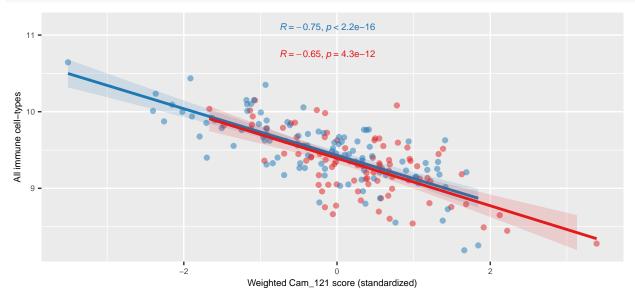
Reproducing Garg et al.'s Figures 4E & 4F

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Garg et al.'s 2021 study identified a gene signature, Cam_121, associated with distant metastases of melanoma and patient survival outcomes. The reproduced figures represent levels of expression for Cam_121 vs a total immune score.

The code found in this repository at \scripts\ImmuneCellAnalysis.Rmd produces the multiple figures. A select figure includes Figure 4E, shown below.

```
g0<- ggplot(data, aes(x=Cam_121,
                      y=ImmuneCell,
                      color=EventMet))+
  geom_point(alpha = 0.5)+
  xlab("Weighted Cam_121 score (standardized)")+
  ylab("All immune cell-types")+
  scale_color_manual(values = c("#1F78B4", "#E31A1C"), name = "Distant\nmetastases")+
  geom_smooth(method = "lm", alpha = .15, aes(fill = EventMet))+
  scale_fill_manual(values = c("#1F78B4", "#E31A1C"), name = "Distant\nmetastases")+
  theme(text=element_text(size=7,
                          family="sans"),
        legend.position = "none")+
  ggpubr::stat_cor(aes(color = EventMet),
                   label.x = -0.75,
                   label.y =c(max(data$ImmuneCell)+0.45, max(data$ImmuneCell)+0.1),
                   size = 2.5,family="sans")+
  ylim(c(min(data$ImmuneCell), max(data$ImmuneCell)+0.5))+
  coord_fixed()
g0
```



To make these figures to a publication standard, the following changes were executed:

- increased text size
- added legend
- removed gridlines and grey background
- introduced axis lines

```
g1<- ggplot(data, aes(x=Cam_121,
                      y=ImmuneCell,
                      color=EventMet))+
  geom_point(alpha = 0.4, size = 1)+
  theme(panel.grid = element_blank(),
       panel.background = element_blank(),
       axis.line = element_line())+
  xlab("Weighted Cam 121 score (standardized)")+
  ylab("All immune cell-types")+
  scale_color_manual(values = c("#1F78B4", "#E31A1C"), name = "Distant\nmetastases")+
  geom_smooth(method = "lm", alpha = .15, aes(fill = EventMet))+
  scale_fill_manual(values = c("#1F78B4", "#E31A1C"), name = "Distant\nmetastases")+
  theme(text=element_text(size=10, family="sans"), legend.position = "top")+
  ggpubr::stat_cor(aes(color = EventMet),
                   label.x = -0.75, label.y =c(max(data$ImmuneCell)+0.15,
                                               max(data$ImmuneCell)), size = 2.5, family="sans")+
  ylim(c(min(8), max(10.5)+0.5))
g1
```

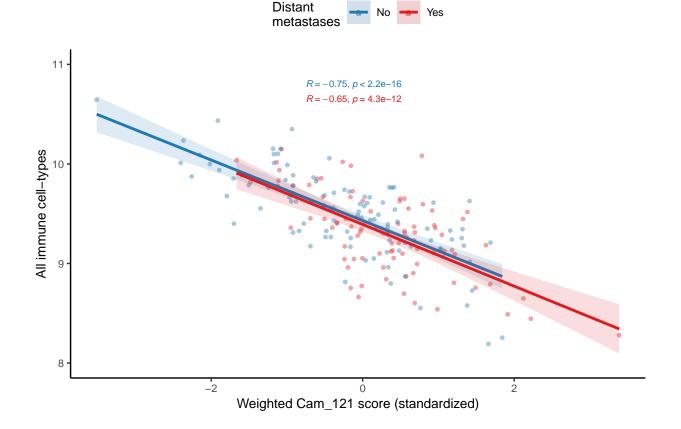
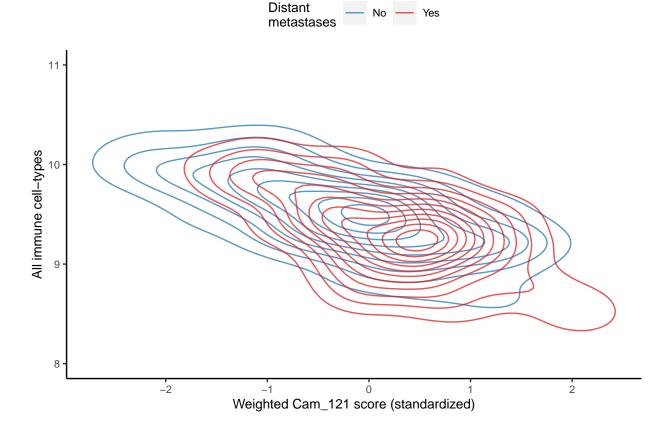


Figure 4F also employs the same set of data, but reconfigures the graph to a density plot to provide further nuance to analysis than just a scatter plot. The following figure is a revised reproduction of the plot.

```
g2<- ggplot(data, aes(x=Cam_121, y=ImmuneCell, color=EventMet))+
    #geom_point()+
    theme(panel.grid = element_blank(),
        panel.background = element_blank(),
        axis.line = element_line())+
    xlab("Weighted Cam_121 score (standardized)")+
    ylab("All immune cell-types")+
    scale_color_manual(values = c("#1F78B4", "#E31A1C"), name = "Distant\nmetastases")+
    geom_density_2d(alpha = 0.75) +
    theme(text=element_text(size=10, family="sans"), legend.position = "top")+
    ylim(c(min(8), max(10.5)+0.5))</pre>
```



The final reproduced figure of the Cam_121 vs immune scores with both figures:

