OP-082 [Bee Biology]
Dietary omega-6:3 fatty acids ratio affects honey bee (Apis mellifera) spatial cognition: maze and landmark learning

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Honey bee foragers have remarkable cognitive abilities, including associative and spatial learning. Malnutrition is a main stressor affecting honey bee health and behavior. Pollen provides many important micronutrients and macronutrients, including lipids. Of these, two essential fatty acids, omega-3 and omega-6, are abundant in pollen, and play a major role in bee health and cognition. However, pollens differ in the relative proportions of these two fatty acids, and bees must maintain a balanced omega-6:3 ratio. High omega-6:3 ratio impairs olfactory and tactile associative learning. Here we tested whether it also affects spatial cognition. Colonies were kept in netted enclosures and fed artificial diets enriched with different concentrations of corn (high in omega-6) and flaxseed (high in omega-3) oils, resulting in a balanced (omega-6:3 ratio = 1) or an unbalanced (omega-6:3 ratio = 5) diet. Newly-emerged bees were marked and tested when they were 3 weeks old. In experiment 1, bees were trained to enter the center of an 8-arm maze (25 cm diameter). Bees had to walk to the end of each arm, where they found sucrose solution reward. A trial was completed upon visiting all 8 arms, and the bee flew back to the colony. Each bee completed 8 such trials. In experiment 2, bees flew into a netted circular arena (90 cm diameter), with 8 feeders aligned on vertical stilts along the circumference. Only one of these feeders contained sucrose solution. Colored cylinder landmarks in a triangular configuration at the center of the arena guided towards the rewarding feeder. A marked bee flew into the arena and visited the feeders until finding the rewarding one, marking the end of a trial, returned to the hive and back to the arena for a total of 8 trials. In both experiments, bees were filmed from above, and detected and followed with custom-built software. In both experiments, bees fed a balanced diet solved the spatial task more quickly, with fewer errors. The implication of these results is that unbalanced forage may impair bee spatial cognition, affecting their foraging and homing abilities, and consequently colony strength.

Dietary omega-6:3 fatty acids ratio affects honey bee (*Apis mellifera*) spatial cognition: maze and landmark learning

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האוניברסיטה העברית בירושלים
THE HEBREW UNIVERSITY OF JERUSALEM

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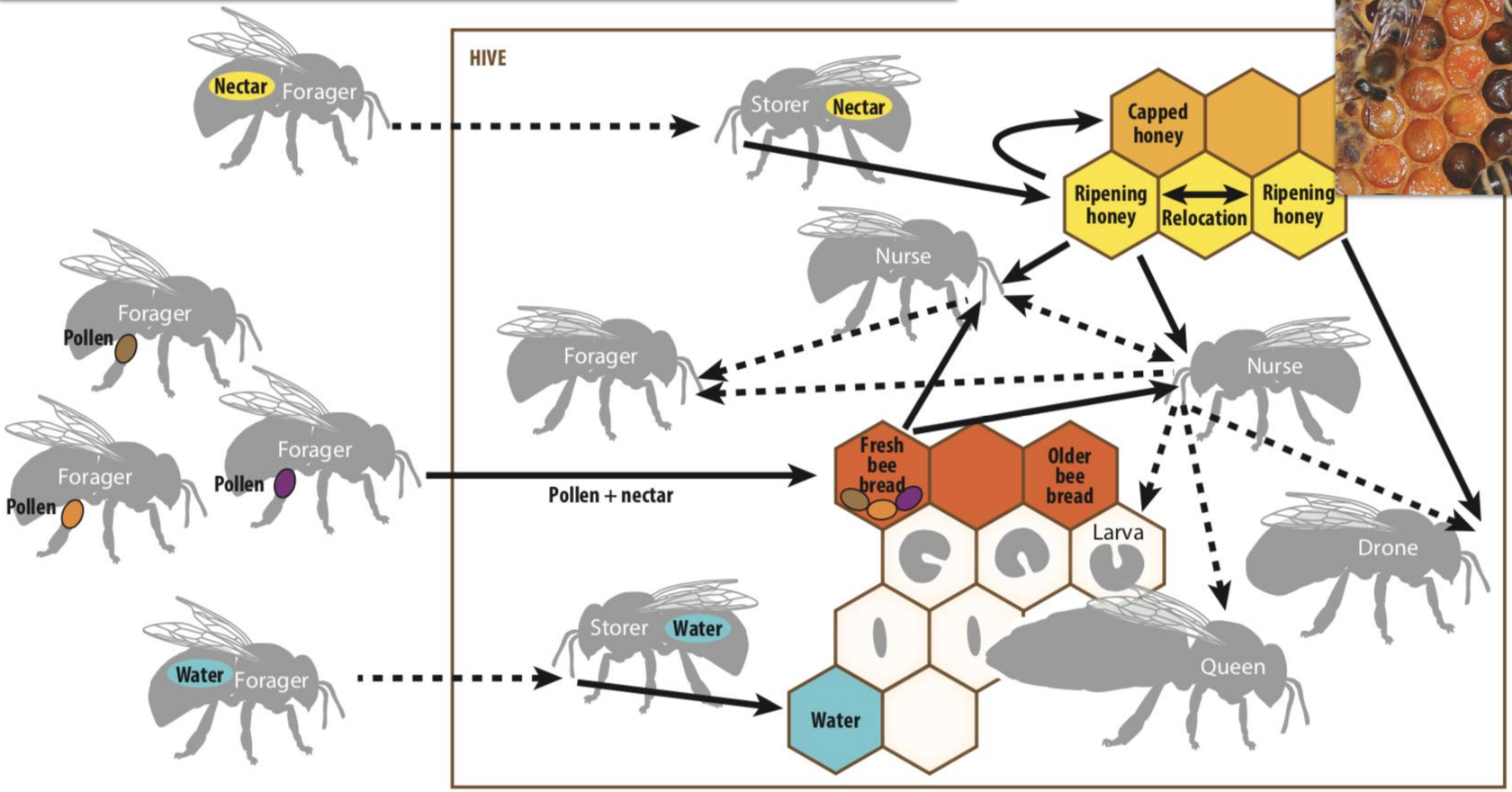
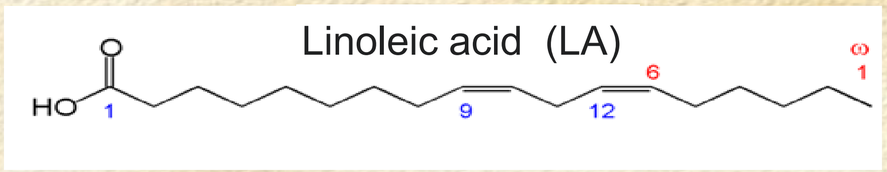
Nutritional Physiology and Ecology of Honey Bees

