Validation of Plasma and Platelet Pathogen Inactivation

Requirements from the UK Standing Advisory Committee on Blood Components/Standing Advisory Committee on Transfusion Transmitted Infection

Background

A number of systems for pathogen inactivation (PI) of plasma and platelet components are CE marked or in development. This document describes data that should be generated and reviewed as part of validation of such systems in the UK, should UKBTS take a strategic decision to procure them.

Consideration of whether UKBTS would implement any such technologies would be made using a framework such as the ABO Risk Based Decision Making Framework. The impact of such technology on the potential potency of the component would be balanced against other factors including the benefits PI may afford. It is therefore difficult to recommend acceptable limits for a reduction in potency in isolation of these considerations regarding benefit, which may differ depending on why PI is being implemented. Therefore the impact of PI on the final component must be considered as part of a wider framework in assessing PI, especially where this may affect cost-effectiveness or customer acceptance of the final component. Stakeholder engagement is essential.

This document is applicable to PI systems applied to units of plasma and platelets collected and treated by licensed Blood Establishments under the Blood Safety & Quality Regulations. It does not cover PI systems where the resulting component is regarded as a licensed medicinal as these are considered under a different regulatory pathway.

This document describes data that must be generated and reviewed as part of the validation process. Data may be provided by manufacturers of PI systems and/or generated by UKBTS. The division of these responsibilities will be defined as part of specifications for procurement of PI systems by the relevant UKBTS.

Minimum requirements for reduction in viable infectious agent

1. Background

This document identifies data that should be reviewed and/or generated and then assessed to ensure that the effectiveness of the system(s) in terms of inactivation of transmissible pathogens is understood. This document also assumes that the current molecular and serological screening procedures remain in place and that PI is an additional risk reduction process.

2. PI systems and their use

There are currently four systems available, two of which can be used for plasma and platelets [amotosalen and UVA (Intercept), riboflavin and broad spectrum UV (Mirasol)], one for platelets only [UVC light and agitation (Theraflex UV)] and one for plasma only [methylene blue and visible light (Theraflex MB - plasma)].

The primary use of PI is to minimise risk of transmission of any pathogen(s) in the products, and in the UK this risk is considered to relate primarily to bacterial contamination of platelet components rather than the presence of infectious agents not detected by the routine screening programme in place. Bacterial contamination is most likely in platelet components as they are produced and stored at temperatures from 20-24°C; lower temperature storage does not take place and at 20-24°C many species of bacteria that are likely contaminants are able to survive and grow.

With the current routine screening of donations, risk of transmission of the blood borne infectious agents for which screening is mandated is very low. Nonetheless, there remains a residual risk of transmission of infection from a donation with a very low level of infectious agent, undetectable on screening - the window period. In the majority of cases the window period reflects an early infection, or a resolving active infection in the case of hepatitis B virus. It is also possible that a low level 'infectious' donation may arise from infection with a blood-borne infectious agent which has sufficient genetic variation to escape detection by the screening algorithm and/or assays in use, while remaining potentially transmissible.

In addition to the infectious agents for which screening is performed routinely, PI may also have value in reducing or eliminating other blood-borne infectious agents which may be present in the products and could cause morbidity in recipients (e.g. HHV-8, HAV). There are infectious agents which are ubiquitous and therefore present in donated products, transmissible, but which have no known associated pathology (e.g. Torque teno virus, GB virus C). Whilst such infectious agents do not appear to have any adverse effect, their removal from products has to be considered to be beneficial.

PI would be applied to plasma and platelet components after their preparation, prior to being released to inventory. Ideally PI would be applied as soon as possible after preparation; if PI is delayed e.g. after freezing/thawing UK Blood Services may want to consider any increased risk of bacterial contamination.

3. Effectiveness

In the presence of effective donor selection and an effective and comprehensive donation screening programme, PI serves to either reduce further any residual risk remaining after screening, or to reduce the level of any other infectious agents present but for which screening is not performed thereby allowing relaxation of deferral times. The main target in the case of PI of platelet components is bacteria which may multiply during storage and lead to serious adverse consequences in the recipient. However, other infectious agents which may be present in plasma or platelet components, and for which screening is not performed, would also be removed/inactivated by PI.

Residual viral risk

The residual risk estimations used by the UK Blood Services provide an estimate of the risk of an infectious donation, containing a blood borne infectious agent for which screening is performed (HCV, HIV, HBV), entering the blood supply despite the donor selection and donation screening processes in place[1]. This residual risk is primarily due to donations collected from donors in the 'window period' of infection, in whom infectious agent is present at a very low level, below that detectable in the sample tested, but when taking into account the overall volume of a particular component there may be sufficient infectious agent present to exceed the minimum infectious dose in a recipient.

