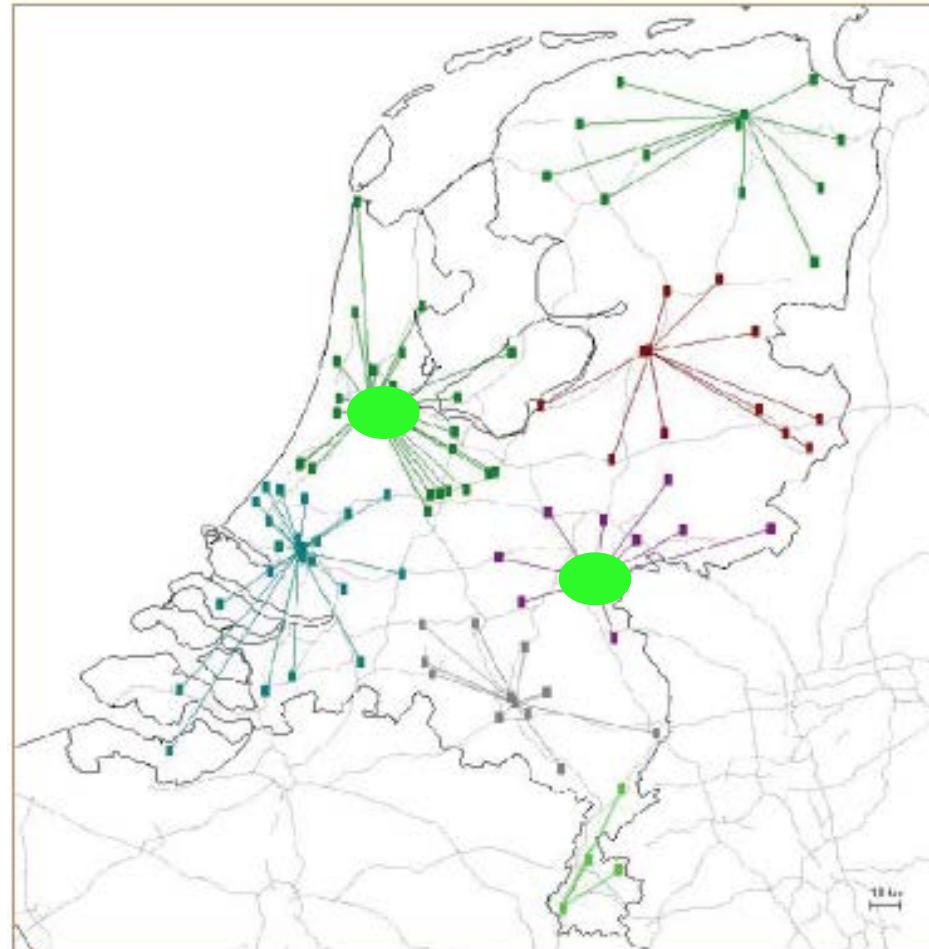




# PAS or plasma?

**Pieter F. van der Meer**  
Sanquin Blood Bank, Unit Production  
Sanquin Research, Clinical Transfusion Research

# Sanquin in 2015



# Collections and issues 2014

441,000

whole blood donations

264,000

apheresis donations (mostly plasma)

433,000

red cell concentrates issued

56,000

platelet concentrates

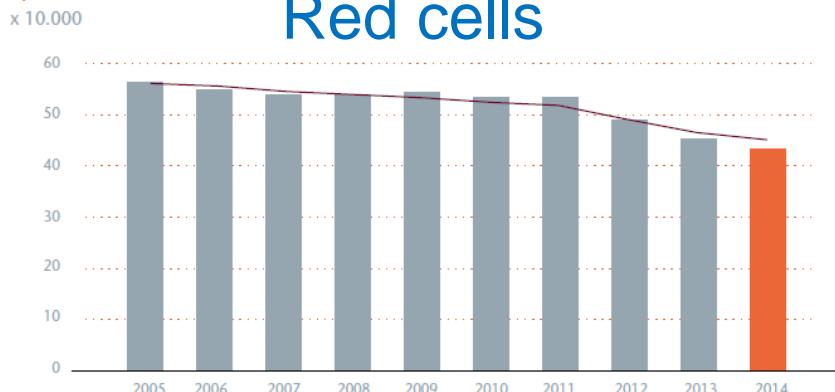
57,000

units FFP

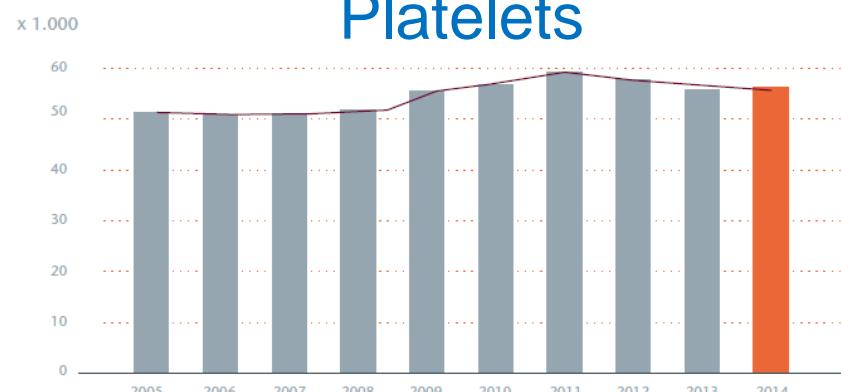
297,000

kilo plasma for fractionation

**Red cells**



**Platelets**



# Platelet production 2014

## Platelet concentrates

- 56,202 buffy coat pools
- About 20% in PAS, 80% in plasma
- 3,230 apheresis units (5.4%)
  - HLA matched for refractory patients
  - HPA 1a-neg for pediatric use

# Platelet additive solution

A balanced electrolyte solution that sustains platelet storage

Originally developed to

- remove plasma as source of proteolytic and glycolytic enzymes; prevent platelet storage lesion
- supplement buffering capacity of plasma; maintain pH>6.0



