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The Sequence Analysis Toolkit for the advanced analysis of EthoVision XT data

Patrick Zimmerman, Wil van Dommelen
Noldus Information Technology
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Instructor

Patrick Zimmerman

- Behavioral Research Consultant at Noldus IT
  - Consultant for behavioral research
  - Documentation specialist
  - Trainer

- Ask questions during Tutorial / contact me at patrick@noldus.nl

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Co-author

Wil van Dommelen

- Wrote the Sequence Analysis Toolkit
- EthoVision project leader at Noldus IT

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Take-home message

Sequence Analysis Toolkit is a useful tool to

- Analyze the sequence of zone visits / behavioral states in EthoVision XT export data files
- Find specific sequences of zone visits / behavioral states
- Get additional information about EthoVision's behavioral states
- Allows export of sequences of zones / behavioral states to The Observer XT

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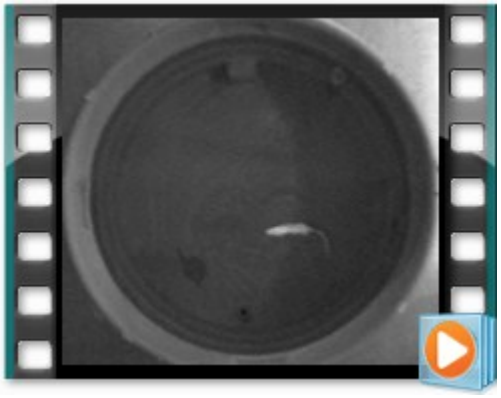
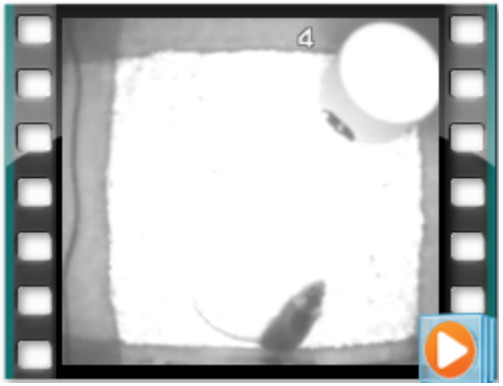
Take-home message

Applications of the Sequence Analysis Toolkit





When do I use the Sequence Analysis Toolkit?

What's the answer to Life, the Universe and Everything?

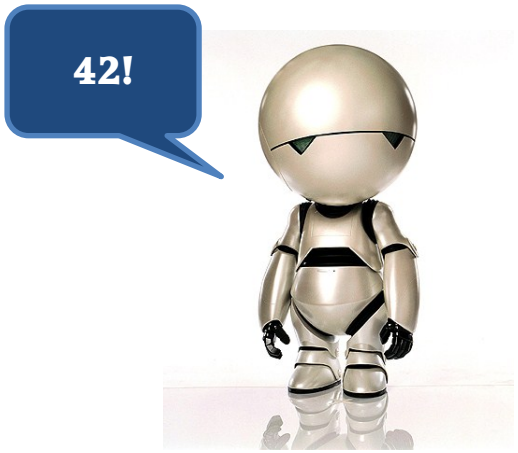
- When you have questions that EthoVision XT cannot answer;

Questions that involve EthoVision XT *state variables* (for example, in Zone, Moving/Not moving) that can translate into *sequences*

Radial maze – How many times were all 8 arms consecutively visited once?

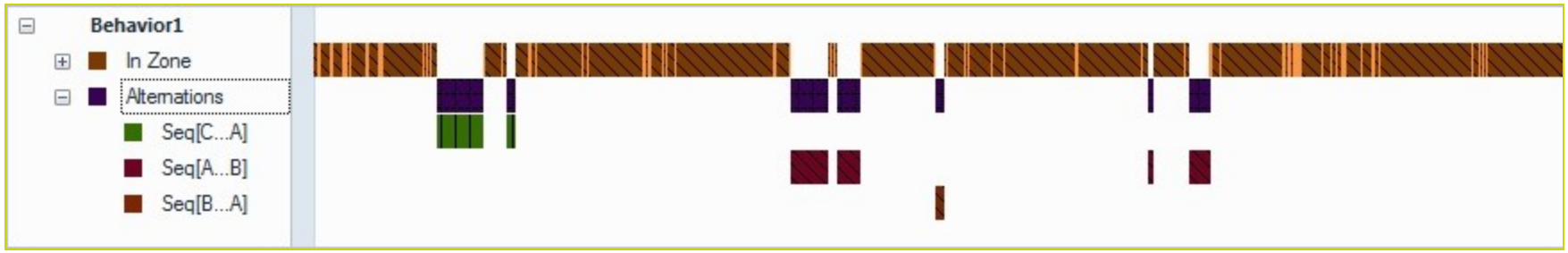
Y-maze – How many 3-arm sequences (1-2-3, 2-3-1, 3-1-2, etc.) were made?

Behavioral state – How many times was the animal not moving in a specific zone for at least 30 seconds ("which means it must have been eating")?



When do I use the Sequence Analysis Toolkit?

- When you want to export behavioral states from EthoVision XT to The Observer XT



EthoVision XT vs. the Sequence Analysis Toolkit

EthoVision XT

- Per zone, behavioral state –
  Frequency, Total / Mean duration, Latency to first, Latency to last
- Transition – From Zone A to Zone B

Sequence Analysis Toolkit

- Per zone, behavioral state –
  Frequency, Minimum / Maximum / Average Latency,
  Minimum / Maximum / Average Duration
- Sequence of zones (e.g., arms, quadrants),
  behavioral states (moving/not moving)

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What is the Sequence Analysis Toolkit?

