**Effects of Noise and Visitor Numbers on Sharks and Baseline Movement Information**

**Aims:**

1. Determine if visitor number has an effect on shark movements and behaviours
2. Determine if decibel level has an effect on shark movements and behaviours
3. Determine if there are significant individual differences in behaviour
4. Identify and outline baseline levels of behaviour

**Methods of data collection:**

Observed 2 species of sharks in 2 separate aquarium environments. Observations were taken in 15 min blocks per individual animal at random times of day from 10am-1pm and 2-5pm for 16 days. Observations were recorded electronically in tge Zoo Monitor application, interval behaviours (interval\_channel\_value) were recorded once every 1 min. and all occurrence behaviours (or rare behaviours; all\_occurrence\_value) were recorded anytime they occurred during the 15 min observation session.

**Datasheets:**

***Datasheet 1: Leopard sharks (lep\_v5) - column titles in bold***

**session**: unique session ID (each session is 15 min long and has one individual/one focal animal)

**date\_time**: date and time of session

**weekday**: WE: weekend, W: weekday

**hour**: hour of the day

**channel**: All Occurrence: measured anytime a behaviour occurred (rare behaviours), Interval: behaviours recorded once every 60 sec (more common behaviours)

**focal:** Name of focal individual (leopard sharks: Katrina, Little girl, Notches, Old girl)

**all\_occurrence\_value**: frequency of rare behaviours measured over the 15 min observation session. Includes: Burst-swim, Face bang, Head-shake, Spy-hop, Surface touch, Turn in water column, Vertical swim, Yawn, Interactions (x8), Routes, Penguin(s) in the water.

- Interactions: Anytime the focal shark interacted with another animal in a positive /negative way (i.e. Pos. Interaction with shark, Neg. Interaction with penguin)

- Routes: No Route: when the shark is not swimming in a pattern, Route 1/2/3: when the focal shark is swimming in a loop pattern repeatedly. (only measured for 1 week out of the full 16 days)

- Penguin(s) in the water: Anytime this all-occurrence value is present in the 15 min. session it means there were penguins in the water. The whole 15-min session should be considered ‘with penguins in the water.’

**frame\_number**: first minute to the 15th min. of observation blocks in the session (1 to 15)

**interval\_channel\_value**: Behaviour individual was partaking in at the exact point of the 60 second intervals. Includes: Glide, Other, Out of view, Rest, Sink, Still, Swim, Vertical.

**speed:** swimming speed,either Swim fast or Swim slow

**depth:** what depth the shark was at, 0 Floor, 1 Low, 2 Mid, 3 High, 4 Surface

**observers**: number of observers present, 0, 1\_5 (1 to 5), 6\_10 (6 to 10), >10

**space\_use\_x:** coordinates for space use on x axis

**space\_use\_y:** coordinates for space use on y axis

**space\_use\_xy:** coordinates for space use on xy axis

**feeding\_day**: whether it was a feeding day or not, Yes (feeding day) or No (not feeding day)

**front\_tank:** yes: at the front of the tank, blank: any other location in the tank

***Datasheet 2: Epaulette sharks (ep\_v5)***

**session**: unique session ID (each session is 15 min long and has one individual/one focal animal)

**date\_time**: date and time of session

**weekday**: WE: weekend, W: weekday

**hour**: hour of the day

**channel**: All Occurrence: measured anytime a behaviour occurred (rare behaviours), Interval: behaviours recorded once every 60 sec. (more common behaviours)

**focal:** Name of focal individual (epaulette sharks: Rebel, Nicole)

**all\_occurrence\_value:** frequency of rare behaviours measured over the 15 min observation session. Includes: Burst swim, Sand blow, Surface swim, Surface touch, Tail-stand, Yawn, Interactions (x7)

- Interactions: Anytime the focal shark interacted with another animal in a positive /negative way (i.e. Pos. Interaction with shark, Neg. Interaction with shark)

**frame\_number**: first minute to the 15th min of observation blocks in the session (1 to 15)

**interval\_channel\_value**: Behaviour individual was partaking in at the exact point of the 60 second intervals. Includes: Glide, Inactive, Other, Out of view, Standing, Stationary active, Swim fast, Swim slow, Tail stand, Walk.

