
American Red Cross

Penn-Jersey ARC

	Distribution	Transfusion Episode
Red Cells	368,000	184,000
Random Donor Plts.	160,000	32,000
Apheresis Plts	21,000	21,000
FFP	120,000	60,000
		297,000

Death Following Blood Transfusion

1. Human error
2. Bacterial contamination of platelets

   Severe morbidity and mortality-1/20,000
3. Transfusion related acute lung injury (TRALI)

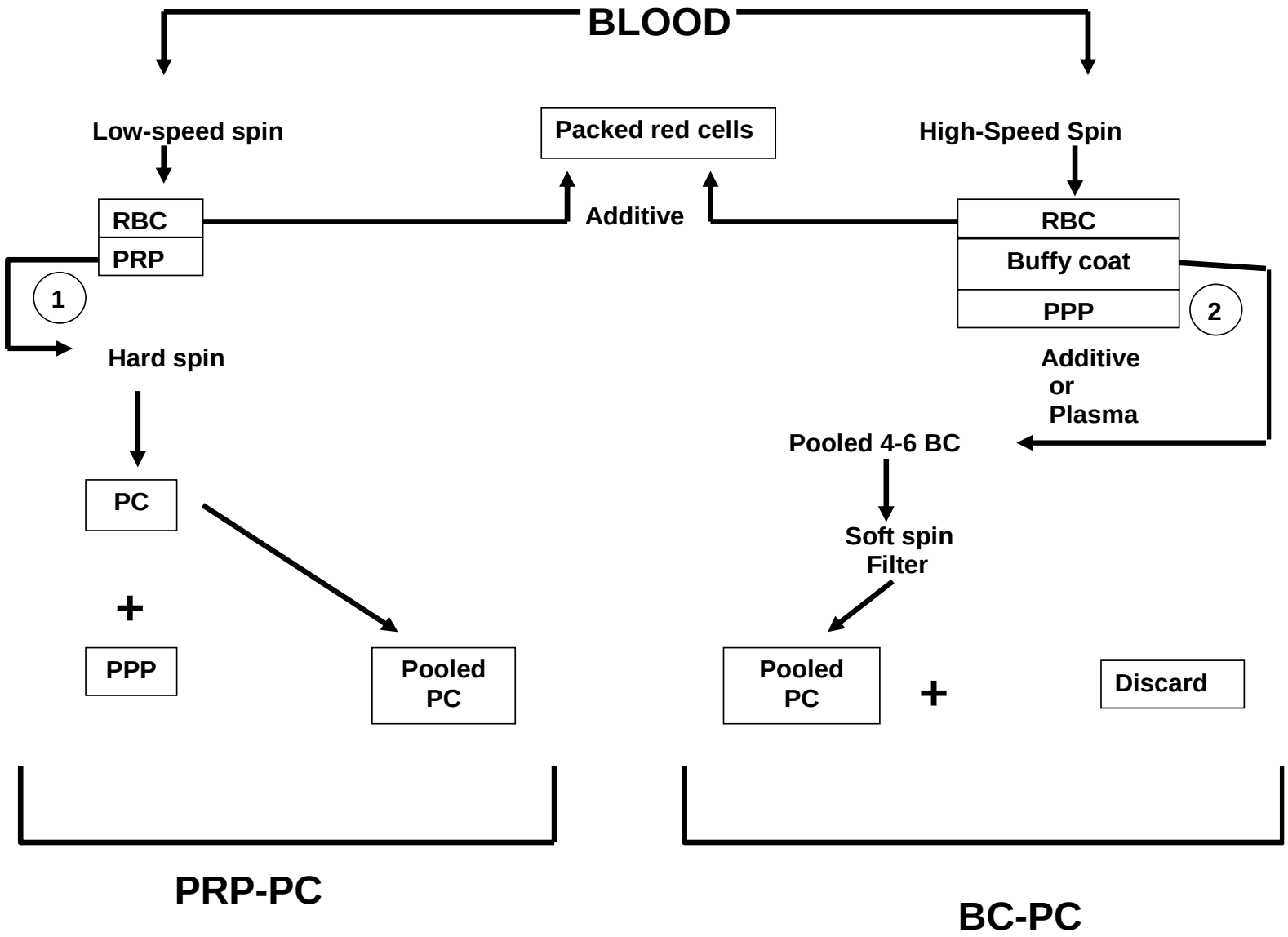
Episodes

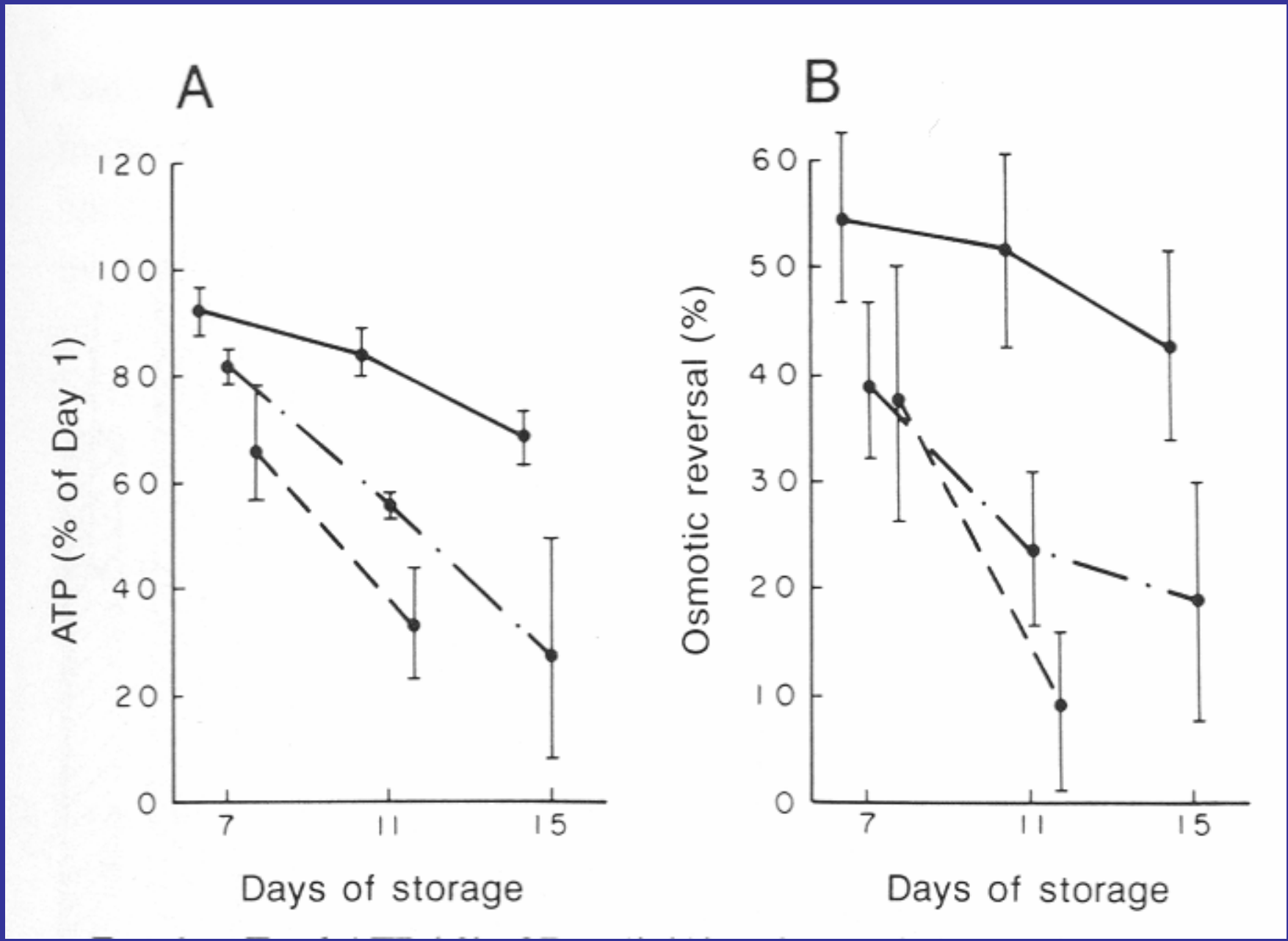
TRALI 2 (? 3)

Sepsis 3

None Fatal

	% Apheresis
Western Europe	42
Denmark, Finland	
Holland, Portugal	<8
Spain	18
France	77


BLOOD
Low-speed spin
Packed red cells
High-Speed Spin
RBC
PRP
Additive
RBC
Buffy coat
PPP
1
2
Hard spin
Additive
or
Plasma
Pooled 4-6 BC
PC
Soft spin
Filter
+
PPP
Pooled
PC
Pooled
PC
+
Discard
PRP-PC
BC-PC


A
120
100
80
60
40
20
0
ATP (% of Day 1)
7
11
15
Days of storage
B
60
50
40
30
20
10
0
Osmotic reversal (%)
7
11
15
Days of storage

UNITED STATES

13,000,000 Blood Donations/year

2,000,000 Platelet Transfusions/year

At 5 PC per Tx, there are enough platelets in the blood donations to meet patient needs.