# Platelet additive solution

## Additional benefits

- More plasma for transfusion/fractionation
- Standardized composition
- Sterile, pathogen-free
- Ability to control storage environment
- Less protein – fewer allergic reactions
- Lower ABO titer
- (Reduction of antibody-mediated TRALI)

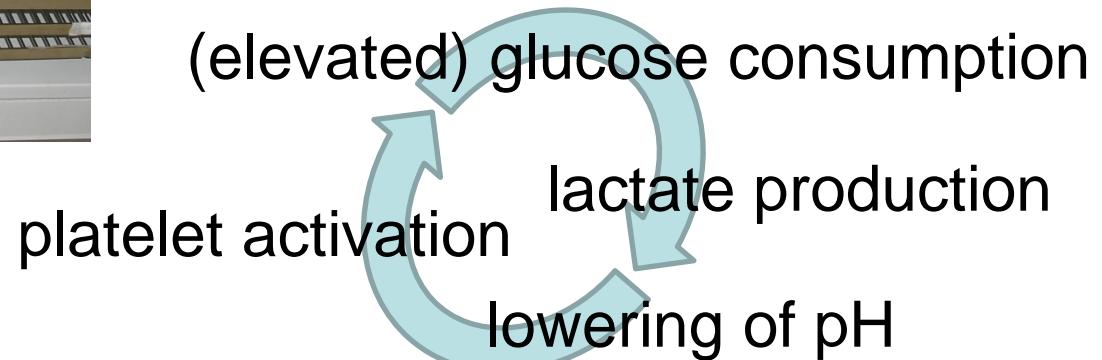
# Nomenclature

International Council for Commonality in Blood Banking Automation

PAS-A	PAS-B	PAS-C	PAS-D	PAS-E	PAS-F	PAS-G
Trade name						
“PAS”	PAS-2	PAS-3	Composol	PAS-IIIM	Plasma-	M-Sol
	PAS-II	PAS-III		SSP+	Lyte A	
	T-Sol	InterSol			Isoplate	
	SSP					
Citrate	X	X	X	X	X	X
Phosphate	X		X		X	X
Acetate		X	X	X	X	X
Magnesium				X	X	X
Potassium	X			X	X	X
Gluconate				X	X	
Glucose						X

# Platelet storage

## Platelet storage lesion (PSL)



# Platelet storage lesion

- All things “bad” happening to a platelet
  - increased activation and metabolism
  - increased signals for removal
  - poorer response to stimuli
  - reduced adhesion
  - ...
- lower recovery, shorter survival
- less able to stop or prevent bleeding

# Platelet additive solutions

Composition of PASs used to ‘tweak’ platelet quality during storage



Citrate  
Acetate  
Potassium  
Magnesium  
Phosphate  
Bicarbonate  
Calcium  
Glucose  
....

# Platelet metabolism

Platelets are extremely metabolically active cells:

- Oxygen consumption rate =  $3 \text{ }\mu\text{mol}/10^{10} \text{ cells/h}$
- Six times as fast as resting muscle; 30% as fast as mammalian brain

Glucose consumption

- into lactate  $3.13 \pm 0.44 \text{ }\mu\text{mol}/10^{11} \text{ platelets/h}$   
(net yield 2 ATP)
- full oxidation  $0.05 \pm 0.01 \text{ }\mu\text{mol}/10^{11} \text{ platelets/h}$   
(net yield ~30 ATP)

# Acetate

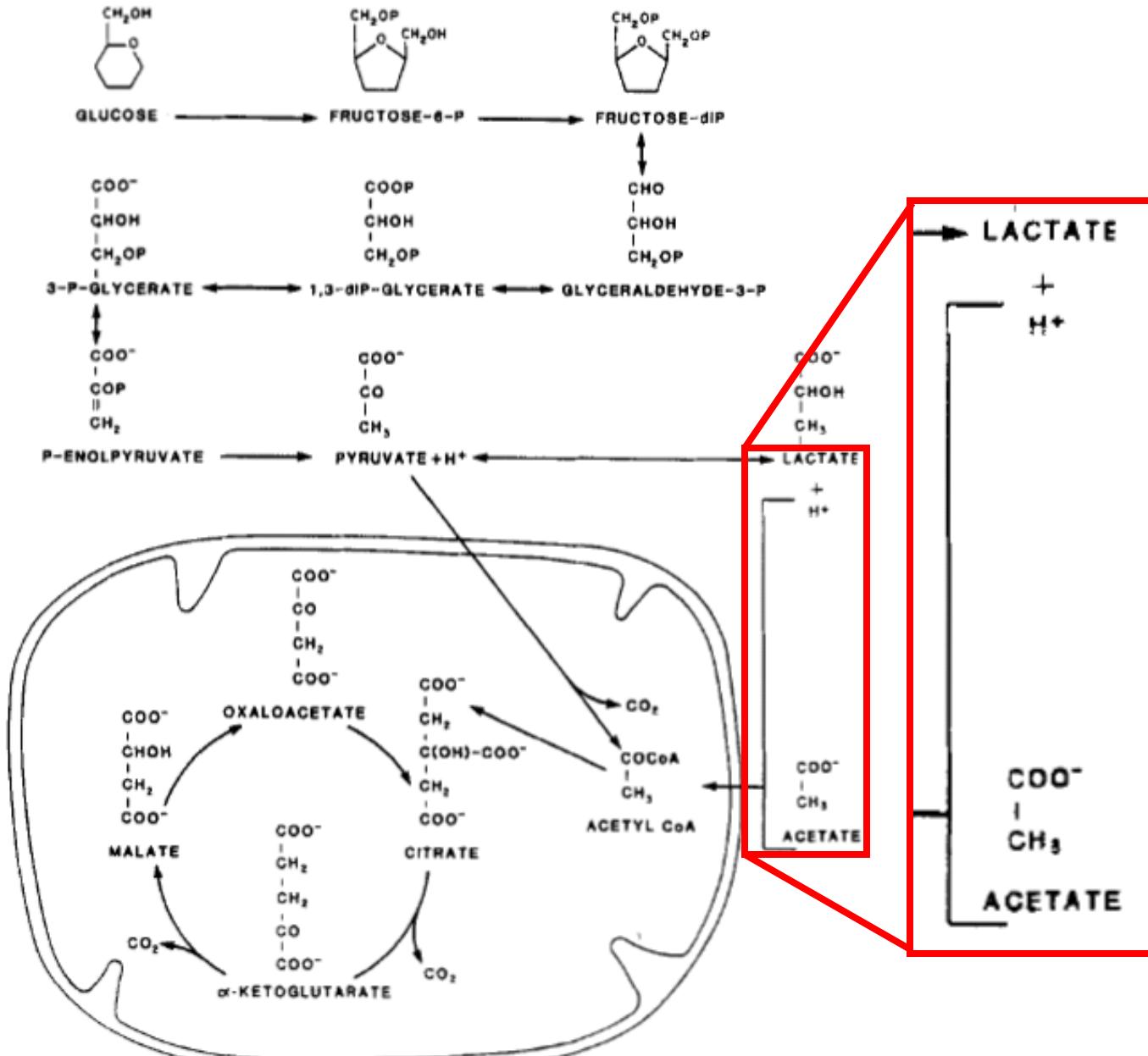
PASs need to provide a fuel that can readily be used by platelets

