Transplant Study

Dr. Nina Guzzetta, Dr. Susana Cruz Beltran
Statisticians: Dr. Renee Moore, Andrea Lane

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1 Summary Tables

Table 1: Demographic and Surgical Data

	All patients (n=64)	PRA- (n=35)	PRA+ (n=29)	p value
Age (Years)	7.5 (2.0,13.0)	8.0 (2.5,13.0)	6.0 (2.0,12.0)	0.39
Gender				
Female	32 (50.0)	18 (51.4)	14 (48.3)	0.80*
Male	32 (50.0)	17 (48.6)	15 (51.7)	
Race				
White	29 (45.3)	14 (40.0)	15 (51.7)	0.29**
African American	26 (40.6)	13 (37.1)	13 (44.8)	
Hispanic	6 (9.4)	5 (14.3)	1 (3.4)	
Asian	2 (3.1)	2 (5.7)	0 (0.0)	
No response	1 (1.6)	1 (2.9)	0 (0.0)	
Weight at transplantation (kg)	26.2 (12.4,44.2)	28.6 (13.5,42.6)	17.6 (12.3,44.4)	0.58
Mechanical support prior to transplantation				
No	44 (68.8)	24 (68.6)	20 (69.0)	0.97*
Yes	20 (31.2)	11 (31.4)	9 (31.0)	
Number of previous surgeries	3.0 (1.8,7.0)	3.0 (2.0,7.5)	3.0 (1.0,7.0)	0.67
ABO Incompatible				
No	59 (92.2)	32 (91.4)	27 (93.1)	0.99**
Yes	5 (7.8)	3 (8.6)	2 (6.9)	
CPB time (min)	138.0 (117.0,180.0)	137.0 (123.0,180.0)	143.0 (111.0,175.0)	0.61
Donor cross clamp time (min)	209.0 (188.0,234.5)	209.0 (195.0,231.0)	210.0 (188.0,236.0)	0.84

Note: Continuous variables: median (Q1,Q3), Wilcoxon rank sum test; Categorical variables: N(%)
* Chi Square test; ** Fisher's exact test

Table 1 displays the demographic variables stratified by PRA status. All continuous variables were evaluated with the nonparametric Wilcoxon rank sum test because of non-normality. Categorical variables were evaluated with either a chi square test or Fisher's exact test depending on expected values. No variables in this table are statistically significant.

Table 2: Study Objectives

	All patients (n=64)	PRA- (n=35)	PRA+ (n=29)	p value
Diagnosis				
ARVD	4 (6.2)	1 (2.9)	3 (10.3)	0.20**
Dilated CM	33 (51.6)	16 (45.7)	17 (58.6)	
Failed SV	18 (28.1)	10 (28.6)	8 (27.6)	
Restrictive CM	7 (10.9)	6 (17.1)	1 (3.4)	
Other	2 (3.1)	2 (5.7)	0 (0.0)	
Number of previous blood product exposures	9.0 (1.0,23.2)	5.0 (0.0,20.5)	10.0 (2.0,25.0)	0.08
Development of donor specific antibody				
No	31 (48.4)	21 (60.0)	10 (34.5)	0.04*
Yes	33 (51.6)	14 (40.0)	19 (65.5)	

Note: Continuous variables: median (Q1,Q3), Wilcoxon rank sum test; Categorical variables: N(%)
* Chi Square test; ** Fisher's exact test

```
## [1] 20.65262
```

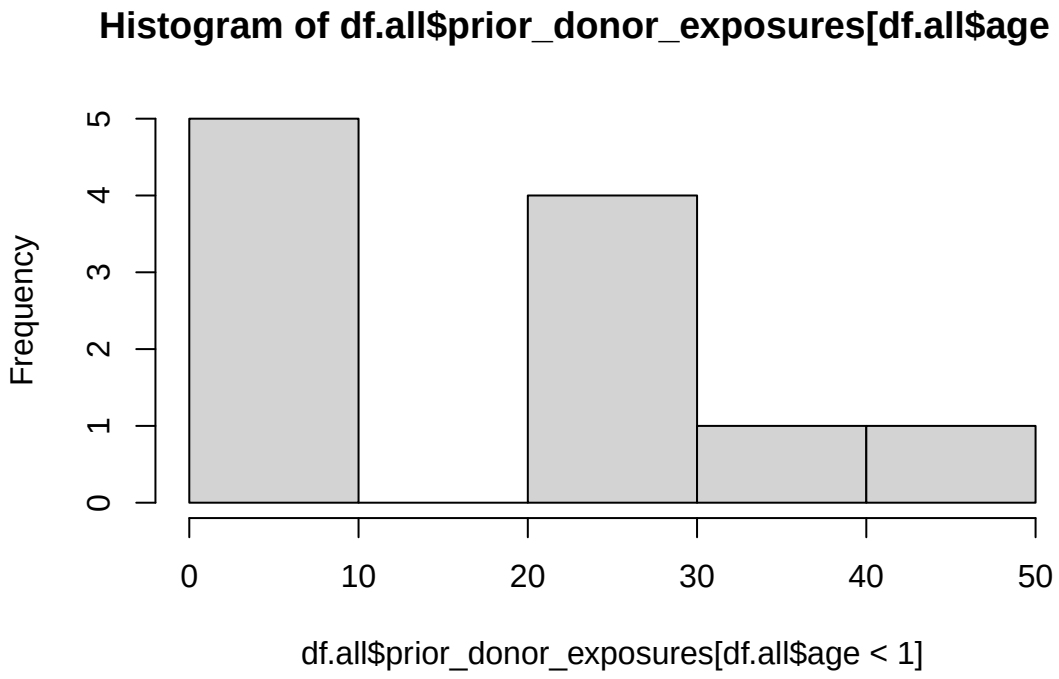
```
## [1] 14.52146
```

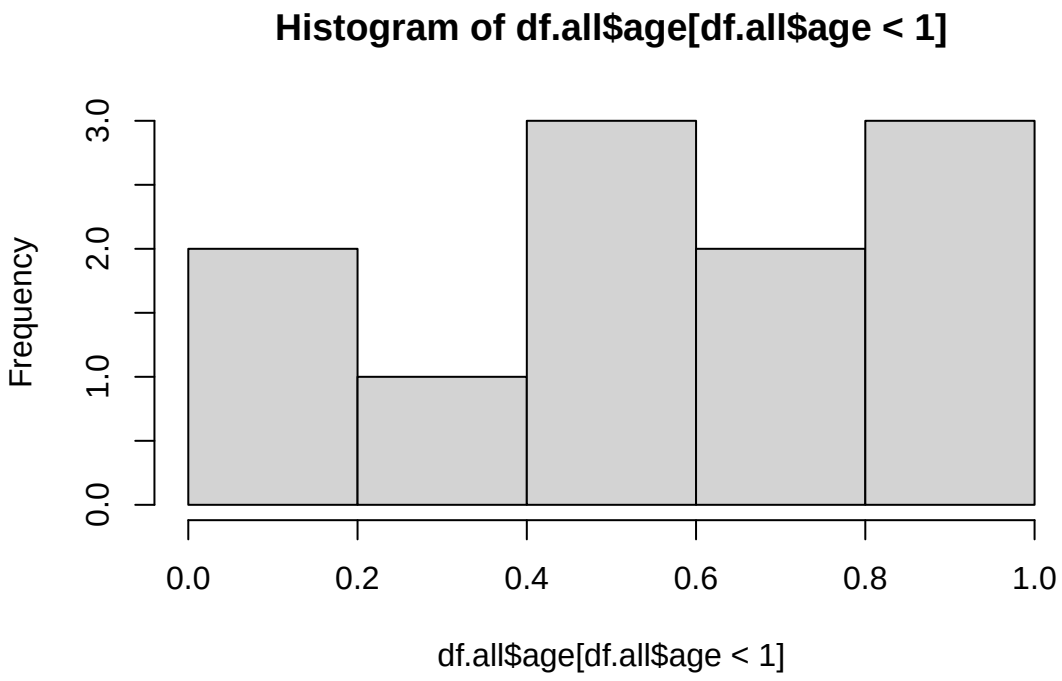
```
## [1] 11
```

```
## [1]  2 43
```

```
## [1] 16.54545
```

```
## [1] 21
```





This table displays variables relevant to the study objectives. Diagnosis categories were combined as follows: "LV Failure" was combined with "Dilated CM", and "RV Failure" and "Shone's complex" were grouped as "Other." As in table 1, continuous variables are evaluated with the nonparametric Wilcoxon rank sum test and categorical variables are evaluated with either the chi square test or Fisher's exact test, depending on expected values. None of the variables are significant at the 0.05 level, but blood product exposures and donor specific antibody are marginally significant.

Table 3: Outcome Variables

	All patients (n=64)	PRA- (n=35)	PRA+ (n=29)	p value
ICU Length of Stay (days)	7.0 (4.0,10.0)	6.0 (4.0,8.0)	7.0 (5.0,12.0)	0.08
VIS	10.0 (8.0,13.0)	10.0 (8.5,12.0)	10.0 (8.0,14.0)	0.47
Plasmapheresis (%)				
No	41 (64.1)	31 (88.6)	10 (34.5)	<0.01*
Yes	23 (35.9)	4 (11.4)	19 (65.5)	
Time to Extubation (hrs)	17.5 (10.8,39.0)	17.0 (9.0,28.0)	24.0 (11.0,86.0)	0.05
Rejection (%)				
No	55 (85.9)	32 (91.4)	23 (79.3)	0.28**
Yes	9 (14.1)	3 (8.6)	6 (20.7)	
ECMO support post op (%)				
No	60 (93.8)	34 (97.1)	26 (89.7)	0.32**
Yes	4 (6.2)	1 (2.9)	3 (10.3)	

Note: Continuous variables: median (Q1,Q3), Wilcoxon rank sum test; Categorical variables: N(%)
* Chi Square test; ** Fisher's exact test

Table 3 displays outcomes of interest. Methodology is the same as in previous tables. Values of rejection other than "No" were grouped as "Yes" ("Yes - cell mediated" and "Yes - Ab mediated"). At the 0.05 significance level, plasmapheresis and time to extubation are significant. Therefore, plasmapheresis and

time to extubation significantly differ between PRA- and PRA+ groups. ICU length of stay is marginally significant.

2 Blood product exposures

2.1 Binary blood product exposures

Now we investigate the relationship between PRA development and blood product exposures prior to transplantation. First, we look at the relationship between those who do and do not develop PRAs. Blood products are divided into four components: RBCs, FFP, platelets, and cryo. The data present blood product exposures as counts, so new variables were created to indicate whether or not that blood product was present in each patient.

Each table presents counts of each combination of blood product exposure and PRA status, as well as a column percentage. This column percentage states the percentage of patients within each PRA status that had each blood product exposure. For example, in Table 4, 37.1% of the PRA- patients had no previous blood product exposures. Comparatively, 6.9% of PRA+ patients had no previous blood product exposures. This could be changed to a row percentage depending on clinical interest. A row percentage would display, for example, the percentage of those without blood product exposures who were and were not PRA+.