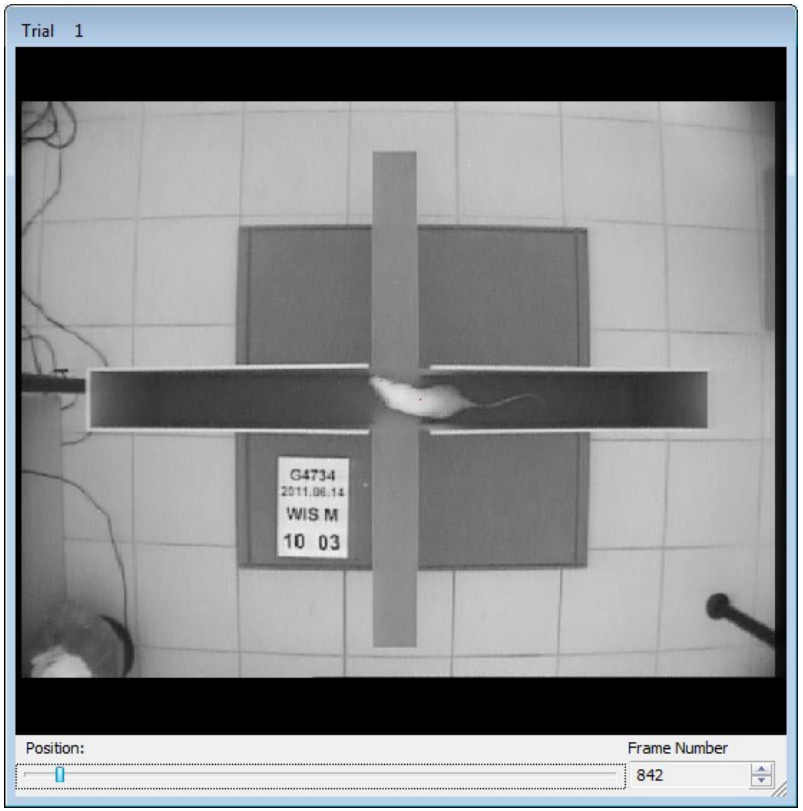
Sequence Analysis Toolkit

- Is a macro in Excel (version 2007 and later)
- Which reads and processes EthoVision XT export files
- To analyze sequences of Zones and Behavioral states

- It is not an 'official' Noldus product
- It ís supported by Noldus' behavioral research consultants

Procedure – EthoVision XT

1. Acquire trials



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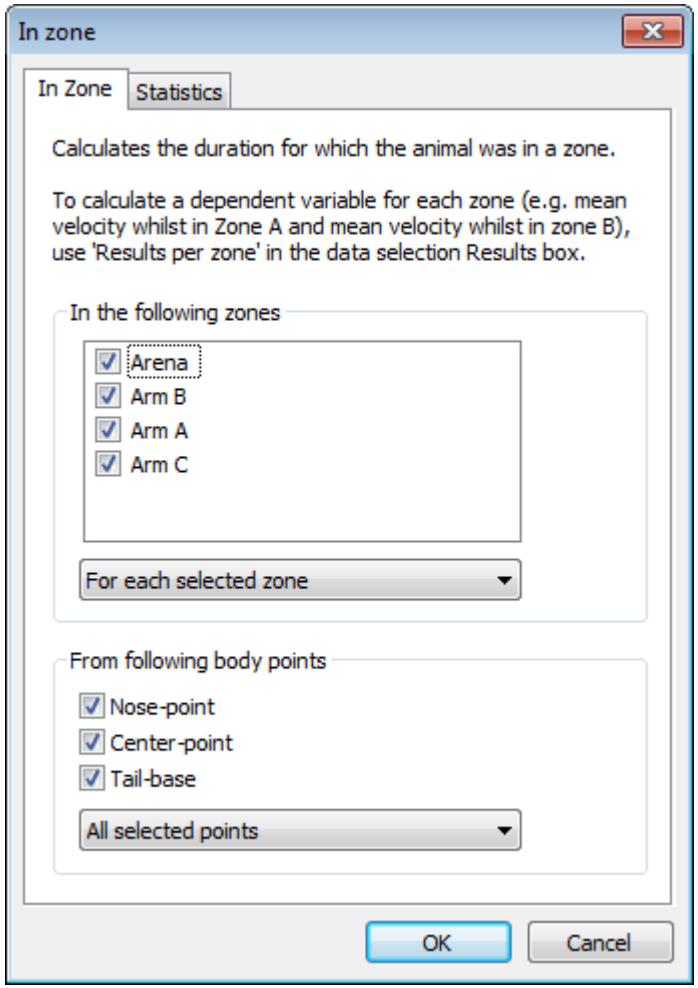
Procedure – EthoVision XT

1. Acquire trials

				User-defined	User-defined	User-defined	System	User-defined
Label				Subject number	Exposure	Treatment	Acquisition status	Day
Description							The current status of acquisition per arena	
Type				Text	Text	Text		Text
Format								
Predefined Values					Acquisition; +	None; Vehicle;	Unknown; Postpo	Day 1; Day 2; D
Scope				Subject	Subject	Subject	Arena	Subject
Trial		Arena	Subject	No.				
Trial	1	Arena 1	Subject 1	1	333 1	+ 48 hours	None	Acquired	Day 6
Trial	2	Arena 1	Subject 1	2	333 2	+ 48 hours	Vehicle	Acquired	Day 6
Trial	3	Arena 1	Subject 1	3	333 3	+ 48 hours	Scopolamine	Acquired	Day 6
Trial	4	Arena 1	Subject 1	4	333 4	+ 48 hours	None	Acquired	Day 6
Trial	5	Arena 1	Subject 1	5	333 5	+ 48 hours	Vehicle	Acquired	Day 6
Trial	6	Arena 1	Subject 1	6	333 6	+ 48 hours	Scopolamine	Acquired	Day 6
Trial	7	Arena 1	Subject 1	7	333 7	+ 48 hours	None	Acquired	Day 6
Trial	8	Arena 1	Subject 1	8	333 8	+ 48 hours	Vehicle	Acquired	Day 6
Trial	9	Arena 1	Subject 1	9	333 9	+ 48 hours	Scopolamine	Acquired	Day 6
Trial	10	Arena 1	Subject 1	10	333 10	+ 48 hours	None	Acquired	Day 6
Trial	11	Arena 1	Subject 1	11	333 11	+ 48 hours	Vehicle	Acquired	Day 6
Trial	12	Arena 1	Subject 1	12	333 12	+ 48 hours	Scopolamine	Acquired	Day 6
Trial	13	Arena 1	Subject 1	13	333 13	+ 48 hours	None	Acquired	Day 6
Trial	14	Arena 1	Subject 1	14	333 14	+ 48 hours	Vehicle	Acquired	Day 6
Trial	15	Arena 1	Subject 1	15	333 15	+ 48 hours	Scopolamine	Acquired	Day 6

