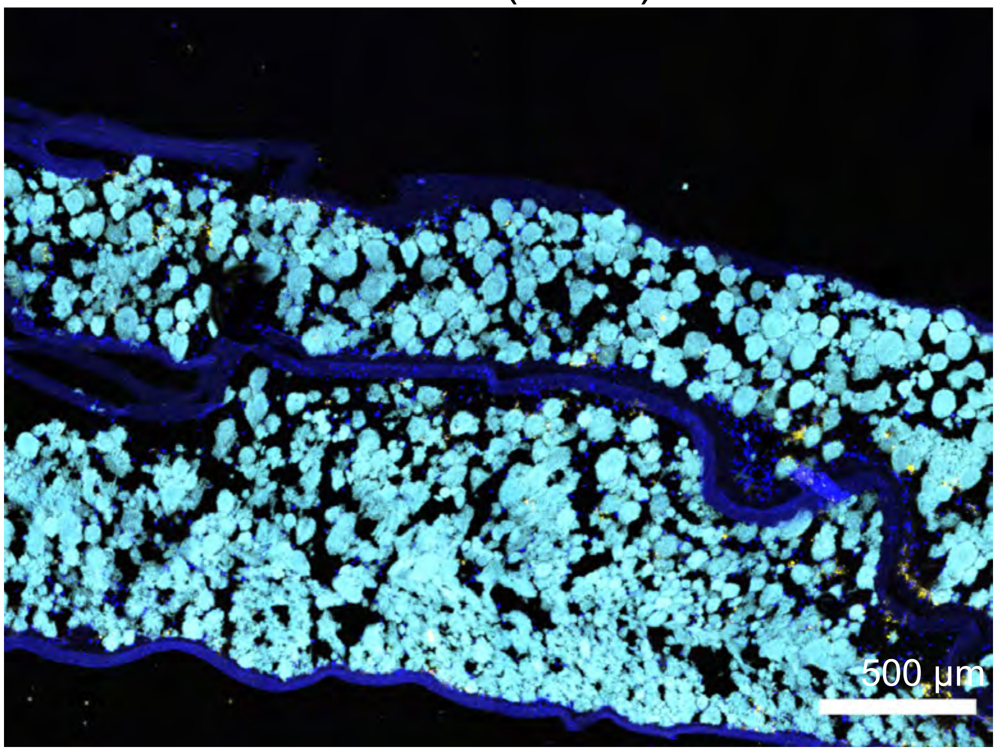
Marbled cultured meat

Kawecki et al, *Manuscript in preparation*

Marbled cultured meat

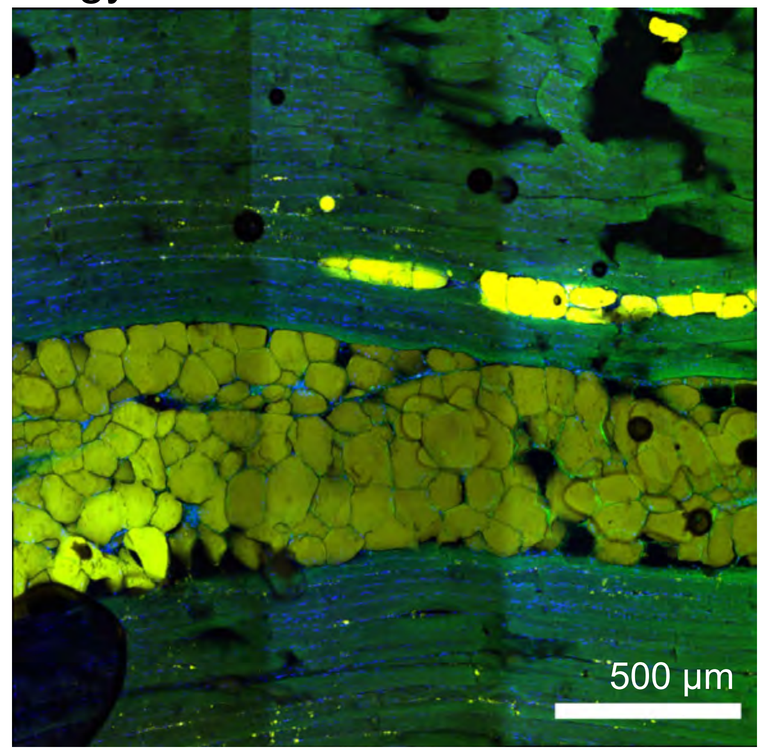
Building multicomponent 3D tissue constructs with muscle and fat microtissue

Cultured marbled meat (rabbit)



Cyan: Microcarriers; Yellow: Nile Red; Blue: DNA (Hoechst)

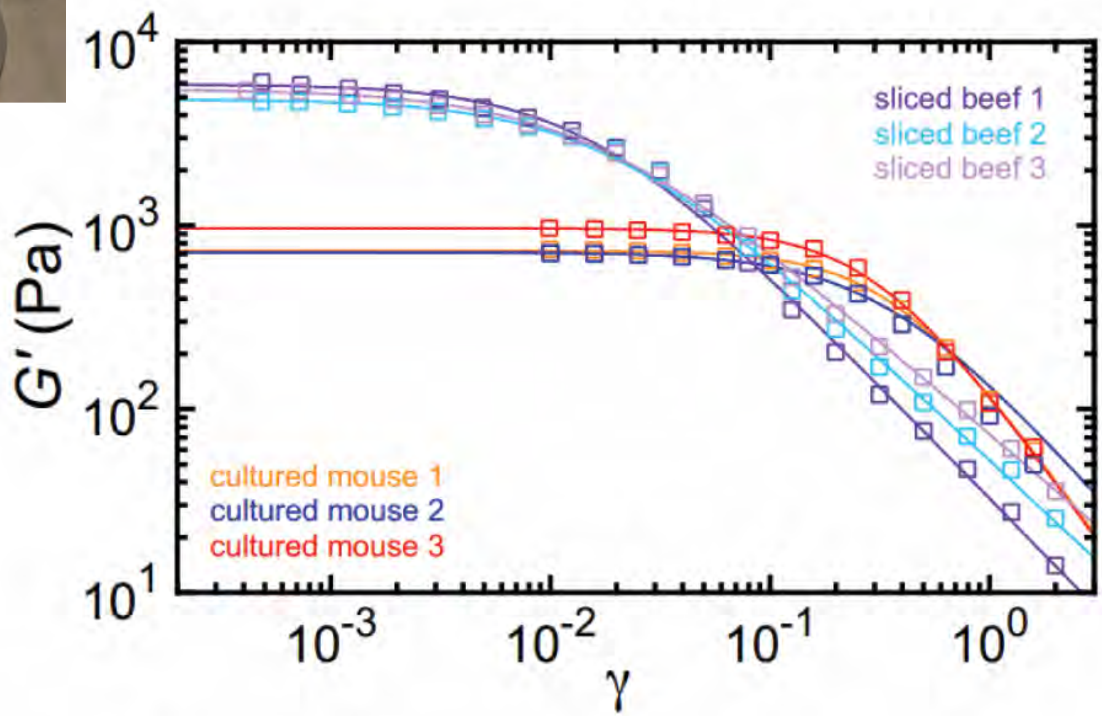
Wagyu steak

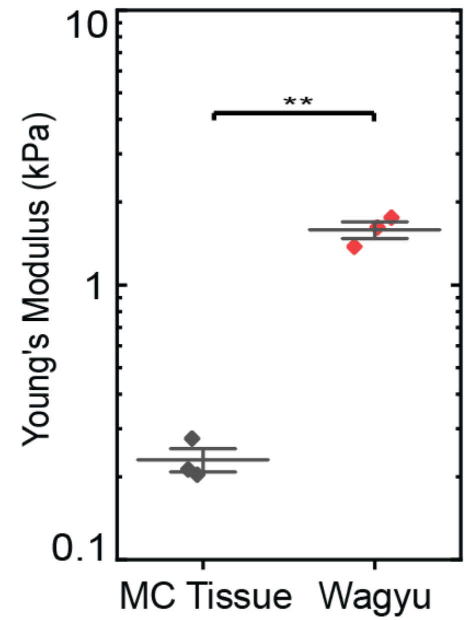


Green: FITC

Kawecki et al, *Manuscript in preparation*

Marbled cultured meat has solid-like behavior, but is more compliant than Wagyu steak