Each table is statistically evaluated with either a chi square test or Fisher's exact test, depending on expected counts. Significance is evaluated at the 0.05 level.

Table 4: Previous blood product exposures

	PRA-	PRA+	Total
No previous blood product exposures	13 (37.1)	2 (6.9)	15
Previous blood product exposures	22 (62.9)	27 (93.1)	49
Total	35	29	64

Note:
p = <0.01 (Chi Square)

In table 4, we see that 62.9% of PRA- patients had previous blood product exposures, compared to 93.1% of PRA+ patients. There is a significant association between total prior blood product exposure and PRA status ($p < 0.01$).

Table 5: Previous RBC exposures

	PRA-	PRA+	Total
No previous RBC	13 (37.1)	3 (10.3)	16
Previous RBC	22 (62.9)	26 (89.7)	48
Total	35	29	64

Note:
p = 0.01 (Chi Square)

62.9% of PRA- patients had prior RBC exposures, whereas 89.7% of PRA+ patients had prior RBC exposures. Ther is a significant association between RBC exposure and PRA status ($p = 0.01$).

48.6% of PRA- patients had previous platelet exposures compared to 62.1% of PRA+ patients. There is not a significant association between prior platelet exposures and PRA status.

51.4% of PRA- patients had prior FFP exposures, and 58.6% of PRA+ patients had prior FFP exposures. There is not a significant association between FFP exposure and PRA status.

48.6% of PRA- patients had prior cryo exposures, and 58.6% of PRA+ patients had prior cryo exposures. There is not a significant association between cryo exposure and PRA status.

Table 6: Previous platelet exposures

	PRA-	PRA+	Total
No previous platelets	18 (51.4)	11 (37.9)	29
Previous platelets	17 (48.6)	18 (62.1)	35
Total	35	29	64

Note:

p = 0.28 (Chi Square)

Table 7: Previous FFP exposures

	PRA-	PRA+	Total
No previous FFP	17 (48.6)	12 (41.4)	29
Previous FFP	18 (51.4)	17 (58.6)	35
Total	35	29	64

Note:

p = 0.57 (Chi Square)

Table 8: Previous Cryo exposure

	PRA-	PRA+	Total
No previous cryo	18 (51.4)	12 (41.4)	30
Previous cryo	17 (48.6)	17 (58.6)	34
Total	35	29	64

Note:

p = 0.42 (Chi Square)

Table 9: Previous cellular (RBC,Platelet) exposure

	PRA-	PRA+	Total
No previous cellular	13 (37.1)	2 (6.9)	15
Previous cellular	22 (62.9)	27 (93.1)	49
Total	35	29	64

Note:

p = <0.01 (Chi Square)

Blood products were grouped into cellular (RBC, platelet) and acellular (FFP, cryo). 62.9% of PRA- patients had prior cellular exposures, and 93.1% of PRA+ patients had prior cellular exposures. This association is significant ($p < 0.01$). Note that these counts are the same as the total prior blood product exposure counts.

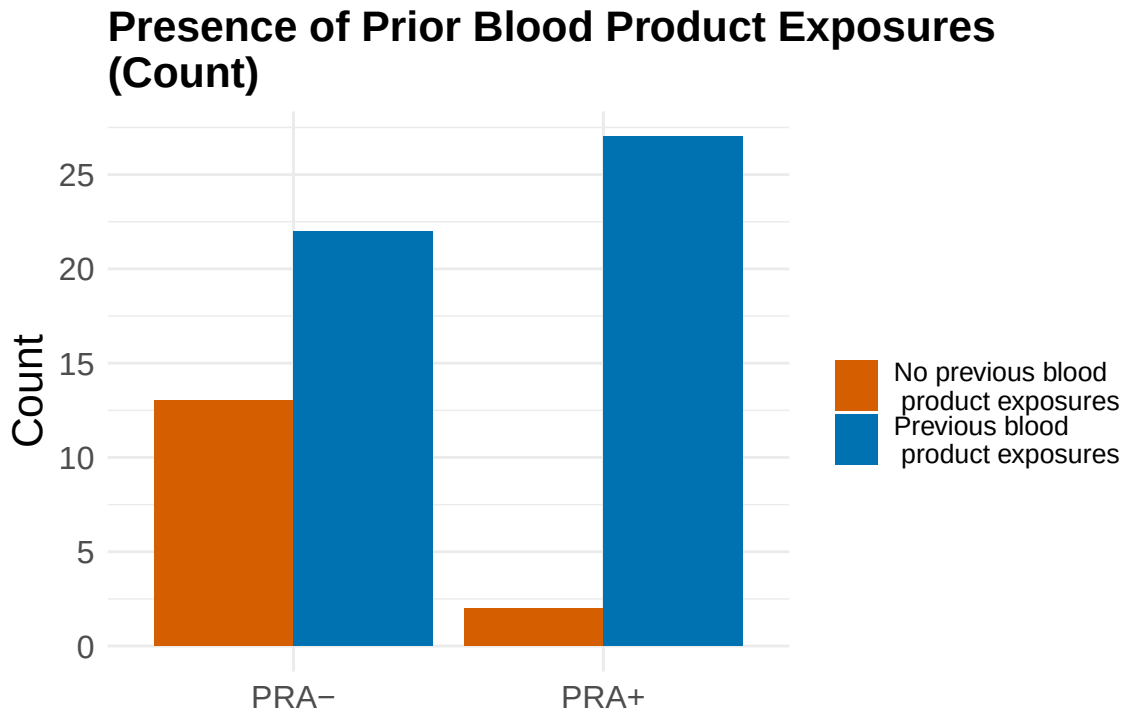
51.4% of PRA- patients had acellular exposures, compared to 72.4% of PRA+ patients. There is not a significant association between acellular exposures and PRA status.

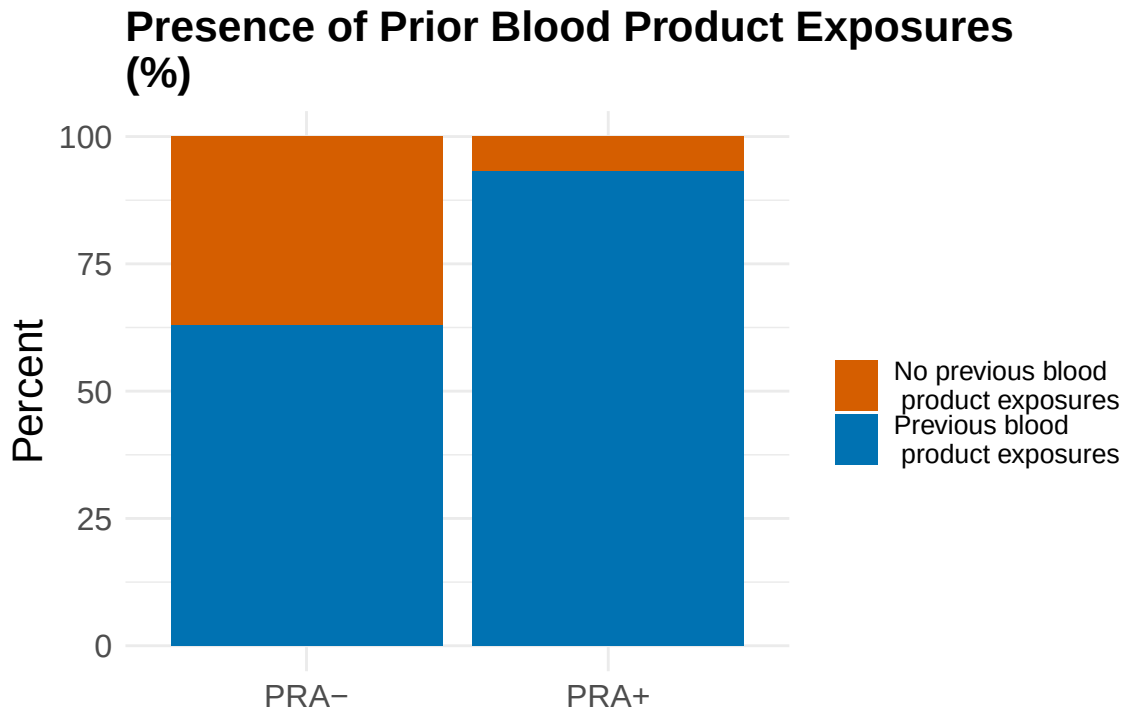
Table 10: Previous acellular (FFP, Cryo) exposure

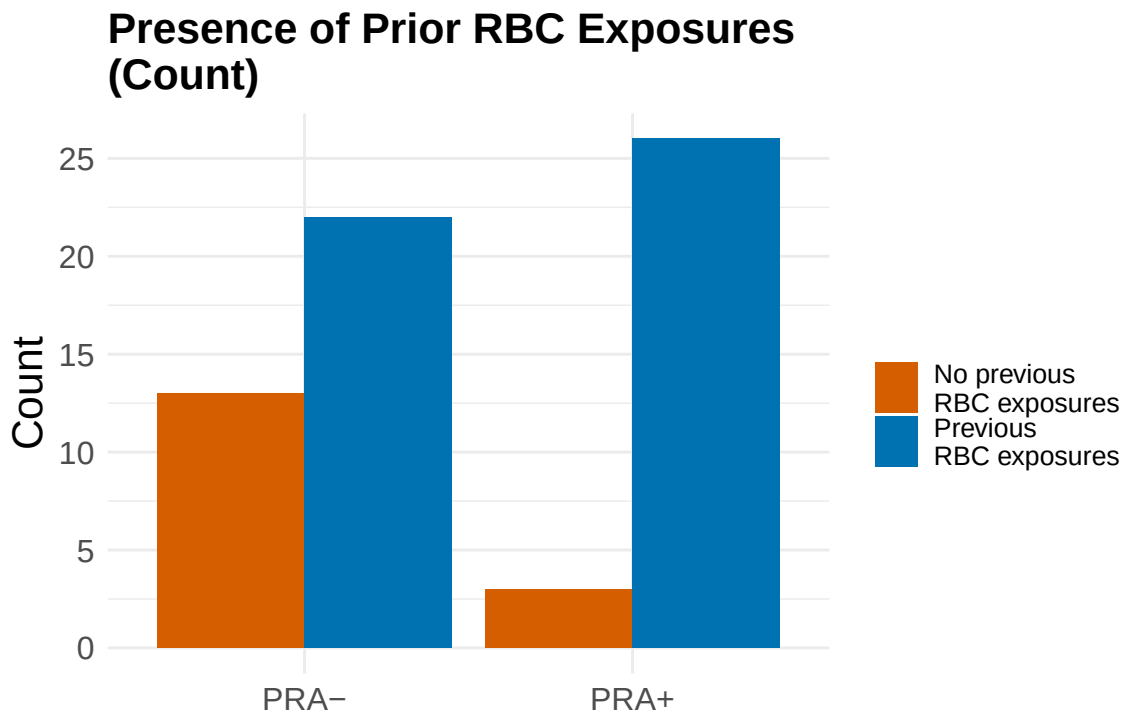
	PRA-	PRA+	Total
No previous acellular	17 (48.6)	8 (27.6)	25
Previous acellular	18 (51.4)	21 (72.4)	39
Total	35	29	64

