

Cardioprotective Effect of the F-36 Alkaloid and DKV-8 Conjugate

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Abstract

The search for new therapeutic strategies for cardiovascular diseases remains an urgent issue. This study investigated the effects of the natural compound 6,7-dimethoxy-1,2,3,4-tetrahydroisoquinoline hydrochloride (F-36) and its conjugate with dihydroquercetin, DKV-8, on the contractile activity of rat heart papillary muscle and their molecular mechanisms. In vitro mechanographic experiments showed that F-36 (5 - 100 μ M) and DKV-8 (5 - 40 μ M) dose-dependently increased contraction force by 75.7 ± 4.7 and $143.6 \pm 5.4\%$, respectively. studies using specific inhibitors (lidocaine, nifedipine, NiCl_2 , caffeine, and cyclopiazonic acid (CPA)) demonstrated that Na^+ channels, L-type Ca^{2+} channels, and sarcoplasmic reticulum Ca^{2+} -ATPase (SERCA2a) play a key role in the observed positive inotropic effect. Overall, F-36 and its DKV-8 conjugate enhance cardiomyocyte Ca^{2+} homeostasis mainly through regulation of SERCA2a and plasma membrane ion channels. Their effects were also confirmed under in vitro hypoxic conditions.

Keywords: Ca^{2+} homeostasis, Cardioprotection, Conjugate, Isoquinoline alkaloid, Papillary muscle, Positive inotropy, SERCA2a

Introduction

Cardioprotection is one of the priority directions in the prevention and treatment of cardiovascular diseases, which remain a major health problem despite significant advances in therapy [1]. According to the World Health Organization, the limited effectiveness and side effects of many currently used drugs contribute to this situation. Disruption of ion transport systems in the sarcolemma [2-4].

Ion channel dysfunction plays an important role in the development of cardiovascular diseases. In particular, the $\text{Na}^+/\text{Ca}^{2+}$ exchanger (NCX) is a key regulator of Ca^{2+} homeostasis and cardiac contractility [5-7]. Under pathological conditions such as ischemia/reperfusion, increased intracellular Na^+ leads to excessive Ca^{2+} accumulation in cardiomyocytes. In addition, SERCA2a plays a crucial role in regulating intracellular Ca^{2+} concentration and in controlling myocardial contraction and relaxation [8-11].

Disturbances in these Ca^{2+} -regulating systems can lead to c. Furthermore, the Na^+/H^+ exchanger (NHE) contributes to intracellular pH regulation and is involved in tissue damage during ischemia and reperfusion [12-14]. Hypoxia significantly reduces myocardial contractile force due to impairment of Ca^{2+} transport systems and Ca^{2+} homeostasis, including Ca^{2+} channels, NCX, Na^+/K^+ -ATPase, KATP channels, ryanodine prescription (RyR2), and SERCA2a [15-17]. Therefore, the study of biologically active substances that can regulate these mechanisms and provide cardioprotective effects with minimal side effects is of great scientific interest.

Material and methods

The study was conducted at the Cell Biophysics Laboratory of the Institute of Biophysics and Biochemistry under the National University of Uzbekistan. Experiments involving animals adhered to the principles outlined in the international Helsinki Declaration as well as the guidelines established by the Council for International Organizations of Medical Sciences (CIOMS, 1985). The regulations of the Institute concerning “Bioethics in the Use of Laboratory Animals for Scientific Research” were strictly observed.

In vitro experiments were performed to investigate the mechanisms by which biologically active compounds affect myocardial functional activity, utilizing a mechanography approach. The contractile activity of papillary muscle preparations was recorded using the SI-BAM21-LC mechanographic device (World Precision Instruments Inc., WPI, USA). In this setup, the muscle preparation was secured with an SI-OHO2F attachment, and the mechanical signal was transmitted via an SI-KG20 piezoelectric sensor to the SI-BAM21-LCB signal amplifier [18]. The signal was then converted through the Lab-Trax-4/16 converter (WPI, USA) and digitized on an IBM PC using the iWorx LabScribe2 software (iWorx Systems, Inc., USA) in a digital format (*.iwxdata) for primary mathematical and statistical processing.

Experiments were conducted using albino rats weighing 200 - 250 g. Animals were anesthetized with sodium pentobarbital (40 mg/kg) prior to the procedure. Under deep anesthesia, the rats were euthanized in accordance with international ethical guidelines, initially by cervical dislocation followed by

decapitation. The thoracic cavity was carefully opened, and the heart was gently excised and immediately placed into a Petri dish containing Krebs solution.