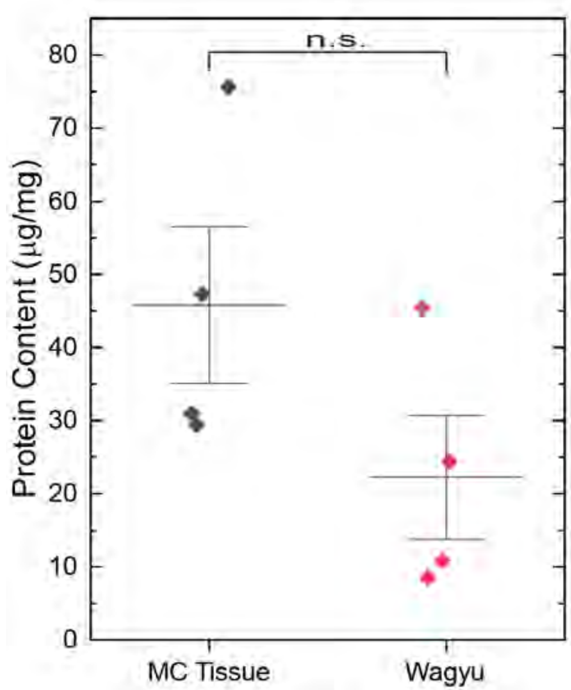
Prof. Tom Mason Yixuan Yu

Kawecki et al, *Manuscript in preparation*

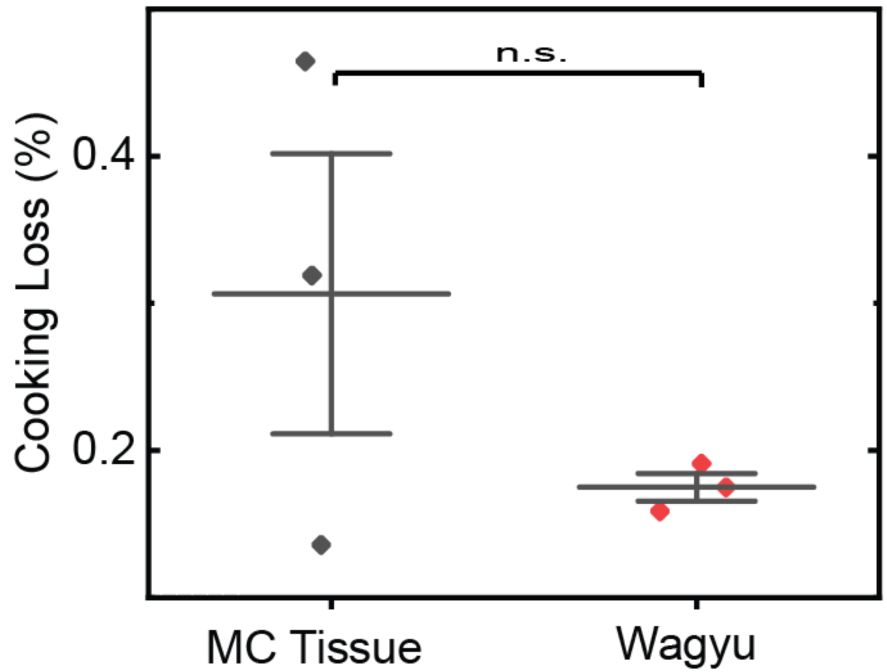
Marbled cultured meat

Marbled cultured meat has desired protein content, but higher cooking loss

Protein content



Cooking loss

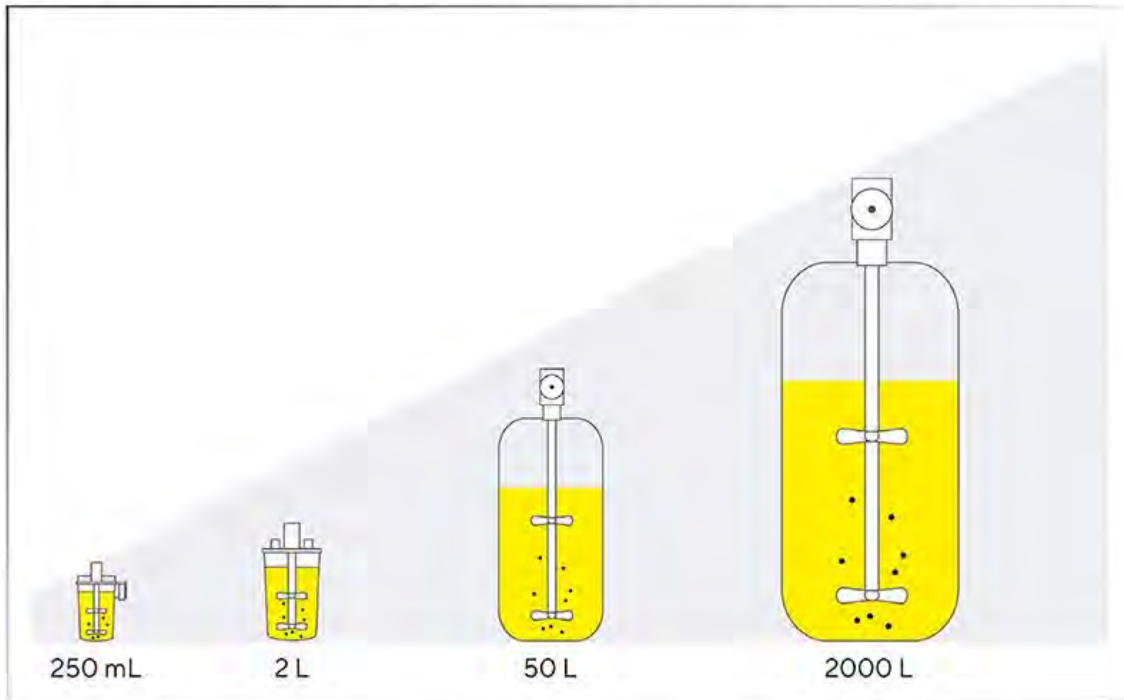


Kawecki et al, *Manuscript in preparation*

Scalability remains a challenge

Marbled cultured meat

How to develop a scalable process for culturing meat with customized scaffolds?



Edible microcarriers

How to develop a scalable process for culturing meat with customized scaffolds?

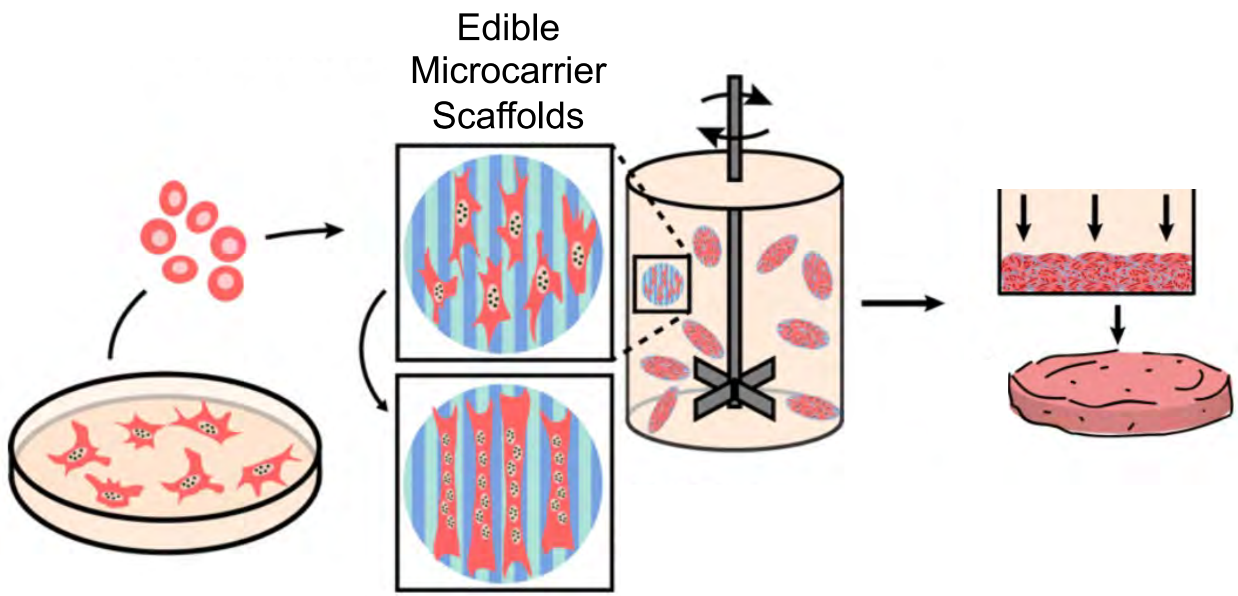
- We recently developed edible microcarrier scaffolds with customized stiffness and topology:


Dr. Sam Norris


Stephanie Kawecki


Kathleen Chen

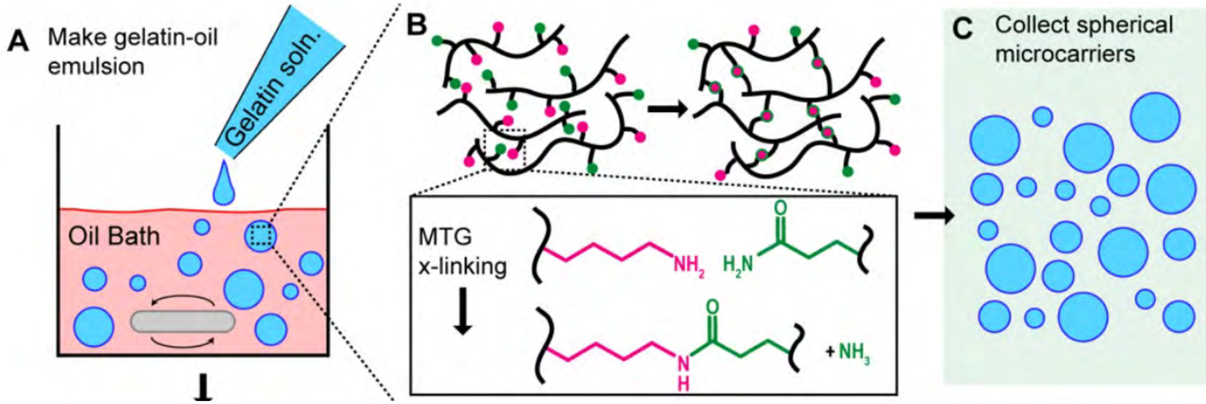

Ashton Davis



Norris, Davis, Kawecki, Chen, Rowat (2022) *Biomaterials*
Rowat, Kawecki et al WO 2020/219755

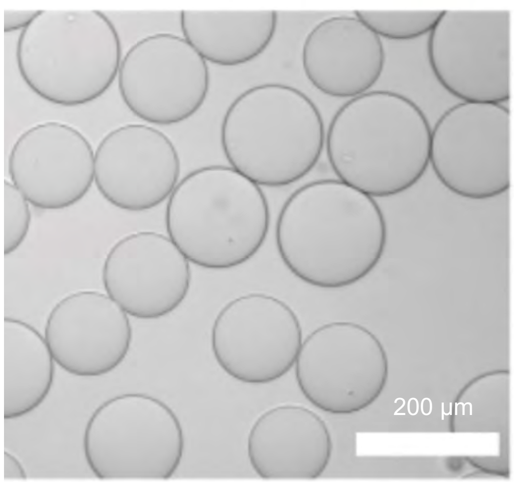
Edible microcarriers

Edible microcarriers with customized stiffness

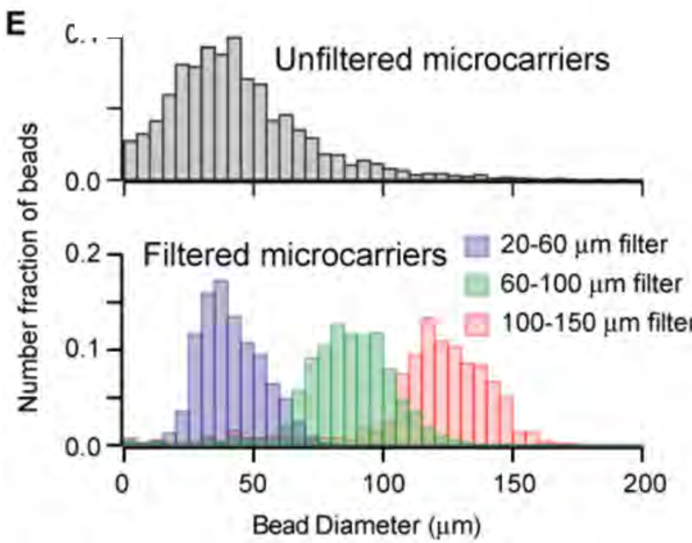


CNSI AT UCLA
Nano and Pico Characterization (NPC) Laboratory

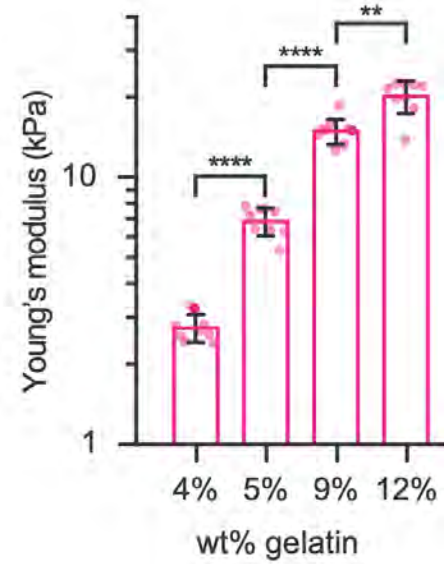
Spherical microcarriers



Tunable size



Tunable stiffness



Engler AJ et al (2004) *J Cell Biol*
McClure et al (2016) *Acta Biomateriala*
Choi JS (2008) *Biomaterials*

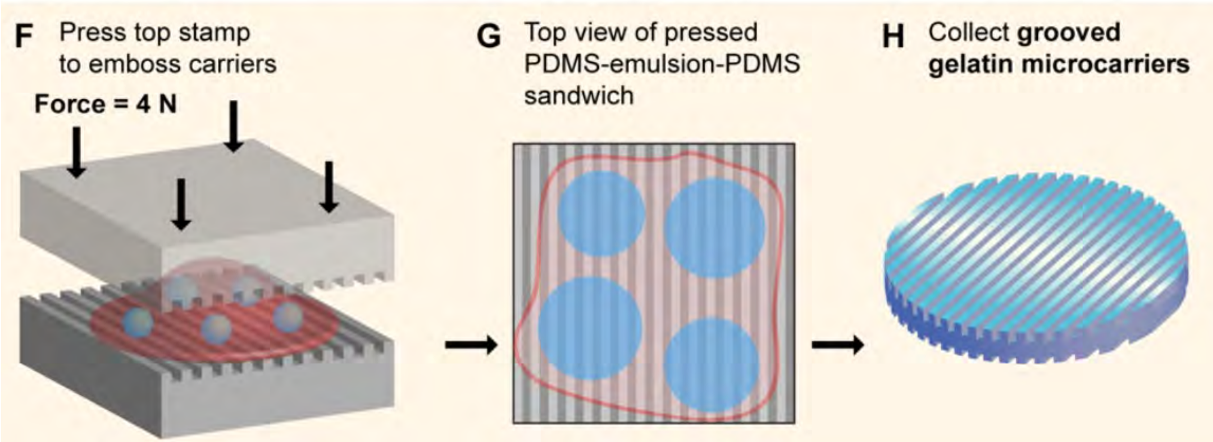
Norris, Davis, Kawecki, Chen, Rowat (2022) *Biomaterials*

Edible microcarriers

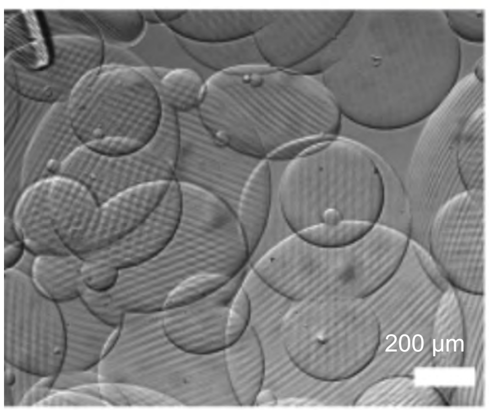
Edible microcarriers

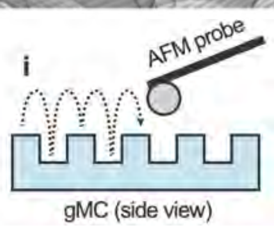
Edible microcarriers with customized stiffness and topology