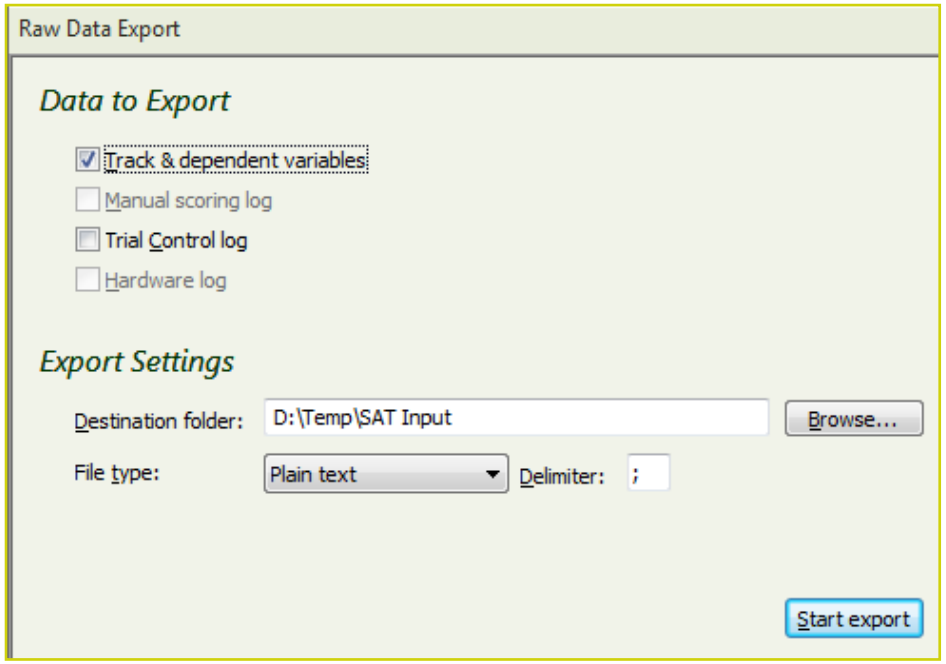
Procedure – EthoVision XT

1. Acquire trials
2. Select Zones in an Analysis Profile



Procedure – EthoVision XT

1. Acquire trials
2. Select Zones in an Analysis Profile
3. Export the raw data to Excel or Plain text



How do I work with the Sequence Analysis Toolkit?

Basic procedure

Sequence Analysis Toolkit

4. Prepare files to be processed with the Sequence Analysis Toolkit
5. Select files and parameters in the Analysis Profile and set criteria to find specific sequences
6. Carry out the sequence analysis
7. Look at the analysis results

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How does the Sequence Analysis Toolkit work?

Sub-sequence *Producer* → *Examiner*

Producer

1. Define a *FromZone* and a *ToZone*
   - Track boundary (first / last zone)
   - A user-specified zone
   - Each zone that occurs in the track
   - A specific sequence or a list of specific zones or sequences
   - FromZone = ToZone
2. Define how to treat multiple encounters of FromZone and ToZone

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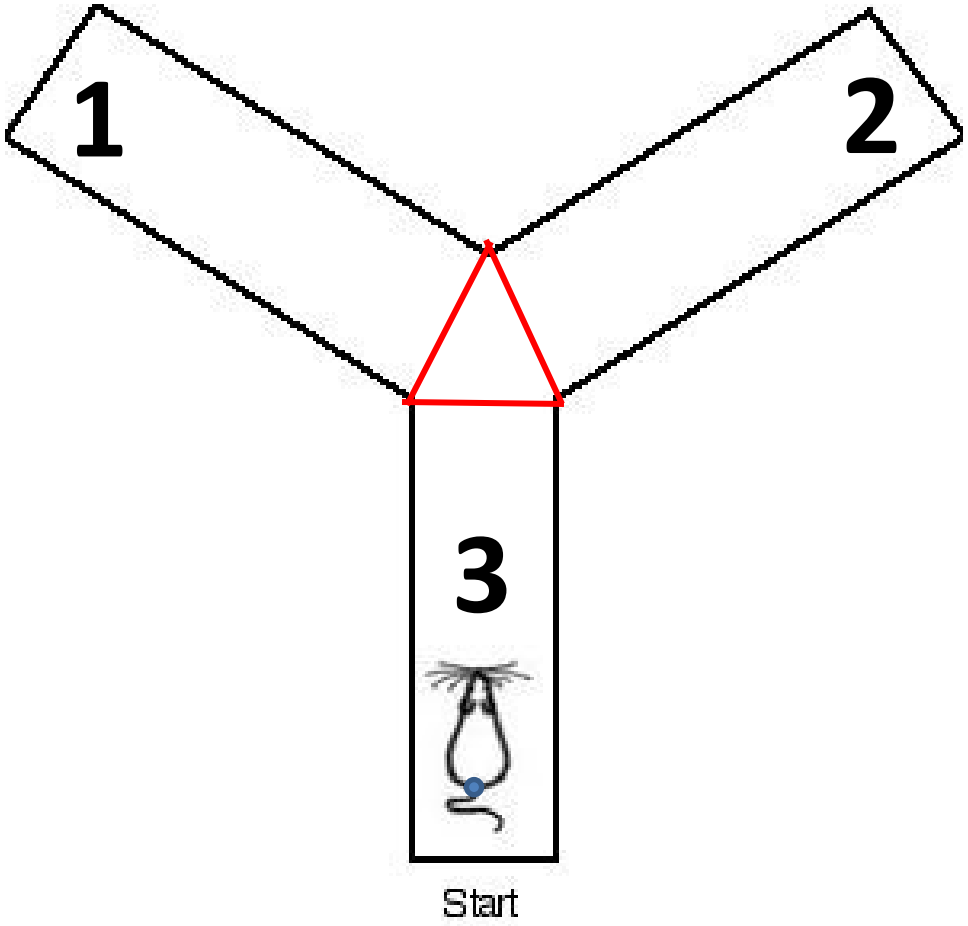
How does the Sequence Analysis Toolkit work?

