Description of sample projects——

Zoology/Entomology/ Neuroscience

for The Observer XT



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The Observer XT Sample Projects

The Observer XT installation USB stick contains a folder **Documentation\Sample projects\Animal Sample Project** with a backup file (*.vpb) of one sample project (**Rat in open field.vpb**).

You can download a large number of other sample projects on the website of Noldus IT. These projects are examples of commonly used tests with The Observer XT.

The following sample experiments are available:

- Rat in open field page 5.
- Parental food provisioning in blue tits page 10.
- Wasp attacking caterpillars page 14.
- Broiler behavior page 18.
- Heart rate variability in horses page 22.
- Drosophila male-male competition page 28.

To download a sample project

- 1. Login to the website. To be able to do so you have to register.
- 2. Browse to www.noldus.com/downloads.
- Choose Downloads and documentation > The Observer XT 15 >
 Sample projects.
- 4. Download the sample project and the accompanying videos.

Install the Sample Projects

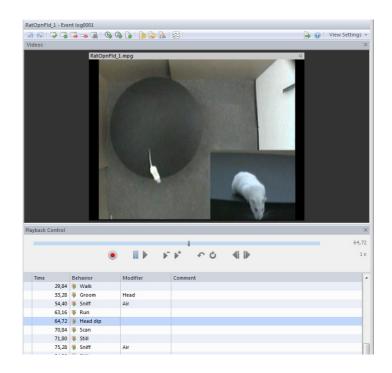
To open the sample experiments you must open the backup files:

- 1. Copy the sample project backup file (.vpb) to your computer.
- Copy the associated video files to the default Video Files folder of The Observer XT:
- 3. In The Observer XT, choose File > Restore Backup.
- 4. Browse to the backup file that you want to restore. Optionally click Browse and select another experiment location. By default the project is saved on
 - C:\Users\Public\Documents\Noldus\The Observer XT\Projects
- 5. Click Open to restore the experiment in this location.

If you open a sample project with external data, but do not have a license for that function, you can open the project, visualize and make calculations with the event data, but not visualize or select or calculate with the external data.

Rat in open field

The behavior of a rat in a wall-less open field is coded from video and analyzed.



Video file

Rat in Open Field.mpg

SETUP

Choose **Setup** > (choose one of the options below).

Coding Scheme

One mutually exclusive behavior group with 14 behaviors. The behavior *Groom* has a modifier group *Body part* with modifiers *Head* and *Body*. The behavior *Sniff* has a modifier group *Object* with modifiers *Substrate* and *Air*.

Independent variables

The values of the independent variables are hypothetical. This sample project serves as an example of how to analyze observational data files by treatment, dosage, etc. The project contains the following independent variables:

- Rat ID.
- Sex of the rat: Female or Male.
- · Age (in weeks).
- Treatment: Control or Treated.
- Drug: name of the drug.
- Dose: dose of the drug.
- Observer: Initials of the person who did the observations.
- Time of day: Morning or Afternoon.
- · Group ID.

OBSERVE

Choose Observe > Observation > Open. Choose one of the following:

- Rat in Open Field_1 Original observation of the video Rat in open field.mpg.
- Rat in Open Field_1_edited This is used for comparison with the original observation in reliability analysis.

This sample project shows a rat in a circular, wall-less open field arena. An overhead camera shows spatial displacement, while a close-up

camera allows scoring of the details of sniffing, grooming, etc. Both signals have been mixed into a single video file.

If the video file is not found

If, after opening an observation, The Observer XT cannot find the corresponding video file, a window appears on top. Browse to the video file, and click **Open**.

To change the default Video Files folder, choose **File > Preferences > Files locations**

ANALYZE

Data profiles

Choose **Analyze** > **Select Data** > **Open Data Profile**. Choose one of the data profiles listed below and click **OK**.