Residual bacterial risk

Residual risk estimates for bacterial infections are not routinely prepared by UK Blood Services. In the last 20 years, as a result of safety measures implemented in donation and manufacturing processes, the risk of bacterial contamination has significantly reduced. All UK Blood Services also currently employ culture systems for bacterial screening (BacT/ALERT) which have further reduced, but not eliminated, the risk of bacterial contamination of blood components. Organisms, undetected by screening, have been reported by quality control outdate testing, 'near misses' and post-transfusion septic reactions. Between 2011 and 2015, NHSBT screened >1.2 million platelet concentrates and noted false negative cultures (all *Staphlococcus aureus*) on four occasions, one of which resulted in patient morbidity[2]. Outdate testing in the Canadian and Welsh Blood Services reported false negative rates in the region of 1 per 1,000 platelet concentrates[3,4], and estimated rates of non-fatal septic reactions and fatal reactions of 1 per 100,000 and 1 per 500,000 concentrates respectively.

Quantitated level of inactivation

The effectiveness of any PI procedure is measured by the reduction in the amount of viable infectious agent remaining after PI has been applied. The manufacturers of the available systems provide data from their own evaluation and validation work. In general the UK Blood Services are not in a position to undertake this work themselves and would use the data provided for viruses however specific work could be performed if necessary. For bacteria, however, the UK Blood Services have the capacity to undertake such inactivation studies and these should be performed to demonstrate the validity of the manufacturer's data. Specimen organisms should be used and seeded into plasma and platelet components at levels likely to be present in bacterially 'contaminated' components.

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[1] The estimated residual risk that a donation made in the infectious window period is not detected on testing: risks specific for HBV, HCV and HIV in the UK. Refer to JPAC website for up to date version.

[2] McDonald *et al*, (2017) Bacterial screening of platelet components by National Health Service Blood and Transplant, an effective risk reduction measure. Transfusion; 57; 1122-1131

[3] Ramirez-Arcos *et al*, (2017) Residual risk of bacterial contamination of platlets: six years of experience with sterility testing. Transfusion; 57; 2174-2181

[4] Pearce *et al*, (2011) Screening of platelets for bacterial contamination at the Welsh Blood Service. Transfus Med; 21 ; 25-32

Required effectiveness

Key to determining the usefulness of any PI system is the definition of the required reduction in levels of infectious agents. This is very much determined by the residual risk associated with the components. For viruses, in the presence of an effective serological and molecular screening programme the role of PI in relation to those infectious agents for which screening is performed is solely to reduce the residual risk to as close to zero as possible. If the screening programme in place leaves a very small residual risk, PI is only required to be able to deal with possibly 1-2 $\log_{10}$ IU/ml of infectious agent, as screening would have identified donations with a high viral load. In relation to the presence of bacteria, because it is likely that PI would replace, rather than be used in addition to, bacterial screening, PI would need to be sufficiently effective to inactivate any bacteria present in the components. Bacteria in the range of 1–100 cfu per unit are the most realistic titres to use when modelling contamination of units of whole blood and the majority of the available data suggest that any bacteria present would be at low level, and unlikely to exceed 20 cfu/ml in an actual donation. For PI to be effective it must be able to prevent growth of these low levels of contaminating bacteria, which may be present soon after donation. The inability of PI systems to perform adequately in this manner might lead to subsequent growth of bacteria during storage with higher bacterial titres developing. Because growth of bacteria may lead to the formation of pyrogenic agents and endotoxin, inactivation of products at time intervals considerably after collection would likely be ineffective in preventing pyrogen- or endotoxin-mediated clinical reactions. As a result, PI methods are usually carried out at short intervals after collection (usually within 24 hours). If PI is not applied close to the time of collection and is applied after a period during which bacteria will have multiplied, the level of bacteria present may be significantly higher: up to 10^5 cfu/ml (depending on organism and sampling time-point). It is expected that PI systems would be suitable for use on products on components soon after collection (within 24 hours) and manufacturers should define a maximum limit for the system (note: current processing practice of time limit from vein to bacterial screening is 36 hours). With respect to different products (e.g. pooled or apheresis platelets) or different storage mediums (e.g. PAS), specific evaluation studies would be required to ensure the PI system achieves the minimum 10^4 cfu/ml inactivation with the product/medium for which the PI use is intended.

4. Pathogens targeted

When assessing the suitability of any PI system, the effectiveness of the system against the following categories of infectious agents should be considered. There needs to be evidence of the effectiveness of PI to inactivate infectious agents present at a level which could lead to transmission from any blood components.

