

# Supplement: Evidence of traumatic brain injury in muskoxen (*Ovibos moschatus*). Code for heatmaps and linegraphs

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## Packages

```
library(ggplot2)
library(readxl)
library(plotly)
library(ggpubr)
library(viridis)
```

## Heatmaps

### Muskox 1 / MO1 / Generic 0191

#### Read in data

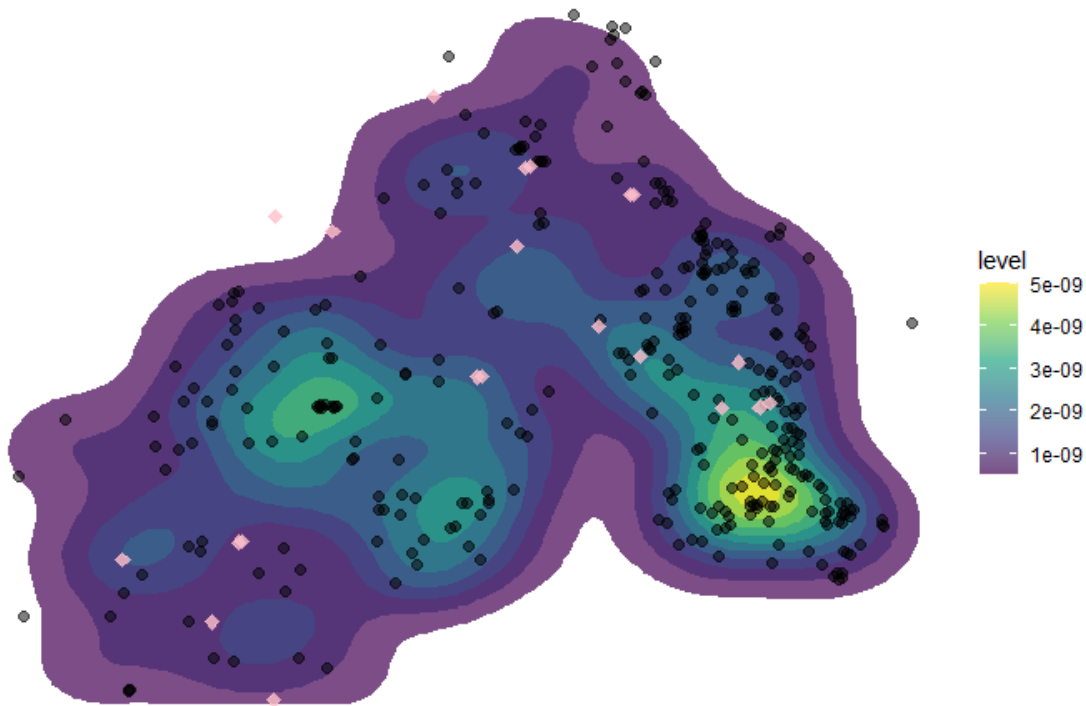
```
NTCs <- read_excel("NA_BHP_Supp_211022.xlsx", sheet=3)
neuropils <- read_excel("NA_BHP_Supp_211022.xlsx", sheet=4)
neurons <- read_excel("NA_BHP_Supp_211022.xlsx", sheet=5)
```

#### Heat Map 1

```
Muskox1_heatmap <- ggplot(neuropils, aes(x=x, y=y))+
  stat_density_2d(aes(fill=..level..), geom="polygon")+
  scale_fill_viridis_c(option = "viridis", alpha=0.7)+
  geom_point(data=NTCs, color="black", size=2, alpha=0.5)+
  geom_point(data=neurons, shape=23, colour="pink", fill="pink", size=2,
alpha=0.8)+
  labs(title="A")+
  theme_void()
```