CNSI at UCLA
Nano and Pico Characterization (NPC) Laboratory
UCLA NanoLab

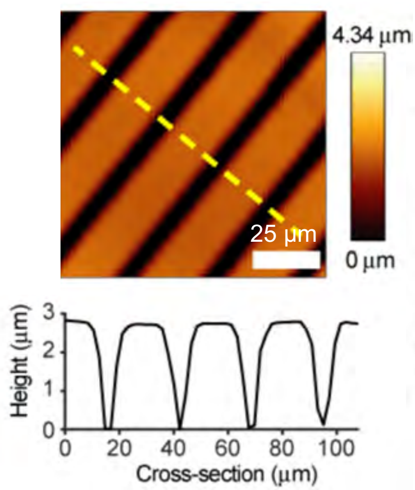


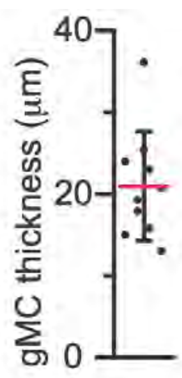
Grooved microcarriers



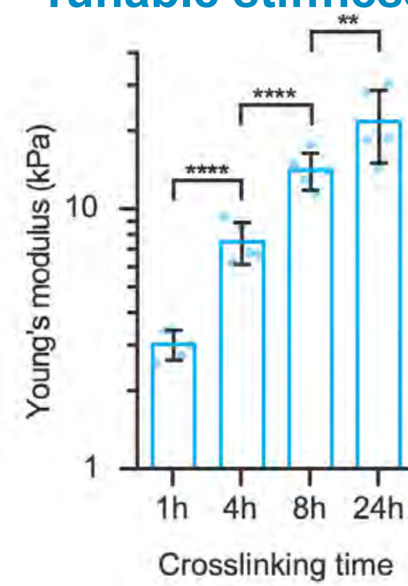


Tunable topology





Tunable stiffness



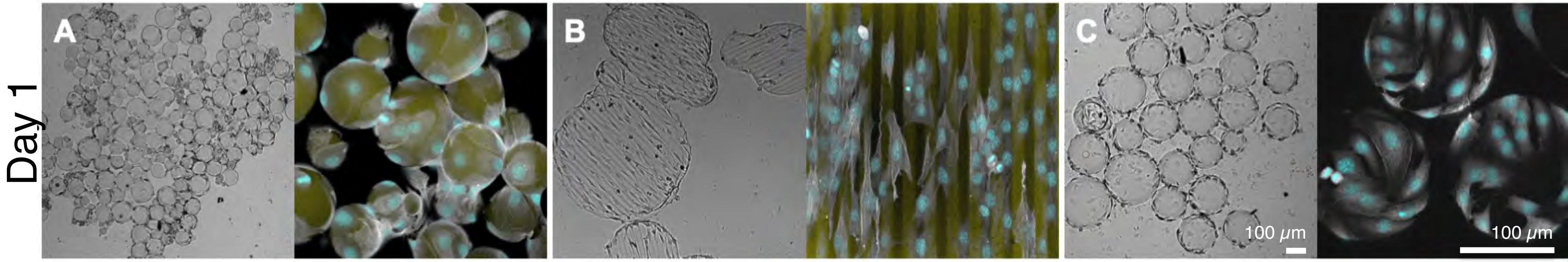
Norris, Davis, Kawecki, Chen, Rowat (2022) *Biomaterials*

Edible microcarriers support cell proliferation

Edible spherical microcarriers (sMCs)

Edible grooved Microcarriers (gMCs)

Commercially-available Cytodex microcarriers



Norris, Davis, Kawecki, Chen, Rowat (2022) *Biomaterials*

Edible microcarriers support cell proliferation

Edible microcarriers

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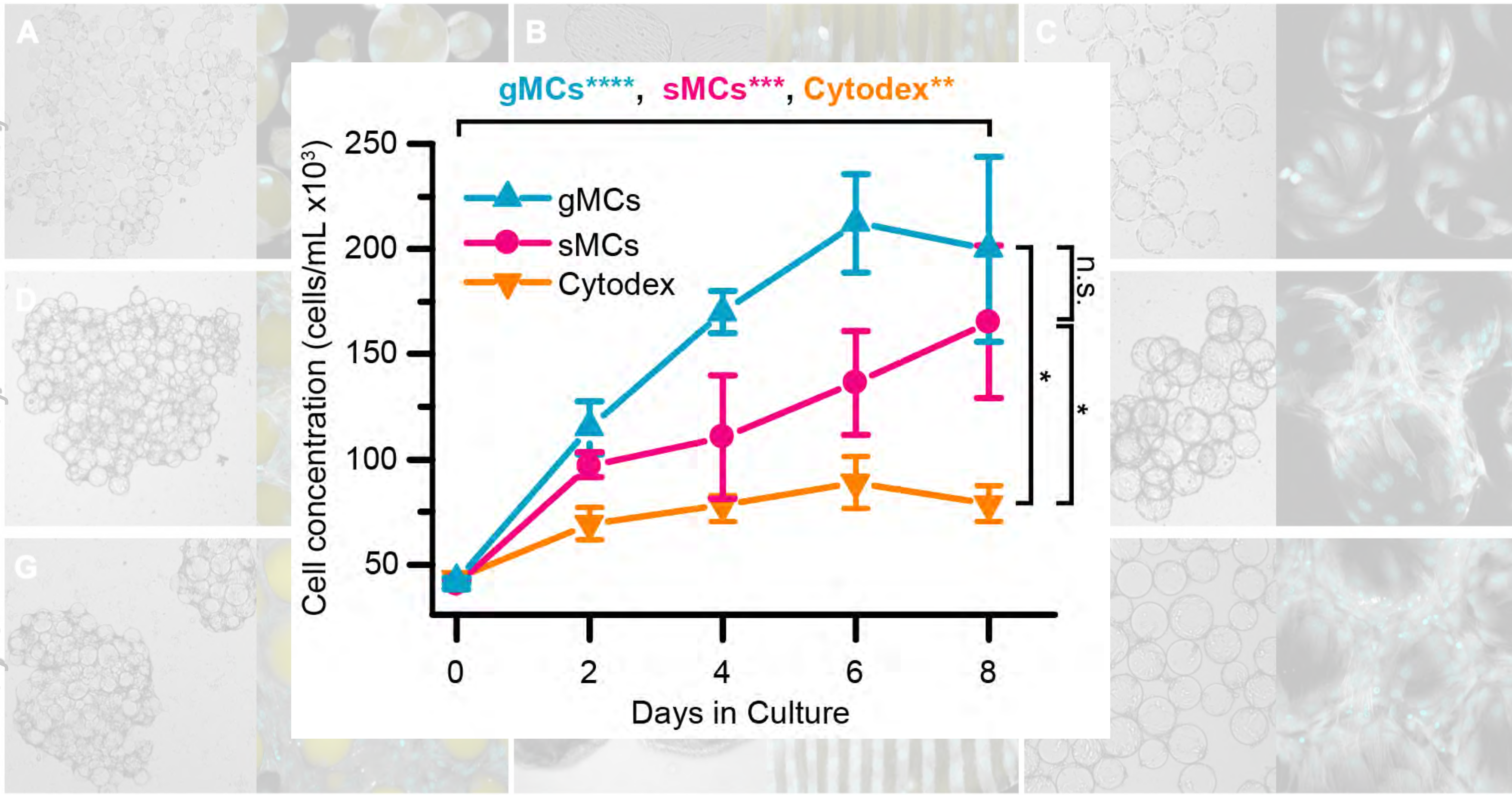
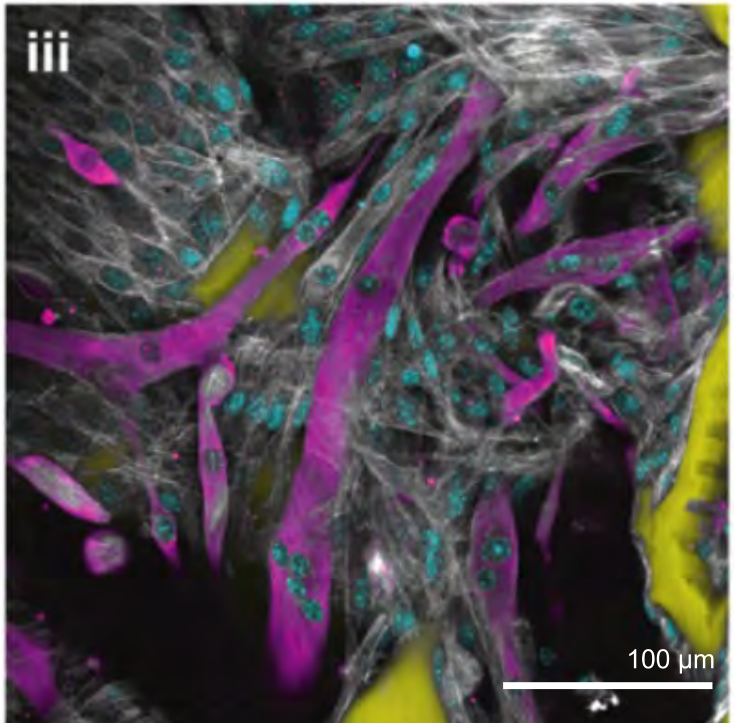
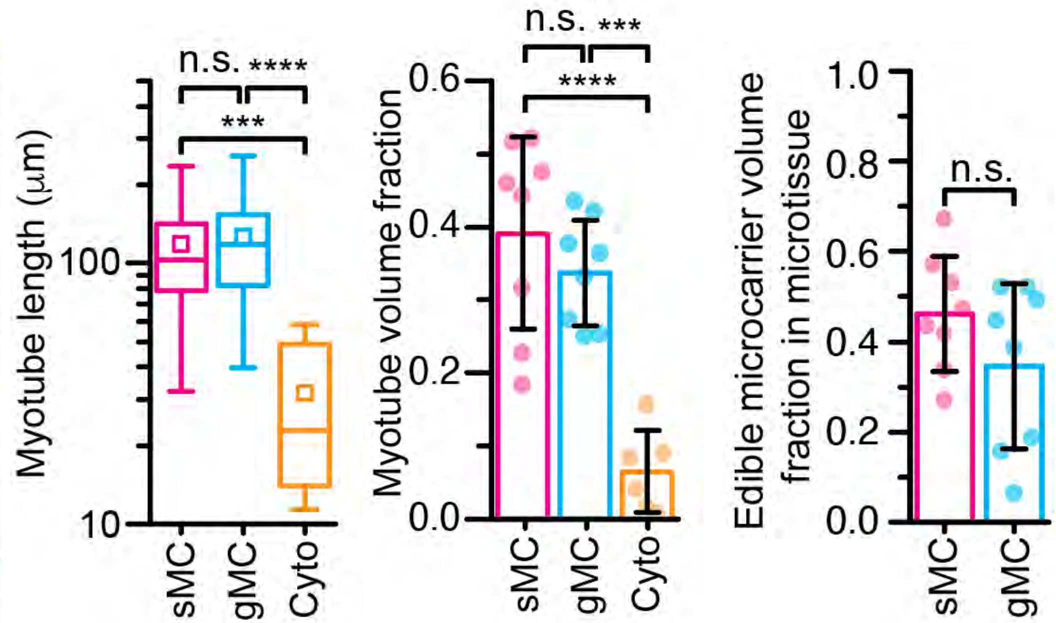
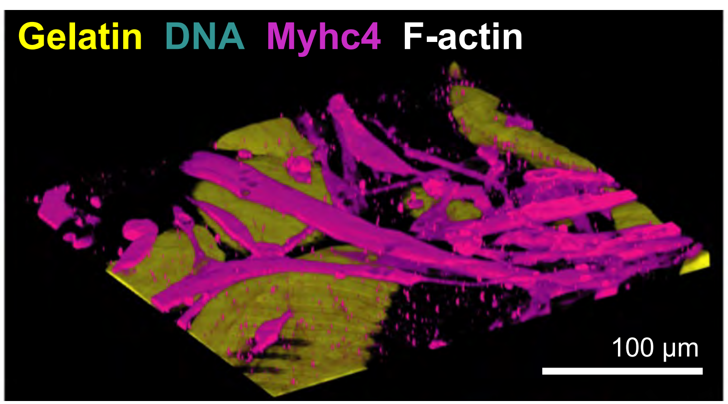


