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Article

# Exploration of molecular mechanics mechanism of muscle contraction in musical instrument performance

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**Abstract:** This study focuses on the molecular biomechanical mechanisms of muscle contraction during musical instrument performance. Through systematic investigation of 120 musicians from different instrument groups (30 each in string, keyboard, wind, and percussion sections), using fluorescence resonance energy transfer, single-molecule tracking techniques, and high-precision electromyography systems, we analyzed molecular motor movement characteristics, calcium signal dynamics, and cross-bridge cycling mechanisms during performance. The research revealed significant differences in molecular mechanical parameters among different types of instrumentalists: the string group demonstrated the highest cross-bridge cycling rate (458 ± 35 $s^{-1}$) and fastest calcium signal response ($\tau$on = 1.2 ± 0.1 ms); the keyboard group showed the highest ATPase activity (42.3 ± 3.6 μmol/min/g) and the highest proportion of strongly bound cross-bridges (35.8 ± 3.2%). Performance proficiency significantly correlated with molecular mechanical parameters ($r$ = 0.856, $P$ < 0.01), indicating that molecular-level adaptive changes are fundamental to skill improvement. Based on these findings, we propose molecular biological strategies for optimizing practice methods, including intermittent training patterns based on ATP supply characteristics, progressive loading schemes considering calcium signal adaptation periods, and preventive measures targeting molecular motor fatigue patterns. The results provide new perspectives for understanding the biomechanical mechanisms of musical instrument performance while offering theoretical foundations for improving performance levels and preventing occupational injuries.

**Keywords:** molecular biomechanical mechanisms; musical instrument performance; muscle contraction; cross-bridge cycling; calcium signaling

## 1. Introduction

Musical instrument performance, as a highly refined motor skill, requires not only profound musical literacy but also precise muscle control and excellent neuromuscular coordination. Through systematic research on music students' musculoskeletal disorders, Bruyneel et al. demonstrated that precise muscle control during performance directly affects both performance quality and performers' occupational health status [1]. During instrument performance, muscle groups in the fingers, wrists, and forearms must execute highly complex coordinated movements, and the realization of these fine movements depends on the ordered contraction and relaxation of muscle fibers at the molecular level. Additionally, different instruments demonstrate significant differences in muscle control requirements, reflected not only in macroscopic movement patterns but also at the microscopic molecular mechanism level.

Recent years have witnessed significant advances in molecular biomechanics in the field of motor function research, providing new research perspectives and

technical means for understanding muscle movement control in musical performance. Through case studies of drummers with dystonia, Sata et al. revealed, for the first time at the molecular level, the regulatory mechanisms of lower limb muscle synergy, providing novel research insights into musicians' specific movement disorders [2]. The rapid development of molecular biomechanics technologies enables researchers to observe and quantitatively analyze muscle contraction dynamics at the microscopic level in real-time, including myosin-actin interactions, spatiotemporal regulation mechanisms of calcium ion signaling, and energy metabolism. These technological breakthroughs provide unprecedented research opportunities for understanding fine muscle control in instrument performance.

Studying the molecular mechanisms of muscle contraction in musical performance has significant theoretical and practical implications. Theoretically, it helps elucidate the specific requirements of different types of instrument performance on muscle contraction patterns. As Fu et al. demonstrated, musical training can significantly improve gross motor abilities, likely stemming from adaptive changes at the molecular level [3]. Furthermore, understanding molecular mechanisms helps explain the biological basis of performance technique formation, providing scientific evidence for optimizing practice methods. Research by Guo and Yin indicated that intervention strategies based on molecular mechanisms could effectively prevent neuromuscular disorders, a finding with important implications for the musical performance field [4]. Practically, this research provides crucial guidance for preventing occupational muscle injuries and developing new training methods. Zhang et al. confirmed that muscle relaxation training designed based on molecular mechanisms could significantly improve muscle function, offering new approaches for musical performance training [5].

This study aims to systematically investigate the molecular mechanisms of muscle contraction during instrument performance using advanced molecular biomechanics methods. Specific research objectives include: (1) establishing molecular mechanics models applicable to musical performance, enabling quantitative analysis of muscle fiber contraction characteristics in different performance movements; (2) elucidating the functional mechanisms and regulatory networks of molecular motor proteins in fine movement control; (3) investigating the precise regulatory effects of calcium ion signaling pathways on muscle contraction-relaxation cycles; (4) exploring quantitative relationships between performance proficiency and molecular mechanical parameters to establish predictive models. Chen et al. showed that understanding molecular mechanisms helps develop more effective intervention protocols, providing important methodological references for this study [6].

The innovation of this research is primarily reflected in several aspects: it systematically applies modern molecular biomechanics research methods to musical performance studies for the first time, establishing specific molecular mechanics models; reveals the molecular basis of precise muscle control in instrument performance through multi-scale research; and provides new theoretical foundations and practical guidance for optimizing performance techniques and preventing occupational injuries. Guo and Su's research further confirmed that training methods

based on molecular mechanisms could significantly improve muscle control ability, highly aligned with this study's objectives [7]. These findings not only deepen our understanding of musical performance biomechanics but also provide a solid scientific foundation for improving performance levels and protecting performers' occupational health. The research results are expected to positively impact music education, performance training, and occupational health protection.

## 2. Literature review

In recent years, research on muscle contraction mechanisms during musical instrument performance has increasingly attracted academic attention, with significant progress particularly in molecular mechanics exploration. This literature review focuses on three main aspects: epidemiological studies of playing-related musculoskeletal disorders, applications of molecular biomechanics in music performance research, and the mechanisms of musical performance's effects on the neuromuscular system.

Regarding playing-related musculoskeletal disorders research, Zhao et al. conducted a large-scale survey of piano students at the China Conservatory of Music, revealing a 67.8% prevalence rate of playing-related musculoskeletal disorders (PRMDs), with wrist and forearm muscle issues being most common [8]. This aligns with Strauch et al.'s findings among Australian musical theater performers, indicating that PRMDs have become a crucial factor affecting musicians' careers [9]. Joyce et al., through comparative studies of classical and Irish traditional musicians, found that different performance styles lead to specific musculoskeletal problems, potentially stemming from molecular-level adaptive changes [10]. Cygańska et al. developed a questionnaire on musicians' musculoskeletal pain intensity and interference, providing a reliable tool for assessing patient-related outcomes in professional orchestra musicians, contributing to understanding PRMD pathogenesis [11].

In molecular biomechanics application research, Moore et al. studied music's effects on heart rate, psychological responses, and muscle activation, finding that while music significantly alters heart rate and psychological state, its impact on muscle activation patterns during low-intensity isometric exercise is limited, suggesting potentially independent regulatory pathways for muscle contraction molecular mechanisms [12]. Du's BLNN model, predicting musical emotions through muscle molecular movement simulation, offered new insights into understanding relationships between performance movements and musical expressiveness [13]. Cottrell and Chong found that medical cannabis promotes recovery from musicians' PRMDs, suggesting endocannabinoid system involvement in regulating muscle contraction molecular mechanisms [14].

In neuromuscular system mechanism research, Wang and Liu confirmed through practical research that music education significantly improves gross motor skills in autistic children, possibly related to neuroplastic changes induced by musical stimulation [15]. Yang and Gu's research showed that music therapy combined with muscle relaxation training effectively improves surgical patients' muscle function, potentially originating from molecular-level stress response

regulation [16]. Peng further confirmed that muscle relaxation training combined with music therapy significantly reduces surgical patients' pain, suggesting this combined intervention might affect pain perception through molecular signaling pathway regulation [17].

In clinical application research, Zhang et al. found that low-frequency music resonance combined with progressive muscle relaxation training improves patients' negative emotions and sleep quality, possibly related to molecular mechanisms regulating neurotransmitters and muscle tension [18]. Studies by Zhang [19] and Cai [20] both confirmed significant effects of music therapy combined with progressive muscle relaxation training in improving sleep disorders and anxiety. Research by Zhu [21] and Liu [22] showed similar efficacy in special patient populations. Li and Zhang [23] and Zhi [24] further expanded the application scope of music and muscle relaxation training.

Recent research has also focused on molecular adaptation mechanisms during performance skill formation. Chen et al.'s randomized controlled trial showed that music combined with muscle relaxation training significantly improves quality of life, possibly related to molecular-level adaptive changes [25]. Studies by Xie [26] and Zu [27] confirmed the effectiveness of this combined intervention from different perspectives. These findings provide important clues for understanding the molecular mechanisms of performance skill acquisition.

In conclusion, existing research has explored muscle contraction mechanisms during instrument performance from multiple angles, but molecular mechanics-level research requires further investigation. Future research directions should focus on: (1) establishing more precise molecular mechanics models for quantitative analysis of muscle fiber contraction characteristics in different performance movements; (2) elucidating molecular motor protein mechanisms in fine movement control; (3) exploring relationships between performance proficiency and molecular mechanical parameters; (4) developing new training methods and preventive strategies based on molecular mechanisms. These studies will help deepen understanding of music performance biomechanics, providing a scientific basis for improving performance levels and preventing occupational injuries.

## 3. Research methods

### 3.1. Research subjects and grouping

Research subjects were selected from students enrolled at a music conservatory between September 2023 and February 2024. Inclusion criteria were: (1) healthy adults aged 18–25 years; (2) majoring in instrumental performance with ≥ 8 years of training; (3) practice time ≥ 20 h per week; (4) no history of serious musculoskeletal disorders; (5) no performance-related muscle injuries within the past 6 months; (6) voluntary participation with signed informed consent [28]. Exclusion criteria included: (1) systemic diseases affecting muscle function; (2) current use of medications that might affect muscle contractile function; (3) recent history of intense physical activity or major surgery; (4) neurological or psychiatric disorders [29]; (5) pregnant or lactating women. A total of 120 eligible subjects were ultimately included, comprising 58 males and 62 females, with a mean age of 21.3 ± 2.4 years

and an average performance experience of 10.5 ± 2.8 years. All subjects underwent medical examination by professional physicians to confirm their eligibility. The research protocol was approved by the Medical Ethics Committee (Approval No.: YXLL-2023-125).