Serendipity:

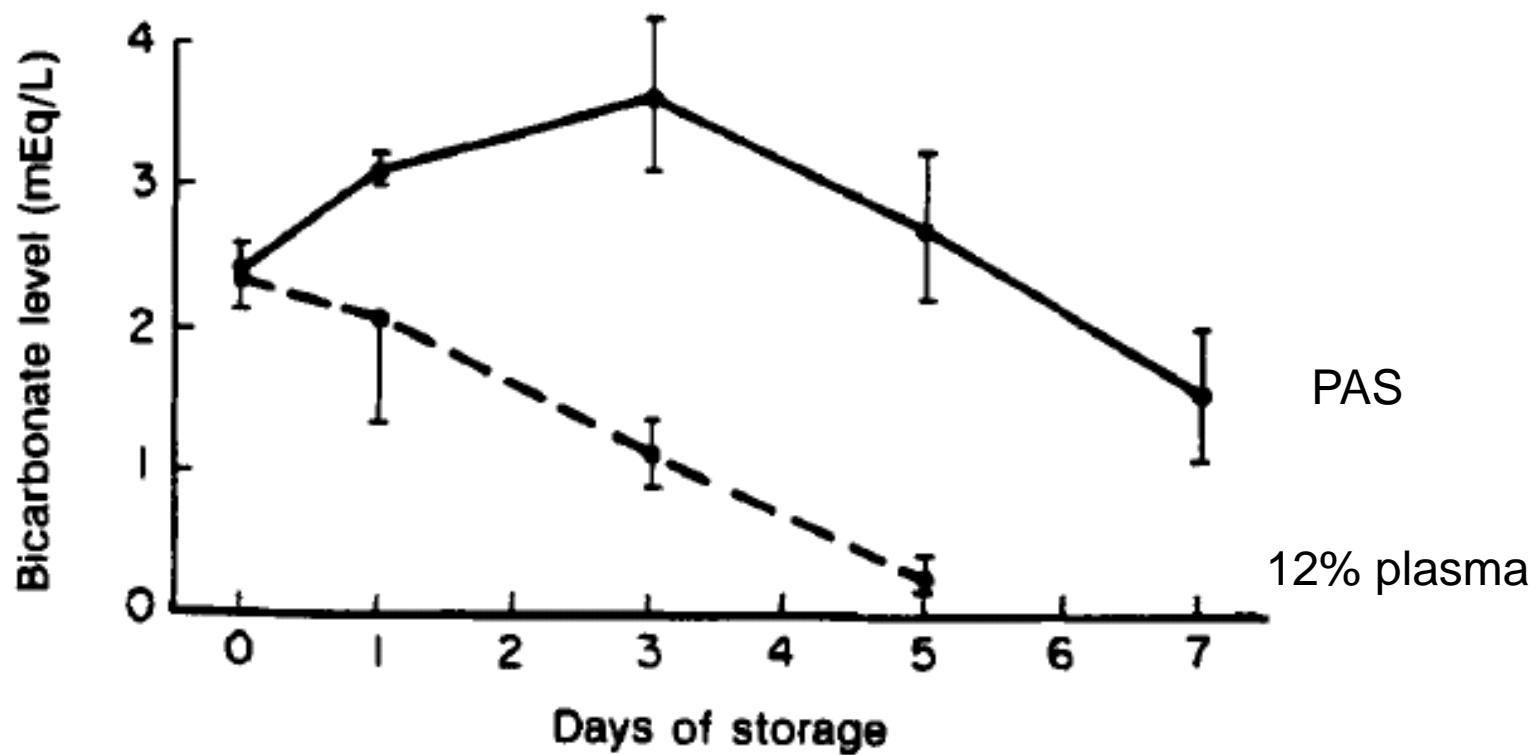
Acetate was present in infusion fluids that happened to be used in the early PAS studies

Lactate production

- No acetate:  $2.4 \pm 0.5 \text{ }\mu\text{mol/day}/10^{11}$  platelets
- 23 mM acetate:  $1.3 \pm 0.3 \text{ }\mu\text{mol/day}/10^{11}$  platelets



# Acetate



Acetate provides it's own buffer

# Effect of K/Mg

<i>Day 7</i>	pH	CD62P
PAS-2	6.98±0.07	49±10
PAS-2 + Mg	7.10±0.07*	41±14

1.5 mM Mg, 4.5 mM K; \*p<0.05; \*\*p<0.01

# Effect of K/Mg

<i>Day 7</i>	pH	CD62P
PAS-2	6.98±0.07	49±10
PAS-2 + Mg	7.10±0.07*	41±14
<hr/>		
PAS-2	6.93±0.04	55±6
PAS-2 + K	7.19±0.03**	35±8*