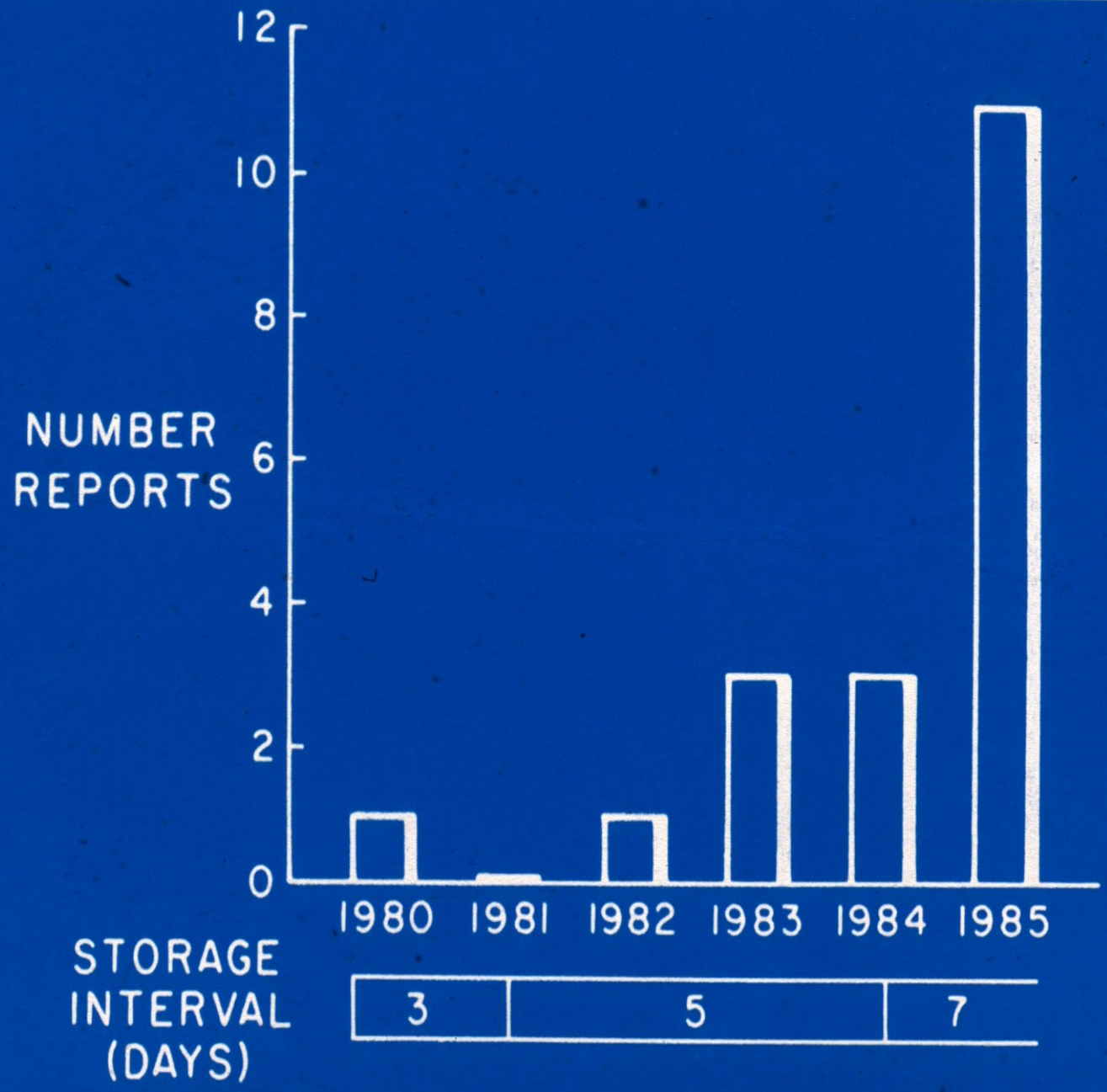
PC are obtained with no extra risk for donor

Improving Platelet Availability

1. Improve donor recruitment and retention
2. More platelets from whole blood - don't throw away gift already given
3. Extend current platelet storage interval – Study 22°C storage interval
4. Other methods

   Hoffmeister's et al

SEPSIS AFTER PLATELET TRANSFUSION



Hoffmeister et al. Science 301:1531, 2003

Storage at 4°C – mouse platelets

New concept of storage lesion –
GP1bœ altered, recognized by liver

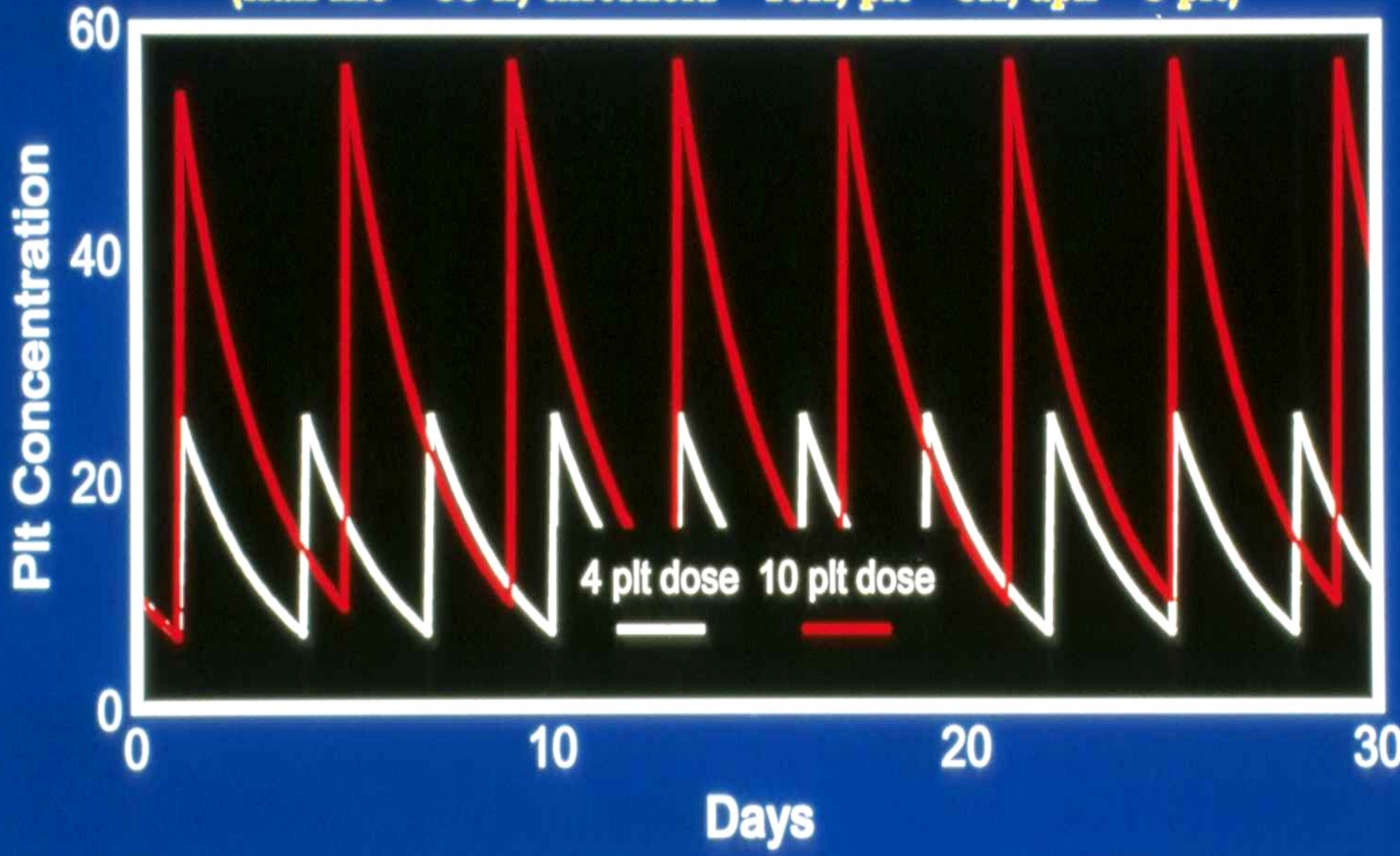
Cover GP1bx with uridine diphosphate galactose – Platelet survival restored

Improving Platelet Availability (Cont.)

