

Ποιότητα πλάσματος

Μαριάννα Πολίτου

Αιματολόγος

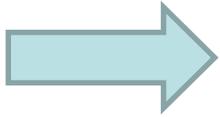
Αν.Καθηγήτρια Ιατρικής Σχολής ΕΚΠΑ
Διεύτρια Αιματολογικού Εργαστηρίου
Ν.Υ.Αιμοδοσίας ΑΡΕΤΑΕΙΟ Νοσοκομείο

	AABB ¹		UK ²		EU ³
	Fresh-Frozen plasma (FFP)	Plasma frozen within 24 hours (FP24)	FFP	MB FFP	
Time from donation to freezing	<8 hours for CPD <6 hours for ACD	<24 hours	No longer stated – must meet FVIII levels stated		Preferably < 6hours <18 hours if unit refrigerated <24 hours if whole blood rapidly cooled and stored 20-24°C
Storage once frozen	≤ -18°C 12 months ≤ -65°C 7 years	≤ -18°C 12 months	≤ -30°C 2 years	≤ -30°C 2 years	-18°C to -25°C 3 months ≤ -25°C 3 years
Storage once thawed	<24 hours 1-6°C ('thawed FFP') <5 days at 1-6°C labelled as 'thawed plasma'	<24 hours 1-6°C ('thawed FP24') <5 days at 1-6°C labelled as 'thawed plasma'	<24 hours 2-6°C	<24 hours 2-6°C	Use immediately
Clotting factor requirement for routine QM	None		>75% units >0.70 IU/ml FVIII	>75% units >0.50 IU/ml FVIII	> 0.70 IU/ml FVIII

- Ολικά αίματα φτάνουν στην Αιμοδοσία από εξόρμηση σε νησί μετά από 48 ώρες από τη συλλογή (συντηρημένα στους 4° C)
- Μπορείτε να παρασκευάσετε φρέσκο κατεψυγμένο πλάσμα?



- Σε πόση ώρα από τη συλλογή του ολικού αίματος πρέπει να καταψυχθεί το πλάσμα για να ονομάζεται ΦΚΠ;
Α. 6 ώρες
Β. 12 ώρες
Γ. 18 ώρες
Δ. 24 ώρες



- Σε τι θερμοκρασία συντηρείται το ολικό αίμα μέχρι τη φυγοκέντρησή του για την παρασκευή πλάσματος;
- 4°C
- 20°C

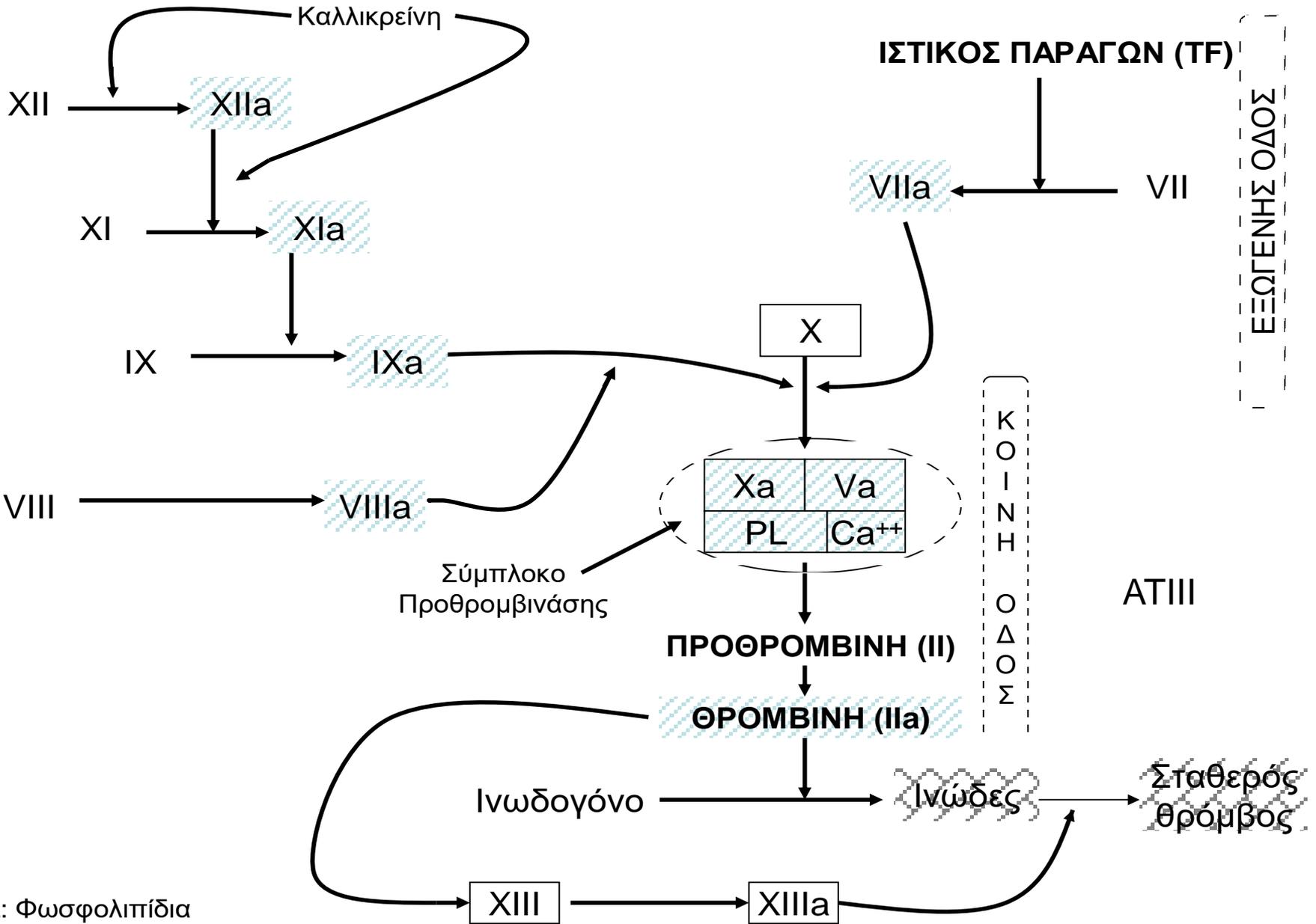
Σύσταση πλάσματος

- Περισσότερες από 700 πρωτεΐνες
- Μεταφορείς
- Ανοσοσφαιρίνες
- Λιποπρωτεΐνες
- Συμπλήρωμα
- Παράγοντες πήξης
- Φυσικοί αναστολείς

- Το πρόσφατα κατεψυγμένο πλάσμα (Fresh Frozen Plasma, FFP) περιέχει
- φυσιολογικά επίπεδα
- όλων των σταθερών παραγόντων πήξης, λευκωματίνη,
- ανοσοσφαιρίνες και
- τουλάχιστον το 70% της αρχικής ποσότητας του FVIII.