- **Default data profile** The default data profile with all data.
- Locomotion behaviors merged In this data profile the behaviors Walk, Run, Rear, and Jump are merged in a Locomotion group. The other behaviors are analyzed separately.
- Head dips This data profile defines intervals based on the behavior Head dips. You can use this profile to visualize the time when Head dips was scored in the observation Rat in Open Field_1.
 This data profile was used to create the Episode selection Head dips.
- Sniffing > 5 s This data profile contains a filter on observation *Rat* in *Open Field_1* (first Filter box) and a filter on the behavior *Sniffing* (second Filter box). A **By duration** filter (third Filter box) is added, to only analyze the events in which *Sniffing* lasted longer than 5 s. This is an example of how you can refine your data selection by taking only events whose duration is significant for your study.

Episode selection

Choose Analyze > Episode Selection > Open.

The episode selection **Head dips** opens. This is based on the data profile with the same name. Play the episode selection to view all video fragments in which head dips took place.

Analysis results

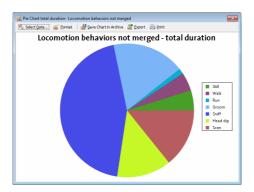
The project contains archived analysis results. To open these analysis results, choose **Analyze** > [analysis type] > **Open Archive**, and open an *.arx file.

- Behavior analysis result Locomotion behaviors merged. This result
 is based on the data profile with the same name (see above). It
 shows statistics of duration (mean duration of instances, and total
 duration) and frequency (total number of occurrences and rate per
 minute) of the behaviors scored. Locomotory behaviors that have
 been scored (Walk and Run) being merged in one result row for
 clarity.
- Behavior analysis result Locomotion behaviors not merged. This result is based on the default data profile. It shows statistics of duration (mean duration of instances, and total duration) and frequency (total number of occurrences and rate per minute) of the behaviors scored. Note that locomotory behaviors scored in the observation Walk and Run are shown in separate rows.
- Sniffing longer than 5 s. This is based on the data profile with the same name. It shows the same statistics as above, but only for Sniff, and only for the events that lasted longer than 5 s.
- Lag sequential analysis result. This result show how many times the behavior listed on the first column is followed by the behaviors indicated in the other columns. For example, *Head dip* is followed by *Sniff* four times. This result is based on the default data profile.
- Reliability analysis result. This quantifies how similar the two observations are. Click Comparison List to see where the mismatches in the scores occur.

Charts

Below each of the two analysis results **Locomotion behaviors merged** and **Locomotion behaviors not merged** you find a chart:

- Pie chart total duration Locomotion behaviors merged Pie chart based on the behavior analysis result with the same name showing the total duration of the behaviors, Still, Groom, Sniff, Head dip, Scan, and Locomotion (Merged).
- Pie chart total duration Locomotion behaviors not merged Pie chart based on the behavior analysis result with the same name showing the total duration of the behaviors, Still, Walk, Run, Groom, Sniff, Head dip and Scan.

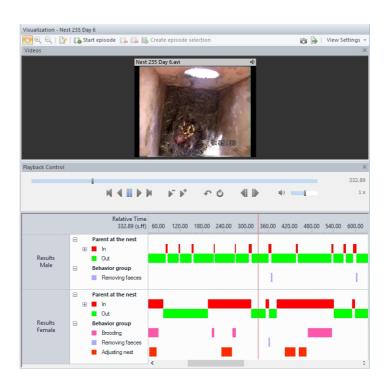


Note

The spatial displacement and behaviors like grooming and rearing can also be tracked automatically with **EthoVision XT**, which is able to acquire data from digital media files or from a live camera image.

Parental food provisioning in blue tits

This sample project deals with the food-provisioning behavior of a small insectivorous bird, the blue tit (*Parus caeruleus*), when raising a brood of hungry nestlings. It illustrates how you can record and analyze numerical data in The Observer XT together with nominal data.



Video files

- Nest 235 Day 6.avi
- Nest 275 Day 12.avi

SETUP

Choose **Setup** > (choose one of the options below).

Independent variables

The project contains the following independent variables:

- Hatching date.
- · Brood size.
- · Age of adult.

