

A case study examining the changes in behaviour and spatial selection in Grey's Kangaroos following the introduction of two wallaby species.

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# Introduction

Intro goes here



# Material and methods

## Study subjects

The study group consisted of 4.2 Eastern Grey kangaroos (GK), 3.0, Bennett’s wallaby (BW), and 2.0 Swamp wallaby (SW) living in an environmentally-enriched exhibit at the Singapore Zoo. Like most macropod-themed exhibits, visitors are permitted to walk through the exhibit via a guided trail while feeding and contact with the animals are largely prohibited. Visitorship was not counted in this study, and all individuals in were adults with no prior co-habitation exposure. The lay out of the exhibit is interspersed with natural foliage and logs (plant types) to provide natural shade and cover while feeding and drinking troughs are fixed in specific areas (see Figure 1).

## Data Collection

As residents of the exhibit, baseline activity and spatial use data were first collected from the GKs for 10 days (2018-12-07 – 2018-12-16) prior to the separate introduction of the BWs and SWs (2018-12-17 and 2019-02-27, respectively); activity and spatial use data were then collected for a period of 10 days after the introduction of each wallaby species. However, due to an unrelated health issue, one SW (‘Mickey’) was removed from the exhibit 7 days post-introduction thus tallying in 25, 15 and 7 days of observational data for the GKs, BWs, and SWs, respectively. As a result, the collected observational data can be grouped into three

Table 1: The ethogram and behavioural key used for examining behaviours in kangaroos and wallabies

| <b>Behaviour</b>      | <b>Key</b> | <b>Description</b>   |
|-----------------------|------------|--|
| Rest                  | Re         | Sitting or lying with body motionless for at least 30 sec, not moving  |
| Vigilance             | Vi         | Head up, ears pointing upwards.  |
| Locomotion            | Lo         | Moving from point A to point B using any method (pentapedal/bipedal)   |
| Drinking              | Dr         | Water consumption  |
| Feeding               | Fe         | Food consumption; grazing and/or browsing                              |
| Grooming              | Gr         | Licking own fur/wiping face with paws                                  |
| Intra-antagonism      | N          | Negative interaction between same species (Fights, Kicks, Etc)         |
| Intra-mutualism       | P          | Positive interaction between same species (Grooming, courtship, Etc)   |
| Negative interaction  | Nso        | Negative interaction between wallaby and kangaroo (Fights, Kicks, Etc) |
| Positive interactions | Pso        | Positive interaction between wallaby and kangaroo (elaborate?)         |
| Others                | O          | Other behaviours not specified in the ethogram.                        |
| Out-of-Sight          | OOS        | Observer is unable to see animal                                       |

treatments designed to characterise inter-species interaction (if applicable): T1 (i.e., solely GKs), T2 (i.e., GKs and BWs) and T3 (i.e., GKs, BWs, and SWs).

Behavioural observations were done using instantaneous scan sampling method where a single observation consisting of the behaviour and approximate zonal location (see Figure 1 for detail) of each individual was recorded. Here, observations were conducted at an hourly interval over a 10 h period that coincides with the operational hours of the exhibit (0900 – 1800 h). Observations were jotted on paper before being transferred onto a excel file. The descriptions of the recorded behaviours during the observation period are described in Table 1.