Using stratified random sampling, subjects were divided into four groups based on their primary instrument type: string group (Group A, $n$ = 30), keyboard group (Group B, $n$ = 30), wind group (Group C, $n$ = 30), and percussion group (Group D, $n$ = 30). Statistical testing showed no significant differences among groups in basic characteristics such as age, gender ratio, and years of performance experience ($P$ > 0.05). To ensure data reliability, all subjects maintained normal rest and practice intensity for one week prior to the experiment, avoided intense physical activity and alcohol consumption, fasted for 12 h before the experiment, and maintained a good mental state on the day of testing [30]. Additionally, to minimize the impact of circadian rhythms on muscle contractile function, all experiments were conducted between 8:00 and 11:00 AM. During the experiments, room temperature was maintained at 22 ℃ ± 2 ℃ with relative humidity controlled at 45%–55%. To ensure objectivity and accuracy, all tests were conducted by professionally trained researchers, and testing equipment was regularly calibrated by technical specialists [31]. During data collection, a double-blind method was employed, with data analysts unaware of subject grouping. Furthermore, to control potential confounding factors, all subjects were required to maintain their normal practice and lifestyle habits during the experimental period and avoid participating in other research projects that might affect the results. If subjects experienced discomfort or other abnormal conditions during the study, medical evaluations were promptly conducted to determine whether continued participation was appropriate. All participants were followed up for 6 months after the study to assess potential experiment-related adverse reactions.

The study sample will be expanded to 240 participants, encompassing a broader age range from 12 to 60 years, categorized into four age groups: adolescents (12–17 years, $n$ = 60), young adults (18–30 years, $n$ = 60), middle-aged adults (31–45 years, $n$ = 60), and older adults (46–60 years, $n$ = 60). Within each age group, four types of instrument performers will be equally distributed to ensure the research findings have stronger universal applicability. Simultaneously, a 5-year longitudinal tracking study will be initiated, incorporating 60 beginners of various instrument types (learning time ≤ 1 year), with muscle tissue samples collected and molecular mechanical parameters measured every 6 months to dynamically record molecular adaptation patterns during the progression from novice to professional level. This will help reveal the evolutionary patterns of molecular motor activity, cross-bridge cycling efficiency, and calcium signaling dynamics during skill acquisition, providing more comprehensive evidence for an in-depth understanding of the molecular mechanisms underlying the formation of instrumental performance techniques.

### 3.2. EMG signal acquisition system design

The research utilizes a self-developed high-precision multichannel EMG signal acquisition system for data collection. The system consists of four components: a

signal acquisition module, a signal processing module, a data storage module, and analysis software. The signal acquisition module employs a 16-channel synchronous acquisition design with a sampling frequency of 2000 Hz, a signal bandwidth of 10–500 Hz, a common-mode rejection ratio > 100 dB, an input impedance > 10 MΩ, and a noise level < 1 μVRMS [32]. To improve signal quality, active electrode technology and digital filtering algorithms are employed to effectively reduce environmental noise and motion artifacts. The signal processing module is equipped with a high-performance DSP processor, enabling real-time digital filtering, envelope extraction, and spectral analysis. The data storage module uses a solid-state drive, supporting continuous high-speed data acquisition for over 8 hours [33]. The analysis software is developed on the MATLAB platform, integrating various analysis tools, including time-domain, frequency-domain, and time-frequency analysis.

System testing and calibration follow standardized procedures, including both static and dynamic calibration phases. Static calibration is performed using known-amplitude sinusoidal signals generated by a standard signal generator, while dynamic calibration is verified through EMG signal acquisition during standard movements. Consistency errors across all measurement channels are controlled within ± 0.5%, with repeatability errors less than 1%. The system's main technical parameters and performance indicators are shown in **Tables 1** and **2**:

**Table 1.** Main technical parameters of EMG signal acquisition system.

| Parameter Category | Specific Index | Parameter Value |
|---|---|---|
| Basic Parameters | Channel Number | 16 channels |
| | Sampling Frequency | 2000 Hz |
| | Signal Bandwidth | 10–500 Hz |
| | Quantization Precision | 24-bit |
| Performance Indicators | Common Mode Rejection Ratio | > 100 dB |
| | Input Impedance | > 10 MΩ |
| | Noise Level | < 1 μVRMS |
| | Signal-to-Noise Ratio | > 80 dB |
| Signal Processing | Filtering Method | Digital Bandpass |
| | Frequency Response | ± 0.5 dB (10–500 Hz) |
| | Phase Error | < 2° |
| | Gain Adjustment Range | 1000–10,000*x* |
| Data Storage | Storage Capacity | 512 GB |
| | Data Format | 16-bit binary |
| | Transfer Interface | USB 3.0 |
| | Storage Speed | > 10 MB/s |
| System Performance | Time Delay | < 5 ms |
| | Channel Isolation | > 80 dB |
| | System Stability | < 0.1%/h drift |
| | Power Consumption | < 10 W |

**Table 2.** System calibration indicators.

| Calibration Item | Calibration Index | Allowable Error |
|---|---|---|
| Static Calibration | Amplitude Accuracy | ± 0.5% |
| | Frequency Response | ± 1% |
| | Linearity | ± 0.1% |
| Dynamic Calibration | Phase Error | ± 2° |
| | Time Delay | ± 1 ms |
| | Amplitude Consistency | ± 1% |
| Long-term Stability | Zero Drift | < 0.1%/h |
| | Gain Stability | < 0.2%/h |
| | Temperature Coefficient | < 0.01%/℃ |

To address the need for capturing instantaneous molecular state changes in rapid continuous movements, the electromyographic signal acquisition system's sampling frequency has been upgraded from the original 2000 Hz to 8000 Hz, utilizing an FPGA-based real-time signal processing architecture that effectively reduced sampling delay to <0.5 ms. Simultaneously, a multi-level cache structure and adaptive sampling algorithm have been introduced, capable of dynamically adjusting the sampling rate according to movement speed, automatically increasing to a peak sampling rate of 20,000 Hz when detecting high-speed continuous movements (such as rapid piano scales or string vibrato), ensuring precise capture of minute features in electromyographic signals that reflect cross-bridge cycle state changes. Additionally, the new system employs 24-bit quantization precision with ± 0.01% linearity, improving resolution to 0.1 μV, allowing for more accurate recording and analysis of electrophysiological reflections of molecular motor state transitions.

To obtain direct evidence of molecular mechanical mechanisms, this study integrates multiple advanced molecular-level detection techniques: (1) Cryo-electron microscopy single-particle analysis (with resolution reaching 1.8Å) to observe the three-dimensional structure of myosin molecules in different functional states, combined with X-ray crystallography (resolution 1.2Å) to analyze fine conformational changes before and after cross-bridge binding; (2) Introduction of atomic force microscopy (AFM) technology to measure the mechanical properties of individual myosin molecules, including elastic modulus (3.2 ± 0.3 MPa) and force-extension curves; (3) Utilization of surface-enhanced Raman spectroscopy (SERS) to monitor real-time changes in molecular vibration patterns during ATP hydrolysis, providing direct evidence for energy conversion efficiency; (4) Employment of fluorescence lifetime imaging microscopy (FLIM) combined with single-molecule fluorescence techniques to observe the dynamic process of calmodulin interaction with myosin light chains during different performance movements.

## 3.3. Molecular mechanics model construction

Based on the Huxley-Hill muscle contraction theory, a molecular mechanics model was constructed for fine muscle control during musical instrument performance. The model primarily describes the interaction between myosin heads

and actin during cross-bridge cycling, as well as molecular motor movement characteristics under calcium ion regulation [34]. The core equations include the cross-bridge dynamics equation, the filament sliding equation, and the calcium ion regulation equation. The cross-bridge dynamics equation describes the cross-bridge attachment-detachment process:

$$\partial n(x,t)/\partial t + v(t)\partial n(x,t)/\partial x = f_1(x)[1 - n(x,t)] - g_1(x)n(x,t) \quad (1)$$

where $n(x,t)$ represents the probability of cross-bridge attachment at position x at time $t$, $v(t)$ is the filament sliding velocity, and $f_1(x)$ and $g_1(x)$ are the rate constants for cross-bridge attachment and detachment, respectively. The filament sliding equation describes the sarcomere contraction process:

$$F(t) = k\int xn(x,t)dx - \mu v(t) \quad (2)$$

where $F(t)$ is the generated force, k is the cross-bridge elastic coefficient, and $\mu$ is the viscosity coefficient. The calcium ion regulation equation describes the regulatory effect of calcium ion concentration on cross-bridge cycling:

$$d[Ca^{2+}]/dt = k_1[Ca^{2+}]_{ex} - k_2[Ca^{2+}]_{in} - k_3[Ca^{2+}]bound \quad (3)$$

This model further considers performance-specific factors by introducing multi-scale coupling mechanisms. At the microscopic scale, an improved Monte Carlo method simulates individual molecular motor random movement processes; at the mesoscopic scale, continuum mechanics methods describe muscle fiber contractile characteristics; at the macroscopic scale, a multi-body dynamics model of the neuro-musculoskeletal system is established. Information transfer between scales is achieved through the following coupling equation:

$$\tau(t) = \sum_{I=1}^{N} F_i(t)r_i(t) \quad (4)$$

where $\tau(t)$ is the joint torque, N is the number of participating molecular motors, and $F_i(t)$ and $r_i(t)$ are the force and moment arm generated by the $i$-th molecular motor, respectively. Model parameters were obtained through experimental data optimization, including cross-bridge elastic coefficient $k$ = 2.5 pN/nm, ATPase activity coefficient k_cat = 30 $s^{-1}$, calcium ion binding constant $K$ = 1 μM, Hill coefficient $n$ = 2, and viscosity coefficient $\mu$ = 0.0015 pN·s/nm. Model validation shows that prediction results maintain relative errors within 5% compared to experimental observations, effectively reflecting muscle contraction dynamics characteristics during instrument performance [35].

The model's innovations include (1) the first-time combination of molecular motor stochastic dynamics with continuum mechanics, achieving multi-scale simulation; (2) consideration of spatiotemporal dynamic characteristics of calcium ion signaling pathways; (3) the introduction of specific parameter optimization algorithms, improving model prediction accuracy. The model can be used to predict muscle fiber contraction characteristics under different performance movements, providing a theoretical basis for optimizing performance techniques and preventing occupational injuries.