Geraldine A. Wright,[1,*] Susan W. Nicolson,[2] and Sharoni Shafir[3]

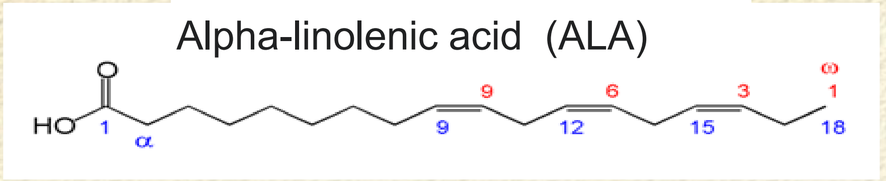


Essential fatty acids

Omega-6



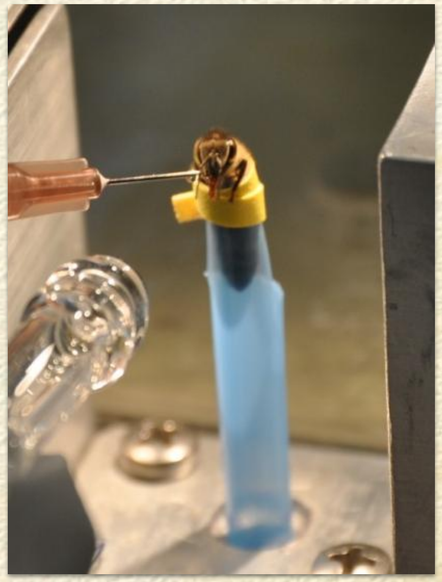
Omega-3

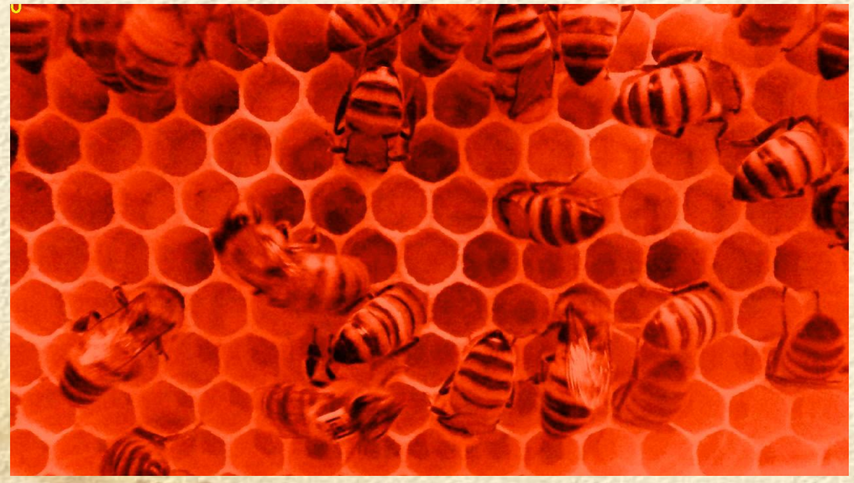


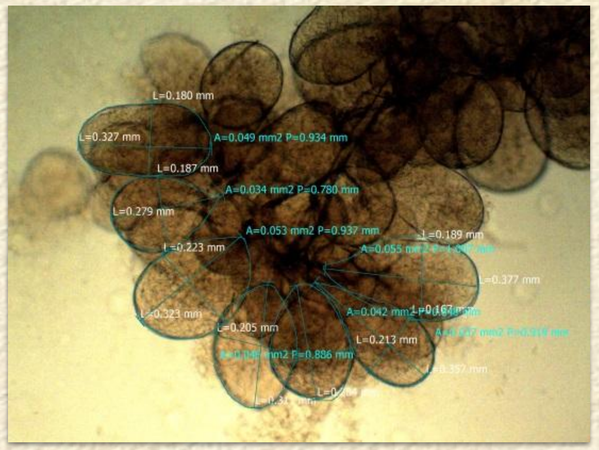
- Colonies collect balanced omega-6:3 ratio pollen (Arien et al. 2015)
- Rosaceae (ratio = 2) and *Eucalyptus* (ratio = 5-10) are unbalanced (Arien et al. 2015)
- Foragers bias pollen collection and recruitment towards balancing omega-6:3 ratio (Zarchin et al. 2017; Hendriksma and Shafir, 2016)

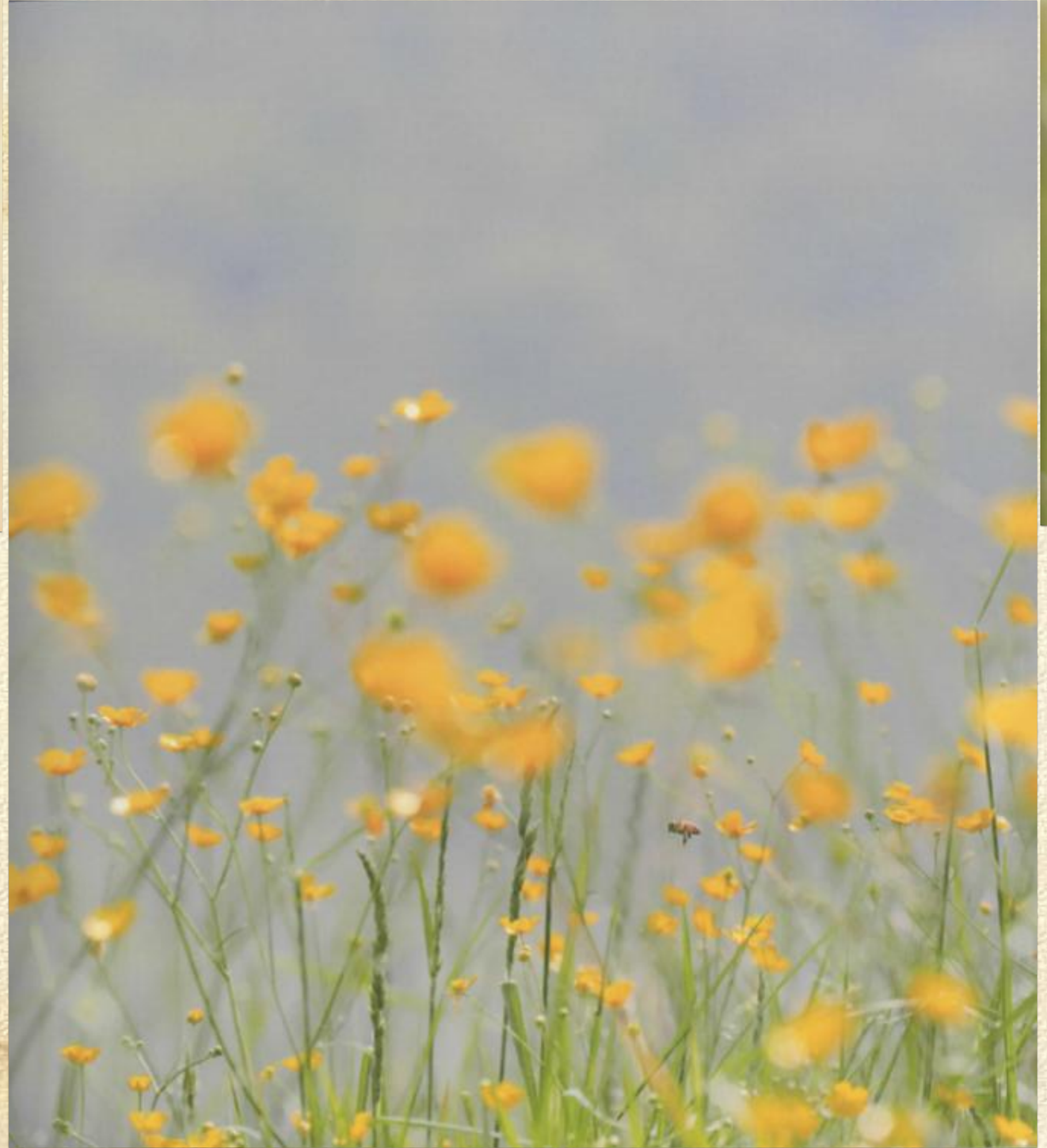
High omega-6:3 ratio in diet impairs:

