# Automated Scanning through the Hair Bundle State Space

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May 2, 2013

## 1 Summary

What follows is a brief description of an experimental system whereby mechanical stimuli are delivered to hair bundles. Using this system, an experimenter may clamp a hair bundle to a particular offset position and deliver mechanical stimuli. In addition, one may independently control offset force delivered to and the apparent stiffness of the coupled fiber-bundle system. Included are protocols for calculation of these control parameters, iteration methods, and calibration techniques. It is assumed in these notes that the basics of the displacement clamp circuit are known.

### 1.1 Pertinent Definitions

- **Control Parameter**: This is any quantitative parameter that will be set to particular values such that the hair bundle may be poised in a particular operating point within its state space. Here, our control parameters are **force** and **stiffness**.
- **Gain**: In a proportional-integral-derivative controller, this refers to the proportional gain ($G$) that controls the strength of negative feedback in a displacement clamp. (For more information on the modified displacement clamp, see additional notes.)
- **Offset**: This is synonymous with the command displacement, $X_C$, of the displacement clamp.
- **Stimulus Fiber:** Our stimulus fiber is a borosilicate glass electrode, pulled orthogonally to its long axis, and coupled to the kinociliary bulb for hair-bundle stimulation.
- **Pulse:** Here, a pulse refers to a steady-state force and stiffness at which the hair bundle is poised for a given period of time, such that **gain** and **offset** are <u>nonzero</u>. Each pulse lasts for $N_P$ seconds.

- **Baseline:** Between each pulse, the hair bundle may be returned to an operating point such that **gain** and **offset** are <u>zero</u>. Each baseline lasts for $N_B$ seconds.
- **Ramp:** Over a period of $N_R$ seconds, the hair bundle is slowly ramped from baseline to pulse.
- **Session:** A session refers to a collection of $M_P$ pulses and $M_B$ baselines over a period of $M_P N_P + M_B N_B + 2N_R$ seconds.
- **Alpha Calibration**: <u>Between</u> each session, the calibration parameter alpha ($\alpha$) is recalibrated via a series of displacements in a piezoelectric transducer coupled with a mirror that translates the shadow of the stimulus fiber over the photodiode without adjusting the position of the stimulus fiber. Here, **gain** and **offset** are <u>zero</u>.
- **Realtime Calibration**: During each baseline, the aforementioned piezoelectric transducer is delivered a sinusoidal signal in order to recalibrate the signal <u>during</u> a session.

# 2 Modified Displacement Clamp

In the case of a displacement clamp with a proportional-integral-derivative (PID) controller, an increase in the proportional gain ($G$) results in an increase int he magnitude of force delivered by the stimulus fiber onto the hair bundle. This increased force reduces the difference between the command displacement ($X_C$) and the hair bundle's position in time ($X$). If one were to consider an imaginary clamp with zero error between $X_C$ and $X$, the system would behave as if the compliance were also zero. However, this case does not follow reality, where PID feedback provides an opportunity to reduce such error. By extension, an increase in $G$ (and thus, feedback) should also reduce the apparent compliance of the coupled fiber-bundle system.

## 2.1 Force-Stiffness Relationship

First, consider the steady-state behavior of a hair bundle. The force exerted by the stimulus fiber ($F_{SF}$) is equal and opposite to the reaction force exerted by the hair bundle ($F_{HB}$).

$$F_{SF} = -F_{HB}$$

This equation can be expanded to include the hair bundle position ($X$), displacement of the stimulus fiber ($\Delta$), stiffness of the hair bundle ($\kappa_{HB}$), and stiffness of the stimulus fiber ($\kappa_{SF}$).

$$\kappa_{SF}(\Delta - X) = \kappa_{HB} X$$

$$\frac{\Delta - X}{X} = \frac{\kappa_{HB}}{\kappa_{SF}}$$

## 2.2 Displacement Clamp

The above equations define a relationship between the terms that will be used in calibration of the displacement clamp. In the clamp circuit, a photodiode acts as the displacement monitor, and the output voltage ($V_D$) is related to the hair bundle position by a coefficient, $\alpha$. The piezoelectric transducer controlling the position of the stimulus fiber receives some command voltage ($V_O$) and displaces the base of the stimulus fiber. This command voltage can be related to the proportional gain $G$ of the PID contorller and the error signal between $X_C$ and $X$.