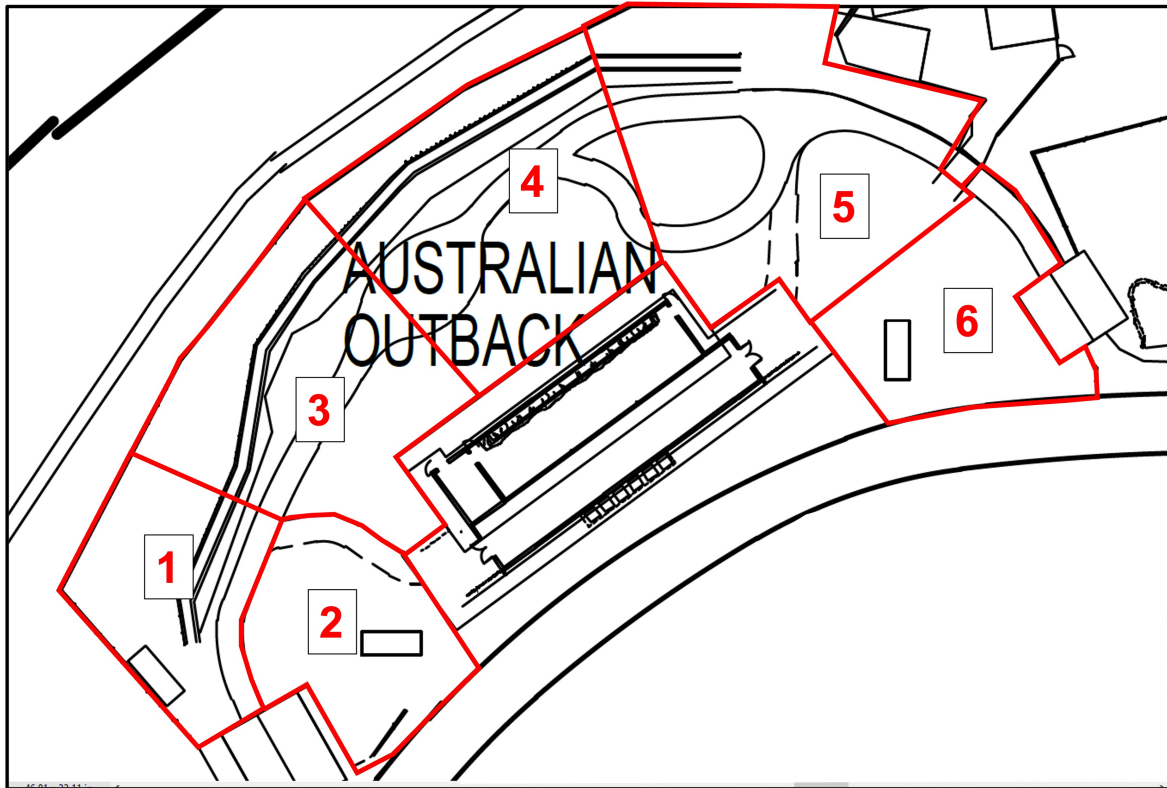


Figure 1: The separate zonation of the exhibit; description of zones are described in-text.  
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## Data analysis

Due to the presence of unequal zone sizes, the modified spread of participation (SPI) index (first described by Plowman 2003) was used to examine exhibit use across each zone for each species. As per Original SPI, the values for Modified SPI also vary between 0 and 1 where 0 describes near-perfect space-use of the enclosure while 1 describes the inverse (i.e., uneven space-use).

The formula for deriving the SPI index is simply as follow:

$$SPI = \sum \frac{|f_o - f_e|}{2(N - f_{e_{min}})}$$

Here,  $f_o$  refers to the number of times an animal was observed in a zone,  $f_e$  is the expected frequency for the zone (based on the respective size of each zone),  $f_{e_{min}}$  is the expected frequency in the exhibit's smallest zone and  $N$  refers to the number of observations for the observation period.

As the Modified SPI only provides a single index that characterises overall exhibit use, an Electivity Index (EI) was also included to describe exhibit use across each identified zone (see Figure 1). Originally conceived for use in ecology (Vanderploeg and Scavia 1979), the EI have been adapted to examine the spatial preference / selection of a broad range of captive animals in exhibits or enclosures under varying degrees of environmental enrichment (see review by Brereton and Fernandez 2021). As such, the inclusion of the EI further elucidates variation or differences in exhibit use following intrinsic (e.g., age) and/or extrinsic (e.g., inclusion of furniture) changes. In its computation, the EI for each identified zone is represented by a value ranging between 1 (indicating sole of the zone) and -1 (indicate no use of the zone) while a value of 0 indicates perfect proportional use of the indicated zone in relation to its size. Much like the modified SPI formula described above, the EI is also designed to overcome the challenges associated with unequal zone sizes (Brereton and Fernandez 2021).