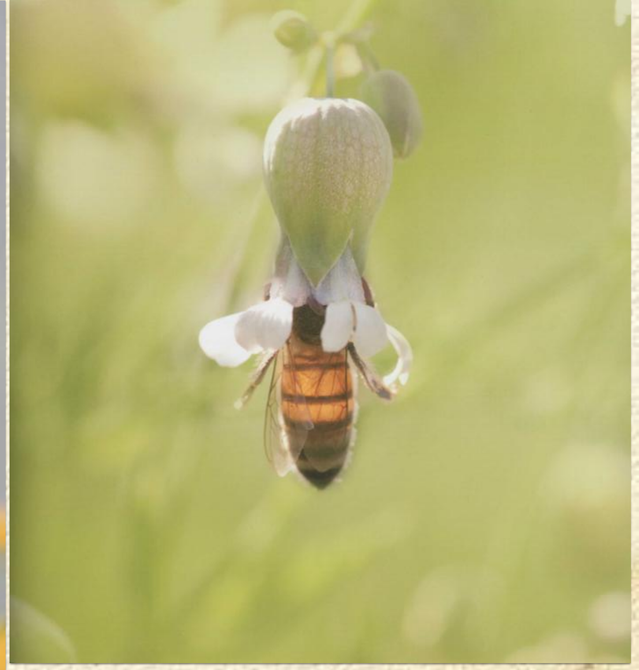
- Learning of foragers (Arien et al. 2015; 2018), nurses (Bennett et al., 2022), and queens (Kanot et al. Unpublished)
- Physiology: hypopharyngeal gland development, survival and reproduction (Arien et al. 2015; 2020)
- Nursing dynamics (Minahan et al. 2024)