**depth:** what depth the shark was at, 0 Floor, 1 Low, 2 Mid, 3 High, 4 Surface

**observers**: number of observers present, 0, 1\_5 (1 to 5), 6\_10 (6 to 10), >10

**space\_use\_x:** coordinates for space use on x axis

**space\_use\_y:** coordinates for space use on y axis

**space\_use\_xy:** coordinates for space use on xy axis

**feeding\_day**: whether it was a feeding day or not, Yes (feeding day) or No (not feeding day)

**vents:** number of ventilations per post session measuring minute (should be correlated with activity and repetitive behaviours because higher ventilations means higher activity)

**shelter**: Yes: shark taking refuge in shelter, blank: shark not taking refuge in shelter

***Datasheet 3: Decibel levels per session (mic\_decibels)***

**session:** session ID number **(*to be used for matching to lep\_v5 and ep\_v5 sheets*)**

**la\_eq:** A-weighted equivalent sound level

**max\_level:** maximum level measured during run time

**lc\_peak:** the C-weighted peak sound pressure level

**Definitions across analysis**

High activity and repetitive behaviours:

- Epaulette sharks (ep\_v5): Burst swim, Surface swim, Surface touch, Tail-stand

- Leopard sharks (lep\_v5): Burst swim, Face bang, Head-shake, Spy-hop, Surface touch, Turn in water column, Vertical swim.

Significance: use a statistical significance level of P < 0.05

1. **Eps and Leps (together) Analysis: (ep\_v5, lep\_v5, and mic\_decibels)**
2. - To calculate cumulative visitor numbers during a 15-min observation period: multiply the occurrence (frequency) of each category in the following manner. The first category of visitors, the researcher only (0), by zero, the second category (1-5) by one, the third category (6-10) by two, and the fourth category (>10) by three; this will produce a scale 0– (?) of accumulative visitor numbers.

B. - Is there a significant difference between:

- Number of observers on the weekdays vs. weekends

- gglplot bar graph with SD

- Level of decibels (3 types of decibels) on the weekdays vs. weekends

- gglplot bar graph with SD

C. - Is number of observers correlated with decibel level (separate test for 3 decibel measurements)

- ggplot line graph with SD (example Figure 1)

D. - Compare decibel levels (3 types) when there are 0 observers (lowest level) in a session to when there are more than 0 (i.e. 1-5, 6-10 and >10). Is there a significant difference here?

- paired t-tests (?)

1. **Separate Analysis (for eps – ep\_v5 combined with mic\_decibels by session):**
2. Baseline Behaviour data

- Convert behavioural data (interval\_channel\_value) into percentages for each session

- Convert behavioural data (all\_occurrence\_value) into frequency counts (or relative frequency) for each session

- visualise with bar graphs

B. Observers:

- Is there a significant difference between frequency of number of observers between leps and eps (0, 1-5, 6-10, and >10)

- ggplot bar graph with SD

C. Observer Numbers and Movement:

- Is there evidence that observer numbers alone affect epaulette shark movement (movement defined as high activity and repetitive behaviours: all\_occurrence\_value)

- Is there a significantly different proportion of time spent doing (interval\_channel\_value) behaviours when there are 0 vs. more than 0 observers.

- visualise with ggplot and SD

D. Decibel Levels and Movement:

- Establish noise threshold for Leq for eps.

- When the equivalent noise levels are higher than the mean, consider the noise higher than usual and expect a behavioural change. In other words, when the Leq is higher than L50, predict a behavioural response.

- Is there evidence that decibel levels alone (Leq only) significantly affect epaulette shark movement (movement defined as high activity and repetitive behaviours)

E. Space Use

- Compare shelter use for sessions with 0 observers and sessions with 1-5, 6-10 and >10 observers. Is there a significant difference?

- Shelter use is occurring when: shelter = yes

- gg plot with SD

F. Vents (number of ventilations – measured for 1 min post session)

- Is number of vents correlated with high activity and repetitive behaviours and what is the relationship?

- Is number of vents correlated with number of visitors and what is the relationship?

- Is number of vents correlated with decibel levels (all 3 types) and what is the relationship?

- ggplot with SD to visualise

G. Feeding Day

- Is there a significant difference between (all\_occurrence\_value) high activity and repetitive behaviours when it is a feeding day (Yes) vs. when it is not (No)

- Is there a significant difference between time spent inactive vs. all other interval behaviours when it is a feeding day?

H. Interactions

- Compare number of positive interactions with number of observers and decibel levels. Is there a significant relationship here between number of positive interactions and observers, or number of positive interactions and decibel level?

I. Depth

- Compare depth (5 types) for sessions with 0 observers vs. sessions with >0 observers. Is there a significant difference with collective time spent at different depth for 0 vs. more than 0 observers.

- Individual variation: did individual sharks spend a significantly different ratio of time at different depth levels?

1. **Separate Analysis (for leps – lep\_v5 combined with mic\_decibels by session)**

A. Baseline behaviour data

- Convert behavioural data (interval\_channel\_value) into percentages (proportions) for each session

- Convert behavioural data (all\_occurrence\_value) into frequency counts (or relative frequencies) for each session

- visualise with bar graphs

B. Observers:

- (covered above, so no need to repeat) Is there a significant difference between frequency of number of observers between leps and eps (0, 1-5, 6-10, >10)

- ggplot bar graph with SD

C. Observer Numbers and Movement:

- Is there evidence that observer numbers alone affect leopard shark movement (movement defined as high activity and repetitive behaviours)

- Is there a significantly different proportion of time spent in (interval\_channel\_value) behaviours when there are 0 vs. more than 0 observers.