1.5 mM Mg, 4.5 mM K; \*p<0.05; \*\*p<0.01

# Effect of K/Mg

<i>Day 7</i>	pH	CD62P
PAS-2	6.98±0.07	49±10
PAS-2 + Mg	7.10±0.07*	41±14
PAS-2	6.93±0.04	55±6
PAS-2 + K	7.19±0.03**	35±8*
Plasma	7.03±0.06	35±8
PAS-2	6.94±0.05*	50±8*
PAS-2 + Mg + K	7.15±0.10*	23±6*

1.5 mM Mg, 4.5 mM K; \*p<0.05; \*\*p<0.01

# Effect of K/Mg

## Potassium

- maintaining membrane potential

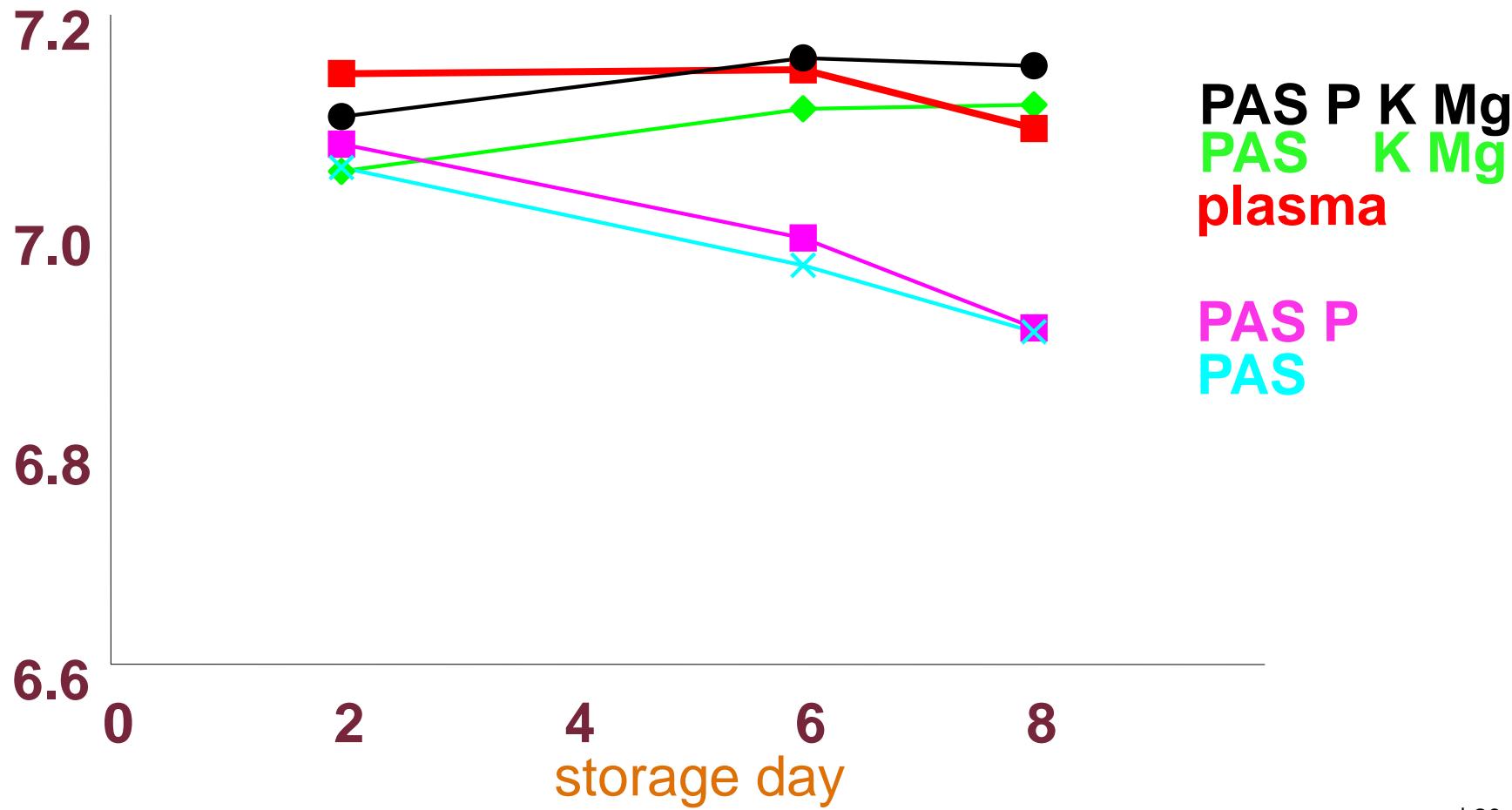
## Magnesium

- activates potassium pumps
- decreases the PLT activation
- influences influx of calcium, thereby intracellular potassium concentration
- inhibits agonist-induced PLT aggregation, by changing membrane fluidity and/or by triggering of cAMP

