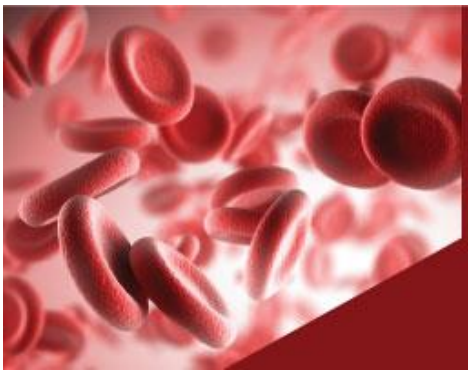

2020 South Central Association of Blood Banks ABSTRACT JOURNAL

2020 Sol Haberman Award Winner

Investigation of incidence and transfusion implications of the (C)ce^{s} type 1 haplotype

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Abstract:
Genotype matching for Sickle Cell Disease (SCD) patients is commonly recommended to avoid transfusion complications and alloimmunization. The (C)ce^{s} haplotype is one of the most common variants seen in the African American (AA) population. This haplotype is not detected by serological phenotyping methods so alloimmunization is common. This adds importance and utility to genotype matching.

Commercially available molecular methods can be helpful in screening donors at a high volume. The C (+)* call on the PreciseType HEA assay indicates a possible (C)ce^{s} haplotype. This call may present as an advantage in helping with the selection of units that could be further characterized for possible genotype matching in the SCD population.

The intentions of this retroactive study were to compare published frequencies of the (C)ce^{s} haplotype with a Southeast AA donor population and to investigate the best way to utilize molecular testing for these donors. The advantages of different testing methodologies and strategies to increase utility of AA inventory are also discussed.

Introduction:
Transfusion concerns: Phenotype matching is recommended for SCD patients. Matching for C, E, and K has been reported to decrease alloimmunization by 18%-75%.[1] Due to the availability of AA units, these patients are commonly transfused with RhD negative units from white donors. This can lead to the depletion of the RhD negative blood supply and also pose greater risk of alloimmunization because of the genetic diversity between these populations. Reports state that in about 85% of SCD patients, at least one RH allele differs between the alleles commonly found in the white donor population.[2]
To add to this concern, one study of 550 patients found that when transfused with Rh matched AA donors, 175 Rh antibodies in 105 individuals were formed.[3] Another report found 82% of SCD patients with Rh antibodies presented serologically positive for the corresponding antigen.[3] A large collection of SCD patients demonstrated that 50 of 242 C+ patients have partial C antigen, making them at risk for forming anti-C if routine Rh phenotype matching is employed. The same study showed 21% of patients were homozygous for altered Rhce antigens. 53% of RHCE alleles were altered in a large group of both patients and AA donors.[3] Analysis of antibody frequency in a group of patients with a partial C phenotype showed 30% produced anti-C when with transfused C positive red blood cells.[5] Patients with homozygous RH variants can also be at risk for alloimmunization to high frequency antigens that these populations lack such as hr^{B} , hr^{S} , and Hr^{B}. [6]

These issues highlight the need for genotyping both AA patients and donors. It has been recommended to genotype all SCD patients that present C positive by serology and transfuse with C negative units if a partial C is predicted. Utilizing RH genotyping when Rh antibodies are present has also been suggested.[1]

(C)ce^s haplotype: The (C)ce^s haplotype (also called r'^{s}) is a result of the genetic exchange of RHCE exons 4 to 7 into the RHD locus. It consists of a hybrid D-CE-D^s allele, as well as the ce^s RHCE allele. The hybrid gene consists of Exons 1, 2, 3, 8, 9, and 10 from RHD and Exons 4 through 7 from RHCE. The following Single Nucleotide Variants (SNV) are present- 186G>T, 410C>T, 455A>C, 733C>G, 1006G>T. The RHCE ce^s allele consists of 48G>C, 733C>G, 1006G>T SNVs. This haplotype yields a weak, partial C (RH2), weak e (RH5), partial c (RH4), VS (RH20) and Rh42 (RH42), and is negative for D (RH1), hr^b (RH31), Hr^b (RH34), and V (RH10).[7,8,9]