Coding scheme

The coding scheme includes two subjects (*Male* and *Female* parents) and two behavior groups:

- Parent at the nest Specifies whether the subject is at the nest box (with mutually-exclusive states In and Out. The behavior In has two modifier groups attached: Prey type (with elements specifying the type of food brought to the nest) and Prey size (numerical, only when Prey type= Caterpillar).
- Behavior group Contains:
 - Brooding The female parent broods the young,
 - **Removing faeces** The parent takes the youngs' faeces out of the nest.
 - Adjusting nest The female parent shakes the nest cup.

OBSERVE

Choose Observe > Observation > Open. Choose one of the following:

- Nest 235 day 6.
- Nest 275 day 12.

ANALYZE

Data profiles

Choose Analyze > Select Data > Open Data Profile. Choose one of the data profiles listed below and click OK.

- All data Includes all events of the two observations.
- Prey size Contains only the events with behavior In (parent entering the nest) and Prey type= Caterpillar. Use this data profile to analyze prey size values.
- Provisioning rate (overall) Like Prey size, with the difference that all prey types are selected and grouped. Use this profile to analyze the frequency of feeding independent of prey type.
- Provisioning rate per prey type All prey types are selected, not grouped.

Analysis results

The project contains archived analysis results. To open these analysis results, choose Analyze > [analysis type] > Open Archive, and open an *.arx file.

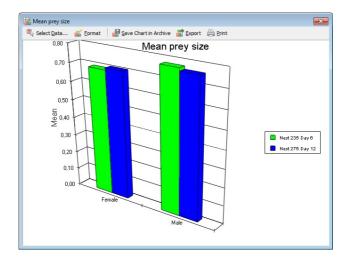
- Behavior analysis result Provisioning rate (overall) based on the data profile with the same name.
- Behavior analysis result Provisioning rate per prey type based on the data profile with the same name.
- Numerical analysis result *Prey size* based on the data profile with the same name.

Charts

The sample project contains four charts:

• Mean prey size – a bar chart based on the numerical analysis result Prey size showing the mean size of the caterpillars which the female and male parent bring to the nest. As you can see the difference between the two nests is very small. In both nests the

male parent brings bigger caterpillars to the nest than the female parent.



- Provisioning rate (overall) a bar chart based on the behavior analysis result with the same name showing the difference in provisioning rate between the male and the female parent for the two nests.
- Nest 235 Day 6 a bar chart based on the behavior analysis result

 Provisioning rate per prey type showing the rate at which the male
 and the female parent of nest 235 provide the different prey types.
- Nest 275 Day 12 similar as the previous one but then for nest 275.

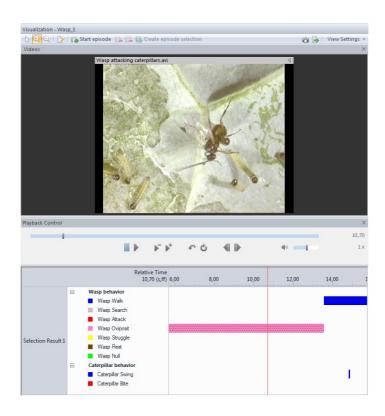
References

Grieco F. 2001. Short-term regulation of provisioning rate and effects on prey size in blue tits. Animal Behaviour 62, 107-116.

Grieco F. 2002. Time constraint on prey choice in breeding blue tits Parus caeruleus: the relationship between provisioning rate and prey size. Animal Behaviour 64, 517-526.

Wasp attacking caterpillars

This sample project deals with the foraging behavior of a parasitic wasp (*Cotesia glomerata*) attacking caterpillars (*Pieris brassicae*). With its distinct "attack-oviposit-attack" cycle, *C. glomerata* shows its adaptation to gregarious hosts. The coding scheme has been slightly modified from that of Wiskerke & Vet (1994).



Video file

Wasp attacking caterpillars.avi.

SETUP

Choose **Setup** > (choose one of the options below).

Coding Scheme

The Coding scheme consists of two behavior groups and one modifier group:

- Wasp behavior Mutually exclusive behavior group with the wasp behaviors as state events.
- Caterpillar behavior Start-Stop behavior group with the caterpillar defense behaviors as point events.
- Target Modifier group linked to the behavior Wasp antennae

Independent variables

The project contains the following independent variables:

- Number of caterpillars.
- Temperature.
- · Humidity.