The left ventricle of the heart was opened to isolate papillary muscles. The prepared cardiac tissues were then placed in a chamber filled with Krebs solution with the following composition (mM): NaCl - 150; KCl - 4; CaCl_2 - 1.8; MgCl_2 - 1; NaHCO_3 - 14; NaH_2PO_4 - 1.8; $\text{C}_6\text{H}_{12}\text{O}_6$ - 11.5; pH = 7.4.

In-vitro hypoxia model

To assess the potential cardioprotective activity of the compounds under hypoxia, rat hearts were perfused with oxygen (95% O_2 /5% CO_2) in a nitrogen (95% N_2 /5% CO_2) gas for 60 min, under conditions that resulted in maximal changes in papillary muscle contractile activity parameters.

Papillary muscle preparation and statistical analysis

The papillary muscle (myocardial) preparations from experimental animals serve as a convenient *in vitro* model for investigating the mechanisms of antiarrhythmic action of pharmacological agents under both physiological and pathological conditions. The papillary muscles of these animals typically measure 1.5 - 4 mm in length, and are widely used in experiments due to their ease of isolation, well-organized striated sarcomeric myofibrillar structure, robust contractile activity, and ability to maintain functional activity for over 6 - 8 h *in vitro*.

For preparation, experimental animals were anesthetized with diethyl ether and euthanized via cervical dislocation. The thoracic cavity was surgically opened, the heart excised, and the papillary muscle preparations (diameter 0.4 - 1.3 mm; length 2.5 - 3.8 mm) were carefully placed into a Petri dish containing Krebs-Henseleit physiological solution.

In these experiments, the contractile force of the papillary muscles was measured at stimulation frequencies of 0.1 - 1 Hz and expressed as a percentage relative to the maximum response. The results represent the arithmetic mean of 5 - 6 independent experiments. All data are presented as mean $\pm$ standard error of the mean (SEM). The datasets were tested for normality using the Shapiro-Wilk test and for homogeneity of variance using Levene's test. Differences were

considered statistically significant at $p < 0.05$. Statistical analyses of the experimental results were performed using OriginPro 2021 software (OriginLab Corporation, USA).

Chemical part

Currently, biologically active compounds isolated from medicinal plants, such as flavonoids and alkaloids, are widely used in the treatment of cardiovascular diseases. In this context, researchers at the Institute of Plant Chemistry of the Academy of Sciences of the Republic of Uzbekistan have investigated the effects of 6,7-dimethoxy-1,2,3,4-tetrahydroisoquinoline hydrochloride (F-36) and the conjugate 2-(3,4-

dihydroxyphenyl)-6-((6,7-dimethoxy-1,2,3,4-tetrahydroisoquinolin-2(1N)-yl)methyl)-3,5,7-trihydroxychroman-4-one (DKV-8) [19] on the contractile force of papillary muscles in rat hearts.

In addition, the effects of various ion channel modulators and regulators were studied, including the Na^+ channel blocker lidocaine ($\text{C}_{14}\text{H}_{22}\text{N}_2\text{O}$, Sigma Ltd), the L-type Ca^{2+} channel blocker nifedipine ($\text{C}_{17}\text{H}_{18}\text{N}_2\text{O}_6$, Sigma Ltd), the $\text{Na}^+/\text{Ca}^{2+}$ exchanger blocker nickel chloride (NiCl_2 , Sigma Ltd), the RyR2 activator caffeine ($\text{C}_8\text{H}_{10}\text{N}_4\text{O}_2$), and the SERCA2a inhibitor cyclopiazonic acid ($\text{C}_{20}\text{H}_{20}\text{N}_2\text{O}_3$, Sigma Ltd) (**Figure 2**).