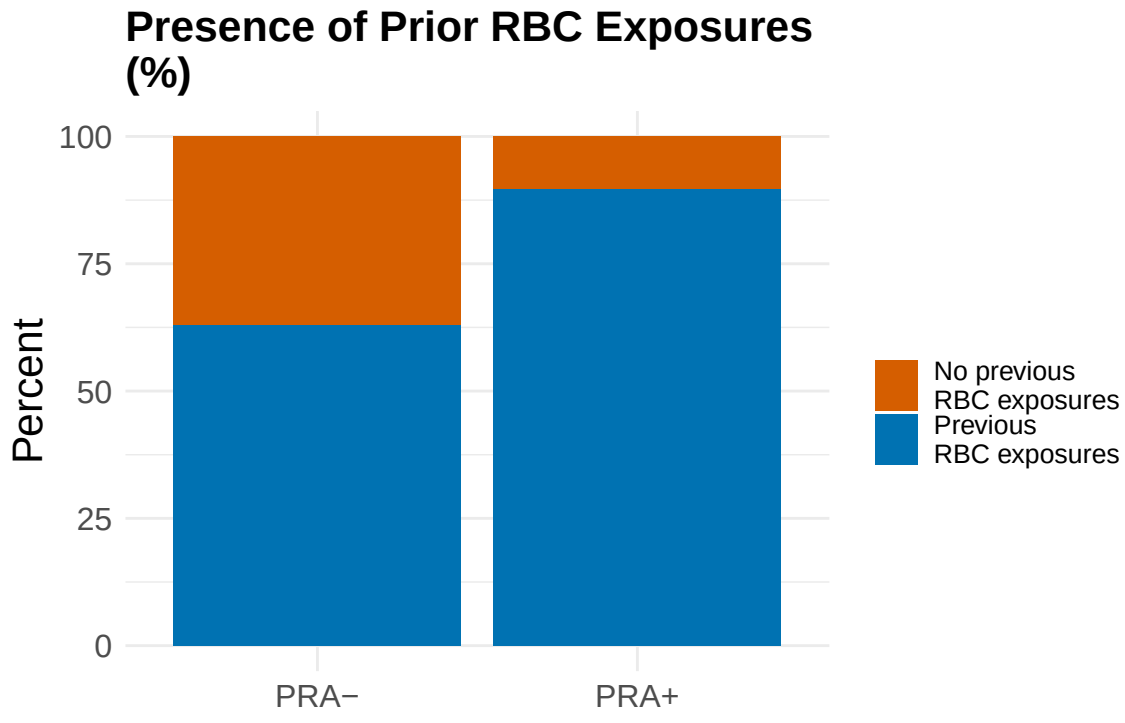
Note:
p = 0.09 (Chi Square)

Here I present graphical illustrations of tables 4 and 5. The plots are presented as both counts and percentages to reflect the information in the tables. For example, in the second plot, we see that 62.9% of PRA- patients had prior blood product exposures, whereas 93.1% of PRA+ patients had prior blood product exposures.






Presence of Prior RBC Exposures (Count)
Count
25
20
15
10
5
0
PRA−
PRA+
No previous RBC exposures
Previous RBC exposures


Presence of Prior RBC Exposures (%)
Percent
100
75
50
25
0
PRA−
PRA+
No previous RBC exposures
Previous RBC exposures

2.2 Blood product exposure categories

For this section, a new variable was created to indicate the number of distinct blood product exposures for each patient. For example, a patient who had a positive count of exposures for RBC and platelets but no FFP or cryo exposures would have 2 distinct exposures. This could be any two exposures, so a patient exposed to only FFP and platelets would also have a value of 2.

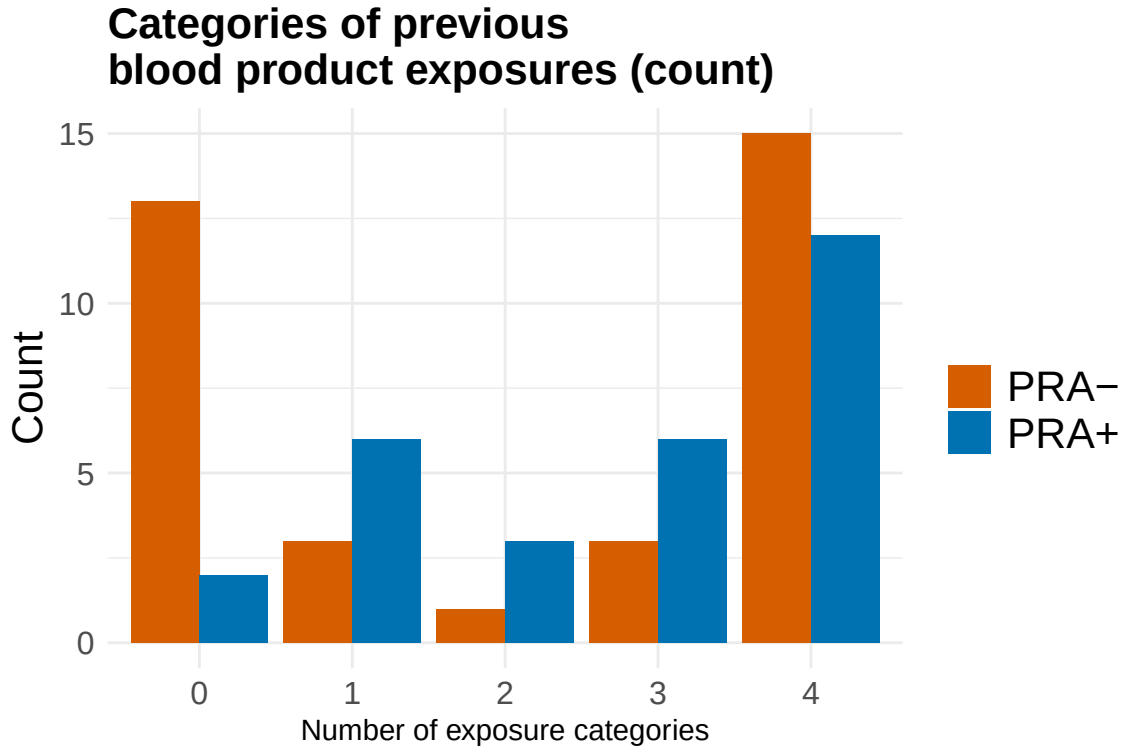
Table 11: Categories of previous blood product exposures

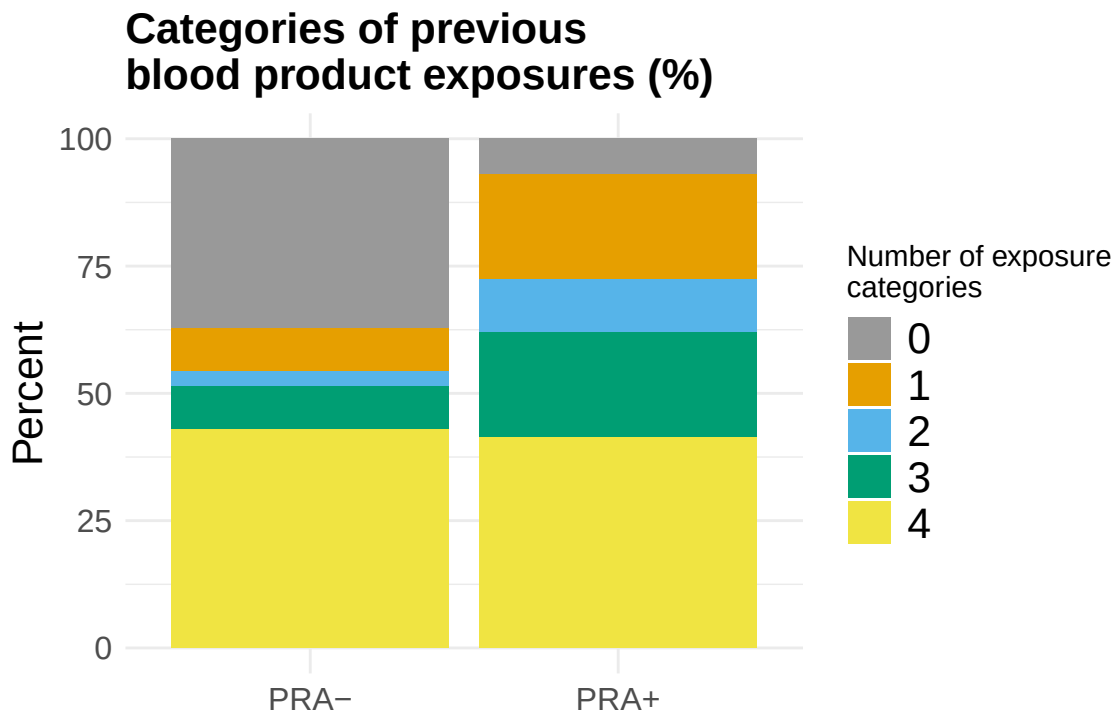
	PRA-	PRA+	Total
0 exposures	13 (37.1)	2 (6.9)	15
1	3 (8.6)	6 (20.7)	9
2	1 (2.9)	3 (10.3)	4
3	3 (8.6)	6 (20.7)	9
4	15 (42.9)	12 (41.4)	27
Total	35	29	64

Note:
p = 0.02 (Fisher's Exact)

In table 11, we see both counts and column percentages of number of distinct exposures for PRA- and PRA+ patients. For example, 42.9% of PRA- patients were exposed to all 4 blood product categories, while 41.4% of PRA+ patients were exposed to all 4 categories. There is a significant association between number of distinct exposures and PRA status ($p = 0.02$).

The figures below display these counts and percentages graphically. The first figure shows the counts of each number of distinct exposures for PRA- and PRA+ patients. The second figure displays the percentages of each number of distinct exposures for PRA- and PRA+ patients.





Here we see that while approximately the same percentage of PRA- and PRA+ patients are exposed to all four categories, PRA+ patients contain a larger percentage of 1, 2, and 3 categories and a lower percentage of 0 categories.

