

Using video playback to study sexual communication

Gil G. Rosenthal

Department of Zoology C0900, University of Texas, Austin, TX 78712, U.S.A. (e-mail: fishman@mail.utexas.edu)

Received 4 February 1997

Accepted 4 January 1999

Key words: sexual selection, mate choice, aggressive behavior, experimental design, animations, visual signals, pseudoreplication

Synopsis

Many sexually-selected phenotypes involve some form of visual communication. Video-playback techniques are a powerful new tool for studying visual signaling systems. Individual aspects of complex stimuli can be reliably manipulated, and stimuli can be repeatedly presented without appreciable variation in their properties. Experimenters can also construct signals which are biologically impossible, but which can be used to ask critical questions about how stimuli are perceived. Video-playback studies of sexual selection are reviewed in the context of how the methodology can be used to extend the range of questions addressed by conventional techniques. Four major issues are discussed with consideration for future studies: (1) the background against which a stimulus is presented; (2) the illumination of the stimulus; (3) the problem of pseudoreplication; and (4) experimental design considerations, including controlling for side biases and order effects and selecting appropriate response assays. Using synthetic animations may address many of these concerns.

Introduction

'The ability to present a putative signal in isolation, without the potential confounding effects of other activities of the signaler, is the main reason for the depth and range of our knowledge of communication systems. To date, playback of sound signals has predominated, but playback of electric signals and even video playback of visual signals suggests that playback will become just as prevalent in studies of communication in other sensory modalities.'

McGregor (1992, p. v)

The study of sexual selection is intimately entangled with that of animal communication. Sexual selection can shape the evolution of signals and the perceptual systems that receive them. Conversely, the design of perceptual systems can constrain the direction of

sexual selection. Any experimental research program aiming to understand mechanisms of sexual selection must therefore include, at some level, an analysis of the appropriate communication processes. As McGregor (1992) points out, this involves presenting a receiver with isolated features of a signal. This approach has been commonly used in several modalities, including vision: presentations of static models and manipulations of traits on live animals are among the classic experiments of early ethology (e.g. Pelkewijk & Tinbergen 1937). It is only with the recent advent of video playbacks, however, that researchers have had the opportunity to play back visual stimuli displaying the complex patterns of temporal change typical of many natural signals.

Visual signals are important in both mate selection and intrasexual competition. The vast majority of studies on fishes cited in Andersson's (1994) comprehensive review of sexually-selected traits demonstrated the salience of one or more visual traits. A typical

courtship or agonistic interaction may involve multiple spatial cues (body size, the presence of an ornament, or patterns of pigmentation), temporal cues (motor patterns), and spectral cues (coloration). These cues are typically dynamic, changing over time in multiple dimensions.

Understanding this spatiotemporal complexity has historically proved intractable. Presentations with static models are useful for testing many hypotheses, but models usually fail to adequately simulate motor displays. Using live exemplars introduces confounding factors when extraneous traits covary with those being studied, and makes many trait manipulations impossible.

Video playback offers a resolution to these problems. Video sequences can be edited to produce realistic exemplars varying precisely in one or more aspects of morphology and behavior, and identical in all other characteristics. Video can also be used to construct stimuli not found in nature, such as 'chimaeras' bearing the behavior of one species and the morphology of another, or disembodied parts of animals performing species-typical motor patterns.

I will briefly review the video-playback studies on sexual selection published to date, illustrate some advantages relative to conventional experiments, and outline some considerations for future studies using the technique. I will discuss aspects of the design of stimuli and experiments that can be readily addressed by most workers in the field. By definition, studies involving sexual selection refer, at least implicitly, to fitness consequences in natural populations. Certain aspects of stimulus design and presentation are thus of critical importance to the external validity of these studies. I will concentrate on four issues relevant to all experiments on visual communication, but particularly to sexual selection studies: (1) the background against which a stimulus is presented; (2) the illumination of the stimulus; (3) the problem of pseudoreplication; and (4) experimental design considerations.

Video-playback studies of sexual selection

To the author's knowledge, all video-playback studies of fishes published to date deal with some aspect of sexual selection, as do several of the video-playback studies published on other organisms. The studies will be discussed according to the characters, if any, which are manipulated.

No manipulation

While most video-playback studies have used the methodology to manipulate stimulus characters, several have used unmanipulated video footage in presentations. These studies have primarily used video techniques to eliminate undesired variation in stimulus characteristics. This is clearly desirable when such variation could confound interpretation of the results. Landmann et al.'s (1999) ontogenetic study addressed changes in female preferences over time. Any change in male behavior over time (e.g. due to seasonal or experiential effects) would have interacted with the effect under consideration.

Concerns can also arise when differences among receivers in an experiment affect the behavior of signalers, or when an interaction between the stimulus individual and the subject could confound interpretation of the results (e.g. conspecific versus heterospecific, Macedonia et al. 1994, Macedonia & Stamps 1994). In such cases, the motivation of individuals to display would be expected to vary substantially according to the identity of the receiver.