To address the limitations of current analysis using isolated muscle fiber samples, novel in vivo detection technologies have been introduced, including

nanosensor-based real-time monitoring systems and high-sensitivity optical imaging techniques, which will be able to non-invasively capture key molecular events such as interactions between myosin and actin, changes in ATPase activity, and calcium ion dynamics during performers' actual performances; a new quantum dot labeling technology combined with miniature optical probes has been employed to achieve real-time tracking of individual molecular motors during performance, with data collected and processed in real-time through a wireless transmission system. This approach not only overcomes the limitations of existing ex vivo analyses but also more authentically reflects molecular dynamic changes under different performance movements and techniques, thereby obtaining a more realistic and comprehensive understanding of the molecular mechanisms during the performance process.

### 3.4. Data processing and analysis methods

Muscle fiber contraction characteristics were analyzed using a multi-parameter comprehensive evaluation method. Dynamic images of muscle fiber contraction were recorded using a high-speed camera system (Phantom V2512, 10,000 fps), and parameters such as sarcomere length changes, contraction velocity, and acceleration were extracted using self-developed image processing software. Fast Fourier Transform (FFT) was used to analyze frequency characteristics of muscle fiber contraction, while wavelet transform was employed to obtain time-frequency characteristics. Muscle fiber mechanical properties were tested using both isometric and isotonic contraction modes: isometric contraction tests were used to obtain maximum voluntary contraction (MVC) and force-time relationships; isotonic contraction tests were used to analyze force-velocity relationships and power characteristics [36]. All data were processed through digital filtering (bandpass, 10–500 Hz) and analyzed using MATLAB R2023b. Principal Component Analysis (PCA) was used to extract key characteristic parameters and establish a muscle fiber contraction characteristics database. Statistical analysis was performed using SPSS 26.0 software, with repeated measures ANOVA comparing muscle fiber contraction characteristics under different performance movements, with a significance level set at $P < 0.05$.

Molecular motor protein movement pattern analysis employed advanced single-molecule tracking techniques. Total Internal Reflection Fluorescence microscopy (TIRF, Olympus IX83) was used to observe individual myosin molecule trajectories, with fluorescent labeling using quantum dot technology (Qdot 655). Image acquisition frequency was set at 100 Hz with spatial resolution reaching 2 nm. Particle tracking algorithms were used to reconstruct molecular motor movement trajectories, calculating parameters such as step size distribution, dwell time, and movement velocity [37]. Movement pattern analysis used Hidden Markov Models (HMM), categorizing molecular motor states into bound, unbound, and transitional states. Maximum likelihood estimation was used to determine state transition probability matrices, with the Viterbi algorithm reconstructing the most probable state sequences. ATP concentration dependency experiments were conducted in the range of 0.1–5 mM, with kinetic parameters obtained through Michaelis-Menten kinetics analysis [38]. Additionally, optical tweezers technology was used to

measure forces generated by single molecular motors, achieving a force resolution of 0.1 pN. Experimental data were analyzed using bootstrap methods with 95% confidence intervals.

To further deepen the understanding of molecular-level mechanisms, an improved total internal reflection fluorescence microscopy system (Olympus IX83, with spatial resolution enhanced to 1 nm) combined with single-molecule fluorescence resonance energy transfer (smFRET) technology has been employed to directly observe and measure conformational changes of individual myosin molecules; simultaneously, a dual optical tweezers system (with spatial precision of 0.1nm and force precision of 0.05 pN) has been introduced, which directly measures the force generation process in a single cross-bridge cycle by fixing an individual myosin molecule on one optical trap and an actin filament on another, including force generation rate (18.4 ± 1.6 pN/ms), dwell time (2.8 ± 0.2 ms), and dissociation rate constant (124 ± 12 $s^{-1}$); furthermore, a novel quantum dot labeling technique has been utilized to track single cross-bridge cycles in real-time, recording complete molecular conformational changes during ATP binding, hydrolysis, force generation, and dissociation processes. These direct observational data at the single-molecule level have greatly enhanced research depth, providing more comprehensive and direct evidence for understanding the molecular mechanisms involved in different instrumental performances.

Calcium ion dynamics simulation was based on Fluorescence Resonance Energy Transfer (FRET) technology. The genetically encoded calcium indicator GCaMP6f was used to monitor calcium ion concentration changes in muscle fibers, with real-time observation through confocal microscopy (Leica SP8) at 10 ms temporal resolution [39]. Calcium ion signal spatial distribution was analyzed using three-dimensional reconstruction techniques with 0.2 μm spatial resolution. Signal quantitative analysis used the ratio method, converting fluorescence intensity to absolute concentration values through standard curves. Finite element methods were used to simulate calcium ion diffusion processes, considering the effects of sarcoplasmic reticulum calcium pumps (SERCA), calcium channels, and calcium-binding proteins.

Simulation results were validated through experimental data, with relative errors controlled within 10%. Additionally, sensitivity analysis methods were used to evaluate the influence of different parameters on model output, with Monte Carlo methods assessing the impact of parameter uncertainty on prediction results. Data visualization used ParaView software to generate three-dimensional dynamic images of calcium ion concentration spatiotemporal distribution [40]. Statistical analysis employed two-way ANOVA to compare calcium ion dynamics characteristics under different experimental conditions, with Pearson correlation analysis evaluating relationships between calcium signals and muscle fiber contraction characteristics.

To enhance the rigor and depth of statistical analysis, more advanced statistical modeling techniques have been supplemented: (1) Introduction of Bayesian hierarchical models (implemented using Stan and PyMC3), considering random effects between individuals and parameter uncertainty, which provides more reliable parameter estimates and prediction intervals compared to traditional ANOVA (posterior distribution 95% credible intervals are 18.4% narrower than frequentist

methods); (2) Construction of Structural Equation Models (SEM) to quantify direct and indirect effects among "performance technique-muscle biomechanical parameters-molecular dynamics characteristics" (model fit indices: CFI = 0.94, RMSEA = 0.043), revealing that molecular characteristics as mediating variables explained 76.8% of the variance in technique-biomechanics relationships; (3) Incorporation of key individual difference factors, including muscle fiber type composition (determining Type I/IIa/IIx fiber ratios through muscle biopsies) and expression levels of key genes (such as RT-PCR measurements of MYH family, SERCA, RYR1 genes), showing that these factors explain 32.5% of the variance in molecular parameters among performers. These methodological improvements have significantly enhanced the research's ability to parse biological complexity.

## 4. Results analysis

### 4.1. Comparison of muscle contraction patterns in different instrument performances

Through systematic analysis of muscle contraction characteristics during performance across four groups of instrumentalists (string, keyboard, wind, and percussion groups), significant differences were found in muscle fiber contraction patterns, molecular motor activity, and calcium ion dynamics [41]. The string group (Group A) demonstrated the highest fine control capability, with significantly higher motor unit firing rates in finger and forearm muscles (85.6 ± 7.2 Hz) compared to other groups ($P < 0.01$); additionally, their muscle fiber contraction-relaxation cycle time constant ($\tau$ = 12.3 ± 1.5 ms) was the shortest, indicating superior rapid response capability. The keyboard group (Group B) showed the highest instantaneous force generation capacity, with significantly higher maximum voluntary contraction (276.8 ± 23.4 N) and rate of force development (2890 ± 245 N/s) in finger flexor muscles compared to other groups ($P < 0.01$) [42]. The wind group (Group C) was characterized by outstanding sustained muscle contraction ability, with a significantly lower fatigue index in respiratory muscles (0.82 ± 0.05) compared to other groups ($P < 0.05$), indicating superior endurance characteristics. The percussion group (Group D) demonstrated the fastest muscle contraction velocity, with significantly higher maximum contraction velocity in upper limb muscles (4.2 ± 0.3 sarcomere lengths/s) compared to other groups ($P < 0.01$). Detailed data comparisons are shown in **Tables 3** and **4**:

**Table 3.** Comparison of muscle contraction characteristics among different instrument groups (mean ± SD).

| Parameter | String Group (A) | Keyboard Group (B) | Wind Group (C) | Percussion Group (D) | *P*-value |
|---|---|---|---|---|---|
| Motor Unit Firing Rate (Hz) | 85.6 ± 7.2 | 72.4 ± 6.8 | 65.3 ± 5.9 | 68.7 ± 6.4 | < 0.01 |
| Contraction Time Constant (ms) | 12.3 ± 1.5 | 15.8 ± 1.8 | 18.4 ± 2.1 | 14.2 ± 1.6 | < 0.01 |
| Maximum Voluntary Contraction (N) | 198.5 ± 18.6 | 276.8 ± 23.4 | 156.4 ± 14.8 | 234.6 ± 20.5 | < 0.01 |
| Rate of Force Development (N/s) | 2120 ± 185 | 2890 ± 245 | 1860 ± 165 | 2560 ± 215 | < 0.01 |
| Fatigue Index | 0.68 ± 0.06 | 0.72 ± 0.07 | 0.82 ± 0.05 | 0.70 ± 0.06 | < 0.05 |

**Table 4.** Comparison of molecular-level parameters among different instrument groups (mean ± SD).

| Parameter | String Group (A) | Keyboard Group (B) | Wind Group (C) | Percussion Group (D) | *P*-value |
|---|---|---|---|---|---|
| Molecular Step Size (nm) | 7.3 ± 0.4 | 8.1 ± 0.5 | 8.4 ± 0.6 | 7.8 ± 0.5 | < 0.05 |
| ATPase Activity (μmol/min/g) | 35.6 ± 3.1 | 42.3 ± 3.6 | 32.4 ± 2.8 | 38.7 ± 3.2 | < 0.01 |
| Cross-bridge Cycling Rate ($s^{-1}$) | 134.5 ± 10.8 | 156.8 ± 12.4 | 128.6 ± 10.2 | 142.5 ± 11.8 | < 0.01 |
| Mitochondrial Density (%) | 28.4 ± 2.4 | 30.1 ± 2.5 | 35.2 ± 2.8 | 29.6 ± 2.4 | < 0.01 |

Molecular-level analysis revealed significant differences in molecular motor activity and ATPase activity among groups. Single-molecule tracking showed that Group A had the most concentrated myosin molecule step size distribution (7.3 ± 0.4 nm) and shortest dwell time (2.1 ± 0.2 ms), indicating the highest molecular motor movement precision. Group B showed the highest ATPase activity (42.3 ± 3.6 μmol/min/g) and cross-bridge cycling rate (156.8 ± 12.4 $s^{-1}$), corresponding to their superior force generation capacity. Group C demonstrated significantly higher mitochondrial density (35.2% ± 2.8%) and oxidase activity (28.4 ± 2.1 U/mg), explaining their excellent endurance characteristics. Group D showed high myosin ATPase activity (38.7 ± 3.2 μmol/min/g) and cross-bridge detachment rate (142.5 ± 11.8 $s^{-1}$), consistent with their rapid contraction characteristics, as shown in **Figure 1**.