OBSERVE

Choose Observe > Observation > Open. Choose one of the following:

- Wasp_1 the original data file, meant for review and data analysis.
 You are advised not to edit this file.
- Wasp_2 an exact copy of Wasp_1, which you can use for event recording, to add extra events, etc.

ANALYZE

Data profiles

Choose **Analyze** > **Select Data** > **Open Data Profile**. Choose one of the data profiles listed below and click **OK**.

- **Default Data Profile** includes all data of the two observations.
- Wasp_1 only contains only the data from Wasp 1.

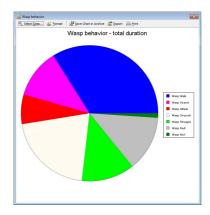
Analysis results

The project contains archived analysis results. To open these analysis results, choose **Analyze** > [analysis type] > **Open Archive**, and open an *.arx file.

- Behavior analysis result Wasp_1 only based on the data profile
 with the same name.
- Lag sequential analysis result Wasp_1 only based on the data profile with the same name.

Chart

Wasp behavior – Pie chart based on the behavior analysis result *Wasp_1 only* showing the duration of the different types of behavior of the wasp during the observation.



Reference

Wiskerke, J.S.C. & Vet, L.E.M. (1994). Foraging for solitary and gregariously feeding caterpillars: a comparison of two related parasitoid species (Hymenoptera: Braconidae). Journal of Insect Behavior 7, 585-603.

Acknowledgment

Video recording by courtesy of Hans Smid, Department of Entomology, Wageningen University, Wageningen, The Netherlands.

Broiler behavior

This sample project shows the behavior of a group of broilers (that is, meat-type chickens) in their home pen. It shows how you can use the Observation method **Combine Continuous and Instantaneous Sampling.** The behavior of the bird indicated by the red arrow in the figure below was scored continuously, to get a complete record of its behavior. The other birds were monitored instantaneously without caring about their identity to get an average time budget per pen.

The aim of this experiment was to monitor the behavioral development of two types (a fast-growing type and a slow-growing type) of broilers in their home pen.

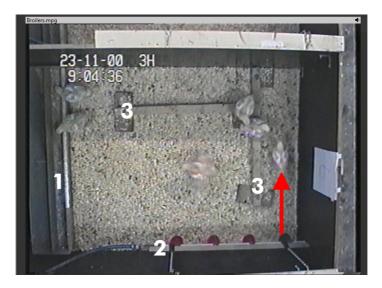


Figure 1 First frame of the media file Broilers.mpg. The red arrow indicates the focal bird. 1=feed trough, 2=drink nipples+cups, 3=perch.

Video file

Broilers.avi.

SETUP

Choose **Setup** > (choose one of the options below).

Project setup

Observation method – Combine Continuous and Instantaneous Sampling.

Observation duration – Observation duration = 3 minutes, Sample interval length = 15 seconds.

Coding Scheme

The Coding Scheme includes 6 subjects: one focal subject which is scored continuously and five subjects which are scored instantaneously.

The Coding Scheme contains three mutually exclusive, exhaustive behavior groups:

- Locomotion With different forms of locomotory behavior.
- Pecking With pecking at different objects.
- Ingestion With eating and drinking.

The three behavior groups all contain state events which are scored as sample points for the five instantaneous subjects.

Independent Variables

The project contains user-defined independent variables:

- Pen number
- Focal bird ID
- · Name of the observer

OBSERVE

Choose Observe > Observation > Open > Observation 1.

ANALYZE

Data Profiles

Choose Analyze > Select Data > Open Data Profile. Choose one of the data profiles listed below and click **OK**.

- All data the default data profile.
- Filter Locomotion with all behaviors from the behavior group Locomotion.

Behavior Analysis

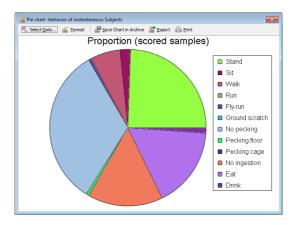
The Behavior Analysis Results contains two archived results. The project contains archived analysis results. To open these analysis results, choose Analyze > Behavior analysis > Open Archive, and open an *.arx file.