While studying D negative, C positive, and VS positive donors, Blunt et al. suggested this phenotype may be due to an RHD/RHCE recombinant gene.[10] Faas et al. described the hybrid allele when investigating VS positive donors with weak C expression. Faas postulated that the VS expression and the weak e expression was due to the 733C>G mutation is Exon 5.[11] It was hypothesized that the weak C expression was due to the presence of 307T near RHCE sequences in combination with 48G on the hybrid allele and the 186T affecting expression as it is close to sites associated with C expression.[11,12] In 1998, the occurrence of 1006T SNV in both the hybrid allele and the ce^s allele was identified. It was also postulated that the molecular background of the V antigen may be linked to the expression of VS due to the presence or absence of the 1006T SNV in combination with the presence of 733G. [12]

A second (C)ce^s haplotype which was termed Type 2 was described. It was defined as a hybrid RHD*D-CE(4-7)-D allele (Exons 1, 2, 3, 8, 9, and 10 from RHD and Exons 4 through 7 from RHCE) along with the ce^s RHCE allele. It also encodes a weak partial C, partial e, partial c, and VS, but does not produce D, V, or Rh42. The conventional RHD background and differing 5' breakpoints in the hybrid allele are the two discriminating features in this haplotype.[7]

It was reported that the ce^s allele also frequently occurs with a non-hybrid DIII type 5 gene. This haplotype does not produce the weak partial C expression or Rh42 and has partial D expression.[7] Later, Westhoff and colleagues explained the 819G>A in the DIII type 5 is silent and does not change amino acid expression thus it is phenotypically identical to the DIIIa allele. They suggested making the DIII type 5 allele designation obsolete to avoid redundant nomenclature. It was also proposed to designate the common hybrid gene RHD*DIIIa-CE(4-7)-D to avoid confusion with the other hybrid. This analysis also verified that the is often associated with the ce^s allele.[13]

Published frequencies show the (C)ce^s type 1 haplotype occurs in 7–15% of black individuals[14] and 7.5% in SCD patients in Afro-Caribbean populations.[15] The ce^s allele has been reported at 9% in South African populations, 12% in a black Dutch population.[12] More recently, it has been found in 4.2% in SCD patients and 3.3% in AA donors , while the RHD*DIIIa-CE(4-7)-D allele was present in 3.1% in SCD patients and 2.6% in AA donors.[3]

Materials and Methods:
Sample selection: Donors who self-identified as black/African American in Florida, Alabama, or Georgia were selected for testing. Informed consent was acquired from all donors. Data was acquired and analyzed in two sets.

The first data set was to establish the incidence of the (C)ce^s type 1 haplotype in this AA donor population and the percentage of C positive AA donors with this haplotype present. 1055 African American donors were tested by PreciseType HEA BeadChip assay (Bioarray Solutions-Immucor, Inc., Warren, NJ). Presence of C antigen was serologically tested. Donors identified by HEA to be possible (C)ce^s haplotype were reflexed to IDCORE XT (Progenika Biopharma, Bizkaia, Spain/ Grifols, Emeryville, CA).

The second data set was to further characterize the frequencies of other genotypes identified from the (C)ce^s call by HEA and determine the number of actual C negative donors in this group. Over a two year period, donors yielding a C (+)* call were tested by serology and reflexed to IDCORE XT. Numbers from data set one were included in data set two.

Molecular Methods: DNA extraction- Genomic DNA was isolated from peripheral blood collected in EDTA by manual or automated methods (QIAsymphony DNA Mini Kit; QIAGEN, Valencia, CA).

Molecular assays: PreciseType HEA Molecular BeadChip Test and the IDCORE XT assay were performed according to manufacturer's recommendations.

Serological methods: Serological testing was performed by one or more of the following methods: automated methods with monoclonal anti-C (Clone MS24) gel cards (Micro Typing Systems, Pompano Beach, Florida) on Ortho Vision (Ortho Clinical Diagnostics, Raritan, NJ), monoclonal anti-C reagent (Clone MS24/P3X25513G8) on the PK7300 system (Beckman Coulter, Brea, CA) or manually by tube method with monoclonal anti-C reagent (Gamma-clone MS24, Immucor, Inc., Norcross, GA).