Muskox1\_heatmap

A



## Muskox 2 / MO2 / Generic 0194

### Read in data

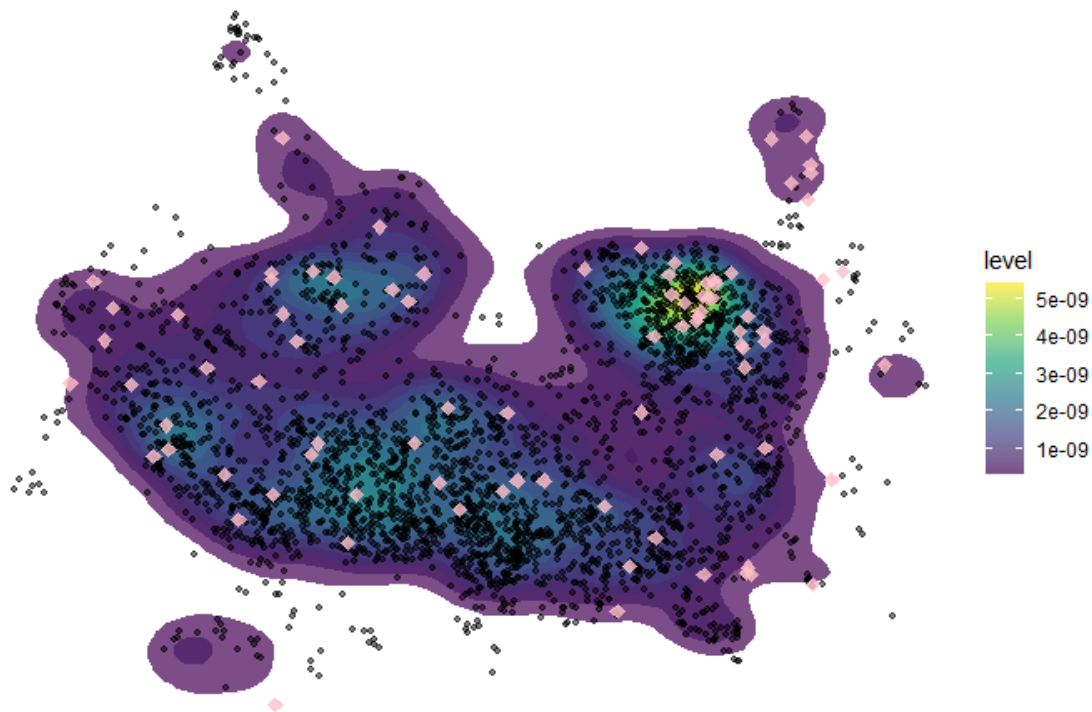
```
NTCs <- read_excel("NA_BHP_Supp_211022.xlsx", sheet=6)
neuropils <- read_excel("NA_BHP_Supp_211022.xlsx", sheet=7)
neurons <- read_excel("NA_BHP_Supp_211022.xlsx", sheet=8)
```

### Heat Map 2

```
Muskox2_heatmap <- ggplot(neuropils, aes(x=x, y=y))+
  stat_density_2d(aes(fill=..level..), geom="polygon", bins=20)+
  scale_fill_viridis_c(option = "viridis", alpha=0.7)+
  geom_point(data=NTCs, color="black", size=1, alpha=0.5)+
  geom_point(data=neurons, shape=23, colour="pink", fill="pink", size=2,
alpha=0.8)+
  labs(title="B")+
  theme_void()
```

Muskox2\_heatmap

B



## Muskox 3 / MO3 / Generic 0193

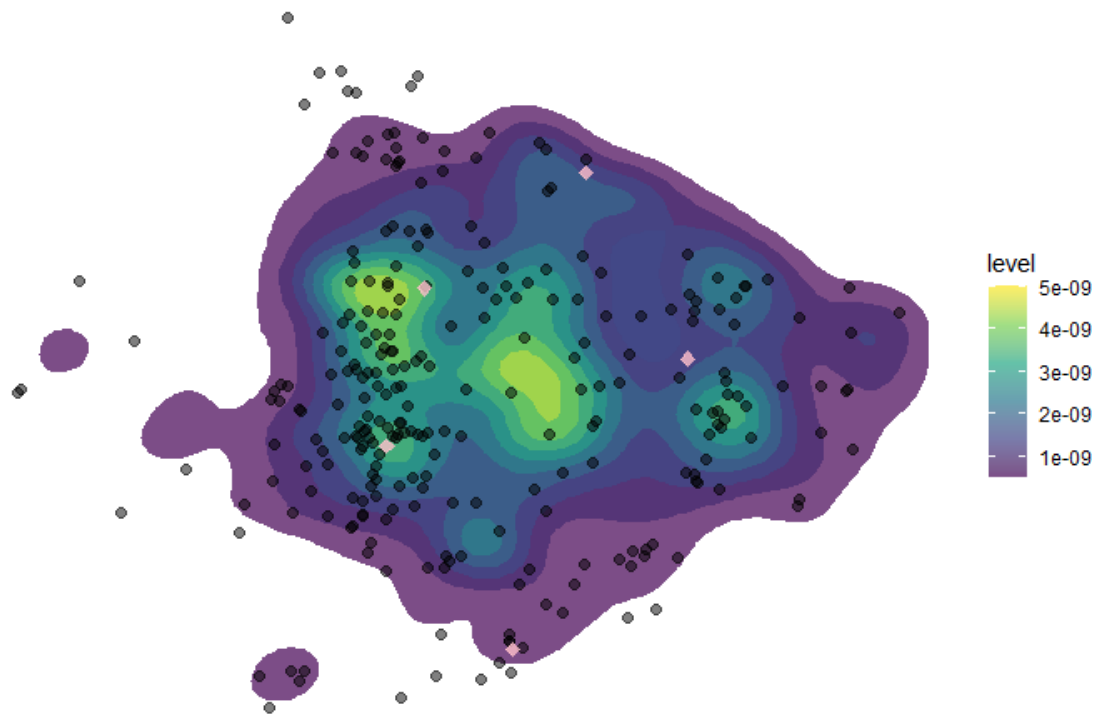
### Read in data

```
NTCs <- read_excel("NA_BHP_Supp_211022.xlsx", sheet=9)
neuropils <- read_excel("NA_BHP_Supp_211022.xlsx", sheet=10)
neurons <- read_excel("NA_BHP_Supp_211022.xlsx", sheet=11)
```

