Wavelet analysis of shuttle motility in P. Polycephalum plasmodium

T.I. Avsieivich, S.G. Proskurin
Biomedical Engineering,
Tambov State Technical University,
Russia
Purpose of research

Slime mold *Physarum polycephalum* is the classic model unicellular organism for amoeboid motility studying. Actin-myosin contractile system generates periodic contractile activity causing shuttle endoplasmic motility in *Physarum* strands. However, the nature and number of oscillators that control the contractile activity of plasmodium remains unknown.

In the present study velocity time dependencies of alternating shuttle streaming of endoplasm in slime mold plasmodium *P. polycephalum* treated by the cellular respiration inhibitors have been investigated by laser Doppler spectroscopy with sign-sensitive mode.
Materials and methods

*P. Polycephalum*

- Slime mold
- Network of cylindrical bands 100-500 µm in diameter and length up to 2 cm
- Model organism for many studies involving amoeboid movement and cell motility

---

**potassium cyanide (KCN):**
- Cytochrome pathway inhibitor

**salicylhydroxamic acid (SHAM):**
- Alternative oxidase inhibitor

1) Inhibition of aerobic ATP synthesis
2) Cessation of breathing
3) Cessation the movement of protoplasm

---

1) Strand of plasmodium was cut out of plasmodium and placed in the buffer solution pH=7.2, velocity time dependence \( V(t) \) was registered;

2) One half of strand was treated by KCN and SHAM (5 and 7 µM respectively) which leads to a cessation the endoplasm streaming;

3) Further inhibitors were removed and strand was placed in buffer. Resuming endoplasm motility through 10 min was recorded.
Velocity time dependencies registration using laser Doppler spectroscopy

Cellular respiration inhibitors
- potassium cyanide (KCN)
- salicylhydroxamic acid (SHAM)

Laser Doppler spectroscopy

He-Ne Laser

λ=638.2 nm, P=1-15 mW

Isolated strand of plasmodium (1 cm) with a stream of particles inside

Results of measurements - Velocity time dependence (20 min)
Sign-sensitive registration principle
Short-time Fourier Transform - STFT of time dependencies (length 600 s)

Frequency spectra of the time dependencies of endoplasmic streaming obtained by STFT in case of velocity module (circles) registration and in an alternating mode registration (squares).

Sign-sensitive registration SNR > 7
Velocity module registration SNR ≈ 1

Two distinct peaks are clearly seen in case of sign-sensitive registration only.
Simulated (red solid line) time dependencies based on the spectral characteristics: frequencies, amplitudes and phases, obtained from STFT.

Velocity module registration $r \approx 0.46$

Sign-sensitive registration $r \approx 0.95$
Velocity time dependence
From LDV measurements

Short-time Fourier transform (STFT) analysis
window (100 sec)
Time resolution is considerably limited

Wavelet transform
gives better visualization of the frequency changes in time
One half of strand treated by KCN and SHAM

10 min after treatment by KCN and SHAM

First oscillator ($\omega_1$) activity suppressing

Power decreasing for both oscillators
Despite the frequencies are differs in first and second signals, the ratio $\omega_2/\omega_1 = 1.96\pm2\%$ in each of them remains constant.
Analytical calculation of the self-oscillation frequencies of harmonics:

\[
\omega = \sqrt{\frac{E(0.5k_1 + k_2 + k_4)}{\eta + \frac{16\mu}{rh} \left(\frac{l}{\pi n}\right)^2} + k_4(0.5k_1 + k_2)}
\]

where \( E \) - Young's modulus of ectoplasm, \( k_1, k_2, k_4 \) - rate constants, \( n \) - harmonic number, \( l, r \) – length and radius of the strand, \( \mu \) – endoplasm viscosity [2].

**Theoretical frequencies:**
\[
\omega_1 = 0.0191 \text{ Hz}, \\
\omega_2 = 0.0123 \text{ Hz}
\]

\[
\frac{\omega_1}{\omega_2} = 1.6
\]

**Experimental frequencies:**
\[
\frac{\omega_2}{\omega_1} = 1.96
\]
Model of initial dependencies

The corresponding simulated (blue solid line) time dependencies based on the spectral characteristics: frequencies, amplitudes and phases, obtained from STFT, implemented in Matlab. Significant correlation \((r \approx 0.95)\) between experimental and simulated data is observed.

Normal conditions, buffer solution

One half treated by KCN and SHAM
10 min after treatment by KCN and SHAM - motion restoring

Harmonic oscillator with $\omega_1$ is still active after treatment – no movement is registered. Activity recovering of the second oscillator with $\omega_2$ means that endoplasm is moving again.
Conclusion

**STFT** reveals two distinct harmonic signals. Despite the frequencies are differs in first and second signals, the ratio $\omega_2/\omega_1=1.97\pm2\%$ in each of them remains constant. The registered oscillators are energetically interconnected – the sum power of oscillators remains constant, only their contributions are changed.

Since method did not provide good enough time resolution, wavelet transform were applied. **Wavelet transform** allowed to track frequency changes in time with high accuracy and clearly identify the number of oscillators. Future work will be aimed to data processing algorithm elaboration for Doppler spectroscopy.
REFERENCES