5. Adhere to trigger
6. Research on dose

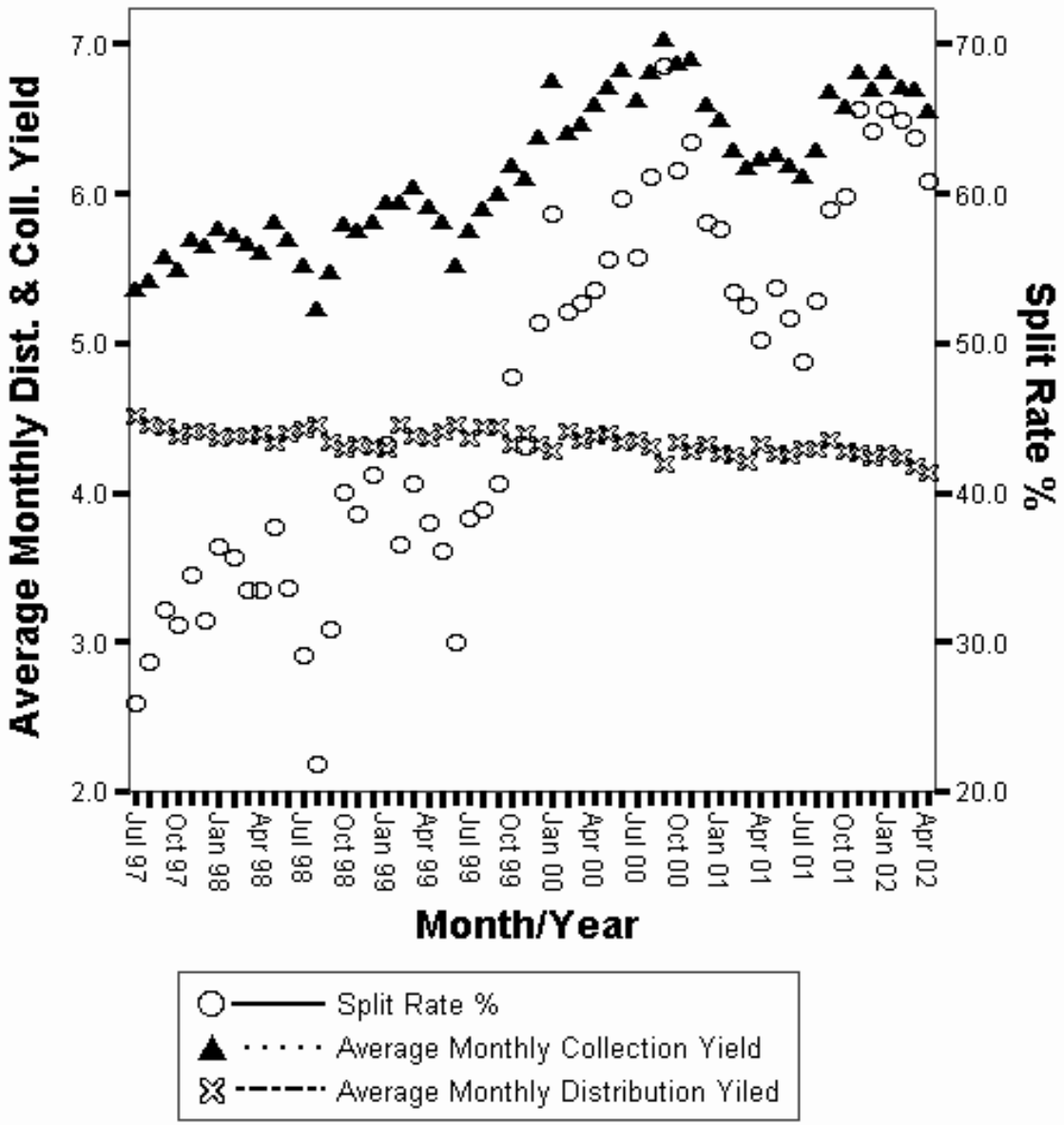
Platelet Transfusion Modeling

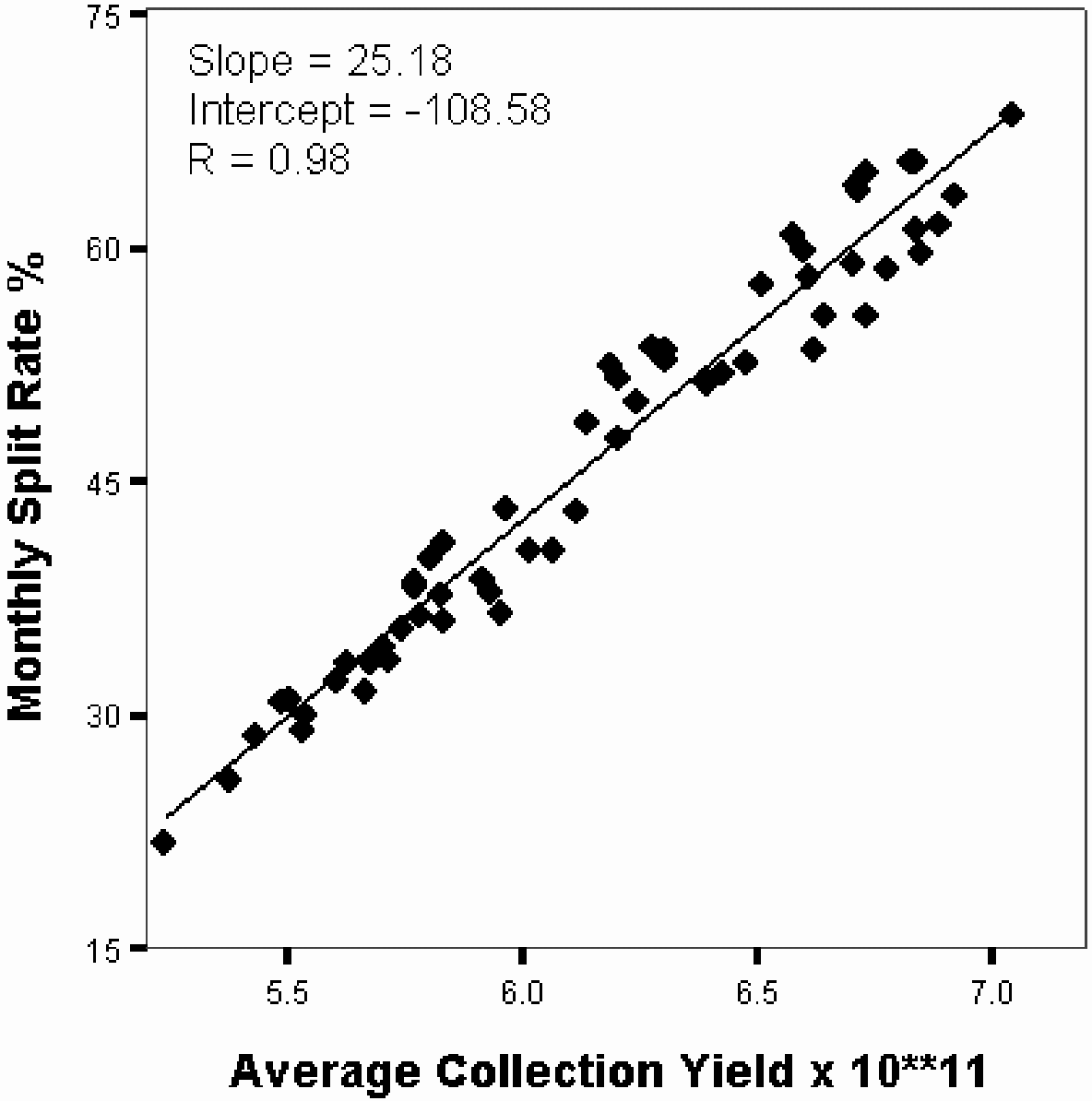
(Half life = 36 h, threshold = 10K, plt = 5K, aph = 8 plt)