For a hypothetical example, females may exhibit a strong preference for males of their own species over those of a closely related species when shown live males displaying behind two-way glass on opposite ends of an aquarium. This result, however, can be interpreted in at least two ways. Females may recognize a conspecific mate signal and thus exhibit a preference for a male of their own species. Alternatively, males of both species may be more motivated to court conspecific females, and females may simply be choosing the male with the higher level of courtship activity. Moreover, if the stimulus males are allowed to communicate with one another, the 'preferred' male may simply have suppressed courtship activity in his counterpart.

Unmanipulated analog video can also be combined with signals from other modalities. Multi-modal playback – the simultaneous presentation of acoustic and video stimuli – has been used with considerable success to study social communication in birds (reviewed in Evans & Marler 1995). This approach was also applied to male-male displays by Baker et al. (1996).

Two studies using unmanipulated video have tested the validity of video techniques. Male and female *Anolis* lizards in the field responded strongly to video playbacks of displaying males (Clark et al. 1997), suggesting that video playbacks can be used to study responses under natural environmental and

motivational conditions. Kodric-Brown & Nicoletto (1997) used analog footage to draw two important conclusions about video playbacks in general. While female guppies, *Poecilia reticulata*, spent more time interacting with a live male behind clear glass than with a video stimulus, they showed no difference in association time between a video stimulus and a live male behind one-way glass, suggesting that the physical characteristics of video were not limiting in this system. Females were more consistent in their choices with video stimuli than with live males behind clear glass, presumably due to the lack of temporal variability in male behavior across presentations. Video techniques thus proved more effective at detecting female preferences than standard methods using live males.

Behavior

A unique, novel property of video playbacks is that they allow the controlled manipulation of complex temporal changes in stimulus characteristics. It is generally impossible to reliably elicit a desired behavior from a live stimulus animal, and static models are inadequate for testing hypotheses involving responses to motor patterns. Two studies have manipulated behavior patterns while holding morphology constant. Rosenthal et al. (1996) presented female green swordtails, *Xiphophorus helleri*, with edited sequences of males performing courtship behavior and of the same males performing a suite of other activities. Rowland (1995) directly manipulated courtship itself. A male performing a characteristic display was videotaped and an edited sequence dubbed onto playback tapes at varying speeds, producing sequences with higher and lower rates of courtship.

Morphology

McClintock & Uetz (1996) used frame-by-frame alteration to study female preference patterns in two closely related species of wolf spiders, *S. ocreata* and *S. rovneri*. Male *S. ocreata* use tufts of bristles on the forelegs in visual courtship displays, while male *S. rovneri* lack tufts. Animation techniques were used to digitally add or remove tufts, thus creating a stimulus male of one species bearing a heterospecific morphological trait. Uetz et al. (1996) used the same technique to test symmetry preference, digitally removing the tufts on one leg only. Allen & Nicoletto (1997) used similar techniques to manipulate fin length within males of one species, the Siamese fighting fish *Betta*

splendens. Fins were lengthened or shortened beyond the typical value for live fish.

Rosenthal & Evans (1998) digitized a portion of one of the courtship stimuli from Rosenthal et al. (1996) and manipulated the presence of two components of the sexually-dimorphic male 'sword'. Females preferred males with full swords over males with one component (the sword extension) removed, and over males with both components removed. Rosenthal & Evans (1998) then addressed the perceptual nature of the female preference for males with long sword, by giving females a choice between males with swords and swordless males of equivalent total length; this abolished the preference for sworded males. Females also failed to show a preference for a disembodied courting sword over a blank screen control, and did not discriminate between a disembodied sword and a swordless male of comparable length.

These studies have manipulated the morphology of realistic stimuli, often outside the range of the biologically possible. The fact that state-of-the-art techniques permit almost any possible manipulation of morphology, however, raises a series of thorny issues for an experimenter designing a stimulus. For example, if a high-contrast ornament, such as a sword in *Xiphophorus*, is digitally erased, should it be replaced with background, or with the lower-contrast pattern of the rest of the animal's body? Rosenthal & Evans (1998) opted for an approach that reflected biological variation in stimulus properties: individuals lacking sword extensions altogether are common in some species, so this component was erased and replaced with background. Males naturally lacking a stripe on the caudal fin, however, are not missing any portion of the fin, so we replaced the stripe with patterns digitally copied from the upper portion of the caudal fin.

Size manipulation is also potentially complicated. If the size of an ornament, or of the whole animal, is being manipulated, should it be uniformly scaled, scaled along one axis, or scaled according to observed allometries? With frame-edited video, all but the first option present a logistical headache for any stimuli varying in three dimensions; synthetic animations (see below) would be more amenable to more sophisticated manipulations of size.