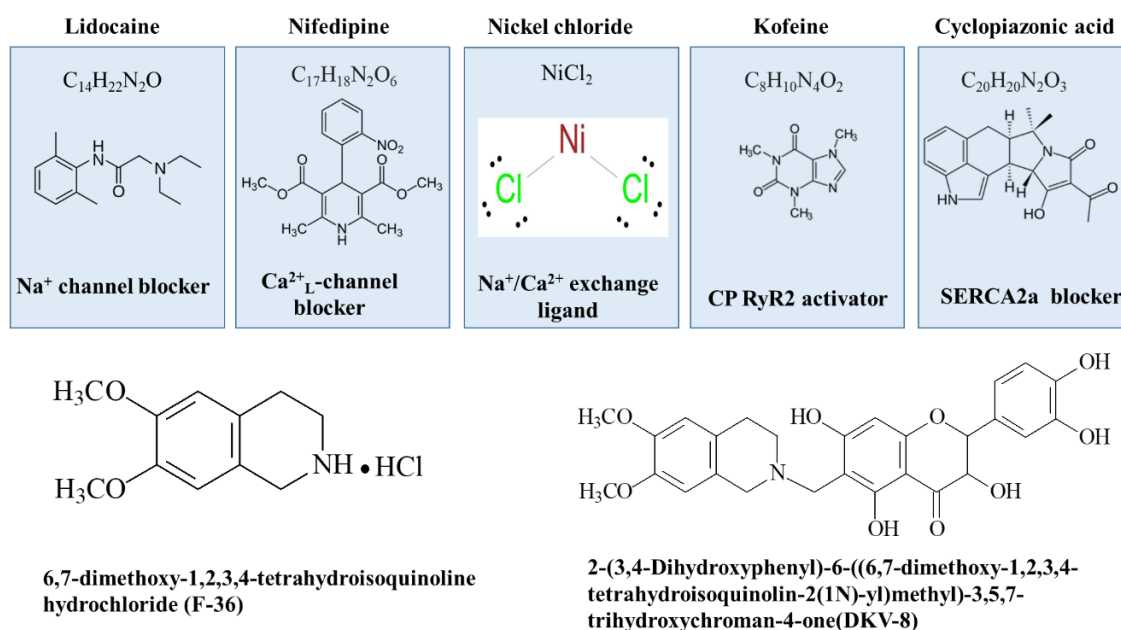


Figure 1 Chemical structure of the conjugate of bilocation and activator, as well as the isoquinoline alkaloid F-36 and DKV-8.

Results and discussion

Dose-dependent effects of the F-36 alkaloid and DKV-8 conjugate

In this context, the dose-dependent effects of the DKV-8 conjugate, derived from the isoquinoline alkaloid F-36 and the flavonoid dihydroquercetin (DHQ), on the contractile activity of rat cardiac papillary muscles were investigated. It was observed

that F-36 (5 - 100 μM) and the DKV-8 conjugate (5 - 40 μM) exhibited a positive inotropic effect at all tested concentrations, enhancing the contractile force of the papillary muscles. The study demonstrated that these biologically active compounds increased the contraction force of rat cardiac papillary muscles by $75.7 \pm 4.7\%$ and $143.6 \pm 5.4\%$, respectively, relative to the control (set as 100%) (**Figure 2**).

