## TAC linearity summary

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11/12/22

```
rm(list = ls())
Load libraries
```

```
library(ggplot2)
library(ggpmisc)
library(plyr)
library(dplyr)
```

Import data file

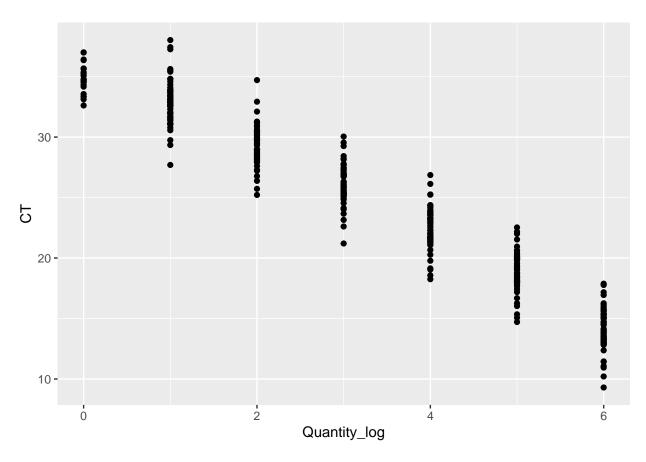
#clear R env

```
TAC<-read.csv("sample_data.csv", na.strings=c("","NA"))
```

Plot a standard curve and summarize efficiency for each target

```
#Format data and add column with logs of quantities
TAC$CT<-as.numeric(gsub(",","",TAC$CT))</pre>
TAC$Quantity<-as.numeric(gsub(",","",TAC$Quantity))</pre>
TAC$Quantity_log<-log10(TAC$Quantity)</pre>
#summarize efficiencies
TAC<-TAC[!is.na(TAC$CT),]
TAC<-TAC[!is.na(TAC$Quantity_log),]</pre>
TAC<-TAC[rowSums(is.na(TAC)) != ncol(TAC), ]
efficiency_summ<-ddply(TAC, "Target.Name", function(x) {
 model <- lm(CT ~ Quantity_log, data = x)</pre>
  coef(model)})
colnames(efficiency_summ) <- c("Target.Name", "Y.intercept", "Slope")</pre>
efficiency_summ$Efficiency<-10^(-1/efficiency_summ$Slope)-1
#create a list of dataframes by target
targets=by(TAC, TAC[,"Target.Name"], function(x) x)
#plot of standard curve points across all targets
```

```
p<-ggplot(TAC, aes(x = Quantity_log, y = CT)) +
  geom_point()
p</pre>
```



```
#plot standard curves for all targets
target_plots <- function(targets){</pre>
  ggplot(targets, aes(x = Quantity_log, y = CT)) +
    geom_point()+
    stat_poly_line(formula = y~x, se=FALSE) +
    stat_poly_eq(aes(label = paste(after_stat(eq.label), after_stat(rr.label), sep = "*\", \"*")), form
q1 <- lapply(targets, target_plots)</pre>
q2 <- lapply(seq_along(q1), function(i) {</pre>
 q1[[i]] + ggtitle(names(targets)[i])
})
#q2 #careful--this command makes a lot of plots!
#faceted plot with all adjusted standard curves for all targets
r<-ggplot(TAC, aes(x = Quantity_log, y = CT)) +
  geom_point() +
 facet_wrap(~ Target.Name)+
  stat_poly_line(formula = y~x, se=FALSE) +
```

```
stat_poly_eq(aes(label = paste(after_stat(eq.label), after_stat(rr.label), sep = "*\", \"*")), formul
r
```

