

Reader-Guigueno Lab Meeting Nov. 4 2020,  
Project Analyses

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2020-11-02



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# Chapter 1

## Background

Based on previous literature we have good reason to believe two things about guppy exploratory behaviour, defined as the propensity to engage with novelty:

- that there is a relatively **strong environmental component to exploratory behaviour** (Burns *et al.* 2016)
- that **exploratory tendencies across contexts are correlated** (De Serrano *et al.* 2016)

My experiments try to examine whether there is a role for experience in the environment in shaping exploratory behaviour and to look at the implications of a potential behavioural correlation across novelty contexts. If exploratory tendencies are correlated across contexts because of shared mechanisms producing both behaviours then shifting exploration in one context should also shift exploratory tendencies in another context even without directly training for exploration in that context.

To tackle these questions I have tried to shift novel object exploration and spatial exploration through reinforcement training.

I took a stepwise approach to this. Before I asked whether I could shift novel object preferences I wanted to confirm that:

- Guppies recognize and respond to novel objects
- Guppies can have their preference for objects shifted

After confirming those I then felt comfortable to go on and see if I could shift *novel* object preferences.



## Chapter 2

# Project 1 — General Methods

**Question:** Can I shift the preference for a particular object by reinforcing an object with a food reward.

I had one set of guppies trained to the blue object (blue-trained guppies) and another set trained to the green object (green-trained guppies).

I performed the following trials:

- 1 baseline object preference test (Guppy in tank with 1 green and 1 blue object, unrewarded)
- 20 training trials (Guppy in tank with 1 green and 1 blue object, rewarded for visiting one or the other based on treatment)
- 1 final object preference test (Guppy in tank with 1 green and 1 blue object, unrewarded)





## Chapter 3

# Model 1 - Preference for the green object at baseline

```
baseline.data.model =  
  lm(green.object.preference ~ 1,  
     data = baseline.data)
```

At baseline, there was no significant difference in green object preference between the treatments ( $p = 0.727$ ).



## Chapter 4

# Model 2 - Preference for the rewarding object during training

To see whether fish were responsive during training our second model asks whether the preference for the rewarding object changes throughout training between the treatments.

```
training.data.model =  
  lmer(rewarding.object.preference ~ rewarding.object.colour * trial + (1 | id),  
        data = training.data)
```

```
## # A tibble: 3 x 6  
##   term                                estimate std.error statistic    df p.value  
##   <chr>                                <dbl>     <dbl>     <dbl> <dbl> <chr>  
## 1 rewarding.object.colourgreen         56.0        27.3      2.05   47.2 0.046  
## 2 trial                               7.70         1.08      7.10  438. < .001  
## 3 rewarding.object.colourgreen:trial  -0.549        1.49     -0.368 435. 0.713
```

Throughout training, over the 20 trials, guppies increased their relative preference for the rewarded object by 7.7 seconds each trial (Figure 4.1,  $p < .001$ ).

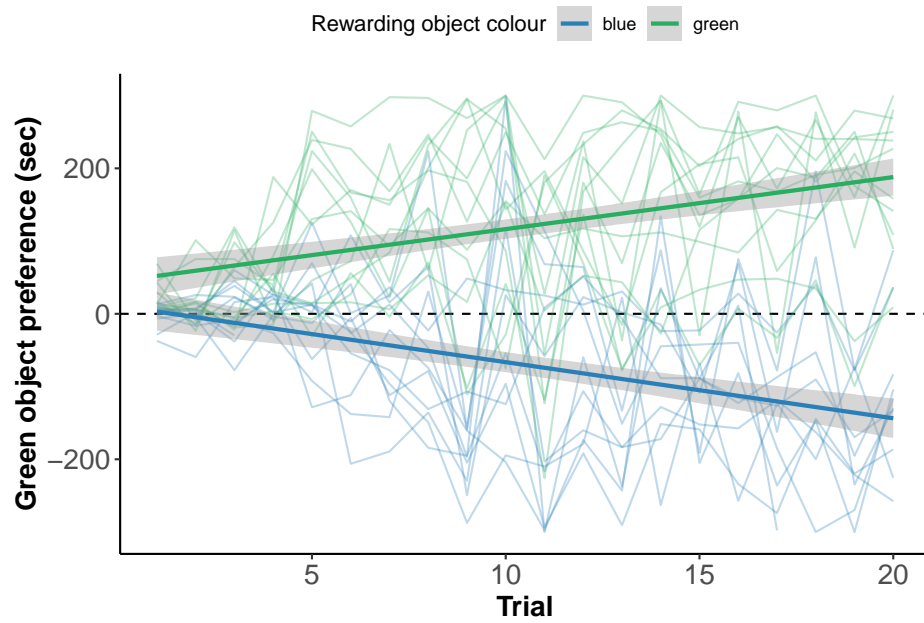


Figure 4.1: Preference for the green object in both treatments. Negative values represent more time spent with the blue object, positive values indicate more time spent with the green object. Faded lines connect individuals across trials and solid lines represents a linear fit with 95% CI (grey shading).