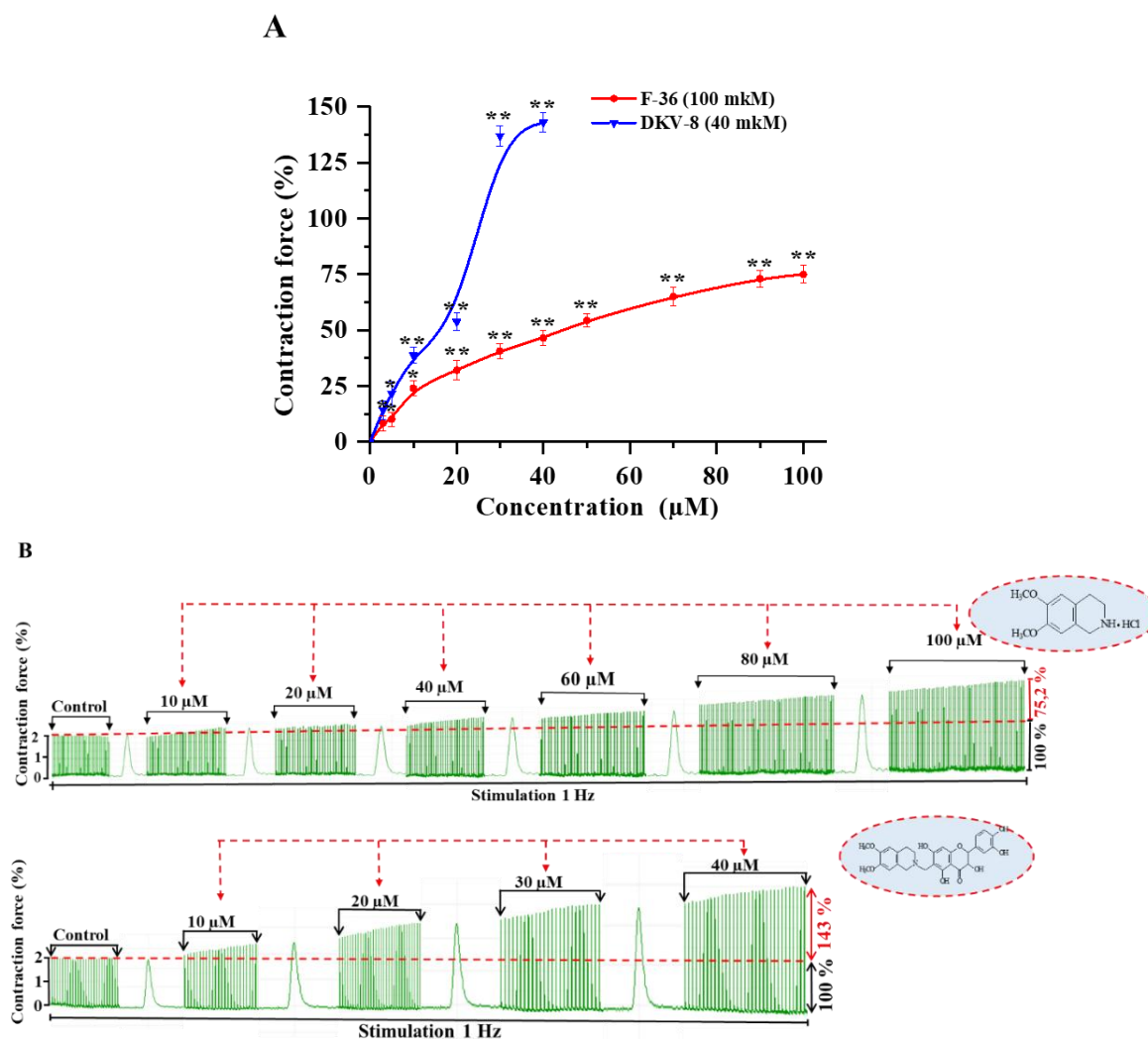


Figure 2 (A) Dose-dependent positive inotropic effect of the isoquinoline alkaloid F-36 and the conjugate DKV-8 on papillary muscle contractile activity. (B) (Under these conditions) The ordinate axis shows the force of contraction of the papillary muscle, expressed as a percentage of the maximum value, which is taken as 100%. The abscissa axis represents the concentration of alkaloids and conjugates. Stimulation frequency 1 Hz ($t = +36 \pm 0.5$ °C); * $-r < 0.05$, ** $-r < 0.01$ $n = 5$.

Under these conditions, it was observed that the DKV-8 conjugate and F-36 significantly increased the force of rat cardiac papillary muscle contraction compared to isoquinoline alkaloids.

Effect of F-36 and DKV-8 on Na^+ channel

During the study, the effects of biologically active compounds on Na^+ channels in the sarcolemma of cardiomyocytes were investigated. Intracellular Na^+ concentration ($[\text{Na}^+]_i$) plays a key role in modulating cardiac contractile function [20-22]. Electrical stimulation of the myocardium activates voltage-gated Na^+ channels, initiating an action potential [23]. During

the plateau phase of this action potential, Ca^{2+} ions enter cardiomyocytes from the extracellular space via L-type Ca^{2+} channels, triggering the release of Ca^{2+} from the sarcoplasmic reticulum (SR) into the cytosol, which ultimately induces contraction [24,25]. Considering the critical role of Na^+ channels in cardiac contraction, subsequent experiments focused on the impact of the tested biologically active substances on cardiomyocyte Na^+ channels.

Preliminary experiments assessed the dose-dependent effects (5 - 30 μM) of lidocaine on papillary muscle contractile activity at a stimulation frequency of 1 Hz. Lidocaine at 30 μM reduced the amplitude of

papillary muscle contraction by $81.7 \pm 4.3\%$ relative to the control. The half-maximal inhibitory concentration

(IC₅₀) of lidocaine on papillary muscle contractility was determined to be $15.1 \mu\text{M}$ (Figure 3).

