MILK PRODUCED BY CELL CULTURES

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FOOD PROTEIN TRANSFORMATION

- ✓ Expect that alternative protein production face an exponential growth where big and research heavy companies will develop, patent, produce and deliver raw materials and ingredients.
- ✓ New start-ups will and are emerging.
- ✓ Will new technologies enable potential partly transition away from production animals?
- ✓ Transformation expected in 3 waves with breakthroughs in the following periods:

2020 - 2025 Plant based proteins

2025 - 2030 Mikrobial produced proteins (precision fermentation)

2030 - 2035 Stem cell based meat and milk

References:

Boston Consulting Group (2021) ”Food for Thought – The Protein Transformation”

EU-analysis ”Farmers of the Future, 2020”

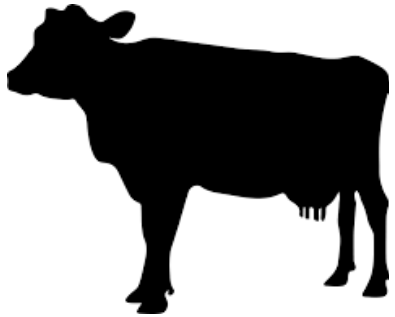
American ”think-tank” RethinkX (2019)

CLIMATEMILK – EFFECT OF FEED ADDITIVES ON MILK QUALITY AND FUN

Controlled feeding trials

WP

Animal feeding t



Feed additives

3-NOP

Nitrate

Farm testing

WE WOULD LIKE TO INVITE YOU TO
GAYANI LOKUGE´S PHD DEFENCE:

SUSTAINABLE DAIRY
- EXPLORING THE LINK BETWEEN CLIMATE-FRIENDLY DIETS AND MILK QUALITY

TUESDAY 25 JUNE 2024 13.00-17.30

Room 5910-214, Department of Food Science
Aarhus University, Agro Food Park 48, 8200 Aarhus N

The defence will be finishing with a reception.

If you wish to partioipate in the reoeption please register no later than 18 June 2024. Sign up for the reception here:
https://events.au.dk/phdreceptionegayanilokuge/signup

PROGRAM

13.00-13.05	Welcome by Chair of the assessment committee, Associate Professor Martin Krøyer Rasmussen and main supervisor, Associate Professor Nina Aagaard Poulsen
13.05-13.50	Gayani Lokuge presents the thesis entitled: *"Sustainable dairy - Exploring the link between climate-friendly diets and milk quality"*
13.50-14.00	Break
14.00-16.00	Defence of the thesis
16.00-16.15	The assessment committee concludes
16.30-17.30	Reception - a light snack will be served

AARHUS UNIVERSITY
DEPARTMENT OF FOOD SCIENCE

ality and composition

WP 2

erall milk composition (Milkoscan)

icle size (Nanoflex, mastersizer)

min B2 (HPLC), B12 (ELISA)

ein profiling (LC-ESI/MS)

y acid composition (GC)

hods for N-compounds in milk

orimetric assay, DUMAS)

nctionality

WP 3

crystallization

ein and fat oxidation

coagulation

Funding: Danish Dairy Research Foundation, Partners: AU-FOOD, AU-ANIS, DSM and Arla Foods amba. Project leader Nina Aa. Poulsen

