



The **CYBERTONGUE**® Company



Optimizing Global Dairy Supply Chains: Enhancing Quality, Reducing Costs Improving Production Efficiency



Dairy Olympics ● 8th April 2025

TEAM



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- Biochemist
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Waste and Losses in the Dairy Supply Chain

Experts estimate that up to one sixth of the world's milk production is lost or wasted* before reaching the consumer

This represents a financial loss of up to USD \$150B per annum

Reducing/eliminating that waste is an opportunity to:

- increase milk supply to meet growing global demand
- take up to one quarter of one percent off all human GHG emissions
- improve dairy's profitability and customer value proposition

* NIZO and Detact Diagnostics 2021. "At-line milk protease testing supports production decision making and waste reduction." Pamphlet. p.1-5.

United Nations Sustainable Development Goals

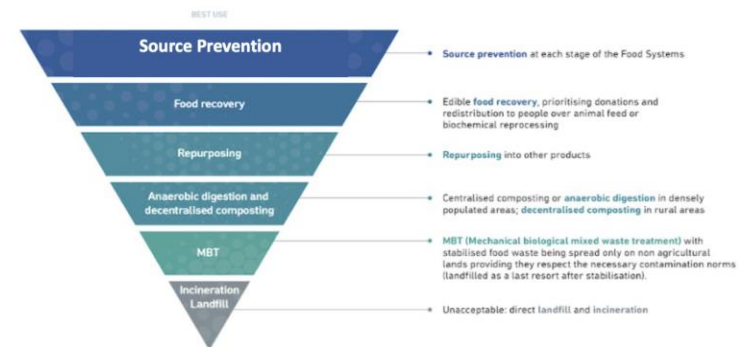
TARGET 12.3



HALVE GLOBAL PER CAPITA FOOD WASTE

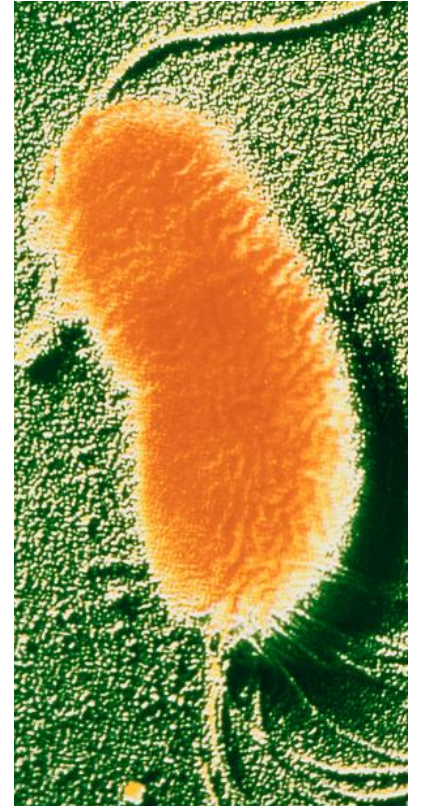
By 2030, halve per capita global food waste at the retail and consumer levels and reduce food losses along production and supply chains, including post-harvest losses.

Food Waste Hierarchy

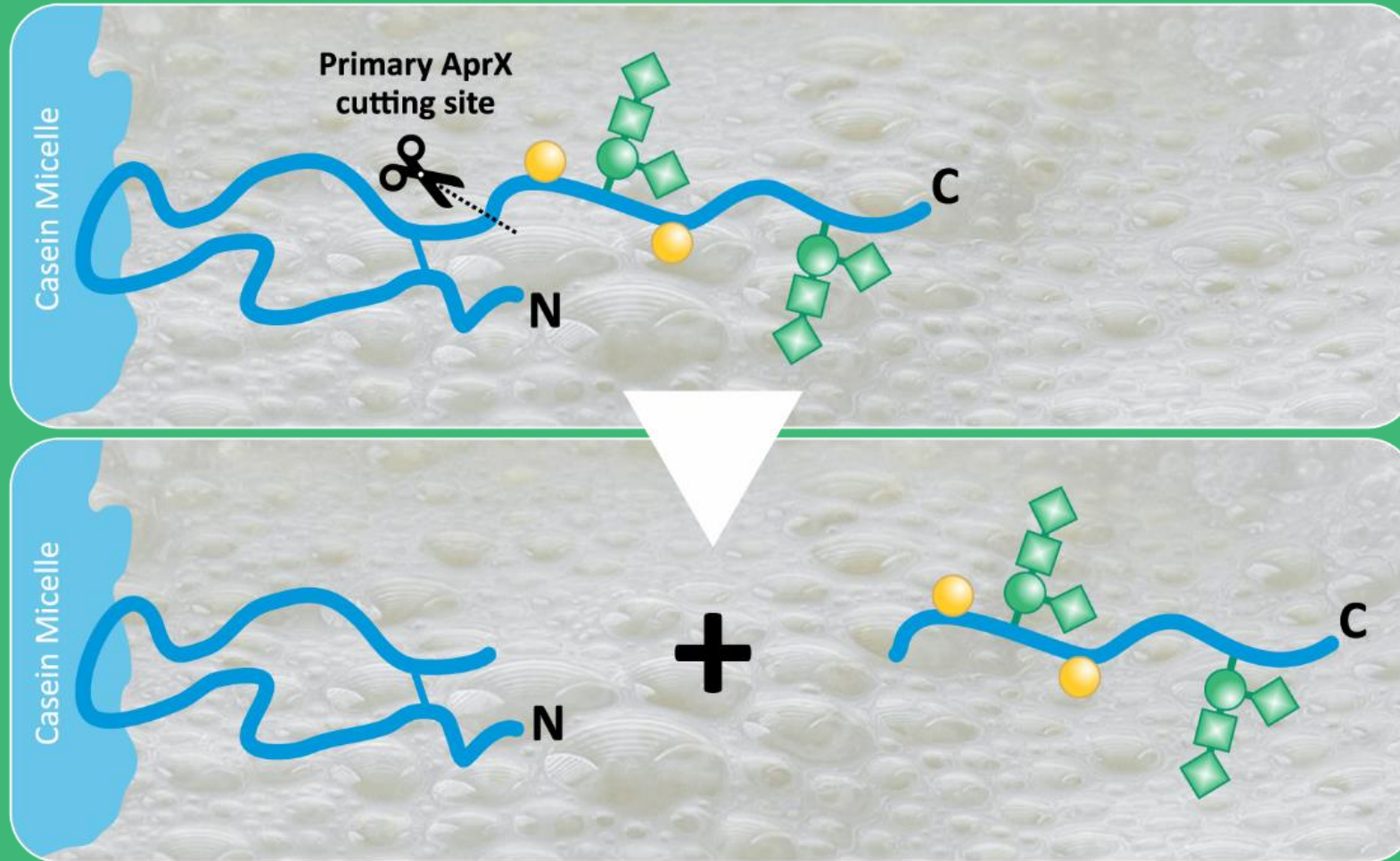


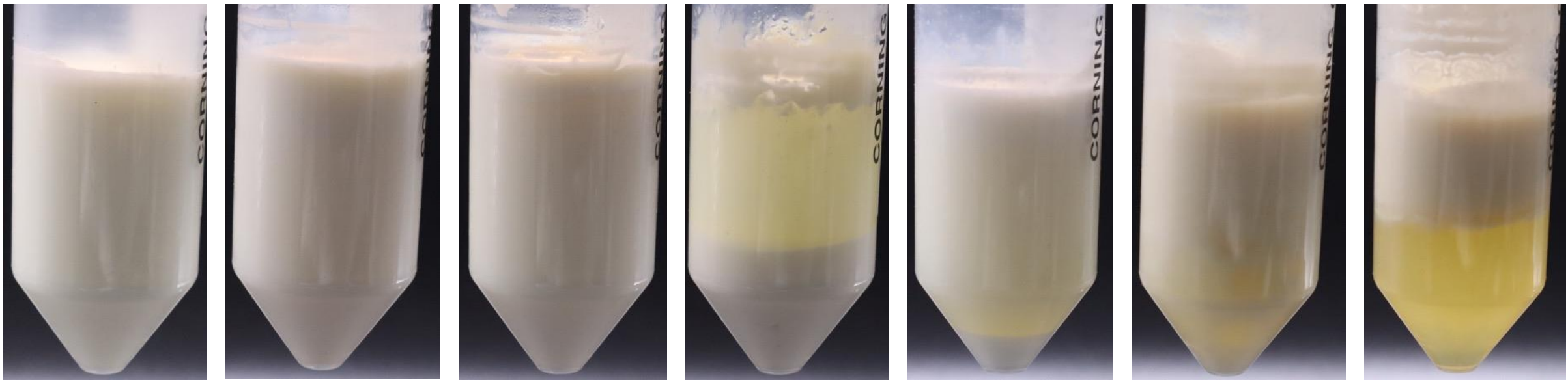
Proteases In Raw Milk Are a Significant Root Cause of Losses and Waste

- Plasmin/Plasminogen (from the cow) damages α - and β -caseins
- AprX (from Pseudomonads - right) damages κ -casein and reduces shelf life of UHT milk
- Subtilase/alcalase (from aerobic bacilli) damages all caseins
- Proteases affect cheese yield and flavour



