

Study on Veronal buffer replacement (CFT)

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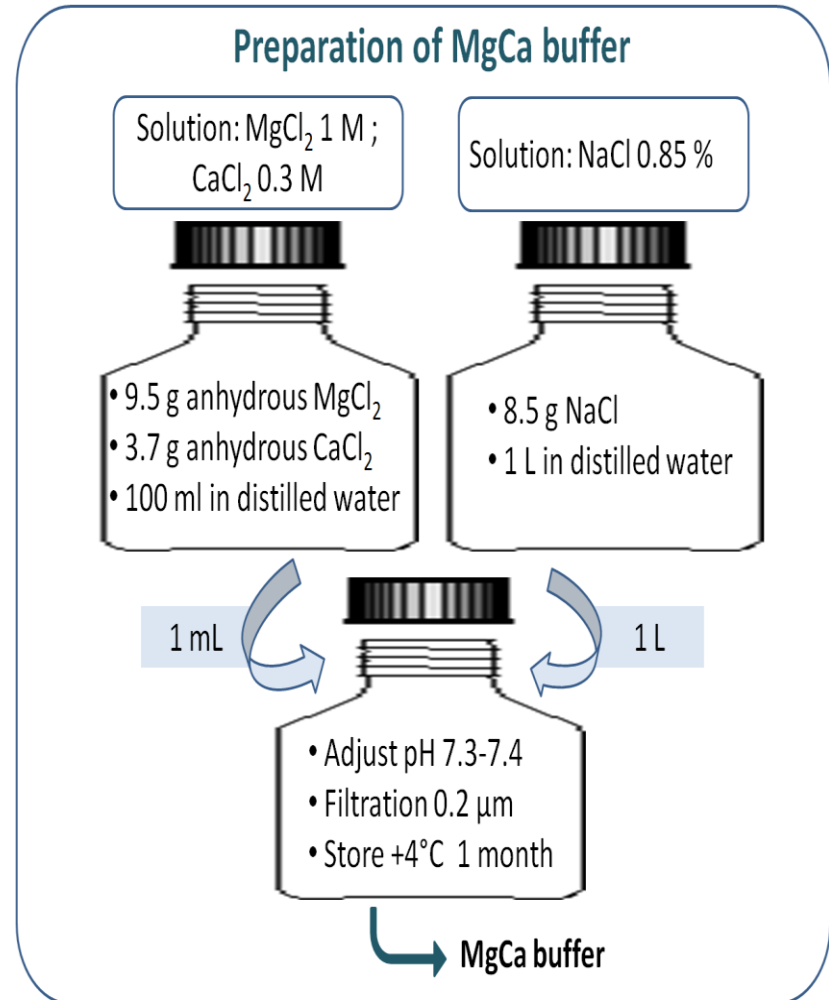
Background

- Recent ban on barbital use
 - ☞ find an alternative of VB for CFT?
- Alternative : **MgCa buffer** (Alton, 1988)
- Contains only magnesium and calcium diluted in NaCl
 - ☞ **non-toxic alternative** 😊
- Assessement of MgCa in comparison to VB