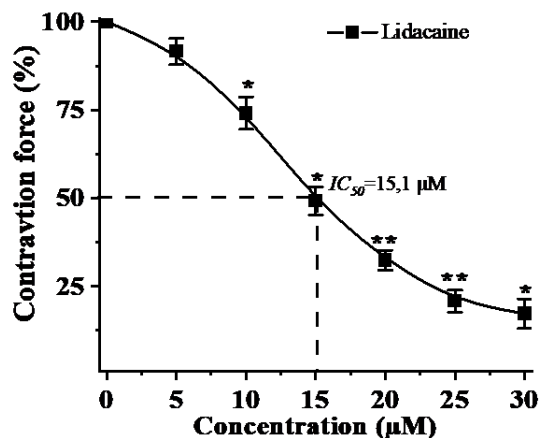


Figure 3 Dose-dependent effect of lidocaine on papillary muscle contractile activity. The ordinate axis shows the amplitude of the contraction force expressed as a percentage (%) of the maximum, and the abscissa axis shows the concentration of lidocaine (μM) (* $-p < 0.05$; ** $-p < 0.01$). The stimulation frequency is 1 Hz ($t = +36 \pm 0.5^\circ\text{C}$); $n = 5$).

To assess whether the DKV-8 conjugate, derived from the isoquinoline alkaloid F-36 and the flavonoid dihydroquercetin (DHQ), exerts a positive inotropic effect through an increase in intracellular Ca^{2+} concentration ($[\text{Ca}^{2+}]_i$) via activation of voltage-gated Na^+ channels, its effect on the contractile activity of rat cardiac papillary muscles was examined in the presence

of lidocaine, a selective Na^+ channel blocker with an IC₅₀ of $15.1 \mu\text{M}$. The results demonstrated that the F-36/DKV-8 conjugate enhanced papillary muscle contraction by $47.8 \pm 3.6\%$, $65.2 \pm 3.4\%$, and $89.3 \pm 3.9\%$, respectively, relative to the control (set at 100%) (Figure 4).

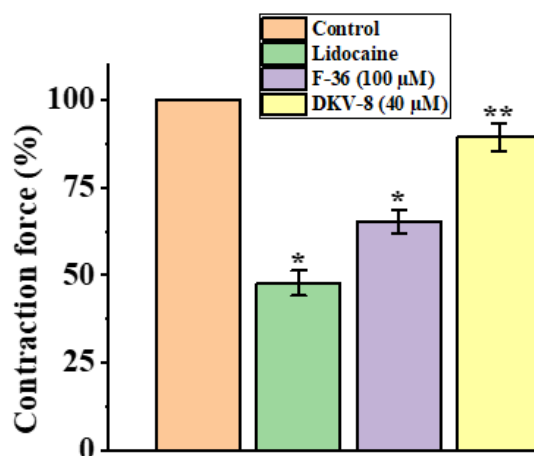


Figure 4 Effect of the DKV-8 conjugate, based on the isoquinoline alkaloid F-36 and the flavonoid DHQ, on the force of papillary muscle contraction in the presence of lidocaine in the medium. The ordinate axis shows the amplitude value of the contraction force, expressed as a percentage (%) of the maximum. In all cases * $-p < 0.05$; ** $-p < 0.01$; $n = 5$.

In this context, activation of Na^+ channels increases intracellular Na^+ levels in cardiomyocytes,

which, through the reverse mode of the $\text{Na}^+/\text{Ca}^{2+}$ exchanger, enhances Ca^{2+} influx into the cytosol [26-

28]. This can lead to an elevation of intracellular Ca^{2+} concentration ($[\text{Ca}^{2+}]_i$) and its storage in the sarcoplasmic reticulum (SR). These findings suggest that the effects of F-36 and DKV-8 on papillary muscle contractility are at least partially mediated via Na^+ channels.

Role of F-36 and DKV-8 in $\text{Na}^+/\text{Ca}^{2+}$ exchange

The $\text{Na}^+/\text{Ca}^{2+}$ exchanger in the heart primarily functions to extrude Ca^{2+} from cardiomyocytes during relaxation, thereby allowing Ca^{2+} entry through L-type Ca^{2+} channels during excitation [29]. The remaining Ca^{2+} ions are largely reabsorbed by SERCA2a. Under conditions of Na^+/K^+ -ATPase inhibition, the $\text{Na}^+/\text{Ca}^{2+}$

exchanger operates in reverse mode, which is considered important for increasing $[\text{Ca}^{2+}]_i$ and producing a positive inotropic effect.