Single cell and fermentation based protein

- Those Vegan Cowboys
- Perfect Day
- Formo
- VIVICI

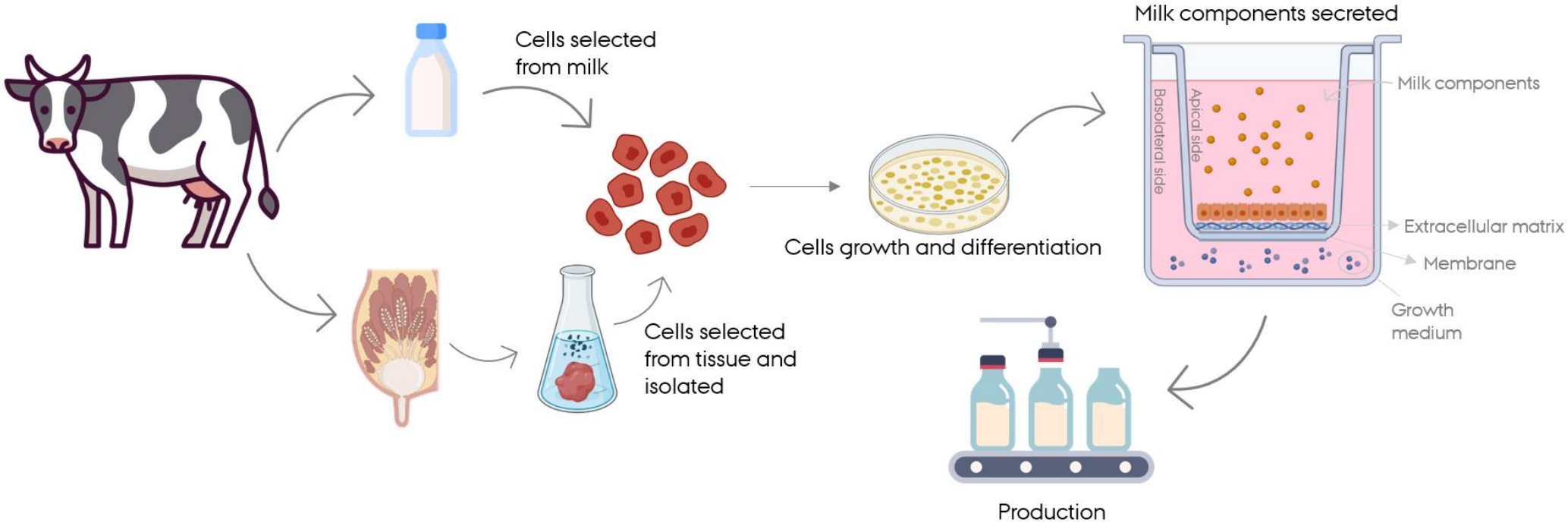
New Culture

- remilk.
- fermify

Cultivated milk (human/bovine)

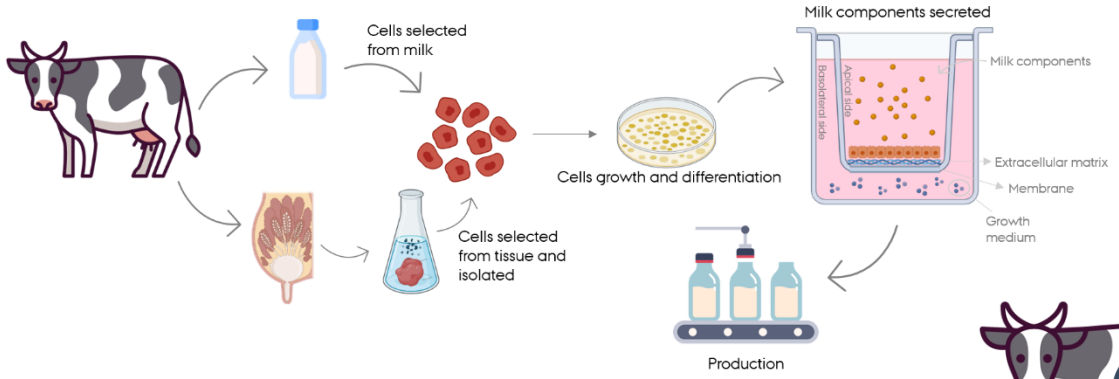
- TurtleTree
- WILK
- BIOMILQ Human milk for babies
- opalia A new era of dairy