The formula for deriving the EI is simply:

$$EI = \frac{W_i - \frac{1}{N}}{W_i + \frac{1}{N}}$$

$$W_i = \frac{\frac{r_i}{p_i}}{\sum \frac{r_i}{p_i}}$$

Here,  $r_i$  refers to the observed use of a resource or zone, and  $p_i$  refers to the expected use of a given resource. The letter  $N$  denotes the total number of zones ( $n = 6$ ; see Figure 1

or resources available to the study species.

To examine the variations and/or shifts in behaviour of each species post-introduction, species- and treatment-specific activity budgets and the Shannon’s Index of Diversity, or Shannon Index, were calculated. Here, the SI is used to examine the diversity of grouped behaviours exhibited by each species and has been applied various other animal-related studies (Menchetti et al. 2019).

The formula for computing the SI is simply:

$$SI = - \sum_{i=1}^B p_i \times \ln \times p_i$$

where  $B$  is the number of behaviour types ( $n = \leq 12$ ; see Table 1) and  $p_i$  is the proportion of behaviour  $i$ . Greater index values demonstrates a greater diversity in exhibited behaviour and index values increases linearly with number of behaviour types and abundances. As an index, the SD index is relatively accurate and useful indicator of positive welfare (see Miller et al. 2020) and can be used to simply compare between treatments. All analysis were performed with the *dplyr* package (Wickham et al. 2015) within the R environment (version 4.1.2).





# Results

## Enclosure use

Although the space use patterns of kangaroos remained relatively un-affected by the introduction of both wallaby species, the BWs exhibited a significant shift towards uneven space use after the introduction of the SWs (Figure 2). Seemingly, the change in space use patterns exhibited by the BWs resulted in SPI values that were comparable to the newly-introduced SWs.

In general, there is very little change observed in the GKs' EI values across the different zones of the exhibit except for the significant increase of utilisation in Zone 5 following the introduction of the SWs (Figure 3). On the other hand, the introduction of the SWs incurred remarkable changes in the BWs zonal use of the exhibit. For instance, the use of Zones 1, 5 and 6 exhibited substantial shifts as the utilisation of the aforementioned zones decreased significantly while the use of Zones 2 and 4 increased significantly (Figure 3). Lastly, the SWs exhibited a clear preference for Zone 1 while the EI value for the remaining zones demonstrated a relatively greater degree of under-utilisation.