## Chapter 5

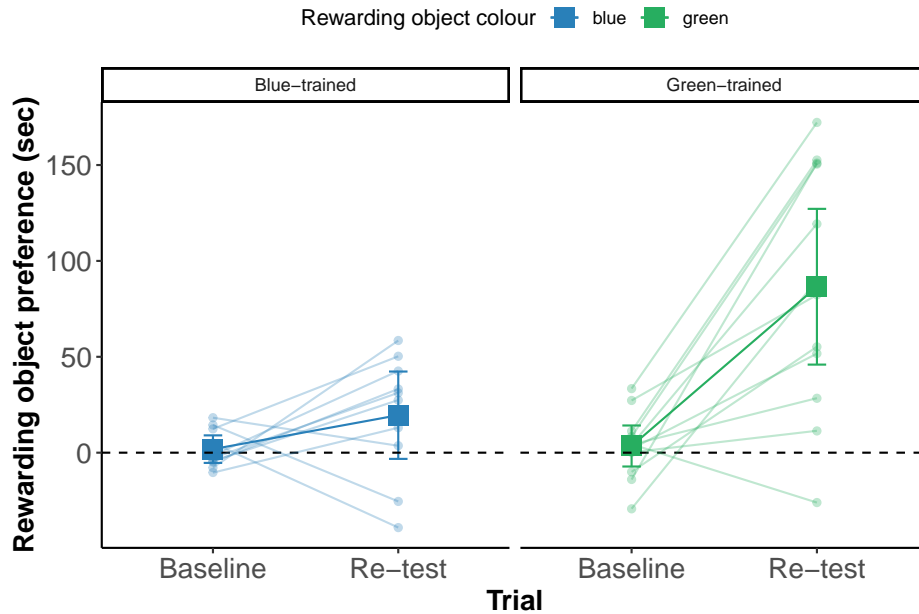
# Model 3 - Preference for the rewarded object during testing depending on treatment

For the main effects of **training** and **rewarding object colour** on object preference I fit a **linear mixed effects model** with **fixed effects of trial** and **rewarding object colour** and a random effect of **individual id**. My third model asks whether the preference for the rewarding object changed between baseline and final test and looks for an interaction with rewarded object colour.

```
test.data.model =  
  lmer(rewarding.object.preference ~ rewarding.object.colour * trial + (1 | id),  
    data = test.data)
```

```
## # A tibble: 3 x 6  
##   term                                estimate std.error statistic    df p.value  
##   <chr>                                <dbl>     <dbl>     <dbl> <dbl> <chr>  
## 1 rewarding.object.colourgreen         1.64       16.3      0.101  39.9 0.92  
## 2 trial21                             17.7       16.6      1.06   20.0 0.3  
## 3 rewarding.object.colourgreen:trial~  65.4       22.5      2.90   20.0 0.009
```

I found a significant interaction effect between trial and rewarding object colour ( $p = 0.009$ ). Guppies that had green rewarded had a rewarded object preference that was 65.4 seconds stronger than the rewarded object preference of guppies trained to blue (Figure ??).



Post-hoc pairwise comparisons investigating the differences between treatments based on whether guppies are untrained or trained reveals that initially there was no difference in the strength of preference for the rewarding object between the treatments (blue-trained guppies had a blue object preference of 1.8 seconds and green-trained guppies had a green object preference of 3.5 seconds,  $p = 1$ ).

Comparing the shift in rewarding object preference between initial and final preference tests in blue-trained and green-trained guppies reveals that the shift in rewarding object preference is significant for green-trained guppies but not for blue-trained guppies. Green trained guppies increased their preference for the green object by 83 seconds (going from a green object preference of 3.5 seconds initially to 86.5 seconds at final test,  $p < .001$ ) whereas blue-trained guppies non-significantly increased their preference for the blue object by 17.7 seconds (going from a blue object preference 1.8 seconds initially to 19.5 seconds at final test,  $p = 0.714$ ). For a full description of post-hoc comparisons see table 5.1.