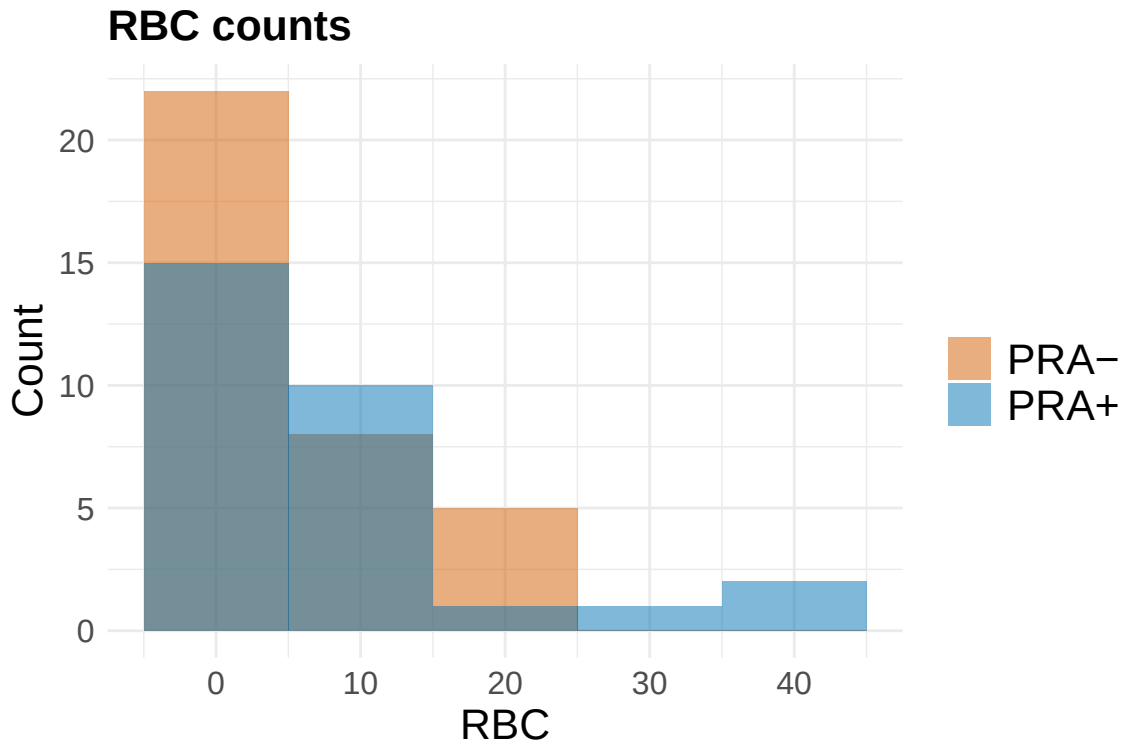
2.3 Blood product exposure counts

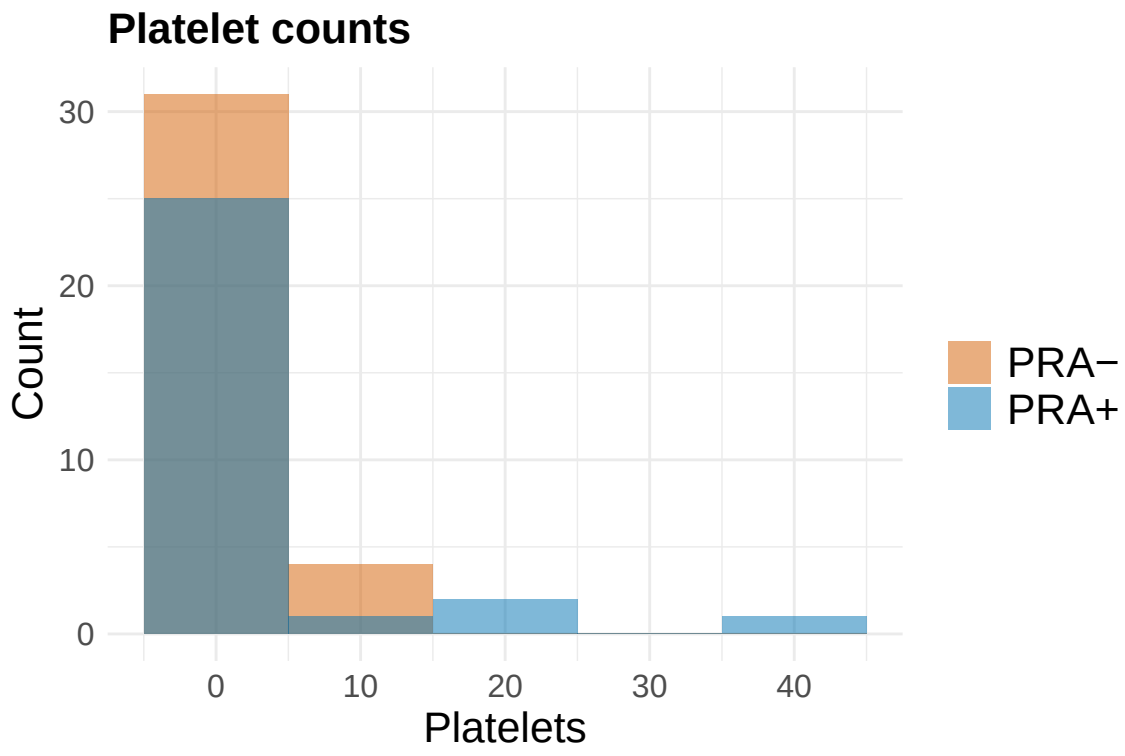
Now we look at **counts** of blood product exposures and the relationship with PRA development. We treat PRA development as binary (PRA+/-). This table was not included in the previous results and none of the exposure categories are statistically significant at the 0.05 level. However, I wanted to include the table as well as the overlapping histograms in case they might be clinically meaningful.

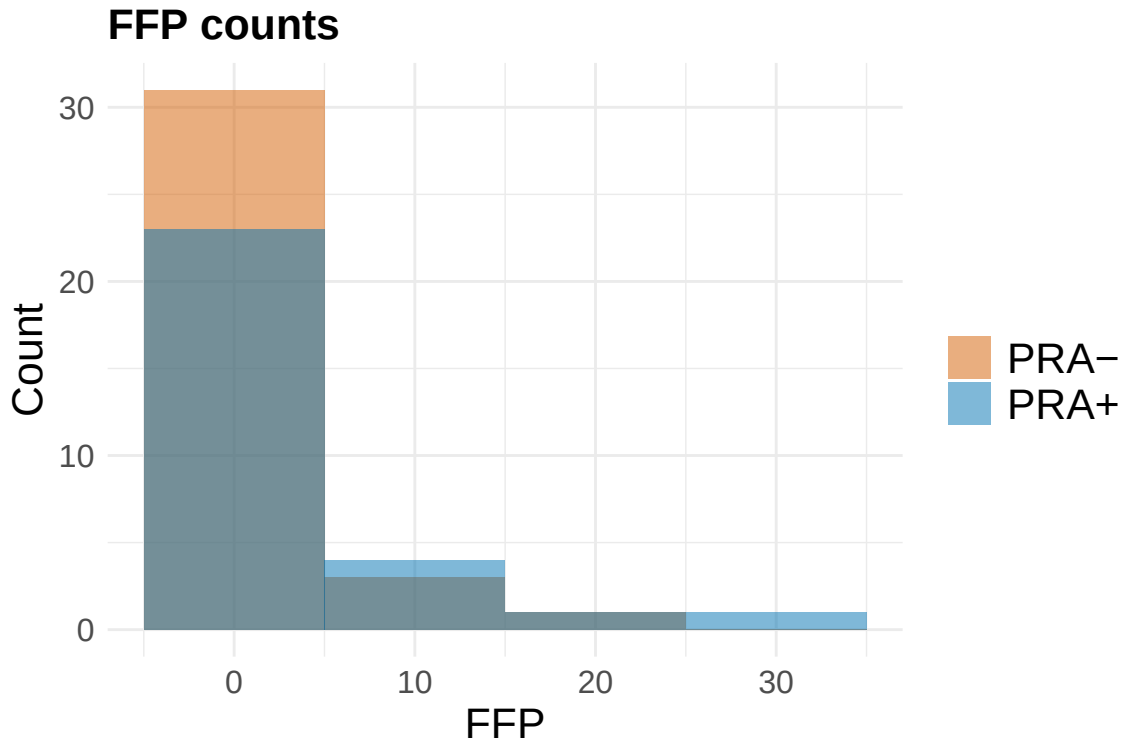
Table 12: Previous blood product exposure categories

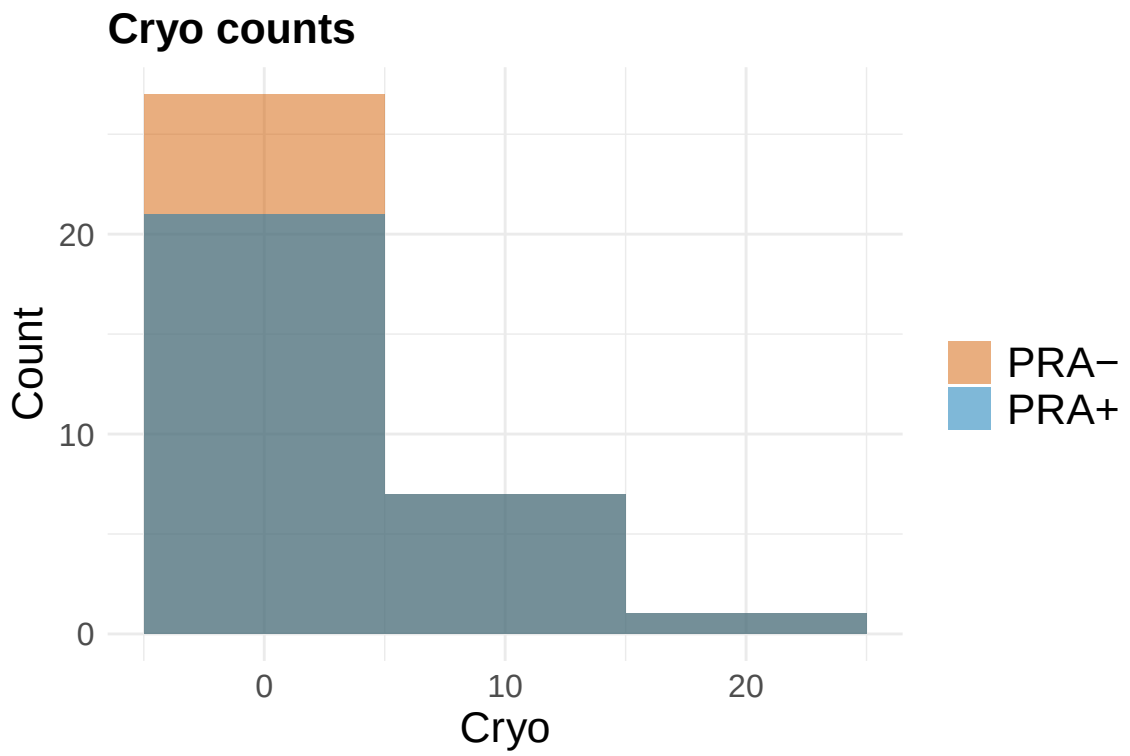
	All patients (n=64)	PRA- (n=35)	PRA+ (n=29)	p value
RBCs	3.0 (0.8,9.2)	3.0 (0.0,8.5)	4.0 (2.0,9.0)	0.15
Platelets	1.0 (0.0,3.0)	0.0 (0.0,3.0)	1.0 (0.0,3.0)	0.30
FFP	1.0 (0.0,4.0)	1.0 (0.0,3.5)	2.0 (0.0,4.0)	0.48
Cryo	1.5 (0.0,5.2)	0.0 (0.0,5.0)	3.0 (0.0,6.0)	0.24