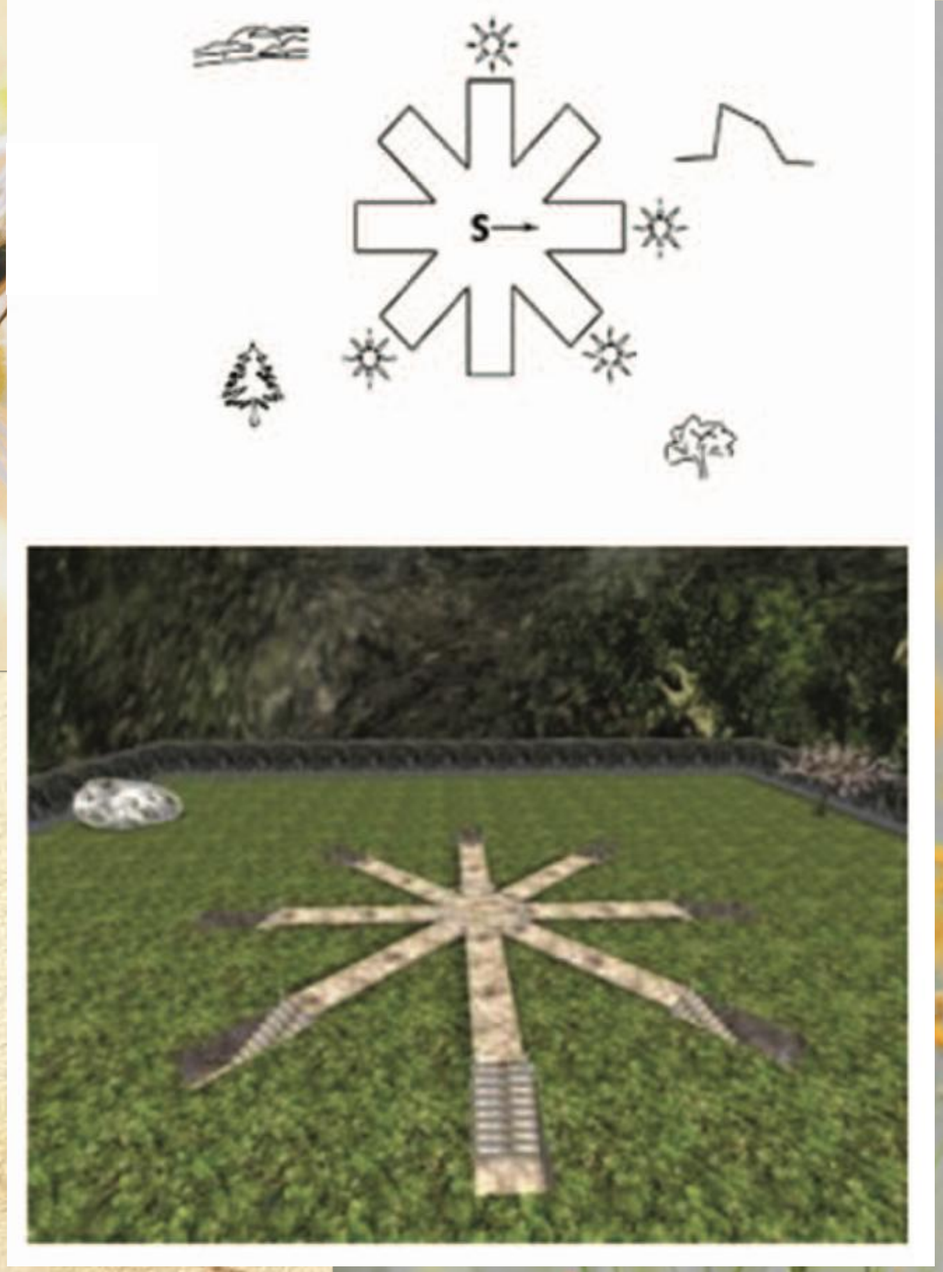


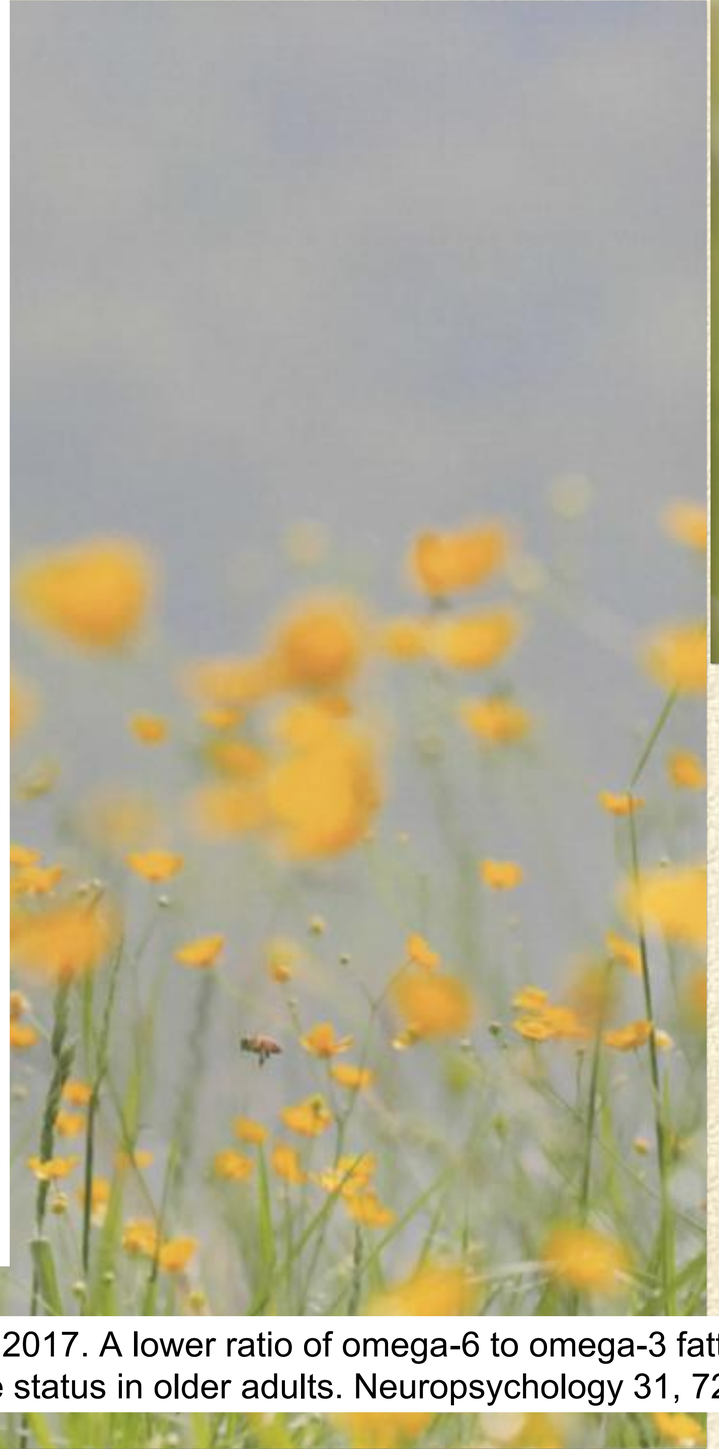


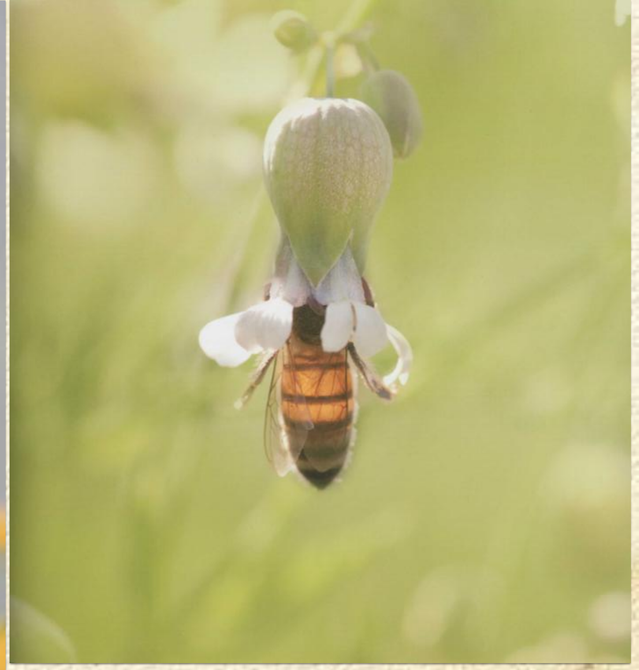


Photos:
Marina Gallandra, La societa delle api.

Helga Heilmann, The buzz about bees,
Jurgen Tautz.