PRINCIPLES OF THE TECHNIQUE

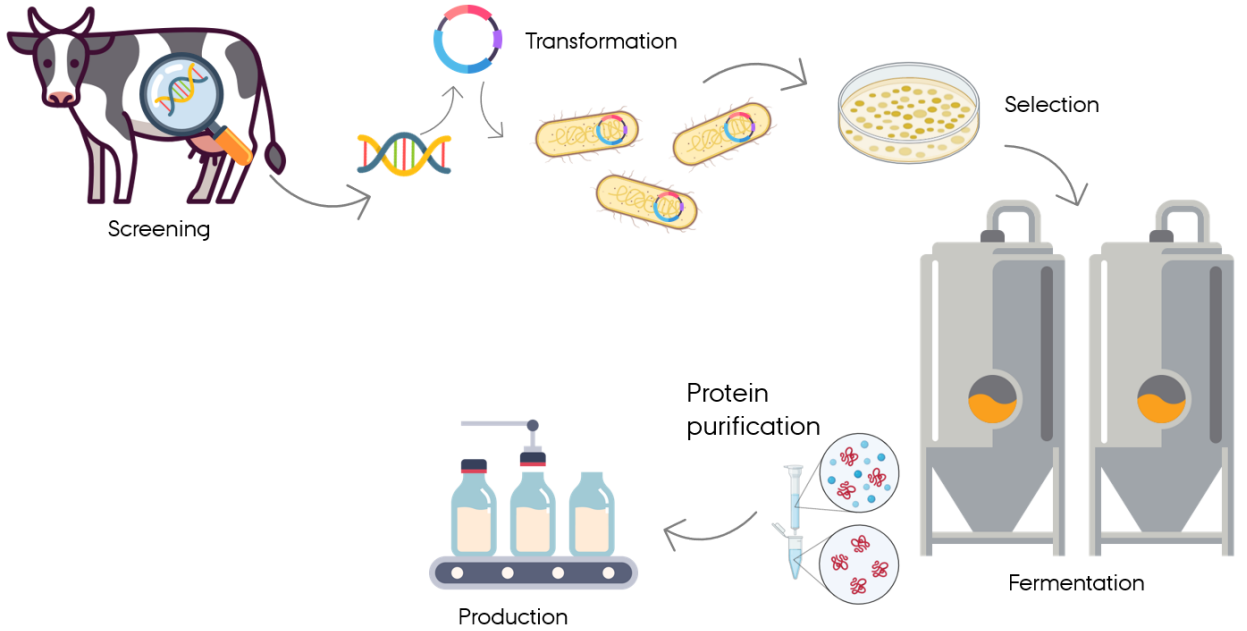


In vitro culture of bovine mammary epithelial cells (MECs) derived from tissue or milk

DIFFERENCE BETWEEN CELLULAR MILK PRODUCTION AND PRECISION FERMENTATION



Single-cell (bacteria/yeast/fungi) genetically modified organisms (GMO) called precision fermentation

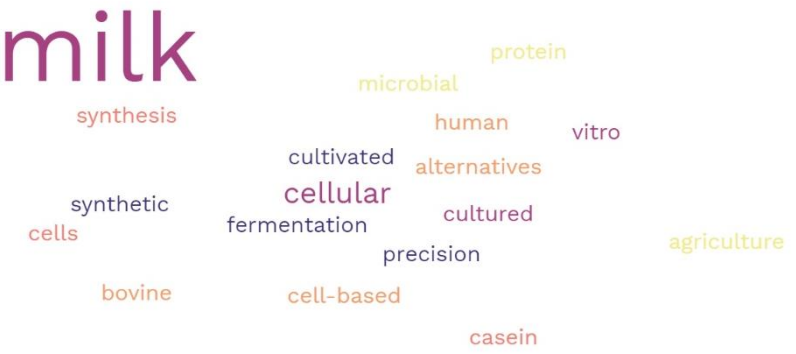


TERMINOLOGIES

Cell-based milk alternative? – no

Cell based protein alternative – yes

According to Codex not ok to use the word milk protein or whey protein in connection with products not coming from a cow, but ok to use the specific protein terminologies, like casein or albumin for example