Morphology and behavior

Three pioneering studies by Clark & Uetz (1990, 1992, 1993) provide perhaps the most striking demonstration

of video techniques to date. Male jumping spiders, *Maevia inclemens*, exhibit extreme dimorphism in morphology and behavior. Gray morphs crouch down in a prone position upon sighting a female, while tufted morphs stand up. Clark & Uetz (1993) used frame-by-frame manipulation to apply the behavior of each morph to the morphology of the other. These studies exemplify the 'impossible', yet biologically realistic, stimuli that can be created with video techniques. Such stimuli can yield important insights into the design of complex signals, in which multiple components interact to produce a response.

Body color

Responses to colors manipulated on videotape are generally difficult to interpret. Color video systems are tuned to human spectral sensitivity functions, and manipulation as perceived by humans should not be assumed to correspond to an analogous manipulation from the study organism's point of view. As an example, a comparison of male responses to digitally-manipulated yellow versus orange abdominal patches in a brooding female cichlid would be effectively meaningless without detailed knowledge about the perceptual biology of the fish. Males might not perceive any difference between the two treatments, and it is unlikely that either manipulated color would be perceptually equivalent to that found in live fish. Fleishman et al. (1998) provide a detailed discussion of the color-matching problem in video playbacks.

Several studies have used video techniques to examine response to nuptial coloration in threespine sticklebacks. The authors of these studies argue that the stickleback peripheral visual system is quite similar to humans'; furthermore, the color being manipulated is usually red, which corresponds very closely to one of the three phosphors on a color television screen.

A variety of techniques has been used to manipulate the presence of red in stickleback stimuli. Bolyard & Rowland (1996) and Rowland et al. (1995a,b) manipulated the entire video signal, presenting subjects with the same video sequence at varying color intensities and in grayscale. McKinnon (1995) used chroma-keying, a technique whereby specific analog-video colors are selectively altered, to manipulate only the throat color of a courting male. McKinnon & McPhail (1996) tested male sticklebacks on synthetic animations of red-, black-, and gray-throated males derived from a single live male exemplar.

The above studies used video techniques to keep behavior and morphology constant while manipulating color within the natural range of variation. McDonald et al. (1995) coupled this manipulation with a complementary alteration of the surrounding background. Sequences of a courting male were altered using a video-processing system that permitted alteration of a selected color in the video signal, and presented to females.

While red coloration in sticklebacks may represent a special case, it is important to note that video playbacks are generally inappropriate for questions involving color. The color-matching problem, however, is not insurmountable. Metamers – in this case, combinations of red, green, and blue which are perceptually indistinguishable from other spectral distributions – can be experimentally determined through conditioning techniques or physiological studies. For many cases (e.g. investigating preferences for orange over yellow patches) this would be necessary only for a subset of the color spectrum.

A cruder, yet less laborious, approach is to use grayscale stimuli in which the color being studied is set to have the highest luminance value; this is easily accomplished with several software applications in which the grayscale conversion parameters can be adjusted. The stimuli are played back on monochrome monitors, and a color filter of known spectral distribution is placed over the image. This procedure is analogous to presenting live stimuli under different lighting conditions (e.g. Long & Houde 1989). This approach would be most suitable for systems in which most of an animal's body was a single color.

The studies reviewed above illustrate two principal advantages over conventional manipulations. Experimenters have much greater control over stimulus characteristics, both in their quantitative properties and in their consistency across presentations and between treatments. Also, video allows the creation of stimuli that are effectively impossible to produce with live animals or models.

The technology available for video image processing and for the construction of synthetic animations is continuously becoming more powerful and less expensive. Video playbacks are often the only alternative for controlled experiments on visual signals. The following sections offer some caveats on the design of video stimuli and video playback experiments.

Stimulus properties

Background

At the most fundamental level, visual perception works by the detection of differences in the spectral properties of an object relative to its background (Sekuler & Blake 1994). Background properties are crucial to the perception of sexually-selected phenotypes. The size of color spots on male guppies, *Poecilia reticulata*, in predator-free experimental populations increased when populations were kept against a fine-grained substrate, and decreased against a coarse-grained substrate (Endler 1983). McDonald et al. (1995) demonstrated that the preference of female sticklebacks for throat patch color depended on the ornament's contrast with the background, rather than on its absolute spectral characteristics. McKinnon & McPhail (1996) reviewed studies on color preferences in sticklebacks and argued that differences in (often unreported) background characteristics might partially account for the inconsistency between studies. Female *Anolis* lizards also express a preference based on the contrast between a male's dewlap and the background (Sigmund 1983).

McKinnon (1995) and McKinnon & McPhail (1996) chose a background for presentations that approximately matched the background against which sticklebacks are viewed in natural populations. In most video-playback studies published to date, however, and in most studies involving manipulation of visual traits in general, background characteristics are biologically rather arbitrary. Background is often selected so as to maximize stimulus conspicuousness and minimize processing artifacts. Such technical constraints should be less limiting in future video playback experiments. State-of-the-art image-capture and display equipment produces images that are in some cases superior, in color and spatial resolution, to analog S-VHS or Hi-8 video. Moreover, video techniques make it easy to adjust background characteristics to the experimenter's specifications.

