**FOR INDIVIDUAL FISH DATA FILES**

file\_prepare : Function to prepare data file for further processing and detect shuttles

calc\_core\_body\_temp: Function to calculate body core temperature if not already done in Shuttlesoft

calc\_Tpref: Function to calculate Tpref using either the mean or median of all body core temperature values

calc\_Tavoid: Function to calculate upper and lower avoidance temperatures

calc\_distance: Function to calculate total distance moved

calc\_shuttling\_frequency: Function to calculate shuttling frequency

calc\_occupancy\_time: Function to calculate occupancy time in each chamber

plot\_body\_core\_temp\_histogram: Function to plot a histogram of time spent at different core body temperatures

calc\_variance: Function to calculate variance in core body temperature experienced throughout the trial

plot\_cumulative\_distance: Function to plot cumulative changes in distance moved over time

calc\_min\_gravitation: Function to calculate minimum gravitation time

calc\_extremes: Function to plot time spent at extreme temperatures, near set limits

plot\_speed\_histogram: Function to plot histogram of movement speeds

plot\_speed\_vs\_core\_temp: Function to plot movement speed against body core temperature

plot\_temperature\_gradient: Function to plot temperatures in each side of the shuttlebox over time

plot\_temp\_segmented: Function to plot core body temperature, Tpref, avoidance temperatures, segmented regression of changes in body core temperature during the trial

plot\_temp: Function to plot changes in core body temperature over time, as well as Tpref, and upper and lower avoidance temperatures, but WITHOUT segmented regression lines

calc\_act\_gravitation: Function to calculate actual gravitation time

shuttling\_aggregated: Function to calculate and plot interval means for shuttling rate over time

speed\_aggregated: Function to calculate and plot interval means for speed over time

Tcore\_aggregated: Function to calculate and plot interval means for body core temperature over time

mean\_correlations\_scatterplots: Function to calculate interval means for speed, shuttling rate, and Tcore, then produces a correlation matrix

animate\_movements: Function to plot a 3D animation of fish movements at selected time intervals, with time as a dimension

plot\_heatmap: Function to produce a heat map of fish locations over the trial

**FOR FILES WITH DATA FROM MULTIPLE FISH**

**(e.g. SUMMARY DATASET FOR A PROJECT):**

plot\_distance\_vs\_shuttles: Function to visualise links between shuttling and activity across individuals in the entire project dataset

plot\_limits\_vs\_distance: Function to visualise links between time near system temperature limits and activity across individuals in the entire project dataset

plot\_limits\_vs\_shuttles: Function to visualise links between time near system temperature limits and shuttles across individuals in the entire project dataset

pca\_and\_plot: Function to perform PCA with shuttles, distance moved, and time near limits, then plot PC scores vs Tpref. Fish that score highly for PC1 are those that are active, perform more shuttles, and spend less time near the limits upper and lower temperature limits set by the user in the shuttlebox, and therefore may not be actively selecting their temperature. In the example dataset, if you plot Tpref versus PC1 Score, fish that score low for PC1 have Tprefs near the extremes. Of course, they may also just prefer these temps but it is also possible they simply were not behaviourally thermoregulating. With the scatterplot points labelled, it’s possible to further inspect there suspicious individuals using some of the other functions (e.g. plot\_temp, shuttling\_aggregated, plot\_heatmap).

plot\_histograms: Function to create frequency distributions for key measures across individuals in the data set