Example – Y-maze



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Example – Y-maze

Complete sequence

[**3-3-3-2-1-3-1-2-2-3-1-2-3-1-2-1-3-**]

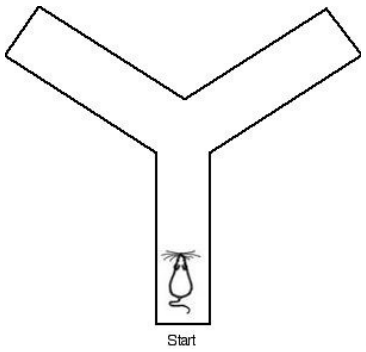
Let FromZone = **1** and ToZone = **2**

[**3-3-3-2-1-3-1-2**-2-3-1-2-3-1-2-1-3-] ?

[**3-3-3-2-1-3-1-2**-2-3-**1-2**-3-**1-2**-1-3-] ?

[**3-3-3-2-1-3-1-2-2-3-1-2-3-1-2**-1-3-] ?

How to treat multiple encounters of FromZone and ToZone?



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Example – Y-maze

Multiple encounters of FromZone and ToZone

[**3-3-3-2-1-3-1-2-2-3-1-2-3-1-2-1-3-**]

Let FromZone = **1** and ToZone = **2**

- **Longest** – First encountered FromZone to last encountered ToZone

  [**3-3-3-2-1-3-1-2-2-3-1-2-3-1-2-1-3-**]

- **Shortest** – Last encountered FromZone to first encountered ToZone

  [**3-3-3-2-1-3-1-2-2-3-1-2-3-1-2-1-3-**]

  [**3-3-3-2-1-3-1-2-2-3-1-2-3-1-2-1-3-**] … etc.

- **All Sequences** – Each FromZone to each ToZone

  [**3-3-3-2-1-3-1-2-2-3-1-2-3-1-2-1-3-**]

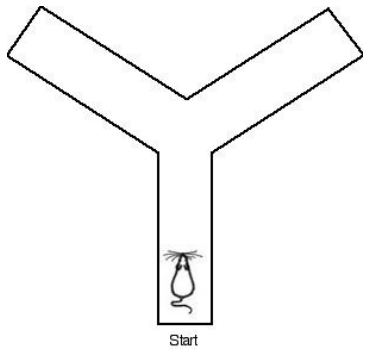
  [**3-3-3-2-1-3-1-2-2-3-1-2-3-1-2-1-3-**]

  [**3-3-3-2-1-3-1-2-2-3-1-2-3-1-2-1-3-**]

  [**3-3-3-2-1-3-1-2-2-3-1-2-3-1-2-1-3-**]

  [**3-3-3-2-1-3-1-2-2-3-1-2-3-1-2-1-3-**]

  [**3-3-3-2-1-3-1-2-2-3-1-2-3-1-2-1-3-**] … etc.



How does the Sequence Analysis Toolkit work?

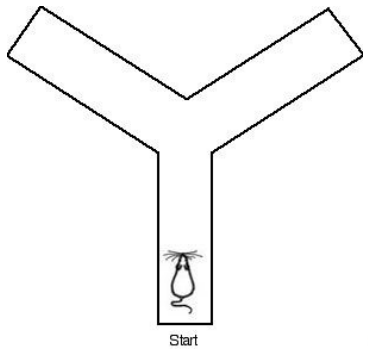
Sub-sequence *Producer* → *Examiner*

Examiner

inspects each sub-sequence given by the *Producer*

- Sub-sequence must exactly match with a specific zone, sequence or list of zones/sequences
- Zones may/may not be revisited in the sub-sequence
- Sub-sequence must **in**clude a specific zone, sequence or list of zones/sequences
- Sub-sequence must **ex**clude a specific zone, sequence or list of zones/sequences
- Subsequence must contain more than, less than or exactly a specific number of zones

and/or



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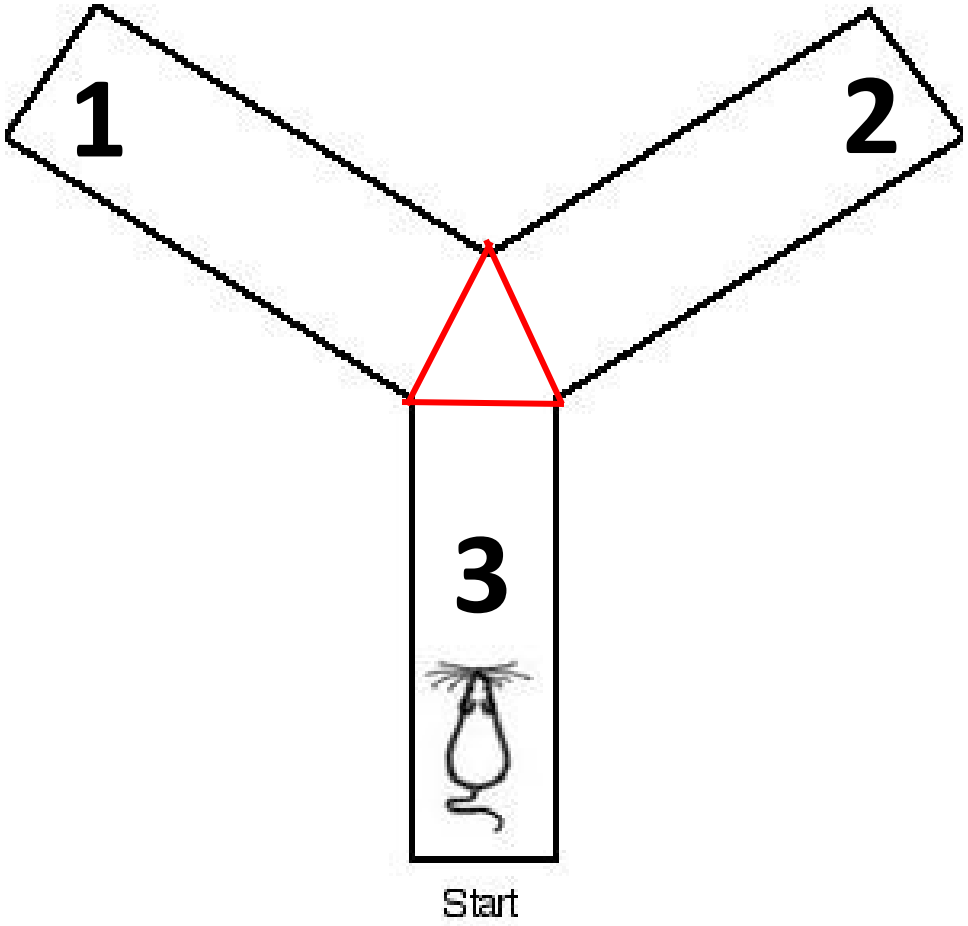
How does the Sequence Analysis Toolkit work?