Testing for (C)ce^{s} haplotype: PreciseType HEA BeadChip assay may cause C typing discrepancies due to the C (+)* on units that had previously been typed by serology as C negative. It can also lead to the demand for C negative units to be placed on D negative supply unnecessarily. This test displays a (+)* for the predicted C phenotype result when 1006G>T and 733C>G variants are detected, as these are two of the SNV present in the ce^{s} allele. This allele commonly is linked to the hybrid RHD*DIIIa-CE(4-7)-D allele that produces a partial C antigen. The assay lacks a probe to verify presence of this haplotype.[16]

Multiple molecular methods for detection of the haplotype have been described. The first uses the 5' breakpoint in RHD intron 3 (IVS3+3100a>g).[17] In 2004, Flegel described an assay that could be used to interrogate the 3' breakpoint found within RHD intron 7.[18]

IDCORE XT uses the 5' breakpoint to predict this haplotype.[19] Because the breakpoint is not shared, IDCORE XT will not detect type 2 haplotypes. It may be worth noting that because IDCORE XT does not interrogate the RHCE c.48C nucleotide, it is unable to distinguish the RHCE*ceVS.03 (ce^{s}) allele from the RHCE*ceVS.05 allele, which possesses the 733C>G and 1006G>T SNVs but not 48G>C. The two are phenotypically similar, the difference being the partial c in the ce^{s} allele.[6] Studies have shown the former to be much more common in AA donors.[3]

Results:
Data set one: Of the 1055 AA donors tested, 48 (4.5%) resulted as C (+)* by HEA, while 1007 (95.5%) did not have the C (+)* call. When tested with the IDCORE XT assay, 27 samples (2.6% of the total 1055 donors) were positive for the (C)ce^{s} type 1 haplotype. The (C)ce^{s} type 1 haplotype was not detected in 21 samples. The genotypes detected and C typing results are presented in Table I. One sample did not have red cells available for serological typing. Only one donor with the (C)ce^{s} haplotype present (RHCE*ce, RHD*r's-RHCE*ce[733G,1006T]) was found to be negative by serology. Sequencing will be performed to investigate the cause of the phenotypic variance. Of the 332 donors that tested C positive by serology, the (C)ce^{s} type 1 haplotype was present in 25 (7.5%).

Table I. Data Set One- Genotypes and Serologic C phenotype

Genotype	Genotype Incidence	C Negative	C Positive	Serology Not tested
RHCE*ce, RHCE*ce[733G,1006T]	16	16		
RHCE*ce, RHD*r's-RHCE*ce[733G,1006T]	16	1	14	1
RHCE*cE, RHCE*ce[733G,1006T]	1	1		
RHCE*ce[712G], RHCE*ce[733G,1006T]	1	1		
RHCE*ce[733G,1006T], RHD*r's-RHCE*ce[733G,1006T]	1		1	
RHCE*ce[733G], RHCE*ce[733G,1006T]	3	3		
RHCE*ce[733G], RHD*r's-RHCE*ce[733G,1006T]	10		10	

Data set two: Over a two year period, 1342 donors' predicted C phenotype by HEA resulted as (+)*. 821 were reflexed to IDCORE XT. Of the 821 tested by IDCORE XT, 10 were unavailable for serological typing. 572 (69.7%) donors had the (C)ce^{s} haplotype present. 229 of those had a variant present in trans. No donors were homozygous for the (C)ce^{s} haplotype. One sample repeatedly resulted as unknown for all RH antigens. Donor will be retested at next donation and sent for sequencing. 3 of the 56 RHCE*ce[733G], RHCE*ce[733G,1006T] donors tested positive for C serologically. One of the two RHCE*ce [733G, 1006T] donors also tested positive for C serologically. Further serological and molecular testing may be performed in the future to investigate these discrepancies.