Norris, Davis, Kawecki, Chen, Rowat (2022) *Biomaterials*

Edible microcarriers support myotube formation







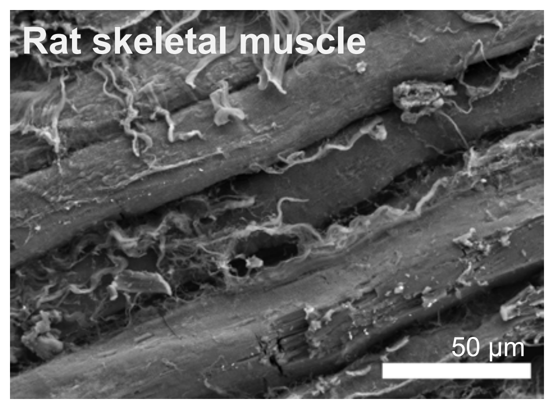
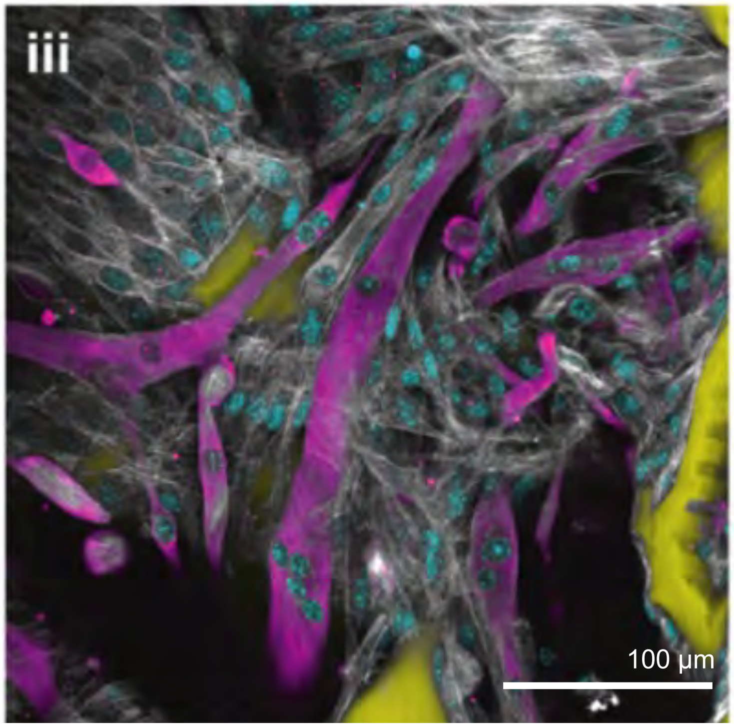


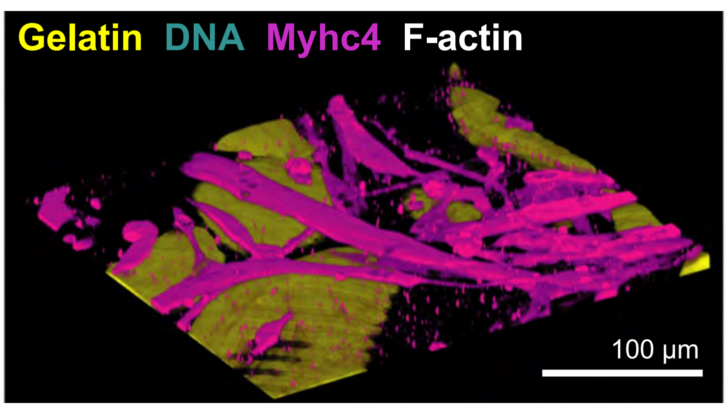
Image: Sam Norris

Norris, Davis, Kawecki, Chen, Rowat (2022) *Biomaterials*

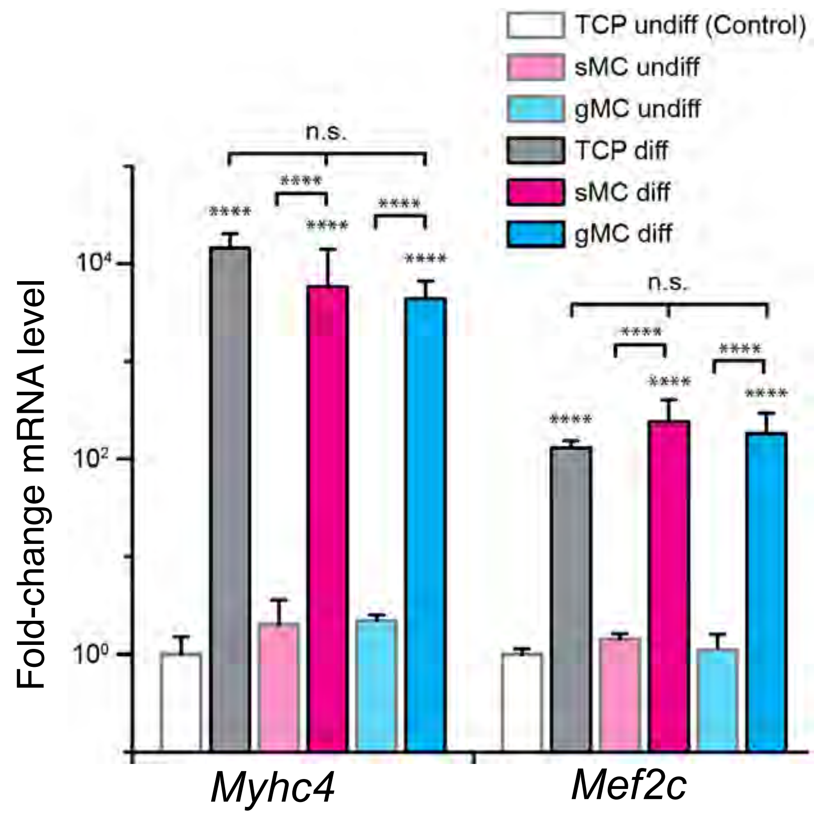
Edible microcarriers

Edible microcarriers support myotube formation





- Similar increase in expression of myogenic markers compared to tissue culture plastic



