

# Zebrafish and learning paradigms

How EthoVision XT can benefit your research

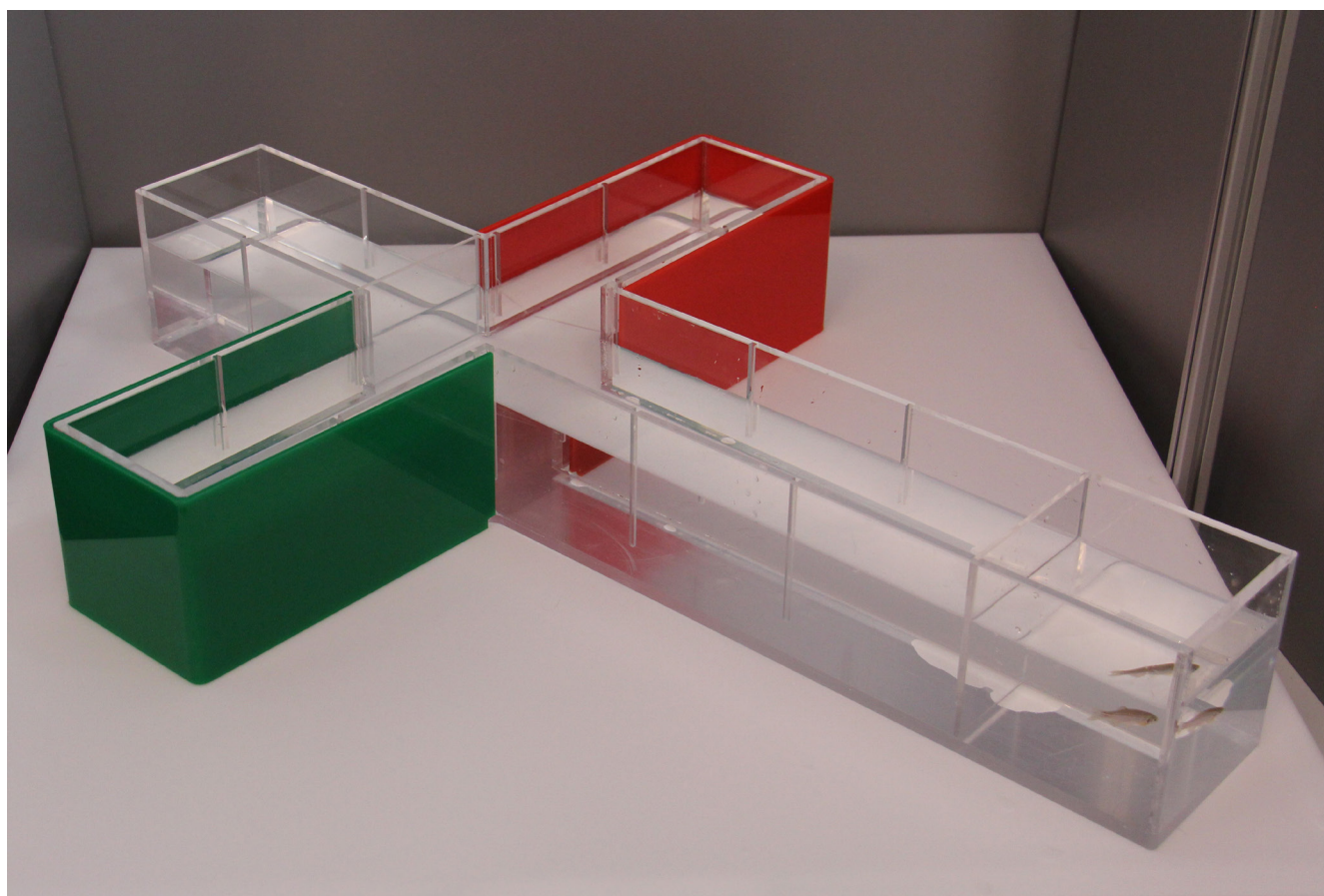


A white paper by Noldus Information Technology

# ZEBRAFISH AND LEARNING PARADIGMS

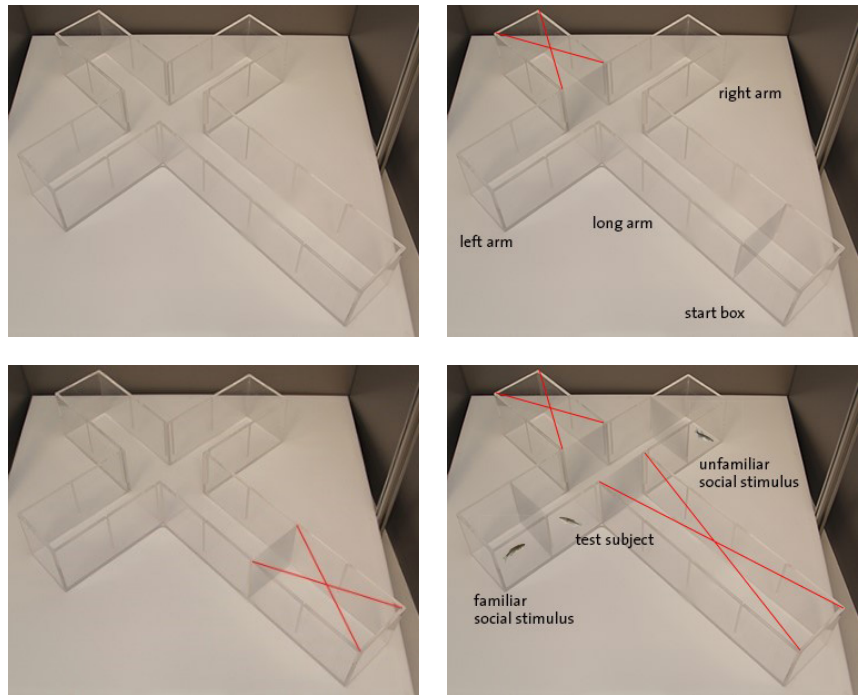
Zebrafish have become an important animal model for studying neurodegenerative diseases. In validation of these models, both pathologically and behaviorally, many standardized paradigms have been translated from rodent models to zebrafish. For learning and memory paradigms, this includes tests such as the T-maze and plus maze.

*Noldus offers the cross maze, a multifunctional maze that can be used for T-maze testing, plus maze testing, place conditioning, contextual learning, social learning, and more. It is designed for video tracking experiments with EthoVision XT.*



# CROSS MAZE

The aquatic cross maze is a multifunctional maze for zebrafish learning and memory testing, but it also allows for other tests such as social preference testing. This is possible because you can adjust the maze any way you want with the use of several inserts.



## TECHNICAL SPECIFICATIONS

### Size

70 x 50 x 10 cm

Perhaps the most straightforward usage of the cross maze is the T-maze, with one long arm and two short ones. Positive and aversive stimuli or environments can be used to teach the zebrafish to swim towards a specific arm, for example, going against natural preference, or to test reversal learning. The arms can be fitted with colored sleeves for color discrimination learning.

Another way to use this maze is to create a plus maze, which is often used for slightly more complex associative learning tasks or anxiety research.

It is even adaptable for social behavior studies, using inserts to physically, but not visually separate familiar and unfamiliar other fish to test social memory or preferences.

# RESEARCH EXAMPLES

Like rodents, zebrafish are quite good at learning tasks like the T-maze, especially when using either food as a motivator or using aversive stimuli. [2,3,8,9,11]. Also, zebrafish see color, making color discrimination tasks another option for testing.