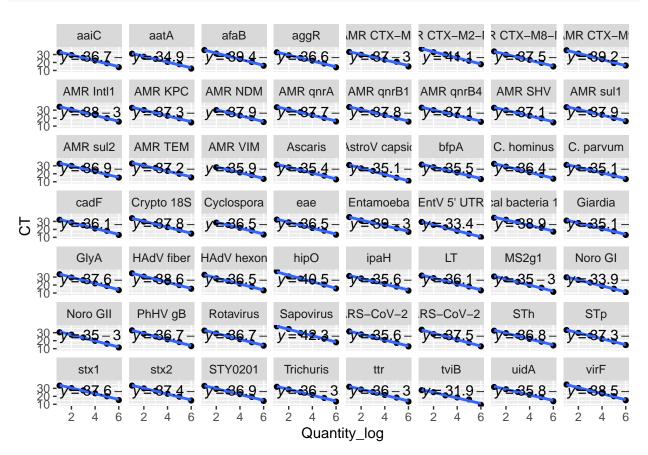
aaiC 40 - 28 - <b>y ≃ 36</b> . 7 -	aatA <b>y'≕34</b> .9 –	afaB y = 39.4 -	33			7  CTX-M8-I $y = 37.5 -$	
10 - AMR Intl1	AMR KPC y=37.3-	AMR NDM	AMR qnrA	AMR qnrB1	AMR qnrB4	AMR SHV	AMR sul1
10 - AMR sul2 40 - 38 - <i>y</i> = 36,9 -	AMR TEM $y = 36.1 -$	AMR VIM	Ascaris	AstroV capsic	bfpA	C. hominus	C. parvum
cadF		Cyclospora	eae	Entamoeba	EntV 5' UTR		Giardia
GlyA		HAdV hexon	hipO	ipaH	LT	MS2g1	Noro GI
Noro GII	PhHV gB y = 36.7 -	Rotavirus	Sapovirus	RS-CoV-2	RS-CoV-2	STh	STp
stx1	stx2 $y = 37 - 3$	STY0201	Trichuris	ttr	tviB	uidA	virF
0 2 4 6	0 2 4 6	0 2 4 6	0 2 4 6	0 2 4 6 ity_log	0 2 4 6	0 2 4 6	0 2 4 6

Plot standard curves and summarize efficiencies without last point in the curve (where it flattens out as it approaches the LOD)

```
#remove last point on standard curve
TAC_adj <- TAC[TAC$Quantity_log != 0, ]</pre>
\#TAC\_adj \leftarrow TAC[TAC\$Quantity\_log != 0, ] \#run this to look at standard curves without last two dilution
#create a list of dataframes by target
targets_adj=by(TAC_adj, TAC_adj[,"Target.Name"], function(x) x)
#plot adjusted standard curves for all targets
target_plots_adj <- function(targets_adj){</pre>
  ggplot(targets\_adj, aes(x = Quantity\_log, y = CT)) +
    geom_point()+
    stat_poly_line(formula = y~x, se=FALSE) +
    stat_poly_eq(aes(label = paste(after_stat(eq.label), after_stat(rr.label), sep = "*\", \"*")), form
  }
s1 <- lapply(targets_adj, target_plots_adj)</pre>
s2 <- lapply(seq_along(s1), function(i) {</pre>
  s1[[i]] + ggtitle(names(targets_adj)[i])
})
```

```
#s2 #careful-this command makes a lot of plots!

#faceted plot with all adjusted standard curves for all targets
t<-ggplot(TAC_adj, aes(x = Quantity_log, y = CT)) +
    geom_point() +
    facet_wrap(~ Target.Name)+
    stat_poly_line(formula = y~x, se=FALSE) +
    stat_poly_eq(aes(label = paste(after_stat(eq.label), after_stat(rr.label), sep = "*\", \"*")), formul
t</pre>
```



Summarize dilutions with detection and high and low CT values for each target FIX THIS

```
TAC_CT<-TAC %>% select(Target.Name, Sample.Name, CT, R.superscript.2.)
high_low<-TAC_CT %>%
group_by(Target.Name) %>%
mutate(
    CT_min=min(CT, na.rm=T),
    CT_max=max(CT, na.rm=T)) %>%
mutate(
    Dilution_min=min(Sample.Name, na.rm=T),
    Dilution_max=max(Sample.Name, na.rm=T)) %>%
select(-Sample.Name) %>%
select(-CT)
```

Summarize adjusted (without lowest dilution) slope, efficiency, and R2 values for each target

Summarize NTC results

```
TAC_NTC<-read.csv("sample_data.csv", na.strings=c("","NA"))

TAC_NTC <- TAC_NTC %>%
    subset(Sample.Name=="NTC")

TAC_NTC$CT<-as.numeric(gsub(",","",TAC_NTC$CT))

TAC_NTC<- TAC_NTC%>%
    select(Target.Name, CT)%>%
    dplyr::rename(NTC_CT=CT)
```