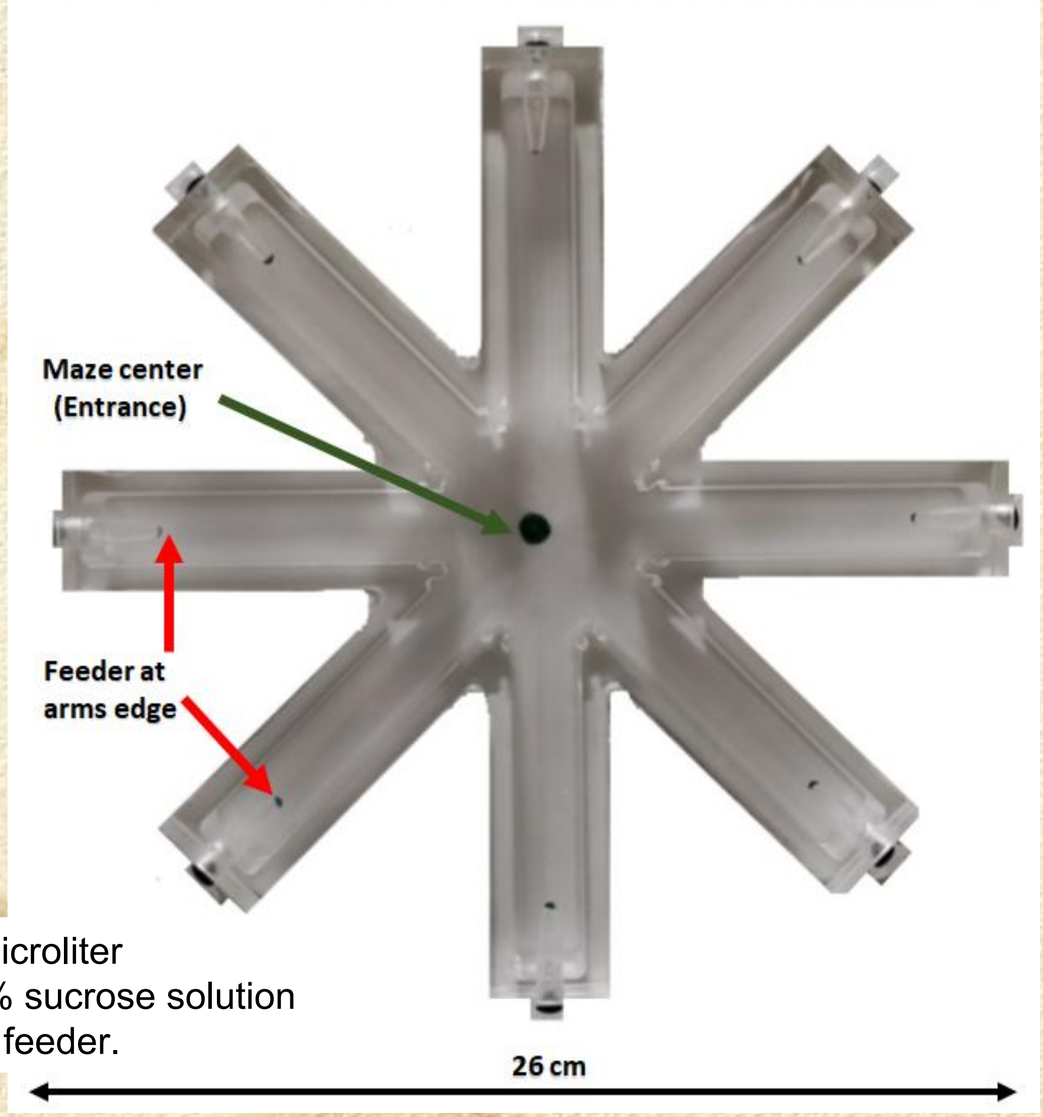






Andruchow, N.D., Konishi, K., Shatenstein, B., Bohbot, V.D., 2017. A lower ratio of omega-6 to omega-3 fatty acids predicts better hippocampus-dependent spatial memory and cognitive status in older adults. Neuropsychology 31, 724-734.

Spatial cognition: 8-arm maze



3 microliter
20% sucrose solution
per feeder.

Rozenbaum E, Shrot T, Daltrophe H, Kunya Y, Shafir S, 2024. Machine learning-based bee recognition and tracking for advancing insect behavior research. Artificial Intelligence Review 57. doi: 10.1007/s10462-024-10879-z.

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Hadassa Daltrophe

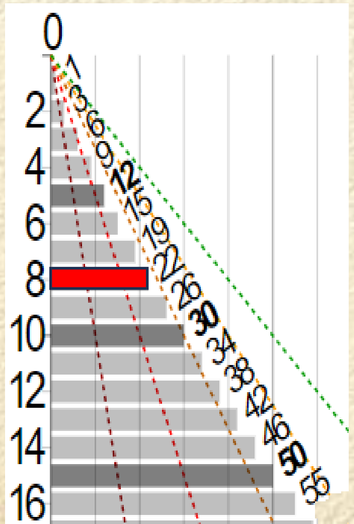
Tammar Shrot

6 bees/colony * 5 colonies = 30 bees/treatment

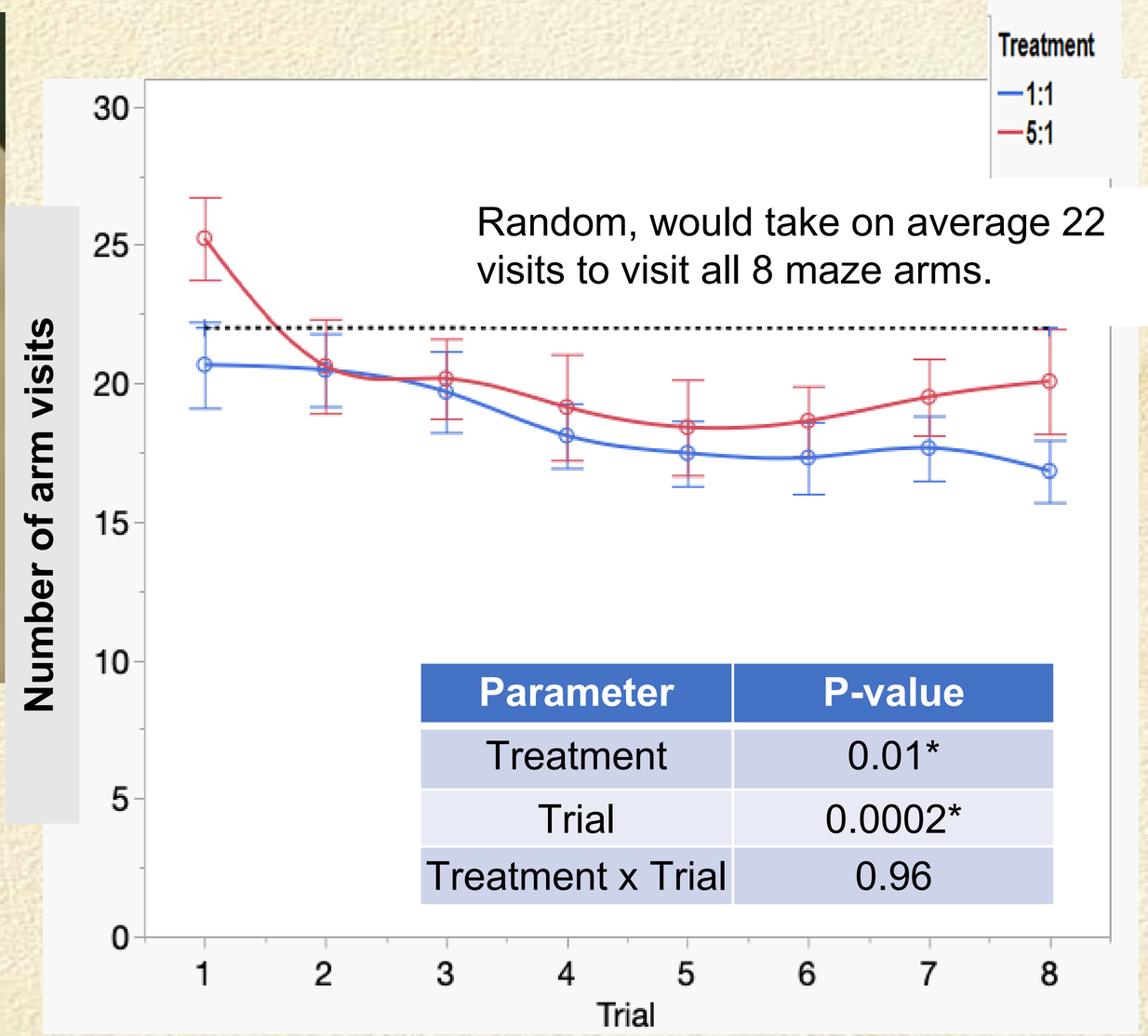
Diet / Ingredients	Balanced 1:1	Unbalanced 5:1
Soy flour (g)	39	39
Honey (g)	57	57
Corn oil (g)	1.1	3.0
Flax oil (g)	2.5	0.6
Omega-6:3 ratio	1	5