$$V_D = \alpha X$$

$$\Delta = \beta V_O = \beta G(V_C - V_D) = \alpha\beta G(X_C - X)$$

## 2.3 Extraction of Control Parameters

An immediate observation is the existence of two components of force. The first is some offset force ($F_C$) that should not change as $X_C$ and $G$ remain constant. In order to hold force constant, as was done in the calibration of this clamp, one must compensate for changes in $G$ with changes in $X_C$. $F_C$ will be employed as one of the control parameters of the hair bundle's state space using this relation.

$$F_C = \alpha\beta G X_C \kappa_{SF}$$

One can further combine the relationships between these terms in order to predict the behavior of $X$ and $\Delta$ as various parameters are varied. Note that the term in the numerator corresponds to offset force, while the denominator corresponds to the total stiffness of the coupled fiber-bundle system. **This allows independent control of two parameters, force ($F_C$), and stiffness ($\kappa_{total}$).**

$$X = \frac{\alpha\beta G X_C \kappa_{SF}}{\kappa_{HB} + \kappa_{SF}(1+\alpha\beta G)} = \frac{F_C}{\kappa_{total}}$$

$$\Delta = \frac{\kappa_{HB} + \kappa_{SF}}{\kappa_{SF}} \frac{F_C}{\kappa_{total}}$$

Emerging from the equation for $X$ is an expanded Hooke's law of elasticity, where displacement and stiffness would have an inverse relationship with constant force. The same is true, though immediately less apparent, in $\Delta$. When the denominator reaches zero, such that $\kappa_{HB} = -\kappa_{SF}$, the system becomes unstable. Both $X$ and $\Delta$ are predicted to follow this inverse relationship between displacement and stiffness with contant offset force. These relationships have been employed in the calibration of the displacement clamp. What can be seen in the denominator is the presence of gain, whose value alters the apparent stiffness of the stimulus fiber.

$$\kappa_{SF,apparent} = \kappa_{SF}(1 + \alpha\beta G)$$

In summary, a displacement clamp would be predicted to provide both independent control of offset force and total stiffness of the system. This can be calibrated with the relationships above. Employing the clamp in an exploration of the two-dimensional hair bundle state space will be accomplished by engaging the relationships for $F_C$ and $\kappa_{SF,apparent}$, the control parameters of import.

# 3 Automated State Space Calculation

We now describe a method by which a protocol employed in both MATLAB and LabVIEW allows a set of input parameters (by the user) in force-stiffness space to be calculated and randomized, if desired, in gain-offset space.

## 3.1 MATLAB Implementation

A MATLAB function allows the user to input the following parameters:

- $F_{min}$, $F_{max}$: the range of forces in the proposed state space
- $\kappa_{min}$,$\kappa_{max}$: the range of stiffnesses in the proposed state space
- $N_F$,$N_\kappa$: the number of force and stiffness parameters, respectively (where the total state space will have dimensions $N_F \times N_\kappa$
- $Slope_{Xc}$, $Slope_G$ : the maximum slope of the ramp from baseline to pulse in $X_C$ and $G$, respectively
- $\alpha$, $\beta$: alpha and beta control parameters, previously described
- $\kappa_{SF}$: stiffness of the stimulus fiber

Using these parameters, the MATLAB script will calculate an array in both $F_C$ and $\kappa$, defined by the range of forces and stiffness above. At each operating point in the state space, gain and command displacement are calculated using the following relationships, where $i$ is the stiffness index and $j$ is the force index.

$$G_{i,j} = \frac{\kappa_i - \kappa_{SF}}{\alpha\beta\kappa_{SF}}$$

$$X_{C,i,j} = \frac{F_j}{\alpha\beta\kappa_{SF}G_{i,j}}$$

This allows a state space in gain and offset to be calculated for implementation in the displacement clamp. Additionally, the ramps between baselines and pulses are defined such that the ramp times are given by the following relationships. The longer ramp time is chosen for all ramps, such that the slope in $X_C$ and $G$ ramps never exceed their maximum values, and all ramps are of equal length.

$$Ramp_G = \frac{max(abs(G))}{Slope_G}$$

$$Ramp_{X_C} = \frac{max(abs(X_C))}{Slope_{Xc}}$$

This function outputs matrices in both gain and offset, and it outputs one ramp time that will be applied to all pulses. These values are fed into LabVIEW for implementation in the state space automation.