### Heat map 3

```
Muskox3_heatmap <- ggplot(neuropils, aes(x=x, y=y))+
  stat_density_2d(aes(fill=..level..), geom="polygon")+
  scale_fill_viridis_c(option = "viridis", alpha=0.7)+
  geom_point(data=NTCs, color="black", size=2, alpha=0.5)+
  geom_point(data=neurons, shape=23, colour="pink", fill="pink", size=2,
alpha=0.8)+
  labs(title="C")+
  theme_void()
Muskox3_heatmap
```

C



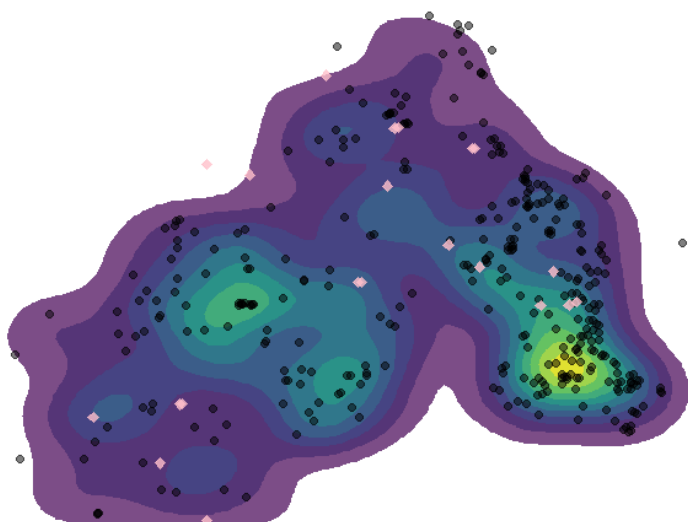
# all

three together

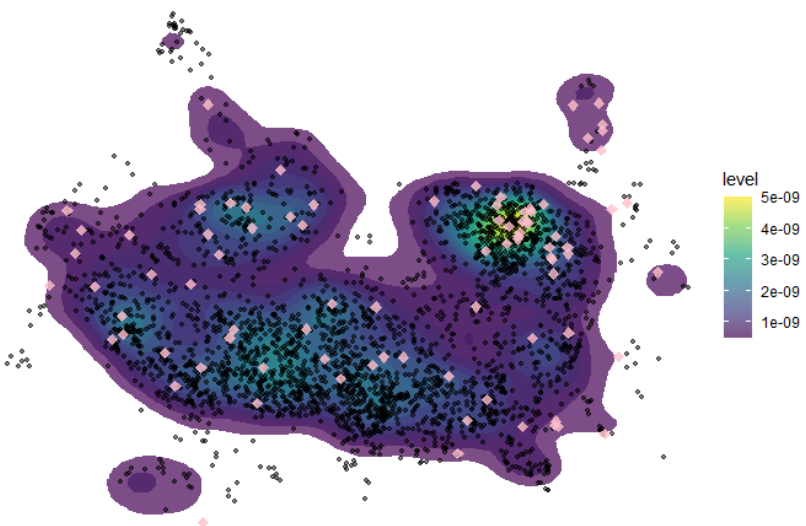
```
AllHeatmaps<-ggarrange(Muskox1_heatmap, Muskox2_heatmap, Muskox3_heatmap,  
nrow=3,  
common.legend = TRUE, legend="right")
```