Summary table

```
summary<-merge(efficiency_summ, R2)
summary<-merge(high_low, summary)
summary<-merge(TAC_NTC, summary)
summary <- summary %>%
    distinct(.keep_all = TRUE) %>%
    select(-model) %>%
    relocate(R.superscript.2., .before=Efficiency) %>%
    relocate(NTC_CT, .before=Y.intercept)%>%
    dplyr::rename(R2=R.superscript.2.) %>%
    dplyr::rename(Y.intercept_adj='(Intercept)') %>%
    dplyr::rename(Slope_adj=Quantity_log)%>%
    mutate_if(is.numeric, round, digits=2)
```

```
##
               Target.Name CT_min CT_max Dilution_min Dilution_max NTC_CT
## 1
                        aaiC 14.07 32.61
                                                   PCP 10<sup>1</sup>
                                                                   PCP 10<sup>6</sup>
## 2
                        aatA 12.37 31.06
                                                   PCP 10<sup>1</sup>
                                                                   PCP 10<sup>6</sup>
                                                                                   NA
## 3
                        afaB 15.97 35.63
                                                   PCP 10<sup>1</sup>
                                                                   PCP 10<sup>6</sup>
                                                                                   NA
                                                                   PCP 10<sup>6</sup>
## 4
                        aggR 13.97
                                       36.36
                                                   PCP 10<sup>0</sup>
                                                                                   NA
## 5
                AMR CTX-M1 14.99 33.21
                                                   PCP 10^1
                                                                   PCP 10<sup>6</sup>
                                                                                  NA
            AMR CTX-M2-M74 17.77 37.26
                                                                   PCP 10<sup>6</sup>
## 6
                                                   PCP 10^1
                                                                                  NA
## 7
            AMR CTX-M8-M25 15.03 33.90
                                                   PCP 10^1
                                                                   PCP 10<sup>6</sup>
                                                                                  NA
## 8
                AMR CTX-M9 16.26 35.41
                                                   PCP 10^1
                                                                                  NA
                                                                   PCP 10<sup>6</sup>
## 9
                 AMR Intl1 15.62 34.34
                                                 PCP 10^1
                                                                   PCP 10<sup>6</sup>
                                                                                  NA
                    AMR KPC 14.57 32.94
## 10
                                                   PCP 10^1
                                                                   PCP 10<sup>6</sup>
                                                                                  NA
## 11
                   AMR NDM 15.10 34.74
                                                   PCP 10^0
                                                                   PCP 10<sup>6</sup>
                                                                                  NA
## 12
                 AMR qnrA 15.82 35.65
                                                   PCP 10^0
                                                                   PCP 10<sup>6</sup>
                                                                                  NA
## 13
                AMR qnrB1 15.64 33.94
                                                   PCP 10<sup>1</sup>
                                                                 PCP 10^6
                                                                                  NA
```

```
## 14
                     AMR gnrB4
                                    15.37
                                               33.16
                                                              PCP 10<sup>1</sup>
                                                                                 PCP 10<sup>6</sup>
                                                                                                    NA
## 15
                        AMR SHV
                                     14.64
                                                              PCP 10<sup>1</sup>
                                                                                 PCP 10<sup>6</sup>
                                                                                                    NΑ
                                               33.00
## 16
                       AMR sul1
                                     16.16
                                                33.74
                                                              PCP 10<sup>1</sup>
                                                                                 PCP 10<sup>6</sup>
                                                                                                    NA
## 17
                       AMR sul2
                                     15.20
                                               32.64
                                                              PCP 10<sup>1</sup>
                                                                                 PCP 10<sup>6</sup>
                                                                                                    NA
## 18
                        AMR TEM
                                     15.43
                                                34.76
                                                              PCP 10<sup>0</sup>
                                                                                 PCP 10<sup>6</sup>
                                                                                                    NΑ
## 19
                        AMR VIM
                                     13.55
                                                              PCP 10<sup>0</sup>
                                                                                 PCP 10<sup>6</sup>
                                               33.12
                                                                                                    NA
## 20
                                                                                 PCP 10<sup>6</sup>
                        Ascaris
                                     12.90
                                               31.42
                                                              PCP 10<sup>1</sup>
                                                                                                    NA
## 21
                AstroV capsid
                                     11.07
                                               34.54
                                                              PCP 10<sup>0</sup>
                                                                                 PCP 10<sup>6</sup>
                                                                                                    NA
## 22
                            bfpA
                                     13.32
                                                33.36
                                                              PCP 10<sup>0</sup>
                                                                                 PCP 10<sup>6</sup>
                                                                                                    NA
## 23
                                                              PCP 10<sup>1</sup>
                                                                                                    NA
                    C. hominus
                                     13.84
                                                32.51
                                                                                 PCP 10<sup>6</sup>
## 24
                     C. parvum
                                     13.27
                                                34.62
                                                              PCP 10<sup>0</sup>