ΕΝΔΟΓΕΝΗΣ ΟΔΟΣ

ΕΞΩΓΕΝΗΣ ΟΔΟΣ



Φυσιολογική διακύμανση

Coagulation factors in FFP

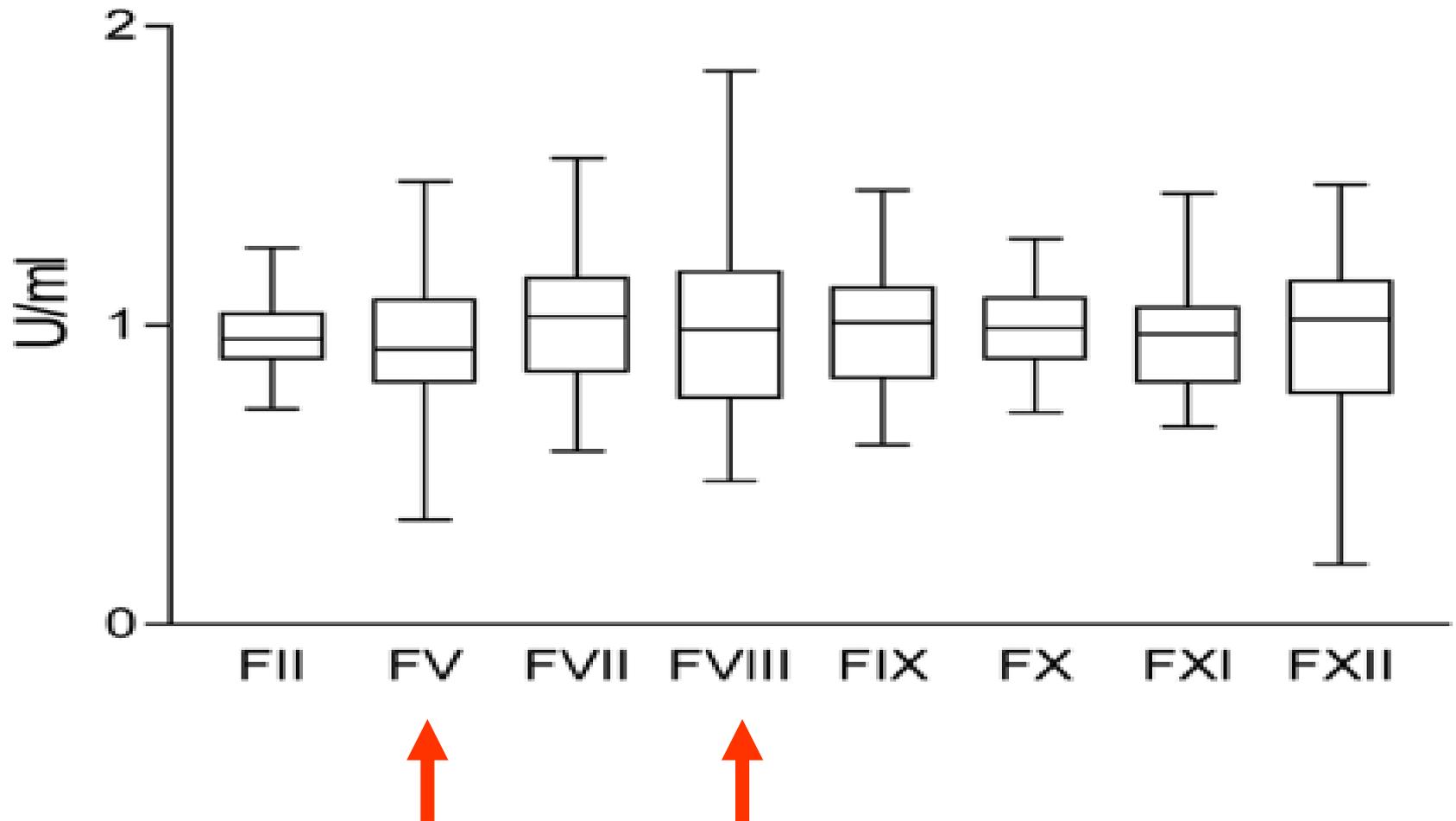
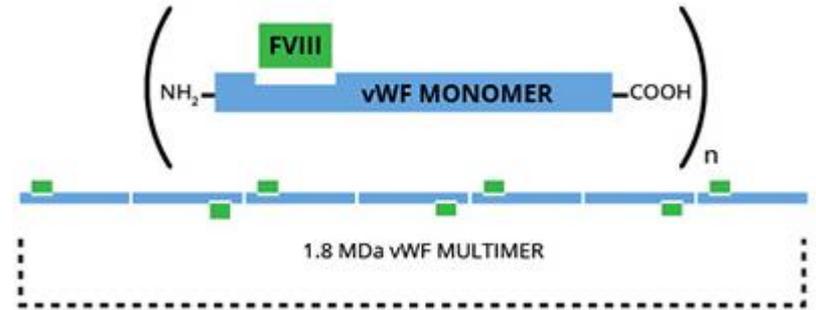
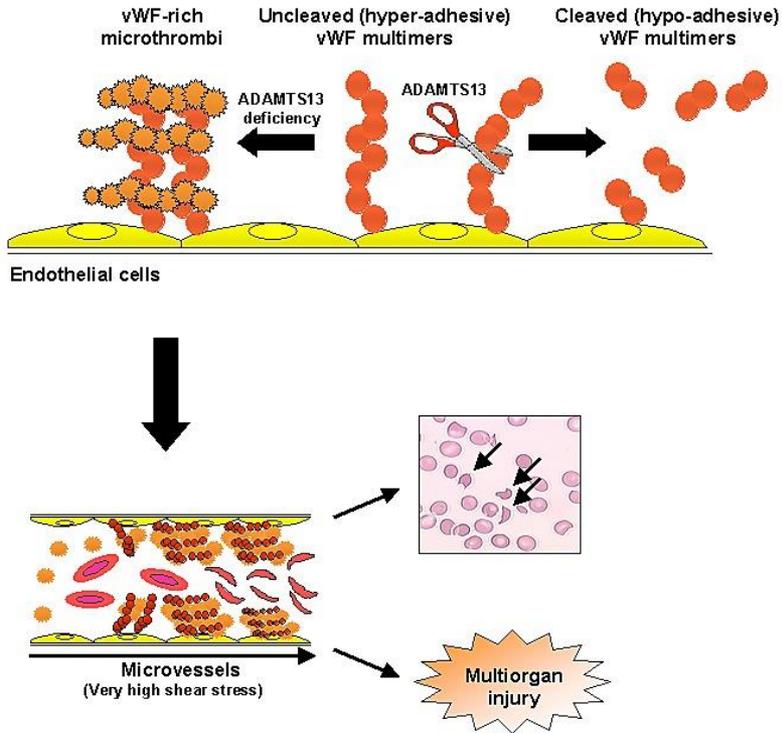
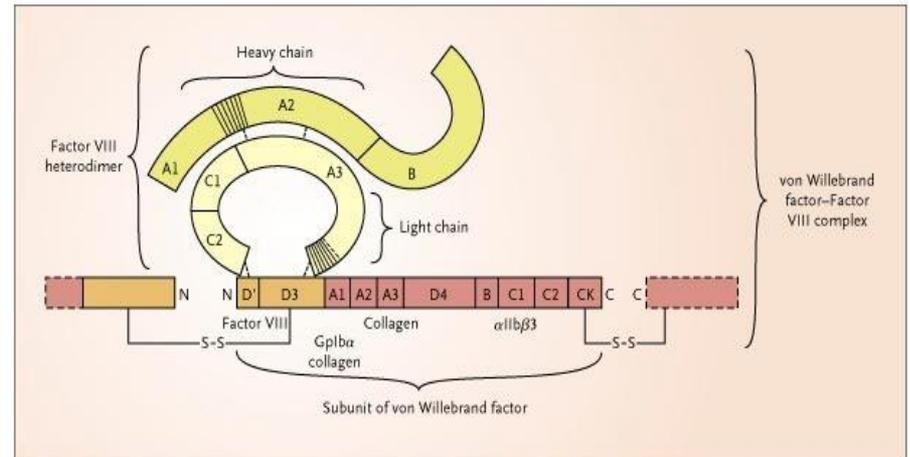


Figure S1



Hyperadhesive high molecular weight von Willebrand factor (vWF) multimers (●); ADAMTS13 (✂); platelets (⊙); erythrocytes (◐).



Υπολειπόμενα κύτταρα στο πλάσμα

- Μπορούν να προκαλέσουν ανοσολογικές αντιδράσεις και απελευθέρωση πρωτεολυτικών ενζύμων

	Απαιτήσεις ποιότητας	
	RBC < $6.0 \times 10^9 / L$ WBC < $0.1 \times 10^9 / L$ PLT < $50 \times 10^9 / L$	1% όλων των μονάδων Τουλάχιστον 4 μονάδες / μήνα

Επαφή του πλάσματος με τα κυτταρικά συστατικά - Δράση της θερμοκρασίας

- Ενεργοποίηση της κίνησης του πλάσματος από το κρύο [Over, 1990].
- [Favaloro, 2004] σημαντική ελάττωση της δραστηριότητας του FVIII και του vWF στο πλάσμα που αποχωρίζεται από το ολικό αίμα μετά από 3.5 hr στους 4°C, σε σχέση με πλάσμα που αποχωρίζεται μετά από 3.5 hr στους 22°C
 - 25% απώλεια FVIII
 - 50% απώλεια vWF:Ag (Non-O blood)
 - 60% απώλεια vWF:CB (Non-O blood)
- Η έκθεση του ολικού αίματος στο κρύο ενεργοποιεί την απελευθέρωση από τα AMT ή άλλα κύτταρα πρωτεασών του vWF (όπως η ADAMTS13) ή ρεδοκτασών του vWF (όπως η θρομβοσπονδίνη), που διασπούν τον vWF.
- Η ελάττωση του vWF οδηγεί και σε ελάττωση του FVIII:C

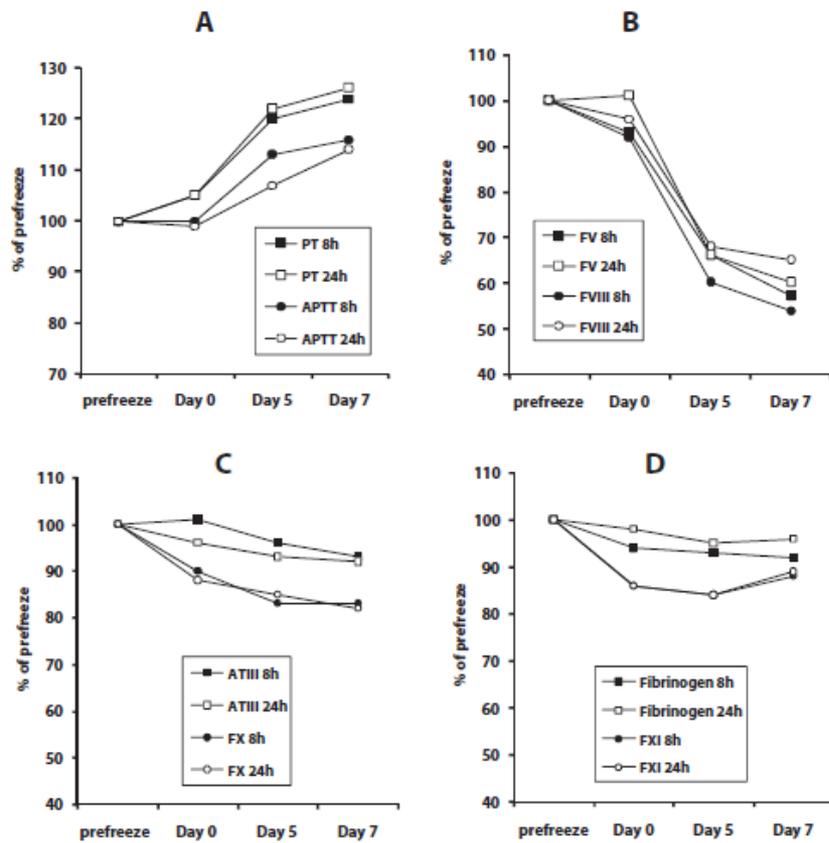
Χρόνος και Θερμοκρασία από την αιμοληψία μέχρι την κατάψυξη

- Ο χρόνος και η θερμοκρασία από τη συλλογή μέχρι την κατάψυξη επηρεάζει τους ασταθείς παράγοντες
- Ο FVIII ο πιο ασταθής και αλλαγές στη δραστικότητά του αντανακλούν μη μετρήσιμες αλλαγές και στους άλλους παράγοντες
- Καλύτερα συντήρηση του αίματος στους 20°C πριν τον αποχωρισμό [Over, 1990]

Χρόνος από τη συλλογή μέχρι τη φυγοκέντρηση

- Ιδανικά σε λιγότερο από 6 ώρες και οπωσδήποτε πριν περάσουν 18 ώρες αν η μονάδα διατηρηθεί σε ψυγείο
- <24 ώρες αν ψυχθεί γρήγορα (από ειδική συσκευή που εξασφαλίζει) και συντηρηθεί στους 20-24°C

- [Transfusion](#). 2010 Sep;50(9):1934-42. doi: 10.1111/j.1537-2995.2010.02648.x.
- **Stability of coagulation factors in plasma prepared after a 24-hour room temperature hold.**
- [Alhumaidan H1](#), [Cheves T](#), [Holme S](#), [Sweeney J](#).
- **BACKGROUND:**
- The manufacture of fresh-frozen plasma (FFP) requires that plasma be frozen within 8 hours of collection and 24-hour frozen plasma requires 1 to 6°C refrigeration before freezing. Manufacture of plasma after a room temperature hold for 24 hours, while convenient, could compromise clotting factor levels.
- **CONCLUSION:**
- Plasma manufactured after a 24-hour room temperature **hold contains coagulation factors comparable to FFP except for a possible reduction of up to 20% in FVIII.**



2. Graphic representations of changes in coagulation factor activity expressed as a percentage of the prefreeze value (Y-axis) in postthawing period up to Day 7 (X-axis). All prefreeze specimens are, by definition, 100%.