- Locomotion focal subject.arx This archive shows the behavior analysis results with data profile Filter Locomotion. The statistics are Mean and Total duration, which are calculated for the focal subject only. The behavior analysis contains an archived chart Pie chart - locomotion focal subject with the mean duration of the locomotion behaviors of the focal subject.
- All behaviors instantaneous subjects.arx This archive contains the results averaged for the instantaneous subjects. This was achieved by de-selecting **Subjects** in the **Layout** tab. The displayed results are:

Proportion (all samples), Scored Samples, Total number (all samples) - These are statistics from the instantaneous sampling category, so they apply to the instantaneous subjects only.

Example — The **Proportion (all samples)** for the behavior *Stand* is 0.73 (= 44 scored samples for Stand for all instantaneous subjects / 60 total number of samples for all instantaneous subjects).

The behavior analysis contains an archived chart *Pie chart - behavior of instantaneous subject.*



References

Bokkers, E.A.M. and Koene, P. (2003). Behaviour of fast- and slow growing broilers to 12 weeks of age and the physical consequences, Applied Animal Behaviour Science 81: 59-72.

Bokkers, E.A.M., Zimmerman, P.H., Rodenburg, T.B., and Koene, P. (2007). Walking behaviour of heavy and light broilers in an operant runway test with varying durations of feed deprivation and feed access, Applied Animal Behaviour Science 108: 129-142.

Acknowledgments

Video recording by courtesy of Dr. E.A.M. Bokkers, Wageningen Institute of Animal Sciences, Wageningen University and Research Centre, The Netherlands.

Heart rate variability in horses

This sample project shows the behavior of a horse in time periods of 5 minutes before, during and after it is exposed to another horse. Simultaneously with collecting the behavioral data, the heart rate of the horse was measured. This project is an example of the possibilities of importing and calculating with external data in The Observer XT. In the Observer XT you can make a selection of your data based on the behavior of your test animals and perform an analysis on the associated external data. And reverse, you can select data based on values of the external data and analyze the behavior of your test animals in those time periods. The name of the subject horse is URL and the observations took place on 3-6 and 15-7.



Figure 2 A novel horse is introduced to the test horse.

Video files

- URL 3-6.mpg
- URL 15-7.mpg

SETUP

Choose **Setup** > (choose one of the options below).

Coding Scheme

The Coding Scheme includes two behavior groups. One is a mutually exclusive, exhaustive group with different behaviors. The second is a group with the behavior *Interbeat interval*. See below under Interbeat intervals for an explanation of this group. Both groups contain state events.

Independent Variables

The project contains the user-defined independent variables:

- Date.
- Name subject horse.
- Name novel horse.

OBSERVE

Choose **Observe** > **Observation** > **Open**. Choose one of the following:

- URL 3-6 a Before encounter with novel horse.
- URL 3-6 b During encounter with novel horse.
- URL 3-6 c After encounter with novel horse.
- URL 15-7 a Before encounter with novel horse.
- URL 15-7 b During encounter with novel horse.
- URL 15-7 c After encounter with novel horse.

Interbeat intervals

The interbeat heart rate interval lengths were imported into the Observer as behaviors. For this purpose the raw data were edited. A column with time stamps was created, based on the interval lengths. A column with the behavior *Interbeat interval* and a column with the

alternating numerical modifiers 1 and 2 was added to the raw data (see Figure 3).

header header header header header header header header header header header	cal Modifier
header header header header	
header header header header	
0.00 3159 Interbeat interval	1
3.16 1665 Interbeat interval	2
4.82 1478 Interbeat interval	1
6.30 1101 Interbeat interval	2
7.40 970 Interbeat interval	1
8.37 937 Interbeat interval	2

Figure 3 Raw data file of interbeat intervals, edited for importing into The Observer.

Subsequently, the data were imported into the observations as observational data. The option **Treat new Behaviors as State events** was selected. Figure 4 shows a visualization of the interbeat intervals in The Observer XT.