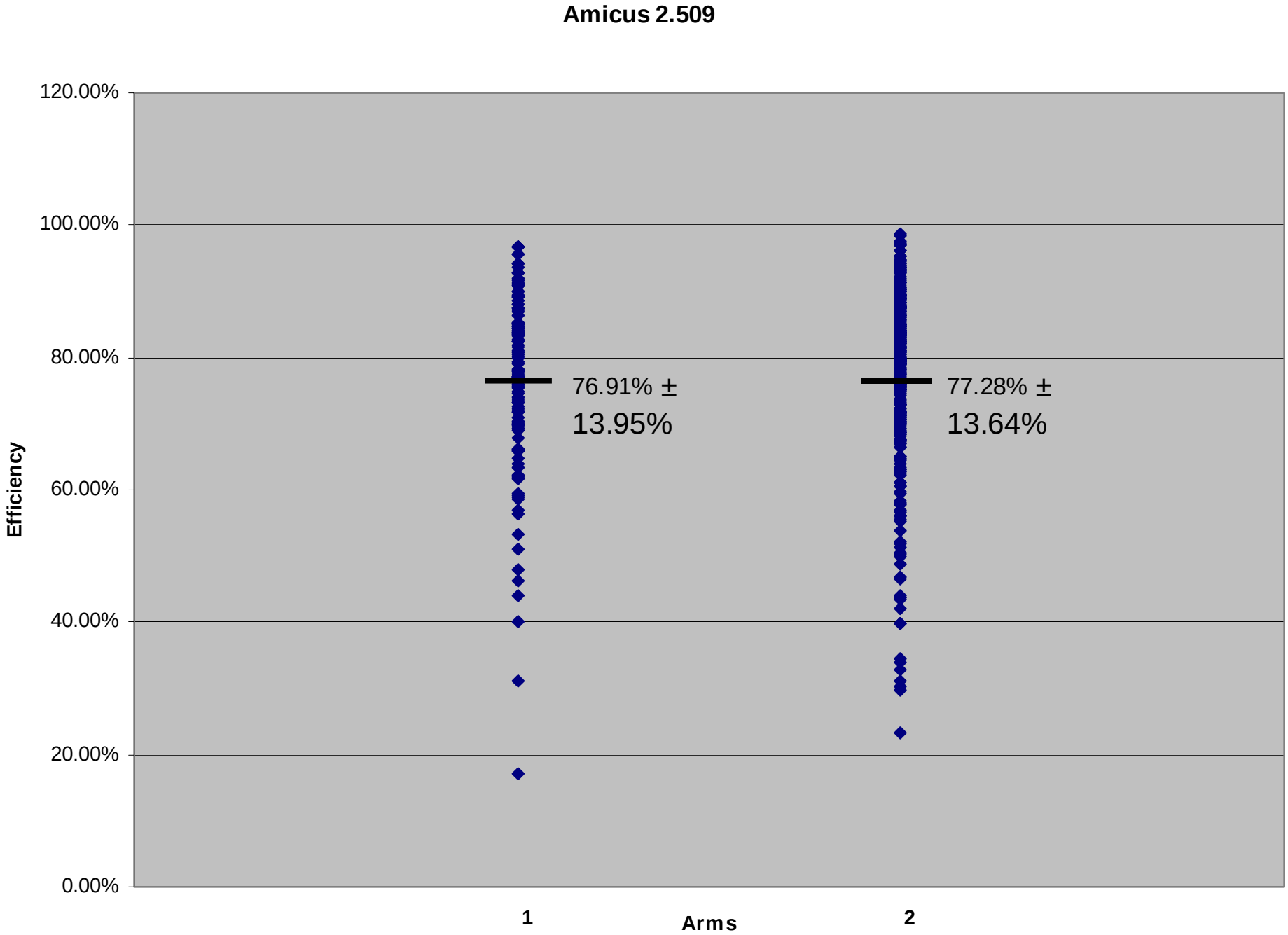


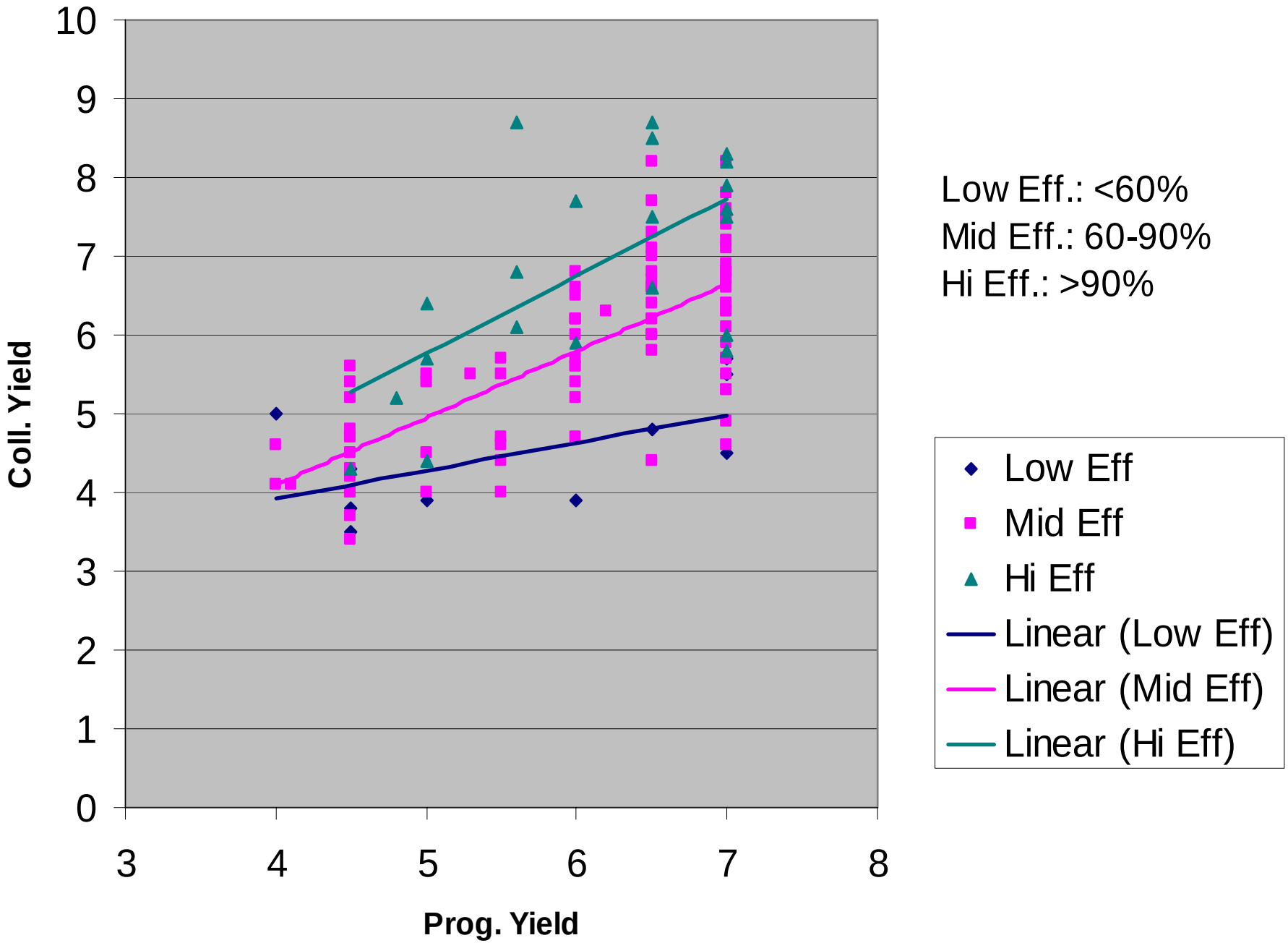
Improving Platelet Availability (cont.)

7. Decrease low yield apheresis collections


Average Monthly Dist. & Coll. Yield
Split Rate %
Month/Year
7.0
6.0
5.0
4.0
3.0
2.0
70.0
60.0
50.0
40.0
30.0
20.0
Jul 97
Oct 97
Jan 98
Apr 98
Jul 98
Oct 98
Jan 99
Apr 99
Jul 99
Oct 99
Jan 00
Apr 00
Jul 00
Oct 00
Jan 01
Apr 01
Jul 01
Oct 01
Jan 02
Apr 02
Split Rate %
Average Monthly Collection Yield
Average Monthly Distribution Yiled


Slope = 25.18
Intercept = -108.58
R = 0.98
Monthly Split Rate %
75
60
45
30
15
5.5
6.0
6.5
7.0
Average Collection Yield x 10**11


Amicus 2.509
120.00%
100.00%
80.00%
60.00%
40.00%
20.00%
0.00%
Efficiency
76.91% ±
13.95%
77.28% ±
13.64%
1
2
Arms


10
9
8
7
6
5
4
3
2
1
0
Coll. Yield
3
4
5
6
7
8
Prog. Yield
Low Eff.: <60%
Mid Eff.: 60-90%
Hi Eff.: >90%
Low Eff
Mid Eff
Hi Eff
Linear (Low Eff)
Linear (Mid Eff)
Linear (Hi Eff)