1. Green-trained guppies increased their preference for the green object.
2. Blue-trained guppies non-significantly increased their preference for the blue object.

Table 5.1: Table of post-hoc tests with Tukey adjustment for multiple comparisons. The numbers represent the initial test trial (0) and the final test trial (21). The colour corresponds to the identity of the rewarding object (blue for blue-trained guppies, green for green-trained guppies). Values are all rounded to 3 decimal places. Significant p-values are bolded.

contrast	estimate	df	lower.CL	upper.CL	p.value
0 blue - 21 blue	-17.697	20.000	-64.236	28.842	0.714
0 blue - 0 green	-1.645	39.908	-45.381	42.092	1
0 blue - 21 green	-84.693	39.908	-128.429	-40.956	<b>** &lt; .001 **</b>
21 blue - 0 green	16.052	39.908	-27.684	59.789	0.759
21 blue - 21 green	-66.996	39.908	-110.732	-23.259	0.001
0 green - 21 green	-83.048	20.000	-125.532	-40.564	<b>** &lt; .001 **</b>





## Chapter 6

# Model 4 - Preference for the rewarded object during training based on feeding

My fourth model asks whether the time spent near the rewarding object during a training session is influenced by whether a fish ate or not.

```
training.data.model.rewarding.object =  
  lmer(rewarding.object.preference ~ ate + (1 | id) + (1 | trial),  
    data = training.data)
```

```
## # A tibble: 1 x 6  
##   term      estimate std.error statistic    df p.value  
##   <chr>      <dbl>    <dbl>    <dbl> <dbl> <chr>  
## 1 ateyes      91.2      11.0      8.33  224. < .001
```

Throughout all of training, fish that ate during a session spent on average 91.2 more seconds near the rewarded object compared to fish that did not eat (Figure, 6.1,  $p < .001$ ). Fish that spent more trials eating may therefore have received more reinforcement for the object-food association.

A discrepancy in reinforcement between treatments may influence performance on a final preference test. To see whether there was a difference in feeding between treatments I counted the number of trials in which an individual fish ate throughout all of training and compared the feeding counts between treatments. To do this I fit a generalized linear model with a Poisson distribution.

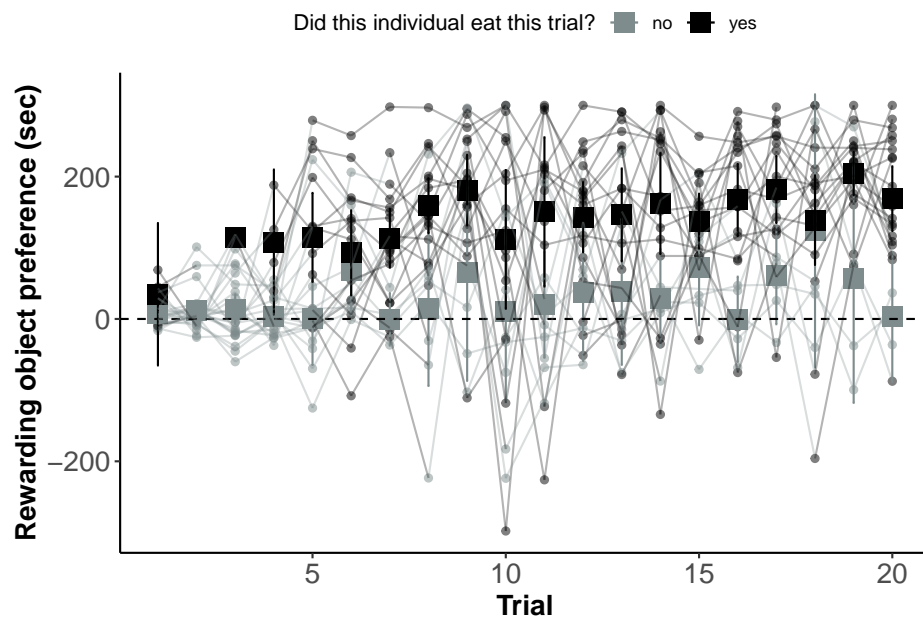


Figure 6.1: Preference for the rewarding object during training based on whether an individual ate during a trial or not. Dashed line represents the no preference value. Data are means  $\pm$  95% CI.

