Validation of Dried Plasma Components

Requirements for the UK Standing Advisory Committee on Blood Components

Background

Currently Lyoplas is the only commercially available dried plasma product in the UK, although it is used 'off label' as it is not licenced. However, the worldwide demand for this product far outstrips its supply. An alternative lyophilised product, FLYP, is available from France, but it is also unlicensed. The US and UK military are pursuing this as a short-medium term alternative to Lyoplas. Systems that blood centres could use to produce UK dried plasma (in a bag form rather than glass bottles that Lyoplas and FlyP are contained in) are in development but also not licensed yet: these are a spray-drying system from Velico and a freeze-drying system from Terumo. NHSBT are assessing the feasibility of supplying the UK military with a UK derived and manufactured product. As the component would be prepared using a device within its intended use, and similar dried products were used historically in the UK as well as currently elsewhere, the degree of novelty as defined in the 'Trial-Provisional component specifications' document on the JPAC website is 'low'.

There is currently no agreed UK or European specification for a dried plasma component, although dried plasma has been produced in the UK in the past (1940's/50's). The purpose of this document is to set out the UK requirements to validate such a component and agree the data required to inform a product specification in the UK on the assumption that it is produced using a medical device and regarded as a blood component. Should UKBTS undertake to validate such a component, data will be submitted to SACBC/JPAC in future for a provisional/full component specification as appropriate.

The impact of such technology on the potential potency of the component would need to be balanced against the benefits dried plasma may bring. 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 a dried plasma component is being implemented. Therefore, the impact of drying on the final component must be considered as part of a wider framework in assessing the technology.

This document is applicable to drying systems applied to units of plasma collected and treated by licensed UK Blood Establishments under the Blood Safety & Quality Regulations. It does not cover systems where the resulting component is regarded as a licensed medicinal product as these are considered under a different regulatory pathway. The regulatory classification of systems in development is not yet known, it is anticipated these will be class II/ III medical devices but this will be subject to submission of further information to the MHRA to decide on their classification when the technology is further along in development and the design of the equipment and consumables frozen.

The data that should be generated and reviewed as part of validation of such systems in the UK, is summarised below. We have based this guidance on the previously agreed values by SACBC (JPAC 19-05) as part of validation process for pathogen inactivation of plasma and universal plasma, along with draft guidance from the FDA for dried plasma. Data may be provided by manufacturers of drying systems, other Blood Services and/or generated by UKBTS. The division of these responsibilities will be defined as part of specifications for validating and implementing drying systems by the relevant UKBTS.

Validation of Plasma Component Quality

Basic information about the system

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

- Principle of drying method including time/temperature/conditions of drying
- Instructions for use
- Excipients added prior to drying (if any) and removal process (if any)
- Regulatory class of device
- Evidence required for the drying system, including accessory equipment and software, to be CE marked and UK CA marked whichever is appropriate, and that the mutual compatibility of the devices has been validated.
- Whether the device is designed to be used on single unit plasma or a mini-pool of 12 or fewer donations.
- The minimum and maximum volume of plasma that can be treated
- Recommended storage temperature and shelf-life
- The type and volume of reconstitution fluid
- Any other component criteria critical to successful treatment e.g. cellular contamination, lipaemia, anticoagulant.
- Evidence of how sterility is maintained during treatment

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 as recommended in section 8.4.2.1 of UK Guidelines 'Evaluation of new fresh frozen plasma/cryoprecipitate components for transfusion'. 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:

- Whether plasma was produced from male or female donors
- The length of time that plasma has been held at 22oC (either as whole blood or plasma) prior to 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 for treatment and production of final dried plasma component
- Freeze-thaw cycles prior to treatment

- The ABO group of units of plasma used, and for data on FVIII and vWF to 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.
- The mix of ABO groups used, if applicable
- The volume of plasma prior to treatment
- The volume of plasma in final dried plasma component
- Whether the plasma is single unit or mini-pool of n donations.
- Whether plasma has been subject to a pathogen inactivation process prior to drying

Data should be provided at the point of manufacture i.e. immediately after dried plasma component has been made and following storage to maximal proposed shelf-life to provide assurance regarding the stability of plasma in the freeze-dried state, and once reconstituted.

a) at point of manufacture

Data outlined in Table 1a and b should be provided to provide a general indication of the effect of the drying system on plasma ideally by paired comparison of the same units of plasma as standard FFP/cryoprecipitate or dried FFP/cryoprecipitate.

In addition, if a mini-pooled product is being considered, viral risk of the finished product must be assessed, including consideration for the need for pathogen inactivation.

If the product is intended to be transfused as ABO universal rather than group specific, then data must be supplied to substantiate this claim and mitigate risk of haemolytic transfusion reactions.