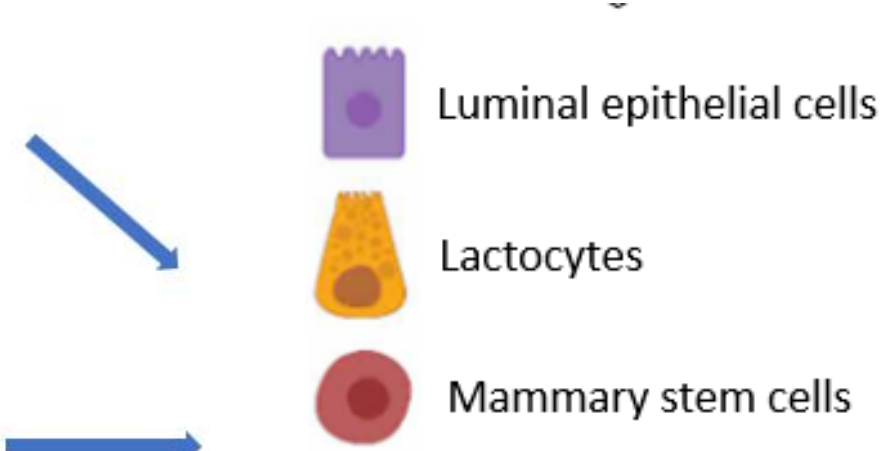
Cell composition of milk from healthy cows (the cell count)

- Macrophages
- Neutrophils (PMN leucocytes)
- Lymphocytes
- Small amount of epithelial cells (mammary epithelial cells=MECs)

Centrifuge to isolate cells

CELL TYPES

1. TISSUE OR MILK COLLECTION

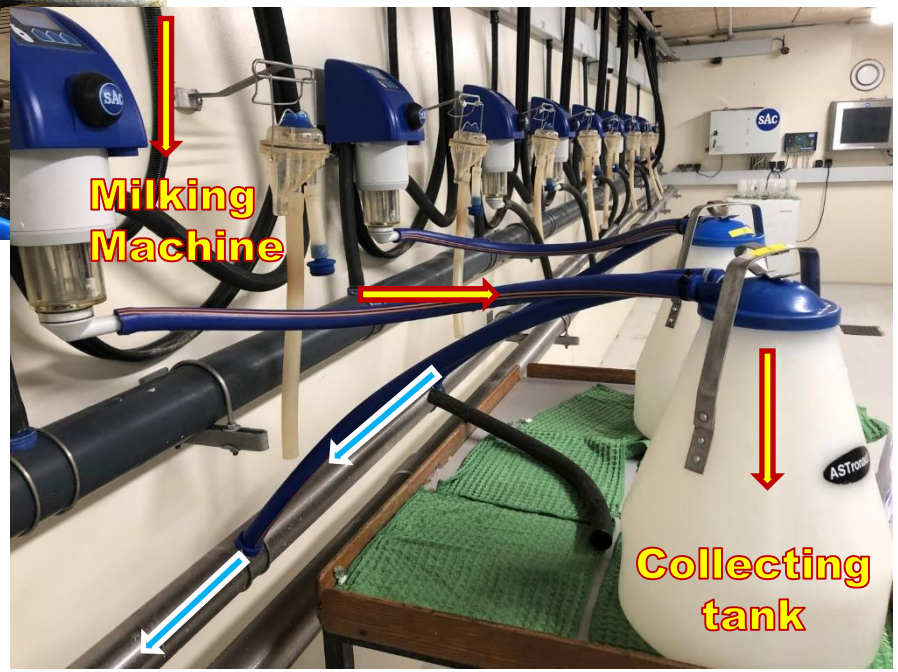


2. CELL ISOLATION

Use marker for luminal epithelial cells but move on with a pool of mixed polulations

Milk cell isolation – milk collection in barn

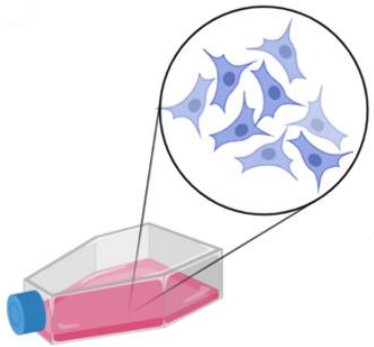
Max. 10 L of milk collected
Extra milk goes to the big tank