## Chapter 7

# Model 5 - Is there a difference in feeding attempts between treatments?

```
feeding.data.model =  
  glm(feeding.count ~ rewarding.object.colour, family = "poisson",  
      data = my.feeding.data)
```

The response variable ‘feeding count’ is a sum of the number of trials in which a guppy ate.

```
## # A tibble: 1 x 5  
##   term                                estimate std.error statistic p.value  
##   <chr>                                <dbl>     <dbl>     <dbl> <chr>  
## 1 rewarding.object.colourgreen    0.0710     0.126     0.561 0.575
```

I found no significant difference in the amount of feeding done by individuals trained to green versus individuals trained to blue (Figure 7.1,  $p = 0.575$ ) which suggests that the observed group-level differences in final test performance between blue-trained guppies versus green-trained guppies cannot be explained by differences in performance during training.

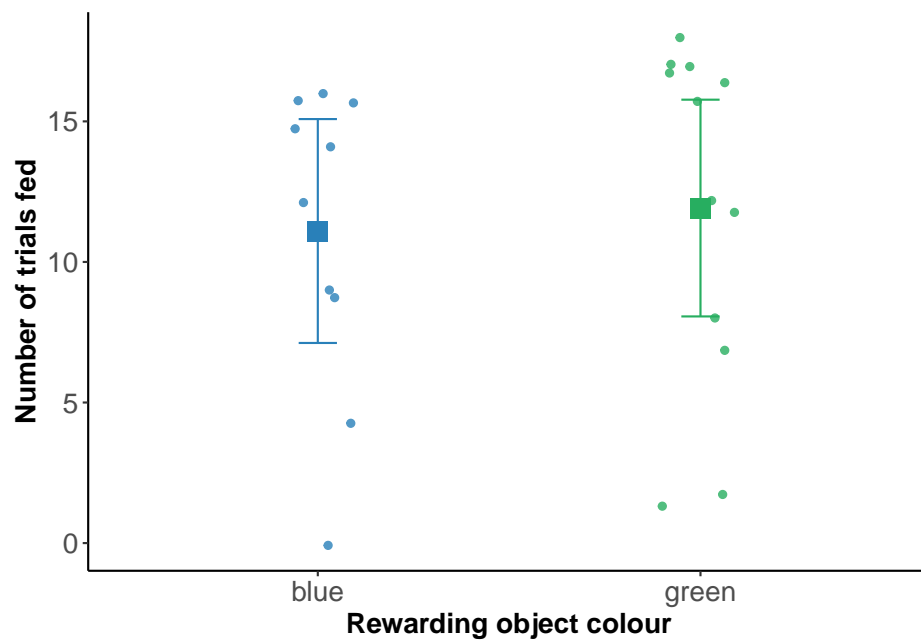


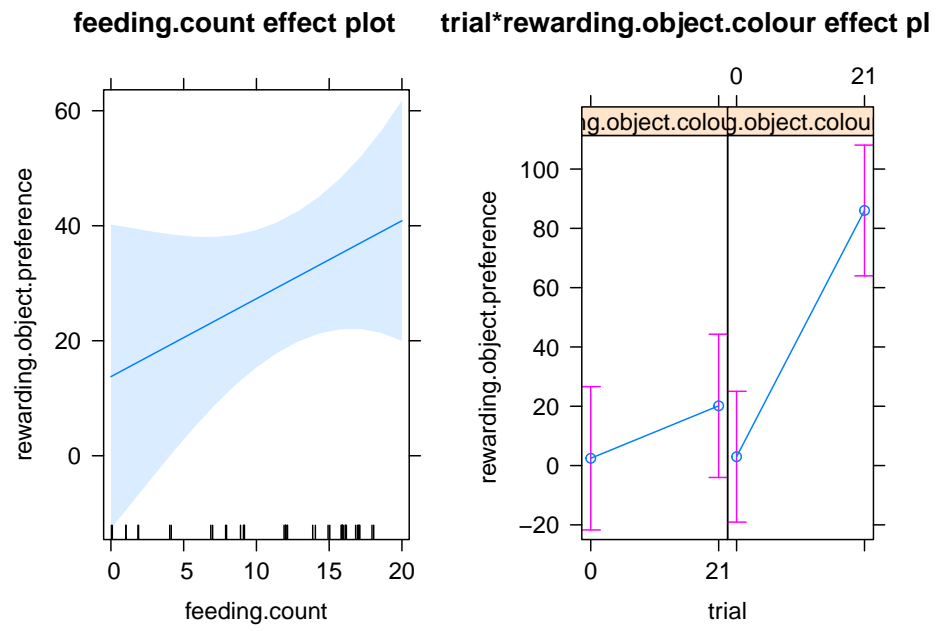
Figure 7.1: Average number of trials in which a fish fed during training. Data are means  $\pm$  95% confidence intervals