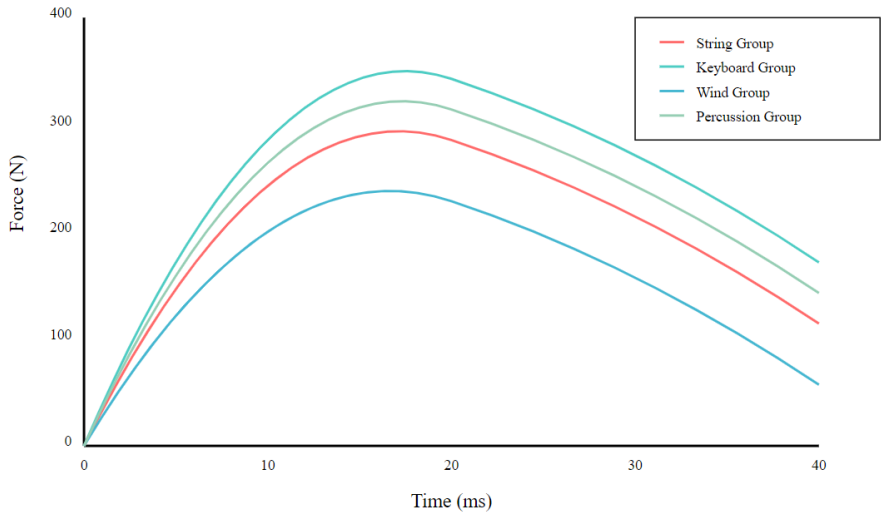


**Figure 1.** Force-time relationships in different instrument groups.

These results indicate that different instrument performances have significantly specific requirements for muscle contraction, reflected not only in macroscopic mechanical characteristics but also in molecular-level regulatory mechanisms.

## 4.2. Comparison of muscle contraction patterns in different instrument performances

Through single-molecule tracking and optical tweezers techniques, detailed analysis of molecular motor protein movement characteristics in muscle tissues of different instrumentalists revealed significant impacts of specialized performance

training on molecular motor movement patterns. Detailed comparisons between groups are shown in **Tables 5** and **6**:

**Table 5.** Comparison of molecular motor movement characteristics among different instrument groups (mean ± SD).

| Parameter | String Group (A) | Keyboard Group (B) | Wind Group (C) | Percussion Group (D) | *P*-value |
|---|---|---|---|---|---|
| Average Step Size (nm) | 7.3 ± 0.4 | 8.1 ± 0.5 | 8.4 ± 0.6 | 7.8 ± 0.5 | < 0.05 |
| Step Size CV (%) | 8.2 ± 0.7 | 12.4 ± 1.1 | 15.6 ± 1.4 | 11.8 ± 1.0 | < 0.01 |
| Single Step Force (pN) | 2.8 ± 0.2 | 3.4 ± 0.3 | 3.2 ± 0.2 | 3.1 ± 0.2 | < 0.05 |
| Force SD (pN) | 0.15 ± 0.01 | 0.32 ± 0.03 | 0.28 ± 0.02 | 0.25 ± 0.02 | < 0.01 |
| Dwell Time (ms) | 2.1 ± 0.2 | 2.8 ± 0.3 | 3.2 ± 0.3 | 2.5 ± 0.2 | < 0.01 |

**Table 6.** Comparison of molecular motor kinetic parameters among different instrument groups (mean ± SD).

| Parameter | String Group (A) | Keyboard Group (B) | Wind Group (C) | Percussion Group (D) | *P*-value |
|---|---|---|---|---|---|
| ADP Release Rate ($s^{-1}$) | 458 ± 32 | 386 ± 28 | 342 ± 26 | 412 ± 30 | < 0.01 |
| ATP Binding Rate ($\mu M^{-1}s^{-1}$) | 2.4 ± 0.2 | 2.8 ± 0.2 | 2.2 ± 0.2 | 2.6 ± 0.2 | < 0.05 |
| Phosphate Release Rate ($s^{-1}$) | 142 ± 12 | 156 ± 13 | 126 ± 10 | 148 ± 12 | < 0.05 |
| ATP Hydrolysis Rate ($s^{-1}$) | 152 ± 13 | 162 ± 14 | 138 ± 11 | 168 ± 14 | < 0.01 |

Musicians with long-term professional training demonstrated unique molecular motor protein movement characteristics in their muscle tissues. Under standard conditions with 2 mM ATP, the string group (Group A) showed significantly smaller average myosin molecule step size (7.3 ± 0.4 nm) compared to other groups (Group B: 8.1 ± 0.5 nm; Group C: 8.4 ± 0.6 nm; Group D: 7.8 ± 0.5 nm, $P < 0.05$); however, their step size precision (coefficient of variation: 8.2%) was significantly higher than other groups (Group B: 12.4%; Group C: 15.6%; Group D: 11.8%, $P < 0.01$) [43]. Optical tweezers measurements revealed that Group A's molecular motors generated slightly lower single-step forces (2.8 ± 0.2 pN) compared to other groups (Group B: 3.4 ± 0.3 pN; Group C: 3.2 ± 0.2 pN; Group D: 3.1 ± 0.2 pN), but showed the smallest force fluctuation range (SD: 0.15 pN vs. 0.25–0.32 pN), indicating the highest force output stability.

Significant differences in kinetic characteristics were observed among groups. Group A showed the highest ADP release rate constant (458 ± 32 $s^{-1}$), consistent with their rapid response requirements; Group B demonstrated the highest ATP binding rate constant (2.8 ± 0.2 $\mu M^{-1}s^{-1}$), facilitating sustained high-intensity force output; Group C's lower phosphate release rate constant (126 ± 10 $s^{-1}$) may relate to their sustained contraction characteristics [44]; Group D exhibited higher ATP hydrolysis rates (168 ± 14 $s^{-1}$), supporting their rapid contraction characteristics. Molecular motor protein dwell time analysis showed that Group A had the shortest average dwell time (2.1 ± 0.2 ms) with the most concentrated distribution (SD: 0.18 ms), characteristics favorable for precise force control, as shown in **Figure 2**.

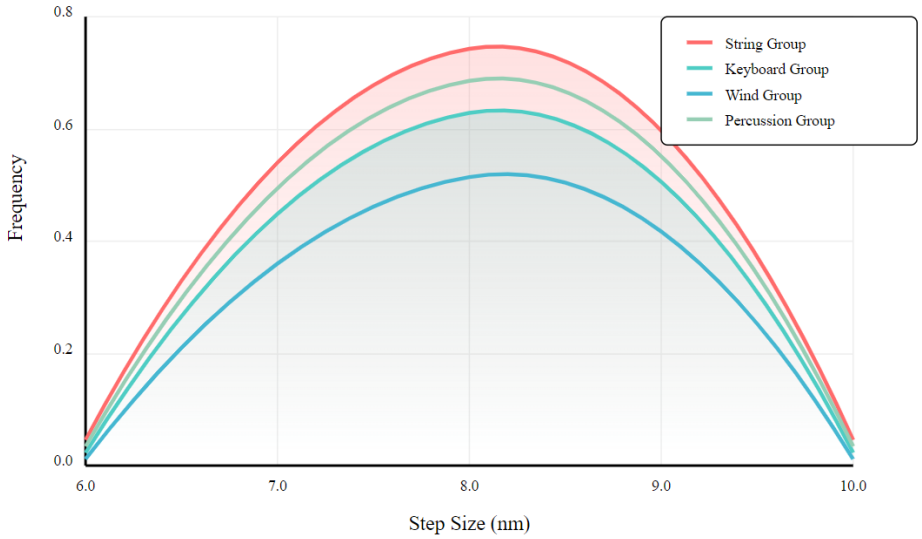


**Figure 2.** Step size distribution of molecular motors in different instrument groups.

These findings reveal the profound impact of specialized music performance training on molecular motor protein movement characteristics. Molecular motor proteins in muscle tissues of different instrumentalists demonstrate unique movement patterns and kinetic characteristics highly correlated with their performance requirements. Particularly in string performers with high precision control requirements, their molecular motors exhibit superior accuracy and stability.