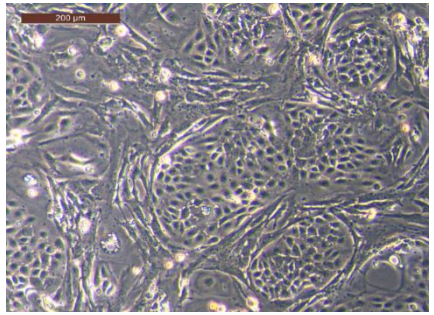
Isolation of mamamry epithalial cells from milk

Milk collection

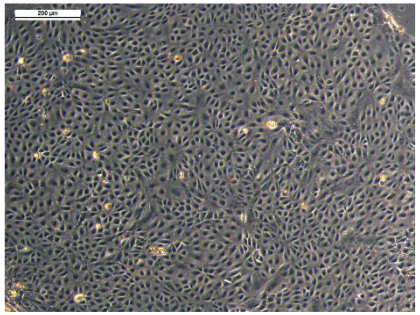
Centrifugation



Isolated cells



Purification &
Characterization



Cell culture

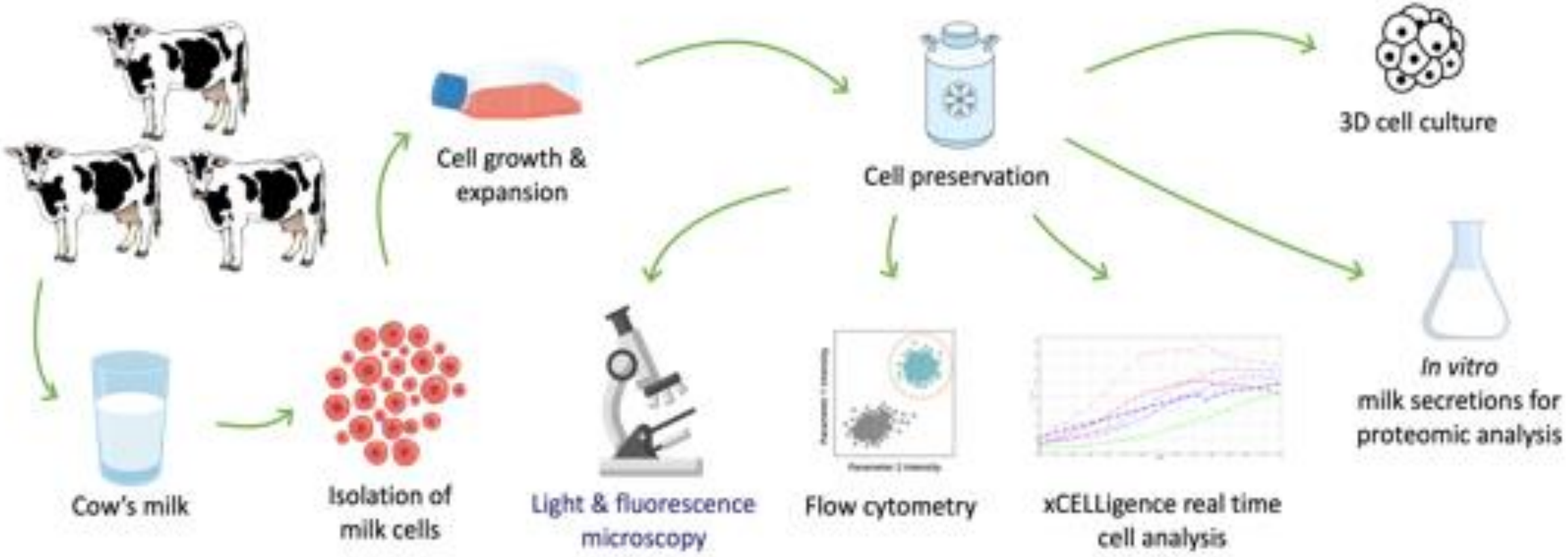
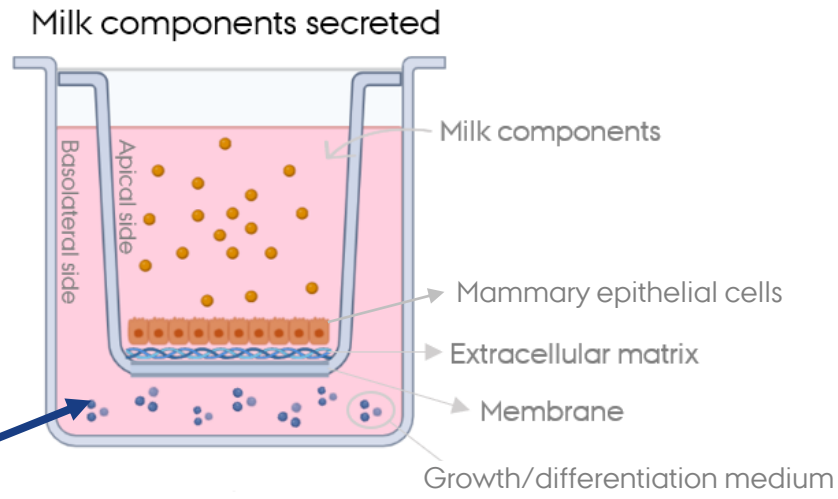