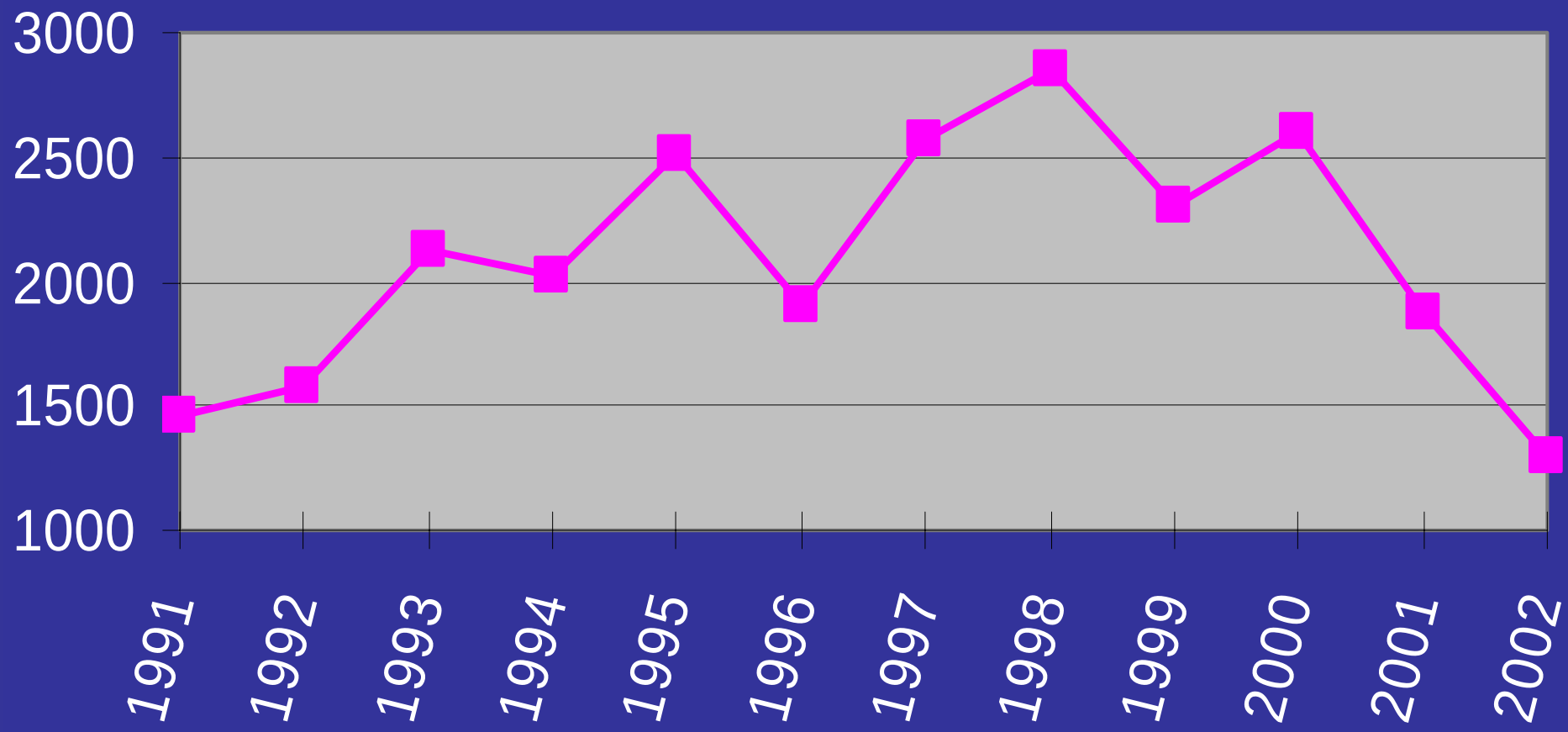
Improving Platelet Availability (cont.)

8. Don’t waste to alloimmunization

Need for Matched Platelets Will Decrease (?50%) But Not Disappear

1. Effect of pregnancy
2. TRAP study showed only 50% efficacy

Yearly Matched Platelet Distribution



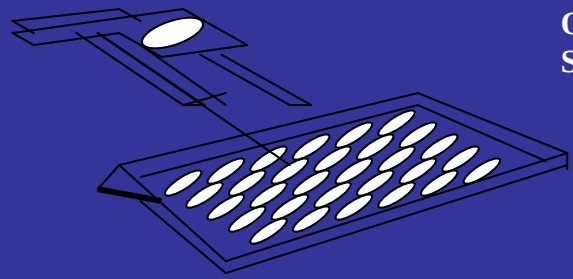
Platelet Matching
American Red Cross

	Number/Month	% Xmatch
Madison	141	<1
Boston	105	4
Baltimore	105	14
Charlotte	433	20
Miami	18	22
Philadelphia	250	32
Connecticut	55	73
Atlanta	212	94

Antibody Specificity Prediction (ASP) Method

1. Perform lymphocytotoxic (LCT) antibody screen
2. Identify HLA antigens to which patient has developed antibody
3. Treat patient with platelets which lack those antigens (i.e. antigen negative platelets)