## 3.2 Limits in Gain

As mentioned before, there exists an inverse relationship between gain and offset, as shown below.

$$F_C = \alpha\beta G X_C \kappa_{SF}$$

When the absolute value of $G$ becomes small (or when $\kappa_{SF,apparent}$ approaches $\kappa_{SF}$), $X_C$ becomes very large for a given $F_C$. This becomes a problem in circuits with limitations in output voltage. If we define, the maximum voltage to be $V_{max}$, we can find cases where the output voltage may exceed $V_{max}$. This is found by finding $F_{max}$, previously defined by the user, and using the relationship $V_c = \alpha X_C$ to find indices where gain at values such that $V_C > V_{max}$.

$$G_{limit} = \frac{F_{max}}{\beta\kappa_{SF}V_{max}}$$

In this case, all indices where $G < G_{limit}$ are set to <u>zero</u> in both $G$ and $X_C$. This ensures that $V \leq V_{max}$.

## 3.3 State Space Exploration

Using a combination of MATLAB scripts and LabVIEW programs, the matrices from Section 3.1 can be reordered in one of the following fashions. The first step is to reshape the 2D matrices into 1D arrays. Then, the order of these arrays may be changed. Note that for all order, a given index referring to some combination of $G$ and $X_C$ is changed such that these two values remain associated with one another.

- **None:** The order of arrays in gain and offset remain unchanged.
- **Randomization:** A pseudo-random number generator is called to reorder the arrays. This allows for random sampling through the two-dimensional state space.
- **Snake:** A "snake" in either the force or stiffness direction is implemented. In these protocols, one parameter is held constant (for example, stiffness), while the other is increased (for example, force). When the maximum value is reached, the first parameter (e.g. stiffness) increases to the next index, and the other parameter (e.g. force) starts at its maximum value and decreases to its minimum. This back-and-forth scanning allows for a raster-type scan without jumping from one extreme value in a control parameter to another.
- **Spiral:** The two-dimensional matrices are sampled, starting at the center and spiraling out to the extreme values, in either a clockwise or counter-clockwise rotation. This provides the benefit of avoiding the extreme values of the state space (very high or very low gain) until the end of the experiment.

## 4 State Space Protocol

A reordered state space with gain and offset as parameters is implemented in LabVIEW. The user may define the following parameters:

- **Pulse Length:** The total time for each pulse is determined manually by the user.
- **Pulse Delay and Total Period:** In combination with the value above, this provides the total baselength time for each operating point.
- **Ramp Time:** Though this is automated in MATLAB, the user can define this value manually.
- **Realtime Alpha Calibration:** During each baseline, a sinusoidal stimulus may be sent to a second piezoelectric transducer that translates the shadow of the stimulus fiber over the photodiode during each baseline. The user can define the number of cycles and amplitude of this calibration. Knowing these parameters, one has a realtime measure relating voltage output from the photodiode and fiber position, which is subject to change throughout a given session.
- **Session Time:** The total session time is defined by the user. LabVIEW then divides the arrays of gain and offset into equal parts, such that the total time for each session ($M_P N_P + M_B N_B + 2N_R$ seconds) is less than or equal to the session time defined by the user. Each session is saved individually and the computer's working memory cleared to avoid memory capacity errors. An alpha calibration is performed between each session.

### 4.1 Interrupts

If the system goes unstable, the bundle is lost, or another error occurs, the user may manually interrupt a given session. Upon interrupting:

- $G$ and $X_C$ are set to zero.
- The computer's memory is cleared.
- A note is appended to the logfile.
- A dialog box for an alpha calibration is opened.
- The current session is appended to the end of the experiment, and the next session is called.

This allows the user to manually recalibrate the system if focus is lost or the hair bundle is no longer coupled to the stimulus fiber. It also allows the system to skip the current session if stability is lost and return to this session later in the experiment.

### 4.2 Additional Note on Timing

Though a given state space may take 5-10 minutes for acquisition as outlined above, a majority of the user's time is spent <u>between</u> sessions during manual readjustment of the system and the ensuing alpha calibration. It is thus in the best interest of the user to reduce the total number of sessions if time is a factor.

## 5 Additional Stimuli

During each pulse, the user may add an additional stimulus. These stimuli include steps, ramps, and trains or sweeps of sinusoids. Full control of these additional stimuli is implemented in LabVIEW.