Fig 1. Illustration of the workflow

CULTIVATED "MILK" SECRETOMES

Mammary epithelial cells from udder or from milk

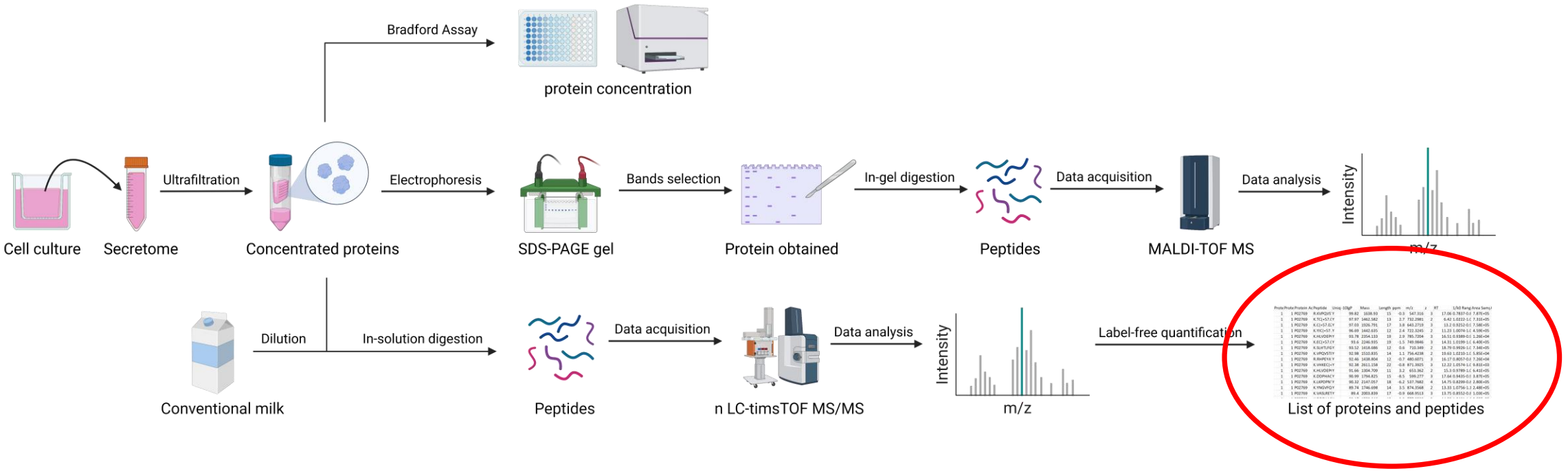
Harvest = secretome = milk secretions + basal medium