$3x10^6$ CELLS/ML
LYMPHOCYTES



DISPENSE 1µL/WELL
ONTO ANTIBODY
SCREEN TRAY

ROOM TEMPERATURE

30 MIN. INCUBATION

WASH SENSITIZED CELLS 3X

ADD 1µL AHG/WELL

ROOM TEMPERATURE

2 MIN. INCUBATION

ADD 5µL COMPLEMENT/WELL

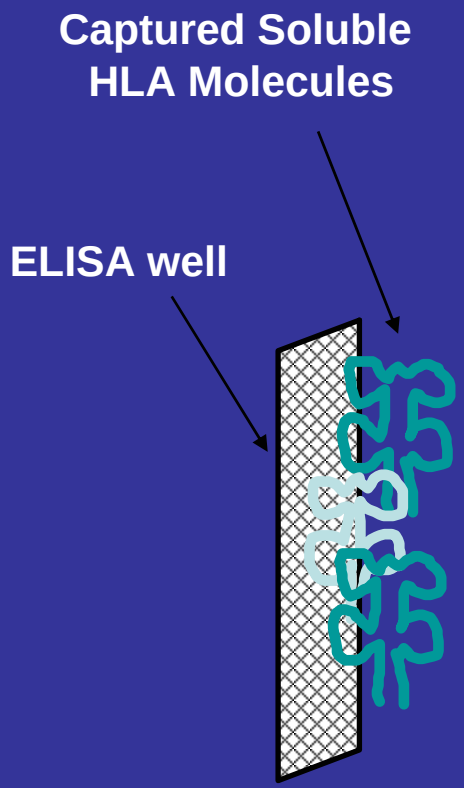
ROOM TEMPERATURE

1 HOUR INCUBATION

STAIN AND FIX WITH EOSIN AND FORMALIN

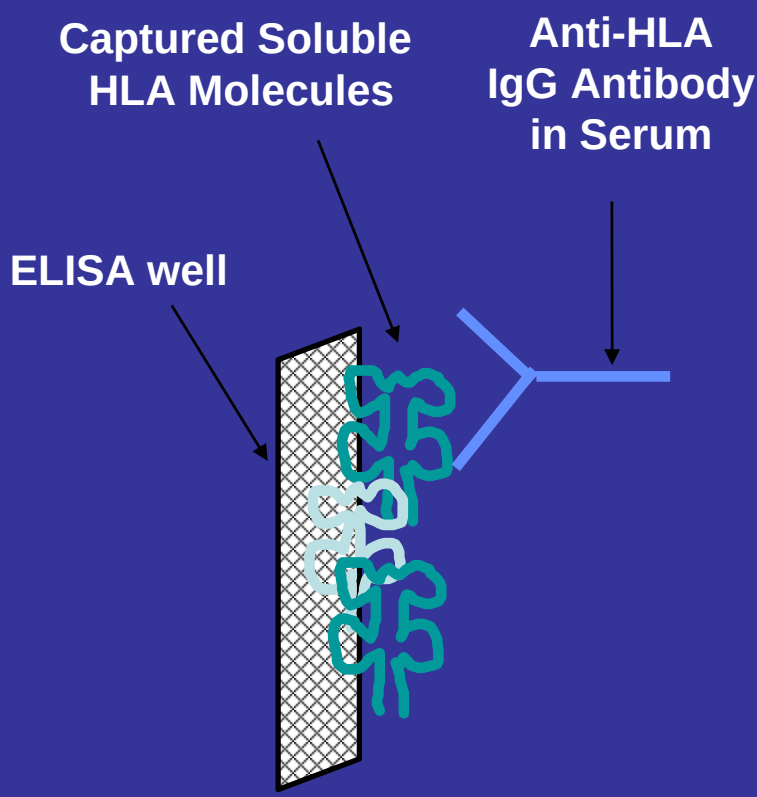
EVALUATE MICROSCOPICALLY FOR DYE EXCLUSION

ELISA based HLA antibody analysis



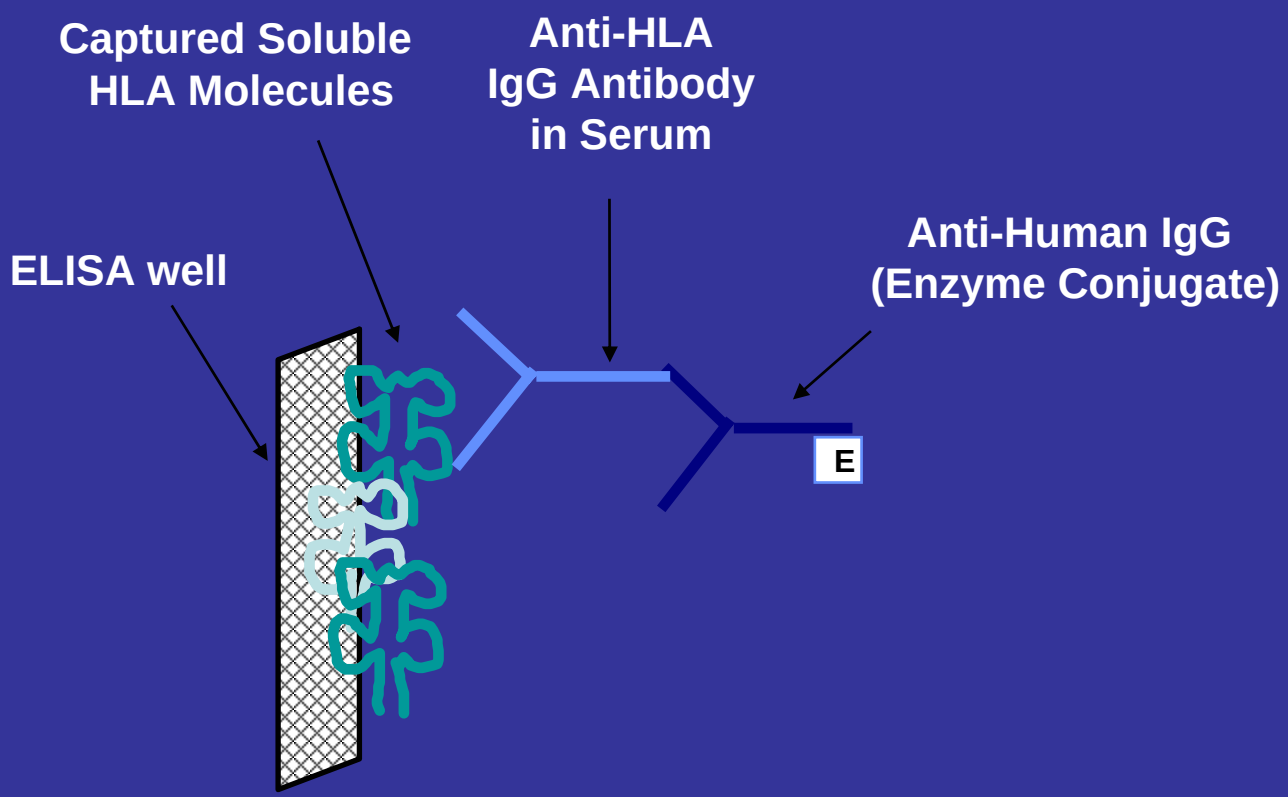
1. sHLA is bound to well of plastic ELISA tray

ELISA based HLA antibody analysis



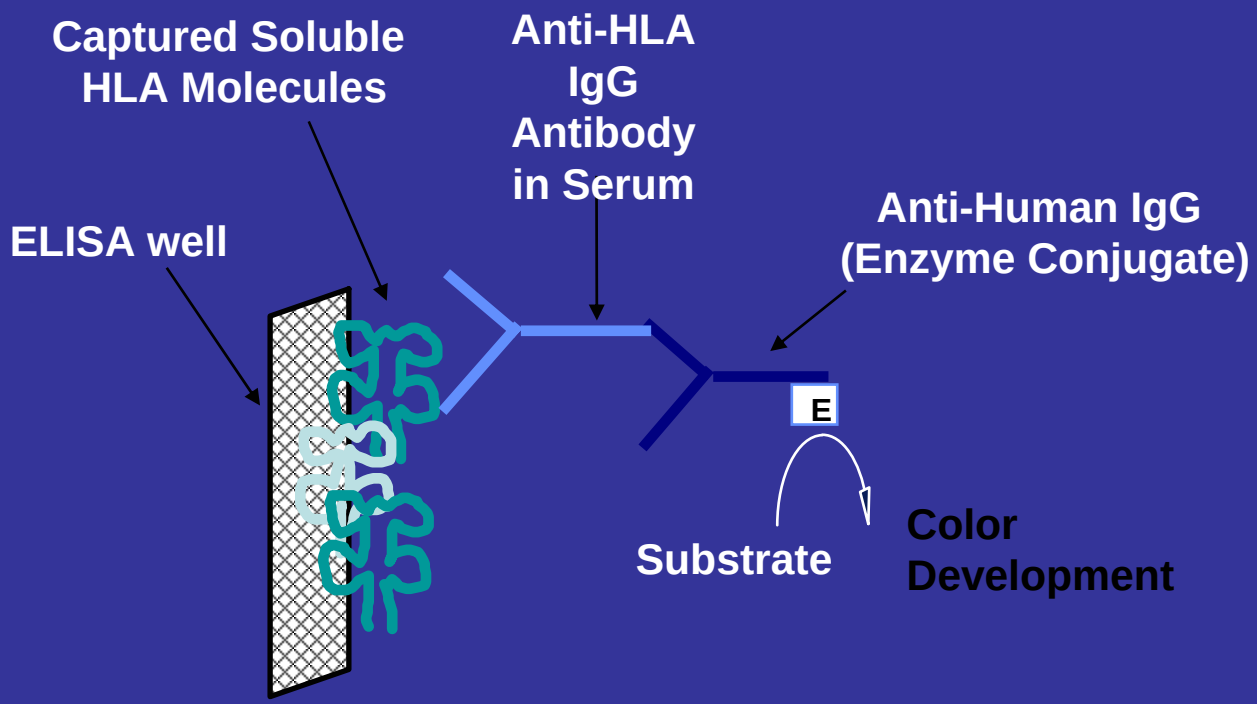
2. Add patient serum to well
   -incubate
   -HLA specific antibodies will bind
   -wash away unbound Ig