Spatial cognition: 8-arm maze

Coupon collector's problem

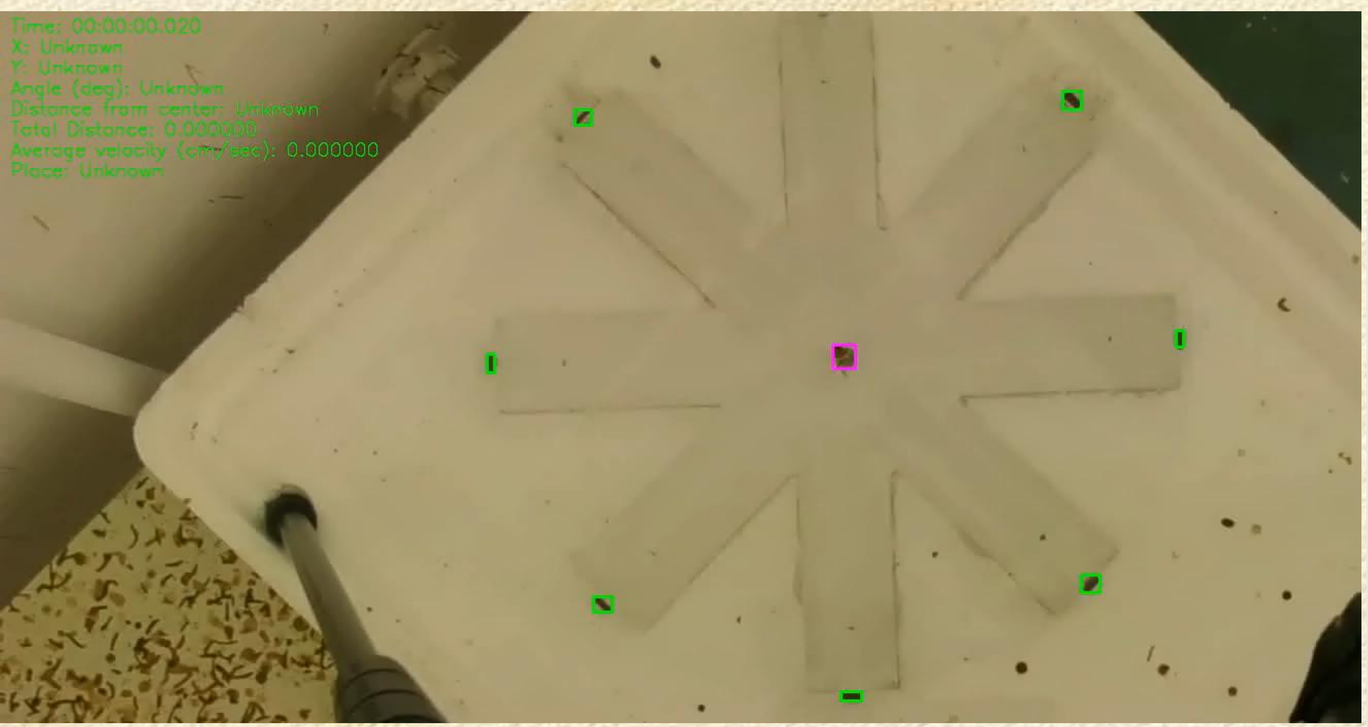


Bees fed a balanced diet solved the maze with fewer total number of visits

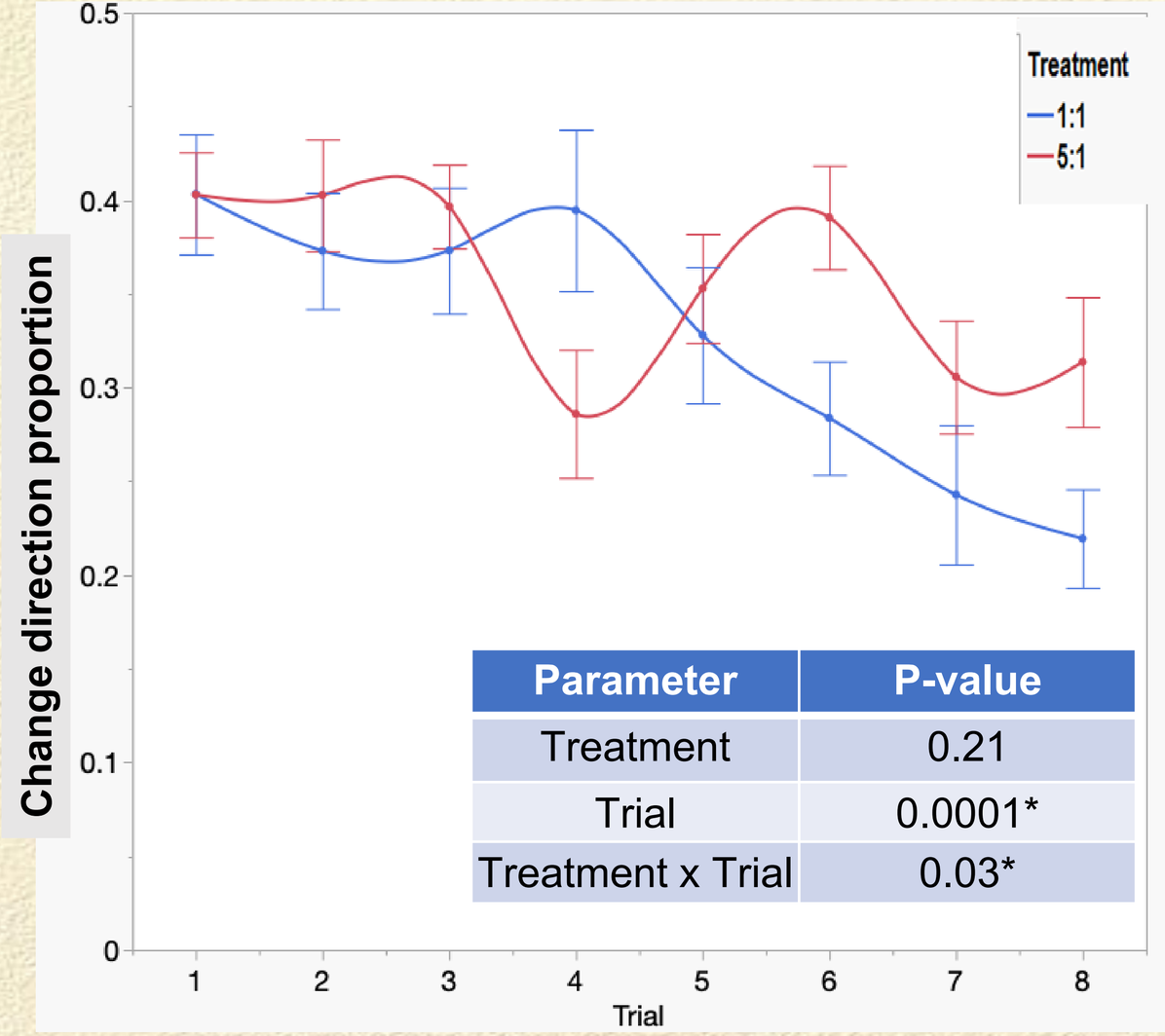


Parameter	P-value
Treatment	0.01*
Trial	0.0002*
Treatment x Trial	0.96