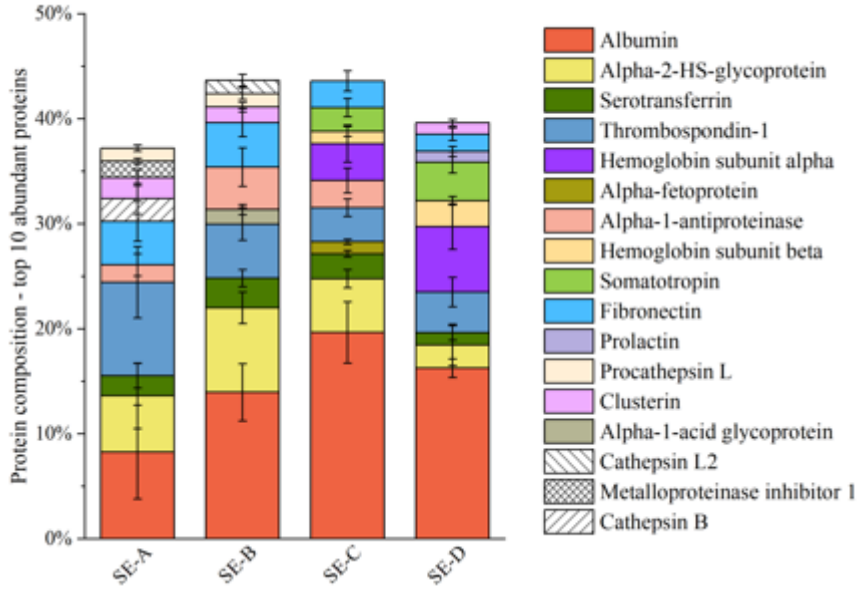
https:/jetbiofil.eu

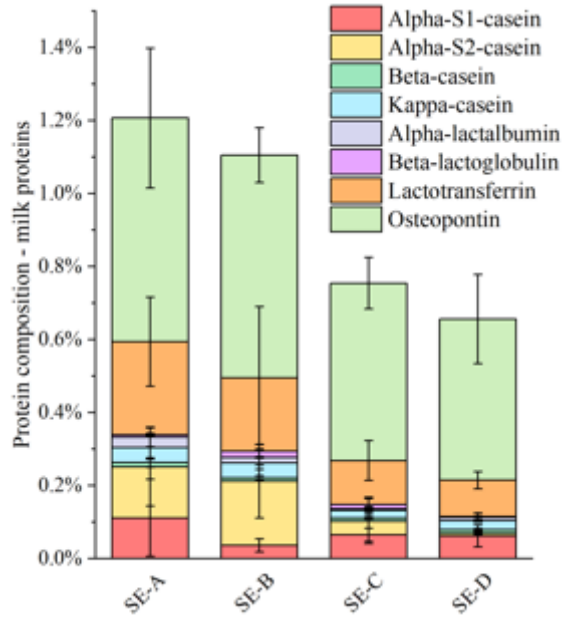
Lactogenic hormones:
Bovine pituitary extract or recombinant prolactin