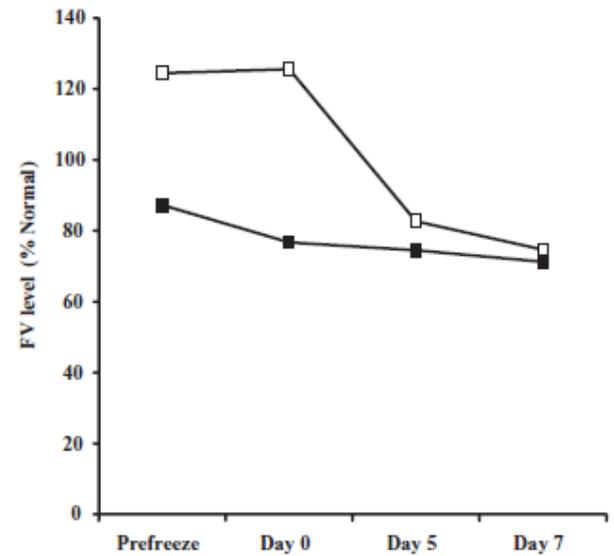


Fig. 4. FV levels (%) in PRP-24HRTFP (□) and WB-24HRTFP (■) over the 7 days of liquid storage. The rate of factor decline in FV activity is noticeably less in the WB-24HRTFP.



- Μπορεί το ολικό αίμα να χρησιμοποιηθεί για την παρασκευή πλάσματος που προορίζεται για κλασματοποίηση

Processing of plasma for fractionation to optimize factor VIII stability

Steps	Recommendations
Whole blood storage before plasma separation	<ul style="list-style-type: none">• Up to 18 to 20 h at 22 °C ± 1 °C• Not more than 8 h at 4 °C
Freezing	<ul style="list-style-type: none">• As soon as possible, within 24 hrs of blood collection or apheresis procedure^a
Freezing rate and temperature	<ul style="list-style-type: none">• As specified by plasma fractionator, following relevant regulations pertaining to the countries where plasma will be fractionated and products will be marketed• < -20 °C or colder
Storage temperature	<ul style="list-style-type: none">• -20 °C or colder, constant
Transportation temperature	<ul style="list-style-type: none">• -20 °C or colder, constant

- Το πλάσμα που προορίζεται για κλασματοποίηση (για σταθερούς παράγοντες, αλβουμίνη, ανοσοσφαιρίνες) μπορεί να φυγοκεντρηθεί σε 72 ώρες από τη συλλογή) και να καταψυχθεί σε θερμοκρασία $-20^{\circ} C$



- Ποιο από τα παρακάτω δεν επηρεάζει την ποιότητα του πλάσματος
- Α. Η λευκαφαίρεση
- Β. Το είδος του αντιπηκτικού
- Γ. Ο όγκος του ολικού αίματος
- Δ. Ο τρόπος παρασκευής από ολικό αίμα/αφαίρεση

Παράγοντες που επηρεάζουν την ποιότητα του πλάσματος

Τεχνική αιμοληψίας-Ικανοποιητική ανάμιξη με το αντιπηκτικό

Αποφυγή ενεργοποίηση της πήξης και άλλων πρωτεϊνών

British Journal of Haematology 2001, 114, 233–240

The effect of leucocyte depletion on the quality of fresh-frozen plasma

- [Br J Haematol.](#) 2001 Jul;114(1):233-40.
- **The effect of leucocyte depletion on the quality of fresh-frozen plasma.**
- [Cardigan R1](#), [Sutherland J](#), [Garwood M](#), [Krailadsiri P](#), [Seghatchian J](#), [Beard M](#), [Beckman N](#), [Williamson LM](#).

Abstract

- The aim of this study was to evaluate the quality of leucodepleted (LD) fresh-frozen plasma (FFP) produced using one of
- five whole blood filters (Baxter RS2000 & RZ2000, NPBI T2926, Macopharma LST1 and Terumo WBSP) or
- two plasma filters (Pall LPS1 and Baxter FGR7014).
- Whole blood or plasma was filtered within 8 h of collection at an ambient temperature. Samples were taken pre- and post filtration for analysis of coagulation factors and complement activation). All filtered units (209--286 ml) contained $< 5 \times 10^6$ residual leucocytes and $< 30 \times 10^9/l$ platelets.
- Statistically significant **losses of factors V, VIII, IX, XI and XII** and increases in markers of coagulation activation were observed which were dependent on filter type. None of the filters had a significant effect on von Willebrand factor (VWF) multimeric distribution or the activity of **VWF and factors II, VII or X**. The effect on levels of C3a appeared to be related to the filter surface charge: positively charged filters resulted in C3a generation, whereas negatively charged resulted in C3a removal. None of the observed changes are likely to be clinically significant unless subsequent processing of plasma (such as pathogen inactivation) results in further losses of coagulation factors.

Parameter	Baxter RS2000 (n = 10)		Baxter RZ2000 (n = 10)		Macopharma LST1 (n = 7)		NPBI T2926 (n = 10)		Terumo WBSP	
	Pre-filter	Post-filter	Pre-filter	Post-filter	Pre-filter	Post-filter	Pre-filter	Post-filter	Pre-filter	Post-filter
PT (s)	13 (12.0-14.4)	13.2* (12.1-15.0)	11.2 (10.4-12.1)	11.2 (10.3-12.3)	13.9 (12.9-15.3)	14.2* (13.1-16.2)	12.9 (12.1-16.6)	13.4** (13.0-17.4)	10.7 (10.1-11.8)	10.7 (10.1-11.6)
APTT (s)	32.3 (28.4-42.4)	32.4 (28.2-42.9)	27.3 (23.9-30.1)	26.8 (23.5-29.0)	33.3 (28.2-34.7)	33.8* (29.0-37.5)	32.2 (27.7-35.3)	32.8* (28.7-36.9)	26.1 (23.3-31.6)	25.8* (22.8-30.5)
Fibrinogen (g/l)	2.76 (1.95-3.81)	2.63 (1.67-3.42)	2.17 (1.55-2.78)	2.13 (1.52-2.73)	3.1 (2.35-4.85)	3.69* (2.67-5.00)	2.25 (2.09-3.06)	2.4 (2.06-3.03)	2.18 (1.87-2.65)	2.17 (1.78-2.61)
Prothrombin (IU/ml)	0.89 (0.70-1.02)	0.9 (0.73-1.04)	1.05 (0.88-1.23)	1.06 (0.86-1.25)	1.04 (0.95-1.09)	0.97 (0.89-1.11)	0.91 (0.76-1.13)	0.89 (0.74-1.03)	1 (0.89-1.12)	1 (0.89-1.10)
Factor V (U/ml)	0.95 (0.75-1.47)	0.96 (0.71-1.40)	0.9 (0.68-1.18)	0.91 (0.69-1.19)	1.14 (1.01-1.26)	0.96* (0.67-1.17)	1.06 (0.42-1.26)	0.85** (0.35-0.97)	0.75 (0.54-1.18)	0.77* (0.54-1.18)
Factor VII (IU/ml)	1.04 (0.60-1.30)	1.03 (0.61-1.38)	1.07 (0.75-1.36)	1.06 (0.74-1.34)	1.04 (0.79-1.48)	0.99 (0.75-1.56)	1.04 (0.74-1.38)	1.02 (0.70-1.41)	1 (0.58-1.33)	1 (0.58-1.33)
Factor VIII (IU/ml)	0.8 (0.56-1.73)	0.79 (0.59-1.70)	1.28 (0.89-1.97)	1.28 (0.87-1.90)	0.78 (0.62-1.50)	0.72 _y (0.48-1.36)	0.83 (0.69-1.28)	0.79* (0.68-1.23)	1.19 (0.94-1.49)	1.15** (0.93-1.49)
Factor IX (IU/ml)	0.91 (0.53-1.24)	0.96 (0.60-1.25)	0.99 (0.81-1.21)	0.97 (0.80-1.17)	0.96 (0.76-1.43)	0.91* (0.69-1.33)	1.06 (0.75-1.21)	1.01* (0.71-1.17)	1.06 (0.70-1.28)	1.05 (0.69-1.31)
Factor X (IU/ml)	0.95 (0.73-1.28)	0.95 (0.74-1.29)	1.04 (0.79-1.25)	1.05 (0.80-1.27)	1.08 (0.90-1.17)	1.05 (0.89-1.20)	0.96 (0.72-1.16)	0.93 (0.72-1.10)	1 (0.70-1.26)	1 (0.71-1.21)
Factor XI (U/ml)	0.94 (0.69-1.23)	0.99 (0.71-1.34)	1.09 (0.94-1.40)	1.06 (0.91-1.33)	0.92 (0.80-1.64)	0.73* (0.66-1.44)	0.98 (0.74-1.21)	0.91* (0.67-1.12)	0.94 (0.69-1.21)	0.95 (0.72-1.13)
Factor XII (U/ml)	0.93 (0.18-1.40)	0.9 (0.20-1.41)	1.11 (0.80-1.32)	1.11 (0.79-1.31)	1.03 (0.71-1.48)	0.90* (0.65-1.35)	0.86 (0.51-1.48)	0.82 (0.50-1.47)	1.11 (0.79-1.40)	1.09 (0.84-1.36)

* $P < 0.05$; ** $P < 0.01$; *** $P < 0.005$; **** $P < 0.0001$ refers to statistical significance from the Wilcoxon rank test between pre- and post-filtration samples.

Data are represented by the median (range).

Table IV. von Willebrand factor, coagulation and complement activation markers in plasma from whole blood filtration.

Parameter	Baxter RS2000 (n = 10) Neutral surface charge of filter		Baxter RZ2000 (n = 10) Positive surface charge of filter		Macopharma LST1 (n = 7) Neutral surface charge of filter		NPBI T2926 (n = 10) Neutral surface charge of filter		Terumo WBSF Neutral surface charge of filter	
	Pre-filter	Post-filter	Pre-filter	Post-filter	Pre-filter	Post-filter	Pre-filter	Post-filter	Pre-filter	Post-filter
VWF antigen (%)	113 (69-171)	114 (74-169)	116 (59-165)	114 (60-157)	103 (0.68-1.43)	98 (0.73-1.47)	123 (81-175)	122 (89-175)	108 (83-135)	108 (79-131)
VWF activity (IU/ml)	0.91 (0.59-1.37)	0.91 (0.57-1.35)	1.04 (0.58-1.37)	1.07 (0.55-1.41)	0.88 (0.55-1.92)	0.93 (0.60-1.58)	1.11 (0.76-1.59)	1.1 (0.67-1.63)	0.88 (0.65-1.15)	0.88 (0.67-1.18)
Factor XIIa (ng/ml)	NA	NA	2.42 (0.90-3.68)	2.77** (0.90-4.25)	NA	NA	NA	NA	1.07 (0.26-2.27)	1.19 (0.41-2.34)
Pro F1 + 2 (nmol/l)	0.63 (0.36-1.04)	0.60* (0.34-1.14)	0.68 (0.40-1.14)	0.73* (0.41-1.22)	0.7 (0.38-0.97)	0.79* (0.52-1.08)	0.55 (0.35-0.75)	0.58 (0.36-0.85)	NA	NA
C3a (ng/ml)	361 (160-623)	333* (160-506)	416 (117-757)	3997*** (665-12010)	347 (73-601)	260* (30-537)	425 (198-700)	291* (< 30-856)	280 (141-510)	389* (172-711)
sC5b-9 (ng/ml)	99 (27-311)	108 (31-320)	NA	NA	63 (38-92)	91 (32-226)	72 (< 25-145)	141* (< 25-234)	NA	NA

* $P < 0.05$; ** $P < 0.01$; *** $P < 0.005$; **** $P < 0.0001$ refers to statistical significance from the Wilcoxon rank test between pre- and post-filtration samples.