Based on this, the subsequent experiments evaluated the effects of the tested compounds on $\text{Na}^+/\text{Ca}^{2+}$ exchange. In the presence of the non-specific $\text{Na}^+/\text{Ca}^{2+}$ exchanger blocker NiCl_2 (10 mM), the positive inotropic effects of F-36 (100 μM) and DKV-8 (40 μM) on papillary muscle twitch force were $69.4 \pm 3.8\%$, $134.3 \pm 3.9\%$, and $188.4 \pm 3.1\%$, respectively. These results indicate that the positive inotropic effect of the F-36 alkaloid is largely associated with significant modulation of $\text{Na}^+/\text{Ca}^{2+}$ exchange function (**Figure 5**).

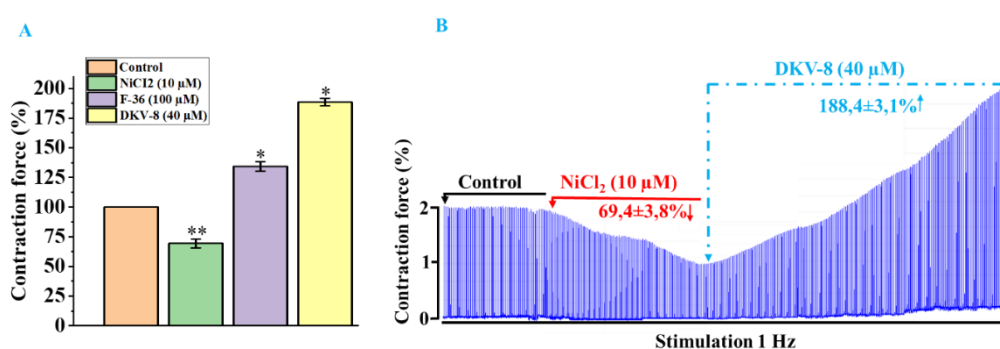


Figure 5 (A) Effect of F-36 and DKV-8 on papillary muscle contraction force in the presence of NiCl_2 . (B) Under these conditions. The ordinate axis shows the amplitude value of the contraction force, expressed as a percentage (%) of the maximum. In all cases * $-p < 0.05$; ** $-p < 0.01$; $n = 5$.

Based on the results of the studies conducted, it was determined that the positive inotropic effect of the isoquinoline alkaloid F-36 and DKV-8 conjugates does not involve $\text{Na}^+/\text{Ca}^{2+}$ exchange.

Effect of F-36 and DKV-8 on L-type Ca^{2+} channels

In subsequent experiments, L-type Ca^{2+} channels, which are widely distributed in cardiomyocytes, were confirmed as the primary pathway for Ca^{2+} entry into the cell [30]. Based on this, the potential involvement of L-type Ca^{2+} channels in the sarcolemma of cardiomyocytes in mediating the positive inotropic effects of the studied

biologically active compounds was evaluated using nifedipine, a selective L-type Ca^{2+} channel blocker.

Preliminary experiments examined the dose-dependent effects of nifedipine on papillary muscle contractility, revealing a half-maximal inhibitory concentration IC_{50} of 0.01 μM . In subsequent experiments, in the presence of nifedipine (0.01 μM) in the incubation medium, the positive inotropic effects of F-36 and DKV-8 on papillary muscle contraction were $52.4 \pm 3.8\%$, $91.9 \pm 3.9\%$, and $131.4 \pm 4.2\%$, respectively, relative to the control (**Figures 6(A) and 6(B)**).