PROTEOMICS OF SECRETOMES FROM CULTIVATED CELLS FROM UDDER TISSUE



PROTEOMICS OF SECRETOMES

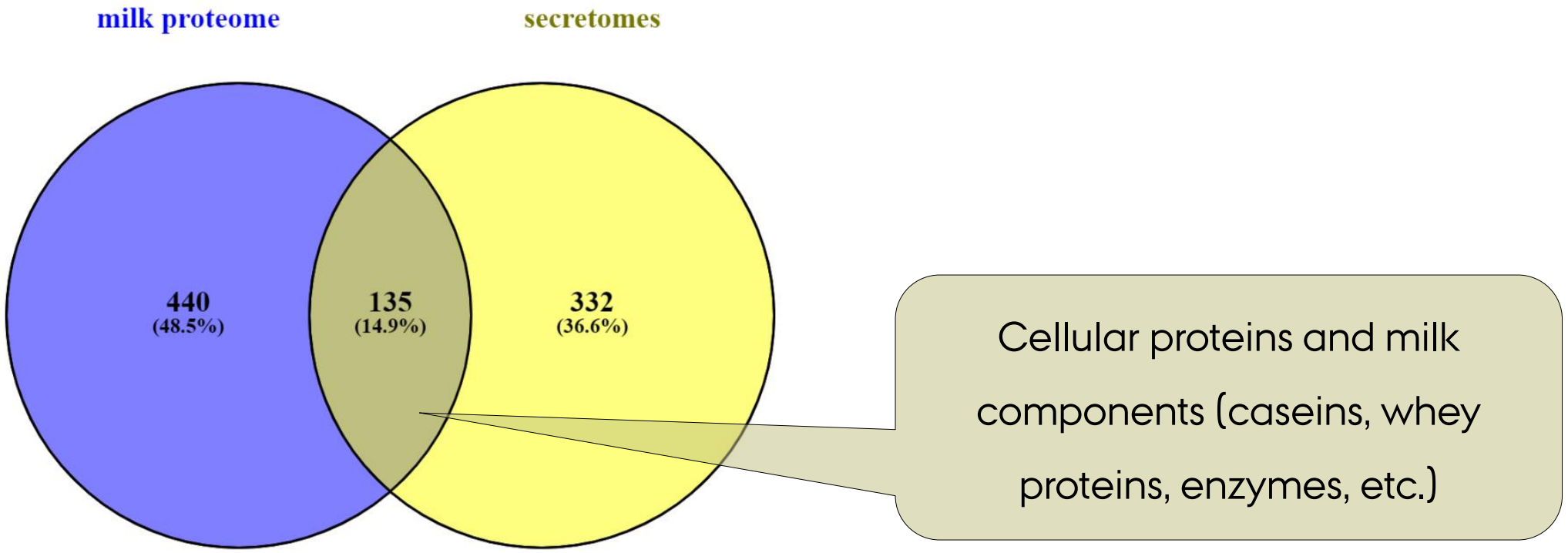




Caseins and whey proteins were also detected, but in low amounts

- The top 10 abundant proteins constituted ~40% of the total proteins.
- Despite the identification of interesting proteins, the total protein contents of the secretomes (approximately 0.05 mg/mL) were much lower than regular milk, with interested milk proteins taking less than 1.5% of the total protein abundance.

SECRETOMES VS. MILK PROTEOME



Venn diagram of milk proteomes (D'Alessandro et al., 2011) compared to common secretomes

Che et al. (2024). Proteomic study of secretomes from cellular agriculture for milk production. Discover Food, in review.

CHALLENGES

- Cell growth and viability
- Induction of lactation and secretion of milk proteins
- Continous growth and secretion
- Sustainable media with growth factors for the cells
- Low protein secretion (currently 0,05 mg/ml)
- Upscaling in bioreactors
- ....

PUBLICATIONS

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DOI: 10.1002/cbin.12116

RESEARCH ARTICLE

Cell Biology International WILEY

Bovine mammary epithelial cells can grow and express milk protein synthesis genes at reduced fetal bovine serum concentration

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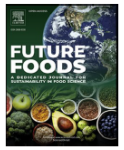

Assessing cellular agriculture potential: Population homogeneity and gene expression in cultured bovine mammary epithelial cells

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Utilising cultured primary bovine mammary epithelial cells for milk components production

PhD thesis
by
Zahra Sattari
November 2023

Department of Food Science
Aarhus University, Denmark

AARHUS UNIVERSITET

Sattari et al. (2024) Lactogenic treatment effects on milk synthesis genes and protein secretion in cultured bovine mammary epithelial cells. Future Foods, accepted.

Che et al. (2024). Proteomic study of secretomes from cellular agriculture for milk production. Discover Food, submitted.

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