Data are represented by the median (range). NA, not available.

- [Vox Sang.](#) 2004 Oct;87(3):156-64.
- **Effects of extended storage of whole blood before leucocyte depletion on coagulation factors in plasma.**
- [Kretzschmar E1](#), [Kruse F](#), [Greiss O](#), [Paunovic D](#), [Kallweit T](#), [Trobisch H](#).
- [Vox Sang.](#) 2005 Jan;88(1):72.
- **Abstract**
- **BACKGROUND AND OBJECTIVES:**
- The aim of this study was to evaluate the quality of leucocyte-depleted plasma produced from leucocyte-depleted whole blood, stored for different periods of times before filtration through polyurethane filters.
- **MATERIALS AND METHODS:**
- Whole blood was collected, from 48 voluntary donors, into quadruple blood bag sets with integrated whole-blood filters, and stored at room temperature for 1, 2, 6, or 18 h before filtration. Five samples were taken: one directly from the donor; one immediately after collection; one before and one after filtration; and one from plasma units before freezing. All samples were analysed for the following parameters: prothrombin time; activated partial thromboplastin time; prothrombin fragments F1+2; fibrinogen; factors VIII, XI and XII; von Willebrand factor antigen; ristocetin cofactor activity; collagen-binding capacity; multimers; and complement C3a-desArg.
- **RESULTS:**
- Different whole-blood storage times before filtration did not have a significant effect on the stability of coagulation factors. The activity of all investigated coagulation factors in plasma was generally above 90 U/dl, even after 18 h of storage of whole blood before filtration. von Willebrand factor multimeric distribution remained stable throughout the process. However, activation of complement did occur during storage.
- **CONCLUSIONS:**
- **Leucodepleted plasma originating from leucodepleted whole blood maintains a satisfactory level of coagulation factors, even after the storage of whole blood for 18 h at room temperature before filtration.**

ΑΝΤΙΠΗΚΤΙΚΟ: CPD ή ACD

- Η βιβλιογραφία είναι αντικρουόμενη για τη δράση των αντιπηκτικών στην ανάκτηση του FVIII
- Η χρήση του ACD μπορεί να οδηγήσει σε λιγότερο FVIII από ότι η χρήση του CPD.

Examples of anticoagulant solutions commonly used in the preparation of plasma for fractionation

	Composition	Recovered plasma	Ratio per 100ml blood	Apheresis plasma
ACD-A	Sodium citrate dihydrate 22.0 g/l Citric acid hydrous 8.0 g/l Dextrose monohydrate 25.38 g/l pH (25 °C) 4.7–5.3	×	15	(×)
ACD-B	Sodium citrate dihydrate 13.2 g/l Citric acid hydrous 8.0 g/l Dextrose monohydrate 15.18 g/l pH (25 °C) 4.7–5.3	×	25	
CPD	Sodium citrate dihydrate 26.3 g/l Citric acid hydrous 3.7 g/l Dextrose monohydrate 25.5 g/l Sodium biphosphate 2.22 g/l Sodium hydroxide 1 N (pH adjustment) pH (25 °C) 5.3–5.9	×	14	(×)
CPD-A	Sodium citrate dihydrate 26.3 g/l Citric acid hydrous 2.99 g/l Dextrose monohydrate 29 g/l Sodium biphosphate 2.22 g/l Adenine 0.27 g/l Sodium hydroxide 1 N (pH adjustment) pH (25 °C) 5.3–5.9	×	14	
CP2D	Sodium citrate dihydrate 26.3 g/l Citric acid hydrous 3.7 g/l Dextrose monohydrate 50.95 g/l Sodium biphosphate 2.22 g/l Sodium hydroxide 1 N (pH adjustment) pH (25 °C) 5.3–5.9	×	14	
4% Citrate	Sodium citrate dihydrate 40 g/l Citric acid hydrous: as required for pH adjustment pH (25 °C) 6.4–7.5		6.25	×

- Αντίστροφη σχέση ανάμεσα στη συγκέντρωση των κιτρικών και τη σταθερότητα του παράγοντα FVIII
- *Transfusion*, 28 (1988), pp. 248-252
- Το πλάσμα από ολικό αίμα που έχει χρησιμοποιηθεί αντιπηκτικό citrate-phosphate-dextrose (CPD) περιέχει διπλάσια ποσότητα κιτρικών από το πλάσμα που έχει συλλεχθεί με πλασμαφαίρεση με 4% sodium citrate (σε αναλογία 1:16 αντιπηκτικό /αίμα)
- Πλάσμα που προέρχεται από ολικό αίμα έχει μικρότερη συγκέντρωση παράγοντα V, IX
- *Transfusion*, 39 (1999), p. 1266

- Ποιο από τα παρακάτω δεν επηρεάζει την ποιότητα του πλάσματος
- Α. Η λευκαφαίρεση
- Β. Το είδος του αντιπηκτικού
- Γ. Ο όγκος του ολικού αίματος
- Δ. Ο τρόπος παρασκευής από ολικό αίμα/αφαίρεση



- Σε πόση ώρα πρέπει το πλάσμα να καταψυχθεί.
- Να φτάσει σε θερμοκρασία $-30^{\circ} C$
- 1 ώρα
- 2ώρες
- 6ώρες
- 12ώρες

Time to Freeze 700 ml, 1500 ml Plasma Under Varying Conditions

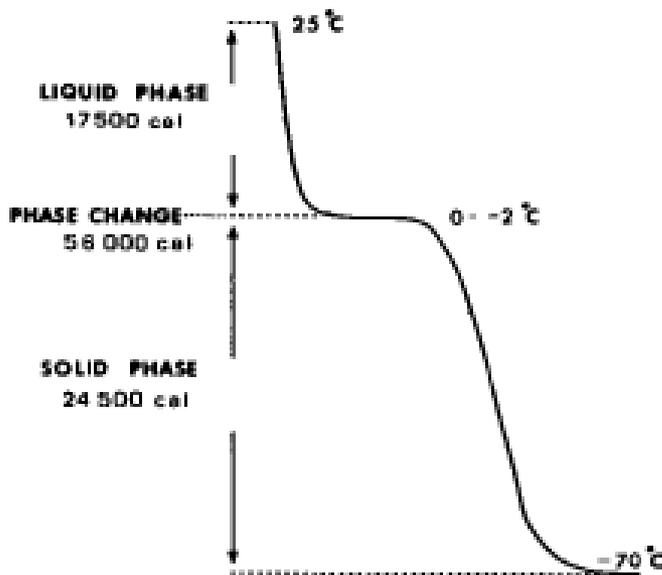


FIG. 2. Freezing of 700 ml plasma. Energy consumption at different stages of freezing.

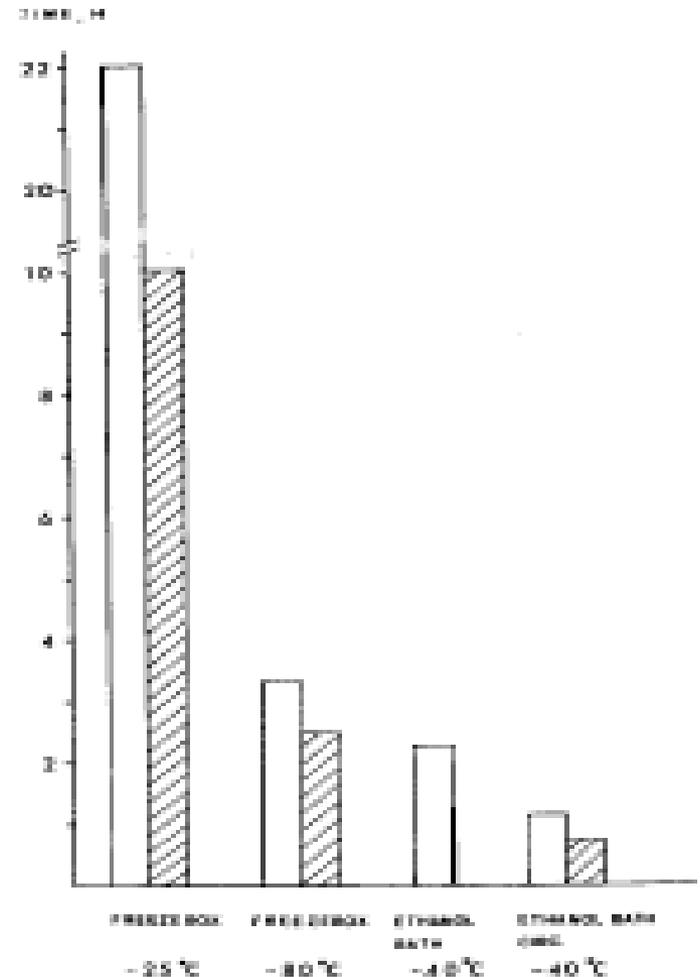
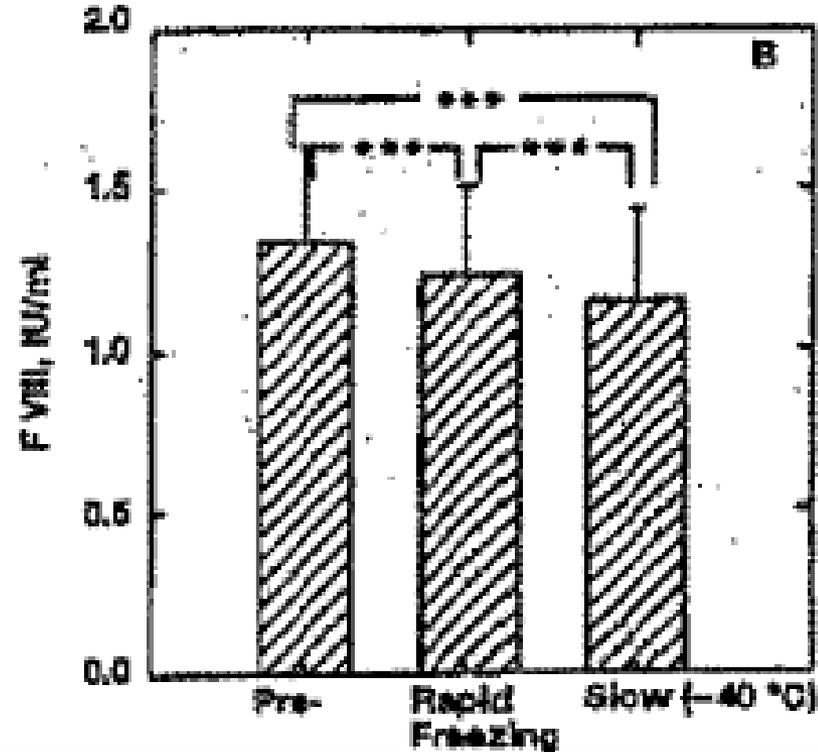
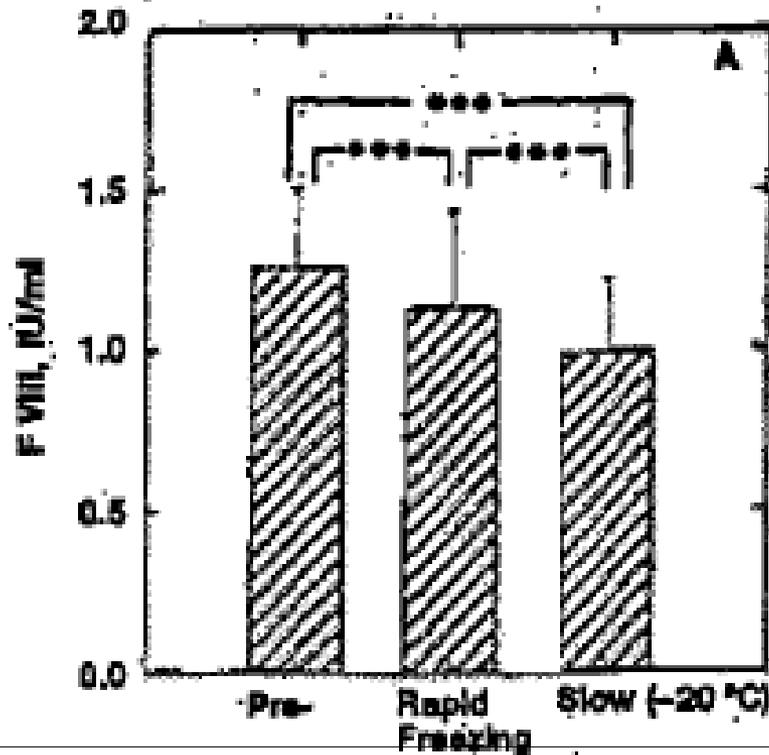