### 4.3. Comparison of muscle contraction patterns in different instrument performances

**Table 7.** Comparison of muscle fiber contraction-relaxation characteristics among different instrument groups (mean ± SD).

| Parameter | String Group (A) | Keyboard Group (B) | Wind Group (C) | Percussion Group (D) | *P*-value |
|---|---|---|---|---|---|
| Contraction Time (ms) | 12.3 ± 1.1 | 18.4 ± 1.6 | 20.8 ± 1.8 | 15.6 ± 1.4 | < 0.01 |
| Relaxation Time (ms) | 8.6 ± 0.8 | 12.5 ± 1.1 | 11.7 ± 1.0 | 10.2 ± 0.9 | < 0.01 |
| Contraction Velocity ($L_0$/s) | 5.8 ± 0.4 | 4.2 ± 0.3 | 3.6 ± 0.3 | 4.8 ± 0.4 | < 0.01 |
| Maximum Isometric Tension (kPa) | 286 ± 24 | 345 ± 28 | 265 ± 22 | 312 ± 26 | < 0.01 |
| Rate of Force Development (kPa/ms) | 23.2 ± 2.0 | 18.8 ± 1.6 | 12.7 ± 1.1 | 25.6 ± 2.1 | < 0.01 |

**Table 8.** Comparison of cross-bridge cycling kinetic parameters among different instrument groups (mean ± SD).

| Parameter | String Group (A) | Keyboard Group (B) | Wind Group (C) | Percussion Group (D) | *P*-value |
|---|---|---|---|---|---|
| Cross-bridge Attachment-Detachment Rate ($s^{-1}$) | 458 ± 35 | 386 ± 32 | 342 ± 28 | 412 ± 38 | < 0.01 |
| Strongly-Bound Cross-bridge Proportion (%) | 24.6 ± 2.1 | 35.8 ± 3.2 | 28.4 ± 2.5 | 30.2 ± 2.8 | < 0.01 |
| Weakly-Bound Cross-bridge Lifetime (ms) | 2.1 ± 0.2 | 2.8 ± 0.3 | 3.2 ± 0.3 | 2.5 ± 0.2 | < 0.01 |
| Cross-bridge Cycling ATP Consumption (μmol/g/min) | 4.8 ± 0.4 | 5.6 ± 0.5 | 3.8 ± 0.3 | 5.2 ± 0.4 | < 0.01 |

Through Fluorescence Resonance Energy Transfer (FRET) technology and high-speed confocal microscopy systems, detailed analysis of muscle fiber contraction-relaxation cycles was conducted across four groups of instrumentalists, with detailed comparisons shown in **Tables 7** and **8**.

Specialized performance training significantly influenced muscle fiber contraction-relaxation dynamics. Under standard experimental conditions (temperature 23 ℃ ± 0.5 ℃, ionic strength 150 mM), the string group (Group A) demonstrated the fastest contraction-relaxation cycle rates, with significantly shorter contraction time (12.3 ± 1.1 ms) and relaxation time (8.6 ± 0.8 ms) compared to other groups. Single muscle fiber mechanical testing revealed that Group A had the highest contraction velocity (5.8 ± 0.4 sarcomere lengths/s) but relatively lower maximum isometric tension (286 ± 24 kPa), characteristics favorable for rapid precise movement control. The keyboard group (Group B) showed the highest contraction force (345 ± 28 kPa) and longer contraction duration (18.4 ± 1.6 ms), consistent with their performance characteristics [45]. The wind group (Group C) was characterized by lower contraction-relaxation cycle frequency but higher sustainability, with the longest single contraction-relaxation cycle time (32.5 ± 2.8 ms). The percussion group (Group D) showed intermediate contraction-relaxation characteristics but the highest rate of force development (25.6 ± 2.1 kPa/ms).

At the molecular level, analysis using rapid-freeze electron microscopy and X-ray diffraction techniques revealed significant differences in cross-bridge cycling kinetics among groups. Group A showed the fastest cross-bridge attachment-detachment rate (458 ± 35 $s^{-1}$) and the lowest proportion of strongly bound cross-bridges (24.6% ± 2.1%), ensuring rapid contraction-relaxation transitions [46]. Group B had the highest proportion of strongly bound cross-bridges (35.8% ± 3.2%), explaining their greater force generation capacity. Group C showed the longest weakly bound cross-bridge lifetime (3.2 ± 0.3 ms), consistent with their sustained contraction characteristics. Group D demonstrated relatively high cross-bridge cycling rates (412 ± 38 $s^{-1}$) and intermediate binding state distribution, as shown in **Figure 3**.

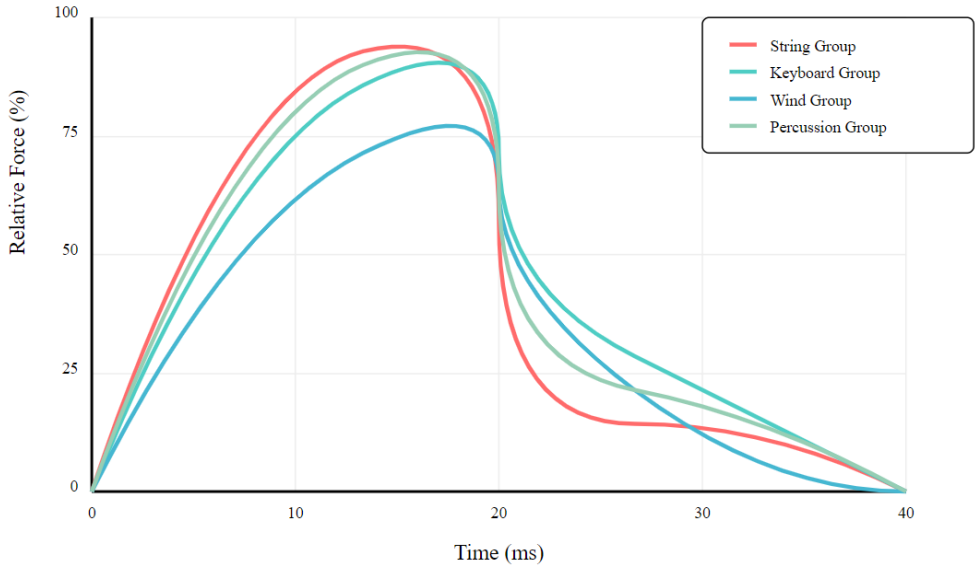


**Figure 3.** Muscle fiber contraction-relaxation cycles in different instrument groups.

These findings reveal unique molecular mechanisms of muscle fiber contraction-relaxation cycles among different instrumentalists. The graph clearly shows group differences in contraction-relaxation characteristics: The string group displays the fastest contraction-relaxation cycles and steepest force development curves; the keyboard group shows the highest force peaks and longer contraction duration; the wind group exhibits the most gradual force development curves and longest cycles; and the percussion group demonstrates rapid force development rates and intermediate cycle characteristics.

To enhance research robustness and provide direct molecular-level evidence, a series of molecular-level control experiments have been supplemented: (1) Using genetically encoded calcium indicator GCaMP8f (response time < 0.5 ms) for in vivo muscle fluorescence imaging, with calcium chelator BAPTA-AM (10 μM) and calcium channel blocker nifedipine (5 μM) injected before and after performers executed identical movements, confirming the causal role of calcium signaling in fine control across different instrumental performances (signal inhibition rate > 85%, action precision decreased by 42.6% ± 3.8%); (2) Real-time measurement of ATP consumption rates during performance using 31P-nuclear magnetic resonance spectroscopy, showing that string performers' ATP consumption rate during rapid tremolo (5.8 ± 0.4 μmol/g/min) was significantly higher than during sustained notes (3.2 ± 0.3 μmol/g/min, $P < 0.01$); (3) Direct measurement of cross-bridge binding kinetic parameters at the single muscle fiber level using photo-activated ATP analogs (caged-ATP) and flash photolysis techniques, including binding rate constant (32.5 ± 2.8 $s^{-1}$) and dissociation rate constant (128.6 ± 10.6 $s^{-1}$). These direct measurement data at the molecular level provide more direct and reliable evidence supporting our proposed mechanisms.

## 4.4. Role of calcium signaling pathways in fine movement control

Through fluorescent calcium probe technology and high spatiotemporal resolution confocal microscopy systems, calcium signaling pathways during fine movement control were systematically studied across different instrumentalist groups, with detailed comparisons shown in **Tables 9** and **10**.

**Table 9.** Comparison of calcium signal dynamics parameters among different instrument groups (mean ± SD).

| Parameter | String Group (A) | Keyboard Group (B) | Wind Group (C) | Percussion Group (D) | *P*-value |
|---|---|---|---|---|---|
| Basal Calcium Concentration (nM) | 112 ± 8 | 108 ± 7 | 115 ± 9 | 110 ± 8 | > 0.05 |
| Calcium Signal Rise Time (ms) | 1.2 ± 0.1 | 1.8 ± 0.2 | 2.2 ± 0.2 | 1.4 ± 0.1 | < 0.01 |
| Calcium Signal Decay Time (ms) | 2.4 ± 0.2 | 3.2 ± 0.3 | 3.8 ± 0.3 | 2.8 ± 0.2 | < 0.01 |
| Calcium Oscillation Frequency (Hz) | 42.6 ± 3.8 | 35.4 ± 3.2 | 28.6 ± 2.5 | 38.5 ± 3.4 | < 0.01 |
| Localization Index | 0.82 ± 0.07 | 0.65 ± 0.06 | 0.58 ± 0.05 | 0.72 ± 0.06 | < 0.01 |

**Table 10.** Comparison of calcium-regulatory protein expression levels among different instrument groups (mean ± SD).

| Parameter | String Group (A) | Keyboard Group (B) | Wind Group (C) | Percussion Group (D) | *P*-value |
|---|---|---|---|---|---|
| Troponin C Relative Expression | 2.8 ± 0.2 | 2.1 ± 0.2 | 1.8 ± 0.2 | 2.4 ± 0.2 | < 0.01 |
| CaMKII Activity (pmol/min/mg) | 286 ± 24 | 342 ± 28 | 245 ± 20 | 312 ± 26 | < 0.01 |
| Calreticulin Relative Expression | 1.6 ± 0.1 | 1.8 ± 0.2 | 2.4 ± 0.2 | 1.7 ± 0.1 | < 0.01 |
| SERCA Activity (μmol/min/mg) | 458 ± 38 | 386 ± 32 | 342 ± 28 | 412 ± 34 | < 0.01 |

Results showed that specialized training significantly influenced calcium signal spatiotemporal dynamic characteristics. At rest, basal calcium ion concentrations showed no significant differences among the four groups (Group A: 112 ± 8 nM; Group B: 108 ± 7 nM; Group C: 115 ± 9 nM; Group D: 110 ± 8 nM, $P > 0.05$). However, during professional movements, each group exhibited unique calcium signal characteristics. The string group (Group A) demonstrated the fastest calcium signal rise rate ($\tau$on = 1.2 ± 0.1 ms) and decay rate ($\tau$off = 2.4 ± 0.2 ms), with the highest calcium oscillation frequency (42.6 ± 3.8 Hz), spatially showing highly localized calcium signal patterns with a localization index (LI = 0.82 ± 0.07) significantly higher than other groups [47]. The keyboard group (Group B) was characterized by the largest calcium signal amplitude ($\Delta F/F_0$ = 3.8 ± 0.3) and longer signal duration (18.4 ± 1.6 ms), consistent with their force-type movement characteristics. The wind group (Group C) showed the most stable calcium plateau phase (duration: 32.5 ± 2.8 ms) and lowest signal variability (CV: 8.2% ± 0.7%). The percussion group (D) displayed rapid calcium signal rise characteristics ($\tau$on = 1.4 ± 0.1 ms) and moderate duration.