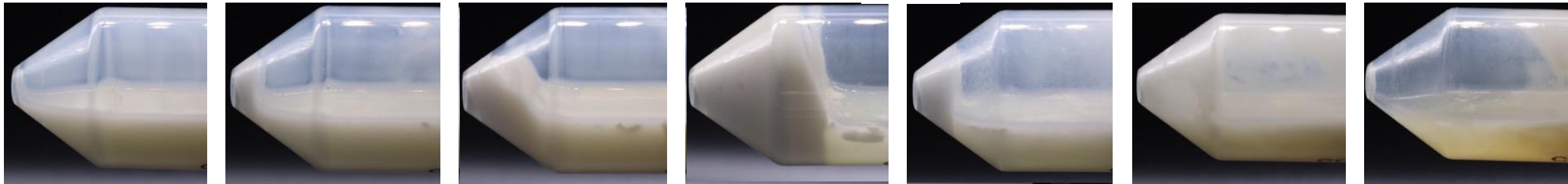
How AprX Damages Milk





4 °C

22 °C



Up to 1 year
Any level of
AprX activity

10 days

2 weeks

14 weeks

1000 μ U/mL
4 wks

100 μ U/mL
12 wks

1000 μ U/mL
12 wks

5 μ U/mL AprX activity

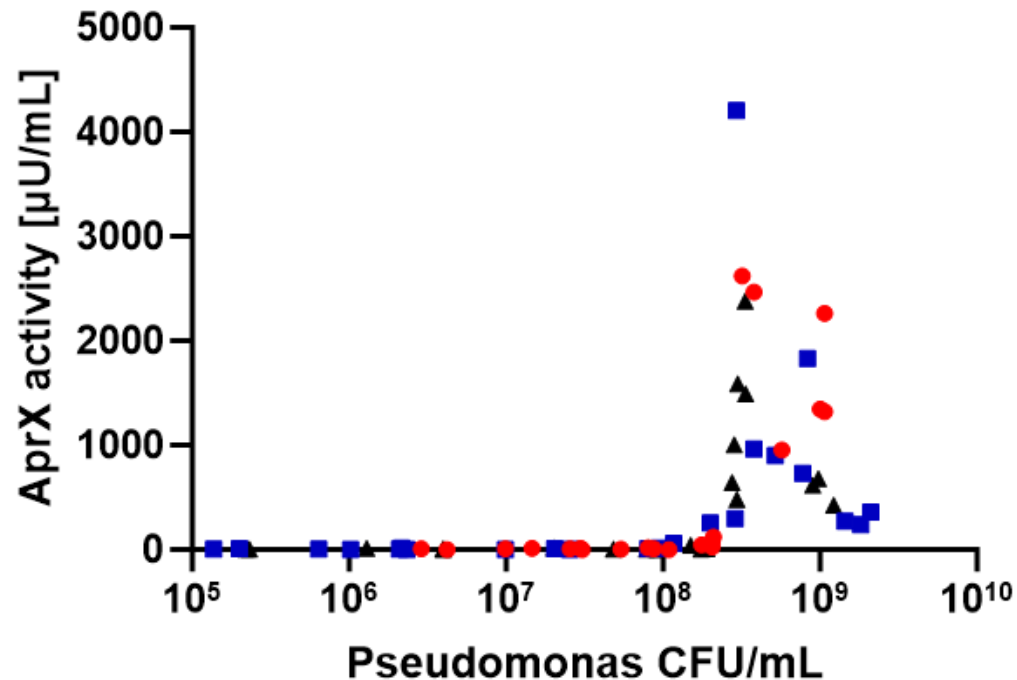
κ -casein selectively &
progressively targetted

at higher protease levels, α - and β -caseins
are also targetted

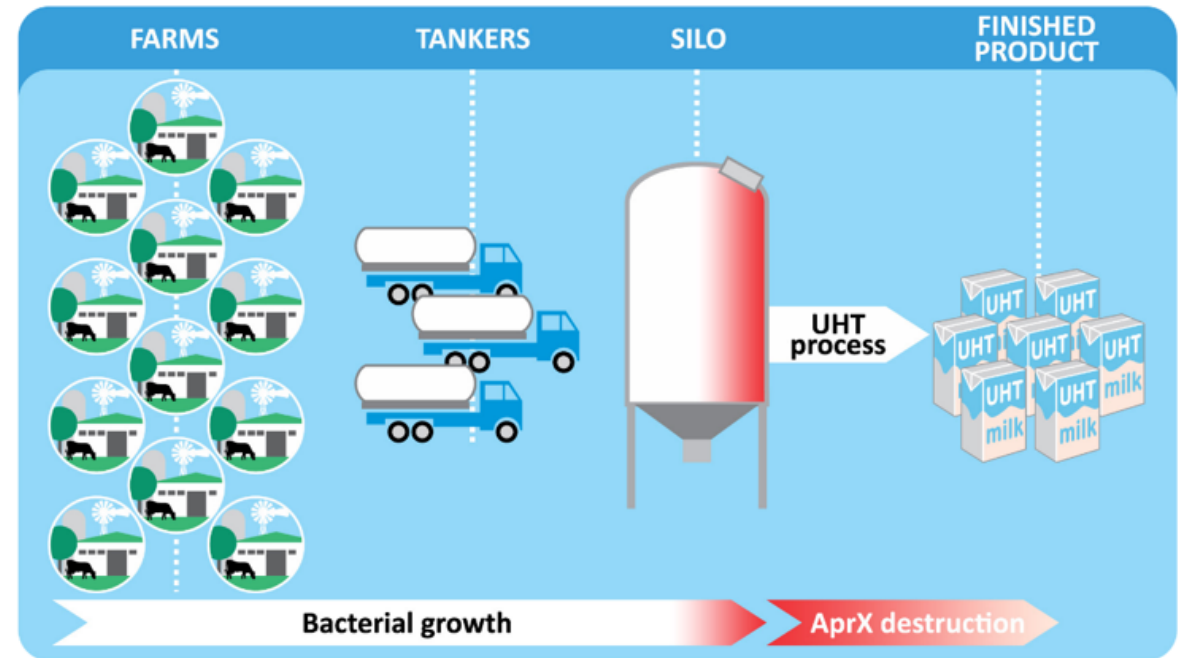
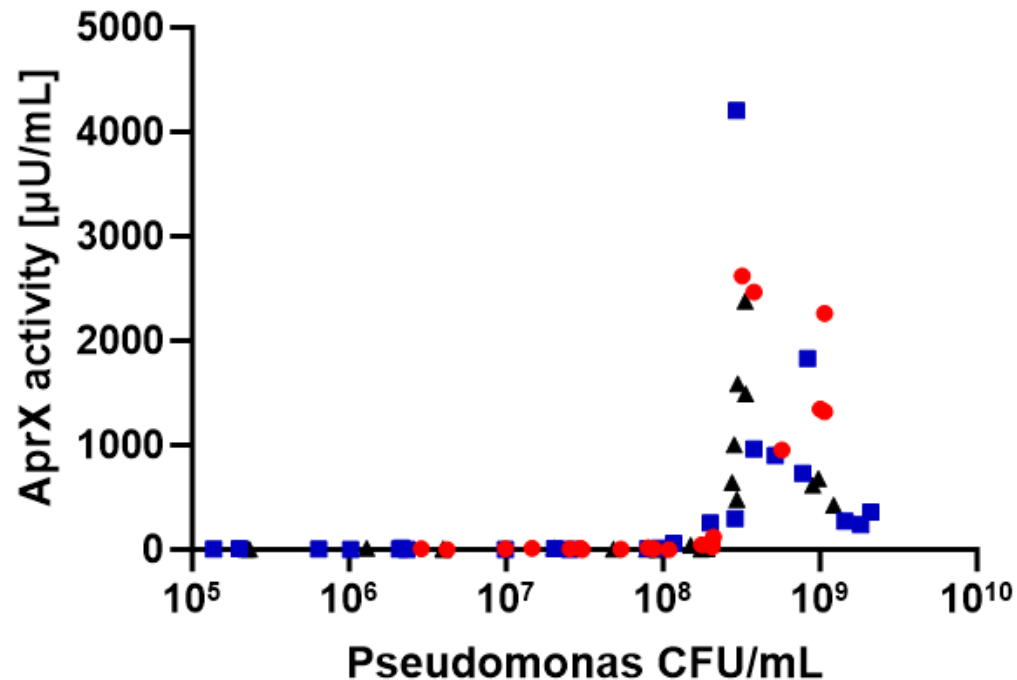
indiFOSS

ppb
technology™
The CYBERTONGUE® Company

Planktonic Growth of *Pseudomonas* spp. Induces AprX expression



Planktonic Growth of *Pseudomonas* spp. Induces AprX expression



Quorum-Induced AprX Expression Leads to Catastrophic Damage to Kappa Casein

- 100s to 1,000s of $\mu\text{U/mL}$ AprX activity in the raw milk
- 10s to 100s of $\mu\text{U/mL}$ AprX activity in UHT milk
- Rapid, catastrophic "sweet curdling", bitterness and/or clearing of UHT milk
- Yield loss and flavour defects in some cheeses
- These levels of protease cannot be cured in the heat exchanger
- You need to manage the quality of your raw milk BEFORE heat treatment

