

#### Video Article

# Testing Visual Sensitivity to the Speed and Direction of Motion in Lizards

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#### **Abstract**

Testing visual sensitivity in any species provides basic information regarding behaviour, evolution, and ecology. However, testing specific features of the visual system provide more empirical evidence for functional applications. Investigation into the sensory system provides information about the sensory capacity, learning and memory ability, and establishes known baseline behaviour in which to gauge deviations (Burghardt, 1977). However, unlike mammalian or avian systems, testing for learning and memory in a reptile species is difficult. Furthermore, using an operant paradigm as a psychophysical measure of sensory ability is likewise as difficult. Historically, reptilian species have responded poorly to conditioning trials because of issues related to motivation, physiology, metabolism, and basic biological characteristics. Here, I demonstrate an operant paradigm used a novel model lizard species, the Jacky dragon (Amphibolurus muricatus) and describe how to test peripheral sensitivity to salient speed and motion characteristics. This method uses an innovative approach to assessing learning and sensory capacity in lizards. I employ the use of random-dot kinematograms (RDKs) to measure sensitivity to speed, and manipulate the level of signal strength by changing the proportion of dots moving in a coherent direction. RDKs do not represent a biologically meaningful stimulus, engages the visual system, and is a classic psychophysical tool used to measure sensitivity in humans and other animals. Here, RDKs are displayed to lizards using three video playback systems. Lizards are to select the direction (left or right) in which they perceive dots to be moving. Selection of the appropriate direction is reinforced by biologically important prey stimuli, simulated by computer-animated invertebrates.

#### Video Link

The video component of this article can be found at http://www.jove.com/video/127/

#### **Protocol**

### **Jacky Dragons**

Jacky lizards (A. muricatus) were held in climate-controlled captive facilities.

Enclosures were constructed of fabricated glass terrariums measuring 60 cm<sup>3</sup>. Enclosures were also filled with Sydney sand substrate, native foliage, and natural wooden perches. Enclosures were placed on top of movable trolleys that allowed for subjects to be placed in the middle of all viewing monitors.

Lizards were placed under a 14L:10D light cycle during simulated summer conditions, and were provided with both heat lamps (125W) and adequate ultraviolet (UV) lighting (300W) during the first half hour of daylight. Lizards were also provided with water ad libitum, fed crickets (*Achetus domesticus*) dusted with vitamins during non-experimental sessions, and routinely wormed for parasites using diluted doses of livermectin.

# **Equipment Used for Playback**

Three video playback rigs are used for stimulus presentation (see Figure 1). The central rig consists of an iMac (Apple Computer Inc.) containing dot stimuli. The two additional rigs consist of an iMac with Final Cut Pro HD for stimulus presentation that contained playback stimuli (i.e., invertebrates).

The digital signal is sent to an advanced digital video converter (Canopus©gt ADVC110, Melbourne, Victoria Australia) for digital-to-analog conversion for video playback and displayed on a Sony Trintron (Sony Corporation, Shinagawa, Tokyo Japan) monitor (Series Nos. PVM-14M2A/PVM-14L2/PVM-14N5A). A Canon (Model No. MV650i) digital camcorder with 22X optical zoom (Cannon Inc., Japan) is mounted directly above the testing area and records responses to stimuli directly to a Sony (Sony Corporation, Shinagawa, Tokyo Japan) video cassette recorder (Serial No. SLV-EZ717AS).

Recorded images are sent to a colour viewfinder (NEC, Serial No. 1892, Kanagawa, Japan). At 7.5 seconds from the start of the dots display, an electronically generated sound marker is sent to a Canopus® ADVC110 and records onto VHS. Video playback is adjusted for PAL-DV standard (5:1 compression; horizontal resolution 575 lines; 25 frames s-1).

# Random-dot Kinematograms (RDKs)

RDKs can be developed using VPixx V1.79 (VPixx V1.79, VPixx Technologies Inc. 2002).

Design stimuli with a 30 x 20 degree field, fill with 200 dots, with each dot measuring 5 x 5 pixels.

Build dot stimuli with eight variations in speed combined with eight levels of coherence and two directions, generating 128 different stimuli.

Speed parameters were designed in degrees per second (°/s). Speed variations were 0.5, 2.5, 5, 10, 20, 40, 80, and 160°/s. Coherence percentages were 0, 2.5, 5, 10, 20, 40, 80, and 100% in either a left or right directional motion.

Export each sequence as a Motion JPEG-B file. However, you can design the RDKs which an array of features.

Each sequence is a total 15 s in duration. The sequence begins with 2.5 s of 0% coherence matched with the trial speed variable. This period initiates the current trial. This is followed by 5 s of the variation in speed and coherence combination. The sequence was completed by 7.5 s of 100% in the appropriate direction.

Import all Motion JPEG-B sequences and edit in Apple Final Cut Pro HD (Apple Computers Inc.). Add a sound marker at each interval. This marker is only audible to VHS, and is used as a cue when scoring behaviour when the changes in stimulus presentation occur. Exported each of the 128 sequences as Final Cut Pro movies.

## **Primary reinforcers - Mealworms**

Place subjects on a randomised reinforcement schedule of five trials per block trial (16 trials) during the experiment. Primary reinforcers are mealworms (Tenebrio molitor).

## **Secondary reinforcers - Animated Invertebrates**

Secondary reinforcers are to be paired with the primary reinforcer. Created biologically meaningful reinforcers, such as three types of computer-animated invertebrates: 1) cricket, 2) mite, and 3) spider.

Create animations using Lightwave v8.3 (NewTek Inc., San Antonio, TX, USA) for 15 s in length, with the initial 7.5 s of still background, and followed by 7.5 s of the invertebrate on the appropriate screen.

Photograph a background of natural vegetation to import as a JPEG in eah clip. Export each sequence into individual JPEG image sequences (375 frames), and compress into a movie file using Final Cut Pro HD for video playback. Design 50 unique clips to prevent habituation to the conditioning paradigm.

### **General Procedure**

Place subjects between the viewing monitors.

Randomize presentation of RDK stimuli for each subject.

Show stimuli in blocks of 16 trials over eight days. Each trial is 15 s with intertrial intervals varying from 30-60 s. Segment each trial interval into 2.5 s of 0% coherence reflective of testing speed, 5 s of testing speed and coherence, and followed by 7.5 s of 100% in appropriate direction matched with speed.

During this 5 s critical-period, subjects are required to respond. Record dependent measures as orientation towards a left or right monitor and latency to respond.

Use only five random trials with a primary reinforcer and reinforce each trial with an animated invertebrate. At the completion of each block trial, remove subject enclosures from the testing area, and introduce another enclosure.

### **Discussion**

Testing the sensory capacity in lizard species has been difficult, and has largely been both a tedious and arduous process. These difficulties stem from finding the correct reinforcer or establishing a functional protocol. Here, I established a successful protocol in which to test motion perception in lizards. This procedure may also be applicable in testing other visual characteristics as well as other sensory mechanisms. In addition, this procedure may be applicable to other species. The usefulness of this protocol not only allows us to understand natural behaviour, but to use lizard behaviour as a model for other scientific endeavours.

### **Disclosures**

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