*EthoVision XT offers a Trial & Hardware control module, which allows you to use the software to automatically send out a signal to a shocker or food-dispenser as soon as the animal enters a specific area.*

## APPLICATIONS

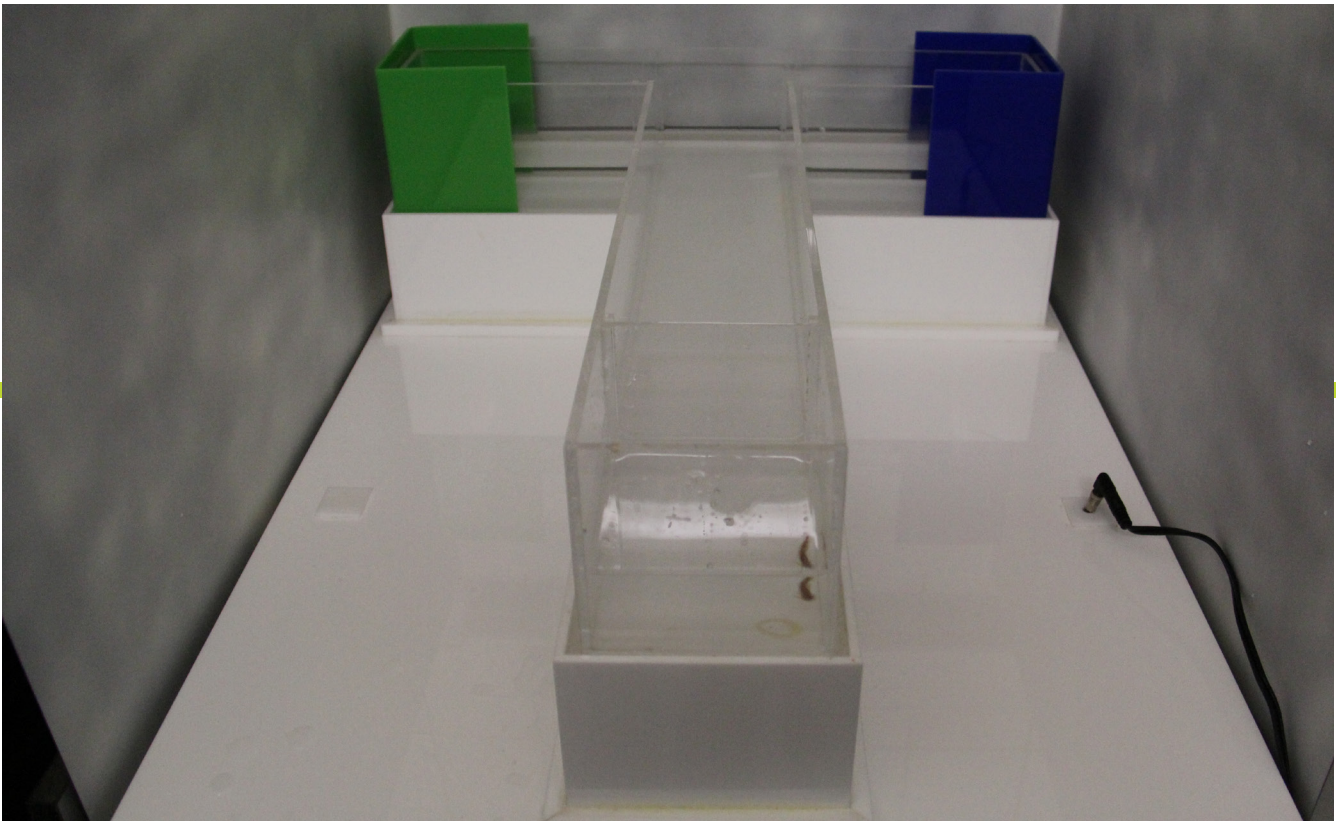
The T-maze and similar setups are used to test behavioral plasticity of certain strains, as well as the effects of genetic manipulation, pharmacological treatment, or (environmental) toxins. For example, the work of Colwill *et al.* [2] describes the behavioral plasticity of zebrafish using learning, reversal learning, and extinction of learned behavior.

Ninkovic *et al.* [8] studied the effect of AChE inhibitors as a promising therapy for addiction, and used the T-maze to test for side-effects influencing learning and memory (they found no negative influence). Peitsaro *et al.* [9] tested the effect of a histamine inhibitor ( $\alpha$ FMH) and found an impairment of long-term (but not short-term) memory. Saili *et al.* [11] investigated the long-term effects of an environmental pollutant called bisphenol A (BPA) and also found impairment of long-term memory using T-maze testing.