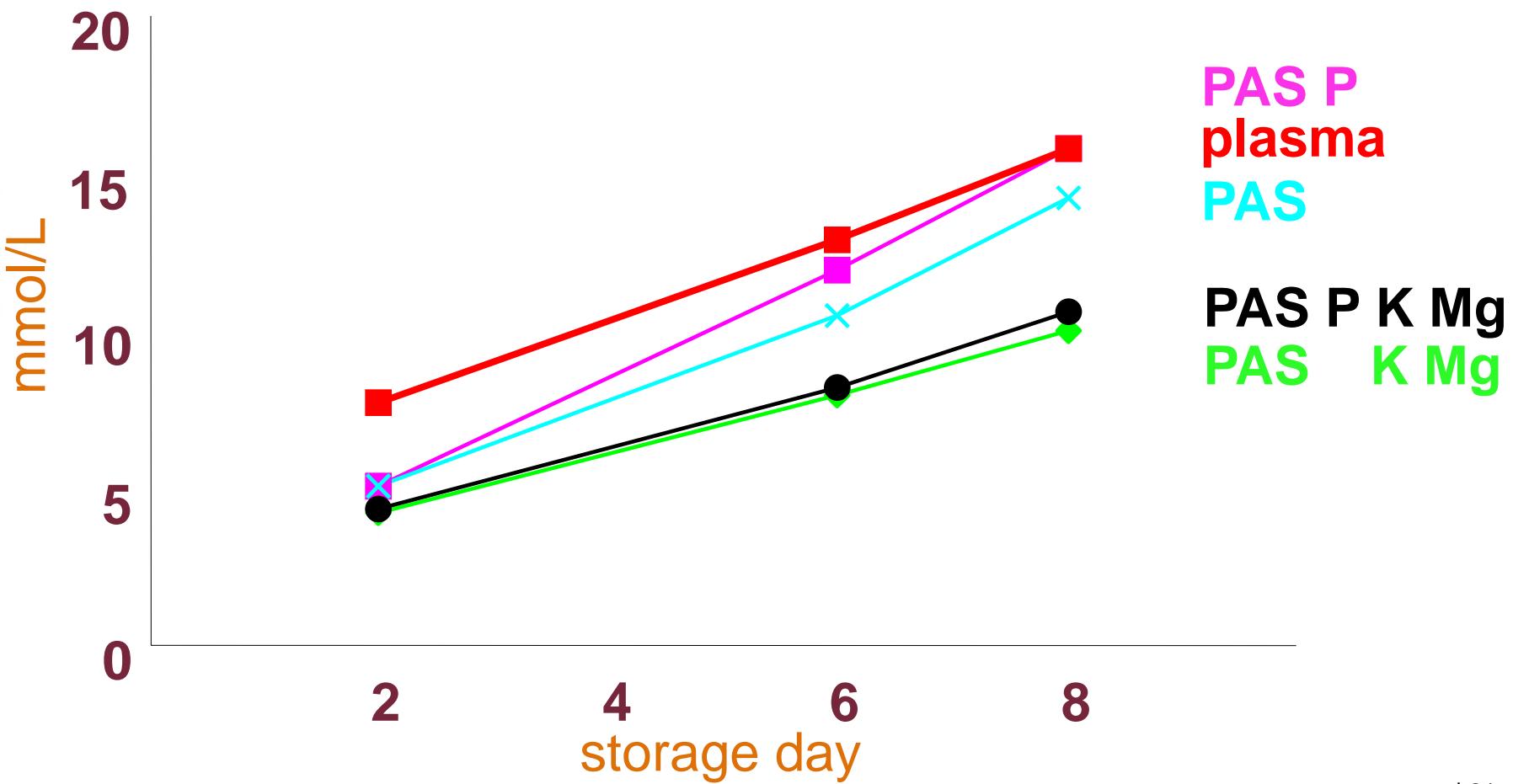
# Comparison of current PASs

- Pool and split buffy coats
- Add plasma or either of 4 PASs
- Centrifuge
- Storage in the same storage containers for 8 days
- *In vitro* analysis