Note:
Median (Q1,Q3), Wilcoxon rank sum test




Platelet counts
30
20
10
0
Count
0
10
20
30
40
Platelets
PRA−
PRA+


FFP counts
30
20
10
0
Count
0
10
20
30
FFP
PRA−
PRA+


Cryo counts
Count
20
10
0
0
10
20
Cryo
PRA−
PRA+

3 Blood product counts and cPRA

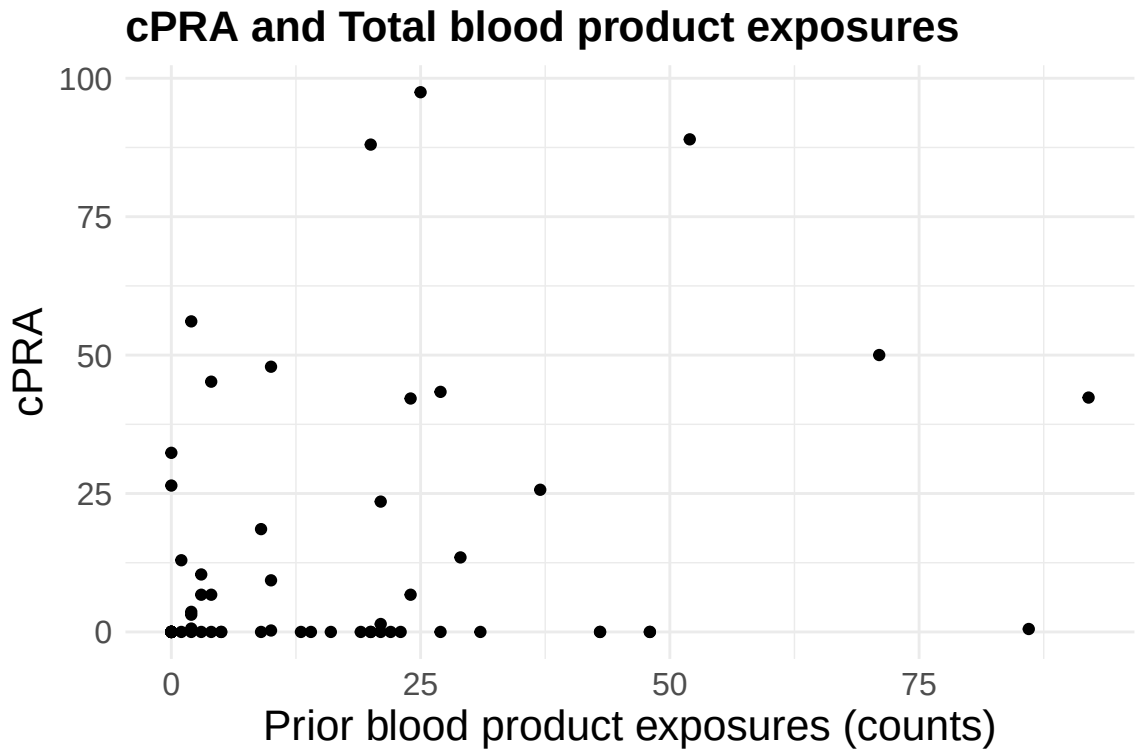
Now we look at counts of blood product exposure and cPRA. Therefore, we are treating both variables as continuous. All subjects are still included, so for PRA- patients, cPRA=0. One observation is excluded from these analyses because its cPRA value is missing, so N=63.

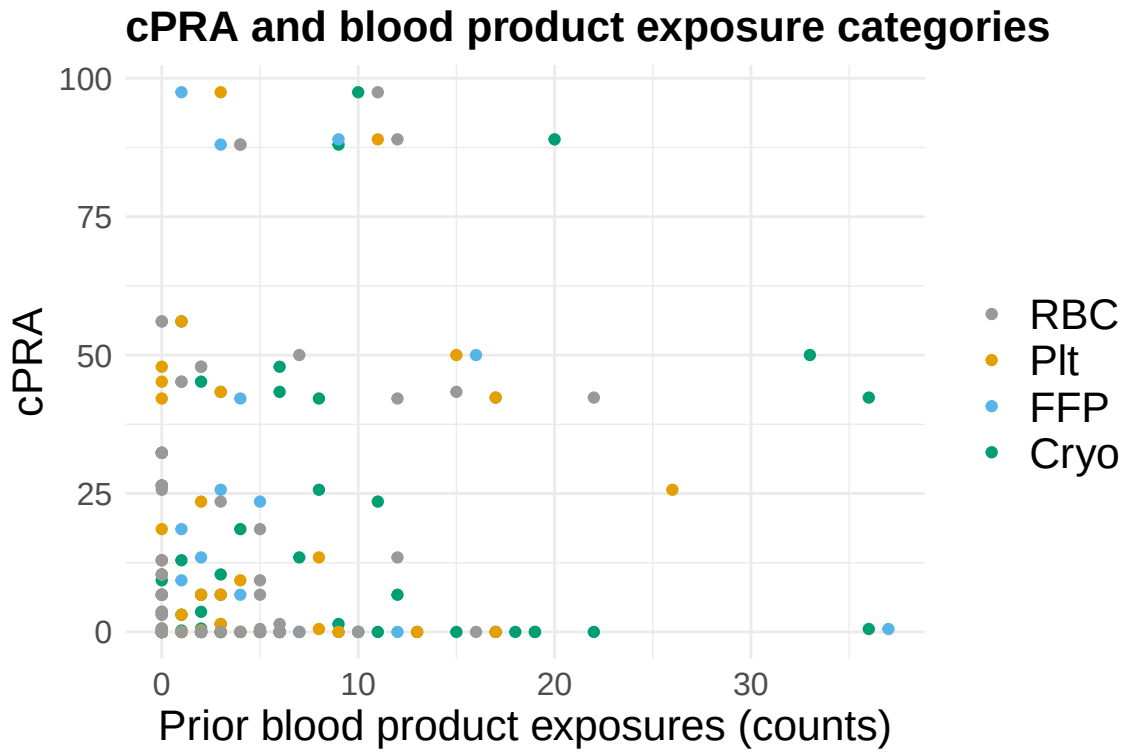
Table 13: Blood products and cPRA correlations

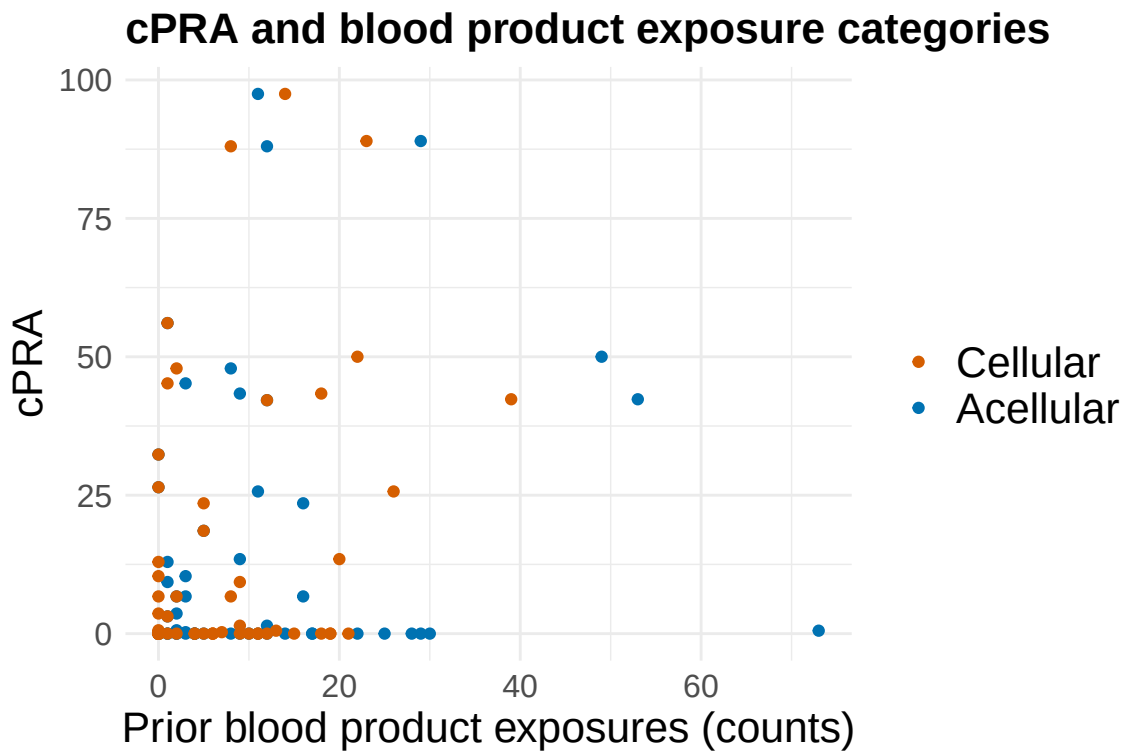
	Spearman rank correlation	p value
Total prior exposures	0.27	0.03
RBC	0.23	0.07
platelets	0.20	0.12
FFP	0.14	0.28
Cryo	0.23	0.07
Cellular	0.23	0.07
Acellular	0.28	0.03

The nonparametric Spearman rank correlation is used because of non-normality. Total prior exposures and acellular exposures are statistically significant, while RBC, cryo, and cellular exposures are marginally significant. These results align with those in section 2.1, with the exception of acellular exposures.

Below, we present scatter plots of blood product exposure counts and cPRA. First, the total blood product exposure counts are presented. The second and third plots differentiate by blood product exposure categories.




cPRA and blood product exposure categories
100
75
50
25
0
cPRA
0
10
20
30
Prior blood product exposures (counts)
RBC
Plt
FFP
Cryo


cPRA and blood product exposure categories
100
75
50
25
0
cPRA
0
20
40
60
Prior blood product exposures (counts)
Cellular
Acellular

4 Secondary outcomes of interest

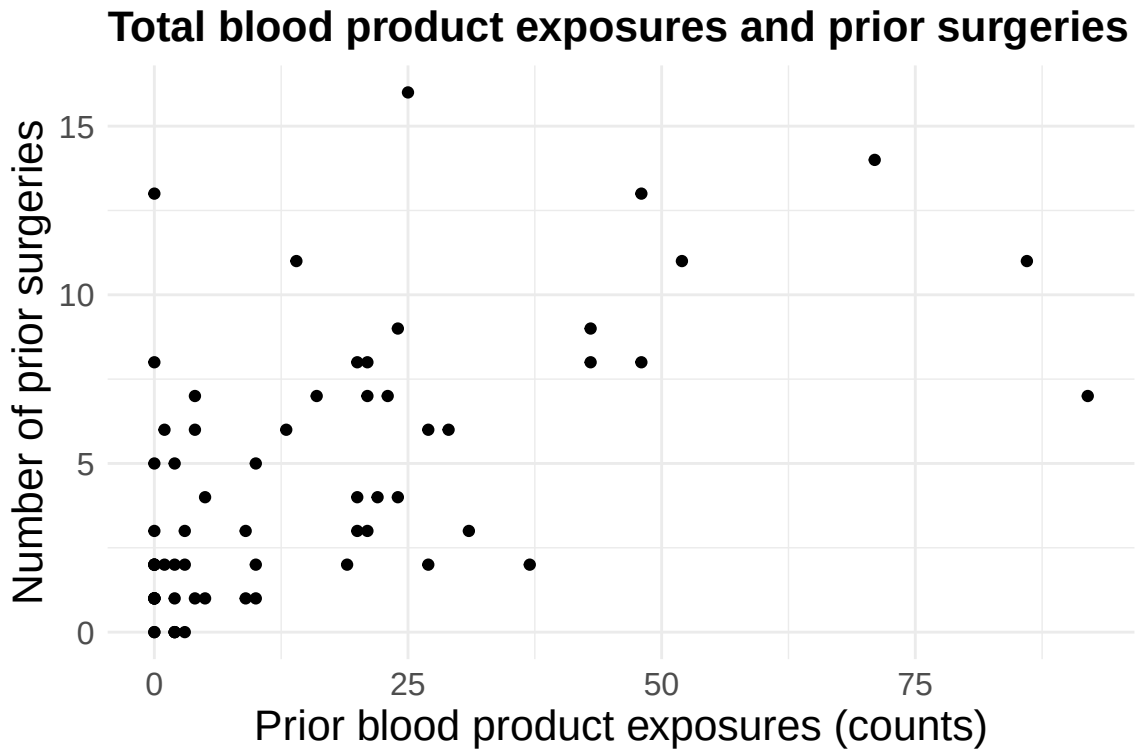
This section explores secondary outcomes of interest, including VIS, previous surgeries, ICU length of stay, and time to extubation.