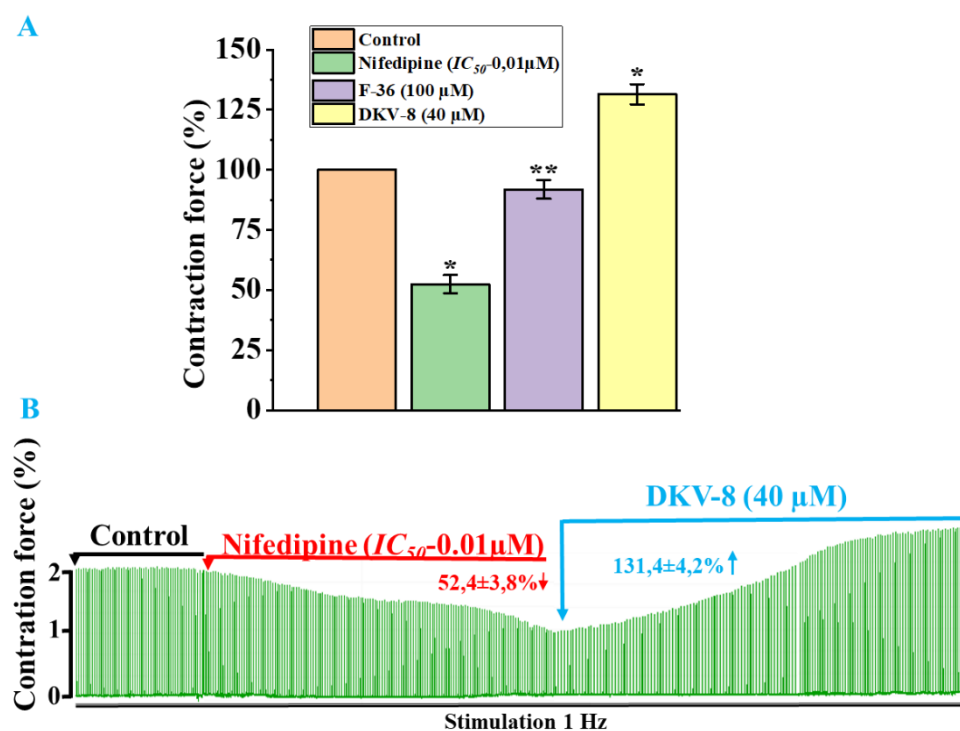


Figure 6 (A) Effect of the F-36 and DKV-8 on the force of papillary muscle contraction in the presence of nifedipine in the medium. The ordinate axis shows the amplitude of the contraction force, expressed as a percentage (%) of the maximum. In all cases, * $-p < 0.05$; ** $-p < 0.01$; $n = 5$.

Based on the results of experiments assessing the effects of the studied biologically active compounds on L-type Ca^{2+} channels in the presence of nifedipine, it was found that the positive inotropic effects of F-36 and DKV-8 on papillary muscle contractility were partially enhanced compared to the control. These findings indicate that the positive inotropic effects of F-36 and DKV-8 involve L-type Ca^{2+} channels in cardiomyocytes.

Effect of F-36 and DKV-8 on RyR2 activity

In the next series of experiments, the effects of the isoquinoline alkaloid F-36 and the conjugate DKV-8 on RyR2 activity were investigated. A method assessing sarcoplasmic reticulum Ca^{2+} ($[Ca^{2+}]_{SR}$) levels was employed, in which the RyR2 activator caffeine (20 μ M) induced a single contraction in unstimulated papillary muscles [31]. Under these conditions, no post-rest potentiation was observed during a 30-second pause after ~15 minutes, which is attributed to the complete release of $[Ca^{2+}]_{SR}$ into the cytosol by caffeine. In this

setup, caffeine (20 μ M) evokes a phasic single contraction by triggering Ca^{2+} release from the SR via RyR2 under unstimulated conditions [32].

The caffeine-induced increase in intracellular Ca^{2+} concentration ($[Ca^{2+}]_i$) is mediated by the Na^+/Ca^{2+} exchanger [33,34]. When the extracellular Na^+ concentration ($[Na^+]_{out}$) is set to 0, the Na^+/Ca^{2+} exchanger is blocked, preventing Ca^{2+} extrusion from the cell. This maintains stable $[Ca^{2+}]_i$ levels and contraction force amplitude in response to caffeine (20 μ M) [35,36].

Under these conditions ($[Na^+]_{out} = 0$), a single caffeine-induced contraction of papillary muscle in the unstimulated state increased the contraction force amplitude by $37.6 \pm 5.3\%$ relative to the control. In the presence of F-36 (100 μ M) and DKV-8 (40 μ M) in the incubation medium, the amplitude of caffeine-induced single contractions increased further to $115.4 \pm 3.6\%$ and $141.1 \pm 3.1\%$, respectively, compared to control (caffeine-only control set at 100%) (Figure 7).