# pH<sub>37°C</sub>



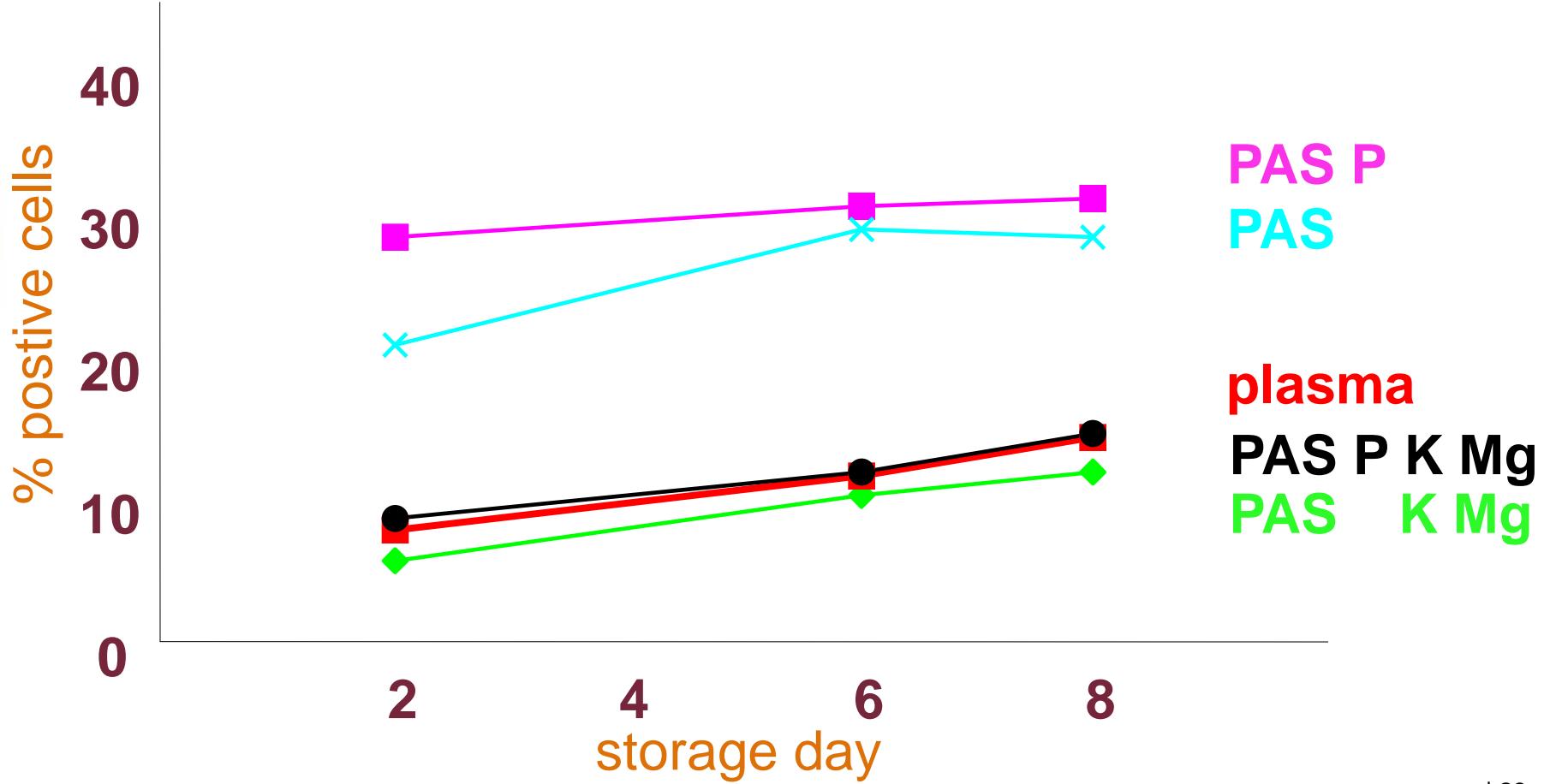
# Lactate



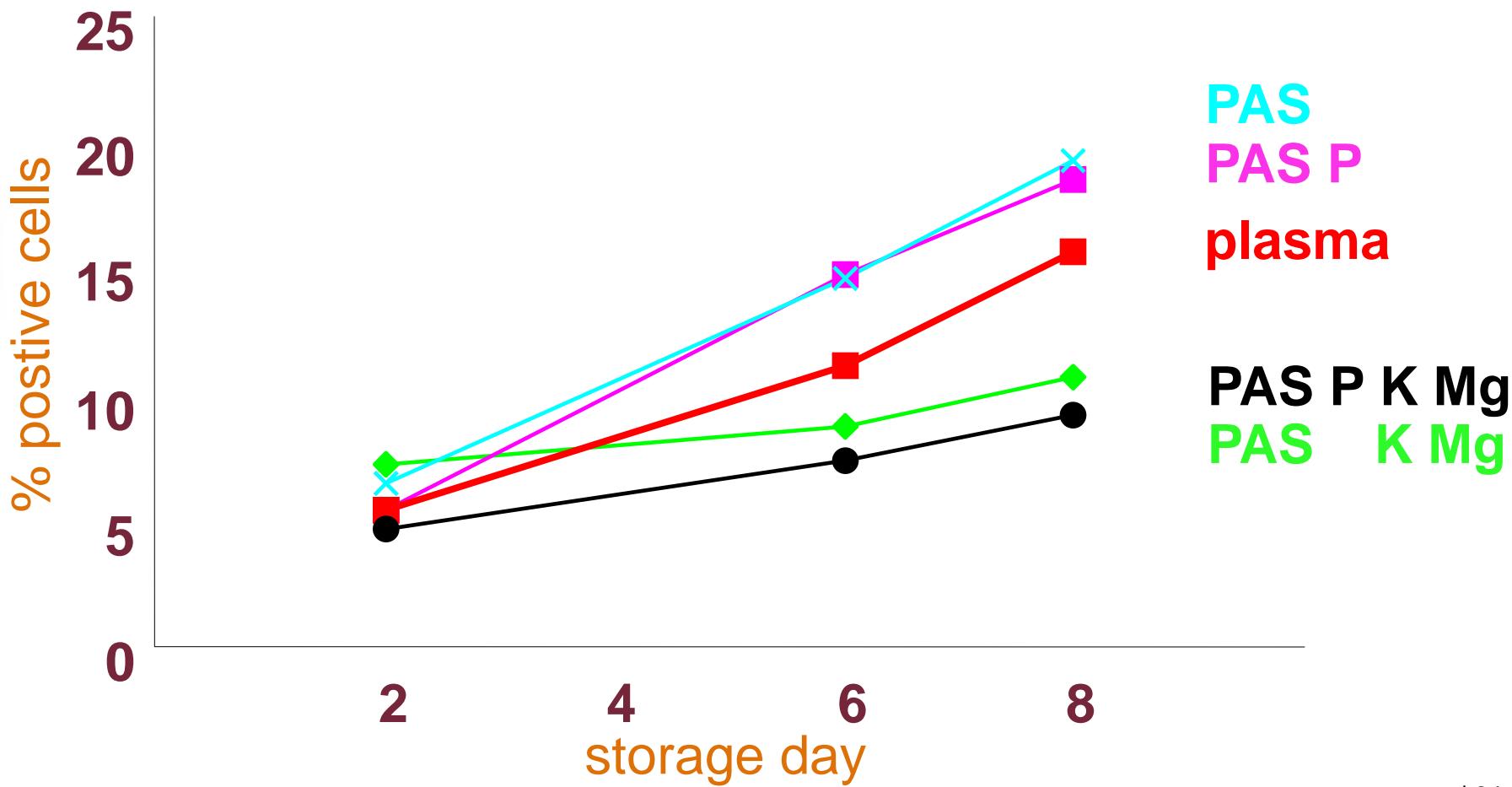
# Lactate production rate

plasma	$0.13 \pm 0.04$	
PAS	$0.14 \pm 0.02$	PAS-2
PAS P	$0.17 \pm 0.03$	PAS-3
PAS P K Mg	$0.11 \pm 0.03$	SSP+
PAS K Mg	$0.10 \pm 0.02$	Composol