Table II. Data Set Two- Genotypes and Serologic C phenotype

Genotype	Genotype Incidence	C Negative	C Positive	Serology Not tested
RHCE*ce, RHCE*ce[733G,1006T]	160	156	3	1
RHCE*cE, RHCE*ce[733G,1006T]	28	27		1
RHCE*ce, RHD*r's-RHCE*ce[733G,1006T]	343	1	337	5
RHCE*cE, RHD*r's-RHCE*ce[733G,1006T]	85		82	3
RHCE*ce[712G], RHCE*ce[733G,1006T]	2	2		
RHCE*ce[712G], RHD*r's-RHCE*ce[733G,1006T]	3		3	
RHCE*ce[733G,1006T], RHD*r's-RHCE*ce[733G,1006T]	18		18	
RHCE*ce[733G], RHCE*ce[733G,1006T]	56	53	3	
RHCE*ce[733G], RHD*r's-RHCE*ce[733G,1006T]	119		119	
RHCE*ce [733G, 1006T]	2	1	1	
RHCE*ceAR, RHD*r's-RHCE*ce[733G,1006T]	4		4	
Unknown	1	1		

Discussion:
Using this algorithm does not allow the possibility to discover those donors that may have the (C)ce^{s} haplotype on one allele when the C gene is present on the other allele as HEA will report the C phenotype as C positive.

It is important to note that it is not possible to differentiate a partial C antigen caused by the (C)ce^{s} haplotypes and a conventional C antigen by serology.[20] This accentuates the importance of genotyping all serologically C positive SCD patients.

Using HEA to discover potential donors to reflex to further RHCE testing to use as genotype matched donors should be considered. The presence of the VS antigen may also be a viable way to screen potential donors.

There are limited commercial options available for RHCE genotyping. The IDCORE XT assay can predict 69 different RHCE genotype combinations and provides that information without additional testing to the RBC genotyping assay. This test is also approved for In Vitro Diagnostics (IVD).[19] The Immucor RHCE Molecular BeadChip Test is another option which uses 25 markers to make allele calls, 17 of which are not included in the IDCORE XT assay. It is currently labeled for Research Use Only (RUO).[21] There are several options for commercial real time polymerase chain reaction RHCE kits available.

The promise of next generation sequencing will no doubt benefit transfusion medicine in many aspects including genotype matching. [22] However, blood centers as well as hospitals will need to address the complication of having an effective way to store and utilize that data.

Summary:
Alloantibody development in patients with the (C)ce^{s} haplotype is a risk that may be avoided with genotyping. Genotyping appropriate donors and being prepared to offer genotyped units may help mitigate transfusion risks in these patients.

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The following Abstracts were submitted for review by the South Central Association of Blood Banks Program Committee chairs and were selected in either ORAL or POSTER categories for the 2020 Abstract Journal. The ORAL Abstracts were presented on July 23, 2020 via webinar, and POSTER Presentations are available at www.scabb.org.

T/S 1 ORAL

A Hemoglobin S Solubility Screen That Conserved Solution

Author: R. Gammon, A. Delk

Background: The need to offer supportive transfusion therapy for patients with sickle cell disease (SCD) continues to grow. Preferred red blood cells (RBC) for SCD patients are hemoglobin-S (HbS) negative and matched for the patient's common Rh antigens and K. There are several methods to test for HbS including electrophoresis, solubility, and molecular testing. Our Immunohematology Reference Lab (IRL) relies primarily on solubility testing. The majority of our patients are D positive and will require C-,E-,K- RBCs. Since most of the compatible blood is from African American donors, 8-10% will be HbS positive. The solubility kit used in the IRL required 2 mL of solution and 10 uL of red blood cells (RBCs) to provide 0.5 tests per mL of solution. As a result, testing is on demand and HbS positive donors are frequently encountered. This can slow the process of finding compatible units. Our IRL attempted to reduce the volume of solution needed to increase the number of units we could test per mL of solution. We did this not to replace the instructions for use (IFU), but rather to potentially use as a screen for known rare donors so that we could identify the HbS positive units in advance of anticipated needs.

Methods: We performed a ratio reduction calculation to determine the volume of RBC solution to add to the microplate. The ratio of 10 uL RBCs/2 mL solution required by the IFU was converted to 10 uL RBC /2000 uL. As the microplate wells will comfortably hold 200 uL, we divided by a factor of 10 to get 1 uL/200 uL solution. We then increased the concentration to 2 uL RBC/200 uL solution since the smallest volume pipette in the IRL was 2 uL**.**

Results: We tested 25 known HbS positive and 25 HbS negative donor RBCs. With the calculated volume approach, all 25 known HbS positive RBC units tested positive and all 25 known HbS negative RBC units tested negative.