AllHeatmaps

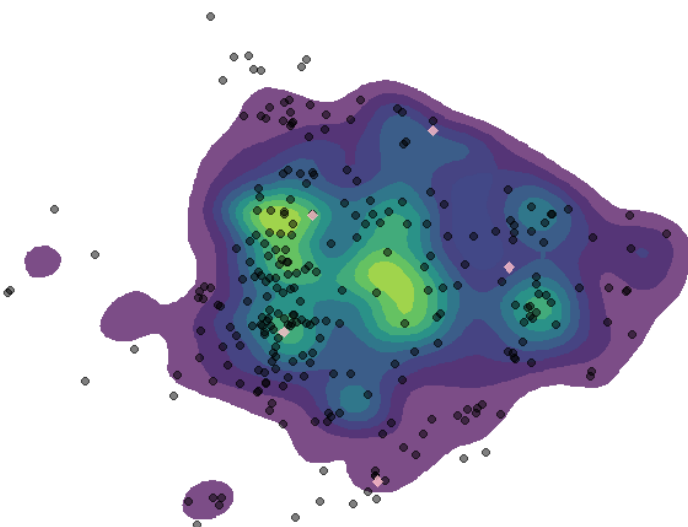
A



B



C



## Linegraphs

### Read in Data

```
AllData <- read_excel("NA_BHP_Supp_211022.xlsx", sheet=2)
```

### make characters non numerical

```
AllData$Individual<-as.character(AllData$Individual)  
AllData$layer<-as.character(AllData$layer)
```

### Subset

```
NTCsub <- subset(AllData, marker=="NTC" & layer=="all")  
Neuropilsub <- subset(AllData, marker=="neuropil" & layer=="all")  
Neuronsub <- subset(AllData, marker=="neuron" & layer=="all")
```

### Marker count per layer per Individual

```
sub_NTC <- subset(AllData, marker=="NTC")  
sub_Neuropil <- subset(AllData, marker=="neuropil")  
sub_Neuron <- subset(AllData, marker=="neuron")
```

## Make line graphs

### 3 Individuals NTC

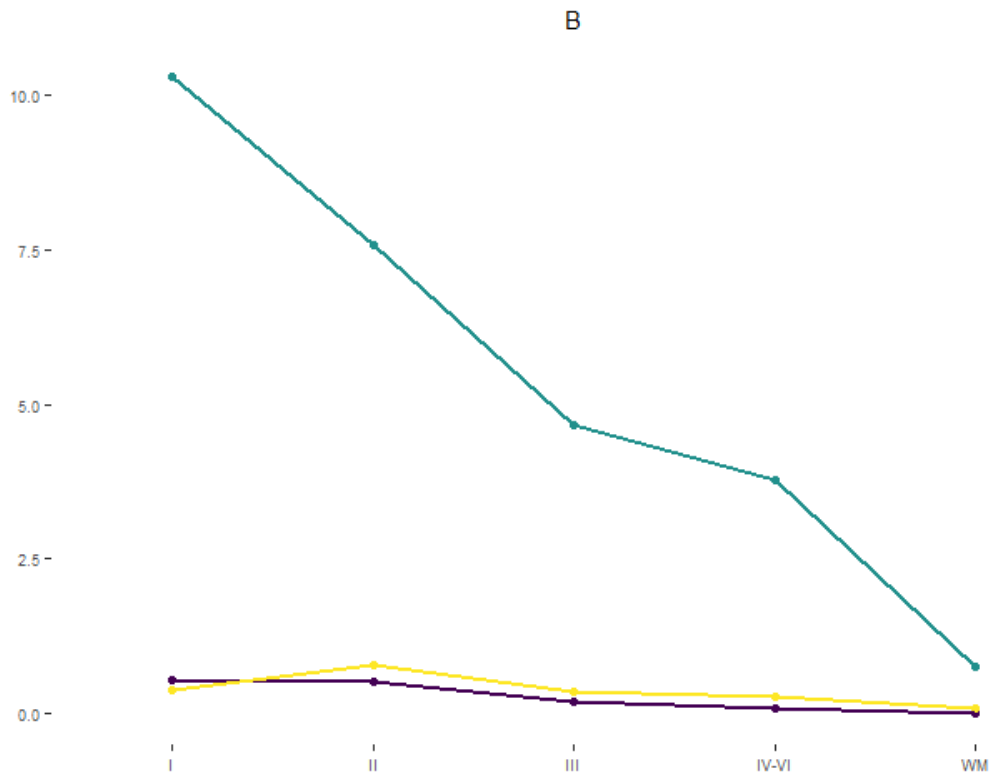
```
p1 <- ggplot(data=sub_NTC, aes(x=layer, y=count, group=Individual,  
colour=Individual)) +  
  geom_line()+ geom_point()+ scale_color_manual(values=viridis(3))+  
  labs(title="B",x=element_blank(), y=element_blank())+  
  theme(  
    plot.title = element_text(size=11, hjust=0.5),  
    panel.border = element_blank(),  
    panel.grid.major = element_blank(),  
    panel.grid.minor = element_blank(),  
    panel.background = element_blank(),  
    axis.line = element_blank(),  
    axis.text=element_text(size=7),  
    legend.position="none") +  
  geom_path(size=1)+  
  scale_x_discrete(limits=c("I","II","III","IV-VI","WM"))
```