Bacteria

Inactivation must be demonstrated for a range of bacteria which are representative of those likely to be present in a donation and which may consequently be present in a platelet component prepared from the donation. Table 1 lists the key bacteria against which PI must demonstrate effective inactivation. Where WHO bacteria reference strains are available they should be used. Although it is considered that the level of bacterial contamination in the original donation which may result in clinically significant levels of bacteria in stored platelet components is no more than 20 cfu/ml, a higher minimum proven level of inactivation must be demonstrated (PI must reduce any bacterial contamination by the amount specified i.e. 10^4) to ensure maximum effectiveness.

Table 1: Required minimum inactivation of potential contaminating bacteria*

Target	Gram +/-	Required minimum log_{10} cfu/ml inactivation by PI
B. cereus	+	10^4
S. aureus	+	10^4
S. epidermidis	+	10^4
S. pyogenes	+	10^4
E. coli	-	10^4
K. pneumoniae	-	10^4
S. marcesens	-	10^4
P. aeruginosa	-	10^4

*with the view the product would be sterile after treatment

Blood Borne Viruses

The viruses that may contaminate blood and blood products encompass many viral types, including viruses with a DNA or RNA genome, with and without a lipid membrane, and ranging in size from small e.g. parvovirus, to larger ones, such as HBV. In addition the pathogenicity of a virus may depend on the patient group and on the product being administered. PI systems should therefore be shown to be able to remove or inactivate a wide range of viruses if they are to be considered satisfactory; typically, validation studies have involved at least three viruses, chosen to represent different kinds of agent. A robust, effective and reliable process will be able to remove or inactivate substantial amounts of virus, typically 4 logs or more.

Blood Borne Viruses for which screening is performed

Inactivation must be demonstrated for HBV, HCV, HIV and WNV. Table 2 provides the minimum level in terms of degree of inactivation (log_{10} kill) required[5]. The minimum required level of effectiveness is dependent upon the residual risk resulting from the screening strategy adopted and has been calculated by taking the lowest claimed sensitivity (95% LoD) of the molecular screening system currently in use within the UK Blood Services for the screening of blood donations for HBV, HCV, HIV and WNV – (Roche 6800/8800 system and assays), and multiplying by the current screening pool size. This figure identifies the actual sensitivities (95% LoD) of the molecular screening assays for these viruses used within the current screening strategy adopted by the UK Blood Services. This therefore represents the level of each virus below which screening would not reliably detect any viral nucleic acid present in a donation, and which would be the actual target of PI applied to donations. The screening programme in place should detect higher levels of viral nucleic acid present in a donation.

In addition to these 4 viruses, molecular screening is also performed for HEV, although the current PI methodologies are not effective against HEV. Using the same calculation, but here using the less sensitive of the 2 commercial screening assays in

[5] minimum level of kill to ensure complete inactivation of currently screened for viruses with current screening methods . Operationally, higher kills (typically >4 log_{10}) for a wider range of viruses (screened and unscreened) would be expected.

use (Roche), the minimum required inactivation level of PI, if it were to be effective, can be determined.

There are infectious agents for which donations are screened routinely, but which are either not inactivated by current PI methodologies (HEV, as above), or where the presence of free infectious agent in the plasma is rare (HTLV, syphilis). In the case of HTLV leucodepletion is considered to be effective at minimising risk of transmission.

Studies that assess viral clearance are required for all PI products. These studies should select viruses that resemble those that may be present in the starting material. For example, the models for HCV include BVDV, Sindbis virus, Semliki forest virus and yellow fever virus as they share many properties, including a lipid membrane, RNA genome and are of a similar size. These studies can only approximate the inactivation and removal that occur during routine manufacture because the model viruses employed in the studies may differ from those present in blood. Therefore the manufacturer should justify the appropriateness of the studies, the choice of viruses and the validation conditions employed. Guidance is available for the selection and assay of model viruses[6]

Other Blood Borne Viruses

Although inactivation of CMV would be beneficial to reduce risk of transmission from viraemic donations, such donations are considered to be rare. CMV has a short viraemic period and is strongly cell-associated and whilst not considered a significant transmission risk in plasma products, the largest risk for CMV transmission is from cell-associated virus (i.e. in products containing cellular components). Leucodepletion and serology screening currently provide an effective approach to minimising CMV risk. To be an effective CMV risk reduction measure (i.e. to replace current serology screening) PI systems would need to be able to inactivate cell-associated virus. Any available data for CMV should be obtained from the PI system manufacturers and assessed.

Inactivation of Dengue, Chikungunya, Zika (and other tropical arthropod-borne viruses) would be attractive, but the importance and any value are dependent upon numbers of imported cases in the UK, which are currently low. If locally acquired cases occur, or there is a large increase in imported cases, in sufficient numbers to present a real risk of an infected individual donating whilst viraemic, PI could be beneficial in reducing risk. Any available data for other blood borne viruses should be obtained from the PI system manufacturers and assessed. Should the driver for PI implementation be mitigation of a specific or unknown emerging pathogen, a log kill sufficient to inactivate the agent beyond its infectivity titre would be required. There are other blood-borne viruses, including parvovirus B19 and hepatitis A virus, which are known to be transmitted but which currently are not screened for in the UK. Donors are deferred if they declare current or recent infection with such viruses. Information about whether any of the PI technologies are effective against such infectious agents, and the level of inactivation obtained, should be obtained from the manufacturer.

[6] CPMP/BWP/268/95 (revised): Note for guidance on viral validation studies: The design, contribution and interpretation of studies validating the inactivation and removal of viruses. Adopted 1996. London, Committee for Proprietary Medicinal Products (Available on the Internet at http://www.emea.eu.int/pdfs/human/bwp/026895en.pdf).

Other infectious agents

Inactivation of other infectious agents may be beneficial, but any benefit is dependent upon the risk of the agent(s), being present in the donor population and consequently in the blood supply. Although inactivation of parasites such as *T.cruzi* and *Plasmodium sp* would be beneficial, the number of donors confirmed as *T.cruzi* infected is extremely low and with effective donor selection and screening risk is already minimal. There are now data demonstrating PI of malaria, but this would only be of relevance if PI of whole blood were to be considered.

Table 2: Required minimum log_{10} inactivation of BBVs

Target	**Screening molecular assay in use**	**Sensitivity of current molecular screening assay (ID screening, manufacturer's 95% LoD)**	**Pool size used for screening**	**Pooled screen sensitivity (calculated from manufacturer's 95% LoD)**	**Required minimum log_{10} inactivation by PI (IU/ml)[1]**
HBV	Roche cobas®	1.4 IU/ml	24	33.6 IU/ml	2.53
HCV	Roche cobas®	7 IU/ml	24	168 IU/ml	3.23
HIV-1	Roche cobas®	25.7 IU/ml	24	616.8 IU/ml	3.8
HIV-2	Roche cobas®	4 IU/ml	24	96 IU/ml	2.98
HEV[2]	Roche cobas® / Grifols Panther	18.6 IU/ml	24	446.4 IU/ml	3.65
WNV L1[3]	Roche cobas®	10.8 cp/ml	6	64.8 cp/ml	2.81
WNV L2[3]	Roche cobas®	4.8 cp/ml	6	28.8 cp/ml	2 .46

[1] Based upon the manufacturer's stated 95% limit of detection (IU/ml) of the screening assays used together with additional 1 log_{10} to take into account any variability affecting the level of inactivation achieved

[2] HEV is not inactivated by any of the current PI technologies

[3] WNV RNA screening is only applied to donations from donors with specific risk exposure

Validation of Component Quality

Plasma

It is anticipated that the system would be used to produce pathogen-reduced fresh-frozen plasma (PR-FFP) and cryoprecipitate from PR-FFP. Currently UKBTS do not produce cryoprecipitate-depleted plasma (CDP) and this is not a requirement for validation currently – see later section on clinical studies.

Basic information about the system

Manufacturers must provide the following to accompany the data set provided:

- Instructions for use
- Active ingredient (if any) and removal process (if any)
- Mechanism of action
- Regulatory class of device (93/42/EEC)
- Evidence that the inactivation system including accessory equipment and software is CE marked under the Medical Devices Directive (93/42/EEC) or the Medical Devices Regulation (2017/745) whichever is appropriate, including toxicology review, and that the mutual compatibility of the devices has been validated.
- Claims made under CE mark in relation to inactivation of pathogens and leucocytes
- The minimum and maximum volume of plasma that can be treated (since the amount of photo-sensitizer added is usually fixed, this will determine its final concentration in plasma and may affect pathogen inactivation and component quality)
- Any other component criteria critical to successful treatment e.g. cellular contamination, lipaemia.

Laboratory Studies

Phase 0

Control data from plasma that has not been treated should be included. The minimum number of units tested should be 16. A paired or pooled and split study design may reduce the number of units required, and advice from SACBC should be sought in this regard.

The number of donations in the final component must not exceed 12.

The following details should be included as part of this process:

- Plasma produced from male donors
- The length of time that plasma that has been held at 22^{o}C (either as whole blood or plasma) prior to PI treatment (data up to 24 hours will be acceptable, ideally units will have been held as whole blood for 18-24 hours.
- The length of time between PI treatment and freezing the final plasma (plasma is expected to be frozen within a maximum of 27 hours of collection).

- Freeze-thawing prior to treatment and then re-freezing after treatment, in addition to the timelines in the bullet point above.
- The ABO group of units of plasma used, and for data on FVIII and vWF data should be separated for blood group O and non-O donations.