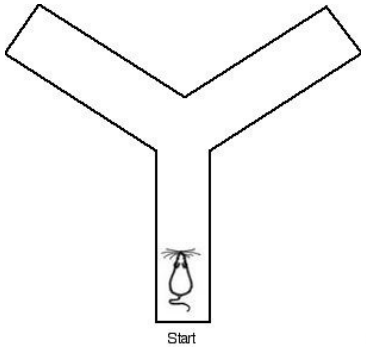
DEMO – Y-maze



Examples – Y-maze

This finds the complete sequence of arm-visits

Range Criteria:			Define your named lists here:	
			List 1	List 2
From zone:	Track boundary		any one of	any one of
To zone:	Track boundary			
Focus on:	longest only			
Additional Criteria:				
Exact sequence:				
Pass through ?	visit each zone any nr of times			
Including:				
Excluding:				
Sequence must be:		zones		



[AnalysisProfile.xlsx]
[Results.xlsx]

Examples – Y-maze

This finds the alternations in the Y-maze

Range Criteria:

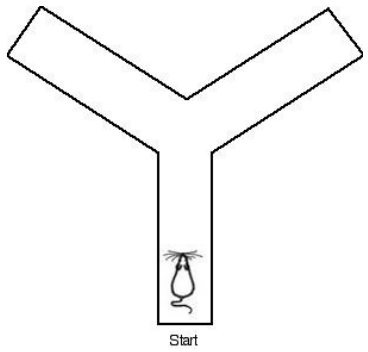
From zone:	Each zone
To zone:	Each zone
Focus on:	shortest only

Additional Criteria:

Exact sequence:	List 1
Pass through ?	visit each zone any nr of times
Including:	
Excluding:	
Sequence must be:	zones

Define your named lists here:

List 1	List 2
any one of	any one of
1-2-3	
1-3-2	
2-1-3	
2-3-1	
3-2-1	
3-1-2	



[AnalysisProfile.xlsx]
[Results.xlsx]

Examples – Y-maze

This finds all the right turns in the Y-maze

Range Criteria:

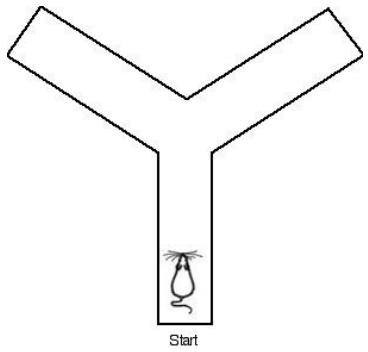
From zone:	Each zone
To zone:	Each zone
Focus on:	shortest only

Additional Criteria:

Exact sequence:	List 1
Pass through ?	visit each zone any nr of times
Including:	
Excluding:	
Sequence must be:	zones

Define your named lists here:

List 1	List 2
any one of	any one of
2-1	
1-3	
3-2	



[AnalysisProfile.xlsx]
[Results.xlsx]

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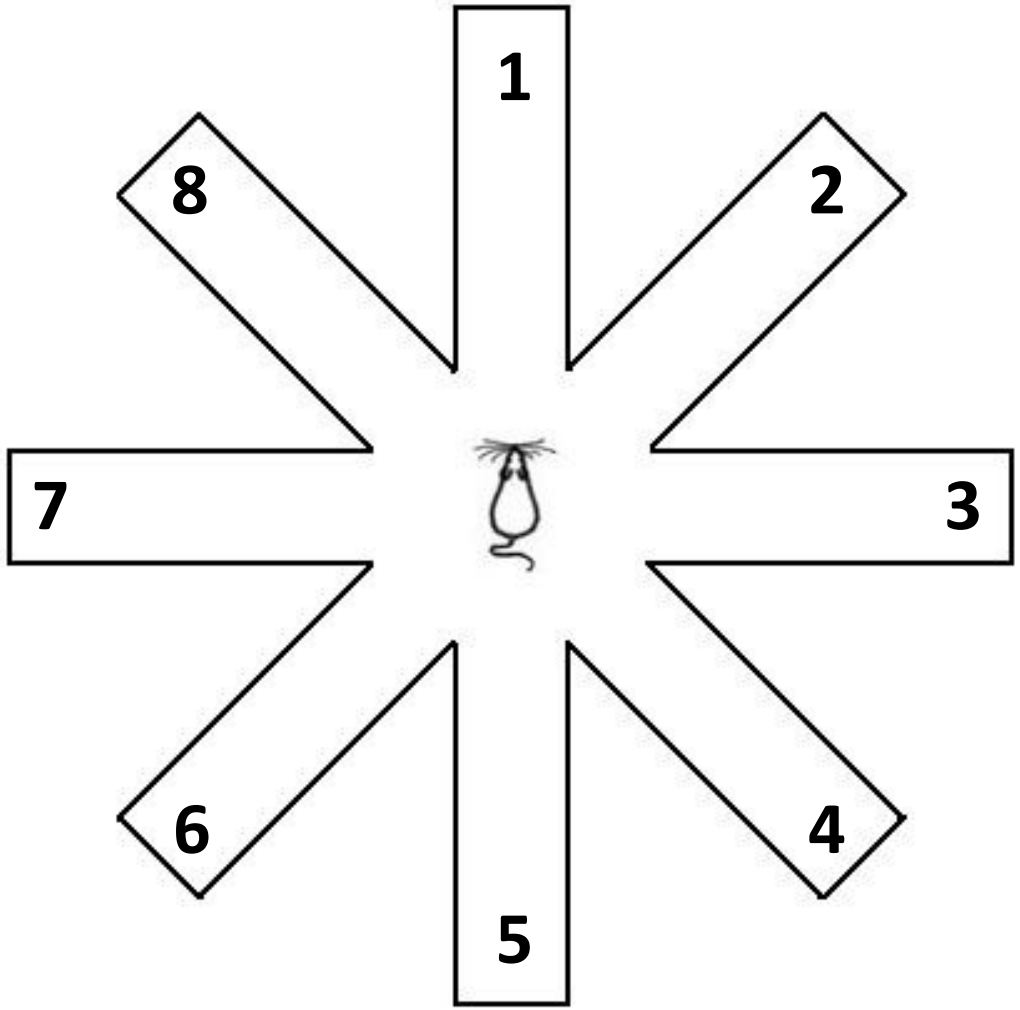
Demo – Sequence Analysis Toolkit

DEMO – Radial maze



Excited, are you?!