Conclusions: Using the ratio reduction calculation approach we achieved 100% concordance with known results. This allowed the IRL to perform ten times as many screening tests or 5 tests per mL of solution vs 0.5 tests per mL per IFU. This approach may be helpful to screen donor RBC units that are HbS screen negative in advance of anticipated needs. When ordered, a screened RBC unit would simply be tested as per the IFU with the likelihood of an unexpected positive minimized. This would serve to reduce turnaround time to optimize patient care.

T/S 2 POSTER

A Snapshot of Donor Genotypes at a Very Large Blood Center in the Southeastern US

Author: M. Gannett, R. Gammon

Background/Case Studies: Many of the phenotype frequencies were performed years ago in populations that do not reflect the heterogeneity seen today. We attempted to determine the genotype frequency of a diverse population serviced by a single blood center in the Southeastern US (Florida, Georgia, North and South Carolina and Alabama) and compare it to previously established frequencies.

Study Design/Methods: The blood center's immunohematology reference laboratory tested select donors using the HEA BeadChip (IVD) methodology using a panel of 35 antigens and for the presence of the hemoglobin S allele. Routine Rh antigens (C, E, c, e) and K were excluded from analysis as a result of our practice of selecting donors in part upon serologic screening for these antigens. Results were retrospectively evaluated by the race the donor self-identified.

Results/Findings: From 2014-2018, 14,072 donors were genotyped (Table). Our African American (AA) donor population had a number of frequency differences than previously reported: V+ (43.5%) and VS+ (46.7%) vs. 30%, Js(a+) 17.3% vs. 20%, Fy(a–b–) 79.1% vs 68%, Jk(b–) 57.6% vs. 51.1%, and Jo(a–) 1.02% vs >99%, with Hy– being four times less common. A relatively high frequency (≈10%) of our Hispanics were V+ and VS+, and 4.1% were Di(a+) vs 1%. Overall, only 4.8% of our donors were Co (b+) vs the 10% reported for most populations. Frequency of donors having least one hemoglobin-S allele in African-Americans was 5.9% vs. 7.3% and 1.5% vs. 0.7% in the Hispanics.

Conclusions: There were differences in current genotype frequencies at this blood center compared to historical phenotype frequencies, in particular for low prevalence antigens. The donor genotype information obtained from this study may be used to target recruitment of a particular ethnicity compatible with recipients located within the blood center's service area.

T/S 3 POSTER

Acute Hemolytic Transfusion Reaction Due To Prepooled Platelets (PLTs)

Author: R. Gammon, S. Cook, A. Trinkle, K. Thomas, K. Benson

Background/Case Studies: A 65 y.o., group A, 52 kg woman with relapsed lymphoma was thrombocytopenic (5,000/uL, 150,000-450,000/uL); prestorage irradiated leukocyte-reduced pooled platelet concentrates (PPLTs), (5 group O donors), were transfused in 47 minutes. There were no adverse events (AE), she was discharged home but 1.5 hours later she returned with fever (103^0F), rigors, dyspnea, cough, chest pressure, nausea, diarrhea and dark urine. Hypotension and tachycardia developed; she was admitted to ICU.

Study Design/Methods: Post-transfusion blood and urine samples were obtained. The PLT bag had been discarded. Serial dilutions (with saline diluent) from the five donor testing tubes and a simulated PLT pool were performed, read at immediate spin (IS) and IgG (1 hr. at 37C incubation).

Results/Findings: Testing confirmed an acute hemolytic transfusion reaction (AHTR): elevated LDH (996 U/L, 135-225 U/L), undetectable haptoglobin (<10 mg/dL, 30-200 mg/dL), prolonged PT (18.3 sec, 10.2-12.9), INR (1.5, 0.8-1.1), aPTT (47.3 sec, 25.1-36.5), no evidence of infection on blood or urine cultures and urinalysis showed dark amber urine with large blood on dipstick but only 0-2 RBCs/high powered field. Pretransfusion hemoglobin of 8.9 g/dL fell to 7.5 g/dL upon admission; PLT count of 5,000/uL increased to 27,000/uL. She developed acidosis, lactate (7.0 mmol/L, 0.5-2.0 mmol/L). Her hemoglobinuria cleared and she required extensive support before discharge home. This was the second AHTR due to PLTs in 32 years of this hospital's operation with 12,195 PLT transfusions in 2018. The first event in 1997 occurred with apheresis PLTs. Both events occurred in smaller–sized recipients. At 37 C the simulated pool and donor 5 had high-titer anti-A. The donor was permanently deferred. (Table)