Spatial cognition: 8-arm maze



Bees fed a balanced diet developed a rule of thumb to move in consistent direction



Parameter	P-value
Treatment	0.21
Trial	0.0001*
Treatment x Trial	0.03*

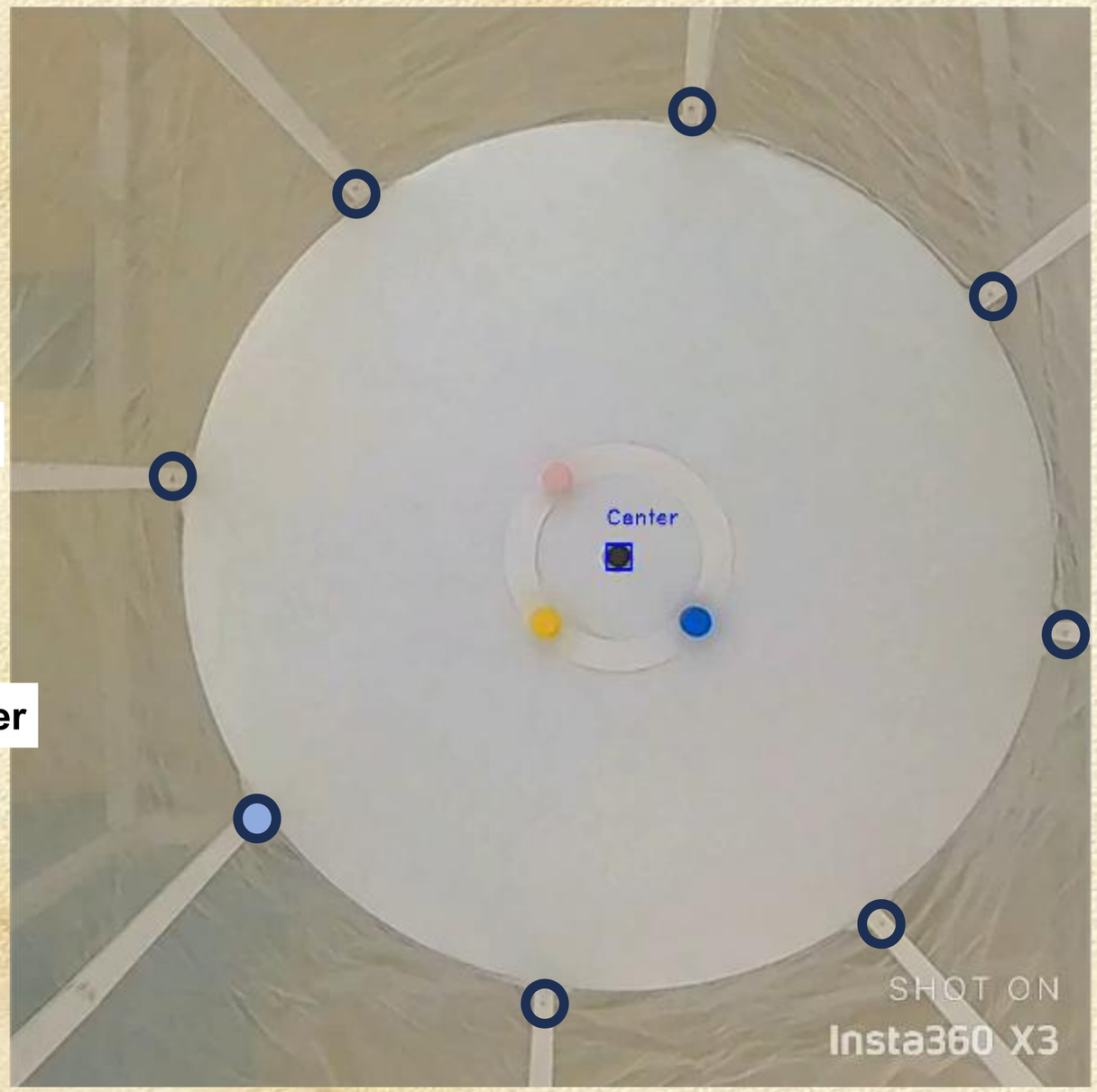
Spatial cognition: landmark learning

Ushitani et al. 2016.