Most playback studies in sexual selection are testing the explicit or implicit hypothesis that variation in one or more characteristics ultimately influences reproductive success in natural populations, whether the characteristic belongs to the sender or the receiver. The representation of a stimulus is thus constructed with reference to what an animal would encounter in nature, even when stimuli are abstracted or have properties outside the natural range of variation. For

example, if one were testing the effect of the size of male body spots on female mating preferences in guppies, the logical first step would be to present females with sequences showing male guppies with varying spot sizes, as opposed to, say, sticklebacks or rectangles exhibiting similar variation. This appears to be a trivial point when one considers the *animal* as the context for a trait, but experimenters almost always place characters in a *background* context that is biologically meaningless. This can potentially have dramatic effects on experimental results. Figure 1 shows a still-frame of a synthetic animation of a courting male swordtail, *Xiphophorus nigrensis*, against a series of backgrounds. To the human eye, and most likely to female swordtails, the sword extending from the caudal fin differs dramatically in apparent length, thickness, and contrast depending on the background.

For researchers with access to natural populations of their study organisms, appropriate background characteristics can be approximated by sampling behavioral interactions from the point of view of the receiver. For example, if male-male aggressive displays typically occur in midwater, signalers are viewed against a diffuse, relatively homogeneous background. If males are defending patches of habitat along a riverbank, displays are more commonly viewed against a complex, variegated background.

There are several ways one could introduce natural backgrounds into playback experiments. For questions which can be addressed with analog-video stimuli (e.g. Rosenthal et al. 1996, Landmann et al. 1999), the easiest approach would be to use edited sequences of animals in the field. Alternatively, one could videotape an animal against a homogeneous background and electronically substitute a natural background. With digitally-manipulated video or synthetic animations, one could either use a single frame of background for the entire stimulus, or, preferably, digitize a brief video sequence of background and use any of a number of software applications to superimpose the manipulated stimulus on the background sequence.

Illumination

The direction and quality of illumination can have a substantial effect on stimulus properties (Sekuler & Blake 1994). In aquatic environments, the strongest illumination comes from overhead. This property is often exploited by fishes to produce a counter-shaded effect that minimizes visibility to predators

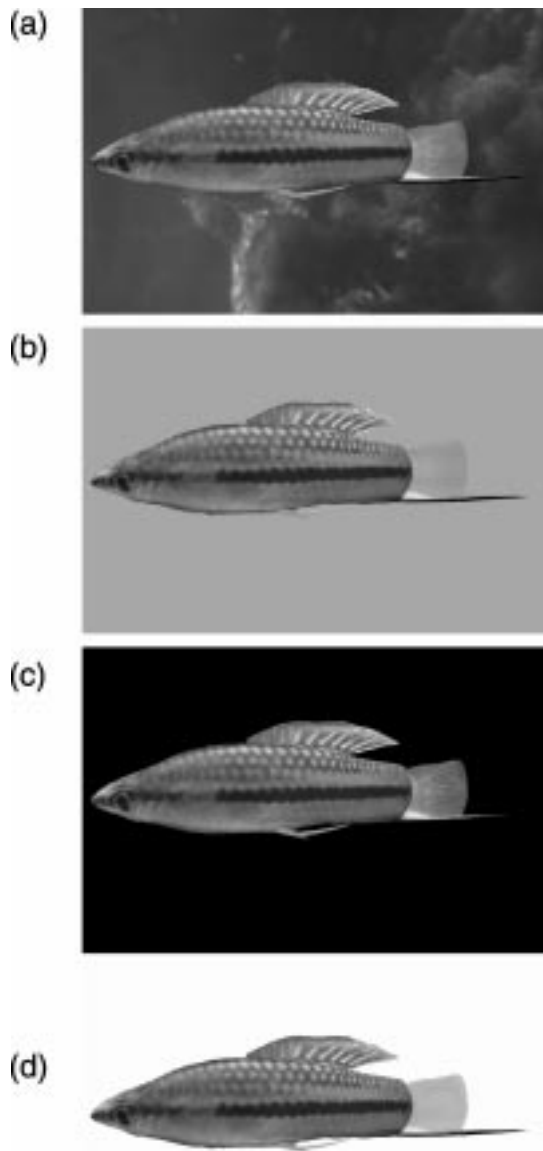


Figure 1. Still from a synthetic animation of a courting male pygmy swordtail, *Xiphophorus nigrensis*, against a – slide of natural background in the Río Choy, San Luis Potosí, Mexico; b – monochrome background based on the mean of 20 sampled RGB values for water ‘spacelight’ color on field videotapes; c – black background; d – white background. Stills are rendered color images converted to grayscale. Note variation in the appearance of the sword across the four images.

(Loew & McFarland 1990). In many taxa, the dorsal surface is dark, blending in with the substrate below, while the ventral surface is lighter, blending in with the surface. Changing the direction of incident light

can substantially alter the perception of a stimulus (Sekuler & Blake 1994). This is perhaps best exemplified by the Halloween trick of shining a light under one’s face to cast sinister shadows.