Conclusions: ABO-mismatched PLT transfusions result in good increments and the majority are free of significant AE. Physicians must be aware of the potential risk of AHTR when platelets are transfused and that post-transfusion fever +/- chills is typically benign but should be rapidly investigated to rule out more serious AE. PLT-associated AHTR has occurred with apheresis PLTs but is very rare with WBD PLTs. These two events could have been prevented by restricting ABO-incompatible plasma volumes in smaller-sized adults (via PLT concentration or use of platelet additive solution), donor anti-A titer screening or avoiding group O PLTs to non-O recipients. As a precaution, we are now volume-reducing ABO-incompatible plasma in platelets to be transfused to all patients less than 60 kg body weight.

Unit	Test	Anti-A	Anti-B
Simulated Pool	IS	256	128
Simulated Pool	37 C	2048	256
Donor 1	IS	32	32
Donor 1	37 C	512	128
Donor 2	IS	128	128
Donor 2	37 C	256	512
Donor 3	IS	128	16
Donor 3	37 C	128	16
Donor 4	IS	64	16
Donor 4	37 C	32	16
Donor 5	IS	256	256
Donor 5	37 C	4096	512

T/S 4 ORAL

Collection of Convalescent Plasma at a Large US Blood Center

Authors: R. Gammon, Y. Wu, R. Reik

Background/Case Studies: COVID-19 convalescent plasma (CCP) is collected from individuals who have recovered from COVID-19. CCP contains antibodies to severe acute respiratory syndrome coronavirus 2 or SARS-CoV-2 (the virus that causes COVID-19). As there are currently no specific therapeutic agents to treat COVID-19, and as CCP has not yet been approved by the US FDA, it is being administered as an investigational product to patients with COVID-19. We describe the experience of CCP collection at a large blood center in the United States.

Study Design/Methods: Per FDA Guidance, CCP was collected from individuals who had a diagnostic test (e.g., nasopharyngeal swab) at the time of illness or a positive serological test for SARS-CoV-2 antibodies after recovery or a physician attestation that this testing was performed and complete resolution of symptoms for at least 14 days before the donation. CCP donors met all allogeneic donor requirements, although the medical directors could make an exception on a case-by-case basis. Collection was either by whole blood or plasmapheresis. All CCP underwent relevant transfusion-transmitted infection testing, females with a history of pregnancy were tested for human leukocyte antigen (HLA) antibodies. The presence of SARS-CoV-2 antibodies using an ELISA methodology were determined on selected donations.

Results/Findings: From 04/14-05/09/20, 494 CCP donors [232 female, 260 males, 2 not provided; 340 first-time, 154 repeat donors, mean age 50.1 years (range 16-85), 111 whole blood, 383 plasmapheresis] presented for CCP donation from which 450 (91.1%) plasma donations were collected. Plasma volume collected from whole blood mean: 341 mL (range 233-403), plasmapheresis mean 643 mL (range 210-680). CCP collection time from initial positive test was mean 33.9 days (range 5-57), CCP collection time from date donor became asymptomatic was mean 28.7 days (range 14-61). 105/450 (23.3%) CCP collections were discarded due to positive test results (73 HLA antibodies, 2 positive red cell antibody screen, 4 anti-HCV, 1 anti-HIV 1/ 2, 13 serologic test for syphilis, 2 anti-*T cruzi*, 10 anti-HBc). In addition, 149 CCP donor plasma samples were tested for total antibodies to SARS-CoV-2 (139 (93.3%) were reactive mean S/CO 62.87 (1.51-454), 10 (6.7%) were nonreactive S/CO mean 0.15 (0.03-0.8).