                                                                                 PCP 10<sup>6</sup>
                                                                                                    NA
## 25
                                     13.05
                                                              PCP 10<sup>1</sup>
                                                                                 PCP 10<sup>6</sup>
                            cadF
                                               32.07
                                                                                                    NA
                    Crypto 18S
## 26
                                     14.72
                                                34.11
                                                              PCP 10<sup>1</sup>
                                                                                 PCP 10<sup>6</sup>
                                                                                                    NA
## 27
                    Cyclospora
                                     13.50
                                                34.17
                                                              PCP 10<sup>0</sup>
                                                                                 PCP 10<sup>6</sup>
                                                                                                    NA
## 28
                                     13.35
                                                32.31
                                                              PCP 10<sup>1</sup>
                                                                                 PCP 10<sup>6</sup>
                              eae
                                                                                                    NΑ
## 29
                     Entamoeba
                                     16.95
                                                35.51
                                                              PCP 10<sup>1</sup>
                                                                                 PCP 10<sup>6</sup>
                                                                                                    NA
## 30
                  EntV 5' UTR
                                                              PCP 10<sup>0</sup>
                                                                                 PCP 10<sup>6</sup>
                                     10.21
                                                32.61
                                                                                                    NA
        Fecal bacteria 16S
                                     17.17
                                                34.60
                                                              PCP 10<sup>1</sup>
                                                                                 PCP 10<sup>6</sup>
                                                                                                    NA
## 32
                                                              PCP 10<sup>0</sup>
                                     13.05
                                                                                 PCP 10<sup>6</sup>
                        Giardia
                                               34.34
                                                                                                    NΑ
## 33
                            GlyA
                                     13.55
                                                34.79
                                                              PCP 10<sup>0</sup>
                                                                                 PCP 10<sup>6</sup>
                                                                                                    NA
## 34
                    HAdV fiber
                                     15.37
                                               34.80
                                                              PCP 10<sup>1</sup>
                                                                                 PCP 10<sup>6</sup>
                                                                                                    NA
## 35
                    HAdV hexon
                                     13.26
                                                32.87
                                                              PCP 10<sup>1</sup>
                                                                                 PCP 10<sup>6</sup>
                                                                                                    NΑ
## 36
                                                              PCP 10<sup>0</sup>
                                                                                 PCP 10<sup>6</sup>
                            hip0
                                     15.69
                                               37.43
                                                                                                    NA
## 37
                                     12.98
                                                              PCP 10<sup>0</sup>
                                                                                 PCP 10<sup>6</sup>
                            ipaH
                                               33.33
                                                                                                    NA
## 38
                                     13.26
                               LT
                                               31.89
                                                              PCP 10<sup>1</sup>
                                                                                 PCP 10<sup>6</sup>
                                                                                                    NA
##
   39
                           MS2g1
                                     11.46
                                               35.07
                                                              PCP 10<sup>0</sup>
                                                                                 PCP 10<sup>6</sup>
                                                                                                    NA
## 40
                       Noro GII
                                     11.43
                                               35.23
                                                              PCP 10<sup>0</sup>
                                                                                 PCP 10<sup>6</sup>
                                                                                                    NA
## 41
                                                                                 PCP 10<sup>6</sup>
                        PhHV gB
                                     13.12
                                                32.97
                                                              PCP 10<sup>1</sup>
                                                                                                    NA
## 42
                                     13.71
                                                35.65
                                                              PCP 10<sup>0</sup>
                                                                                 PCP 10<sup>6</sup>
                                                                                                    NA
                     Rotavirus
## 43
                     Sapovirus
                                     17.88
                                                38.02
                                                              PCP 10<sup>1</sup>
                                                                                 PCP 10<sup>6</sup>
                                                                                                    NA
## 44
               SARS-CoV-2 N1
                                     12.83
                                                34.63
                                                              PCP 10<sup>0</sup>
                                                                                 PCP 10<sup>6</sup>
                                                                                               10.66
## 45
                SARS-CoV-2 N2
                                     12.91