p1

```
## Warning: Removed 3 row(s) containing missing values (geom_path).
```

```
## Warning: Removed 3 rows containing missing values (geom_point).
```

```
## Warning: Removed 3 row(s) containing missing values (geom_path).
```



### 3 Individuals Neuropil

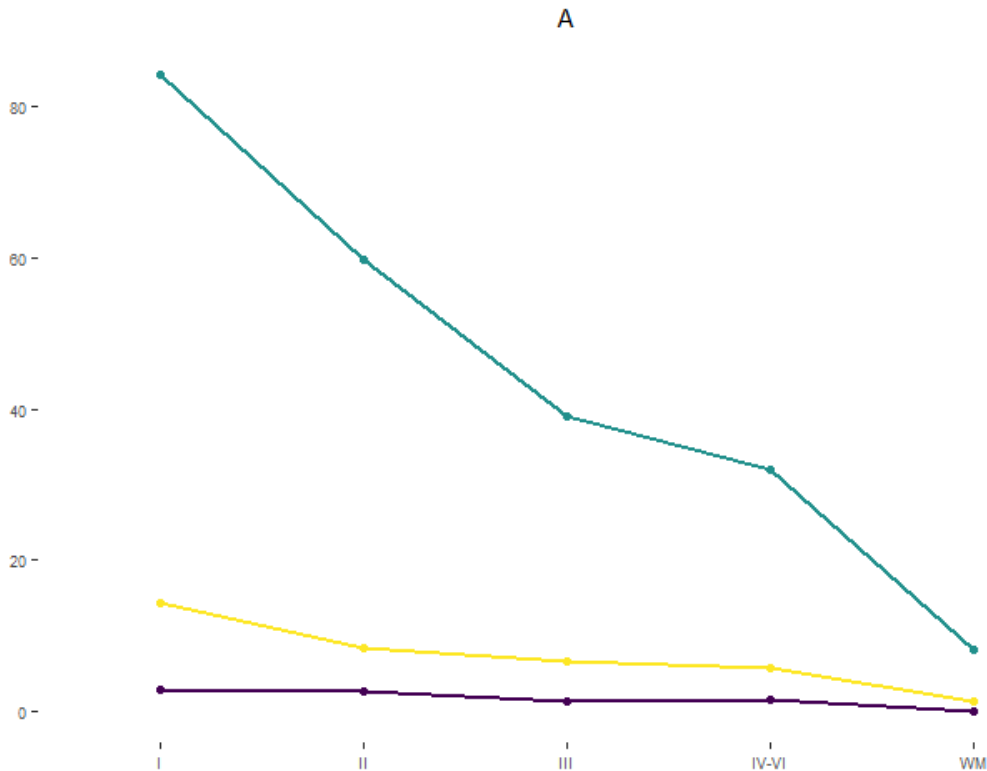
```
p2 <- ggplot(data=sub_Neuropil, aes(x=layer, y=count, group=Individual,
colour=Individual)) +
  geom_line()+ geom_point()+ scale_color_manual(values=viridis(3))+
  labs(title="A",x=element_blank(), y=element_blank())+
  theme(
    plot.title = element_text(size=11, hjust=0.5),
    panel.border = element_blank(),
    panel.grid.major = element_blank(),
    panel.grid.minor = element_blank(),
    panel.background = element_blank(),
    axis.line = element_blank(),
    axis.text=element_text(size=7),
    legend.position="none") +
  geom_path(size=1)+
  scale_x_discrete(limits=c("I","II","III","IV-VI","WM"))
```