## Chapter 8

### Model 6 - What if I control for feeding count?

```
test.data.feeding.controlled.model =  
  lm(rewarding.object.preference ~ trial * rewarding.object.colour + feeding.count,  
      data = test.feeding.data)
```

```
## # A tibble: 4 x 5  
##   term                                estimate std.error statistic p.value  
##   <chr>                                <dbl>    <dbl>    <dbl> <chr>  
## 1 trial21                             17.7      16.9      1.05  0.301  
## 2 rewarding.object.colourgreen         0.538     16.2     0.0332 0.974  
## 3 feeding.count                       1.36      1.02     1.33  0.192  
## 4 trial21:rewarding.object.colourgreen 65.4      22.9     2.86  0.007
```



Nothing changes if I control for feeding count.

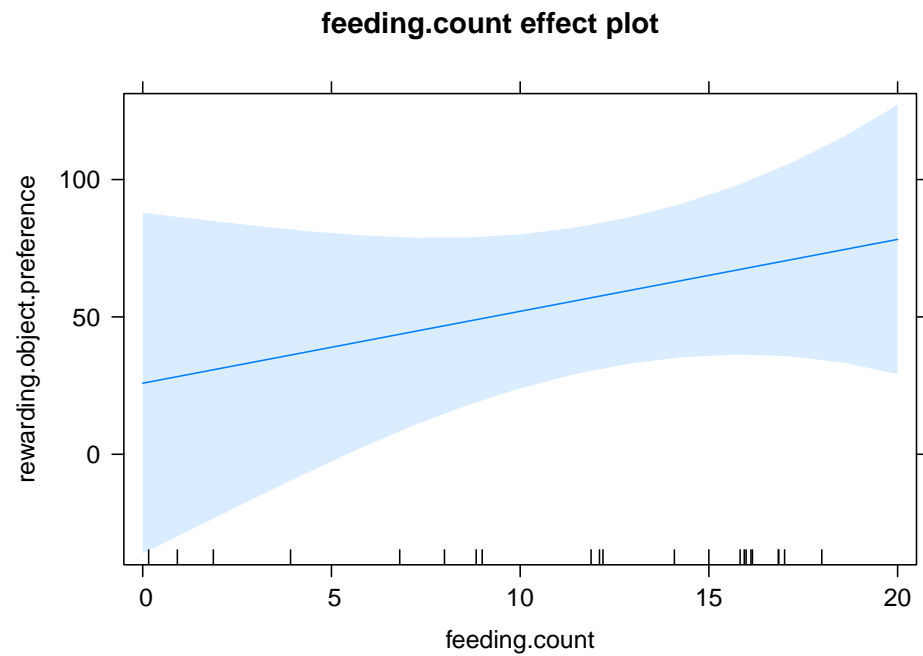
## Chapter 9

# Model 7 - Does feeding count predict anything?

```
test.data.feeding.controlled.model1 =  
  lm(rewarding.object.preference ~ feeding.count,  
     data = retest.feeding.data)
```

```
## # A tibble: 1 x 5  
##   term          estimate std.error statistic p.value  
##   <chr>          <dbl>    <dbl>    <dbl> <chr>  
## 1 feeding.count    2.62      2.32     1.13 0.272
```