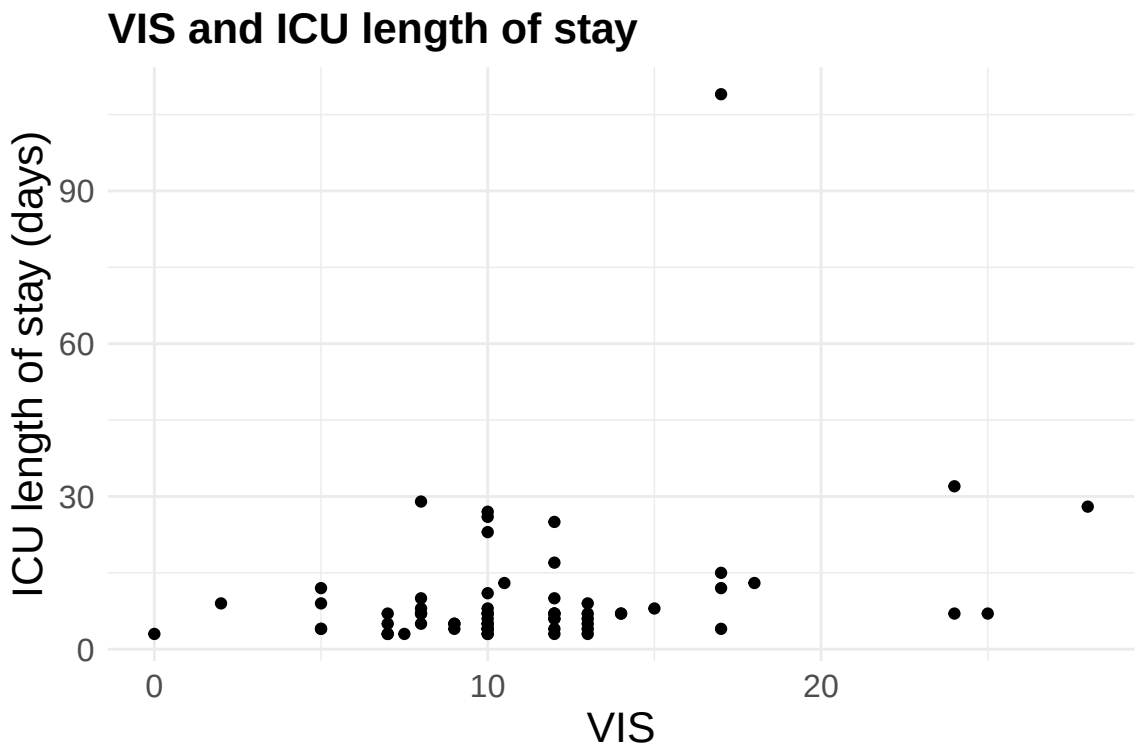
Table 14: Secondary correlations of interest

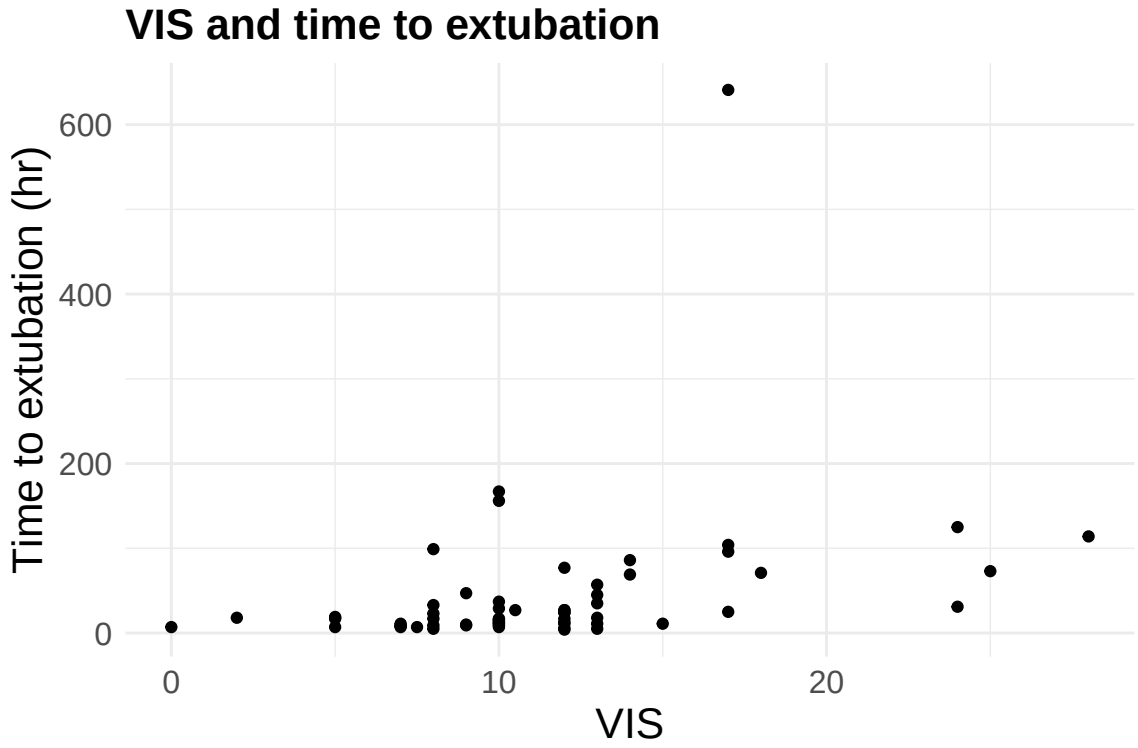
	Spearman rank correlation	p value
Previous surgeries, total prior blood product exposures	0.59	<0.01
VIS, ICU length of stay	0.29	0.02
VIS, time to extubation	0.48	<0.01
ICU length of stay, time to extubation	0.68	<0.01

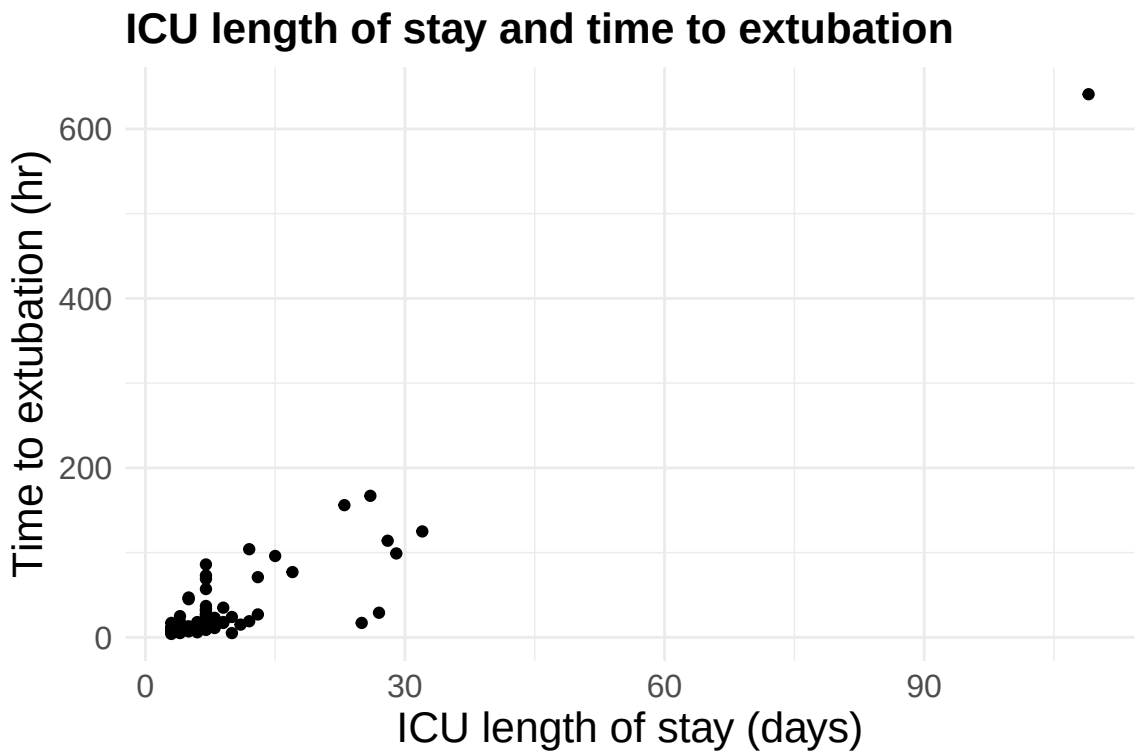
Table 14 shows correlations among secondary outcomes of interest. All of these correlations are statistically significant. Therefore, patients who have more blood product exposures are also likely to have had more previous surgeries, those with a higher VIS are likely to have had a longer ICU stay and longer time to extubation, and those with a longer ICU stay are likely to also have a longer time to extubation.

Below we present scatter plots of these linear relationships.