                                                37.00
                                                              PCP 10<sup>0</sup>
                                                                                 PCP 10<sup>6</sup>
                                                                                               15.00
## 46
                              STh
                                     13.98
                                                35.67
                                                              PCP 10<sup>0</sup>
                                                                                 PCP 10<sup>6</sup>
                                                                                                    NA
## 47
                                               35.35
                                                              PCP 10<sup>0</sup>
                                                                                 PCP 10<sup>6</sup>
                              STp
                                     14.46
                                                                                                    NA
## 48
                            stx1
                                     15.32
                                                33.46
                                                              PCP 10<sup>1</sup>
                                                                                 PCP 10<sup>6</sup>
                                                                                                    NA
## 49
                                     14.74
                                               36.41
                                                              PCP 10<sup>0</sup>
                                                                                 PCP 10<sup>6</sup>
                                                                                                    NA
                            stx2
## 50
                        STY0201
                                     14.15
                                                33.11
                                                              PCP 10<sup>1</sup>
                                                                                 PCP 10<sup>6</sup>
## 51
                     Trichuris
                                     13.40
                                               31.99
                                                              PCP 10<sup>1</sup>
                                                                                 PCP 10<sup>6</sup>
                                                                                                    NΑ
## 52
                                     13.37
                                                32.00
                                                              PCP 10<sup>1</sup>
                                                                                 PCP 10<sup>6</sup>
                              ttr
                                                                                                    NΑ
## 53
                                       9.30
                                                              PCP 10<sup>1</sup>
                                                                                 PCP 10<sup>6</sup>
                                                                                                    NA
                            {\tt tviB}
                                               27.70
## 54
                                                              PCP 10<sup>1</sup>
                                                                                 PCP 10<sup>6</sup>
                            uidA
                                     13.79
                                               31.71
                                                                                                    NA
## 55
                            virF
                                     15.04
                                               34.82
                                                              PCP 10<sup>1</sup>
                                                                                 PCP 10<sup>6</sup>
                                                                                                    NA
##
        Y.intercept Slope
                                     R2 Efficiency Y.intercept_adj Slope_adj R2_adj
## 1
                 36.67 -3.63 0.99
                                                  0.88
                                                                        36.67
                                                                                       -3.63
                                                                                                  0.99
## 2
                 34.86 -3.59 0.99
                                                                        34.86
                                                                                       -3.59
                                                                                                  0.99
                                                  0.90
## 3
                 39.42 -3.83 1.00
                                                  0.82
                                                                        39.42
                                                                                       -3.83
                                                                                                  1.00
## 4
                 36.50 -3.64 1.00
                                                  0.88
                                                                        36.63
                                                                                       -3.67
                                                                                                  1.00
## 5
                 36.98 -3.45 0.98
                                                  0.95
                                                                                                  0.98
                                                                        36.98
                                                                                       -3.45
## 6
                 41.08 -3.82 1.00
                                                  0.83
                                                                        41.08
                                                                                       -3.82
                                                                                                  1.00
## 7
                 37.53 -3.66 1.00
                                                  0.88
                                                                        37.53
                                                                                       -3.66
                                                                                                  1.00
## 8
                 39.17 -3.86 1.00
                                                                                                  1.00
                                                  0.82
                                                                        39.17
                                                                                       -3.86
## 9
                 38.04 -3.66 1.00
                                                  0.88
                                                                        38.04
                                                                                       -3.66
                                                                                                  1.00
## 10
                 37.26 -3.63 0.99
                                                  0.89
                                                                        37.26
                                                                                       -3.63
                                                                                                  0.99
## 11
                 35.88 -3.26 0.98
                                                  1.03
                                                                        37.94
                                                                                       -3.71
                                                                                                  0.99
```