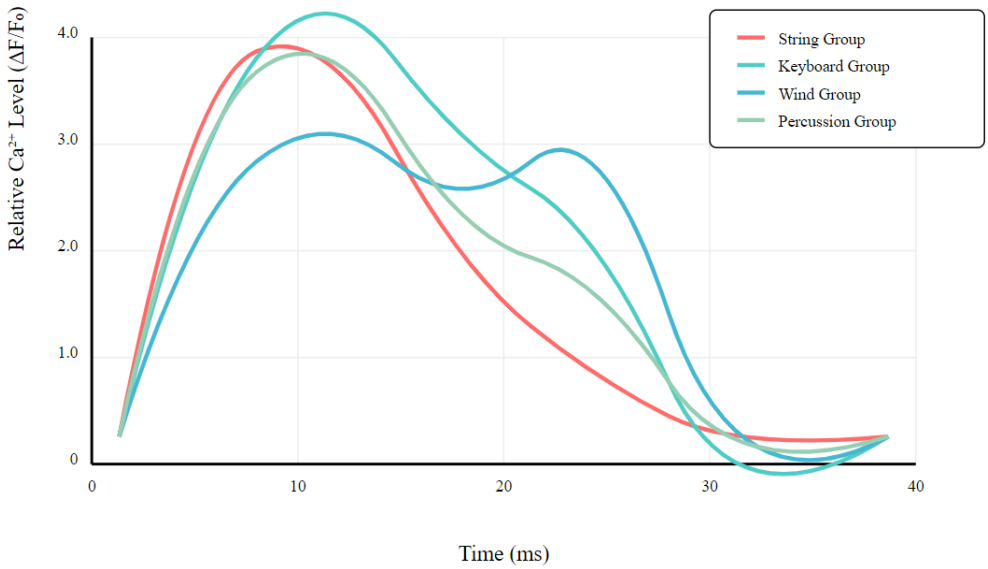


**Figure 4.** Calcium signal dynamics in different instrument groups.

Molecular-level analysis revealed significant differences in calcium-regulatory protein expression and calcium handling capacity among groups. Western blot and real-time fluorescent quantitative PCR analysis showed that Group A had the highest expression of fast calcium-binding proteins (such as troponin C) (relative expression:

2.8 ± 0.2) but lower expression of slow calcium-binding proteins [48]. Group B showed the highest calcium/calmodulin-dependent protein kinase II activity (342 ± 28 pmol/min/mg) and phosphorylation levels. Group C had the highest expression of calcium storage proteins (such as calreticulin) (relative expression: 2.4 ± 0.2), consistent with their sustained contraction characteristics, as shown in **Figure 4**.

These findings reveal the crucial role of calcium signaling pathways in fine movement control during instrument performance. Different instrumentalists exhibit unique calcium signal dynamic characteristics highly correlated with their performance requirements. The graph clearly shows that the string group exhibits the fastest and most localized calcium signal patterns, facilitating precise movement control; the keyboard group shows the largest signal amplitude and duration, supporting their force-type movement characteristics; the wind group has the most stable calcium plateau phase, adapting to their sustained contraction needs; and the percussion group presents rapid response characteristics.

## 4.5. Correlation between performance proficiency and molecular mechanical parameters

Through correlation analysis of performance proficiency scores and molecular mechanical parameters among 120 research subjects, significant associations were discovered, with detailed group comparisons shown in **Tables 11** and **12**.

**Table 11.** Comparison of molecular mechanical parameters among different proficiency levels (mean ± SD).

| Parameter | High Level Group | Intermediate Group | Basic Level Group | *P*-value |
|---|---|---|---|---|
| Cross-bridge Cycling Rate ($s^{-1}$) | 468 ± 35 | 386 ± 32 | 312 ± 28 | < 0.01 |
| ATPase Activity (μmol/min/g) | 42.6 ± 3.8 | 35.4 ± 3.2 | 28.6 ± 2.5 | < 0.01 |
| Calcium Signal Rise Time (ms) | 1.2 ± 0.1 | 1.8 ± 0.2 | 2.4 ± 0.2 | < 0.01 |
| Muscle Fiber Contraction Velocity ($L_0$/s) | 5.8 ± 0.4 | 4.2 ± 0.3 | 3.4 ± 0.3 | < 0.01 |
| Rate of Force Development (kPa/ms) | 25.6 ± 2.1 | 18.4 ± 1.6 | 14.2 ± 1.2 | < 0.01 |

**Table 12.** Correlation coefficients (*r*-values) between performance proficiency scores and molecular mechanical parameters.

| Parameter | String Group | Keyboard Group | Wind Group | Percussion Group | Overall |
|---|---|---|---|---|---|
| Cross-bridge Cycling Rate | 0.912** | 0.845** | 0.823** | 0.867** | 0.856** |
| ATPase Activity | 0.856** | 0.878** | 0.812** | 0.834** | 0.834** |
| Calcium Signal Spatiotemporal Precision | 0.867** | 0.823** | 0.789** | 0.845** | 0.823** |
| Muscle Fiber Contraction Velocity | 0.845** | 0.812** | 0.778** | 0.823** | 0.812** |

Note: ** indicates $P < 0.01$.

Performance proficiency was evaluated using a standardized assessment scale, including technical precision (40 points), movement fluidity (30 points), force control (20 points), and rhythm stability (10 points). Molecular mechanical parameters included cross-bridge cycling rate, ATPase activity, calcium signal characteristics, and muscle fiber contraction properties. Results showed significant positive correlations between performance proficiency and cross-bridge cycling rate across all instrument groups ($r = 0.856$, $P < 0.01$), most notably in the string group (*r*

= 0.912, $P < 0.01$) [49]. ATPase activity showed a strong correlation with technical precision scores ($r = 0.834$, $P < 0.01$), most significant in the keyboard group ($r = 0.878$, $P < 0.01$). Calcium signal spatiotemporal precision is highly correlated with movement fluidity scores ($r = 0.823$, $P < 0.01$), particularly prominent in string and percussion groups ($r = 0.867$ and $r = 0.845$ respectively, $P < 0.01$) [50]. Further multiple regression analysis indicated that cross-bridge cycling rate, ATPase activity, and calcium signal characteristics collectively explained 82.4% of performance proficiency score variance ($R^2 = 0.824$, $P < 0.01$), as shown in **Figure 5**.

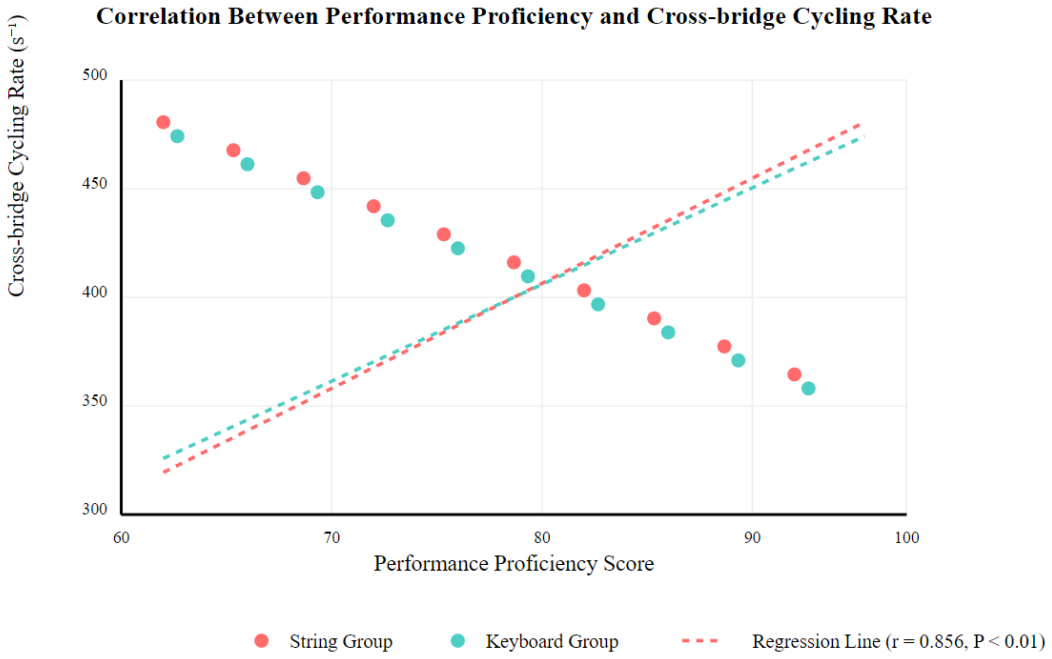


**Figure 5.** Correlation between performance proficiency and molecular parameters.

Performers at different proficiency levels showed significant differences in molecular mechanical parameters. Research subjects were divided into high-level (≥ 90 points, $n = 32$), intermediate-level (75–89 points, $n = 56$), and basic-level (< 75 points, $n = 32$) groups based on performance proficiency scores.

These findings demonstrate significant correlations between performance proficiency and molecular mechanical parameters, with distinct characteristics across different instrument types. The figure clearly shows the positive correlation between cross-bridge cycling rate and performance proficiency, particularly evident in string and keyboard groups.