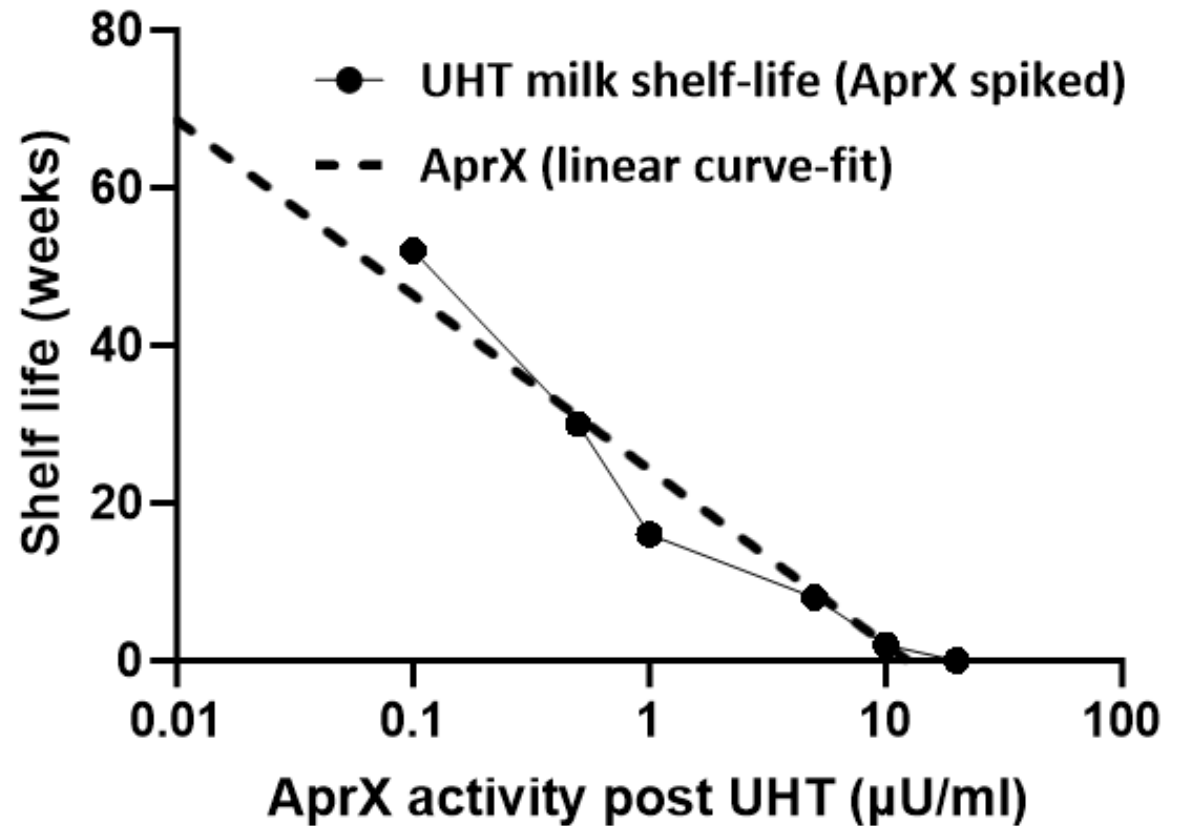


Biofilms Lead to **More Subtle** Loss of Shelf-Life

- At shelf-life over 3 months, planktonic expression of AprX is likely “too much” to have been the cause - BUT
- AprX expression is 100-fold higher in Pseudomonas biofilms than in the planktonic phase (Gieschler, 2022, PhD thesis)
- In collaboration with commercial partners, CYBERTONGUE® detected AprX activities of 10-40 $\mu\text{U/mL}$ in farm samples. Follow-up inspection of those farms revealed biofilms
- In-house experiments show that Pseudomonas biofilms express detectable levels of AprX protease

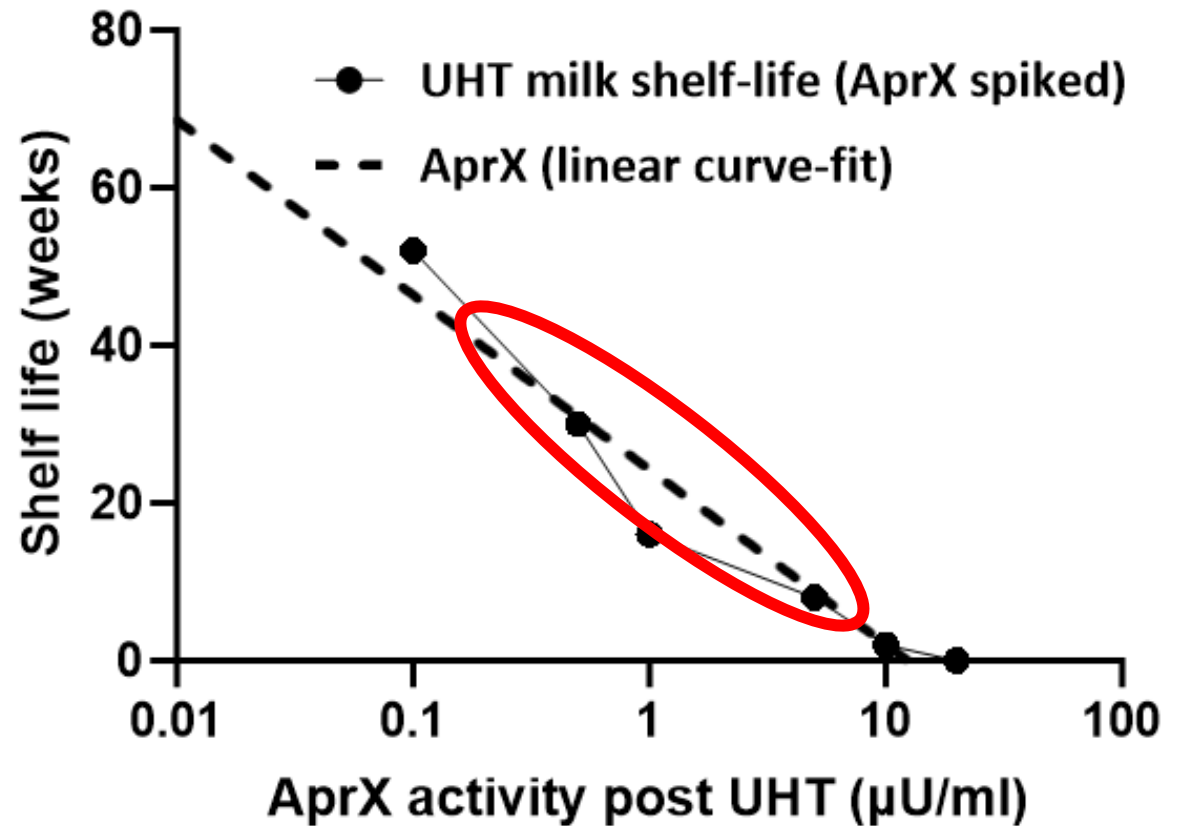
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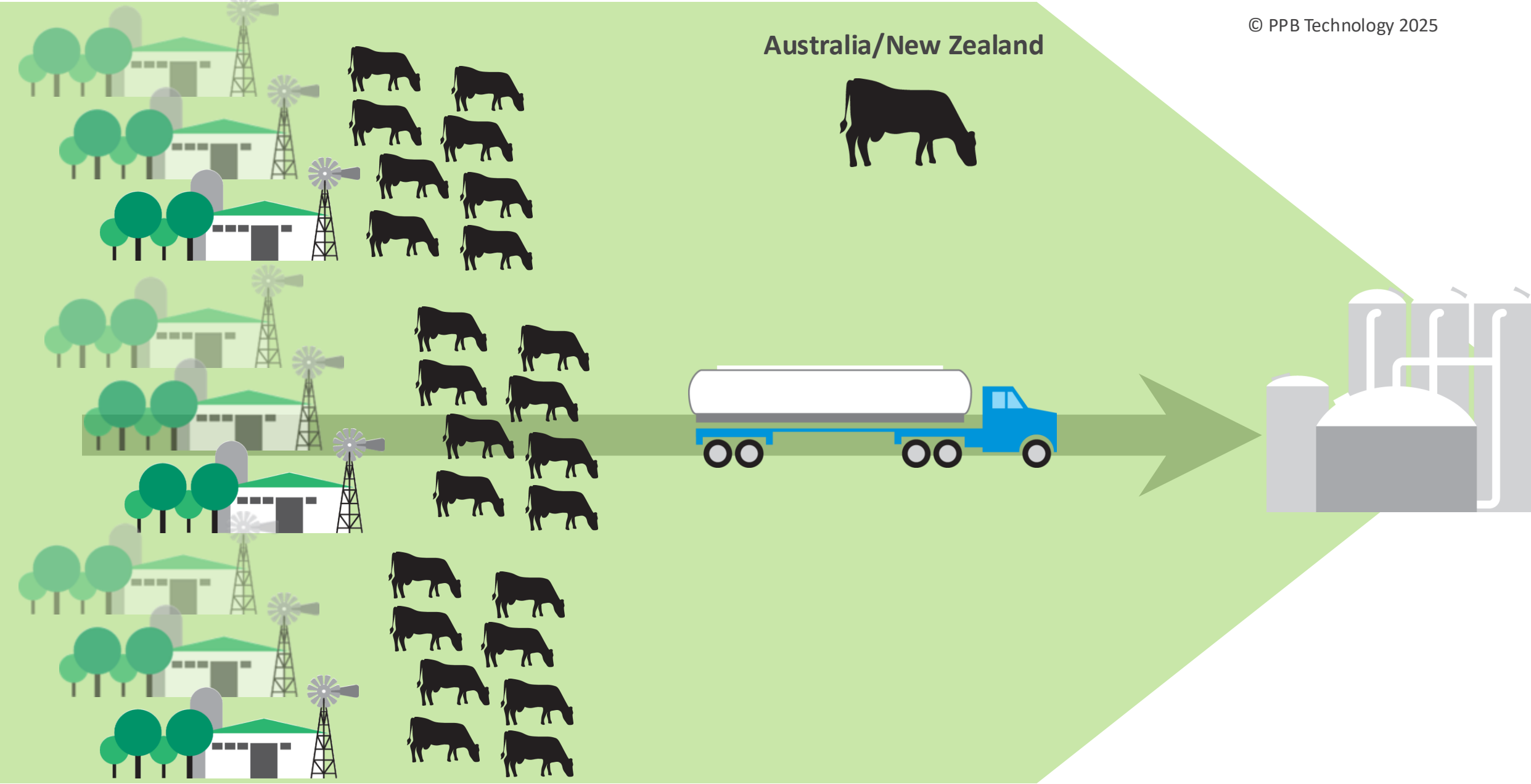
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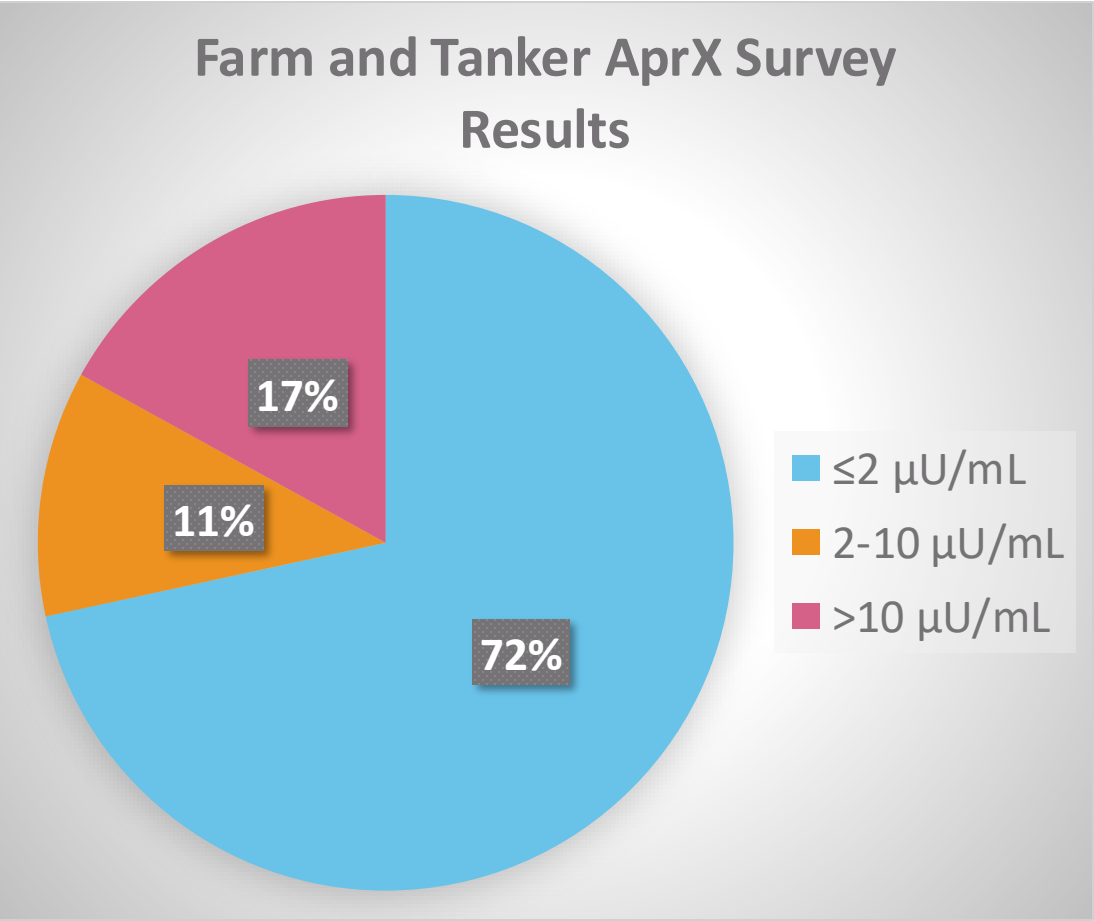
Australia/New Zealand