# CD62P expression

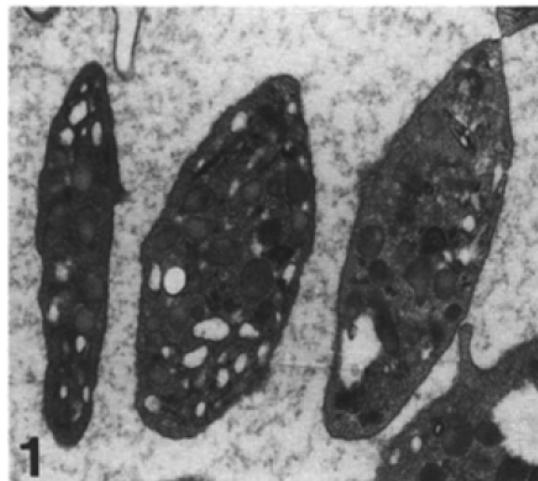


# Annexin A5 binding



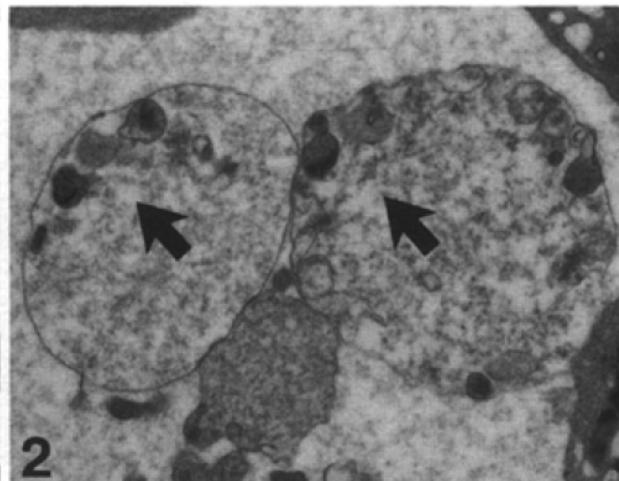
# Percent residual plasma

At least  $\pm 30\%$  needed to preserve structural integrity



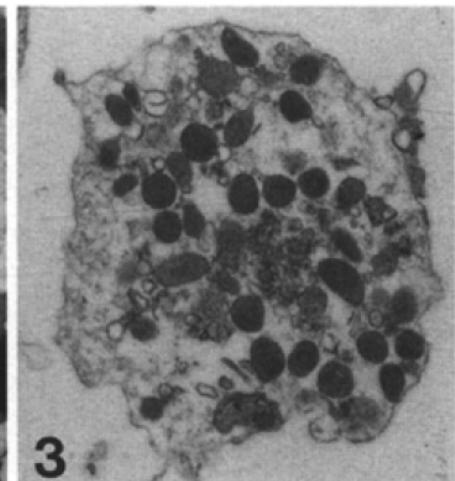
1

Day 1



2

Day 8 in plasma



3

Day 8 in 80% PAS-2

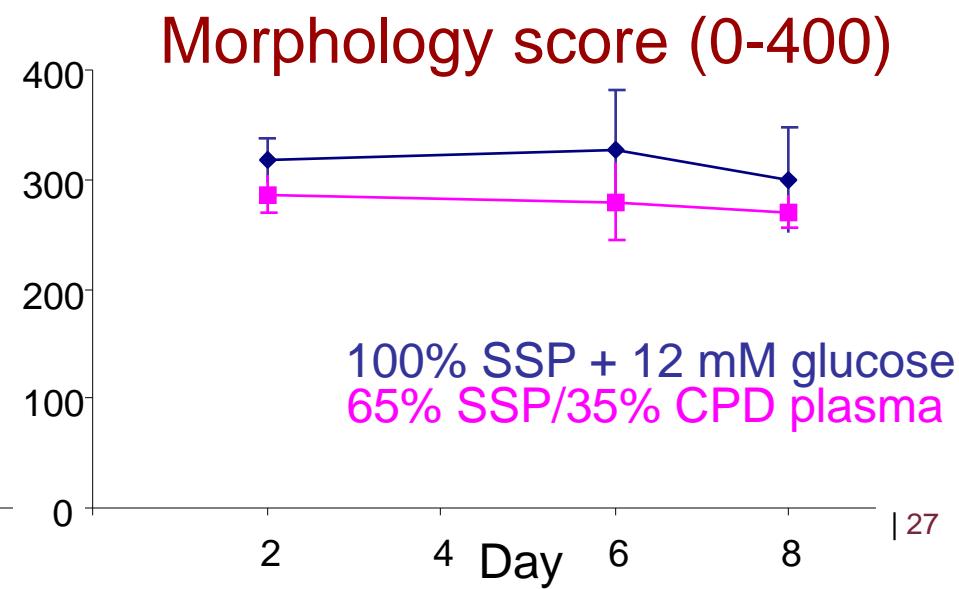
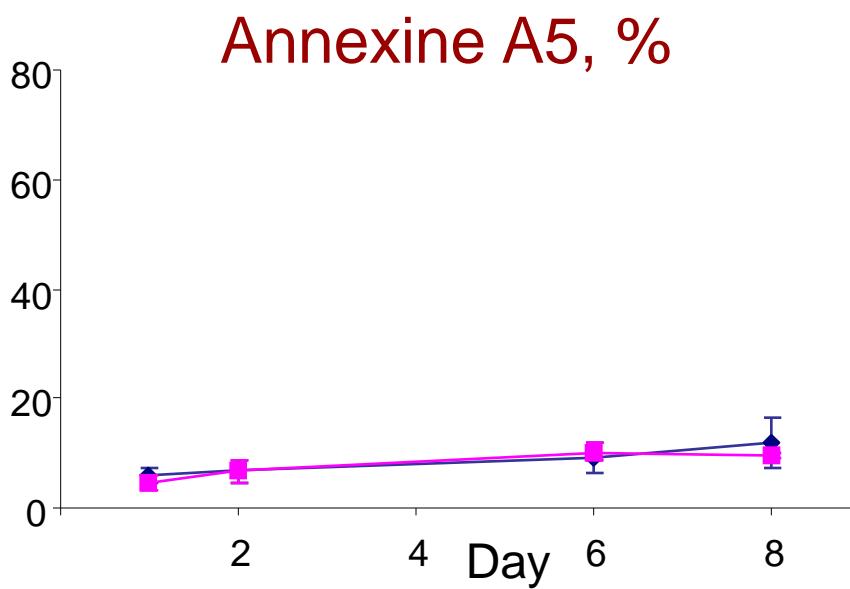
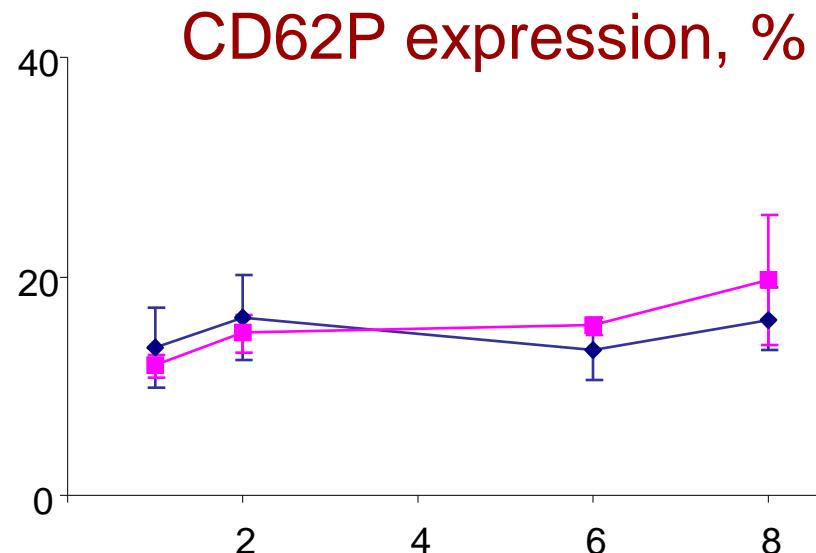
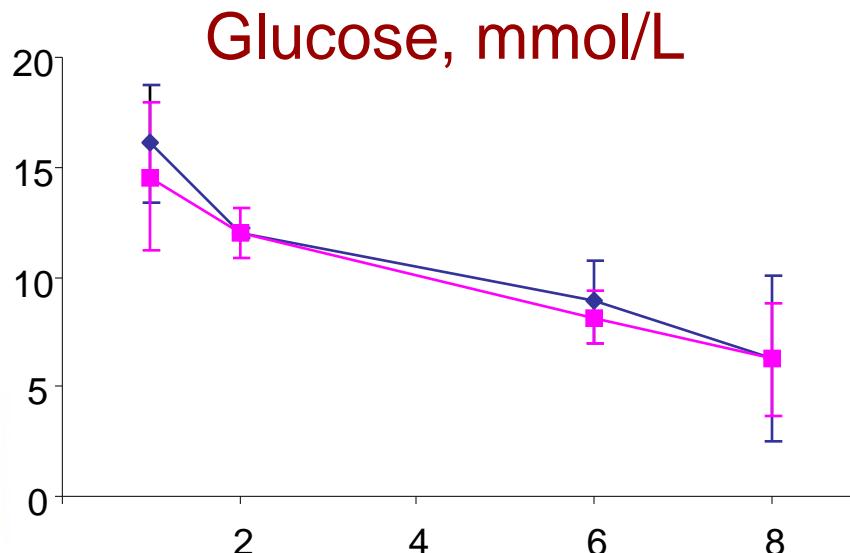
20% of platelets