The following variables must be stated in the study report/data provided

- Whether the plasma has been collected via apheresis or whole blood donation. For plasma produced from whole blood this must be collected into CPD anticoagulant.
- Whether the plasma was leucocyte-depleted prior to treatment
- The mix of ABO groups used
- The volume of plasma treated

Data should be provided at the point of manufacture i.e. prior to freezing or shortly after being frozen, and also after freeze-thawing to provide assurance regarding the stability of plasma in the frozen state and following thawing

a) at point of manufacture

Manufacturers of PI systems are expected to provide a data set as outlined in Table 3, that provides a general indication of the effect of the PI system on plasma.

b) following frozen storage and subsequent thawing

Data should be provided by the manufacturer to support the stability of PR plasma. The same variables as above should be considered in the choice of plasma for validation to assess:

- The stability of PR-plasma when stored at a stated temperature between -20 ^{o}C and -40 ^{o}C for 36 months
- The stability of PR plasma once thawed and stored at 4 ±2 ^{o}C, this must be for a minimum of 24 hours, but preferably longer.

The parameters for which data must be provided are as follows:

- During storage once frozen for 36 months: FVIII, fibrinogen
- Stability once thawed and stored for a minimum of 24 hours: fibrinogen, FVIII, PS (free antigen and activity tested on the same samples), FV, FVII, thrombin generation tests.

These data are required to demonstrate the effect of the PI system on plasma. It is anticipated that some of the variables listed above may differ in the data set provided and how UKBTS may use these technologies. It is therefore expected that each UKBTS would perform its own limited validation to ensure that the system as applied by them produces satisfactory results.

It is acknowledged that it is difficult to define the content of plasma required for it to be clinically effective. This is in part because it is not known what levels of clotting factors must be present in plasma for it to be effective, and partly because a reduction in some but not other factors in combination could be more or less of concern depending on the clinical scenario in which plasma is being transfused.

The basis for the minimum acceptable values in Table 3 is therefore a concept of ‘no worse than current’, which includes components such as extended thawed plasma

and PR-FFP. Due to the wide variation in values for coagulation factors in normal plasma, for single-unit PR-plasma the specified values are given as a maximum change from pre-treatment values, the minimum mean value and the value that greater than 90% of units are expected to satisfy. The data on PR-plasma are required to satisfy the criteria in Table 3 on this basis.

However, the acceptability of the loss of component quality would need to be considered as part of the overall framework decision in relation to implementation of pathogen inactivation, and balanced against the benefits PI may bring in the specific context in which it is being considered. Therefore the data will be considered as a whole, and an opinion from SACBC and other stakeholders will be taken into account in the decision as to whether this is considered acceptable as part of the UKBTS procurement process.

Table 3: Parameters to be validated and expected minimum values based on a concept of no worse than current

		Should meet the specified values below			
		Required?	Mean loss due to treatment process (%; pre v post or control v test)	Mean in final component	90% of units should be above
Basic	Volume	Y	NA	To meet specification	
Coagulation screening tests	PT ratio	Y	NA	NA	NA
	APTT ratio	Y	NA	NA	NA
Global tests	Thrombin Generation (1 or 5pM TF)	Y	NA	NA	NA
	ROTEM/ROTEG	D	NA	NA	NA
Coag. factors	Fibrinogen (Clauss)	Y	≤40	≥1.70g/l	1.50g/l
	Fibrinogen antigen	Y	<5%	≥ 2.50g/l	2.00g/l
	Factor II	Y	≤20	≥0.8 U/ml	0.70 U/ml
	V	Y	≤20	≥0.7 U/ml*	0.60 U/ml
	VII	Y	≤20	≥0.8 U/ml	0.60U/ml
	VIII**	Y	≤30	≥0.5 U/ml*	0.50 IU/ml
	IX	Y	≤20	≥0.8 U/ml	0.70 U/ml
	X	Y	≤20	≥0.8 U/ml	0.70 U/ml
	XI	Y	≤40	≥0.6 U/ml	0.60 U/ml
	XII	Y	≤20	≥0.8 U/ml	0.60 U/ml
	XIII	Y	≤20	≥0.8 U/ml	0.70 U/ml
vWF	Ag	Y	≤20	≥0.8 U/ml	0.70 U/ml
	RiCof/CBA	Y	≤20	≥0.8 U/ml	0.70 U/ml
	Multimers	D	NA	NA	NA
	Cleaving protease	Y	≤20	≥0.8 U/ml	0.70 U/ml
Inhibitors	AT III	Y	≤20	≥0.8 U/ml	0.70 U/ml
	Prot C	Y	≤20	≥0.8 U/ml	0.70 U/ml
	Prot S free antigen & activity	Y	≤20	≥0.8 U/ml	0.60 U/ml
	Alpha-2 antiplasmin	Y	≤20	≥0.8 U/ml	0.70 U/ml
Activation	TAT/Frag1.2/FPA	Y	NA	NA	NA
	FXIIa/S2302	Y	NA	NA	NA
	C1 Inhibitor	Y	≤20	≥0.8 U/ml	0.70 U/ml
Residual photosensitiser (if added)		Y	NA	NA	NA

All assays are expected to be functional (i.e clotting or chromogenic assay), unless otherwise indicated. * allows for loss of each prior to treatment due to whole blood or plasma storage. ** based on equal mix of group O and A donations, value may need to be adjusted for other group mix.