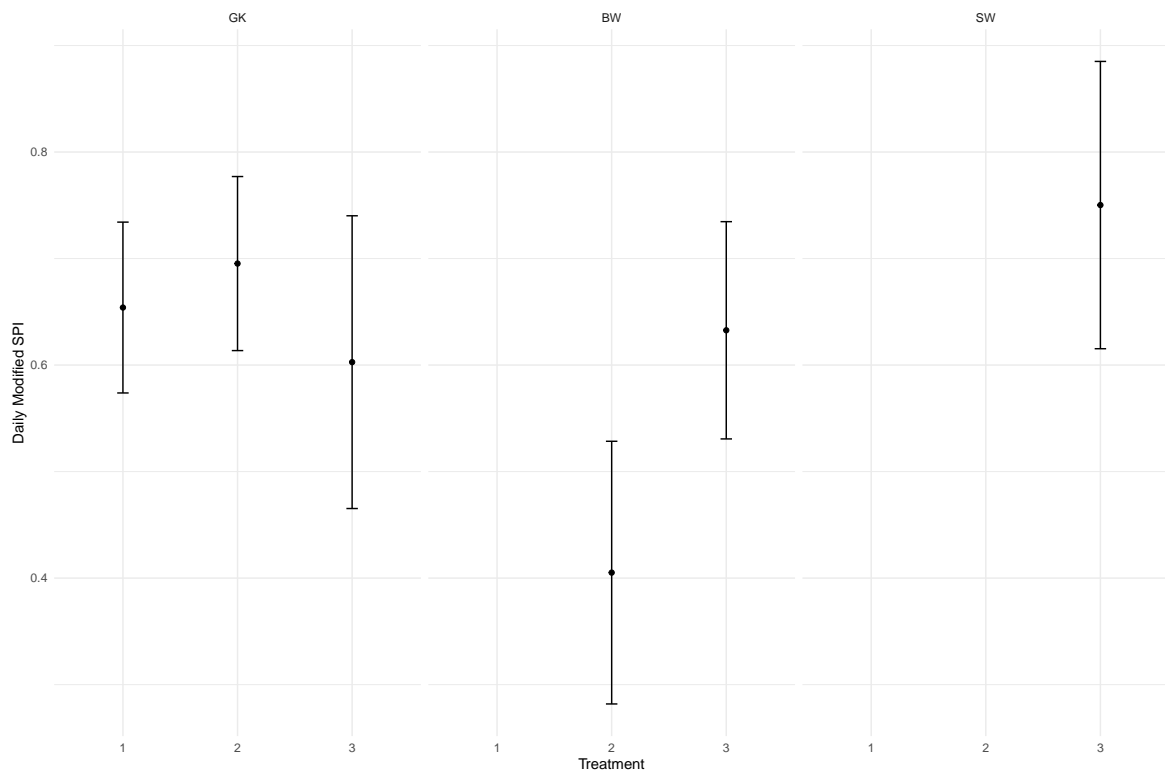


Figure 2: The Modified Spread of Participation Index (SPI) of Grey Kangaroos ( $n = 6$ ; GKs), Bennett's wallabies ( $n = 4$ ; BWs) and Swamp wallabies ( $n = 2$ ; SWs) describing spatial use of a mixed-species exhibit in the Singapore Zoo. Treatment levels are indicated on the x-axis.