Dairy Farms

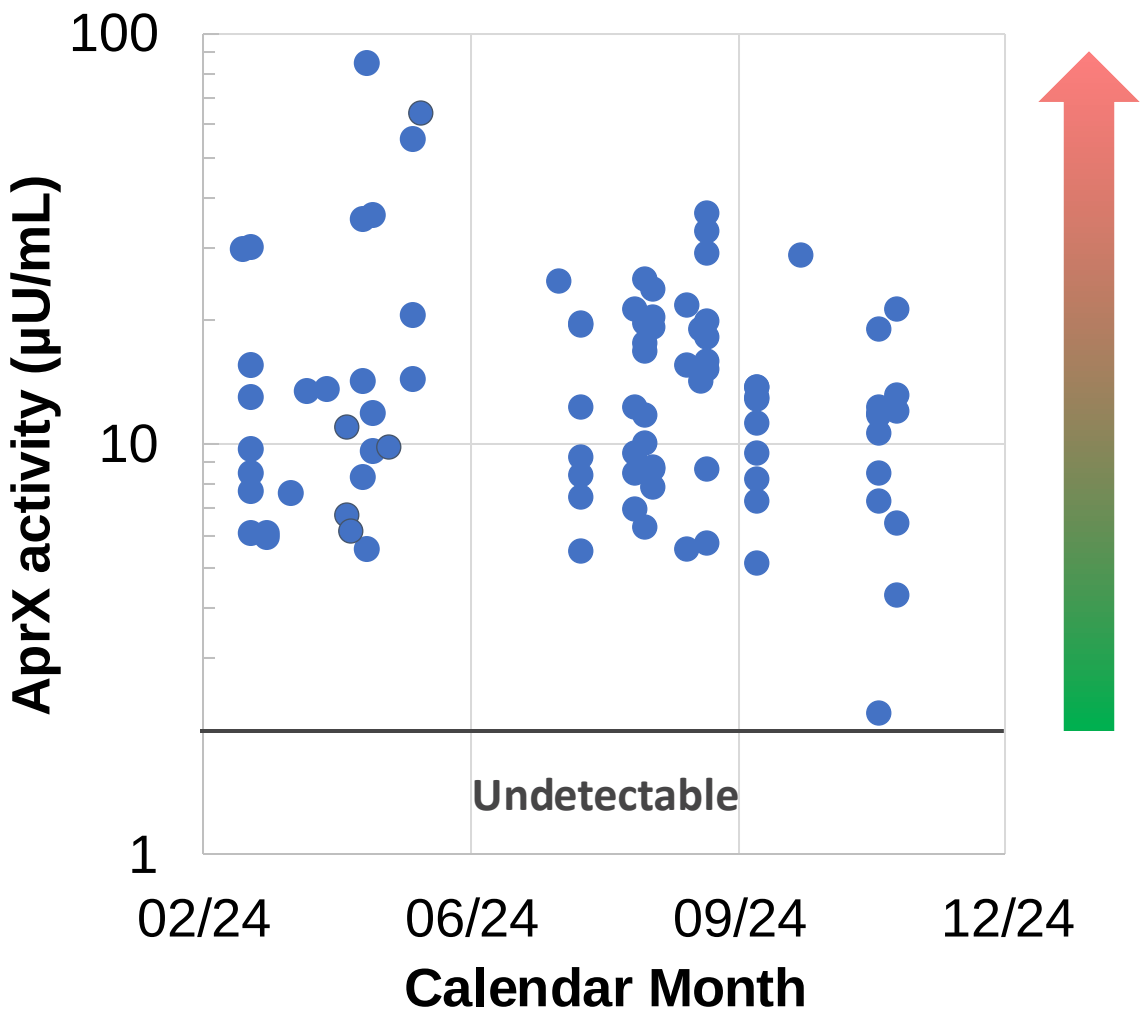
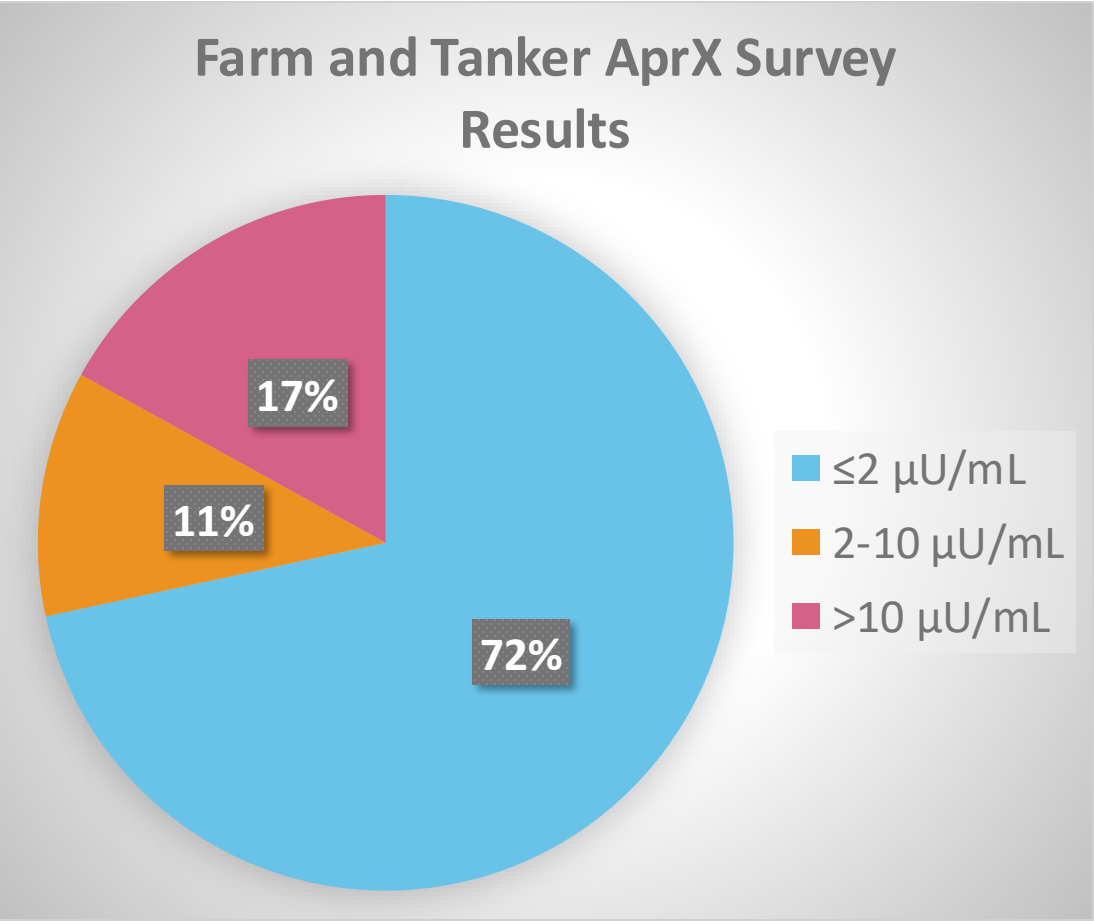
Tankers