nearly all

# Percent residual plasma

- Pool and split 2 platelet concentrates
- Add 10% ACD, centrifuge, remove all supernatant
- Unit A: SSP+ and 12 mM glucose     $16.1 \pm 2.7$  mM
- Unit B: 35% plasma/65% SSP+        $14.6 \pm 3.3$  mM
- Store for 8 days
- Various in vitro measures
- n=3

# Percent residual plasma



# Percent residual plasma

- No difference between 100% PAS with added glucose versus 65% PAS/35% CPD plasma
- Therefore, the plasma carry over was necessitated by the glucose requirement; protein is not compulsory

# Allergic reactions

	Tx, n	Total, %	FNTR			Solution
				Allergic	Others	
Oksanen, 1994	23	78%	22%	52%	4%	plasma PAS-A
	86	23%	5%	19%	0	
Bertolini, 1989	448	1.4%	0.5%	0.89%	0	plasma PAS-F
	129	0%	0	0	0	
De Wildt-Eggen, 2000	192	12%	4.2%	5.2%	3.0%	plasma PAS-B
	132	5.3%	4.5%	0	0.8%	
Kerkhoffs, 2006	311	5.5%	n.r.	n.r.	n.r.	plasma PAS-B
	373	2.3%	n.r.	n.r.	n.r.	
Andreu, 2007	1275	n.r.	0.08%	0.16%	n.r.	plasma PAS-B
	8206	n.r.	0.15%	0.02%	n.r.	
	25698	n.r.	0.16%	0.60%	n.r.	plasma PAS-B
	3525	n.r.	0.14%	0.31%	n.r.	

	Plasma	PAS-C	Difference
Kerkhoffs, 2010			
CCI-1 h	17.1±7.3	15.3±6.5	-9%
CCI-24 h	12.8±7.8	11.6±7.6	-7%
Tobian, 2014			
CCI-1h	4.9	3.7	-24%
CCI-24h	2.1	1.7	-19%

	Plasma	PAS-C	PAS-E
CCI-1-24h	10.2	8.6	10
Difference		-16%	-2%

# Recovery and survival

	n	Source	Solution	Storage, d	Recovery, %	Survival, d
Slichter, 2014b	6	Apheresis	plasma	5	<b>59±7</b>	<b>6.5±0.6</b>
	10	Apheresis	plasma	7	<b>44±5</b>	<b>4.9±0.7</b>
	6	Apheresis	PAS-F	5	<b>59±5</b>	<b>6.3±0.8</b>
	10	Apheresis	PAS-F	7	<b>52±3</b>	<b>6.0±0.3</b>
	4	Apheresis	PAS-F	9	<b>55±5</b>	<b>6.6±0.6</b>
	10	Apheresis	PAS-F	13	<b>49±3</b>	<b>4.6±0.3</b>
	10	Apheresis	PAS-F	14	<b>43±3</b>	<b>4.2±0.5</b>

# Conclusions

Over the past decades, numerous PASs have been developed

Some were good, some not so good

With the ‘newer’ PASs, *in vitro* quality of platelets is not worse than when stored in plasma, probably even better

Acetate partially replaces glucose consumption, limiting lactate formation and thereby the platelet storage lesion

# Conclusions

Various modifications have been done to further optimize platelet quality, notably the addition of potassium and magnesium

Lower protein content gives fewer allergic transfusion reactions; further optimization possible?

Recovery, survival as well as CCI are good if not better for platelets in PAS than in plasma

# PAS or plasma?

With the current generation of PAS, because

- storage quality (judged *in vitro* and *in vivo*) is at least as good

- allergic reactions are fewer

the use of PAS for the storage of platelet concentrates should be preferred.