Conclusions: This blood center was able to demonstrate the success and feasibility of a CCP collection program. In addition, the majority of CCP demonstrated the presence of SARS-CoV-2 antibodies.

T/S 5 ORAL

Convalescent Plasma with Negative Antibodies Effects on Patients

Authors: R. Gammon, Y. Wu, R. Reik

Background/Case Studies: COVID-19 convalescent plasma (CCP) is collected from recovered patients. While most develop antibodies, testing post CCP collection found some negative for SARS-CoV-2 antibodies. We reviewed CCP recipient outcomes to determine if there was improvement and donor intake records for a common cause.

Study Design/Methods: All met US FDA allogeneic blood donor and CCP requirements. Per US FDA Guidance (05/01/20) CCP was collected from individuals who had diagnostic test at time of illness or positive serological test for SARS-CoV-2 antibodies after recovery. SARS-CoV-2 antibodies by ELISA was determined on selected CCP donations. Hospitals were surveyed on CCP recipient's clinical course, number of doses, concurrent medications and status. Blood center intake files of donors' screening criteria were reviewed.

Results/Findings: 7 CCP donors of 11 plasma products tested negative for SARS-CoV-2 antibodies during test validation. 4/11 were discarded due to HLA antibodies, 7 were transfused at six hospitals. Time from admission until CCP transfusion: mean 8.7 days (range 1-16 days). Time in ICU until CCP transfusion: mean 6.9 days (2-16 days), 3 patients never in ICU. Time patient had been on ventilator until CCP transfusion: mean 12.0 days (2-29 days), 1 patient never on ventilator. 5 hospitals reported 2 patients weaned from ventilator, 1 was not and 2 were never on ventilator. Changes in clinical course within 72 hours from three patients showed 2 normalized body temperature, 2 increased PAO_2/FIO_2, but none reported resolution of cough, shortness of breath or chest pain. Changes in laboratory values reported from 4 patients showed 3 decreased C-reactive protein, 1 decreased procalcitonin and 1 developed an elevated white blood cell count. 2/7 of (28.6%) patients received a second CCP. Time one patient had been on ventilator until second CCP transfusion was 10 days. Changes in clinical course within 72 hours posttransfusion reported none normalized body temperature nor reported resolution of cough, shortness of breath or chest pain and 1 increased PAO_2/FIO_2. For 3 patients it was reported at any point during their hospital stay 2 received antivirals, 2 steroids, 1 hydroxychloroquine, and 2 zithromycin. Current status of 4 patients: 1 mechanical ventilation, 2 oxygen, and 1 room air. 4/7 (40%) CCP donors tested positive for HLA antibodies vs. 73/450 (16.2%) all blood center CCP donors. 3/7 (42.9%) antibody negative donors tested positive for SARS-CoV-2 by PCR at one facility. 2/7 (28.6%) tested positive for SARS-CoV-2 antibodies prior to donating CCP.

Conclusions: In this uncontrolled case series of 7 patients with COIVD-19 who received CCP without demonstrable SARS-CoV-2 antibodies, some experienced improvement in clinical course. Limited sample size and study design precluded a definitive statement about the potential effectiveness of CCP.

T/S 8 POSTER

The Importance of Multi-Stakeholder Approaches for Permanent Risk Reduction

Authors: M. Keskula

Objective/Background: While Cesium-137 (Cs-137)-based blood irradiators are fundamental to avoiding transfusion-associated graft vs. host disease (TA-GVHD), they also present a security risk; cesium chloride, a highly dispersible form of Cs-137 used in these irradiators, can be used by malicious actors to build a radiological dispersion device (RDD) or "dirty bomb". Detonation of an RDD, according to numerous studies, would likely result in substantial economic and psychological damage. In recent years, X-ray-based blood irradiators have provided a clinically effective and risk-reducing replacement for Cs-137 irradiators. Since 2014, the National Nuclear Security Administration Office of Radiological Security (ORS), through its Cesium Irradiator Replacement Program (CIRP), has worked with sites that have voluntarily elected to replace their Cs-137 based irradiators with x-ray irradiators. Using experiences garnered through this program, we seek to outline the wide variety of stakeholders and interests involved in the transition from Cs-137-based blood irradiation to X-ray irradiation, and to assist sites in developing a roadmap for making that transition.