Processing Plant

AprX Farm & Tanker Survey 2024 - Australia

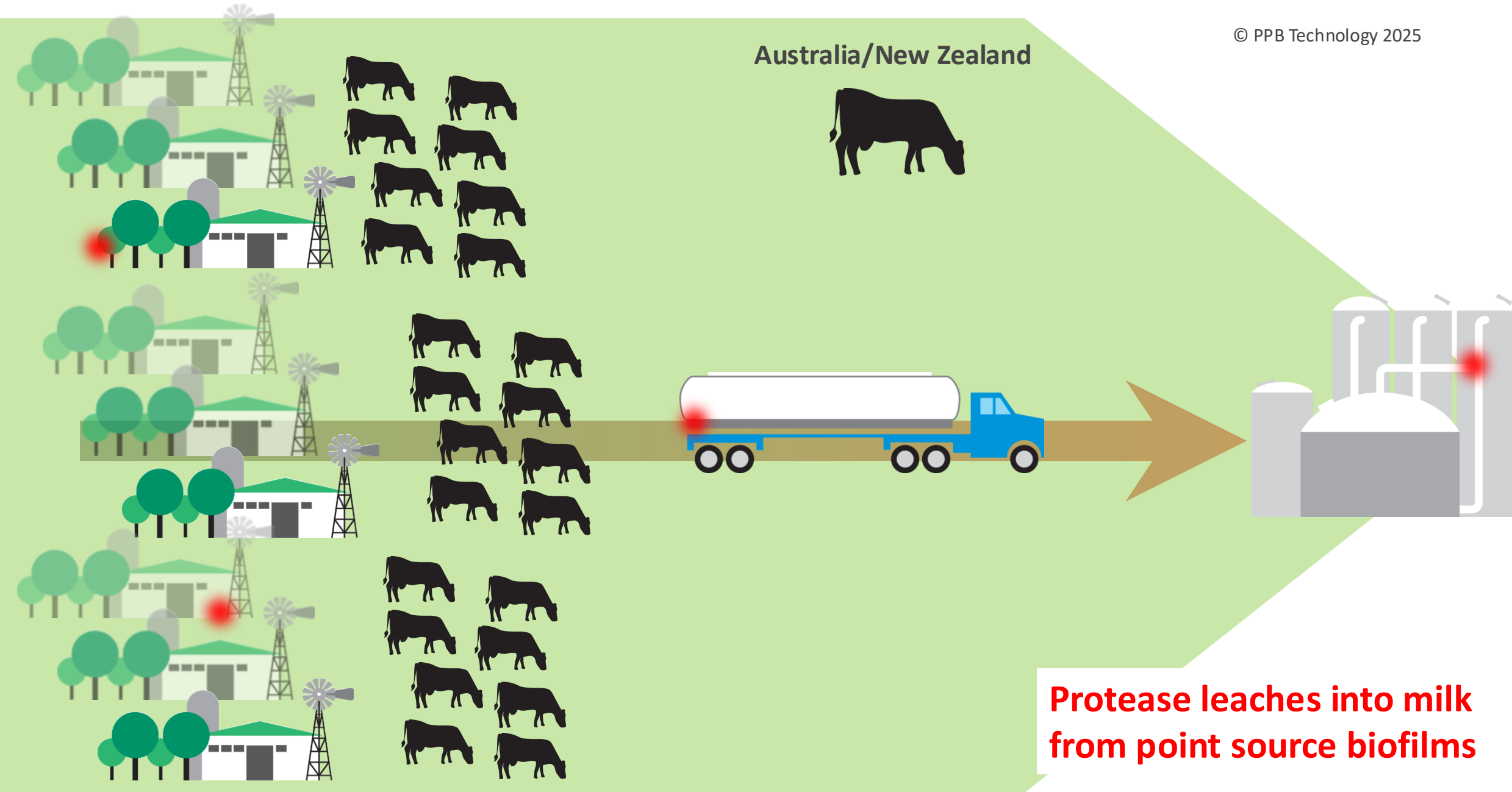


AprX Farm & Tanker Survey 2024 - Australia



Dilution and Destruction of AprX Also Matters

		v 1.2 02032025
Dilution of AprX into Finished UHT Product		
CYBERTONGUE® Limit of Detection (Standard Test)	100	μU/ml
CYBERTONGUE® Limit of Detection (Ultrasensitive Test)	2	μU/ml
% Uncontaminated Milk	92	%
% Contaminated Milk	8	%
% Effectiveness of UHT process (70-90%)	80	%
Highest Level of AprX in Finished Product (Standard)	1.74	μU/ml
Highest Level of AprX in Finished Product (Ultrasensitive)	0.032	μU/ml