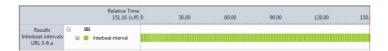


Figure 4 Visualization of Interbeat intervals in the Observer.

External data

From the interbeat interval data, the heart rate (beats per minute) over intervals of 10 seconds was calculated. These data were imported into

the Observer as external data. Figure 5 shows a visualization of the heart rate data.



Figure 5 Visualization of heart rate data in the Observer.

ANALYZE

Data profiles

Choose Analyze > Select Data > Open Data Profile. Choose one of the data profiles listed below and click **OK**.

- All data with all data selected.
- Before, during and after encounter IBI From the list of behaviors, interbeat interval has been selected. The data profile contains three result containers:
 - Before encounter with novel horse.
 - During encounter with novel horse.
 - After encounter with novel horse.
- **Before, during and after encounter behavior** –This data profile is similar to the previous one. Other behaviors have been selected.
- **Heart rate > 100 beats per minute** with the time intervals in which the heart rate of the horse was above 100 bpm.
- Intervals with Frolic locomotion with the time intervals in which the horse showed frolic locomotion.

Episode selection

Choose Analyze > Episode Selection > Open.

The episodes were selected in which the heart rate of the horse was above 100 bpm.

Analyses

The project contains archived analysis results. To open these analysis results, choose Analyze > [analysis type] > Open Archive, and open an * arx file

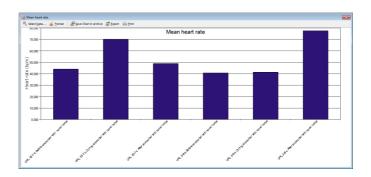
The project contains four archived behavior analysis results:

- Behavior when the heart rate was above 100 bpm contains the Mean duration. Standard deviation and Total duration of the behavior of the horse over time periods in which its heart rate was above 100 bpm. This archived analysis also contains a bar chart of the behavior.
- Effect of novel horse on behavior contains the Mean duration, Standard deviation and Total duration of the behavior of the horse in the time periods before, during and after a novel horse was introduced.
- Effect of novel horse on heart rate contains an analysis result on the interbeat interval data in the time periods before, during and after a novel horse was introduced. This archived analysis also contains bar charts of the mean interbeat interval lengths and the standard deviation of the interval lengths. The results show that the interbeat interval length is lower during and after exposure to the novel horse. This indicates that the horse is most at ease in the periods before introduction of the novel horse.
- Interbeat interval length during frolic locomotion contains the Mean, Total duration, Standard deviation and the Total number of intervals of the interbeat interval length of the horse over time periods when the horse showed frolic locomotion. This archived analysis also contains a bar chart of the interbeat interval length.

The project contains two archived numerical analysis results:

• Heart rate during frolic locomotion – contains the minimum value, maximum value, mean value and total number of samples of the heart rate (bpm) over time periods when the horse showed frolic locomotion. This archived analysis also contains a bar chart of the mean heart rates.

• Numerical analysis of heart rate data – contains the minimum value, maximum value, mean value and total number of samples of the heart rate (bpm) of all observations. This archived analysis also contains a bar chart of the mean heart rates.



Exporting selections of data, based on the behavior of the horse

With a data profile you select fragments of your external data and export these data to another program. With the data profile *Intervals by Frolic locomotion* you can for example select the interbeat interval data for the time periods in which the horse showed frolic locomotion. You can export these data and import them into another program to calculate other statistics like the heart rate variability or the LF/HF ratio.

Reference

E. de Graaf-Roelfsema, 2007. Endocrinological and behavioural adaptations to experimentally induced physical stress in horses.

Dissertation Utrecht University, Faculty of Veterinary Medicine

Acknowledgements

Noldus IT is thankful to Dr. M.C. van Dierendonck (Equus Research) for letting us use video material, observational data and heart rate data of her research

Drosophila male-male competition

In this project the behavior of two strains of Drosophila males towards females is followed. The males were bred for over a hundred generations under one of the following two conditions.

- Males M were kept Monogamous: 1 male with 1 female.
- Males P were kept Polygamous: 6 males with 1 female.

