Influence of release pad impregnation on latex conjugate release

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Objective

=> To identify release pad impregnation conditions that ensure quantitative conjugate release as well as membrane blocking "on the fly"

Method

- => Impregnation buffer components are systematically varied
- => release, background, as well as test and control line intensity are quantified
- => statistical evaluation of the results (Design of Experiment)
- => detailed study of main factor(s)



Factors for screening experiment:

Factor	Buffer	Tween 20	BSA	Succrose
	Molarity	Conc.	Conc.	Conc.
	[mMol]	[%]	[%]	[%]
high	200	0,5	2	3
low	10	0,01	0	0



Experimental procedure:

- Soak release pad with different buffer mixtures => dry @20°C for at least 4 hours
- Add conjugate particles to release pad => dry @20°C for 4 hours
- 3. Build hCG test strips
- 4. Evaluate performance <u>during the run:</u>
 => observe running time and flow front separation
- 5. Evaluate performance <u>on the dry strip</u>:
 - => scan dry strips and use image analysis tools to quantify conjugate release, background, and line intensity
- 6. Use statistical evaluation program to identify main factors and interactions (Design of Experiment)



Main effects plot for release and background





Main effects plot for test- and control-line intensity





Influence of BSA (detailed investigation)



Influence of BSA on release and background





Influence of BSA on test- and control-line intensity





Influence of BSA on running time







=> good latex conjugate release and low background only for impregnation with high BSA and high succrose. Tween 20 alone is not sufficient.



Comparison to gold conjugate test strips:



=> good gold conjugate release already at through addition of Tween 20. BSA and succrose nor mandatory for efficient release and low background.



Dry strips



=> Low test line intensity for low BSA concentration

Hypothesis: Analyte is faster than conjugate particles and blocks antibody before conjugate arrives at test line.



Influence of BSA on flow front separation





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