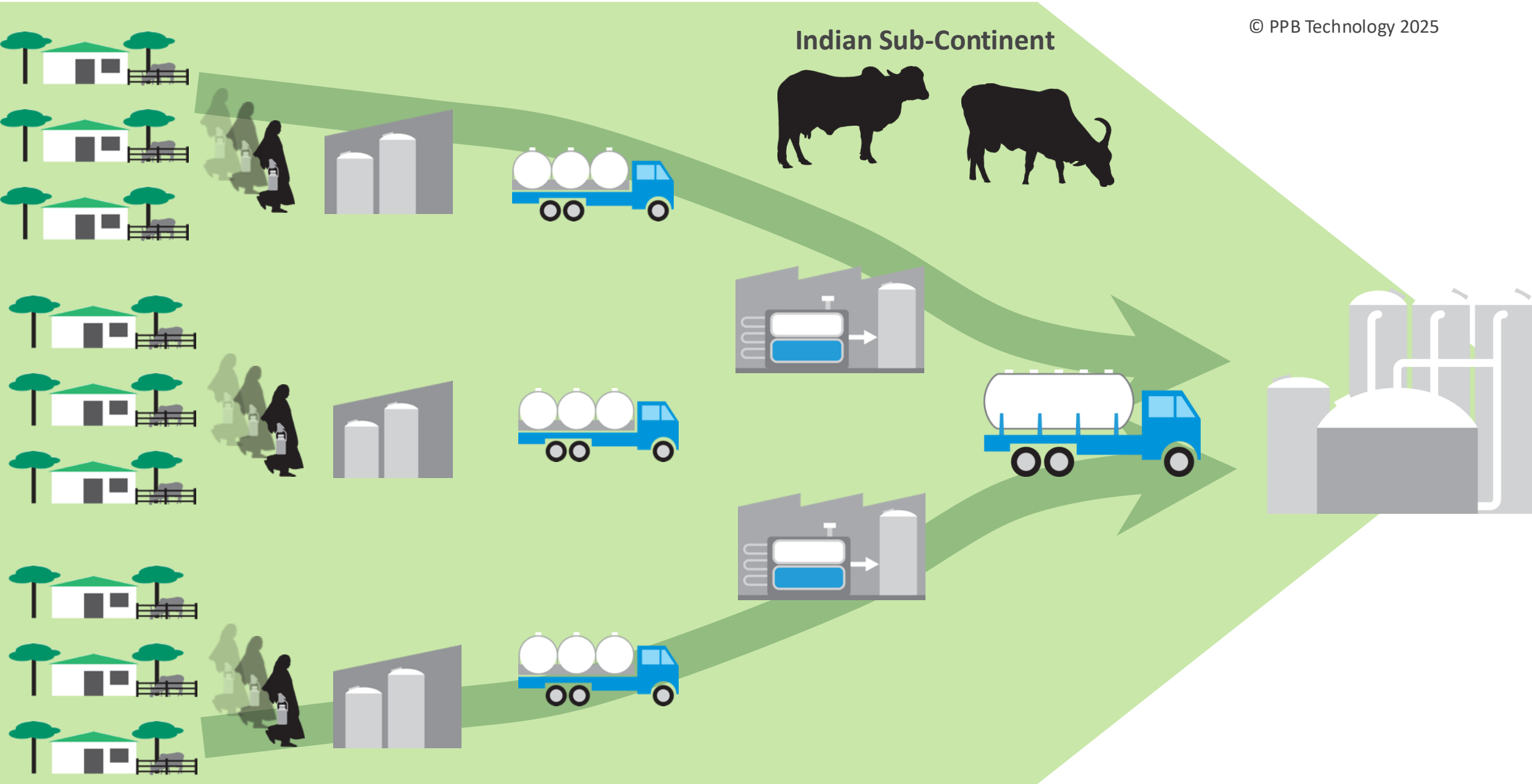
Australia/New Zealand

Dairy Farms

Tankers

Processing Plant

Protease leaches into milk from point source biofilms



Indian Sub-Continent

Dairy Farms

Churns
Carried

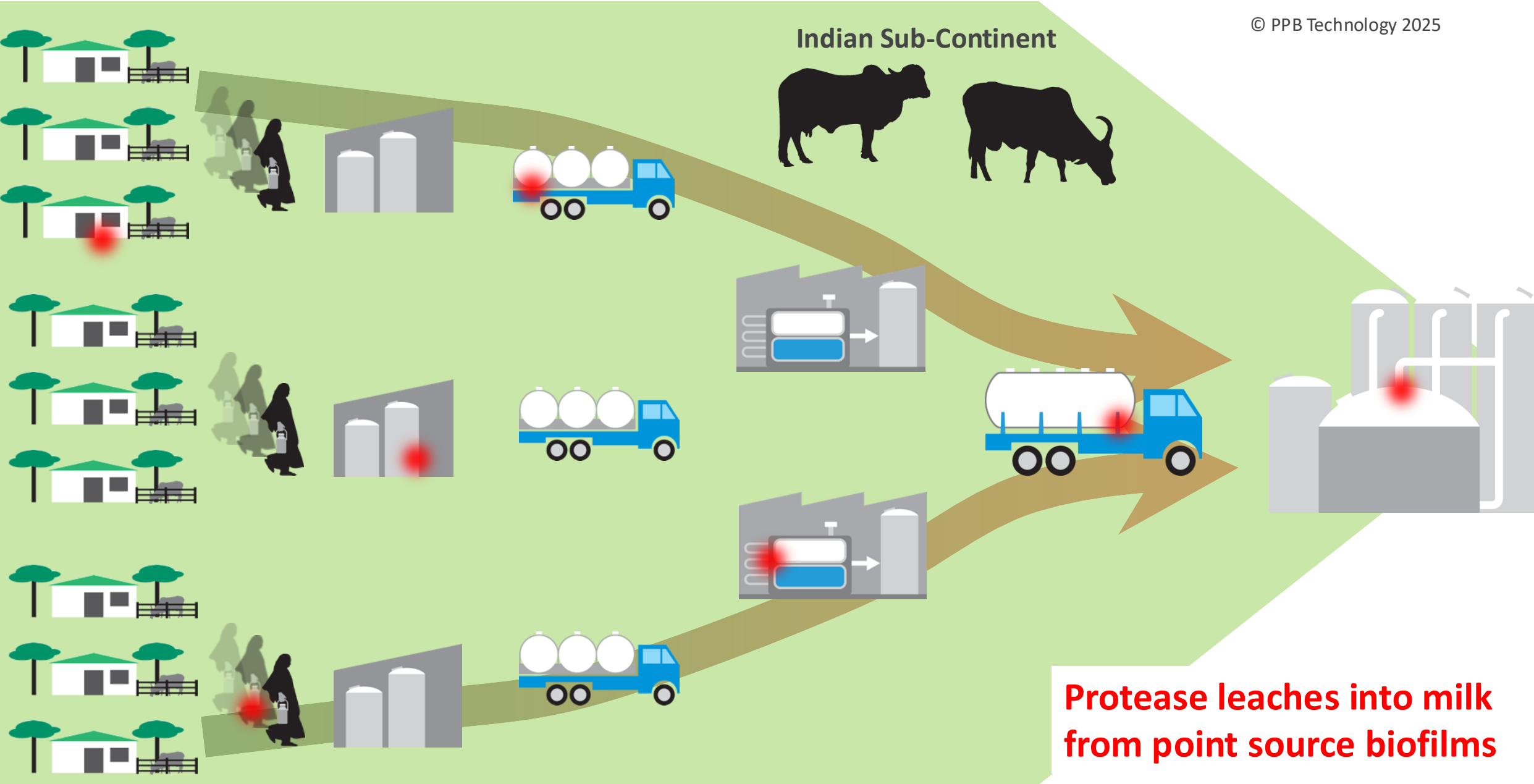
Collection
Centres

Tankers

Bulk Milk
Coolers

Tankers

Processing Plant



Dairy Farms

Churns Carried

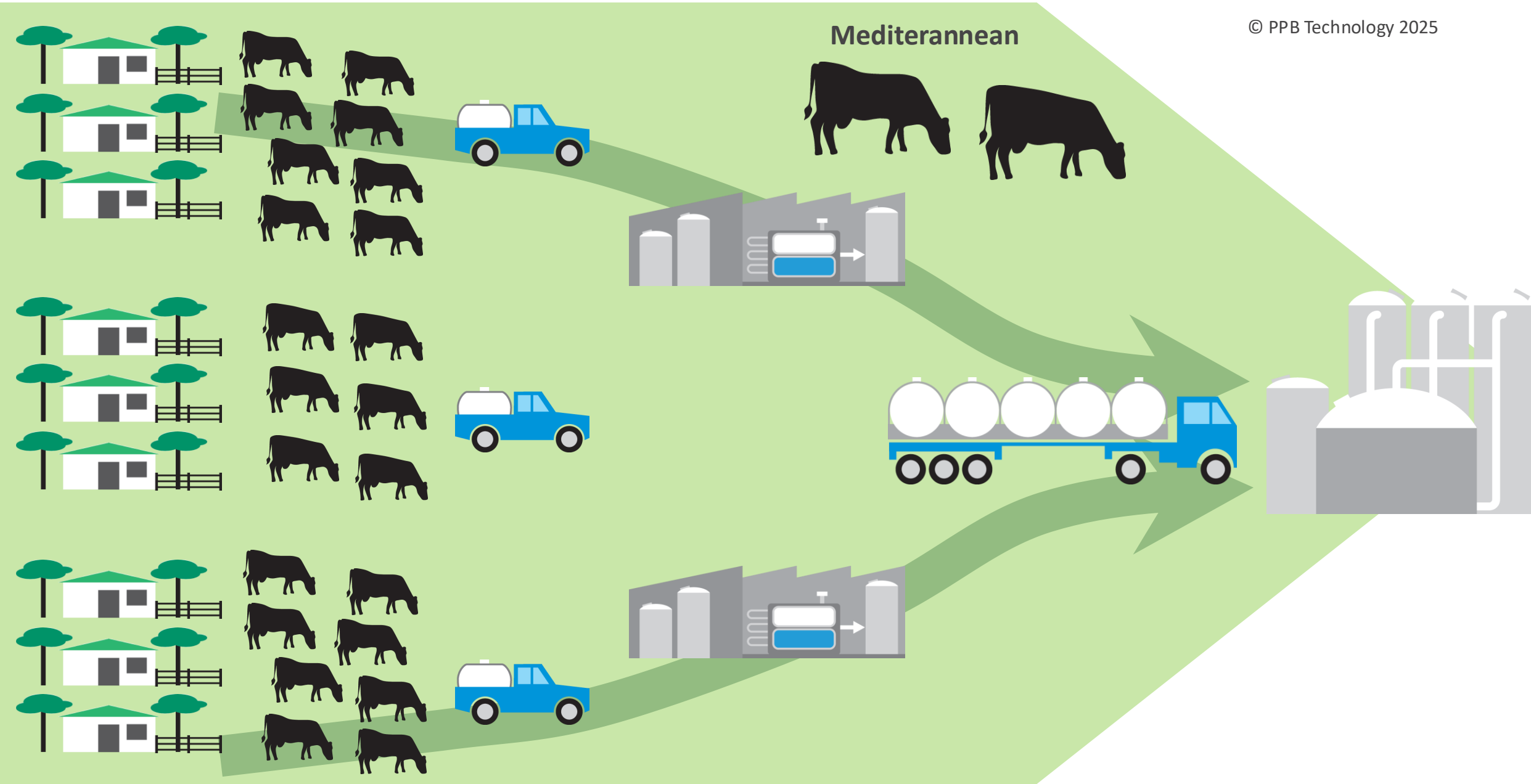
Collection Centres

Tankers

Bulk Milk Coolers

Tankers

Processing Plant



Mediterranean

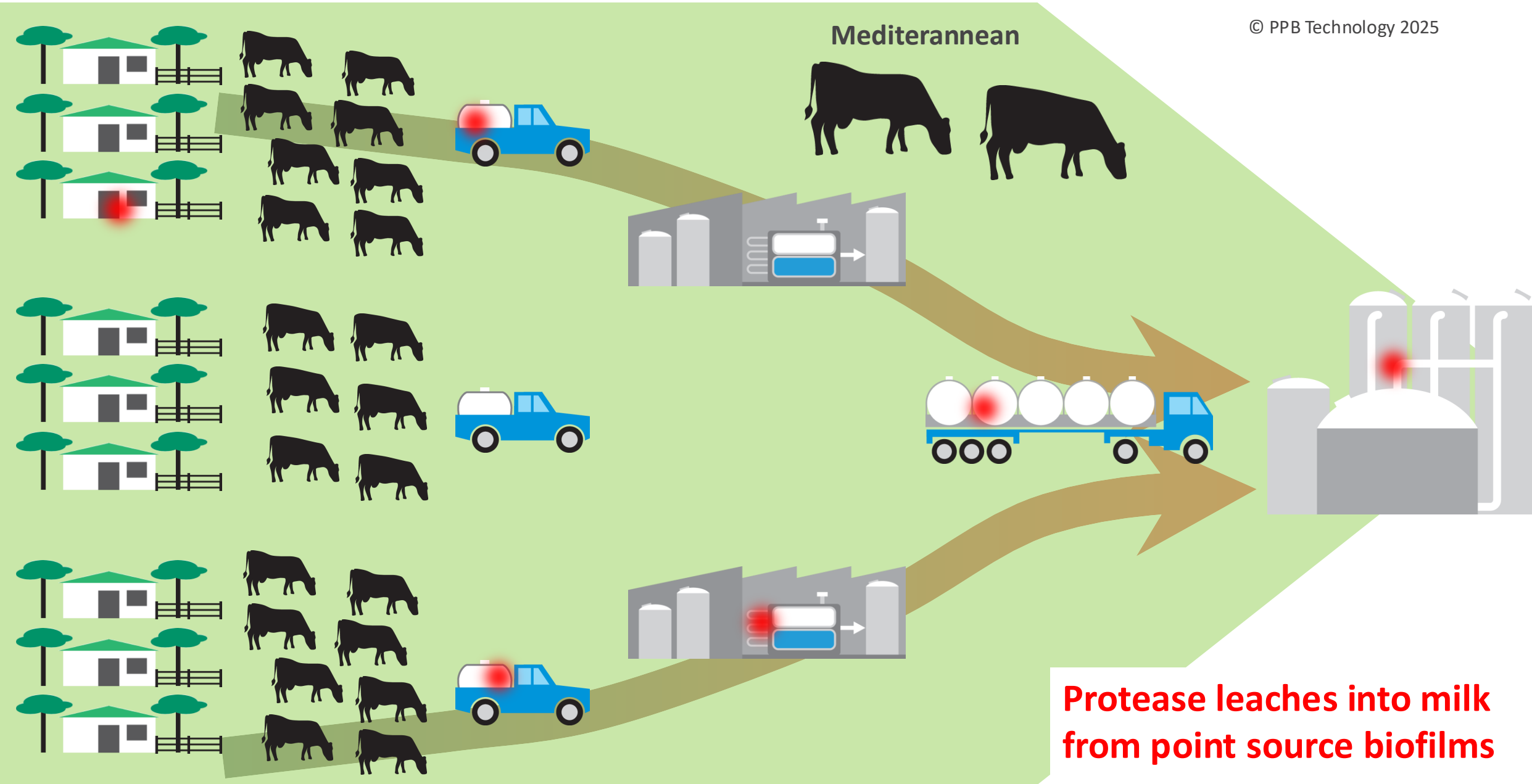
Dairy Farms

Mini-tankers

Collection
Centres/Bulk Milk
Coolers

Tankers

Processing Plant



Mediterranean

**Protease leaches into milk
from point source biofilms**