Examples – Radial maze

Radial maze

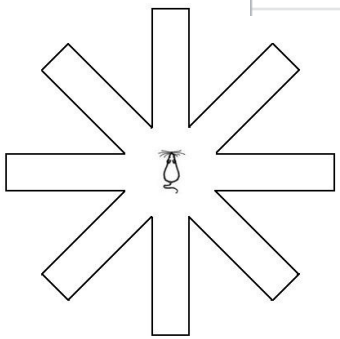
- Short-term memory – How do animals search the for baited arms? Do they revisit arms?
- Treatments – How do different treatments (drugs, housing, stress) affect performance in the maze and therefore memory?

Examples – Radial Maze

This finds the sequence in which all 8 arms have been exactly once without re-visits

Range Criteria:				**Define your named lists here:**	
				List 1	List 2
From zone:	Each zone			any one of	any one of
To zone:	Each zone				
Focus on:	longest only				
Additional Criteria:					
Exact sequence:					
Pass through ?	visit each zone only once				
Including:					
Excluding:					
Sequence must be:	exactly	8	zones		

[AnalysisProfile.xlsx](#)
[Results.xlsx](#)



Examples – Radial Maze

How many times did I visit each arm exactly once in a specific order?

Range Criteria:

From zone:	Track boundary
To zone:	Track boundary
Focus on:	longest only

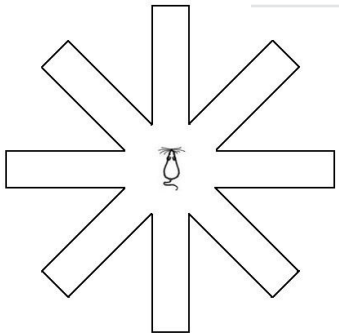
Additional Criteria:

Exact sequence:	List 1	
Pass through ?	visit each zone any nr of times	
Including:		
Excluding:		
Sequence must be:		zones

Define your named lists here:

List 1	List 2
any one of	any one of
1-2-3-4	

[AnalysisProfile.xlsx](#)
[Results.xlsx](#)



Examples – Radial Maze

How many times did I re-visit an arm?

Range Criteria:

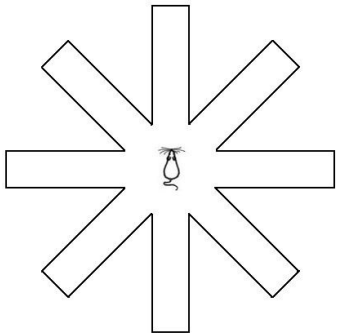
From zone:	From zone equals To zone
To zone:	From zone equals To zone
Focus on:	shortest only

Additional Criteria:

Exact sequence:			
Pass through ?	visit each zone any nr of times		
Including:			
Excluding:			
Sequence must be:	exactly	2	zones

Define your named lists here:

List 1	List 2
any one of	any one of



[AnalysisProfile.xlsx]
[Results.xlsx]

Examples – Radial Maze

How many times did I re-visit a specific arm?

Range Criteria:

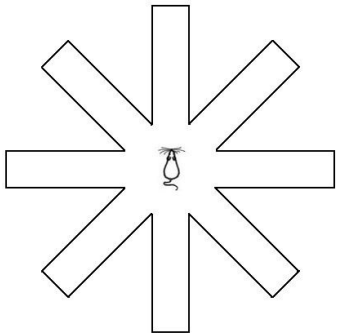
From zone:	From zone equals To zone
To zone:	From zone equals To zone
Focus on:	shortest only

Additional Criteria:

Exact sequence:	List 1
Pass through ?	visit each zone any nr of times
Including:	
Excluding:	
Sequence must be:	zones

Define your named lists here:

List 1	List 2
any one of	any one of
1-1	
3-3	
5-5	
7-7	



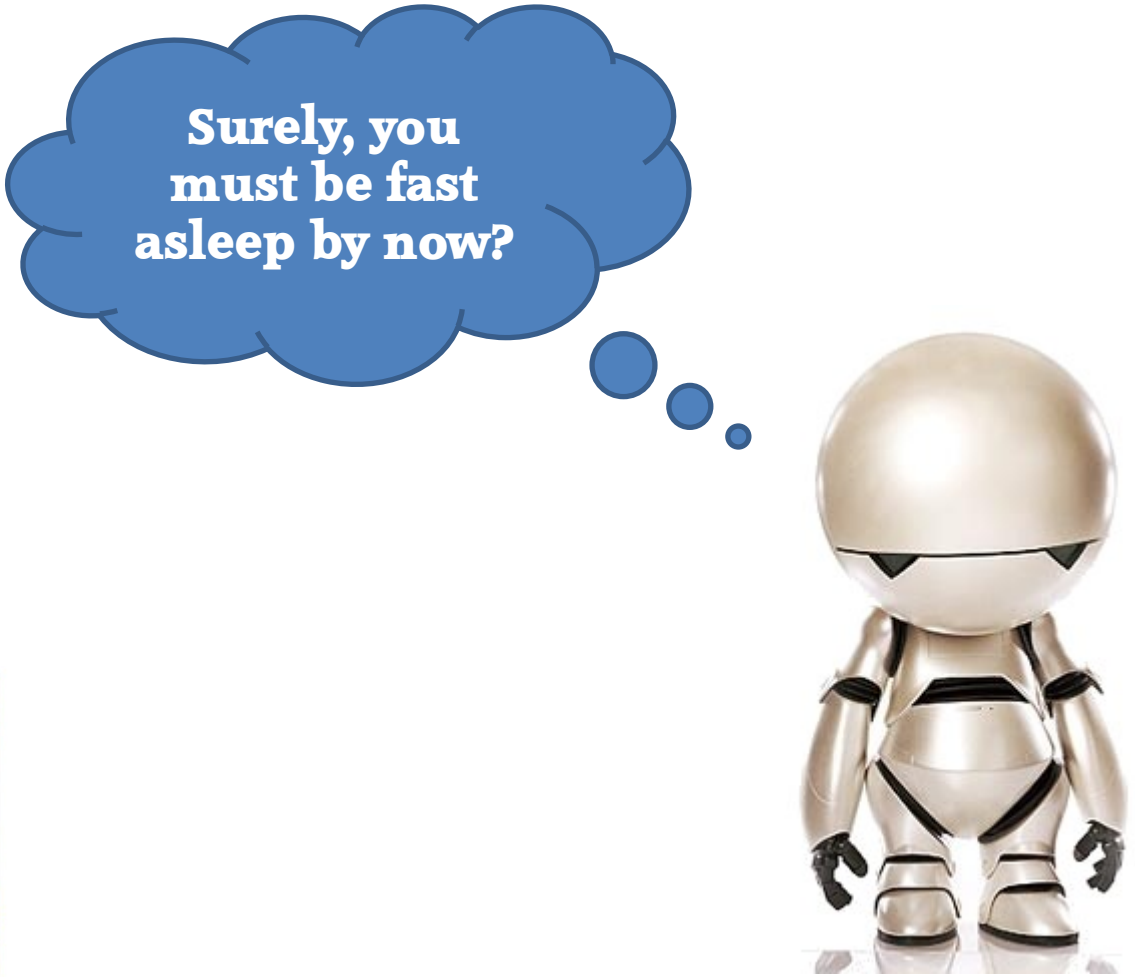
Examples – Analyzing behavioral states

Dreamy 'state'

- 'Dreamy' defined as the animal not moving for at least 10 seconds as defined in EthoVision XT

Procedure – Sequence Analysis Toolkit

DEMO – Behavioral state 'Dreamy'



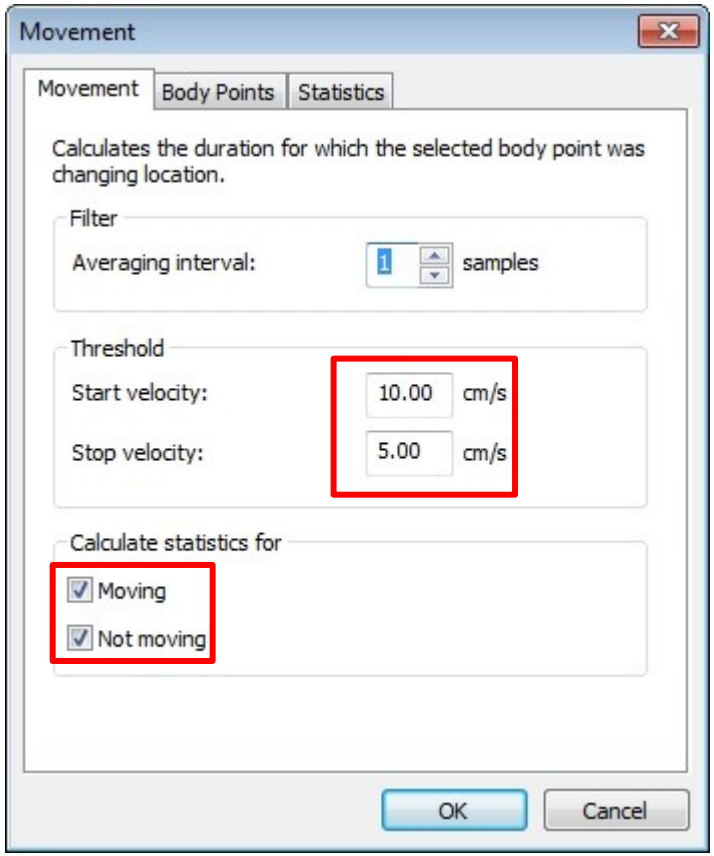
Example – Exporting Zones / Behavioral states to The Observer XT

EthoVision XT

1. Acquire trials

2. Select Movement states in an Analysis Profile
   - Set Threshold
   - Select all states

3. Export the raw data to Excel or Plain text



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Examples – Analyzing behavioral states

How many times was the animal 'dreamy' for at least 10 seconds?

Range Criteria:

From zone:	D
To zone:	A
Focus on:	shortest only

Additional Criteria:

Exact sequence:		
Pass through ?	visit each zone any nr of times	
Including:		
Excluding:		
Sequence must be:		zones

Define your named lists here:

List 1	List 2
any one of	any one of

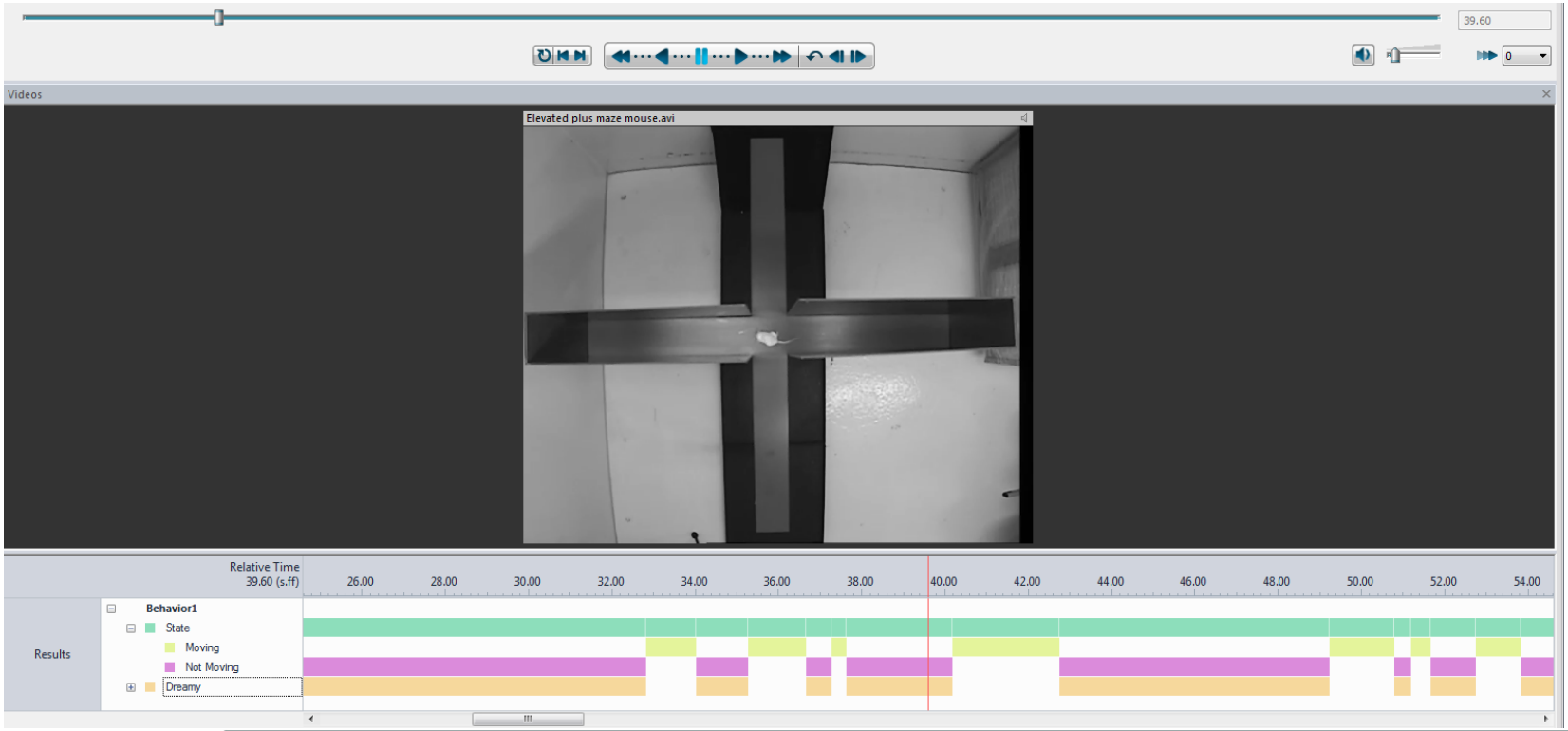
[AnalysisProfile.xlsx]
[Results.xlsx]