ELISA based HLA antibody analysis



3. Add “second step” antibody
   -enzyme conjugate
   -binds human Ig
   -incubate
   -wash away unbound antibody

ELISA based HLA antibody analysis

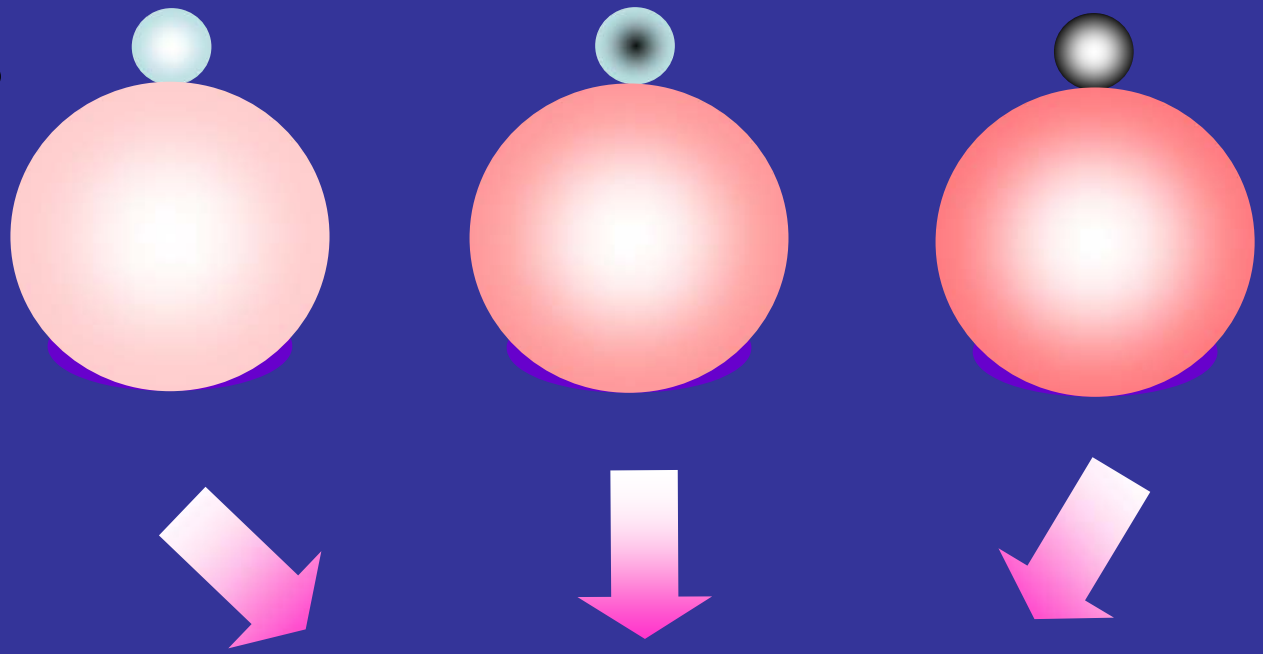


4. Add enzyme substrate
   -will turn color in positive wells
   -read on ELISA reader

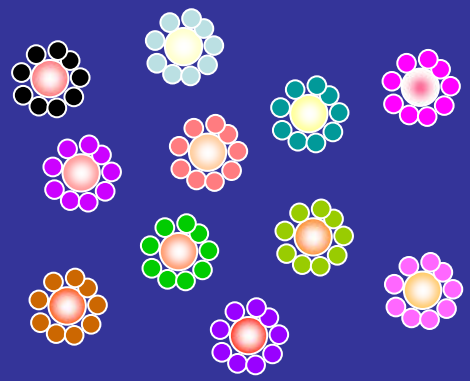
Concept of FlowPRA™ Specific Test

HLA antigens

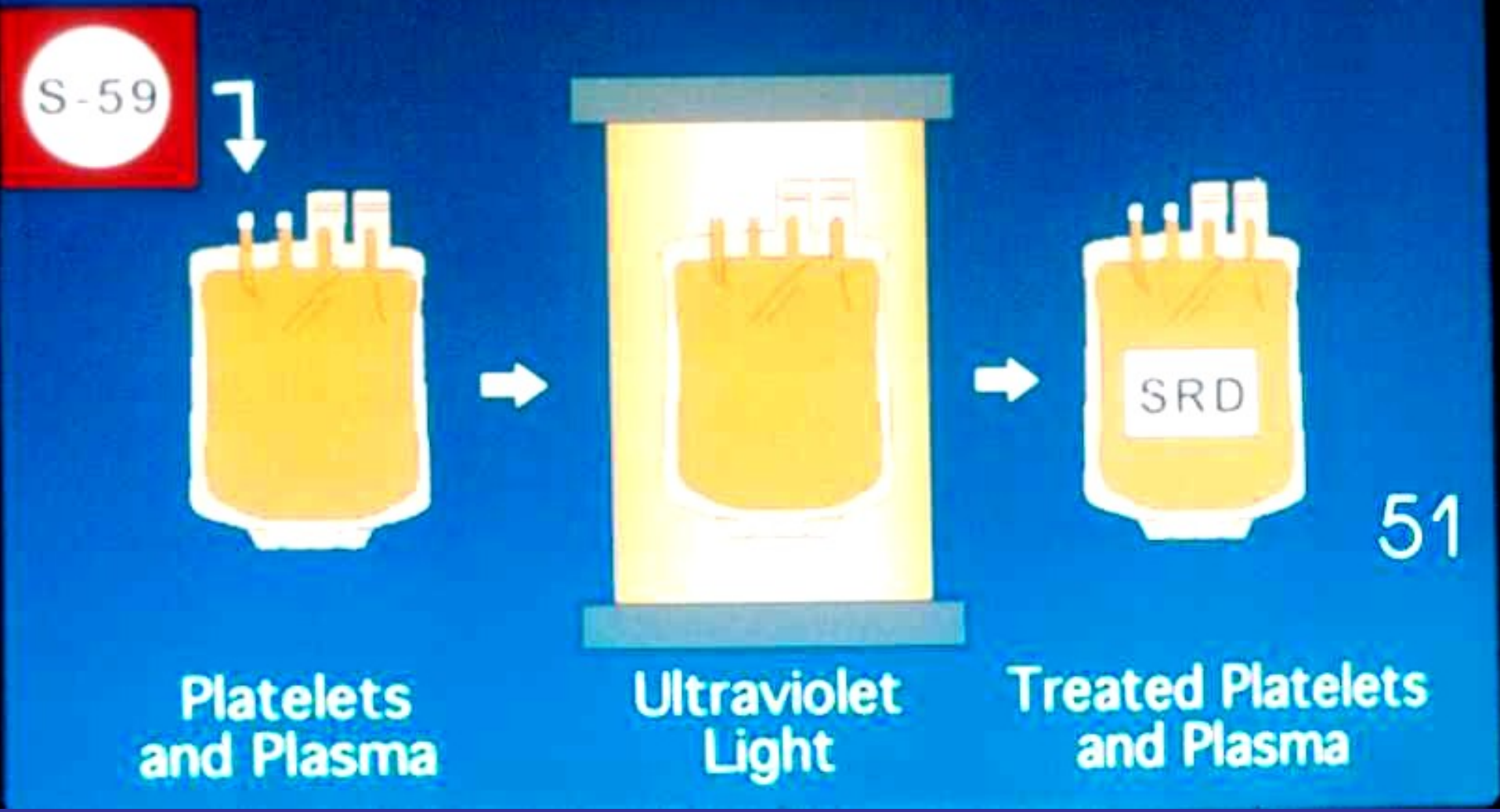
Latex beads



Mixture of 8-11 beads

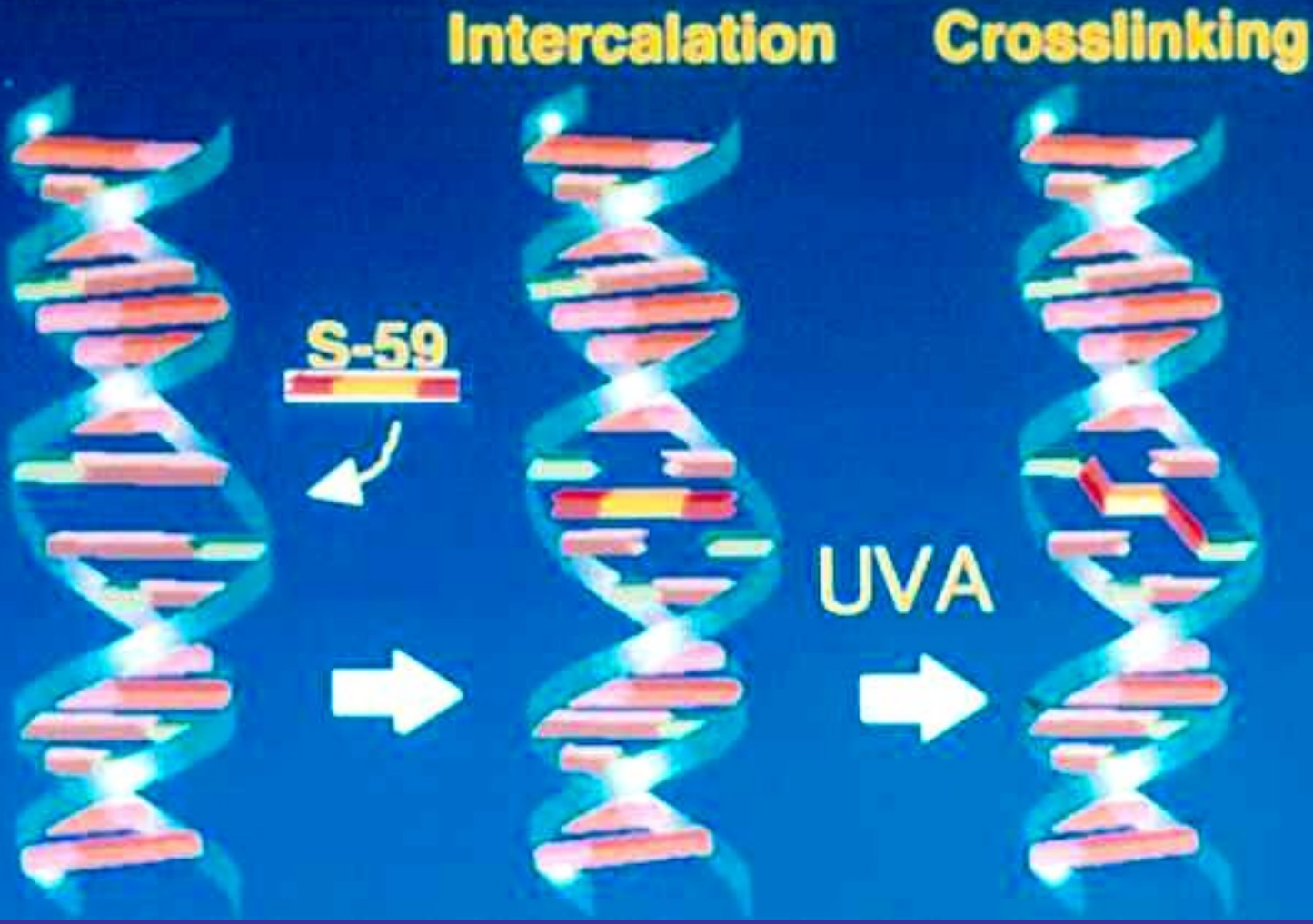


Process For Platelet Decontamination



51

S-59 Reaction Mechanism



Elimination of Pathogen-Contaminated Apheresis Platelets

(Post-Implementation of WNV NAT Testing & Bacterial Screening)

Pathogen	Risk Eliminated with PCT Platelets	Contaminated Units Eliminated Annually
HIV-1 and 2[a]	**1:2,135,000**	**0.6**
HBV[a]	**1:205,000**	**6.2**
HCV[a]	**1:1,935,000**	**0.7**
HTLV-I and II[a]	**1:2,993,000**	**0.4**
West Nile Virus[b]	**1:560,000 to 1:370,000**	**2.3 to 3.4**
Bacteria[c]	**1:29,000**	**43.6**
***T. cruzi* (Chagas' Disease)[d]**	**1:42,000**	**30.1**
Plasmodium sp (malaria)[d]	**1:4,000,000**	**0.3**
Cumulative Total (Post-Testing)		84.2 to 85.3

a. Dodd et al. 2002
b. Biggerstaff and Peterson 2002
c. Blajchman 2002
d. AABB Technical Manual 2002