Continuous sampling was used as observation method. In each observation a female Drosophila was kept together with a monogamous male and a polygamous male. At random one of the males was marked with a yellow spot. The behavior of both males was observed. This way it was investigated whether natural selection on monogamy or polygamy led to differences in behavior of males towards females and competing males.

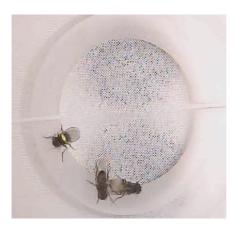


Figure 6 Set-up with two males and a female (on the right). One of the males is marked with a yellow spot.

Video files

- Video12.avi
- Video29.avi
- Video32.avi

SETUP

Choose Setup > (choose one of the options below).

Coding Scheme

The coding scheme includes two subjects, the polygamous male *Male Poly* and the monogamous male *Male Mono*. There is one mutually exclusive behavioral group *Males* with the following behaviors:

- · Approach/follow.
- · Courtship song.
- Intimidation.
- Attack.
- · Nothing.

There is a modifier group *Receiver* linked to the behaviors *Approach/follow*, *Courtship song*, *Intimidation* and *Attack*. This modifier group contains the following receivers:

- · Male Marked.
- Male Not Marked.
- Female.

Independent Variables

In each observation, one of the males was marked at random with a yellow spot. The independent variables list contains the user defined variable *Treatment*. This column shows whether the monogamous or polygamous male was marked.

OBSERVE

Choose Observe > Observation > Open. Choose one of the following:

- Male-Male Observation 12.
- Male-Male Observation 29.
- Male-Male Observation 32.

Data Profiles

Choose Analyze > Select Data > Open Data Profile. Choose one of the data profiles listed below and click **OK**.

- All data the default data profile
- Approach/follow to male/female in which the data were filtered by Approach/follow to Male and Approach/follow to Female
- Attack to male/female in which the data were filtered by Attack Male and Attack Female
- Courtship song to male/female in which the data were filtered by Courtship song to Male and Courtship song to Female
- Intimidation to male/female in which the data were filtered by Intimidation to Male and Intimidation to Female
- Filter Approach/follow in which all Approach/follow behavior of both males was included.
- Filter Attack in which all Attacks of both males were included.
- Filter Courtship song in which all Courtship songs of both males were included.
- Filter Intimidation in which all Intimidating behavior of both males was included.

Episode selection

Choose **Analyze** > **Episode Selection** > **Open** and choose one of the episode selections.

- · Approach-follow.
- Attack.
- · Courtship song.
- Intimidation.

ANALYZE

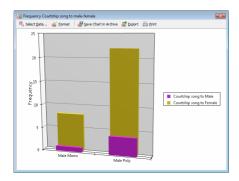
The project contains archived analysis results. To open these analysis results, choose **Analyze** > [analysis type] > **Open Archive**, and open an * arx file.

Behavior analysis

The project contains four archived behavior analyses:

- Approach-follow to male-female with the Mean and Total duration, Frequency and Latency of the events Approach/follow to either the other male or the female. This archive also contains a chart of the analysis results. The polygamous males Approached/ followed the females more often, but the approaches were shorter in comparison to the monogamous males.
- Attack to male-female with the Mean and Total duration,
 Frequency and Latency of the events Attack to either the other male or the female. Only the polygamous males attacked and they attacked only the other males.
- Courtship song to male-female with the ean and Total duration,
 Frequency and Latency of the events Courtship song to either the
 Male or the Female. This archive also contains two charts of the

analysis results. The polygamous males sang to the females more often than the monogamous males.



• Intimidation to male-female – with the Mean and Total duration, Frequency and Latency of the events Intimidation to either the Male or the Female. This archive also contains a chart of the analysis results. Both males only intimidated the other male. The polygamous male intimidated more often.

Lag sequential analysis

The project contains an archived lag sequential analysis which, for example, shows that the behavior *Approach/follow* to female is often followed by *Courthsip song* to female.

Acknowledgments

Acknowledgements for use of project to Allan Debelle, University of Sheffield, United Kingdom.