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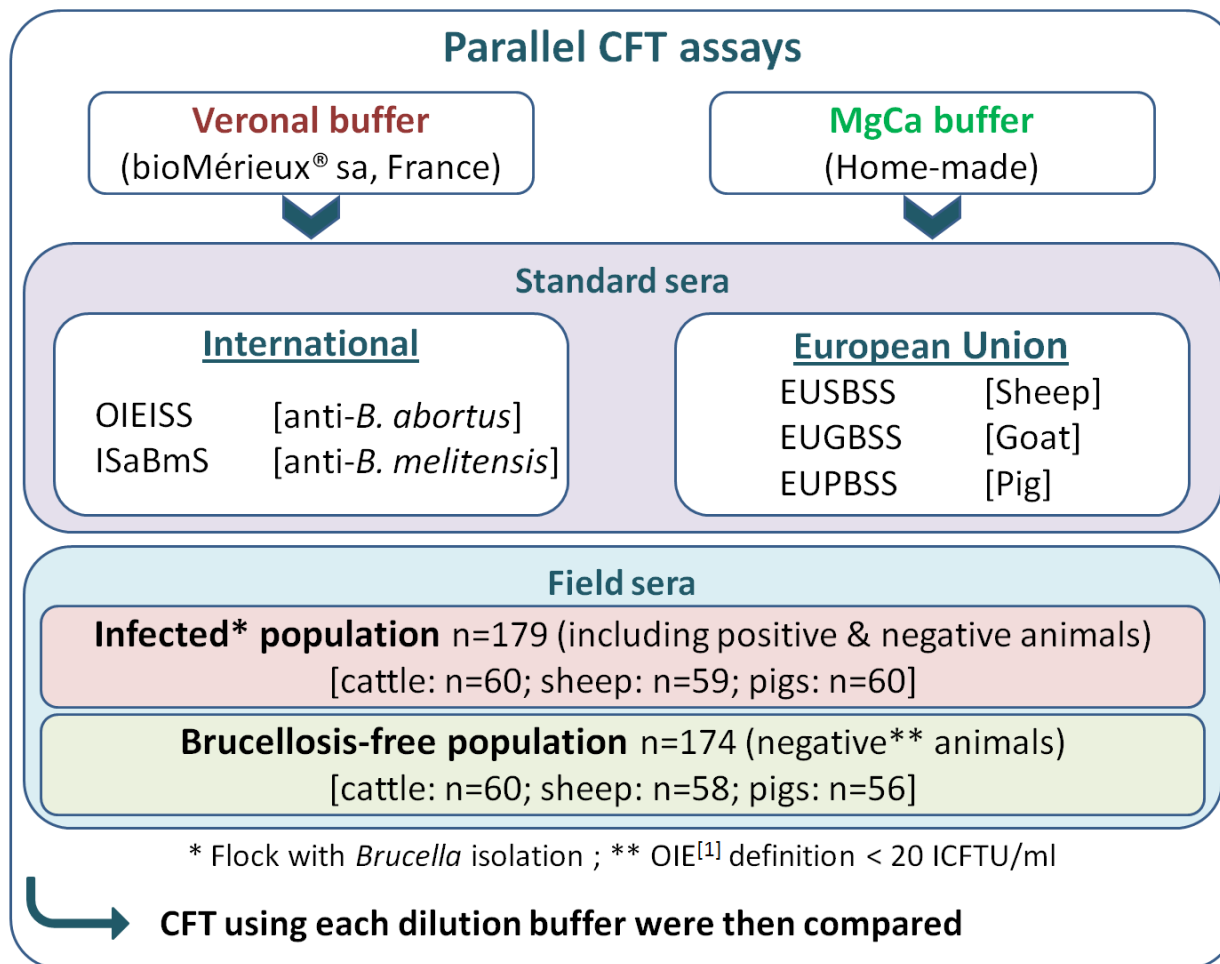
Composition & preparation

- 1 L of NaCl 0.85%
- 1 ml of stock solution MgCl₂ [1M] and CaCl₂ (2H₂O) [0.3M]
- Adjust the pH strictly between 7.3 and 7.4 (critical !!!!)
- Filtration on 0.22 µm (replacement of sodium azide) → control of bacterial contamination
- Storage at 5 ± 3° C 1 month
 - ☞ Safe and simple composition
 - ☞ Critical adjustment of the pH



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Protocol



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Results

Standard sera

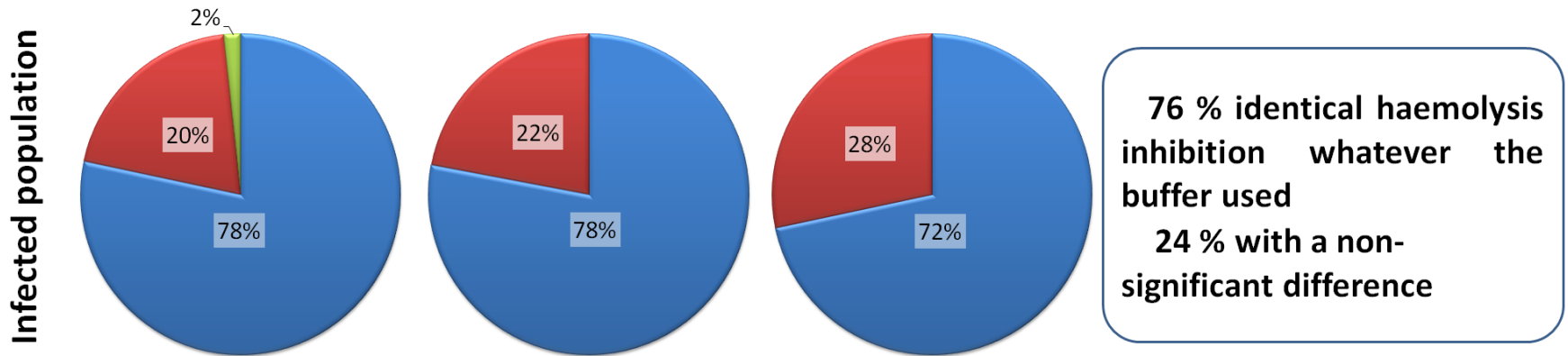
	International		European		
	OIEISS	ISaBmS	EUSBSS	EUGBSS	EUPBSS
VB	1493	533	426	267	853
MgCa	1493	640	426	320	533