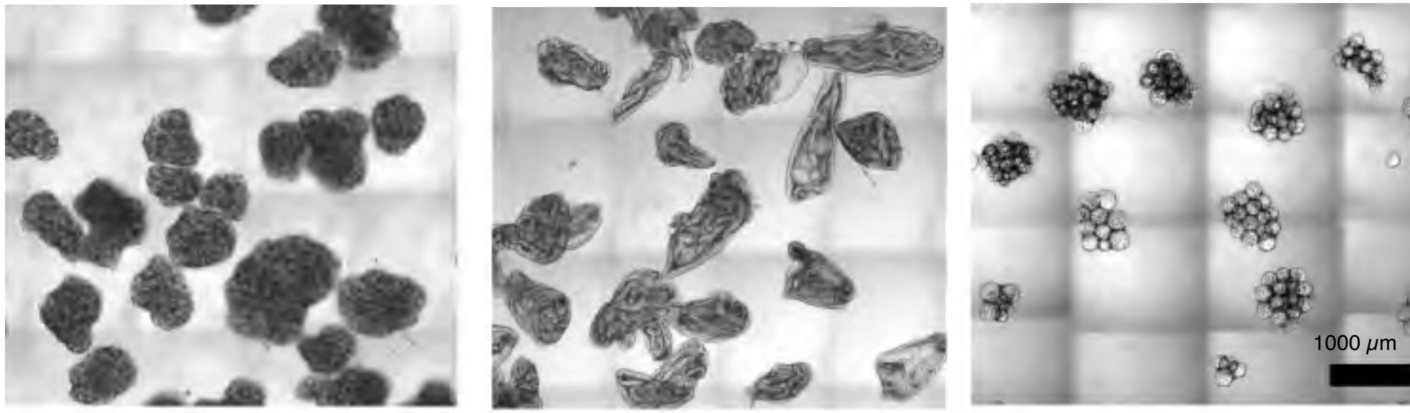
Norris, Davis, Kawecki, Chen, Rowat (2022) *Biomaterials*

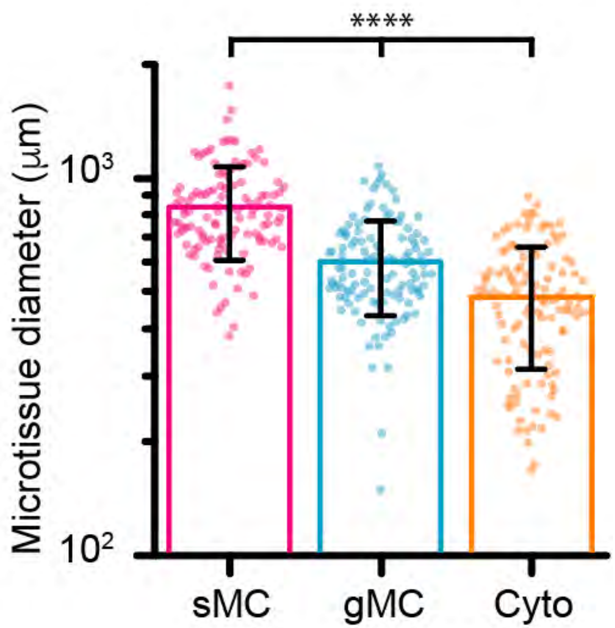
Edible microcarriers

Edible microcarriers support microtissue production using a scalable process

- Cells and microcarriers aggregate in suspension culture

C2C12/sMCs C2C12/gMCs C2C12/Cytodex



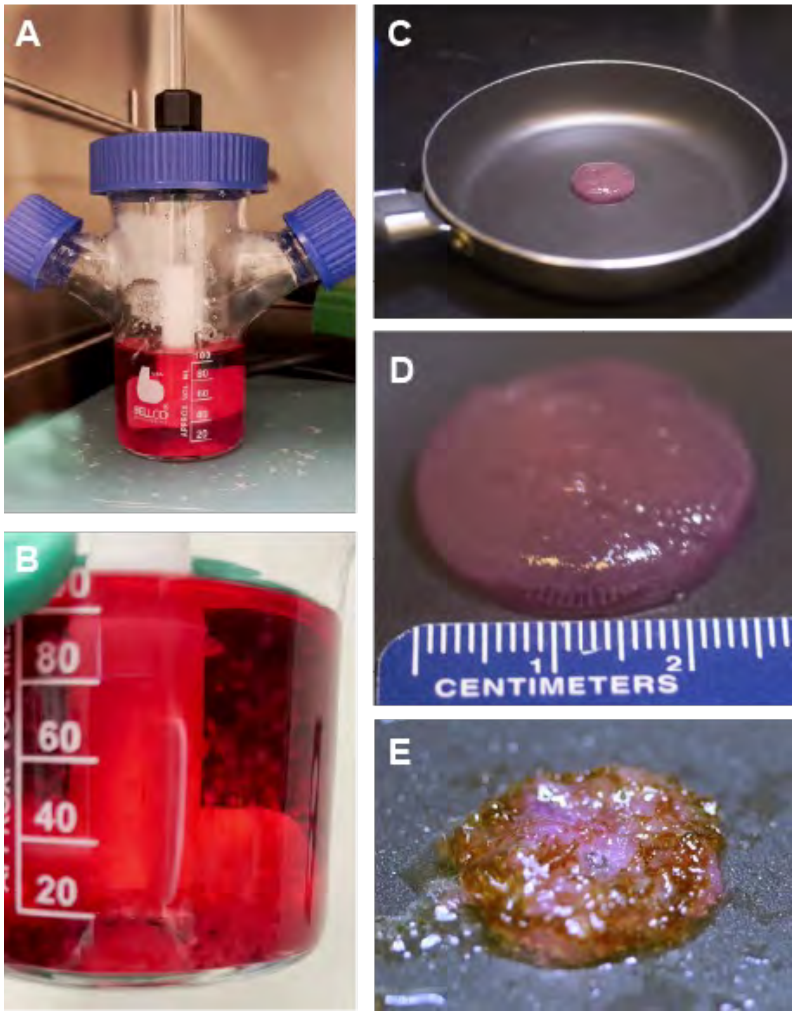


Norris, Davis, Kawecki, Chen, Rowat (2022) *Biomaterials*

Edible microcarriers

Edible microcarriers

Edible microcarriers support bovine satellite muscle cell growth and cookable cultured meat



!!!

www.youtube.com/watch?v=6LhA7F0s5PU

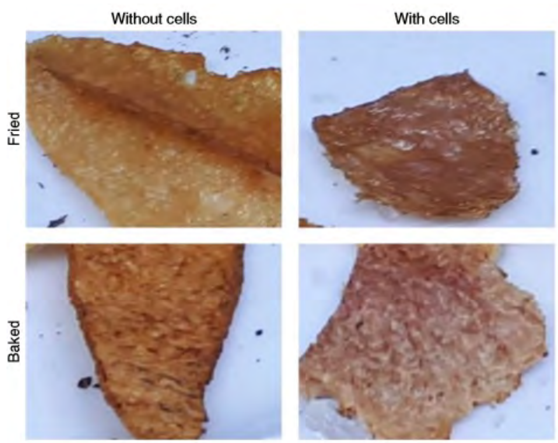
Norris, Davis, Kawecki, Chen, Rowat (2022) *Biomaterials*

Approaches in culturing meat

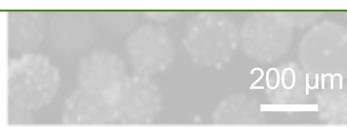
Scalability

?