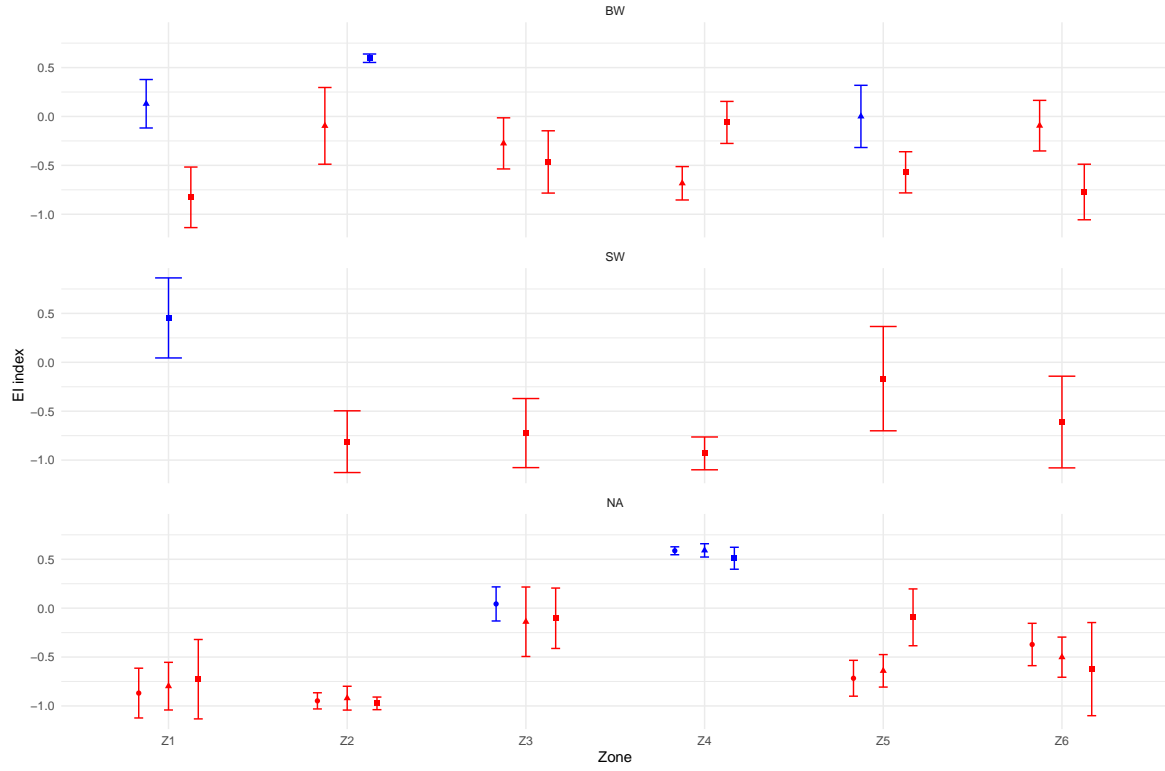


Figure 3: The Electivity Index values (EI; -1 to 1) of resident Ks and introduced BWs and SWs across six separate zones within a mixed-species exhibit. Different shapes are used to represent the different treatments: circles, triangles and squares are used to represent Treatments 1, 2 and 3 respectively. Here, degree of utilisation corresponds to blue and red color; EI values above 0 describes extent of utilisation (i.e., coloured blue) while EI values below 0 describes extent of under-utilisation (i.e., coloured red).

## Behavioural shifts

In general, all three macropod species were mostly engaged in resting behaviour with feeding following closely (Figure 4). Here, the introduction of the BWs did not incur any significant changes in the behaviour repertoire of the GKs. On the other hand, the introduction of the SWs resulted in a general decrease in locomotive and grooming behaviour in both GKs and BWs; BWs were also observed to rest less and engage in greater feeding and vigilant behaviours (Figure 4).

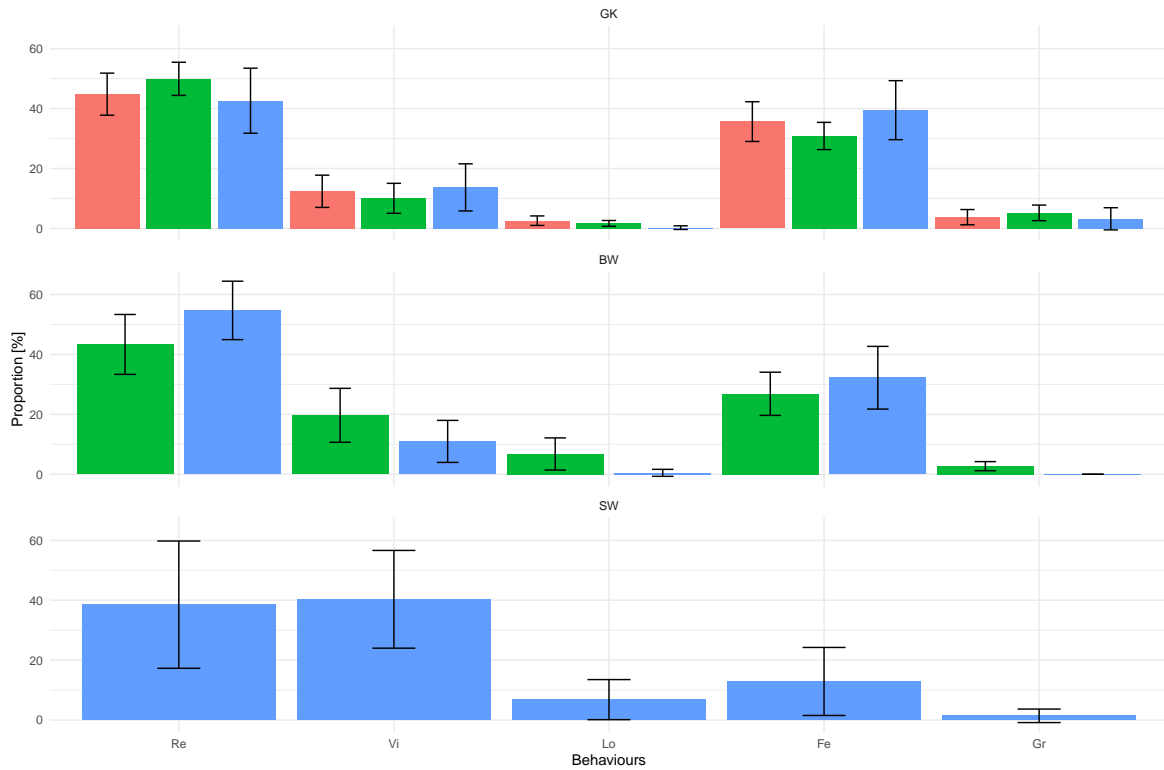


Figure 4: The activity budgets of Grey kangaroos (GK), Bennett's wallabies (BW) and Swamp wallabies (SWs) as computed across the different treatments (i.e., T1–T3). Treatment 1, 2 and 3 are colored red, green and blue, respectively. Simultaneous behaviours that occur  $< 1\%$  across all three treatments were removed (i.e., Dr, N, P, Nso, Pso, O).