VIS and ICU length of stay
ICU length of stay (days)
90
60
30
0
0
10
20
VIS


VIS and time to extubation
Time to extubation (hr)
600
400
200
0
0
10
20
VIS

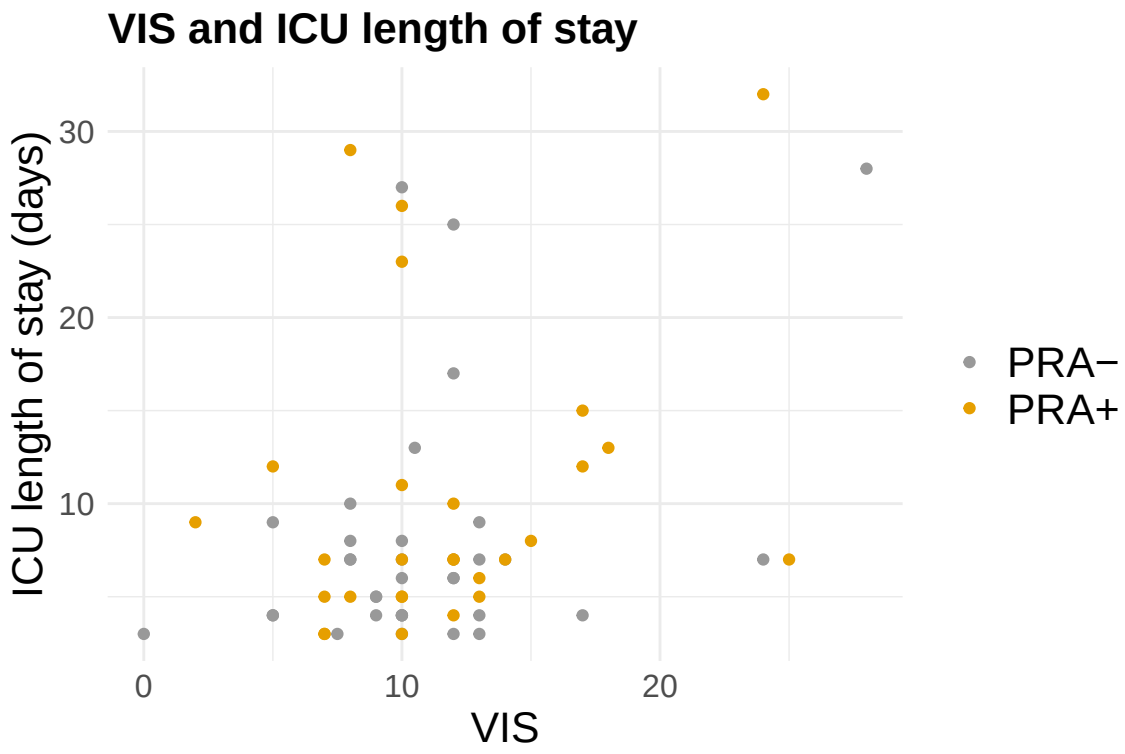


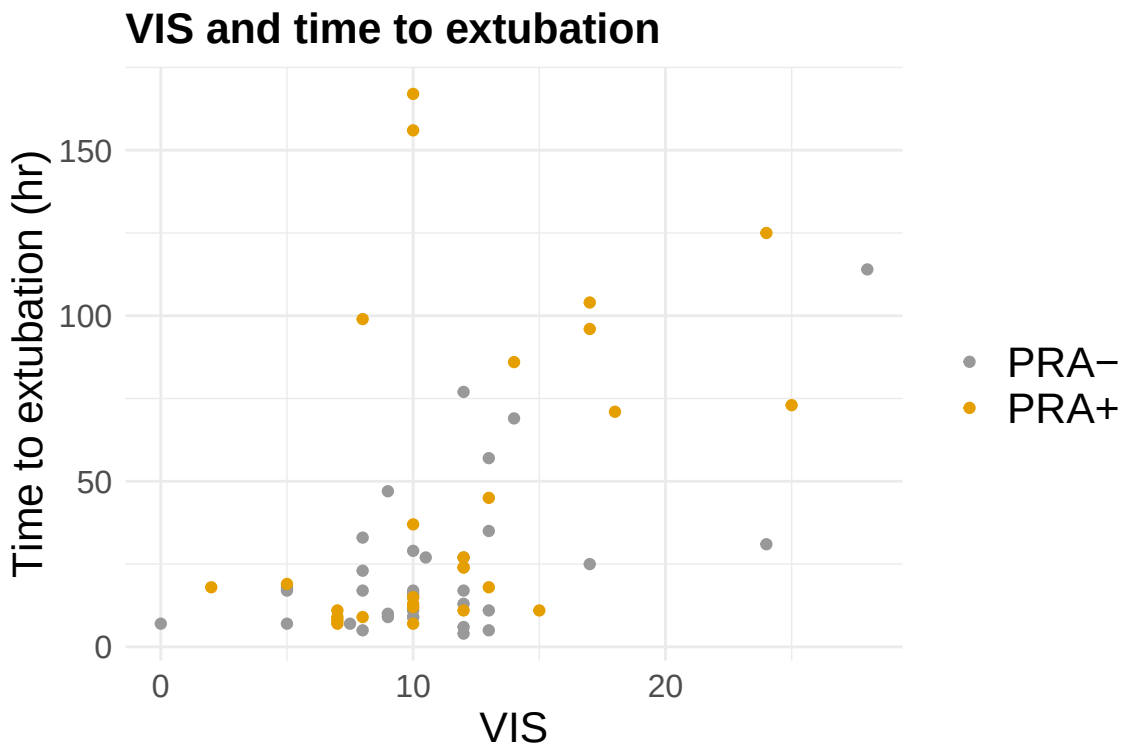
We see that there is an outlier in these variables, namely one patient who experienced a long ICU stay and time to extubation. I removed this outlier from the datset to see if these secondary relationships were strongly influenced by this single data point. The following table and plots present the same relationships but without this outlier and differentiated by PRA status.

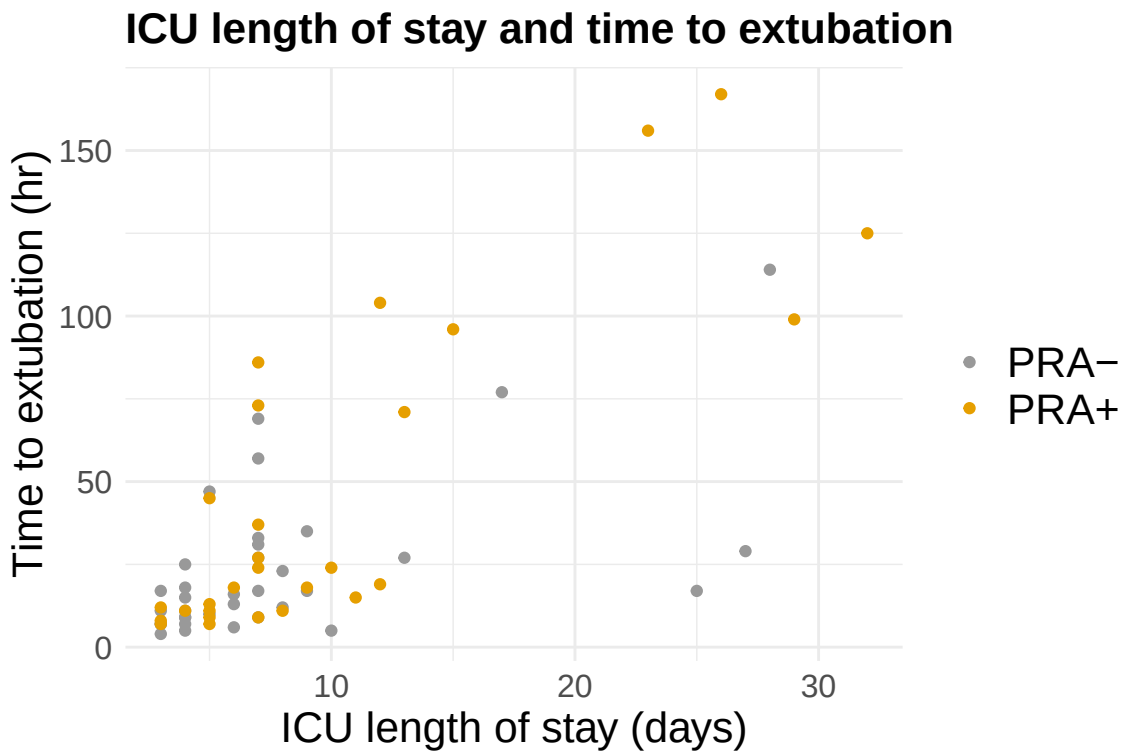
We see that the relationships continue to be statistically significant without the outlier.

Table 15: Secondary correlations of interest

	Spearman rank correlation	p value
VIS, ICU length of stay	0.26	0.04
VIS, time to extubation	0.46	<0.01
ICU length of stay, time to extubation	0.66	<0.01


VIS and ICU length of stay
ICU length of stay (days)
30
20
10
0
10
20
VIS
PRA−
PRA+


VIS and time to extubation
Time to extubation (hr)
150
100
50
0
0
10
20
VIS
PRA−
PRA+


ICU length of stay and time to extubation
Time to extubation (hr)
150
100
50
0
10
20
30
ICU length of stay (days)
PRA−
PRA+

The following tables present other relationships of interest among categorical variables.

Table 16: Allele class and rejection

	No rejection	Rejection	Total
Class I	15 (65.2)	3 (50.0)	18
Class II	6 (26.1)	1 (16.7)	7
Class I and II	2 (8.7)	2 (33.3)	4
Total	23	6	29

Note:
p = 0.41 (Fisher's Exact)

Table 16 includes only PRA+ patients and shows those who have class I, class II, or both class I and II alleles. There is not a significant association between allele class and rejection status.

Table 17: Allele class and donor specific antibody

	No donor specific antibody	Donor specific antibody	Total
Class I	10 (100.0)	8 (42.1)	18
Class II	0 (0.0)	7 (36.8)	7
Class I and II	0 (0.0)	4 (21.1)	4
Total	10	19	29

Note:
p = 0.01 (Fisher's Exact)

Table 17 includes only PRA+ patients and explores the relationship between allele class and development of a donor specific antibody. 100% of those who did not develop a donor specific antibody had a class I allele only, while those who did develop a donor specific antibody were fairly evenly spread among class I, class II, or both. There is a significant relationship between allele class and development of a donor specific antibody ($p = 0.01$).

Table 18: Rejection and PRA status

	PRA-	PRA+	Total
No rejection	32 (91.4)	23 (79.3)	55
Rejection	3 (8.6)	6 (20.7)	9
Total	35	29	64

Note:
p = 0.28 (Fisher's Exact)

Table 18 explores the relationship between rejection status and PRA status. 8.6% of PRA- patients had a positive rejection status, while 20.7% of PRA+ patients had a positive rejection status. This relationship is not statistically significant.

Table 19 explores the relationship between rejection status and developing a donor specific antibody. 6.5% of those who did not develop a donor specific antibody had a positive rejection status, while 21.2% of those who did develop a donor specific antibody had a positive rejection status. This relationship is not statistically significant.