Dairy Farms

Mini-tankers

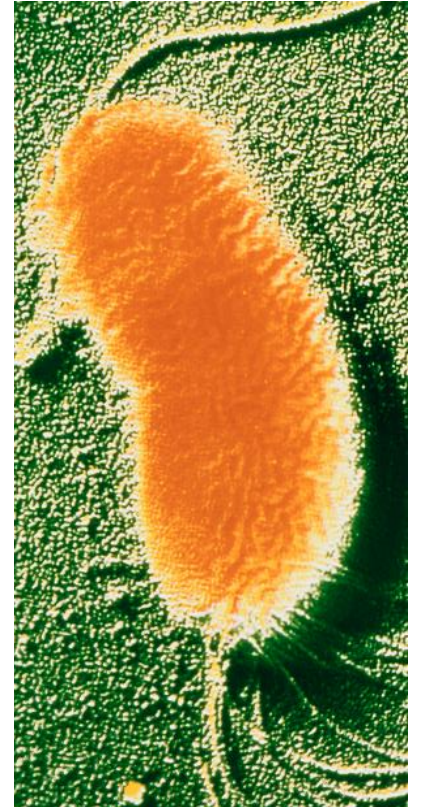
Collection
Centres/Bulk Milk
Coolers

Tankers

Processing Plant

Summary

- Heat stable proteases are a major cause of losses and waste in the dairy sector
- AprX protease from *Pseudomonas* is the main culprit in chilled milk supplies
- AprX protease activity must be held at low levels in raw milk destined for UHT milk or cheese-making, because...
- Heat processing on its own is not sufficient to eliminate the proteases
- What gets measured can be managed



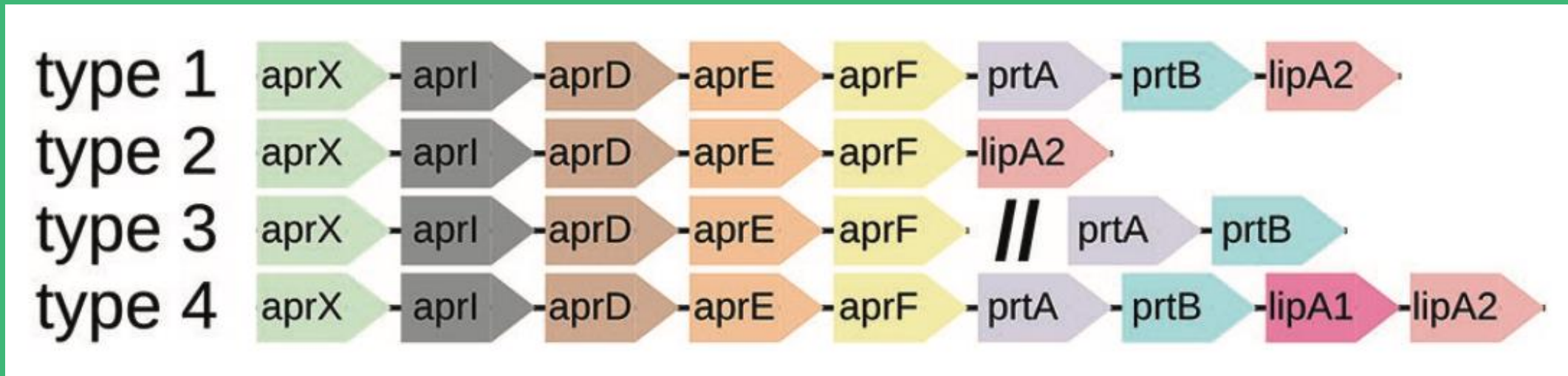
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Genetics of AprX Expression in *Pseudomonas* spp.