FIG. 3. Plasma freezing time to -25°C with different equipment. Plasma frozen in 750-ml (filled symbols) and in 1500-ml (hatched symbols) containers.

Carlebjork, 1986

Δραστικότητα του FVIII στο FFP μετά από αργή και ταχεία κατάψυξη

Akerblom, 1992



Ταχεία κατάψυξη: $-40\text{ }^{\circ}\text{C}$, $<40\text{ min}$ (απώλεια 8% FVIII)

Αργή κατάψυξη: in $-40\text{ }^{\circ}\text{C}$ or $-20\text{ }^{\circ}\text{C}$ (απώλεια 12 και 20% FVIII αντίστοιχα)

• Καμία διαφορά FVII, vWF, TAT complex, C1-esterase inhibitor

Η ανάκτηση του FVIII στο FFP επηρεάζεται από την ταχύτητα/ρυθμό κατάψυξης

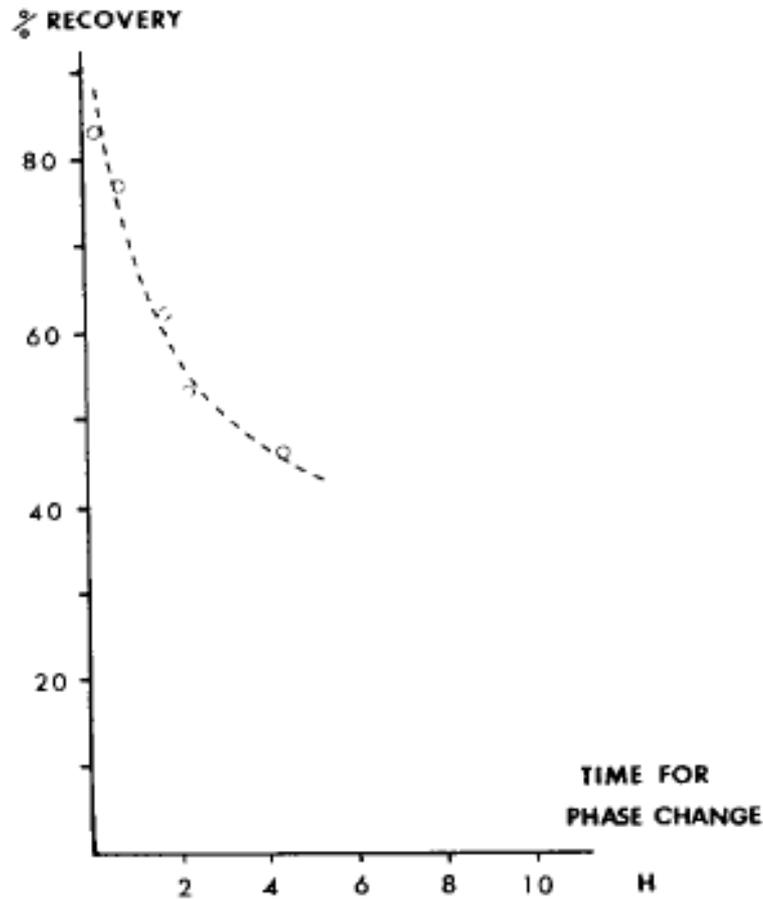


FIG. 6. Plasma factor VIII recovery correlated to phase change time at the freezing point.

- Σε πόση ώρα πρέπει να καταψυχθεί.
- Να φτάσει σε θερμοκρασία $-30^{\circ} C$
- 1 ώρα
- 2 ώρες
- 6 ώρες
- 12 ώρες



**MBF 12
High Performance Contact
Shock Freezer**

0+

FF(0)

AB B



ANERETA FFC
ANERETA FFC





- Για πόσο χρονικό διάστημα συντηρείται το ΦΚΠ
- 6 μήνες
- 1 χρόνο
- 2 χρόνια
- 3 χρόνια

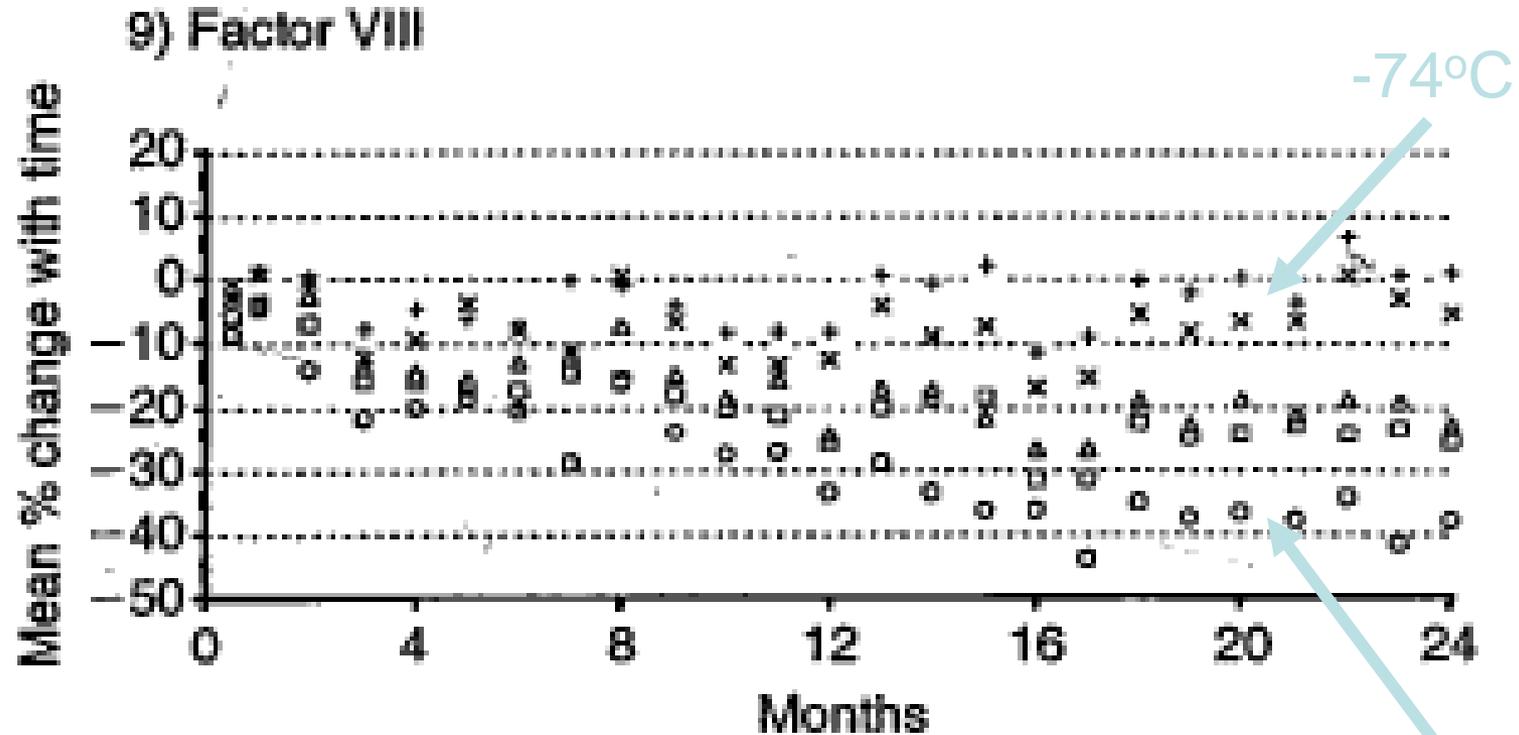
Blood Coagulation and Fibrinolysis 2001, 12:229-236

Stability of coagulation proteins in frozen plasma

B. Woodhams, O. Girardot, M.-J. Blanco, G. Colesse and Y. Gourmelin

- [Blood Coagul Fibrinolysis](#). 2001 Jun;12(4):229-36.
- **Stability of coagulation proteins in frozen plasma.**
- [Woodhams B1](#), [Girardot O](#), [Blanco MJ](#), [Colesse G](#), [Gourmelin Y](#).
- [Author information](#)
- **Abstract**
- This study reports on the frozen stability of all commonly measured coagulation proteins in normal citrated plasma: activated partial thromboplastin time, prothrombin time (%), thrombin time and fibrinogen (Clauss); clotting assays for factors II, V, VII, VIII, IX, X, XI and XII; functional assays for protein C (clotting), protein S (clotting), antithrombin (chromogenic) and plasminogen (chromogenic); and immunological assays for von Willebrand factor and D-dimer.
- All these factors listed are stable for up to 3 months if frozen at -24 degrees C or lower.
- At -74 degrees C, all these factors (allowing for 10% variation) were stable for at least 18 months, most were stable for 24 months.
- The number of proteins showing > 5% variation over baseline after 6 months storage indicates that some decay does occur even at -74 degrees C.
- The best stability, especially at -24 degrees C, was obtained when small samples (1 ml) were stored in screw-cap tubes with a minimum dead space.
- **The decrease in stability of the coagulation proteins directly correlates with the effect of temperature and time.**