Natural material scaffolds

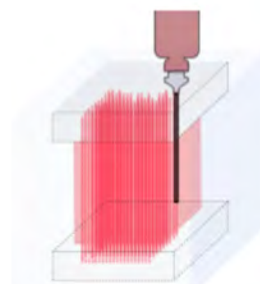


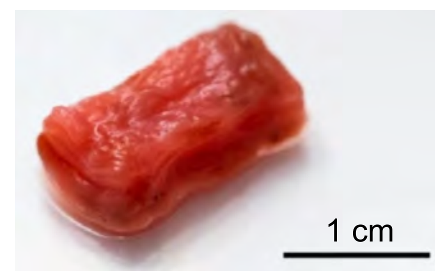
Ben-Arye et al (2020) *Nature Food*



Verbruggen et al (2018) *Cytotechnology*

Bioprinting



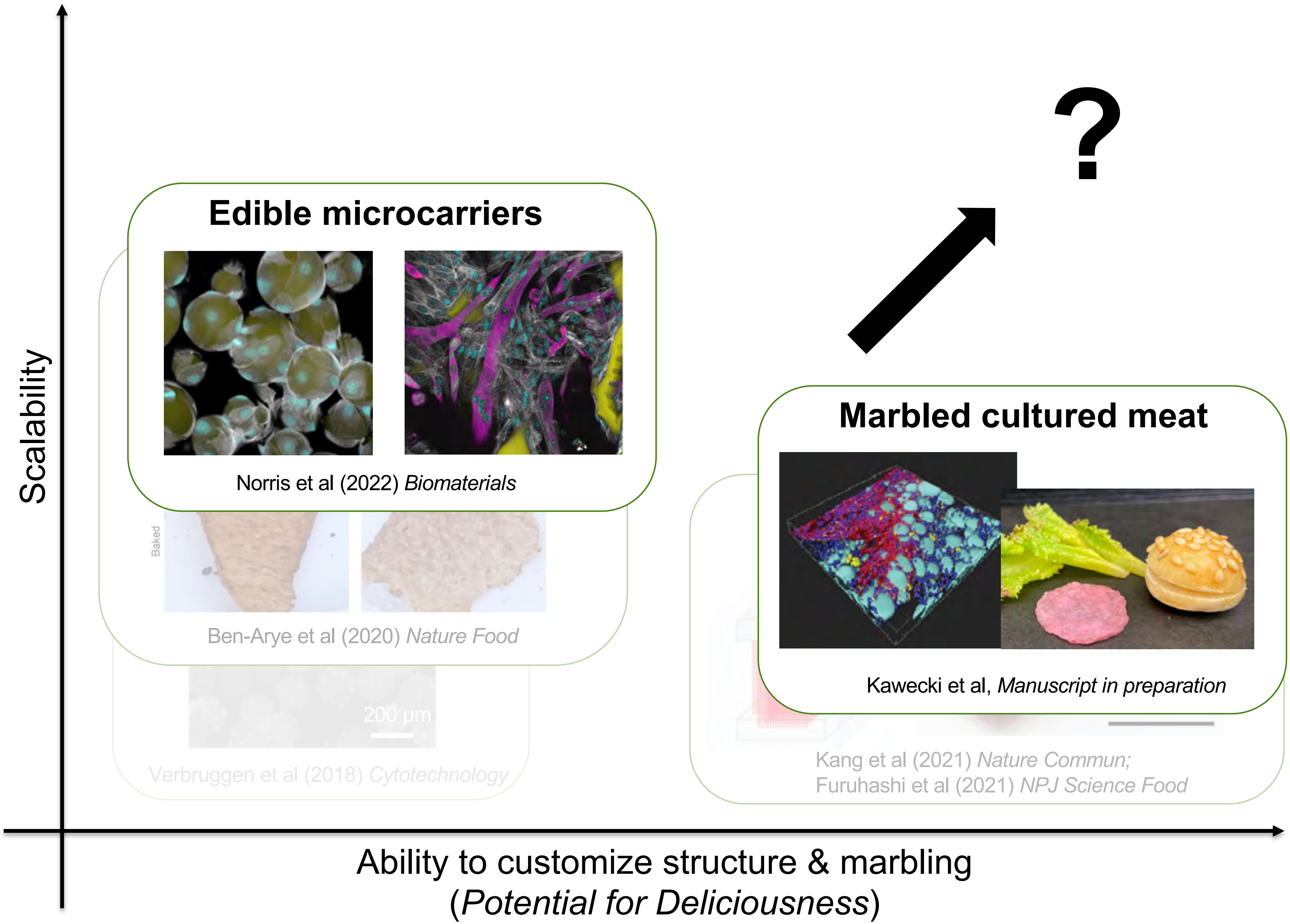


Kang et al (2021) *Nature Commun;*
Furuhashi et al (2021) *NPJ Science Food*

Ability to customize structure & marbling
(*Potential for Deliciousness*)

Our approach

Summary of our cultured meat approaches

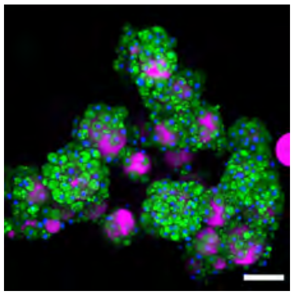


Our approach

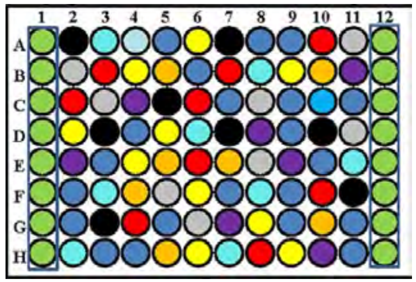
Towards delicious cultured meat

- CNSI Noble Family Innovation Fund Seed project:

Goal: Identify small molecules that accelerate lipid accumulation in edible, engineered adipose tissue



+



Top 12 compounds
+
Tunable microcarrier stiffness

Identified media additives & microcarrier stiffness for accelerated lipid accumulation

CNSI AT UCLA
Molecular Screening Shared Resource

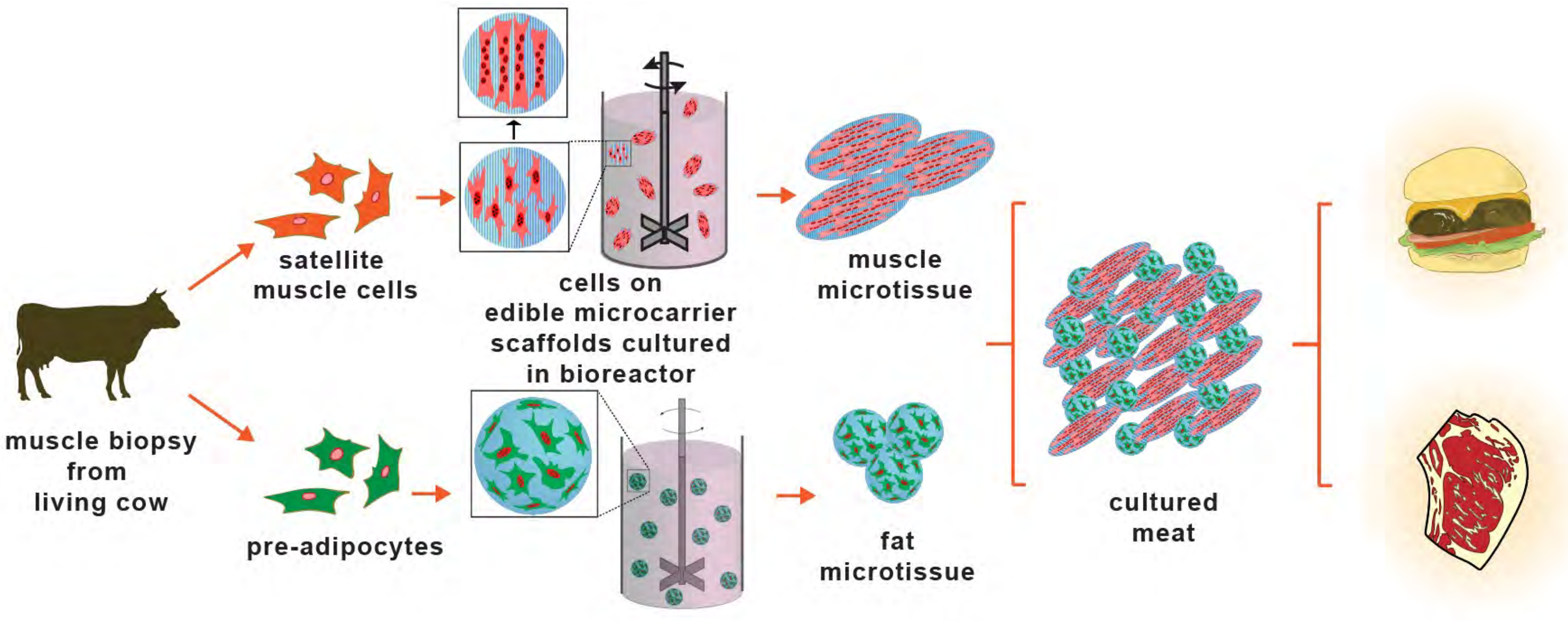
Robert Damoiseaux, Scientific Director, MSSR