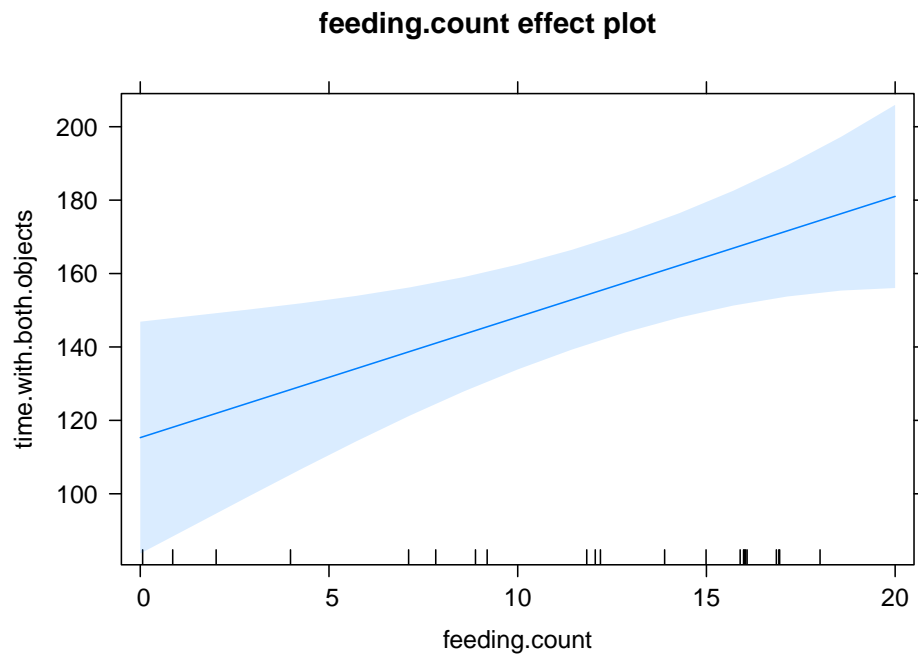
Testing for an effect of feeding count on rewarding object preference finds that there is no significant effect but the effect is in the expected direction.



However, there is an effect of feeding on the time spent near **both** objects at re-test.

```
test.data.feeding.controlled.model2 =  
  lm(time.with.both.objects ~ feeding.count,  
     data = retest.feeding.data)
```





During the final test, a fish that had 0 feedings spent 115.299 seconds near both objects whereas fish that fed in 20 trials spent 181.009 seconds near both objects, a 1.6-fold increase.



## Chapter 10

# An interesting trend from a very early pilot

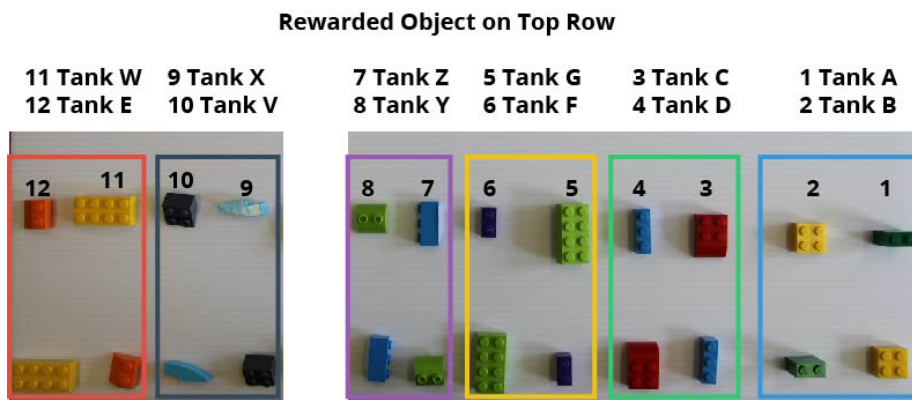


Figure 10.1: Objects from a very early experiment

```
pilot.model =  
  lmer(time.with.trained.object ~ trial * object.colour + (1 | id),  
        data = pilot.data)  
  
## # A tibble: 7 x 6  
##   term                                estimate std.error statistic    df p.value  
##   <chr>                                <dbl>     <dbl>     <dbl> <dbl> <chr>  
## 1 trial11:object.colourgreen         24.5        12.0      2.04    26.6 0.052  
## 2 trial11:object.colourgrey          0.300        16.1     0.0186    26.6 0.985
```

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```
## 3 trial11:object.colourorange 12.7      16.1    0.787   26.6 0.438
## 4 trial11:object.colourpurple  2.50      18.6    0.134   26.6 0.894
## 5 trial11:object.colourred    25.9      16.1    1.60    26.6 0.121
## 6 trial11:object.colourwhite  11.6      17.7    0.659   30.4 0.515
## 7 trial11:object.colouryellow -1.63     13.2   -0.124   26.6 0.902

## model: time.with.trained.object ~ trial * object.colour
##
## trial*object.colour effect
##      object.colour
## trial    blue   green   grey  orange  purple      red   white  yellow
##    0 56.22172 47.98031 39.63883 53.18540 46.2453 43.50927 54.78697 64.86343
##    11 46.11182 62.37222 29.82920 55.78797 38.6378 59.25800 56.31596 53.11867
```