Ελάττωση της δραστικότητας FVIII

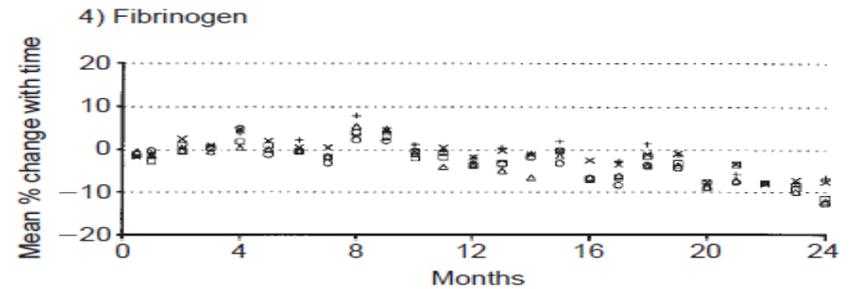
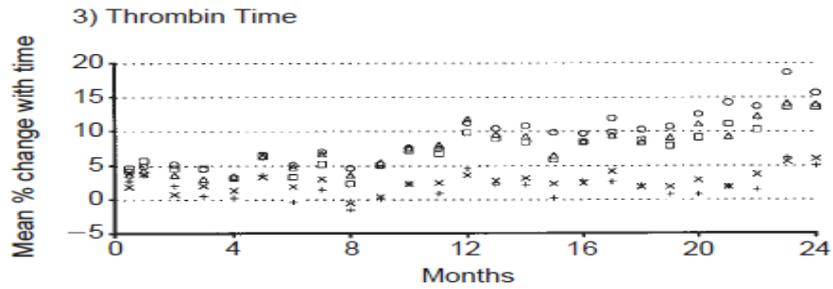
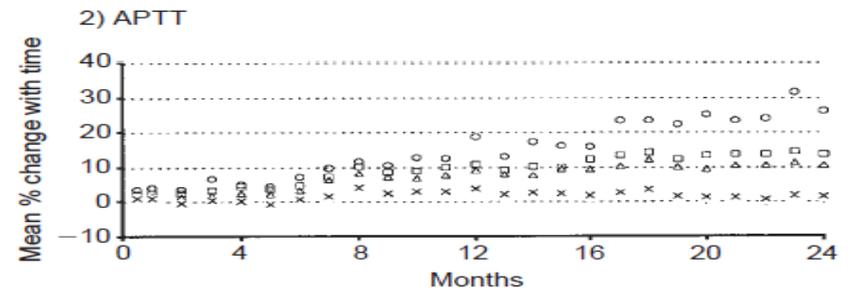
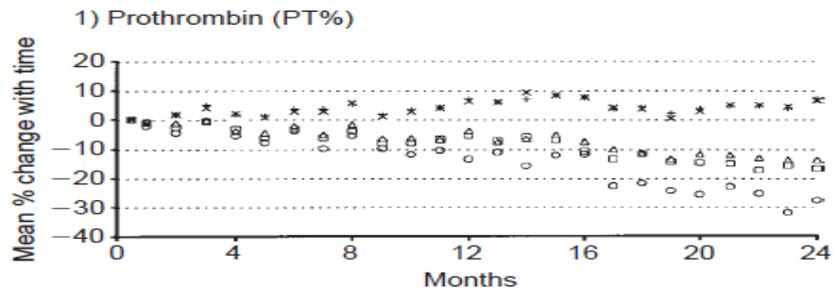


5 ml aliquots of plasmapheresis plasma

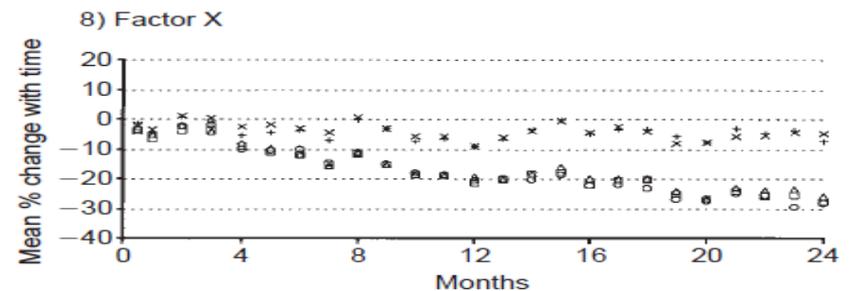
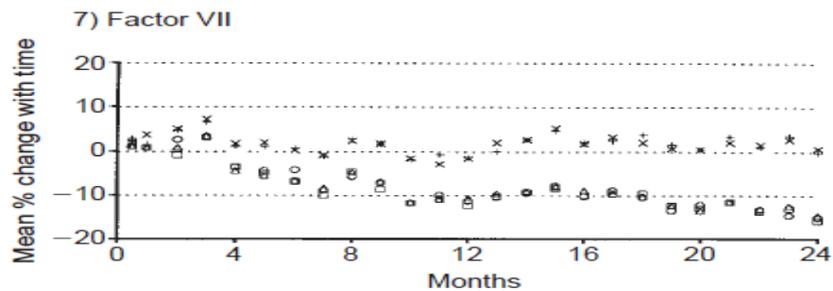
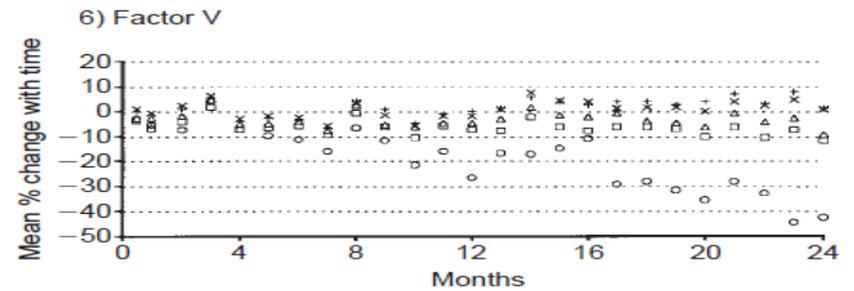
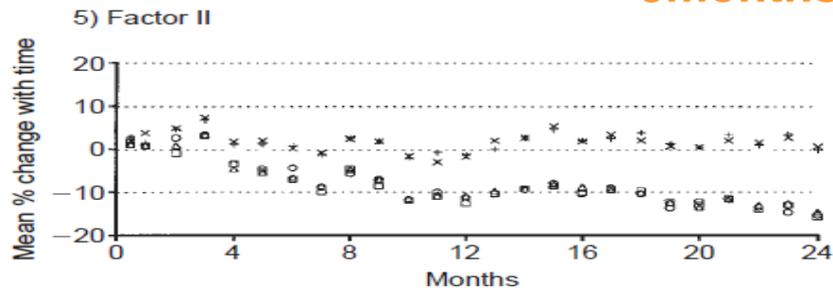
Frozen and stored at -74°C (x)

Frozen and stored at -24°C (o)

Woodhams, 2001



3months



Συντήρηση/ Αποθήκευση

- 36 μήνες σε θερμοκρασία χαμηλότερη από -25°C
- 3 μήνες σε θερμοκρασία από -18 - 25°C

Σε πόση ώρα πρέπει να γίνει η απόψυξη



Απόψυξη : ≤ 30 min



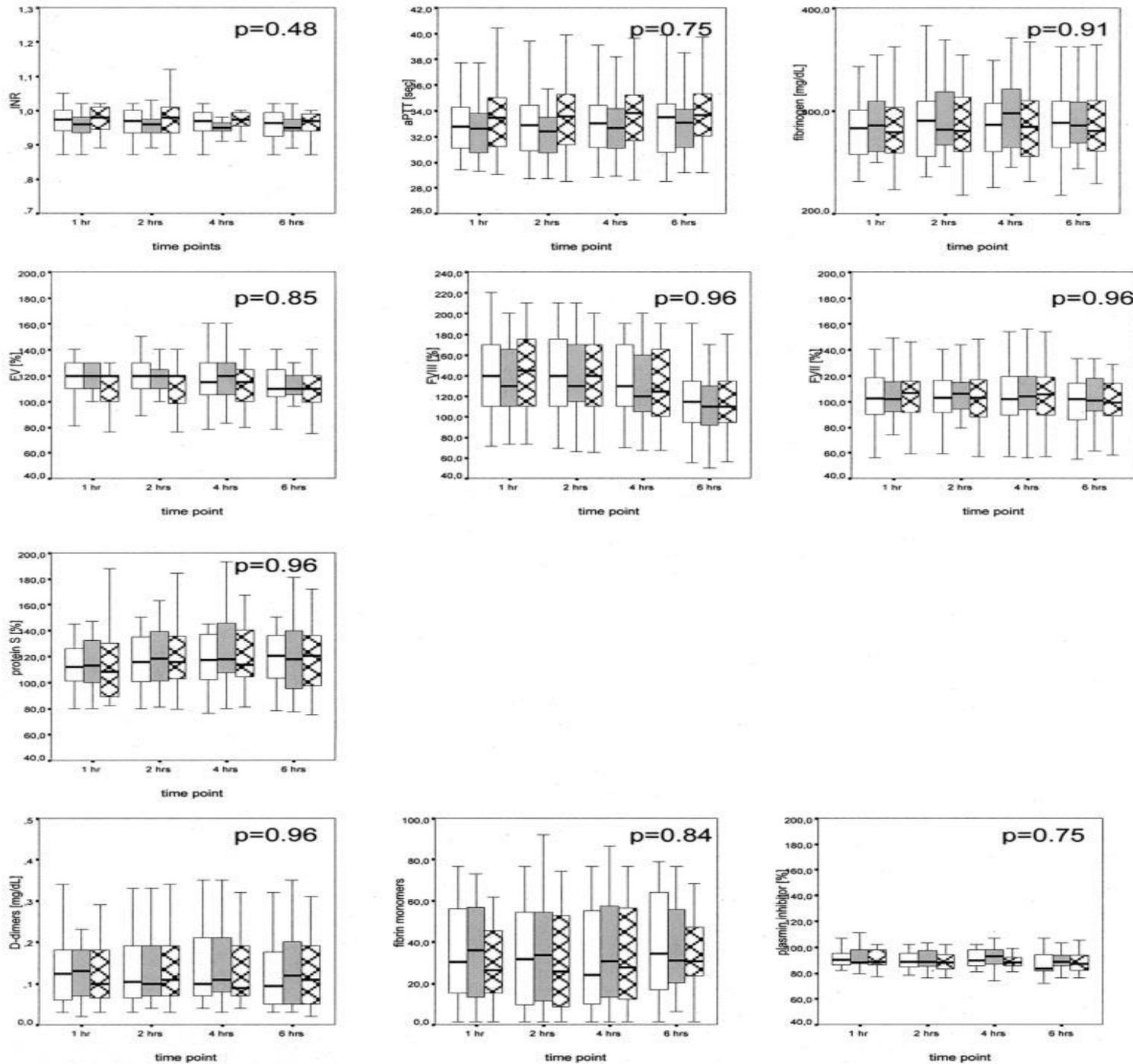


Figure 1. Impact of the thawing procedure on the activity of clotting factors and inhibitors between 1 h and 6 h after thawing. White boxes r unning warm water; grey boxes transfusion-therm 2000 ® ; hatched boxes: plasmatherm III ® ; FV factor V; FVII factor VII; FVIII factor VIII; DD d-dimers; FM fibrin monomers; 2-AP 2-antiplasmin; PS protein S.