p2

## Warning: Removed 3 row(s) containing missing values (geom\_path).

## Warning: Removed 3 rows containing missing values (geom\_point).

## Warning: Removed 3 row(s) containing missing values (geom\_path).



### 3 Individuals Neuron

```
p3 <- ggplot(data=sub_Neuron, aes(x=layer, y=count, group=Individual,
colour=Individual)) +
  geom_line() + geom_point()+ scale_color_manual(values=viridis(3))+
  labs(title="C",x="Brain layers", y=element_blank())+
  theme(
    plot.title = element_text(size=11, hjust=0.5),
    panel.border = element_blank(),
    panel.grid.major = element_blank(),
    panel.grid.minor = element_blank(),
    panel.background = element_blank(),
    axis.line = element_blank(),
    axis.title.x = element_text(size=8),
    axis.text=element_text(size=7)) +
  geom_path(size=1)+
  scale_x_discrete(limits=c("I","II","III","IV-VI","WM"))
```

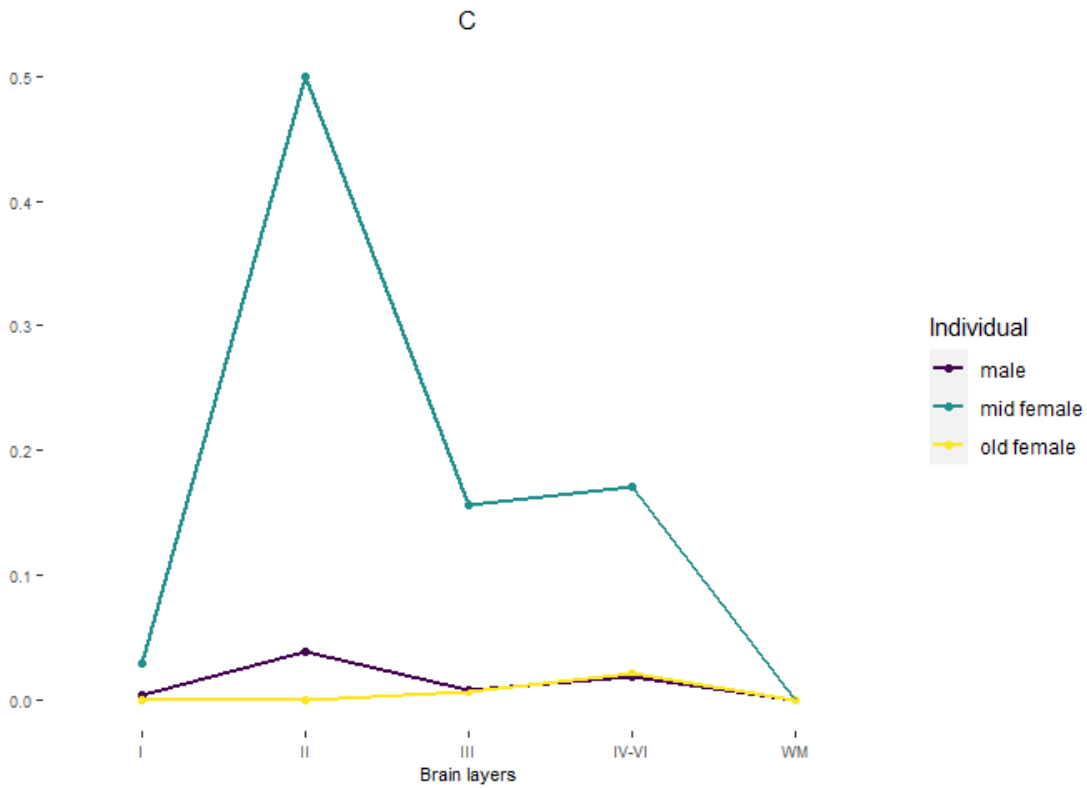
p3

## Warning: Removed 3 row(s) containing missing values (geom\_path).

## Warning: Removed 3 rows containing missing values (geom\_point).

## Warning: Removed 3 row(s) containing missing values (geom\_path).





##arrange on page

```
all3<-ggarrange(p2, p1, p3, nrow=3, common.legend = TRUE, legend="right")
```

```
all3<-annotate_figure(all3, left=text_grob("Estimated population / volume  
"~(mm^3),rot = 90))
```

```
all3
```