Consideration should be given to performing a proteomic analysis to ascertain the impact of drying on plasma proteins and additionally the effect of plasma in maintaining endothelial integrity using appropriate models.

b) following storage and subsequent reconstitution

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

- The stability of dried-plasma when stored at the manufacturers recommended temperature for prolonged periods. It is anticipated that this will be at 4oC. If the product can be stored at ambient temperature, this must be assessed at temperatures likely to be encountered in routine use – for example this may exceed 25oC in austere environments. It is expected that the shelf life will be shorter at higher storage temperatures.
- The stability of dried-plasma once reconstituted and stored at 4 ±2 oC or other temperature, for the maximum recommended time cited by the manufacturer prior to administration.

The minimum parameters for which data must be provided are as follows (these should be supplemented with other factors shown to be most affected by the drying treatment):

- Following storage in the dried state for the maximal recommended shelf-life: FVIII, fibrinogen
- Once reconstituted for the maximal period of time recommended prior to administration: fibrinogen, FVIII, PS (free antigen tested on the same samples), FV, FVII, thrombin generation tests.

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

It is acknowledged that it is difficult to define the optimal quality 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 of concern depending on the clinical scenario in which plasma is being transfused.

The basis for the minimum acceptable values in Table 1 is therefore a concept of 'no worse than current', which includes components in use in the EU such as extended thawed plasma and pathogen inactivated plasma. Due to the wide variation in values for coagulation factors in normal plasma, for single-unit dried-plasma the specified values are given as a maximum change from pre-treatment values, the minimum mean value that must be achieved and the criteria that at least 90% of units are expected to satisfy. The data on dried plasma are required to satisfy the criteria in Table 1 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 dried plasma, and balanced against the benefits that dried plasma may bring in the specific context in which it is being considered. Therefore, the data will be considered and an opinion from SACBC and other stakeholders will be considered in the decision as to whether this is considered acceptable as part of the development process.

Phase 1 and 2 studies

Routine quality monitoring for dried plasma will focus on those factors that are usually most affected by manufacturing and drying systems as well as standard monitoring of cellular content and volume.

A minimum of 125 units (according to routine UK Blood Services' practice) are expected to be produced and tested in Phase 1, and a larger number of units produced in phase 2 (to be defined following completion of phase 1), with 1% of units tested or a proportion determined by statistical process control.

Consideration will need to be given to how the drying process is controlled and whether there is an ongoing requirement to measure moisture content of the plasma for example.

Animal models of major haemorrhage

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

Clinical data

Clinical data required will depend upon the indications for the dried plasma product. In the first instance it is likely that the product would be developed for use in austere military environments where supply of FFP/cryoprecipitate is challenging, with possible expansion to civilian use for the treatment of major haemorrhage.

Current guidance from the FDA for indications for dried plasma where conventional plasma is unavailable is that a dose escalation study (1,2 3 units) should be completed in normal volunteers with end points of coagulation factor measurement and adverse events. FDA guidance if the dried plasma is used to replace conventional plasma states that well conducted clinical studies are required but do not specify what these should be. Manufacturers are required to discuss these in pre-submission meetings. The requirements for clinical studies for CE marking are not yet known, especially given the recent changes to directives in May 2020 and changes in UK regulations/requirements following exit from the EU.

Precedent for the clinical use of dried plasma already exists. The UK produced dried plasma in the 1940's routinely, but this practiced stopped due to concerns over viral transmission prior to modern standards for virology testing. Dried plasma produced by Germany (Lyoplas) and South Africa (Biopharma FDP) have been in use since the 1990's with the same clinical indications as for FFP and an excellent safety record (Pusateri et al 2019). Additionally, some UK air ambulances now use Lyoplas in their trauma packs. Likewise, the French military have produced and used dried plasma since the mid 1990's, and this was approved by the FDA for military use in 2018. Therefore, if the final devices used were CE marked, the degree of novelty of the type of product is considered 'low' as defined in Chapter 8 of the UK guidelines.

It therefore stands to reason that the main focus of new technology to produce dried plasma is to demonstrate in laboratory studies that the manufacturing process does not have unexpected effects. As the main indication for dried plasma would be expected to be for the treatment of major haemorrhage, it follows that the relevant comparator would be extended thawed FFP, or alternative dried plasma preparations in use. If laboratory studies demonstrate that dried plasma is similar to these preparations, then there is no rationale to perform phase 1 safety studies in volunteers as there is no evidence of increased adverse events with dried plasma to date, and studies in patients to address safety would be the next step. This is consistent with systems licenced for pathogen inactivation of plasma, where phase 1 safety studies have not been a feature – solvent-detergent treated, and methylene blue treated and riboflavin-treated FFP went to phase 2 clinical studies or clinical use following on from laboratory studies. In addition, phase 1 studies for novel red cells and platelet products usually focus on assessing the ability of the cell to survive in normal volunteers using a small aliquot of the product, and usually do not assess safety endpoints. Dose escalation studies for Intercept treated FFP were performed, and it is not clear whether this was required by regulators who regarded this as a higher class device than MB-FFP, this might be in part due to toxicological concerns relating to the mechanism of action of amotosalen in causing permanent cross linking of nucleic acid.