Kathleen Chen, PhD candidate, Chemistry

Future outlook

Towards sustainable and delicious cultured meat

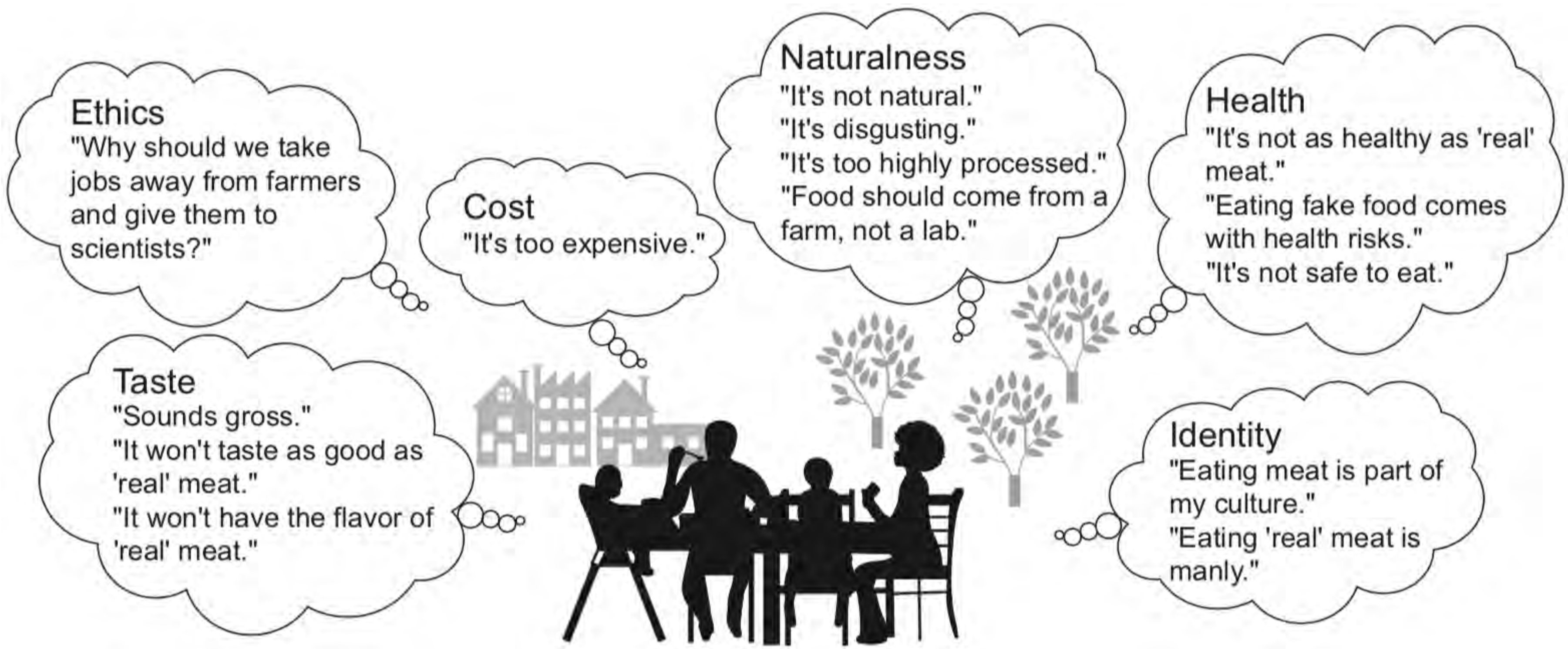
• *The vision*: to grow a marbled, cultured steak where each protein-rich bite is tender and juicy



Future outlook

Furuhashi et al (2021) *Nature Food*; Rubio et al (2020) *Nat Comm*; Tomiyama et al (2020) *Trends Food Sci Tech;* MacQueen et al (2019) *npj Science of Food;* Campuzano & Pelling (2019) *Front Sustain Food Syst;* Springmann et al (2018) *Nature;* Verbruggen et al (2018) *Cytotechnol;* LCA by Odegard et al (2021) *CE Delft;* TEA by Vergeer et al (2021) *CE Delft*

Challenge: consumer perceptions of cultured meat

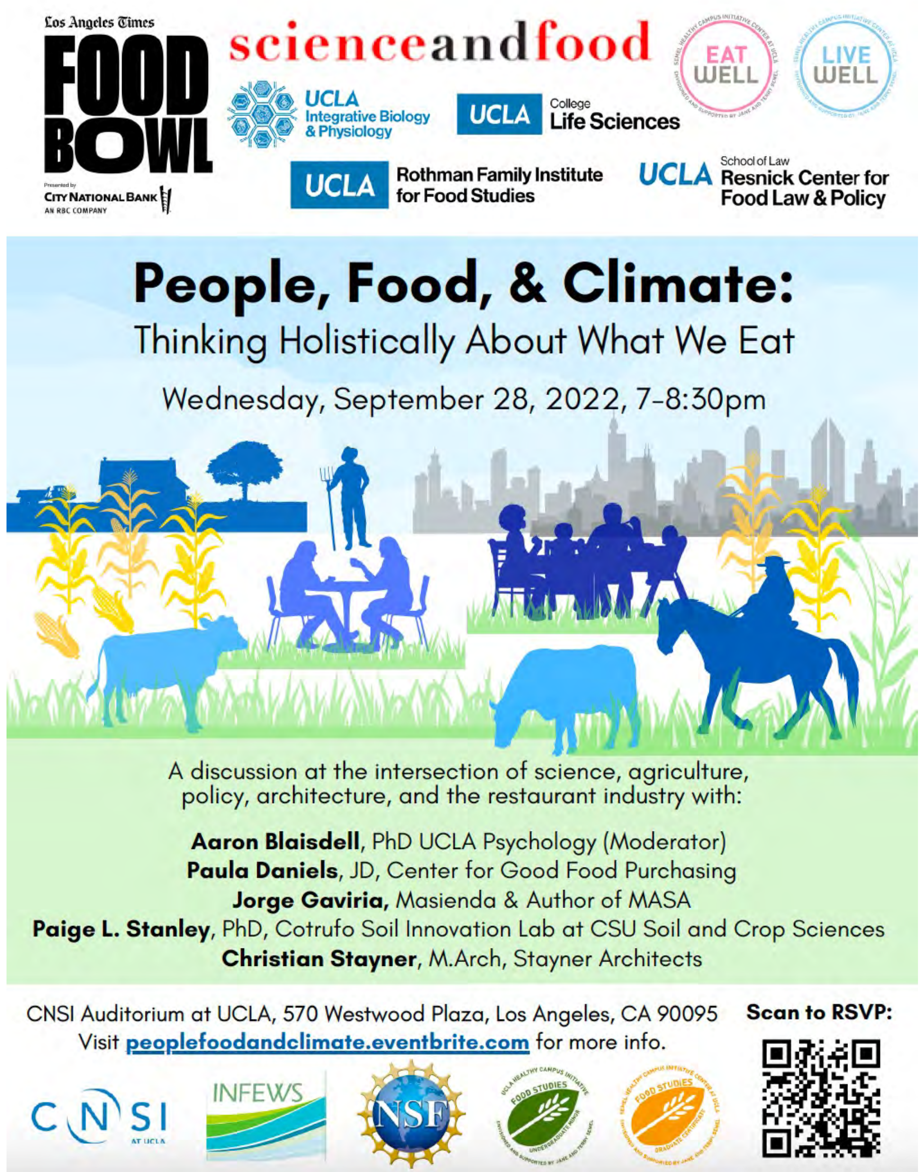


Prof. Janet Tomiyama, Psychology

Tomiyama, Kawecki, Rosenfeld, Jay, Rajagopal, Rowat (2020) *Trends Food Sci Tech*

Future outlook

Opportunities to enrich the public understanding of science and food



Future outlook

THE ROWAT LAB
UNIVERSITY OF CALIFORNIA LOS ANGELES

SEMEL HCI Center at UCLA

NSF

NATIONAL INSTITUTES OF HEALTH

USDA

NIFA

UCLA Integrative Biology & Physiology

CNSI AT UCLA

Noble Family Innovation Fund

THE GOOD FOOD INSTITUTE

NEW HARVEST

+Marcie H. Rothman Presidential Chair in Food Studies
+Farber Family Foundation

Sam Norris, PhD

Stephanie Kawecki

Kathleen Chen

Ashton Davis

Ester Fridman

Angelina Flores

Don Lamkin, PhD

Bryanna Chavez

Jennifer Soto, PhD

Chau Ly

Layal Suboh

Adhvaith Vijay

Pancho Alvarez

Dr. Navjot Kaur Gill (PhD), now Postdoc, U British Columbia
Dr. David Hoelze (postdoc), now Assc Prof, Ohio State U
Dr. Tae-Hyung Kim (postdoc), now Asst Prof, U New Mexico
Dr. Kendra Nyberg (PhD), now R&D lead, Calico

Collaborators

Manish Butte, UCLA
Robert Damoiseaux, UCLA
Rachelle Crosbie, UCLA
Arjun Deb, UCLA
Beth Karlan, UCLA
Jennifer Fenton, Michigan State U
Andrea Garmyn, Michigan State U
David Kaplan, Tufts
Parag Katira, San Diego State U
Song Li, UCLA
Sandra Orsulic, UCLA
Deepak Rajagopal, UCLA
Jason Rowntree, Michigan State U
Gale Strasburg, Michigan State U
Wendy Slusser, UCLA
Janet Tomiyama, UCLA
Claudio Villaneuva, UCLA
Tom Vondriska, UCLA
Xia Yang, UCLA