The positioning of lighting used in filming stimuli is mentioned in only two published video-playback studies of sexual selection (McDonald et al. 1995, Rosenthal et al. 1996). In both of these, lights were placed so as to maximize contrast with the background, in order to facilitate image processing. The stimuli may therefore not have exhibited illumination patterns comparable to those found in nature. Most of the studies reviewed manipulate color or behavior, rather than the arrangement of patterns on the body, so the placement of illumination may not be critical. Nevertheless, it is clearly desirable to have the illumination properties of the stimulus reflect patterns encountered in the field.

The spectral distribution of incident light used when filming stimuli is also a concern. Fishes in a natural environment are illuminated by sunlight, which may be filtered through any combination of clouds, terrestrial vegetation, aquatic vegetation, and particles dissolved or suspended in the water (Endler 1990). Long & Houde (1989) showed that the spectral distribution of ambient light could eliminate female mate choice patterns in guppies. All of the studies reviewed (except McKinnon & McPhail’s (1995) synthetic animation) presumably used artificial indoor light, directly striking clear water, in stimulus production.

Even if one were to use field footage – videotape of animals displaying under some subset of natural light conditions – in the preparation of stimuli, the original spectral distribution would be distorted at least twice – once as the light reflecting off the displaying animal was transduced to videotape, and once as the playback equipment outputted the video signal. Nevertheless, it is reasonable to expect that using natural light regimes, or an approximation thereof, would generally produce more biologically relevant stimuli than the usual artificial-light approach.

There are several properties of video that may introduce confounding factors into an experiment if they interact with the perception of stimuli. These include poor color and brightness fidelity, low spatial and temporal resolution, and the absence of many depth cues. As the above sections point out, however, video can be used to more easily minimize other potential confounding effects peculiar to visual traits, by introducing background and lighting characteristics appropriate to the hypotheses being tested. Moreover, these techniques offer experimenters the opportunity to explicitly test

the effects of these characteristics on the perception of stimuli (e.g. McDonald et al. 1995).

Experimental design

Pseudoreplication

The problem of pseudoreplication in playback experiments was the focus of a major symposium on acoustic playback studies (McGregor et al. 1992). I will briefly summarize the 17 participants' consensus paper, which suggests how pseudoreplication should be dealt with in playback experiments, and then propose an analogous approach for video playbacks.

The term *pseudoreplication* was originally proposed by Hurlbert (1984) in the context of field ecology. McGregor et al. (1992) modified the original definition somewhat to better apply to playback studies: 'the use of an n (sample size) in a statistical test that is not appropriate to the hypothesis being tested'.

If, for example, female guppies were being tested for their response to population-level differences in spot patterns, a typical study might run as follows: (1) females are tested for their response to live males from their own population, X, and another population, Y; (2) after finding a significant preference for sympatric males, the experimenter prepares four digitally-altered video sequences, showing a sympatric male with his normal spot pattern; the same, sympatric male with an allopatric pattern; and the complementary sequences for an allopatric male. This is perhaps a two-month procedure, but using manipulated video is necessary in order to determine whether variation in spot pattern per se, independent of behavior and other factors, influences female preference.

The sample size to be used in a statistical test for this experiment depends on the hypothesis. If the alternative hypothesis is extremely narrow – that Male X1's spot patterns are more attractive to females from population X than are Male Y1's, then one could treat each female's response as a data point (McGregor et al. 1992). If, however, the hypothesis is that population X females prefer male spot patterns from population X over population Y, the sample size is effectively equivalent to the number of exemplars – in this case, one. In order to test the more general hypothesis – namely, that all female guppies prefer the spot patterns of males from their own populations – one would have to test multiple subjects from multiple populations on multiple exemplars from multiple populations, a project

which would surely require multiple generations of multiple video-manipulators.

McGregor et al. (1992) point out that the use of synthetic stimuli can often circumvent the pseudoreplication problem. A strength of video techniques is that one can artificially modify stimuli so that subjects are presented exemplars varying only in the traits under consideration. One could redesign the above guppy study slightly and present females with manipulations of a male from their own population, in which the spot pattern was varied between the sympatric and allopatric averages. Every aspect of the stimulus would be controlled, except for spot size. This would allow the experimenter to use an n in inferential statistics that was equal to the number of subjects, provided the hypothesis was that manipulation of *this particular male* affected female response (McGregor et al. 1992). This approach has been used in the majority of the video-playback studies reviewed.

A potentially more powerful approach than manipulation of live males is the *de novo* synthesis of artificial stimuli from population data, a technique which has been used with considerable success in studies of acoustic signals. Signal parameters are measured in samples of individuals, and their average values are used to generate synthetic stimuli. Ryan & Rand (1995) constructed frog calls using data on eight call variables gathered from 10 individuals in each of eight species; females did not show a preference for natural calls over these synthetic calls. Using such stimuli can augment the external validity of the results. Rather than having to restrict the alternative hypothesis to manipulated differences in one, arbitrarily-chosen exemplar, one can test for the effects of manipulating traits in the context of phenotypic variation in a population or species.