More sophisticated setups might include the testing of episodic memory. By combining context (color), placement, and (familiar and novel) objects, you can investigate whether zebrafish can detect a difference when a familiar object is placed in a familiar context (environmental color) but in a different place. This is what Hamilton *et al.* [5] did.

## COLOR AND PATTERN DISCRIMINATION

Avdesh *et al.* [1] gave the two shorter arms of their T-maze different colors and tested color preference. This seems to differ between strains – while their strain preferred red and green, Colwill *et al.* [2] found a preference for blue and purple.



Kim et al. [6] found that using colors on the short arms of a T-maze actually aid in memory testing. After establishing a natural preference for the color red, they found that this was not enough for memory formation; fish did not automatically swim to the previously red colored arm during memory testing. However, a food reward did have this effect on learning, and interestingly, pairing the color red and a food reward did increase this learning effect. Rimstad et al. [10] show that zebrafish do not only show color discrimination, but also pattern discrimination when presented with horizontal stripes, vertical stripes, and squares of different sizes.

### **SOCIAL BEHAVIOR TESTING**

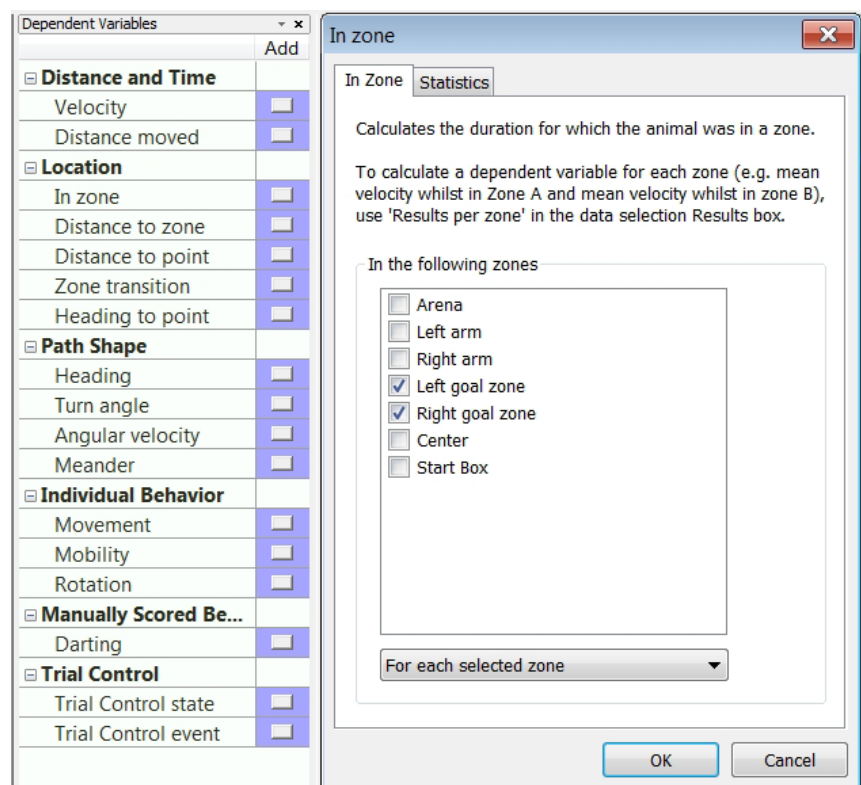
A social context also has an effect on learning, McAroe *et al.* [7] found. As a shoal (5 fish, in this case), fish learn to find a food reward in a modified T-maze (with three arms of the same length) much more quickly. Interestingly, this is not transferred to the individual, as they did not show improved learning in individual testing. Also, shoals adapted a consistent place navigation strategy, whereas individuals did not show a specific strategy to get to the food.