Y – Test / data is required, N – Test / data is not required, D – Test / data is desirable, NA not applicable.For some tests we have not specified values, since there are not relevant international standards to permit comparison of data across laboratories in a meaningful way.

Phase 1 and 2 studies

Routine quality monitoring for pathogen reduced plasma will focus on those factors that are usually the most affected by PI systems i.e fibrinogen and FVIII, as well as standard monitoring of cellular content and volume.

A minimum of 125 units are expected to be produced and tested in Phase 1, and 2000 units produced in phase 2, with 1% of units tested or a proportion determined by statistical process control.

In phase 1 and 2 studies the following requirements (see Table 4) shall be met in the final component as produced for transfusion.

Table 4: PR-FFP phase 1 and 2 study component requirements

			PR-Fresh frozen plasma				
Parameter		*Unit*	*Current Specification (for PR-plasma, 2018)*				
			Minimum	*Maximum*	*Required min %*		*Required min %*
					pass rate in routine use		*pass rate in validation*
Volume		mL	200	300	75		95
Platelets		$x10^9$/L	N/A	30	75		75
Red cells		$x10^9$/L	N/A	6*			95
FVIII		IU/mL	0.5	N/A	75		75
WBC		$x10^6$/unit	N/A	1	90		90
Fibrinogen		g/l	1.50 g/l		NA		90
Fibrinogen		g/l	above 60% of starting material on average		NA		90
Visual inspection							100%

*or as recommended by manufacturer of PI system

Cryoprecipitate

Manufacturers of PI systems are expected to provide data to demonstrate that plasma treated using their system can be used as a starting material to produce cryoprecipitate (i.e the criteria for PRplasma listed in Table 3 are satisfied). In addition, the final component shall meet the following criteria (Table 5). As for plasma, these are based on the concept of no worse than current. Processes recommended by manufacturers of PI systems for producing single or pooled cryoprecipitate that are non-standard for UK Blood services will be considered and may necessitate an amendment to the values listed in the tables below. This would be agreed with the manufacturer.

Table 5: PR-Cryo Phase 1 and 2 component requirements

5a Single cryoprecipitate

		PR-Single cryoprecipitate				
Parameter	*Unit*	*Current Specification(for PR-cryoprecipitate)*				
		Minimum	*Maximum*	*Required min %*		*Required min %*
				pass rate in routine use		*pass rate in validation*
Volume	mL	20	60	75		90
FVIII	IU/unit	50		75		75
WBC	x10^6/unit		1	90		90
Fibrinogen	mg/unit	140		75		90
Visual inspection 100%						100%

5b Pooled cryoprecipitate

		PR-Pooled cryoprecipitate				
Parameter	*Unit*	*Current Specification for PR-cryoprecipitate*				
		Minimum	*Maximum*	*Required min %*		*Required min %*
				pass rate in routine use		*pass rate in validation*
Volume	mL	100	300	75		75
FVIII	IU/unit	250		75		75
WBC	x10^6/unit		1	90		90
Fibrinogen	mg/unit	700		75		75
Visual inspection						100%

UKBTS will investigate the suitability of PR plasma for cryoprecipitate production during phase 1 and 2 studies.

Clinical data on plasma/cryoprecipitate

Clinical studies/observations should include human data on allergic and other infusion reactions, other safety considerations, and efficacy as below.

Clinical studies comparing PR plasma with untreated or an alternative PR methodology are desirable for indications such as thrombotic thombocytopenic purpura (TTP), single factor deficiencies (e.g. factor V deficiency, C1-esterase inhibitor deficiency) and any acquired coagulopathies.
However, it is recognised that in many settings large randomised clinical studies to define the optimal role of standard or untreated plasma are lacking. Peer-reviewed observational data and data from post-marketing surveillance including national haemovigilence reports must be available or in collection.