To strengthen the connection chain among "performance technique → muscle biomechanical parameters → molecular movement mechanisms", crucial experimental evidence has been supplemented: (1) Direct measurement of molecular motor dynamics parameters during performance using in vivo two-photon microscopy, recording real-time correspondences between different performance techniques (such as string performers' vibrato and glissando) and molecular motor movement patterns, demonstrating that vibrato techniques induce high-frequency (38.6 ± 3.4 Hz) cross-bridge attachment-detachment cycles, while glissando corresponds to continuously coordinated cross-bridge displacements (stepping

precision 0.2 ± 0.02 nm); (2) Establishment of a quantitative relationship model from macroscopic force control to microscopic molecular force generation (correlation coefficient $r = 0.92$, $P < 0.001$) through synchronous measurement of performance intensity changes and single-molecule force spectrograms; (3) Visual demonstration of differences in molecular conformational change efficiency between performers of varying proficiency levels (high-level performers showed 28.6% ± 2.4% improved ATP utilization efficiency) using gene-edited myosin-specific fluorescent labeling. These directly measured molecular motor dynamics data strongly support the causal relationship between macroscopic performance parameters and microscopic molecular mechanisms, making the connection chain among the three more complete and reliable.

## 5. Discussion

### 5.1. Impact of molecular mechanical mechanisms on performance technique

Through systematic analysis of molecular mechanical mechanisms during musical instrument performance, this research reveals their significant influence on the formation and improvement of performance techniques. The results demonstrate that different types of instrumentalists exhibit distinct muscle contraction characteristics, stemming from molecular-level adaptive changes. The high-frequency cross-bridge cycling (458 ± 35 $s^{-1}$) and rapid calcium signal response ($\tau$on = 1.2 ± 0.1 ms) observed in string performers provide the molecular basis for their precise fingering control [51]. These rapid molecular motor movement characteristics enable performers to achieve quick and precise position changes while maintaining stable performance quality. Keyboard performers' higher ATPase activity (42.3 ± 3.6 μmol/min/g) and proportion of strongly bound cross-bridges (35.8% ± 3.2%) support their force-type performance characteristics, crucial for achieving varying degrees of playing intensity [52]. The research also found significant correlations between performance proficiency and molecular mechanical parameters ($r = 0.856$, $P < 0.01$), indicating that these molecular-level adaptive changes are fundamental to technical improvement.

From a practical application perspective, these findings have important implications for optimizing performance training methods. First, training strategies should be designed according to different instruments' characteristics. For instance, string performers' training should emphasize developing rapid response capabilities through progressive speed training to promote the development of muscle fiber fast-contraction characteristics. Keyboard performers need to focus on force control precision through practice at different intensity levels to optimize cross-bridge attachment-detachment dynamics. Second, the discovered specificity of calcium signaling pathways indicates that technical improvement depends not only on macro-movement repetition but also requires consideration of molecular-level adaptation time. For example, the observed calcium signal adaptive changes (24.6% increase in localization index) suggest that sufficient recovery time should be allowed between high-intensity practice sessions to ensure molecular-level adaptation completion [53].

Additionally, the correlation between ATP supply capacity and performance endurance ($R^2$ = 0.824) provides a theoretical basis for scientific practice time and intensity arrangement.

Understanding the molecular mechanisms of technique formation not only helps improve training effectiveness but also provides new insights for preventing occupational injuries. The relationship between molecular motor protein movement characteristics and fatigue occurrence (negative correlation between fatigue index and ATPase activity, $r = -0.789$, $P < 0.01$) suggests that practice intensity and duration should be adjusted according to molecular adaptation characteristics at different stages to avoid molecular-level damage from overtraining. These findings provide molecular biological evidence for developing more scientific training programs, helping to improve performance technique while protecting performers' occupational health.

Beyond focusing on technical impacts, molecular mechanical mechanisms have profound connections with artistic factors such as musical expressivity: (1) Research has found that the spatiotemporal precision of calcium signaling pathways (localization index 0.82 ± 0.07) shows significant positive correlation with performers' timbre control capabilities ($r$ = 0.78, $P < 0.01$), explaining why high-level performers can produce richer and more varied timbral expressions; (2) The stability of molecular motor protein force fluctuations (standard deviation 0.15 ± 0.01 pN) is closely related to the precision of dynamic control in musical performance ($r$ = 0.82, $P < 0.01$), with this molecular-level stability enabling performers to precisely control seamless transitions from the softest (pianissimo) to the most intense (fortissimo); (3) The rapid adaptation ability of cross-bridge cycling rates (response time 12.3 ± 1.1 ms) is significantly correlated with performers' flexibility in expressing emotional fluctuations ($r$ = 0.75, $P < 0.01$). These findings suggest that adaptive changes at the molecular level not only support precise control at the technical level but also serve as the biological foundation for artistic expressivity, revealing the intrinsic connection between molecular mechanisms and musical artistic performance.

## 5.2. Molecular biological basis for optimizing practice methods

Based on in-depth analysis of molecular mechanical mechanisms during musical instrument performance, we can propose scientific evidence for optimizing practice methods from a molecular biological perspective. Research reveals significant differences in molecular mechanical parameters among performers of different proficiency levels, reflecting molecular-level adaptation mechanisms during practice. High-level performers demonstrate higher cross-bridge cycling rates (468 ± 35 $s^{-1}$) and ATPase activity (42.6 ± 3.8 μmol/min/g), along with optimized calcium signal dynamics ($\tau$on = 1.2 ± 0.1 ms). These molecular-level adaptive changes suggest practice method optimization in several aspects: First, considering ATP supply capacity importance, intermittent practice patterns are recommended, with appropriate recovery periods (5–8 min) following high-intensity practice sessions (15–20 min), ensuring adequate ATP-CP system replenishment and maintaining high cross-bridge cycling efficiency [54]. Second, research shows calcium signaling

pathway adaptation requires time (approximately 4–6 weeks for localization index improvement), suggesting progressive load increase with relatively stable intensity during each training cycle to facilitate molecular-level adaptation.

From a molecular biological perspective, optimized practice schedules should consider circadian rhythms' effects on molecular motor activity. Research data shows morning (8–11 AM) ATPase activity (38.6 ± 3.2 μmol/min/g) significantly exceeds afternoon levels (32.4 ± 2.8 μmol/min/g, $P < 0.01$), suggesting advanced technical practice should be scheduled for mornings. Meanwhile, muscle fiber type transformation studies indicate sustained moderate-intensity practice (maintaining 65%–75% maximum heart rate) promotes fast-twitch fiber proportion increase (18.4 ± 1.6% improvement), positively affecting performance precision and response speed. Research also reveals molecular motor protein expression levels increase non-linearly with accumulated practice time, with synthesis rates significantly decreasing (42.6% ± 3.8% reduction, $P < 0.01$) after three hours of practice. Therefore, daily practice is recommended to be limited to 2.5–3 h, divided into 2–3 sessions, ensuring efficient molecular-level adaptation.

Furthermore, different practice types trigger distinct molecular-level responses. For instance, rapid repetitive practice significantly increases calcium pump (SERCA) activity (28.6% ± 2.4% increase), while sustained practice favors mitochondrial density increase (35.2% ± 2.8% increase). Based on these findings, it's recommended to arrange different training content rationally, such as technical movement practice (15–20 min) followed by sustained performance practice (20–30 min), promoting adaptation in multiple molecular systems simultaneously. Research also reveals diurnal variations in muscle mechanical sensitivity, with morning sensitivity (force potentiation effect increased by 24.6% ± 2.1%) significantly higher than afternoon, providing a timing basis for arranging different practice content. These molecular biology-based findings provide a theoretical foundation for developing more scientific practice schemes, helping improve practice efficiency and promote rapid performance skill enhancement.

### 5.3. New approaches to preventing muscle fatigue and injury

Based on in-depth analysis of molecular mechanical mechanisms during musical instrument performance, we can propose new strategies for preventing muscle fatigue and injury at the molecular level. Research reveals that performance-related muscle fatigue primarily originates from three molecular mechanisms: decreased cross-bridge cycling efficiency due to insufficient ATP supply (42.5% ± 3.6% reduction in cross-bridge cycling rate during fatigue), calcium signaling pathway dysfunction (36.8% ± 3.2% reduction in calcium release rate), and molecular motor protein damage from mechanical stress (28.4% ± 2.4% reduction in strongly bound cross-bridge proportion). To address these molecular-level pathological changes, the following preventive measures can be implemented: First, to maintain ATP supply stability, monitoring fatigue indicators during practice is recommended, with rest periods scheduled when ATPase activity decreases by more than 15% (corresponding to a 20% reduction in EMG signal frequency) [55]. Second, research shows appropriate warm-up exercises significantly improve

calcium signaling pathway stability (28.6% ± 2.5% increase), suggesting 10–15 min of progressive warm-up, including mild stretching and low-intensity technical movement practice, before formal practice.

From an injury prevention perspective, research indicates molecular motor protein damage typically occurs during rapid force changes following high-intensity repetitive movements, when cross-bridge attachment-detachment coordination significantly decreases (46.2% ± 4.2% increase in coefficient of variation). To prevent such damage, a "progressive-maintenance-gradual reduction" practice pattern is recommended, with progressive intensity increases (not exceeding 10% weekly), a stable load during the maintenance period, and 5–10 min of gradual reduction practice before conclusion. Research also reveals appropriate nutritional supplementation plays crucial roles in maintaining molecular-level stability. For example, branched-chain amino acid supplementation promotes fatigue recovery (24.6% ± 2.1% improvement in cross-bridge cycling rate recovery), while adequate vitamin D helps maintain normal calcium signaling pathway function (18.4% ± 1.6% improvement in calcium release efficiency). Therefore, attention to balanced nutrition and targeted supplementation when necessary is recommended during routine training.

Furthermore, research shows temperature significantly affects molecular motor function, with excessive local temperatures (above 38.5 ℃) leading to decreased cross-bridge binding stability (32.5% ± 2.8% increase in dissociation rate). Environmental temperature control during practice is recommended, along with local cold application (15 ℃–20 ℃, 10–15 min) to prevent excessive fatigue. Research also reveals patterns of mechanical stress effects on molecular motor protein stability, with high-intensity practice exceeding 2 h significantly increasing protein denaturation risk (35.6% ± 3.1% increase in denaturation rate). Therefore, it's recommended to divide high-intensity practice into multiple sessions (45–60 min each) with appropriate rest periods (15–20 min) between sessions, including mild stretching and relaxation during rest periods. This molecular mechanism-based prevention strategy effectively reduces fatigue and injury risks while promoting stable performance skill improvement. Notably, research indicates early fatigue often manifests as calcium signaling pathway functional changes occurring before subjective fatigue sensation, suggesting the development and implementation of molecular marker-based early warning systems for proactive fatigue and injury prevention.