##	10	36.75 -3.32 0.99	1.00	37.71	-3.54	0.99
						1.00
##		37.77 -3.57 1.00	0.91	37.77	-3.57	
##		37.06 -3.47 0.99	0.94	37.06	-3.47	0.99
##		37.06 -3.59 0.99	0.90	37.06	-3.59	0.99
##	16	37.91 -3.54 0.99	0.92	37.91	-3.54	0.99
##	17	36.90 -3.57 0.99	0.91	36.90	-3.57	0.99
##	18	36.05 -3.24 0.98	1.04	37.18	-3.50	0.99
##	19	34.11 -3.22 0.98	1.04	35.89	-3.62	0.99
##	20	35.44 -3.64 1.00	0.88	35.44	-3.64	1.00
##	21	34.83 -3.93 1.00	0.80	35.08	-3.99	1.00
##	22	34.52 -3.35 0.99	0.99	35.52	-3.58	0.99
##		36.45 -3.68 1.00	0.87	36.45	-3.68	1.00
##		34.89 -3.46 1.00	0.95	35.13	-3.52	0.99
##		36.14 -3.73 0.99	0.85	36.14	-3.73	0.99
##						
		37.83 -3.78 1.00	0.84	37.83	-3.78	1.00
##		34.99 -3.38 0.99	0.98	36.47	-3.71	0.99
##		36.47 -3.73 0.99	0.86	36.47	-3.73	0.99
##		38.99 -3.57 0.99	0.90	38.99	-3.57	0.99
##		33.06 -3.71 1.00	0.86	33.45	-3.80	1.00
##		38.88 -3.47 0.99	0.94	38.88	-3.47	0.99
##	32	34.76 -3.47 1.00	0.94	35.13	-3.56	0.99
##	33	36.28 -3.60 0.98	0.90	37.58	-3.90	1.00
##	34	38.58 -3.80 1.00	0.83	38.58	-3.80	1.00
##	35	36.50 -3.79 1.00	0.84	36.50	-3.79	1.00
##	36	38.88 -3.72 0.97	0.86	40.51	-4.09	0.99
##	37	34.52 -3.38 0.99	0.98	35.56	-3.61	0.99
##	38	36.05 -3.67 0.99	0.87	36.05	-3.67	0.99
##	39	35.01 -3.79 1.00	0.84	34.96	-3.77	0.99
##		35.11 -3.85 1.00	0.82	35.01	-3.83	1.00
##		36.73 -3.83 1.00	0.82	36.73	-3.83	1.00
##		36.22 -3.65 1.00	0.88	36.70	-3.76	1.00
##		42.25 -4.00 1.00	0.78	42.25	-4.00	1.00
##		35.17 -3.61 1.00	0.89	35.64	-3.72	1.00
##		37.26 -4.00 1.00	0.78	37.48	-4.05	0.99
##		36.28 -3.57 0.99	0.91	36.81	-3.69	0.99
##		36.38 -3.52 0.99	0.92	37.27	-3.73	1.00
##	48	37.57 -3.58 0.99	0.90	37.57	-3.58	0.99
##	49	36.96 -3.56 1.00	0.91	37.44	-3.66	1.00
##	50	36.94 -3.67 0.99	0.87	36.94	-3.67	0.99
##	51	36.02 -3.67 1.00	0.87	36.02	-3.67	1.00
##	52	36.00 -3.64 0.99	0.88	36.00	-3.64	0.99
##	53	31.89 -3.49 0.97	0.93	31.89	-3.49	0.97
##		35.75 -3.51 0.99	0.93	35.75	-3.51	0.99
##	55	38.45 -3.81 1.00	0.83	38.45	-3.81	1.00
##		Efficiency_adj				
##	1	0.88				
##		0.90				
##		0.82				
##						
		0.87				
##		0.95				
##		0.83				
##		0.88				
##		0.82				
##	9	0.88				

```
## 10
                 0.89
## 11
                 0.86
## 12
                 0.92
## 13
                 0.91
## 14
                 0.94
## 15
                 0.90
## 16
                 0.92
                 0.91
## 17
## 18
                 0.93
## 19
                 0.89
## 20
                 0.88
## 21
                 0.78
## 22
                 0.90
## 23
                 0.87
## 24
                 0.93
## 25
                 0.85
## 26
                 0.84
## 27
                 0.86
## 28
                 0.86
## 29
                 0.90
## 30
                 0.83
## 31
                 0.94
## 32
                 0.91
## 33
                 0.81
## 34
                 0.83
## 35
                 0.84
## 36
                 0.76
## 37
                 0.89
## 38
                 0.87
## 39
                 0.84
## 40
                 0.82
## 41
                 0.82
## 42
                 0.84
## 43
                 0.78
## 44
                 0.86
## 45
                 0.77
## 46
                 0.87
## 47
                 0.85
## 48
                 0.90
## 49
                 0.87
## 50
                 0.87
                 0.87
## 51
## 52
                 0.88
## 53
                 0.93
## 54
                 0.93
## 55
                 0.83
```

write.csv(summary,file="TAC\_sample\_linearity\_summary.csv")