Plasma exchange for TTP

Currently in the UK the recommended plasma of choice for treatment of TTP is solvent-detergent treated plasma produced from blood donations from a country of low vCJD risk (BCSH 2012). If an alternative PR-plasma is to be considered instead of SDFFP for TTP then its efficacy would need to be demonstrated in clinical studies and the acceptability of these data reviewed by experts in the treatment of TTP (via BSCH/BSHT). There is evidence that methylene blue FFP, despite having normal levels of ADAMTS13 activity, is less effective in this setting[7] (de la Rubia *et al*, 2001; Rio-Garma *et al*, 2008). Therefore the efficacy of plasma for this indication is not predicted by the results of laboratory testing.

Information on post marketing surveillance data and /or on-going programmes should be provided. This should include national haemovigilence data where available.

The data from such studies will be used as part of the overall decision making framework.

Platelets

Basic information about the system

Manufacturers must provide the following to accompany the data set provided:

- Instructions for use
- Active ingredient (if any) and removal process (if any)
- information relating to type of platelet storage media
- Mechanism of action
- Regulatory class of device (93/42/EEC)
- Evidence that the inactivation system including accessory equipment and software is CE marked under the Medical Devices Directive (93/42/EEC), or the Medical Devices Regulation (2017/745), whichever is appropriate including toxicology review, and that the mutual compatibility of the devices has been validated.
- Claims made under CE mark in relation to inactivation of pathogens and leucocytes and the suitability of process as an alternative to bacterial screening, irradiation or CMV screening.
- The minimum and maximum volume and platelet content/concentration that can be treated
- Any other component criteria critical to successful treatment e.g cellular contamination, lipaemia.

[7] de la Rubia, J., Arriaga, F., Linares, D., Larrea, L., Carpio, N., Marty, M.L. & Sanz, M.A. (2001) Role of methylene blue-treated or fresh-frozen plasma in the response to plasma exchange in patients with thrombotic thrombocytopenic purpura. British Journal of Haematology, 114, 721– 723.

Rio-Garma, J., Alvarez-Larran, A., Martinez, C., Muncunill, J., Castella, D., la, R.J., Zamora, C., Corral, M., Viejo, A., Pena, F., Rodriguez-Vicente, P., Contreras, E., Arbona, C., Ramirez, C., Garcia-Erce, J.A.,Alegre, A., Mateo, J. & Pereira, A. (2008) Methylene blue-photoinactivated plasma versus quarantine fresh frozen plasma in thrombotic thrombocytopenic purpura: a multicentric, prospective cohort study. British Journal of Haematology, 143, 39– 45.

a) laboratory studies

Laboratory data on platelet function of pathogen reduced platelets in plasma, and in PAS-E / plasma mixture must be provided as these are the current standard in the UK if the system is designed for use with both media. The recommended storage media for use with the system must be specified by the manufacturer. Laboratory data for platelets in PASE must be provided unless the system is not recommended for use with PASE. Other PAS may be considered should data suggest a more optimal PAS in conjunction with the PI system. Clinical data using earlier generation PAS may be acceptable, provided there is laboratory data linking this to PASE.

Data must be provided for each of the following combinations for which a claim is made:

- Buffy coat derived pooled platelets in plasma (collected in CPD anticoagulant)
- Buffy coat derived platelets in plasma and PAS-E mix
- Apheresis platelets in plasma (collected in ACD-A anticoagulant)
- Apheresis platelets in plasma and PAS-E mix

- Note: PI systems should be suitable for treatment of platelet components collected on a range of apheresis platforms. A list of apheresis devices compatible with the system must be supplied along with supporting documentation for any combinations that are claimed by the manufacturer to be compatible (laboratory data and details of CE marking and compatibility of the medical devices) and details of any restrictions or specific requirements on suspension medium or shelf-life.

In each case, data must be obtained from platelets:

- treated at the latest permitted time point post-collection (or manufacture), and
- subjected to periods of interruption of agitation of up to 24 hours, with the maximum period permitted to maintain platelet quality defined.
- at the end of the claimed shelf life, and (if a claim is made)
- subjected to secondary processing if these components are to be manufactured using PI:
    - resuspension in 100% PAS, or
    - split into paediatric sized components, or
    - IUT

It is anticipated that data in relation to interruption of agitation, washing and use for IUT would be obtained by UKBTS as this is specific to each institution.

Data must be provided in accordance with Table 6.