D. Decibel Levels and Movement:

- Establish noise threshold for Leq for leps.

- When the equivalent noise levels are higher than the mean, consider the noise higher than usual and expect a behavioural change. In other words, when the Leq is higher than L50, predict a behavioural response.

- Is there evidence that decibel level alone (Leq) significantly affects leopard shark movement (movement defined as high activity and repetitive behaviours)?

E. Space Use

- Compare space-use for sessions with 0 observers and sessions with 1-5, 6-10 and >10 observers

- Did use of the rest of the tank (not front; front\_tank = ‘blank’) increase significantly when there were more than 0 observers?

- Did use of the front of the tank increase significantly when penguins were in the tank? (front\_tank = yes)

- Penguins in tank defined as “Penguin(s) in the water” occurring at any time during the session in *all\_occurrence\_value* column.

F. Depth

- Compare depth (5 types) for sessions with 0 observers vs. sessions with >0 observers. Is there a significant difference with collective time spent at different depth for 0 vs. more than 0 observers.

- Individual variation: did individual sharks spend a significantly different ratio of time at different depth levels?

- visualise with ggplot and SD

G. Speed

- Compare swim speed (fast or slow) for sessions with 0 observers and sessions with >0 observers, is there a significant difference here?

- visualise with ggplot and SD

H. Routes

- Compare routes taken (no route vs. Route 1/2/3) for sessions with 0 observers and sessions with >0 observers, is there a significant difference here?

- Was there a significant difference between individuals in ratio of time spent in no route vs. in Route 1/2/3?

- Route frequency found in *all\_occurence\_value*

I. Penguins in the water

- Compare swim speed (fast or slow) to sessions with penguins in the water (‘Penguin(s) in the water’ occurs at any time during the session) to sessions with no penguins in the water. Is there a significant difference here?

- Compare frequency or high activity and repetitive behaviours to sessions with penguins in the water (occurs at any time during the session) to sessions with no penguins in the water. Is there a significant difference here?

- Compare Space use (front of the tank vs. rest of the tank) is there a significant difference in space-use when penguins are in the water (occurs at any time during the session) to sessions with no penguins in the water?

- visualise with ggplot and SD

J. Feeding Day

- Is there a significant difference between (all\_occurrence\_value) high activity and repetitive behaviours when it is a feeding day (Yes) vs. when it is not (No)

- Is there a significant difference between (interval\_channel\_value) proportion of time spent doing behaviours on feeding day vs. not feeding day?

K. Interactions

- Compare number of negative interactions with number of observers and decibel levels. Is there a significant relationship here between number of negative interactions and observers, or number of negative interactions and decibel level?

Examples:

Figure 1:

**Example methods from similar study: (Quadros et al. 2014)**

We tested whether the data met the requirements for parametric statistics by an Anderson–Darling normality test. Noise levels follow a parametric distribution (P > 0.05), but the behavioural data did not (P < 0.05), even after attempting data transformations; therefore, parametric tests were used for noise levels analysis and non-parametric statistical tests were used for behavioural analysis.

For noise levels analysis, we performed a linear regression to verify the relation of visitors (independent) and Leq (dependent). We also compared Leq values between intense visitation days and Mondays (day closed for visitation) for each enclosure by a Paired T test.

Behavioural data were converted into percentages for each session per species group (N = 15). Behavioural and shelter use data were compared for days with and with-out the presence of visitors using Wilcoxon matched pair tests. This was performed for each species group as well as for enclosure type. As noise levels can be similar for days with and without visitors, we established a noise threshold for Leq. When the equivalent noise levels were higher than the mean, we considered the noise higher than usual and expected a behavioural change. In other words, when the Leq was higher than L50, we predict a behavioural response. We compared the behaviours shown by each species group with higher Leq and lower Leq employing the Wilcoxon matched pair tests. The same procedure was used for comparing expressed behaviours in louder and quieter samples at each enclosure type. To calculate cumulative visitor numbers during a 20-min observation period: we multiplied the occurrence (frequency) of each category in the following manner. The first category of visitors, the researcher only (A), by zero, the second category by one (B), the third category by two (C) and so on: this produced a 0–50 scale of accumulative visitor numbers. We did this because zoo visitor numbers varied during our 10 consecutive sample points in each 20-min observation session. The sound pressure levels measured were correlated with behavioural expression per observation session for species and by individuals liable to identification, using a Spearman rank correlation. All statistical tests used a statistical significance level of P < 0.05 and were carried out in the software Minitab version 16 and IBM SPSS20.