Although the change in behavioural diversity have been largely insignificant across treatments, there is some evidence to suggest that the introduction of the SWs effected a notable decrease in the behavioural diversity of GKs and BWs (Figure 5). On the other

hand, the transition from T1 to T2 (i.e., introduction of the BWs) observed an small, albeit insignificant, increase in behavioural diversity in the GKs.

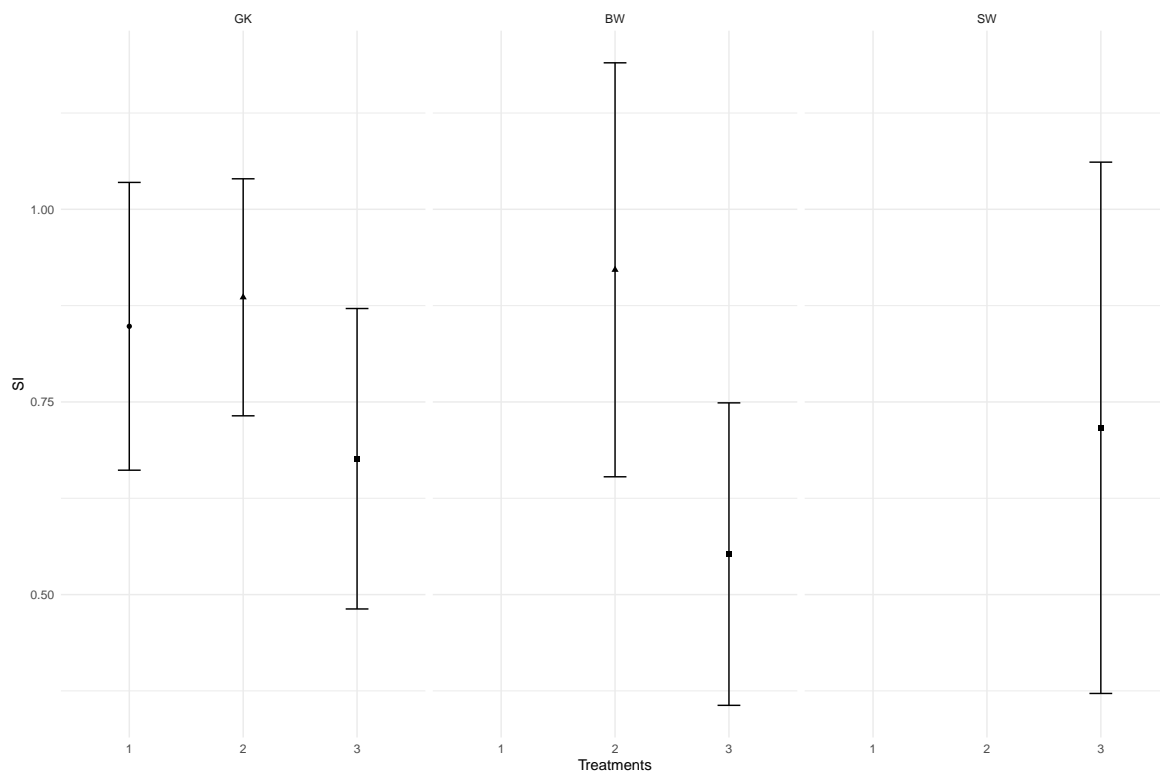


Figure 5: The daily Shannon Index (SI) adapted to examine behavioural diversity in Grey Kangaroos ( $n = 6$ ; GKs), Bennett's wallabies ( $n = 4$ ; BWs) and Swamp wallabies ( $n = 2$ ; SWs). Here, greater SI values describes a greater diversity in exhibited behaviour and is usually indicative of better animal welfare.



# Discussion

Discussion goes here

Some food for thought:

- Are there reports of aggression between species?
- Any change in BCS or increase in stress-related health issues?





# Conclusion

Conclusion goes here



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