Elimination of CMV Transmission

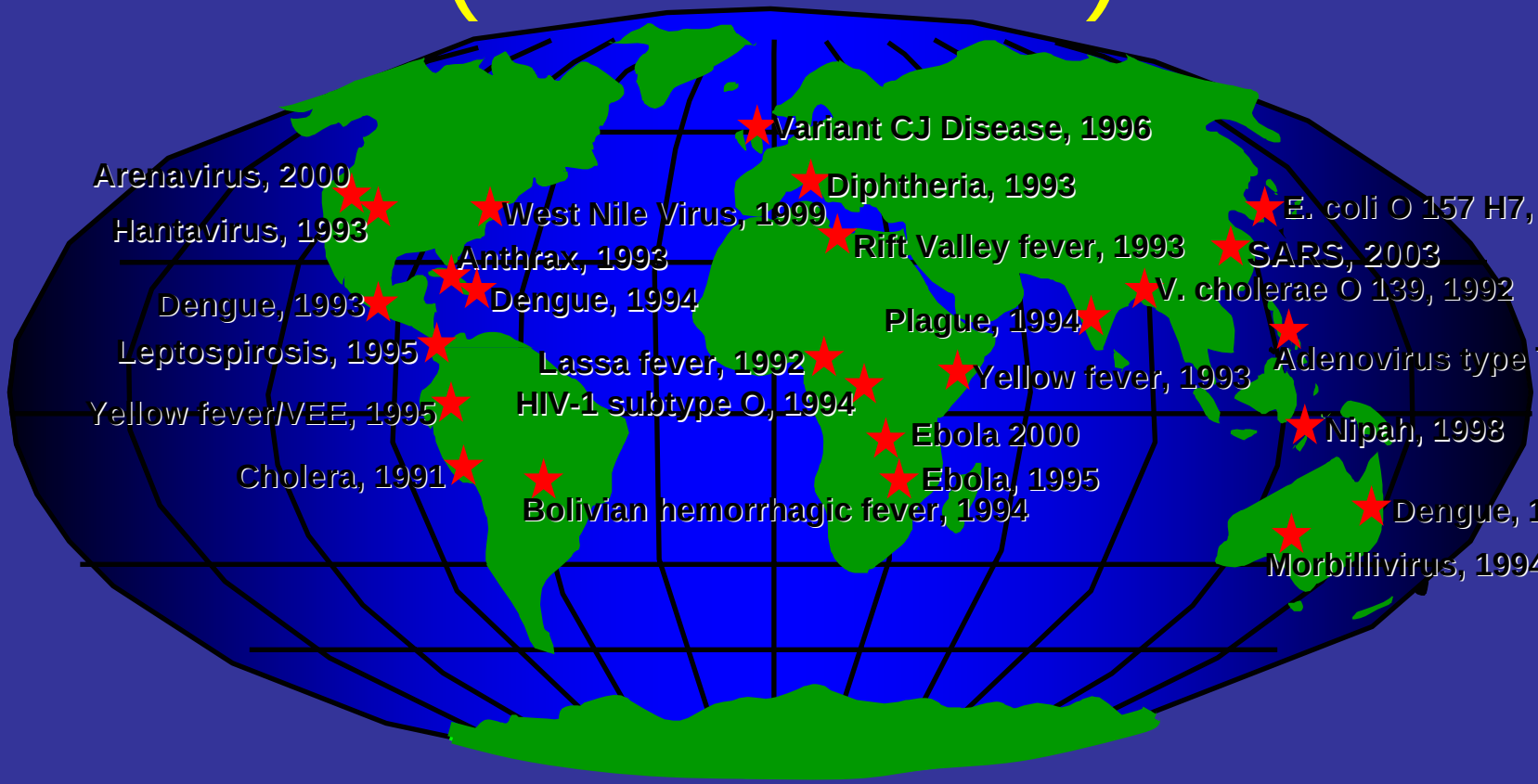
- Serious problem for CMV-negative patients undergoing myeloblative therapy
- Measureable failure rate to reduce risk of CMV transmission
  - 1% in CMV sero-negative products
  - 2.4% in leukoreduced products
- Symptomatic disease requiring treatment in ~30% of patients infected

a. Bowden et al. 1995
b. Laupacis et al. 2001

Disadvantages of Testing

- Sensitivity, specificity
- Delay in testing results
- Window periods
- Elimination of donors
- Lag time between pathogen identification and development of a screening assay

Recent Emerging Agents/Diseases (1991 – 2000)



kindly provided by Dr. B Horowitz

Paradigm - 2002

Has to be a paired control in same donor

Typical control - “ROP” - regular old platelets

- at end of licensed storage interval

- perhaps worst case scenario

Problems With Paradigm

- No “line in the sand” - ? 40% Rec., 5 day MCL
- No delineation of acceptable inferiority for test vs control, if any
- ROP will vary widely from study to study
- Creeping inferiority: $X_O \longrightarrow X_1 \longrightarrow X_2 \ldots X_n$

A Proposal

- Control should be fresh platelets
- Experimental results should be expressed as % of control
- Acceptable after storage:
  - Recovery: 2/3 fresh
  - Survival: 1/2 fresh
- Acceptable to have a predetermined reduction for experimental relative to extent of patient benefit

Methodologic Issues in the Use of Bleeding As An Outcome in Transfusion Studies

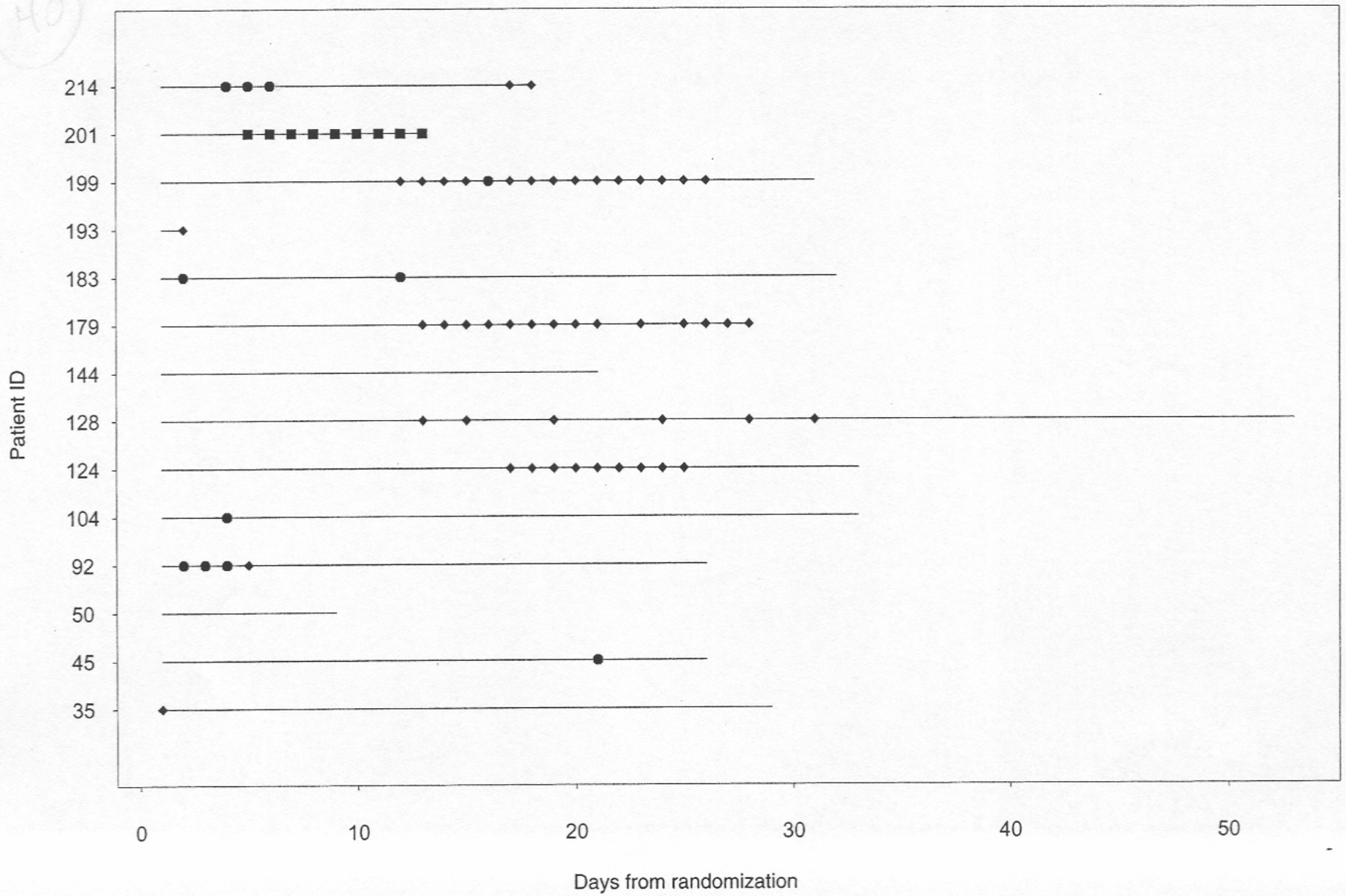
Heddle et al. Transfusion 43, 2003

Questioning by trained, blinded observer

Classification

Grade

1. Petechiae
2. Mild bleeding
3. Gross blood loss
4. Debilitating, fatal bleeding


Patient ID
214
201
199
193
183
179
144
128
124
104
92
50
45
35
0
10
20
30
40
50
Days from randomization