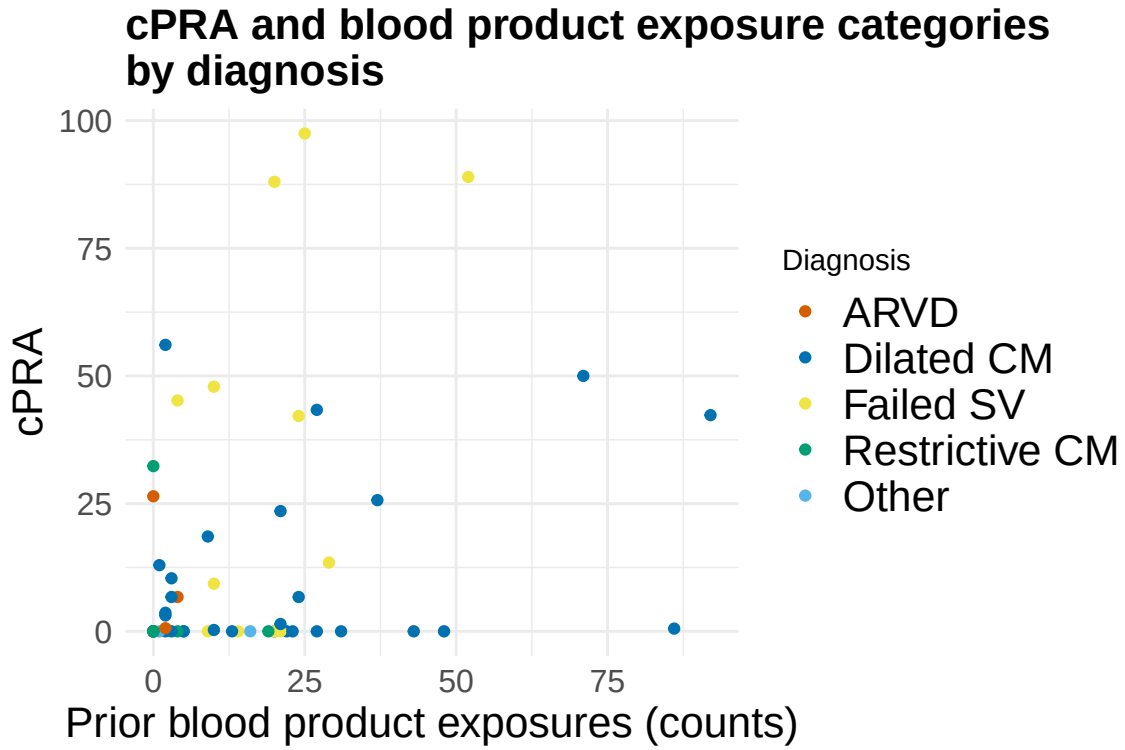
Table 19: Rejection and donor specific antibody

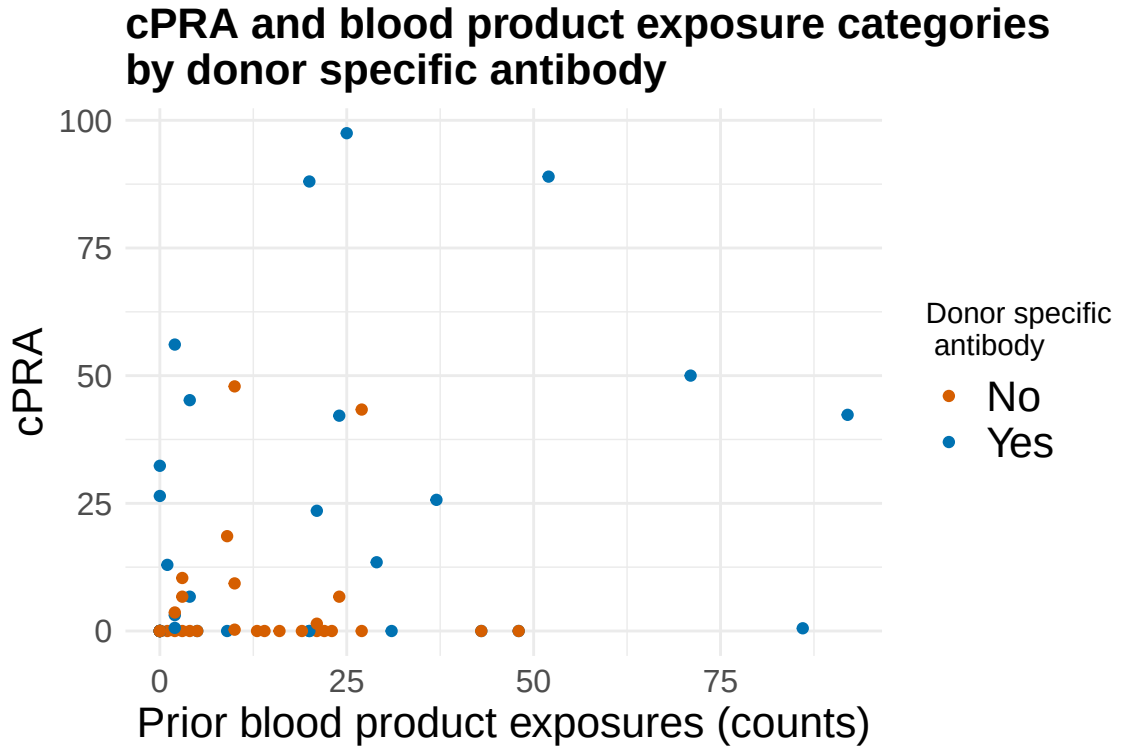
	No donor specific antibody	Donor specific antibody	Total
No rejection	29 (93.5)	26 (78.8)	55
Rejection	2 (6.5)	7 (21.2)	9
Total	31	33	64

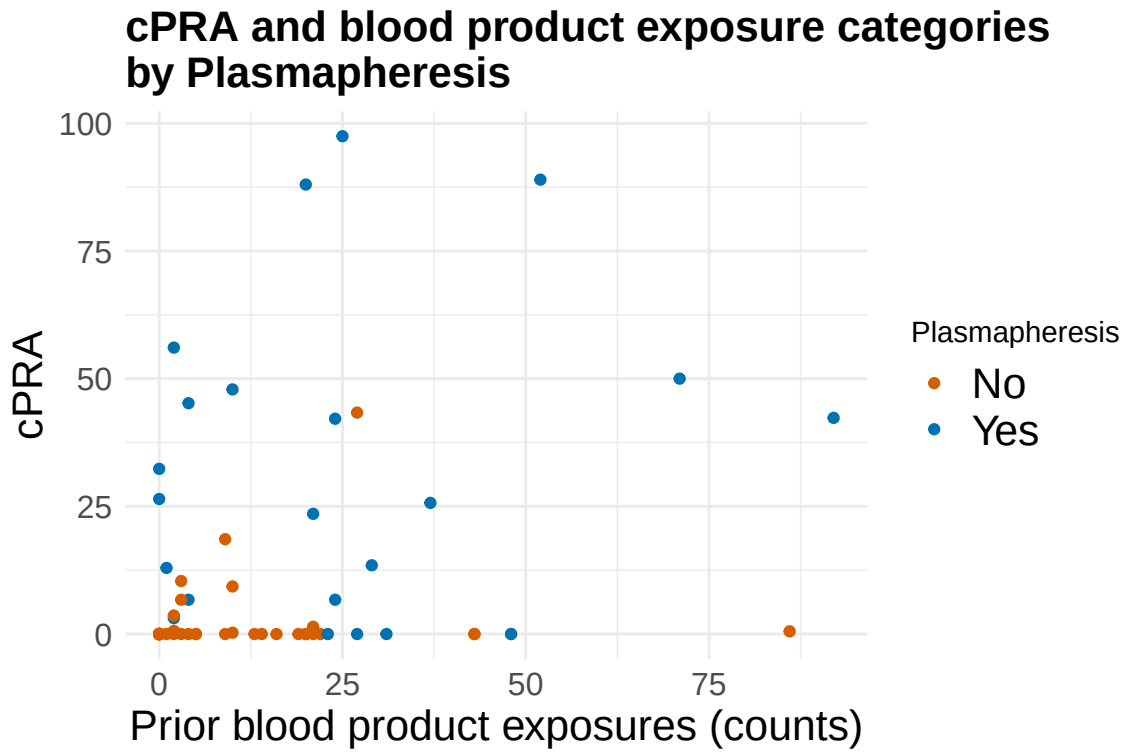
Note:
p = 0.15 (Fisher's Exact)

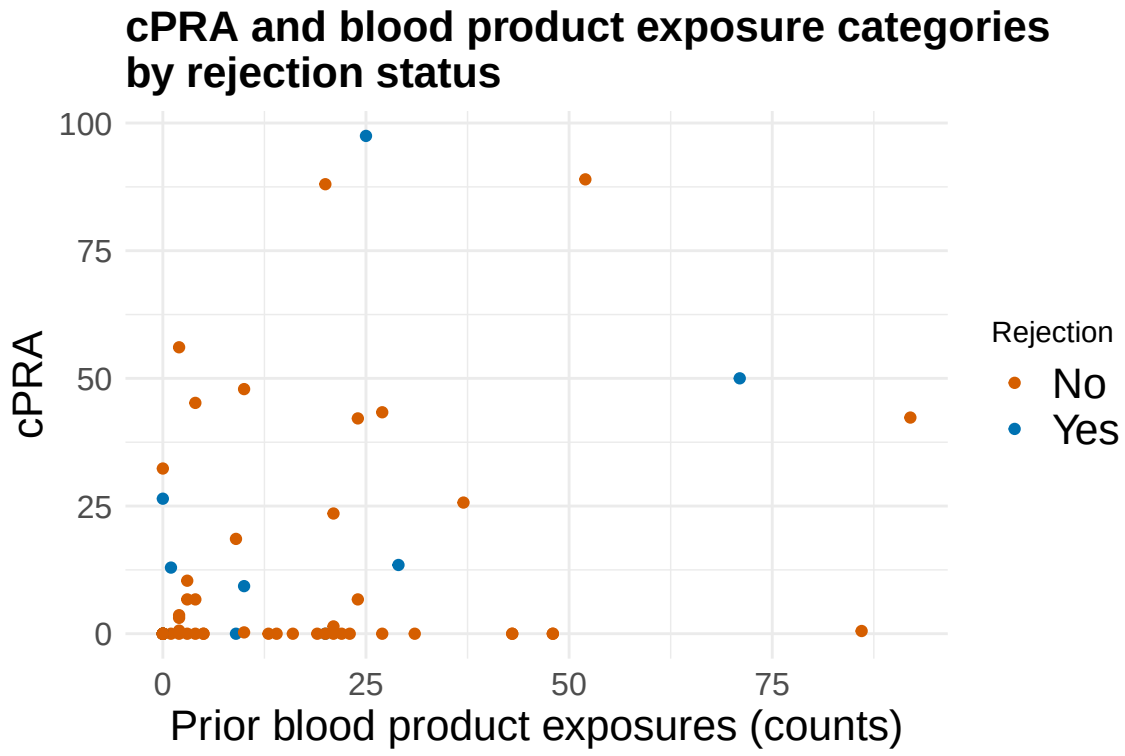
5 Other figures of (possible) interest

Here I present scatter plots of cPRA and total blood product exposures, but differentiated by other variables of interest. The variables of interest were based on the significant or nearly significant variables from the "Study Objectives" and "Outcomes" tables.






cPRA and blood product exposure categories by Plasmapheresis
cPRA
100
75
50
25
0
0
25
50
75
Prior blood product exposures (counts)
Plasmapheresis
No
Yes


cPRA and blood product exposure categories by rejection status
cPRA
100
75
50
25
0
0
25
50
75
Prior blood product exposures (counts)
Rejection
No
Yes