Such techniques can now readily be employed with video. Clark & Uetz (1992) used the mean values for several behavioral parameters to construct 'abnormal' morphs in their study. McKinnon & McPhail (1996) recently demonstrated that female sticklebacks will respond to a synthetic animation of a male. Current animation and rendering software makes it feasible to generate similar animations based on estimates of population values for morphological and behavioral parameters. This would have an additional advantage in terms of the time necessary to make stimuli. There would be a considerable initial time investment in constructing a synthetic sequence which elicited the same response as an analog-video sequence. Once this sequence was created, however, behavioral and morphological manipulations of chosen traits would be

very easy to generate; one would just have to change those particular parameter values once, and they would be automatically applied throughout the animation.

Controlling for order effects and side biases

It is of course always desirable to control for effects of presentation order and possible side biases (Martin & Bateson 1993), but these factors are of particular concern for video-playback experiments. When an experiment involves the manipulation of a few traits in a sequence and subjects are serially presented with many variations on the same sequence, the potential for habituation across trials is very high. Conversely, subjects might respond *more* strongly to a given sequence if they are familiar with the stimulus or its context after repeated presentations. Experimenters should take care to vary presentation order across subjects, either systematically (when the number of possible presentation sequences is less than or equal to the number of subjects) or randomly (when the number of possible presentation sequences is much greater than the number of subjects). Depending on the statistical design of the experiment and the availability of study animals, it may be preferable to use a naïve subject for each presentation.

Side biases, in which a subject shows a stimulus-independent preference for one side of an aquarium or arena, are a hobgoblin of playback experiments in which stimuli are presented simultaneously. With video playbacks, there may be subtle between-side differences in monitor tuning or in the playback speed or signal quality of the videocassette player; this is often the case when one piece of equipment has suffered more previous use than another. Care should be taken to vary stimulus presentation at least between subjects, and ideally within subjects. The nature of video playbacks makes the latter very straightforward: in the simplest case, all the experimenter need do is construct two tapes, one with the sequence 'stimulus A → interval → stimulus B' and one with the sequence 'stimulus B → interval → stimulus A'. Successive subjects are shown the stimulus tapes on alternate sides (Rosenthal & Evans 1998). This design thus controls for side biases *within* subjects, by reversing the sides on which the stimuli are presented; and *across* subjects, by varying the initial arrangement of stimulus presentation. By showing each stimulus in identical proportion on each side's equipment, any biases due to differences in playback apparatus are eliminated.

Response assays

Some video-playback studies lend themselves readily to the criticism that it is difficult to be sure that animals are responding in a biologically-meaningful way. This can be easily avoided if subjects perform behaviors which are specific to aggressive or sexual interactions, such as bite-butts in sticklebacks or head-bobs in anoles. Some systems, such as those using side-association measures of preference, may lack such clear-cut assays of response; in these cases, one solution is to use video techniques to replicate previous studies using live stimuli. If subjects exhibit a pattern of responses consistent with previous results, they are likely responding to video sequences as biologically salient stimuli. Alternatively, experimenters could use an independent assay of sexual or aggressive motivation, such as a physiological measure or a behavioral response to a set of stimuli other than those under consideration, to screen for responsive individuals (Rosenthal & Evans 1998).

Conclusions

While the study of visual communication has been a major focus of ethology since its origins (e.g. Pelkewijk & Tinbergen 1937), this area has lagged in terms of the rigor with which studies can be conducted. A major limitation has been the difficulty of designing well-controlled experiments testing responses to stimuli. For all their drawbacks, video techniques have the potential to substantially extend the validity of experiments on visual signals, as well as to extend the range of questions which can be experimentally addressed. The studies reviewed illustrate this potential quite well. Subjects can be presented with standardized stimuli varying in behavior, but identical in morphology; varying in morphology, but identical in behavior; displaying identical behavior at varying rates; displaying combinations of morphology and behavior not normally encountered; or displaying normal behavior and form, but atypical background contrast properties. Moreover, even when a study does not call for manipulation of traits, video provides a stimulus which is unvarying over time: the same individuals can be presented the same stimulus at different points in ontogeny.

Further video-playback studies should take advantage of currently available methods which make it possible to reduce the likelihood of confounding effects

caused by arbitrary background or lighting characteristics, and to increase statistical power by avoiding pseudoreplication. While it may be difficult to always address these factors, they should be given priority over the temptation to produce the stimuli which look 'best' to a human observer. Information on background and lighting characteristics, and the number of exemplars used to produce stimuli, should be included in the methods section of future publications. Experimenters should also consider the physical limitations of video playback, which render many possible studies uninterpretable without detailed knowledge of the perceptual biology of the study organism.