Exporting Zones / Behavioral states to The Observer XT

Sequence Analysis Toolkit

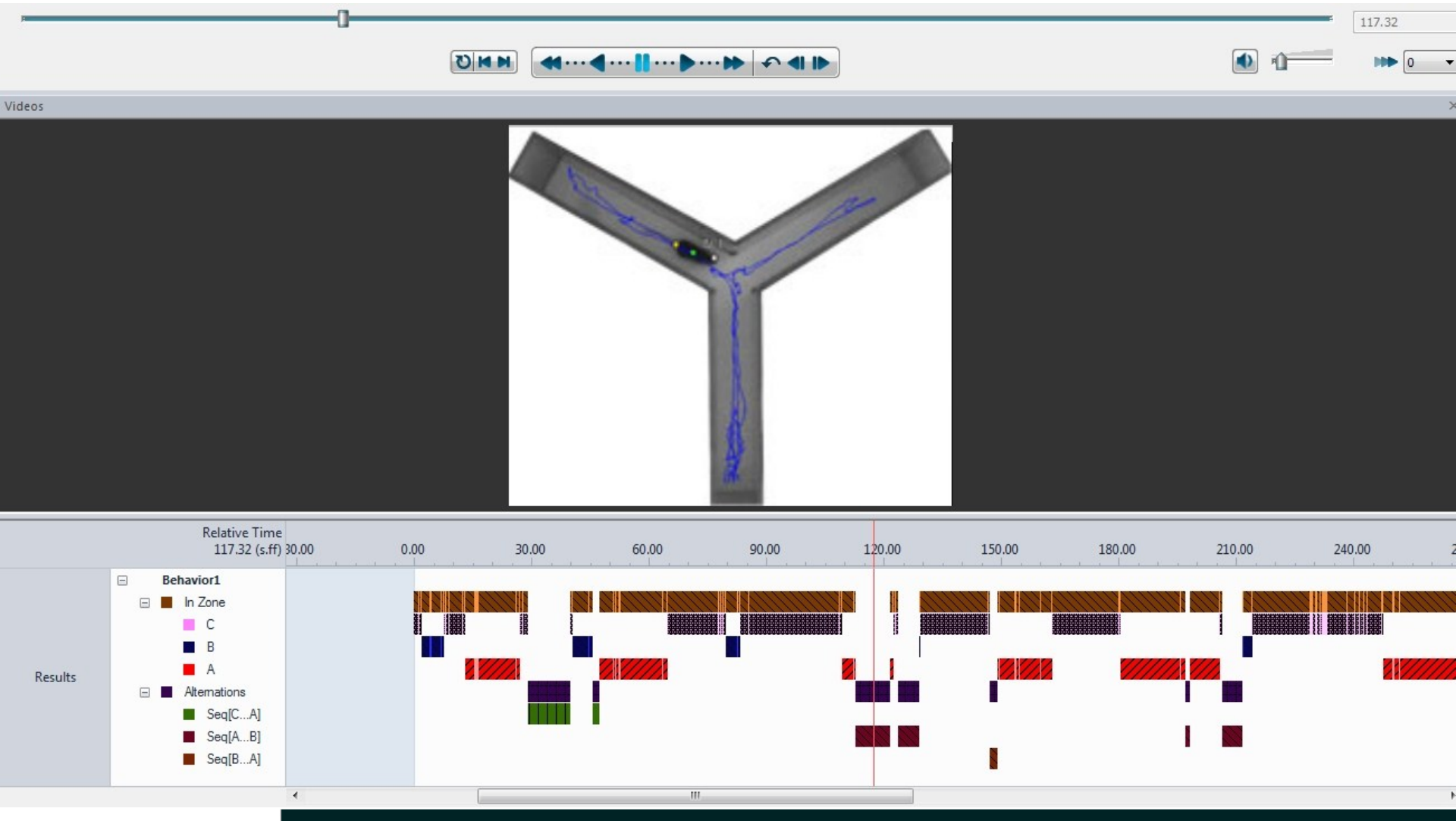
1. Click the *Export Event Logs* button

2. Import the export file (*.txt) into The Observer XT



Exporting Zones / Behavioral states to The Observer XT

Y-maze - Alternations



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Take-home message

Sequence Analysis Toolkit is a useful tool to

- Analyze the sequence of zone visits / behavioral states
- Find specific sequences of zone visits / behavioral states
- Get additional information about EthoVision's behavioral states
- Allows export of sequences of zones / behavioral states to The Observer XT

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Thank you for your attention

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Questions?