Methods: The search strategy focused on identifying experiential data from past ORS engagements with sites through the CIRP and identifying lessons learned through engaging with blood bank operators, hospital and university administrators, Radiation Safety Officers(RSOs), and other relevant stakeholders. In addition, we searched for white papers and academic research focused on the efficacy of X-ray irradiators, the cost considerations for changing devices, and the considerations inherent in planning a transition, to provide relevant context. Through this, we identified consistencies in motivations and roles among a variety of involved stakeholders.

Results/Findings: While different stakeholders (including site-level management, users, regulators and RSOs) may have different priorities when considering alternative technologies, we have identified several common trends among these stakeholders that have proven prominent in consideration of replacement efforts. The regulatory requirements of operating a Cs-137 device have proven to be especially important in incentivizing transitions; making sure that stakeholders are involved in the decision making process (and all parties involved are aware of the security ramifications of using cesium) is necessary to create momentum. Cost concerns are central to most stakeholders involved in operating a blood irradiation facility when considering the cost of procuring and operating an X-ray irradiator, disposing of the old Cs-137 irradiator, thinking about end-of-life costs for cesium, the cost of implementing new security systems or trainings to stay current on regulations for cesium devices, and liability for sources as licensees. Ensuring that alternative technologies can provide sufficient clinical results relative to radiological devices is also a notable concern to users. ORS has seen greater success when all of these interests are taken into consideration in a site's transition to x-ray irradiation.

Conclusion: When planning for a transition from Cs-137 to X-ray irradiation, stakeholder concerns, including costs associated with both technologies, the liabilities involved in maintaining Cs-137 irradiators, and the need to provide quality care to patients, need to be considered. Addressing these concerns in a holistic manner is the best mechanism for creating consensus in considering a Cs-137 to X-ray transition that would achieve permanent risk reduction. Efforts to incentivize replacement of Cs-137 based irradiators to achieve permanent risk reduction are more likely to be effective if they are able to address these key concerns sufficiently for all stakeholders. ORS can assist in this process through outreach, engaging with all involved stakeholders, awareness building, and providing cost assistance to improve the viability of transition.

T/S 9 ORAL

Using RHCE*ce Variants Paired With RHCE*cE Haplotypes to Conserve the R2R2 Blood Supply

Authors: K. Billingsley, S. Rinaudo, K. Bowman, M. Kalvelage

Background/Case Studies: RHCE*ce variants and the accompanying anti-e like antibodies are commonly identified in previously transfused patients of African descent. While RHCE*cE (R2R2) units are not the product of choice for a patient who does not have an RHCE*cE haplotype, R2R2 units are often used in lieu of genotype matched blood for transfusion if the patient is RHCE*cE paired with an RHCE*ce variant as these units are frequently more easily obtained. Though the R2 haplotype prevalence in African Americans and Caucasians is 11% and 14%, respectively; the R2R2 phenotype has a frequency of just 1- 2%. Therefore, the use of R2R2 units for patients with an RHCE*cE/RHCE*ce variant genotype puts an undue burden on the R2R2 donor base and reduces blood availability for those patients producing true anti-e.

Study Design/Methods: A 25-month retrospective study was conducted to evaluate the frequency of RHCE*ce variants paired with an RHCE*cE haplotype. Samples were submitted for genotyping using an in-house algorithm. DNA was extracted using previously published methods and tested using the IDCore XT assay (Progenika Biopharma, Bizkaia, Spain). Data was derived from self-identified African American donors and patients submitted for RBC genotyping.

Results/Findings: Of the 1303 samples assayed from African American sources, 523 (40%) yielded RHCE*ce variants paired with common RHCE haplotypes (RHCE*ce, RHCE*Ce, RHCE*cE). See table 1. Further evaluation of these haplotypes revealed 71 (13.6%) samples with an RHCE*cE haplotype pairing. See table.

Table 1: Heterozygous RHCE*ce variants with all common RHCE haplotypes Variant Type	Number detected	Percent detected
RHCE*ce(733G)	413	31.7
RHCE*ce(733G,1006T)	88	6.8
RHCE*ceAR	15	1.2
RHCE*ce(712G)	7	0.5