Despite these caveats, video playback remains one of the most powerful new techniques in the study of animal behavior. Among the canonical model systems in studies of sexual selection are taxa for which there is abundant information on genetics, distribution, physiology, and evolutionary history, but whose reliance on visual signaling has often made it difficult to rigorously test hypotheses about the evolution and design of communication systems. Notable among these are small freshwater fishes such as guppies, mollies, swordtails, and sticklebacks, which together are the subjects of over half the studies in this review. Video techniques offer an exciting opportunity to address complex questions about sexual communication in such systems.

For example, the ability to manipulate the presence and behavior of stimulus exemplars provides a new array of tools for studies of mate-choice copying. The ability to 'mix and match' conspecific and heterospecific signals, or reconstruct ancestral phenotypes, will surely prove invaluable to students of species recognition and the role of preexisting biases in signal evolution. The long-neglected role of experiential and ontogenetic effects in the expression of mate preferences can now be examined with a methodology that keeps stimuli constant over time. Video can be used to derive a detailed picture of the perceptual landscape of receivers, yielding insights on some of the proximate constraints on the design of the signal-receiver dyad and on the interpretability of playback experiments themselves.

Complex sexual phenotypes consist of many interacting traits. In order to characterize the selective regimes acting on these phenotypes, and in order to understand the constraints underlying these selective regimes, we need a methodology that allows traits to be controlled, exaggerated, abolished, or abstracted, all the while preserving any necessary complexity in the

stimulus as a whole. Cinema was revolutionized, for better or for worse, by the advent of sophisticated visual effects in 'Star Wars'. Such 'special effects' have finally trickled down to the scientific community, where it is hoped that their impact will be just as revolutionary and decidedly positive.

Acknowledgements

This issue and the symposium that prompted it represent an important contribution to a new field. A. Kodric-Brown and P.F. Nicoletto deserve special thanks for their efforts in planning and organizing them both. W. McClintock's excellent Video Playback Webserver (<http://sensorydrive.lscf.ucsb.edu/>) was of considerable help in surveying the burgeoning literature in this area. I am indebted to C.S. Evans and J.M. Macedonia for introducing me to video-playback techniques and rigorous experimental design. L. Dries, N. Guda, L. Higgins, D. Johnson, N. Kime, A. Kodric-Brown, J.M. Macedonia, S. Phelps, M.J. Ryan, I. Schlupp, R. Thompson, K. Warkentin, P. Warren, and two anonymous reviewers provided valuable comments on drafts of this manuscript.

References cited

- Allen, J.M. & P.F. Nicoletto. 1997. Response of *Betta splendens* to computer animations of males with fins of different length. *Copeia* 1997: 195–199.
- Andersson, M. 1994. Sexual selection. Princeton University Press, Princeton. 599 pp.
- Baker, M.C., T.T. Tracy & L.E. Miyasato. 1996. Gargle vocalizations of black-capped chickadees: test of repertoire and video stimuli. *Anim. Behav.* 52: 1171–1175.
- Bolyard, K.J. & W.J. Rowland. 1996. Context-dependent response to red coloration in stickleback. *Anim. Behav.* 52: 923–927.
- Clark, D.L., J.M. Macedonia & G.G. Rosenthal. 1997. Testing video playback to lizards in the field. *Copeia* 1997: 421–423.
- Clark, D.L. & G.W. Uetz. 1990. Video image recognition by the jumping spider, *Maevia inclemens* (Araneae: Salticidae). *Anim. Behav.* 40: 884–890.
- Clark, D.L. & G.W. Uetz. 1992. Morph-independent mate selection in a dimorphic jumping spider: demonstration of movement bias in female choice using video-controlled courtship behavior. *Anim. Behav.* 43: 247–254.
- Clark, D.L. & G.W. Uetz. 1993. Signal efficacy and the evolution of male dimorphism in the jumping spider, *Maevia inclemens*. *Proc. Nat. Acad. Sci. USA* 90: 11954–11957.
- Endler, J. A. 1983. Natural and sexual selection on colour patterns in poeciliid fishes. *Env. Biol. Fish.* 9: 173–190.