A key consideration for dried plasma is in relation to any excipients added during the manufacture process and the likely impact of these in vivo. Further, in the assessment of PI systems, some manufacturers tried to assess the likelihood of neoantigen formation, which is notoriously difficult to do, but a proteomic approach could be applied to plasma to demonstrate that the drying process does not significantly alter plasma proteins.

In addition to the requirements from SACBC/JPAC for validation outlined here, as part of the CE marking process manufacturers developing these technologies will need to discuss requirements with the relevant regulator and these may also be affected by the regulatory classification of the device once known.

Phase 2/3 Clinical studies/observations should include human data on allergic and other infusion reactions, other safety considerations, and efficacy as below.

Clinical studies comparing dried plasma with untreated or an alternative dried plasma 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 compare the efficacy of standard or untreated plasma are lacking, because the effect size between different plasmas with regards to clinical efficacy is likely to be very small, and this means that large trials will take a very long time to run, in order to demonstrate any difference. Hence, peer-reviewed observational data and data from post-marketing surveillance including national haemovigilence reports are the best way to collect information on efficacy and safety of plasma products.

It is expected that controlled studies in a suitable patient group such as cardiac surgery would be undertaken to assure that there are no unexpected adverse events or effect on efficacy prior to routine use. Such studies might be designed as non-inferiority studies compared to standard of care with laboratory endpoints indicative of efficacy. Manufacturers should consult SACBC for further advice.

Regulatory considerations

Based on the current medical devices regulations, it is expected that dried plasma will be produced using class II/III medical devices and considered a blood component. However, this will be discussed with the MHRA by commercial companies developing such technology in conjunction with UKBTS. The above validation criteria are based on the assumption that these are blood component. If that is not the case, further dialogue will be needed to define the relevant pathway for validation and approval in the UK.

Blood components produced from single blood donations or small pools of donations using medical devices must comply with the Blood & Safety Quality Regulations (BSQR) in the UK. Currently all plasma produced by NHSBT for transfusion is derived from a single donor, except for cryoprecipitate where 5-6 donations are pooled to provide a sufficient therapeutic dose. NHSBT centres hold a Blood Establishment Licence and are inspected and regulated by the MHRA. Novel components produced using medical devices must be validated and produced in accordance with the BSQR. In addition to the requirement to CE mark medical devices to market them in the UK, UK Blood Services have established mechanisms for the validation and approval of novel blood components produced using medical devices. The route to approval of a new specification for dried plasma would be through SACBC/JPAC if the product is produce using a medical device and is considered a blood component.

Table 1a: Coagulation Parameters to be validated and expected minimum values following manufacture 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	PT ratio	Y	NA	NA	NA
screening tests	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	Fibrinogen (Clauss)	Y	≤40	≥1.70g/l	1.50g/l
factors	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	0.60 U/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	≥0.50 U/ml	0.40 U/ml
	Multimers	Y	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

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.

Table 1b: Biochemical Parameters to be validated (acceptable ranges yet to be determined)

Residual excipients (if added)	
Particulates	
Moisture content	
Immunoglobulins	Total, IgG, IgM, IgA
Lipoproteins	Total triglycerides, total cholesterol, HDL, LDL
pH	
Osmolality	
Total protein	
Albumin	
Sodium	

References

Backholer, L., Green, L., Huish, S., Platton, S., Wiltshire, M., Doughty, H., Curnow, E. and Cardigan, R. (2017) A paired comparison of thawed and liquid plasma. *Transfusion*, 57: 881-889

Huish, S., Green, L., Curnow, E., Wiltshire, M. and Cardigan, R. (2019) Effect of storage of plasma in the presence of red blood cells and platelets: re-evaluating the shelf life of whole blood (Editorial, p. 3293). *Transfusion*, 59: 3468-3477

Martinaud C., Civadier C., Ausset, S., Verret, C., Deshayes, A-V., Sailliol, A. (2012) *In Vitro* Hemostatic Properties of French Lyophilized Plasma. *Anesthesiology*, 117:339-346

Mohr H, Knuver-Hopf J, Lambrecht B, Scheidecker H, Schmitt H. No evidence for neoantigens in human plasma after photochemical virus inactivation. *Annals of Hematology* 1992;65:224-228.

Pusateri, A.E., Given, M.B., Schreiber, M.A., Spinella, P.C., Pati, S., Kozar, R.A., Khan, A., Dacorta, J.A., Kupferer, K.R., Prat, N., Pidcoke, H.F., Macdonald, V.W., Malloy, W.W., Sailliol, A. and Cap, A.P. (2016) Dried plasma: state of the science and recent developments. *Transfusion*, 56: S128-S139

Zur, M., Glassberg, E., Gorenbein, P., Epstein, E., Eisenkraft, A., Misgav, M. and Avramovich, E. (2019) Freeze-dried plasma stability under prehospital field conditions. *Transfusion*, 59: 3485-3490

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