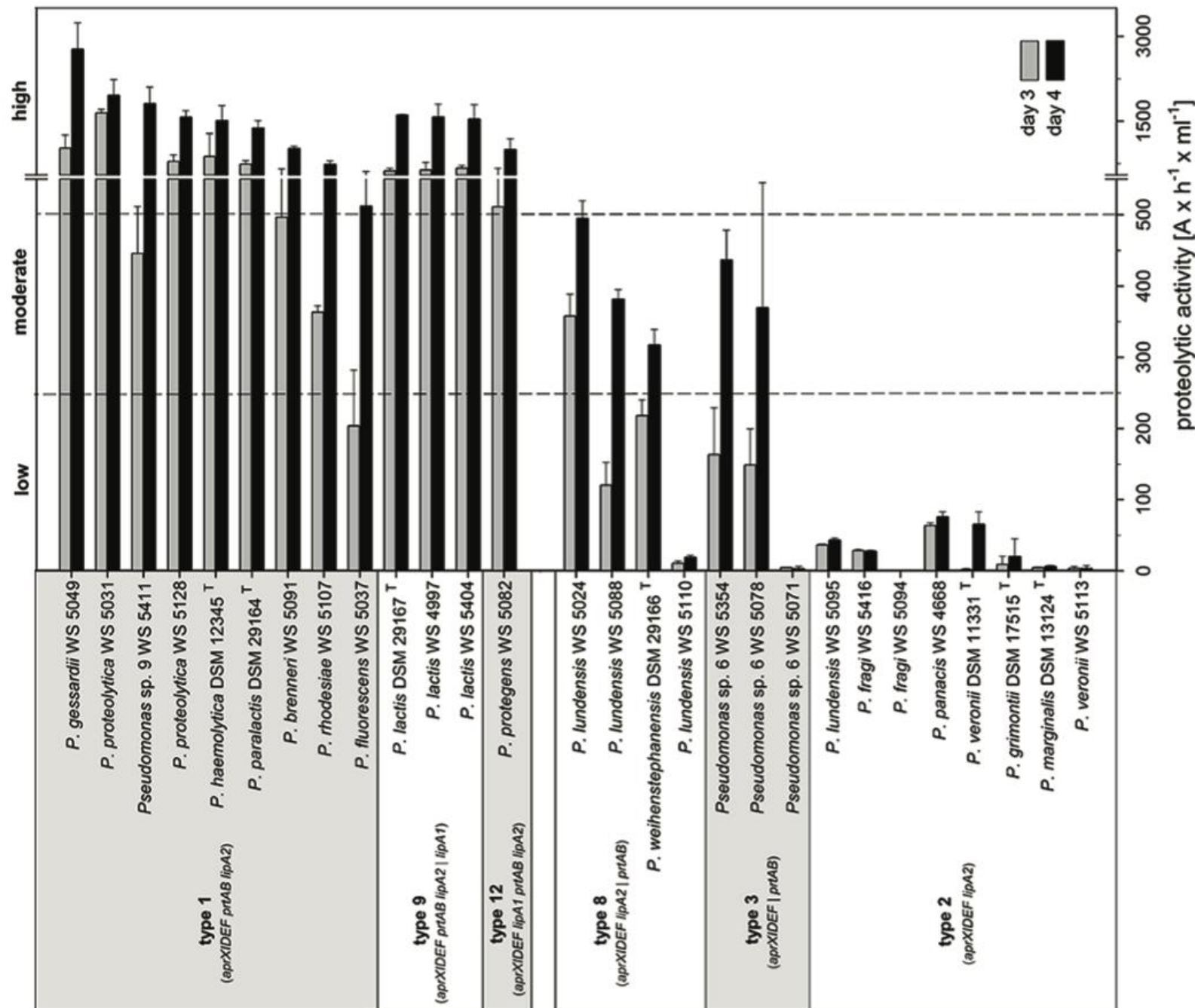


Genetic Organization of the *aprX-lipA2* Operon Affects the Proteolytic Potential of *Pseudomonas* Species in Milk

Christopher Maier^{1†}, Christopher Huptas^{2†}, Mario von Neubeck^{1‡}, Siegfried Scherer^{1,2}, Mareike Wenning³ and Genia Lücking^{1*}

 **frontiers**
in Microbiology

ORIGINAL RESEARCH
published: 10 June 2020
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ORIGINAL RESEARCH

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Review

Heat stability of indigenous milk plasmin and proteases from *Pseudomonas*: A challenge in the production of ultra-high temperature milk products

Marina Stoeckel ^{a,*}, Melanie Lidolt ^a, Timo Stressler ^b, Lutz Fischer ^b, Mareike Wenning ^c, Jörg Hinrichs ^a

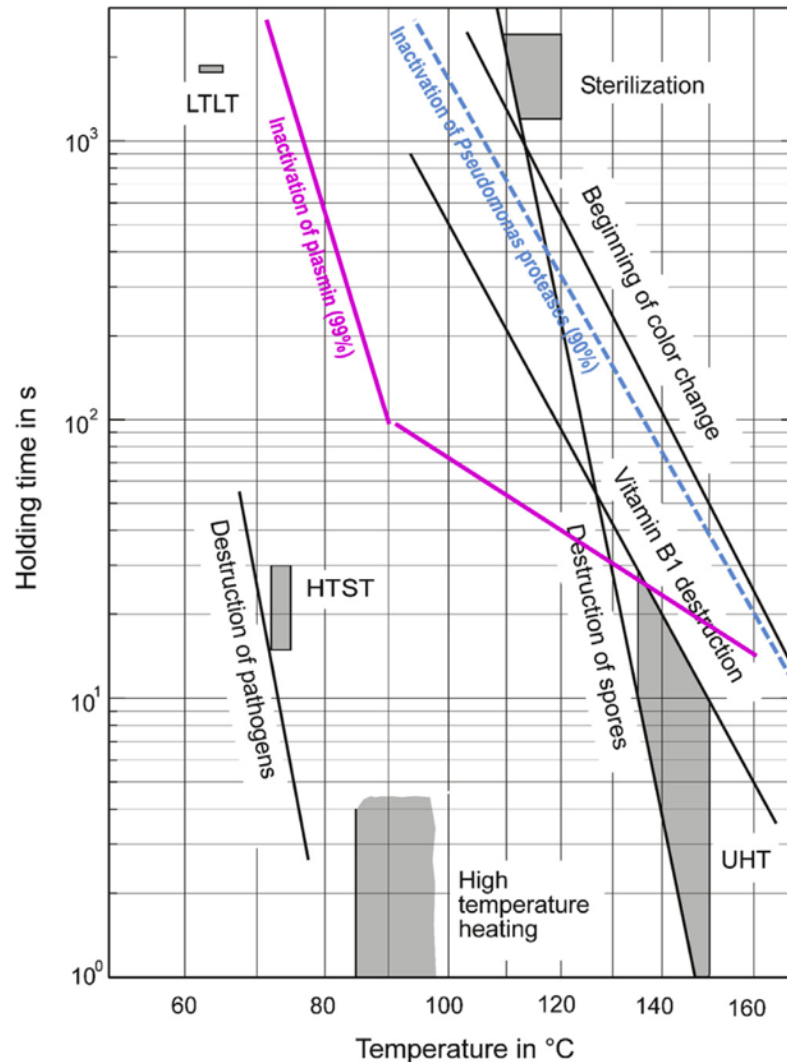
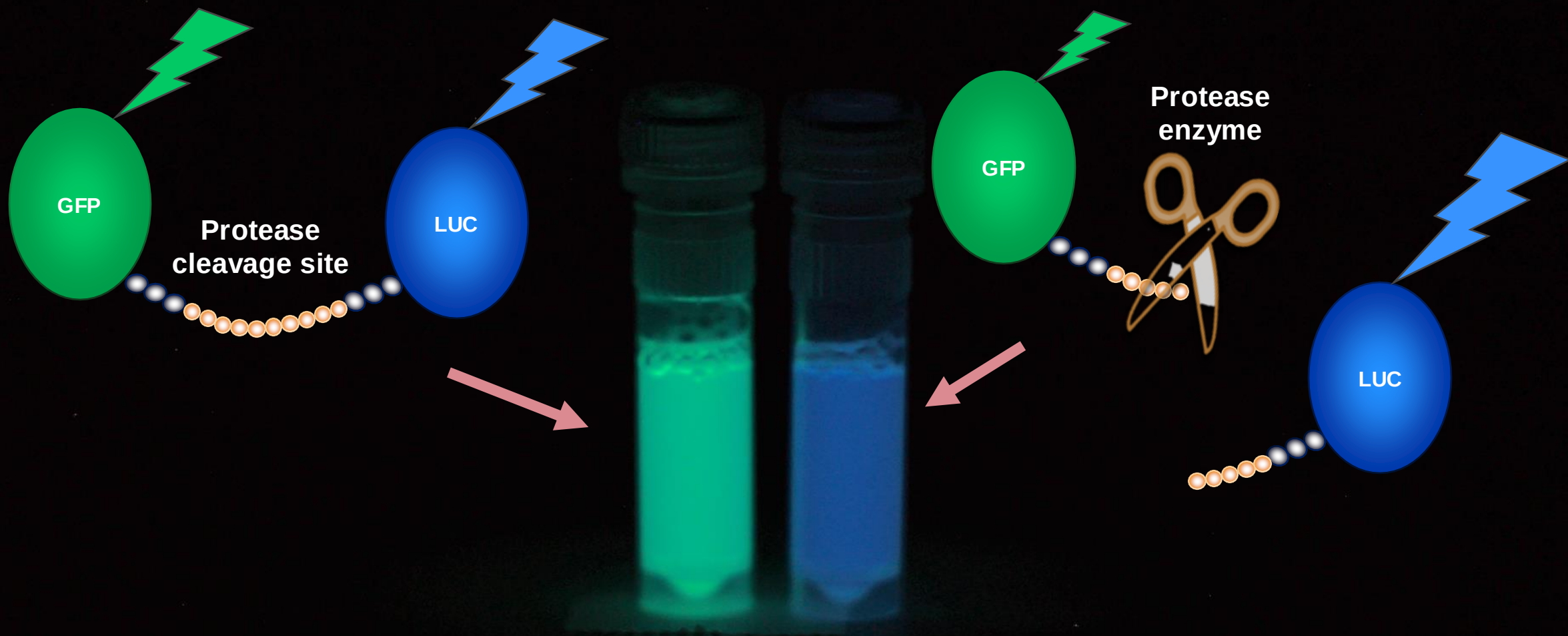


Fig. 1. Inactivation of plasmin by 99% (solid line) and of the proteases from *Pseudomonas* by 90% (dashed line) in milk (modified from Kessler, 2002). The mean values from Tables 1 and 2 were used for the kinetic data.



cream or oil
assay tube using the
ed volume pipette

next

back



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