*No matter what the aim of your T-maze study is, EthoVision XT accurately tracks your zebrafish as it swims through the maze and gets you objective data.*

# BEHAVIORS OF INTEREST

During the trials, different behavioral endpoints were measured automatically by EthoVision XT, such as:

- Latency to reach the goal arm
- Which arm (and which color) was entered
- Which arm received first preference
- Entries into different zones
- Duration of swimming in the different zones
- Swim speed
- Distance swum
- Freezing behavior
- Mobility



# REFERENCES

1. Avdesh, A.; Martin-Iverson, M.T.; Mondal, A.; Chen, M.; Askraba, S.; Morgan, N.; Lardelli, M.; Groth, D.M.; Verdile, G.; Martins, R.N. (2012). Evaluation of color preference in zebrafish for learning and memory. *Journal of Alzheimer's Disease*, **28**, 459-469.
2. Colwill, R.M.; Raymond, M.P.; Ferreira, L.; Escudero, H. (2005). Visual discrimination learning in zebrafish (*Danio rerio*). *Behavioural Processes*, **70**, 19-31.
3. Grella, S.; Kapur, N.; Gerlai, R. (2010). A Y-maze choice task fails to detect alcohol avoidance or alcohol preference in zebrafish. *International Journal of Comparative Psychology*, **23**, 26-42.
4. Grossman, L.; Utterback, E.; Stewart, A.; Gaikwad, S.; Chung, K.M.; Suci, C.; Wong, K.; Elegante, M.; Elkhayat, S.; Tan, J.; Gilder, T.; Wu, N.; DiLeo, J.; Chachat, J.; Kalueff, A.V. (2010). Characterization of behavioral and endocrine effects of LSD on zebra-fish. *Behavioural Brain Research*, **214**, 277-284.
5. Hamilton, T.J.; Myggland, A.; Duperreault, E.; May, Z.; Gallup, J.; Powell, R.A.; Schalomon, M.; Digweed, S.M. (2016). Episodic-like memory in zebrafish. *Animal Cognition*, **19**, 1071-1079.
6. Kim, Y.H.; Lee, K.S.; Park, E.R.; Min, T.J. (2017). Adding preferred color to a conventional reward method improves the memory of zebrafish in the T-maze behavior model. *Animal Cells and Systems*, doi: 10.1080/19768354.2017.1383938.
7. McAroe, C.L.; Craig, C.M.; Holland, R.A. (2017). Shoaling promotes place over response learning but does not facilitate individual learning of that strategy in zebrafish (*Danio rerio*). *BMC Zoology*, **2**, 10.
8. Ninkovic, J.; Folchert, A.; Makhankov, Y.V.; Neuhaus, S.C.F.; Sillaber, I.; Straehle, U.; Bally-Cuif, L. (2006). Genetic identification of AChE as a positive modulator of addiction to the psychostimulant D-Amphetamine in zebrafish. *Journal of Neurobiology*, **66(5)**, 463-475.
9. Peitsaro, N.; Kaslin, J.; Anichtchik, O.V.; Panula, P. (2003). Modulation of the histaminergic system and behavior by  $\alpha$ -fluoromethylhistidine in zebrafish. *Journal of Neurochemistry*, **86**, 432-441.
10. Rimstad, L.A.; Holcombe, A.; Pope, A.; Hamilton, T.J.; Schalomon, M.P. (2017). Preferences for achromatic horizontal, vertical, and square patterns in zebrafish (*Danio rerio*). *PeerJ*, doi: 10.7717/peerj.3748.
11. Saili, K.S.; Corvi, M.M.; Weber, D.N.; Patel, A.U.; Das, S.R.; Przybyla, J.; Anderson, K.A.; Tanguay, R.L. (2012). Neurodevelopmental low-dose bisphenol A exposure leads to early life-stage hyperactivity and learning deficits in adult zebrafish. *Toxicology*, **291**, 83-92.
12. Zheng, S.; Liu, C.; Huang, Y.; Bao, M.; Huang, Y.; Wu, K. (2017). Effects of 2,2',4,4'-tetrabromodiphenyl ether on neurobehavior and memory change and bcl-2, c-fos, grin1b and lingo1b gene expression in male zebrafish (*Danio rerio*). *Toxicology and Applied Pharmacology*, **333**, 10-16.



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