- Τι περιλαμβάνει ο ποιοτικό έλεγχος

- Κατά μέσο όρο μετά την ψύξη και την απόψυξη >70% της ποσότητα VIII
- Κάθε 3 μήνες 10 μονάδες κατά τον πρώτο μήνα της αποθήκευσης
- Απουσία διαρροής
- Απουσία χρωματικών αλλοιώσεων ή ορατών θρόμβων



- Ξεπαγώνετε 3 μονάδες ΦΚΤΠ
- Ποια από τις παρακάτω μονάδες θα χορηγήσετε ποιές θα τα απορρίψετε

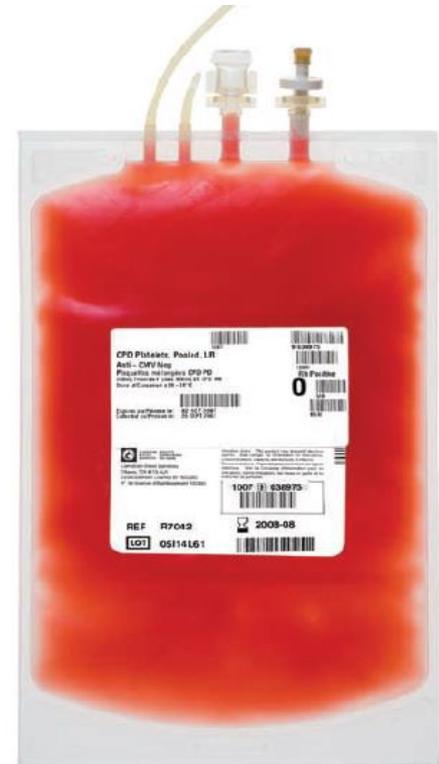


Lipemic Plasma



Icteric Plasma

mination in Pooled
latelets prepared
Coat method)
tion = 2.15 mL RBC.



nt

y

mination in Pooled
latelets prepared
Coat method)
tion = 2.15 mL RBC.





Plasma with Green Colour (green colour is commonly a result of donor taking oral contraceptive pill)

bright orange – Vitamin A and large quantities of carrots.

• • • • •



- Σας ζητούν από την κλινική 2 πλάσματα. Τα ξεπαγώνετε.
- Ο ασθενής πυρέσει. Πόση ώρα μετά αν τα διαθέσετε παραμένουν ΦΚΠ;
- 2 ώρες
- 4 ώρες
- 8 ώρες
- 12 ώρες
- Πού πρέπει να τα συντηρήσετε;
- 4° C
- 20° C

Table IV. Haemostatic factor content of thawed fresh-frozen plasma (FFP), and after storage at 4°C. A typical unit of 300 ml includes (IU/ml), except fibrinogen (g/l).

	Levels when freshly thawed	Levels at 24 h	Levels at 5 d
Fibrinogen	2.67	2.25	2.25
FII	80	80	80
FV	80	75	66
FVII	90	80	72
FVIII	92	51	41
FIX	100		
FX	85	85	80
FXI	100		
FXII	83		
FXIII	100		
Antithrombin III	100		
VWF	80*		

These values were determined in the Pathology Laboratories of Southampton University Hospitals Trust.

Protein C and antithrombin levels are in the 'normal range'.

*With some loss of HMW multimers, particularly if SD-treated.

Table 5. Average levels of FVIII in plasma following thawing and storage at 4°C

Paper	Day 0/1	After 24 hours	After 5 days
Sheffield et al 2011	0.86	0.63	0.51
Von Heymann et al 2009	1.00	0.80	0.70
Yazer et al 2008	0.72	0.60	0.69
Donwes et al 2001	Group A 1.07 Group B 1.03 Group O 0.70	0.76 0.74 0.51	0.63 0.67 0.41

	AABB ¹		UK ²		EU ³
	Fresh-Frozen plasma (FFP)	Plasma frozen within 24 hours (FP24)	FFP	MB FFP	
Time from donation to freezing	<8 hours for CPD <6 hours for ACD	<24 hours	No longer stated – must meet FVIII levels stated		Preferably < 6hours <18 hours if unit refrigerated <24 hours if whole blood rapidly cooled and stored 20-24°C
Storage once frozen	≤ -18°C 12 months ≤ -65°C 7 years	≤ -18°C 12 months	≤ -30°C 2 years	≤ -30°C 2 years	-18°C to -25°C 3 months ≤ -25°C 3 years
Storage once thawed	<24 hours 1-6°C ('thawed FFP') <5 days at 1-6°C labelled as 'thawed plasma'	<24 hours 1-6°C ('thawed FP24') <5 days at 1-6°C labelled as 'thawed plasma'	<24 hours 2-6°C	<24 hours 2-6°C	Use immediately
Clotting factor requirement for routine QM	None		>75% units >0.70 IU/ml FVIII	>75% units >0.50 IU/ml FVIII	> 0.70 IU/ml FVIII

Απόψυξη

Αποθήκευση μετά την απόψυξη

8 ώρες 4°C

4 ώρες σε θερμοκρασία δωματίου



- Οι μέθοδοι αδρανοποίησης ιών επηρεάζουν την ποιότητα του πλάσματος;
- Ναι
- Όχι

Αδρανοποίηση ιών

- η προσθήκη κυανού του μεθυλενίου
 - ή ψωραλένιων και έκθεση σε ορατό φως και UVA αντίστοιχα μονήρων μονάδων ή
 - η επεξεργασία δεξαμενών πλάσματος με απορρυπαντικό (solvent detergent).
- Ελαττωμένη δραστικότητα: Ινωδογόνου (24-39%),
 - FVIII (13-33%)
 - Ελαττωμένη δραστικότητα Παραγόντων I, V, VII, VIII, X (17-30%)
- Ελαττωμένη δραστικότητα πρωτεΐνης S 50% και παραγόντων V31% ,VIII28%

Maximum increases or decreases in coagulation parameters due to solvent/detergent or methylene blue/light treatment

Parameter	SDP maximum decrease/increase	MBP maximum decrease/increase
PT (s)	0.3	1.5
APTT (s)	2	8
Thrombin time (s)	1	3
Clottable fibrinogen (g/l)	0.2	1.0
Factor V (U/100 ml)	9	32
Factor VIII (U/100 ml)	20	33
Factor IX (U/100 ml)	3	23
Factor XI (U/100 ml)	17	27
Factor XIII (U/100 ml)	15	16
Von Willebrand Factor (VWF:Rco) (U/100 ml)	6	29
Antithrombin (U/100 ml)	9	2
Protein C (U/100 ml)	11	7
Protein S (U/100 ml)	51	0
C1-esterase inhibitor (U/100 ml)	7	27
Plasminogen (U/100 ml)	7	1
Plasmin inhibitor (U/100 ml)	76	5
Antitrypsin (U/100 ml)	25	no data
VWF-cleaving protease (U/100 ml)	0	no data
Markers of activated hemostasis: F1 + 2, TAT, D-dimers	not increased	not increased

SDP = solvent/detergent-treated plasma; MBP = methylene blue/light-treated plasma; plasminogen and all inhibitors of blood coagulation were measured by activity assays.

- [Transfusion](#). 2008 Jan;48(1):108-17. Epub 2007 Sep 27.
- **Coagulation function in fresh-frozen plasma prepared with two photochemical treatment methods: methylene blue and amotosalen.**
- [Osselaer JC](#)¹, [Debry C](#), [Goffaux M](#), [Pineau J](#), [Calomme G](#), [Dubuc E](#), [Chatelain B](#), [Vandendaele MC](#), [Hsu J](#), [Rheinschmidt M](#), [Lin L](#).
- **Author information**
- **Abstract**
- **BACKGROUND:**
- Pathogen inactivation of plasma intended for transfusion is now the standard of care in Belgium. Two methods for treatment of single plasma units are available: amotosalen plus ultraviolet A light and methylene blue plus visible light. This study compared the quality and stability of plasma treated with these two methods.
- **CONCLUSION:**
- There is adequate preservation of therapeutic coagulation factor activities in both PCT-FFP and MB-FFP.
- The overall coagulation factor levels and stability of PCT-FFP were better preserved than MB-FFP.

- [Transfusion](#). 2014 Aug;54(8):1935-44. doi: 10.1111/trf.12602. Epub 2014 Mar 18.
- **Microparticle profile and procoagulant activity of fresh-frozen plasma is affected by whole blood leukoreduction rather than 24-hour room temperature hold.**
- [Chan KS1](#), [Sparrow RL](#).
- [Author information](#)
- **Abstract**
- **BACKGROUND:**
- Microparticles (MPs) are small phospholipid-containing vesicles that have procoagulant properties. MPs are thought to contribute to the hemostatic potential of plasma. This study investigated the effects of whole blood (WB) hold time and leukoreduction (LR) on the MP profile and hemostatic potential of fresh-frozen plasma (FFP).
- **STUDY DESIGN AND METHODS:**
- WB units (n=12) from healthy donors were divided into two pairs and each pair was held at 20 to 24°C for 6 or 24 hours. At the designated hold time, 1 unit from the pair was LR while the other unit was not LR. FFP was prepared by standard procedures, aliquoted, and frozen. The MP content was determined by flow cytometry using an absolute count assay and specific labels for red blood cells (CD235a), platelets (CD41), and phosphatidylserine (PS). The hemostatic potential was determined by thrombelastography (TEG) and coagulation factor assays.
- **RESULTS:**
- Compared to non-LR-FFP, LR-FFP had significantly lower numbers of MPs, particularly CD41+ MPs and PS-positive MPs (p<0.03). LR-FFP, compared to non-LR-FFP, had a slower clot formation time (p=0.002); lower clot strength (p<0.001); and lower Factor (F)VIII, FXII, and fibrinogen levels (p<0.01). With longer WB hold time, the TEG profile was unchanged, although FVIII levels were decreased as expected (p<0.01). On average FFP units met quality requirements.
- **CONCLUSION:**
- LR of WB resulted in lower hemostatic potential of FFP in conjunction with depletion of MPs and coagulation factors. Longer WB hold time did not significantly affect the hemostatic potential of FFP as measured by TEG. Acceptable hemostatic quality was maintained for all FFP processing conditions studied.