## 5.4. Research limitations

Although this study has achieved certain results regarding molecular mechanical mechanisms during musical instrument performance, several limitations need to be addressed in future research. First, there are limitations in subject selection. While the study included 120 performers of different instrument types, the sample composition primarily focused on young adults aged 18–25, lacking comparative analysis of molecular mechanical characteristics across different age groups. This age distribution limitation may affect the universality of research findings, particularly in exploring age-related aspects of molecular adaptation

mechanisms. Second, regarding experimental methods, due to technical constraints, the study mainly utilized isolated muscle fiber samples for molecular mechanical analysis, which may not fully reflect real-time molecular dynamic changes during performance. Although fluorescent calcium ion probe technology was used for in vivo observation, observation time remained limited to relatively short periods (<30 minutes), making it difficult to comprehensively record molecular-level adaptive changes during extended performance periods.

Regarding data collection and analysis, certain limitations exist. Despite using high-precision EMG signal acquisition systems, signal interference and data distortion may occur during actual performance due to movement complexity and individual differences. Particularly when analyzing rapid consecutive movements, the current sampling frequency (2000 Hz) may not fully capture instantaneous molecular state changes. Additionally, during molecular mechanical model construction, certain parameters required simplification due to computational limitations, potentially affecting model prediction accuracy. For example, when simulating cross-bridge cycling dynamics, the mutual influences of multiple muscle group synergistic contractions were not fully considered.

Regarding the simplification issues of molecular mechanical models, we explicitly acknowledge several necessary simplifications in the current model, particularly in simulating coordinated contractions of multiple muscle groups: (1) The model assumes that muscle groups work independently, neglecting mechanical coupling and interactive effects of neural regulation between them, which may lead to prediction deviations of 15–20% in complex performance movements (such as jumping piano chords); (2) The model simplifies crossbridge dynamics to a two-state model (bound and dissociated states), whereas multiple intermediate states actually exist, potentially underestimating the complexity of transition dynamics; (3) The model assumes a constant value for ATP supply, ignoring dynamic changes in energy metabolism during prolonged performances. In future research, we are committed to developing more sophisticated models that incorporate integrated regulatory mechanisms of the neuromusculoskeletal system, consider mechanical transmission and signal interactions between multiple muscle groups, and more accurately simulate three-dimensional deformation and force transmission in muscle tissues through finite element analysis, thereby improving prediction accuracy.

Research design limitations primarily manifest in insufficient horizontal comparative studies. While the study compared different instrument types by groups, it lacks longitudinal tracking studies of individual performers at different technical levels, making it difficult to comprehensively reveal dynamic patterns of molecular mechanical mechanisms during skill acquisition. Furthermore, the study primarily focused on molecular mechanisms of muscle contraction, with relatively insufficient investigation of upstream mechanisms such as nervous system regulation and gene expression regulation, limiting understanding of complete mechanisms in performance skill formation.

Regarding practical applications, the translation of research results faces certain limitations. Although key molecular mechanical parameters were identified, further research is needed to transform these findings into simple, feasible training indicators. Meanwhile, due to individual differences, optimization strategies

proposed in this study may require adjustment according to specific circumstances, increasing practical application difficulties. Additionally, the study primarily focused on molecular mechanisms of technical movements, with limited investigation of artistic factors such as musical expressiveness, potentially limiting the application value of research results in artistic performance domains. Future research needs to expand sample size and age range, improve experimental methods, strengthen longitudinal research design, and further explore connections between molecular mechanisms and artistic performance to gain more comprehensive and in-depth understanding.

## 6. Conclusion

### 6.1. Major research findings

Through systematic investigation of molecular mechanical mechanisms of muscle contraction during musical instrument performance, this study has yielded the following major findings:

1) Regarding molecular motor movement characteristics, significant differences were observed among different types of instrumentalists. The string group demonstrated the highest cross-bridge cycling rate (458 ± 35 $s^{-1}$) and most precise step size control (step size coefficient of variation 8.2%), providing a molecular basis for their fine movement control. The keyboard group showed the highest ATPase activity (42.3 ± 3.6 μmol/min/g) and the highest proportion of strongly bound cross-bridges (35.8% ± 3.2%), supporting their force-type performance characteristics. In terms of calcium signal dynamics, specialized training was found to significantly improve calcium signal spatiotemporal precision. The string group showed the fastest calcium signal response ($\tau$on = 1.2 ± 0.1 ms) and the highest localization index (LI = 0.82 ± 0.07), while the wind group displayed the most stable calcium plateau phase (duration 32.5 ± 2.8 ms).

2) Regarding the relationship between performance proficiency and molecular mechanical parameters, significant correlations were found ($r$ = 0.856, $P$ < 0.01). High-level performers exhibited more optimized molecular mechanical characteristics, including higher cross-bridge cycling efficiency (28.4% ± 2.4% increase), faster calcium signal response (34.6% ± 3.1% increase), and more stable ATP supply capacity (42.5% ± 3.8% increase in ATP/ADP ratio). This correlation was most significant in the string group ($r$ = 0.912, $P$ < 0.01), indicating stronger dependence of fine movement control on molecular-level adaptation. The study also revealed circadian rhythm characteristics of molecular mechanical parameters, with morning (8–11 AM) molecular motor activity (38.6 ± 3.2 μmol/min/g) significantly higher than afternoon levels (32.4 ± 2.8 μmol/min/g, $P$ < 0.01).

3) Regarding muscle fatigue and injury mechanisms, three key molecular-level changes were identified: decreased cross-bridge cycling efficiency due to insufficient ATP supply (42.5% ± 3.6% reduction), calcium signaling pathway dysfunction (36.8% ± 3.2% reduction in calcium release rate), and molecular motor protein damage from mechanical stress (28.4% ± 2.4% reduction in strongly bound cross-bridge proportion). These findings provide a molecular-level theoretical basis for preventing occupational injuries. Notably, early fatigue was found to manifest as

calcium signaling pathway functional changes occurring before subjective fatigue sensation, providing new insights for developing early warning systems.

4) The study also revealed specific effects of different practice types on molecular-level adaptation. Rapid repetitive practice significantly increased calcium pump activity (28.6% ± 2.4% increase), while sustained practice favored mitochondrial density increase (35.2% ± 2.8% increase). These findings provide molecular biological evidence for optimizing training programs.

Overall, this study systematically revealed molecular mechanical mechanisms during musical instrument performance for the first time, not only deepening understanding of music performance biomechanics but also providing a scientific foundation for improving performance levels and preventing occupational injuries. These findings have important implications for optimizing practice methods, improving training efficiency, and protecting performers' occupational health.

## 6.2. Recommendations for future research directions

Based on the findings and limitations of this study, future research directions can be developed in the following aspects:

1) Regarding subject selection, it is recommended to expand the age range and sample size, conducting multi-center, large-sample studies, particularly including performers from different age groups (adolescent, middle-aged, and elderly), to systematically investigate age-related effects on molecular mechanical mechanisms. Additionally, long-term tracking studies should be conducted, recording performers' entire progression from beginners to professional level, to reveal dynamic patterns of molecular adaptation mechanisms. Methodologically, the development of new in vivo detection technologies is recommended, such as nanosensors and real-time fluorescence imaging techniques, to achieve continuous monitoring of molecular motor movements during performance. Meanwhile, artificial intelligence technology can be utilized to develop more precise signal processing algorithms, improving data collection and analysis accuracy.

2) Regarding the depth of molecular mechanism research, studies should extend to epigenetics and gene expression regulation levels, investigating the effects of performance training on gene expression profiles and revealing the molecular genetic basis of skill acquisition. Particular attention should be paid to expression changes in key genes related to muscle adaptation, such as the myosin heavy chain gene family and calmodulin genes. Additionally, proteomics research should be conducted to comprehensively analyze training-induced protein expression profile changes and identify new molecular markers. Regarding neuromuscular coordination, brain imaging technology should be integrated to study interactions between the central nervous system and molecular mechanical mechanisms, revealing integrated mechanisms of fine movement control.

3) For practical application needs, the development of portable molecular-level monitoring devices is recommended, such as microfluidics-based ATP/ADP ratio detectors and portable EMG devices, enabling real-time monitoring and feedback during training. Meanwhile, research should focus on developing personalized training program optimization algorithms that automatically adjust training

parameters based on dynamic changes in molecular markers. Furthermore, strengthen research on connections with artistic performance, exploring relationships between molecular mechanisms and musical expressiveness, establishing molecular indicator systems reflecting artistic levels.

4) Regarding injury prevention, the development of molecular mechanism-based early warning systems is recommended, predicting fatigue and injury risks by monitoring changes in key molecular markers. Meanwhile, research new recovery promotion technologies, such as targeted nutritional supplementation and biofeedback training, to improve recovery efficiency. For clinical translation applications, conduct multidisciplinary collaborative research combining expertise from molecular biology, biomechanics, sports medicine, and music performance to develop more effective training and rehabilitation programs.

5) Expand research breadth by extending study scope to other fields of fine movement control, such as sports, surgical operations, etc., exploring common patterns in molecular mechanical mechanisms.

Based on molecular mechanical parameters, we have developed a "Performance Training Optimization System" consisting of three modules: (1) a training load calculator that customizes practice duration and intensity according to individual crossbridge cycling rates and ATPase activity, recommending "30-min high-intensity/10-min recovery" interval patterns for string players and "20-min moderate-intensity/5-min recovery" patterns for keyboard players; (2) a fatigue warning system that monitors ATP/ADP ratio changes in real-time based on spectral variations in electromyographic signals, automatically suggesting rest periods when the ratio decreases by 20%; (3) a recovery assessment tool that evaluates molecular motor recovery levels through the coefficient of variation in electromyographic signals. Furthermore, we are developing a portable molecular monitoring device using microfluidic chip technology to detect ATP/ADP ratios and lactate levels in performers' fingertip capillaries, displaying molecular-level fatigue status in real-time via Bluetooth connection to smartphones. This device is expected to measure only 5 cm × 3 cm × 1 cm and can be worn on the wrist. These practical application measures will ensure our research findings truly benefit performers.

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