Spatial cognition: landmark learning

Empty Feeders

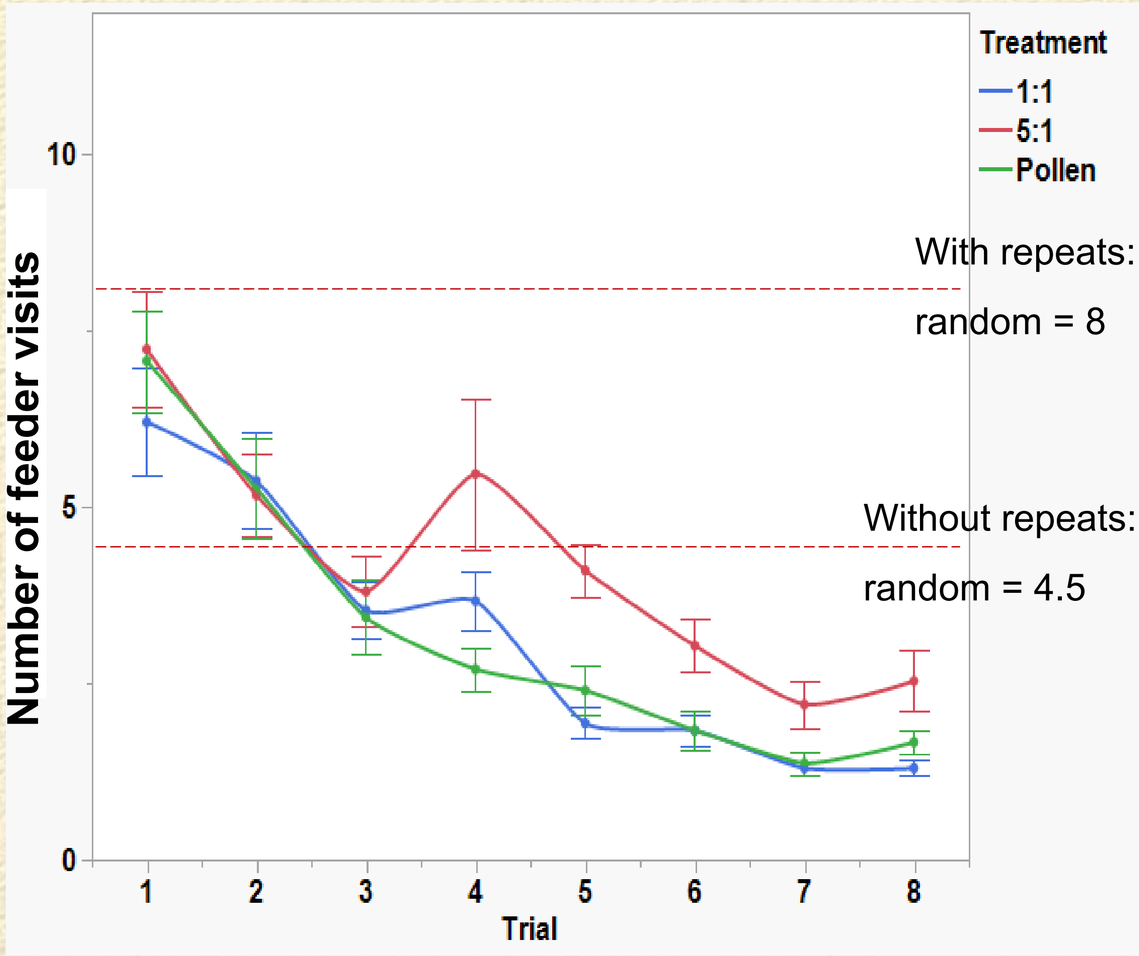
Rewarding Feeder



- Rewarding feeder changed, and location moved, every trial.
- Landmarks were rotated to face new feeder.

Trial 1

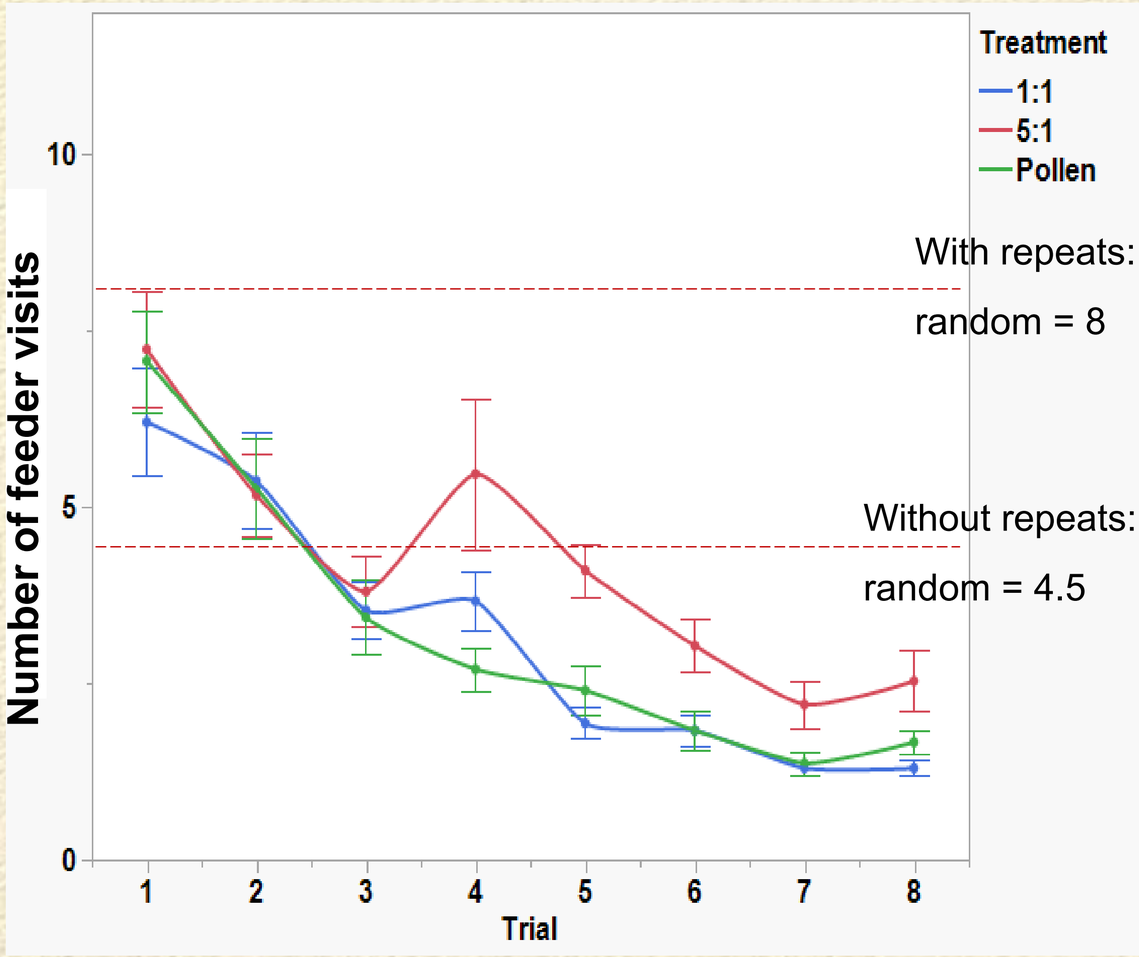
Bees fed a balanced diet found the rewarding feeder with fewer total number of visits



Parameter	P-value
Treatment	0.03*
Trial	<0.0001*
Treatment x Trial	0.06

Trial 8

Bees fed a balanced diet found the rewarding feeder with fewer total number of visits



Parameter	P-value
Treatment	0.03*
Trial	<0.0001*
Treatment x Trial	0.06

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- Diet affects honey bee cognition, including in spatial tasks.
- Colony strength would be affected by spatial cognition abilities.

Thanks

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