- Endler, J. A. 1990. On the measurement and classification of animal colour patterns. *Biol. J. Linn. Soc.* 41: 315–352.
- Evans, C. S. & P. Marler. 1995. Language and animal communication: parallels and contrasts. pp. 341–382. *In*: H. Roitblat & J. Arcady-Meyer (ed.) *Comparative Approaches to Cognitive Science*, MIT Press, Cambridge.
- Fleishman, L.J., W.J. McClintock, R.B. Death, D.H. Brainard & J.A. Endler. 1998. Colour perception and the use of video playback experiments in animal behaviour. *Anim. Behav.* 56: 1035–1040.
- Hurlbert, S.H. 1984. Pseudoreplication and the design of ecological field experiments. *Ecological Monographs* 54: 187–211.
- Kodric-Brown, A. & P.F. Nicoletto. 1997. Repeatability of female choice in the guppy: response to live and videotaped males. *Anim. Behav.* 54: 369–376.
- Landmann, K., J. Parzefall & I. Schlupp. 1999. A sexual preference in the Amazon molly, *Poecilia formosa*. *Env. Biol. Fish.* 56: 327–333 (this issue).
- Loew, E.R. & W.N. McFarland. 1990. The underwater visual environment. pp. 1–43. *In*: R. Douglas & M. Djamgoz (ed.) *The Visual System of Fish*, Chapman and Hall, Cambridge.
- Long, K. D. & A. E. Houde. 1989. Orange spots as a visual cue for female mate choice in the guppy (*Poecilia reticulata*). *Ethology* 82: 316–324.
- Macedonia, J.M., C.S. Evans & J.B. Losos. 1994. Male *Anolis* lizards discriminate video-recorded conspecific and heterospecific displays. *Anim. Behav.* 47: 1220–1223.
- Macedonia, J.M. & J.A. Stamps. 1994. Species recognition in *Anolis grahami* (Sauria, Iguanidae): evidence from responses to video playbacks of conspecific and heterospecific displays. *Ethology* 98: 246–264.
- Martin, P. & P. Bateson. 1993. *Measuring behaviour: an introductory guide*, 2nd ed. Cambridge University Press, Cambridge. 222 pp.
- McClintock, W.J. & G.W. Uetz. 1996. Female choice and pre-existing bias: visual cues during courtship in two *Schizocosa* wolf spiders (Araneae: Lycosidae). *Anim. Behav.* 52: 167–181.
- McDonald, C.G., T.E. Reimchen & C.W. Hawryshyn. 1995. Nuptial colour loss and signal masking in *Gasterosteus*: an analysis using video imaging. *Behaviour* 132: 965–977.
- McGregor, P.K. 1992. Preface. pp. v–vii. *In*: P. McGregor (ed.) *Playback and Studies of Animal Communication*, Plenum Press, New York.
- McGregor, P.K., C.K. Catchpole, T. Dabelsteen, J.B. Falls, L. Fusani, H.C. Gerhardt, F. Gilbert, A.G. Horn, G.M. Klump, D.E. Kroodsma, M.M. Lambrechts, K.E. McComb, D.A. Nelson, I.M. Pepperberg, L. Ratcliffe, W.A. Searcy & D.M. Weary 1992. Design of playback experiments: the Thornbridge Hall NATO ARW consensus. pp. 1–9. *In*: P. McGregor (ed.) *Playback and Studies of Animal Communication*, Plenum Press, New York.
- McKinnon, J.S. 1995. Video mate preferences of female three-spined sticklebacks from populations with divergent male coloration. *Anim. Behav.* 50: 1645–1655.
- McKinnon, J.S. & J.D. McPhail. 1996. Male aggression and colour in divergent populations of the threespine stickleback – experiments with animations. *Can. J. Zool.* 74: 1727–1733.
- Pelkewijk, J.J.T. & N. Tinbergen. 1937. Eine reizbiologische Analyse einiger Verhaltensweisen von *Gasterosteus aculeatus* L. *Z. Tierpsychol.* 1: 193–204.
- Rosenthal, G.G. & C.S. Evans. 1998. Female preference for swords in *Xiphophorus helleri* reflects a bias for large apparent size. *Proc. Nat. Acad. Sci. USA* 95: 4431–4436.
- Rosenthal, G.G., C.S. Evans & W.L. Miller. 1996. Female preference for a dynamic trait in the green swordtail, *Xiphophorus helleri*. *Anim. Behav.* 51: 811–820.
- Rowland, W.J. 1995. Do female stickleback care about male courtship vigour – manipulation of display tempo using video playback. *Behaviour* 132: 951–961.
- Rowland, W.J., K.J. Bolyard, J.J. Jenkins & J. Fowler 1995a. Video playback experiments on stickleback mate choice: female motivation and attentiveness to male colour cues. *Anim. Behav.* 49: 1559–1567.
- Rowland, W.J., K.J. Bolyard & A.D. Halpern. 1995b. The dual effect of stickleback nuptial coloration on rivals: manipulation of a graded signal using video playback. *Anim. Behav.* 50: 267–272.
- Ryan, M.J. & A.S. Rand. 1995. Female responses to ancestral advertisement calls in túngara frogs. *Science* 269: 390–392.
- Sekuler, R. & R. Blake. 1994. *Perception*. McGraw-Hill, New York. 572 pp.
- Sigmund, W.R. 1983. Female preference for *Anolis carolinensis* males as a function of dewlap color and background coloration. *J. Herp.* 17: 137–143.
- Uetz, G.W., W.J. McClintock, D. Miller, E.I. Smith & K.K. Cook. 1996. Limb regeneration and subsequent asymmetry in a male secondary sexual character influences sexual selection in wolf spiders. *Behav. Ecol. Sociobiol.* 38: 253–257.