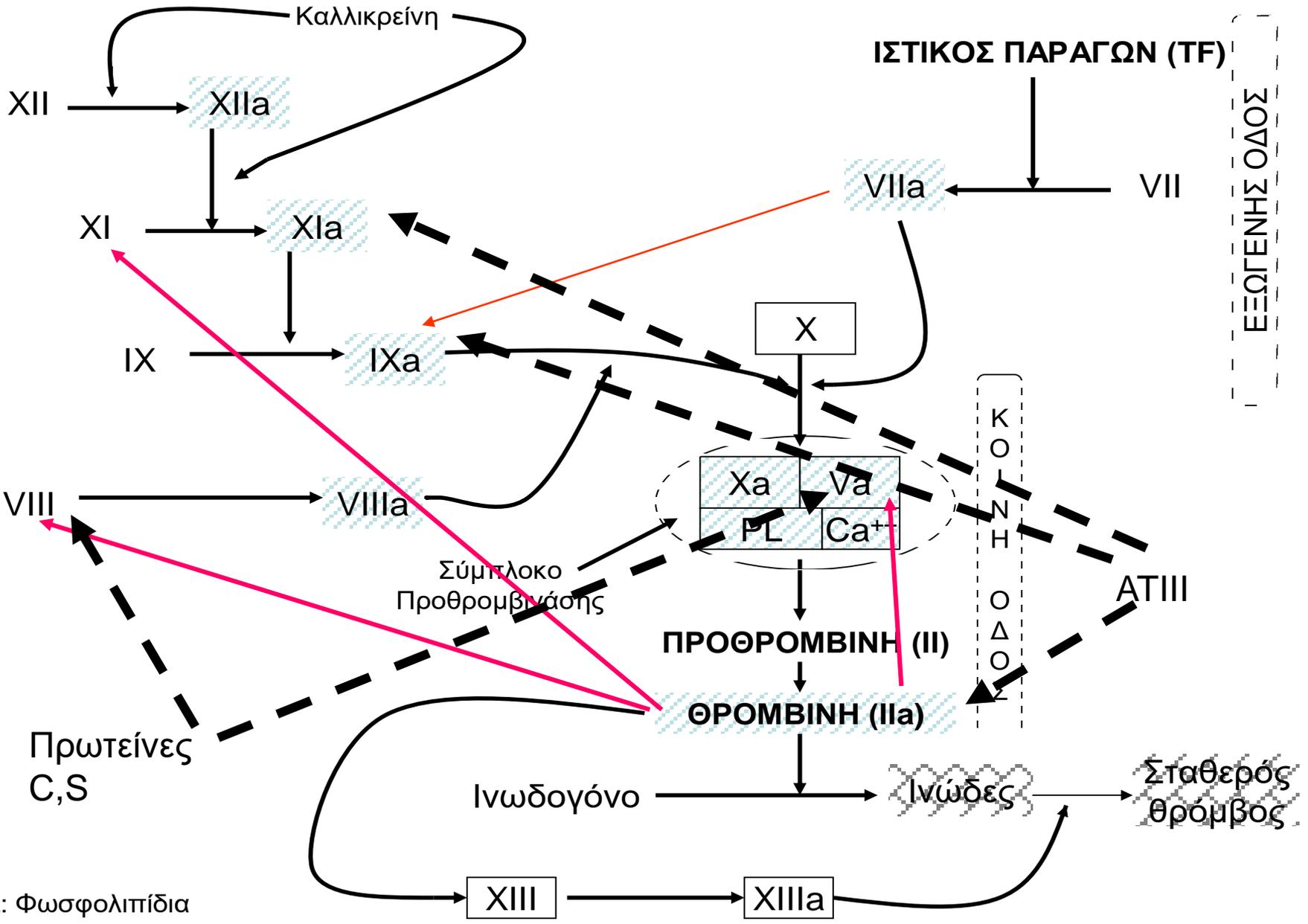
- Μικροκυστίδια
- Θρομβοελαστογραφία

ΕΝΔΟΓΕΝΗΣ ΟΔΟΣ

ΕΞΩΓΕΝΗΣ ΟΔΟΣ

TFPI

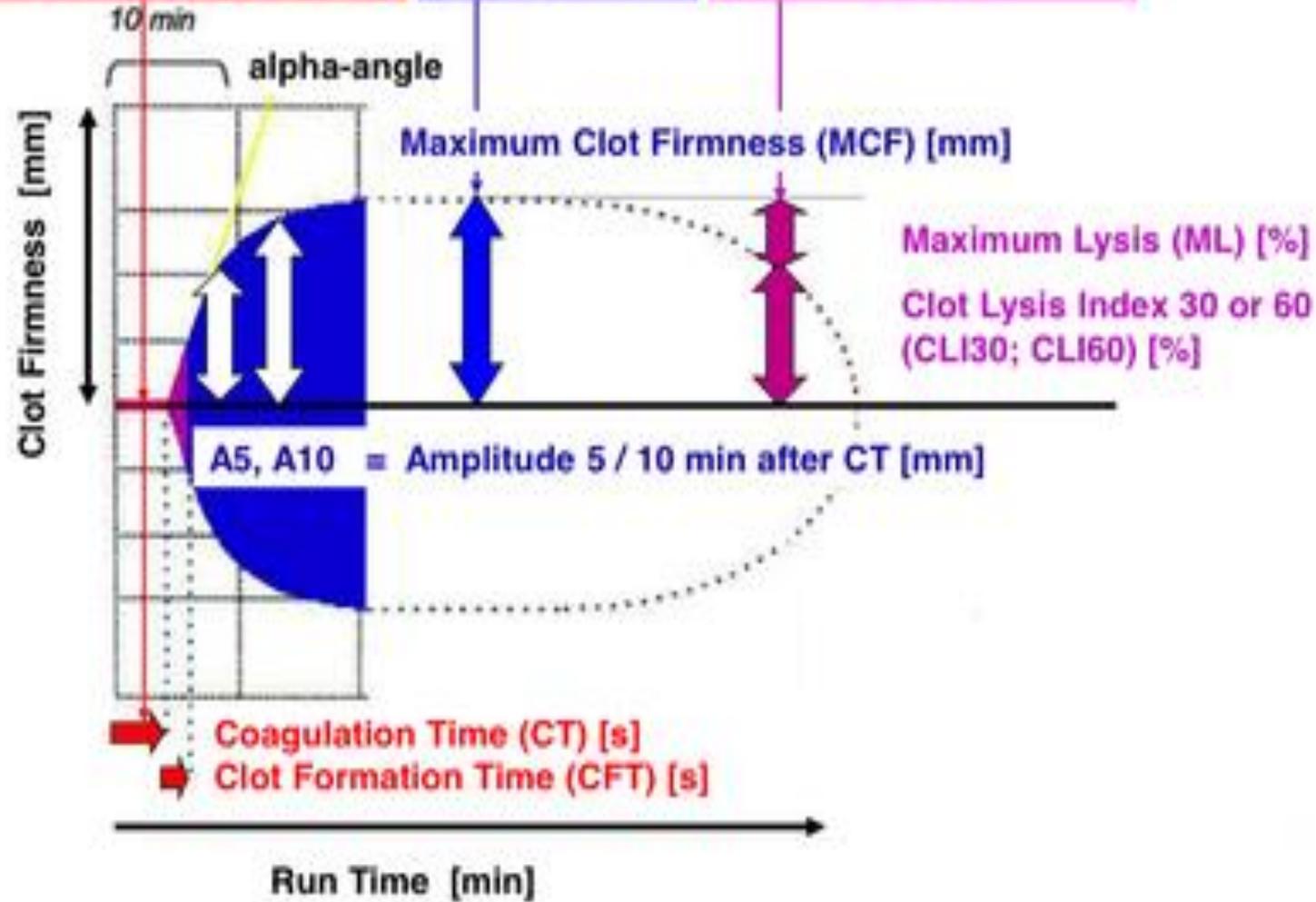
ΙΣΤΙΚΟΣ ΠΑΡΑΓΩΝ (TF)



Coagulation factors, anticoagulants, FDPs, tissue factor expression

Platelets, fibrinogen, F XIII, colloids

Fibrinolytic enzymes, fibrinolysis inhibitors, F XIII



- [Vox Sang.](#) 2016 Jul;111(1):33-42. doi: 10.1111/vox.12395. Epub 2016 Feb 29.
- **Increased coagulation and fibrinolytic potential of solvent-detergent plasma: a comparative study between Omniplasma and fresh frozen plasma.**
- [van Beers JJ1](#), [van Egmond LT1](#), [Wetzels RJ1](#), [Verhezen PW1](#), [Beckers EA2](#), [van Oerle R1,3](#), [Spronk HM3](#), [Straat RJ1](#), [Henskens YM1](#).
- [Author information](#)
- **Abstract**
- **BACKGROUND AND OBJECTIVES:**
- In this study, differences in levels of proteins involved in coagulation and fibrinolysis were compared between fresh frozen (quarantine plasma) and Omniplasma. Furthermore, thawing conditions and plasma stability after thawing were studied.
- **MATERIALS AND METHODS:**
- 10 Omniplasma and 10 quarantine plasma units were used to study different procoagulation, anticoagulation and fibrinolytic parameters. Analysis took place at different time-points during plasma storage at 2-6°C.
- **RESULTS:**
- At baseline, significant reduced levels of factor V, free protein S, α 2-antiplasmin and tPA-induced ROTEM lysis time were observed in Omniplasma as compared to quarantine plasma. Moreover, thrombin generation, IXa-AT complex levels and factor XIa were significantly increased in Omniplasma. The majority of the parameters studied remained stable in Omniplasma 48 h after thawing, with the exception of factor VIII (decrease) and IXa-AT (increase).
- **CONCLUSION:**
- Our results suggest an increased coagulation potential, presumably as a result of contact activation during the production process and also, an increased fibrinolytic potential in Omniplasma. The stability of Omniplasma, based upon the different parameters studied, is comparable to Q-plasma. A maximum post-thawing time of 48 h for Omniplasma can be suggested.