Table 6: In vitro assessment of platelet quality

Variable	Day 0/1 Pre treatment	Day 0/1 Post treatment	Day 5	Day 7*	Day 8/9*
Volume (mL)	✓	✓			
Red cell count (x10^9/L)	✓	✓			
Platelet count (x10^9/L)	✓	✓			
Plasma:PAS ratio*	✓				
Minimum plasma volume (mL)	✓				
Leucocyte count	✓	✓			
Leucocyte subsets	D	D			
Morphology eg swirling	✓	✓	✓	✓	✓
Activation markers inc CD62P	√	✓	✓	✓	✓
Lysis eg LDH	D	D	D	D	D
Glucose/lactate	✓	✓	✓	✓	✓
pH	✓	✓	✓	✓	✓
ATP or HSR	✓	✓	✓	✓	✓
Function	D	D	D	D	D
Cytokines	D	D	D	D	D
Chemokines	D	D	D	D	D
Coagulation activation markers (eg FXIIa)	D	D	D	D	D
Microvesicles	D	D	D	D	D
Residual photosensitiser (if added)	✓				

*data should be supplied for is the day of claimed shelf life. It is desirable to also provide data 1 or 2 days beyond the claimed shelf-life and this would become essential if data provided on the day of shelf-life indicate that platelet quality is borderline acceptable at that point in storage. D=Desirable.

These data are required to demonstrate the effect of the PI system on platelet quality. It is anticipated that some of the variables listed above may differ in the data set provided and how UKBTS may use these technologies. It is therefore expected that each UKBTS would perform its own limited validation to ensure that the system as applied by them produces satisfactory results. Results from phase 1 operational studies should comply with the table below (Table 7).

Irradiation

If the system is a suitable alternative to irradiation for the prevention of TA-GvHD, then evidence of CE marking and data must be provided to support this claim. This should take the form of: Notified Body evaluations, publications from peer reviewed scientific journals and reports from other Blood Services or other organisations in the world that define and support the claimed efficacy and overall performance of the system. Additionally, data on PI of the component plus irradiation should be generated and reviewed, if both PI and irradiation are to be used in combination (this is considered unlikely and would be unnecessary if the data to substantiate the claim regarding use as an alternative to irradiation is accepted). In the framework for considering implementation of PI, up to date information should be included on the effect of standard irradiation on platelet quality since approximately 50% of the platelet supply in the UK is currently irradiated and it is anticipated that PI would replace this practice.

Table 7: PI treated platelets Phase 1 requirements

		Platelets			
Parameter	*Unit*				
		Minimum	*Maximum*	*Required min % pass rate in routine use*	*Required min % pass rate in validation*
Volume	mL	150	380	75	95
Platelets	$x10^9$/unit	240	-	75	80
WBC	$x10^6$/unit		1	90	90
pH at expiry		6.4		95	95
Plasma:PAS	Ratio	Must meet PI system specifications*			
Platelets	$x10^9$/L	Must meet PI system specifications*			
Visual abnormality					Max 0%

b) <u>animal models of haemorrhage</u>

Data may be provided in support of claims for efficacy but will not replace the need for data in human studies.

c) <u>recovery and survival studies in normal subjects</u>

Data must be provided from recovery and survival studies, in healthy volunteers, of pathogen reduced platelets compared to untreated platelets. Data must be provided at a minimum shelf-life of day 5.

Due to variation in values for recovery and survival between individual donors, it is preferable to compare platelets at the end of shelf-life with fresh platelets from the same individual according to BEST recommended procedures[8]. Values at end of shelf-life are expected to be >66% of fresh for recovery and >58% for survival whether PI treated or not. Alternatively, If treated platelets are compared to untreated platelets at end of shelf-life in an unpaired study then sufficient numbers of individuals must have been studied to detect a difference of 20%. The difference in recovery and survival between treated and untreated platelets at end of shelf-life should not exceed 30% based on current figures for CE marked PI systems.

As for plasma, any reduction in component quality must be taken into consideration in the decision making framework used to consider implementation of PI. This is particularly pertinent for measures such as platelet recovery or survival, which could alter the number of platelet components transfused per patient depending on the indication for transfusion.

d) <u>clinical studies</u>

Information should be provided relating to the product by publication in peer-reviewed journals, and/or preferably through the incorporation of the data into systematic reviews.

[8] AuBuchon JP, Herschel L, Roger J, Murphy S Preliminary validation of a new standard of efficacy for stored platelets. Transfusion. 2004 Jan;44(1):36-41.

These data may be provided from platelets in plasma or in PAS / plasma mixture but there must be laboratory data that clearly links the data between storage in the two media.

Randomised clinical studies comparing untreated platelets with PI treated platelets should include data on corrected count increments at 1 hour and 24 hours (or equivalent measure), repeat transfusion interval, total units transfused, adverse events and bleeding events grades 2-4 (WHO score). The interpretation of these studies should recognise the evolving evidence base on the role of platelet transfusions highlighted in trials in patients with haematology malignancies, neonates and intracerebral bleeding, where in some trials the findings demonstrate evidence of harm.

Some end of shelf life clinical data should also be provided, ideally from controlled clinical studies and include count increments and clinical bleeding scores.

Information on post-marketing surveillance data and /or on-going programme should be provided. This should include national haemovigilence data where available.

The data from such studies will be used as part of the decision making framework.

Dr Helen New
SAC on Blood Components
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