[in ICFTU/ml]

- **Similar titres** whatever the dilution buffer used
- The difference between results with both buffers **never exceeded one dilution factor ($\frac{1}{2}$)**, i.e. not significant for CFT.

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Results



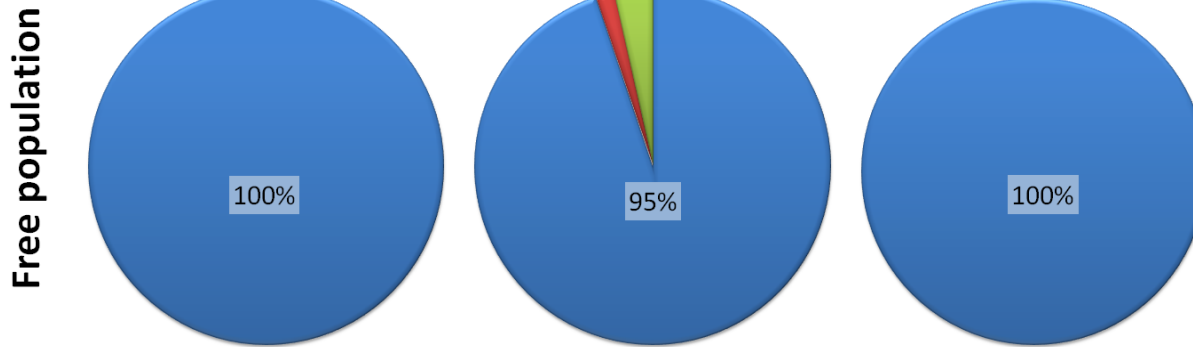
■ Exactly the same titre (ICFTU/ml)

■ Difference < 1 dilution factor

■ Anti-complementary in both buffers

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Results



98 % similar haemolysis inhibition whatever the buffer used
2 ovine sera were anti-complementary with both buffers



■ Exactly the same titre (ICFTU/ml) ■ Difference < 1 dilution factor ■ Anti-complementary in both buffers

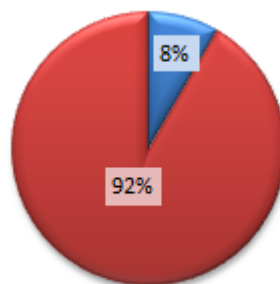
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Results *B. ovis* CFT

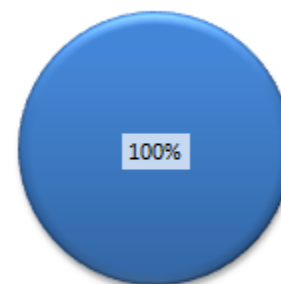
Standard	
	ISaBoS
VB	100
MgCa	167

[in ICFTU/ml]

Infected population



Free population



■ Exactly the same titre (ICFTU/ml) ■ Difference < 1 dilution factor



Difference observed probably linked to different results of complement titration with VB and with Mg Ca

Study on Veronal buffer replacement

- No significant difference was observed between veronal or MgCa buffers:
 - ✓ International or EU standard sera,
 - ✓ field sera,
 - ✓ whatever the host species (cattle, sheep or pigs).

MgCa buffer = non-toxic, easy to produce & cheap

☞ suitable alternative to the hazardous VB in the OIE approved CFT.

- Validity period

(1 month at $5 \pm 3^{\circ}$ C)

Further work : proposed for inclusion in the next revision of the OIE Terrestrial Manual.

☞ *If possible should be confirmed in as many NRLs as possible on serum collection of each animal species (especially if SOP \neq EURL SOP)*