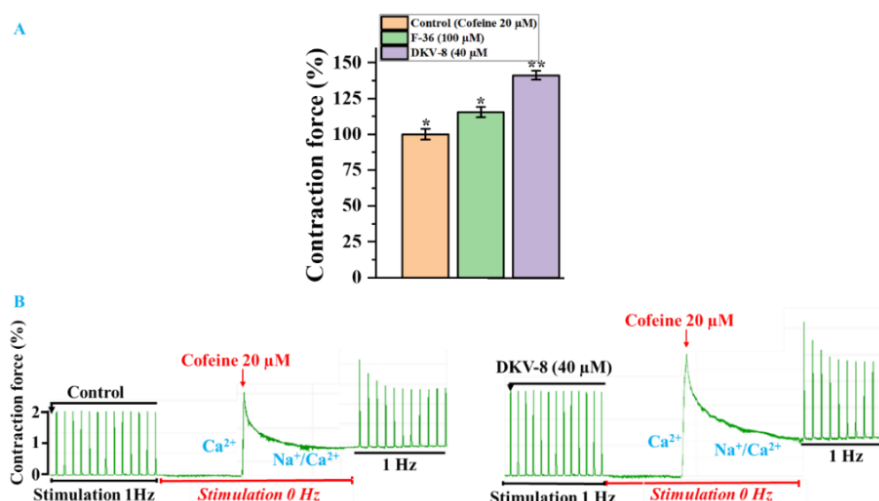


Figure 7 (A) Inotropic effect of the F-36 and DKV-8 under the conditions of incubation with the RyR2 activator - caffeine. (B) Under these conditions. The ordinate axis shows the contraction force of the papillary muscle, expressed as a percentage of the maximum value, taken as 100%. The stimulation frequency is 1 Hz ($t = +36 \pm 0.5$ °C). * $-p < 0.05$, ** $-p < 0.01$, ($n = 5$).

The results indicate that the positive inotropic effects of the studied compounds are associated with an increase in cytosolic Ca^{2+} concentration. These compounds significantly enhance the amplitude of single papillary muscle contractions induced by caffeine under unstimulated conditions.

To further investigate these effects, the influence of F-36 and DKV-8 on sarcoplasmic reticulum (SR) Ca^{2+} dynamics was analyzed using the rapid-cooling contraction (RCC) method. In this approach, cardiomyocytes are exposed to rapid cooling of the incubation medium ($+37 \rightarrow +2$ °C) for 30 s without

electrical stimulation [37,38]. RCC is associated with increased $[\text{Ca}^{2+}]_i$ via RyR2 activation and allows assessment of both SR Ca^{2+} dynamics and changes in myofilament Ca^{2+} sensitivity. The “peak” observed as a rapid rise in contractile force upon rewarming of the incubation medium ($+2 \rightarrow +37$ °C) reflects enhanced myofilament Ca^{2+} sensitivity [39,40]. In the experiments, the presence of F-36 (100 µM) and DKV-8 (40 µM) increased the contraction force under rapid cooling by $21.6 \pm 3.3\%$ and $28.7 \pm 3.1\%$, respectively, compared to control (control RCC = $16.4 \pm 3.8\%$) (Figure 8).

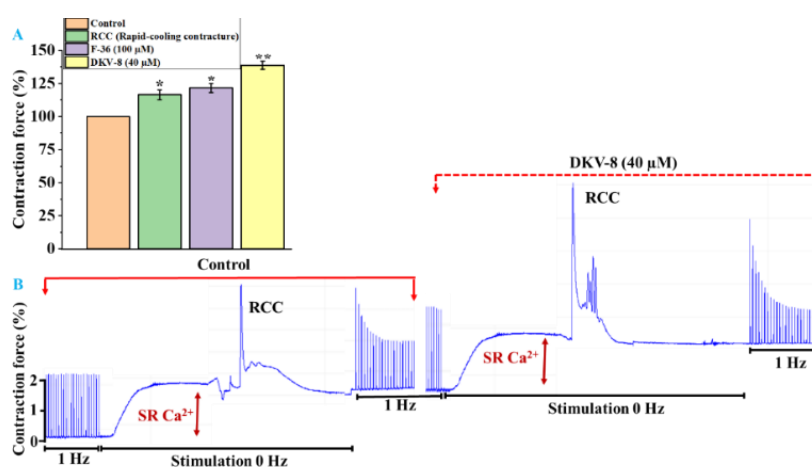


Figure 8 (A) Effect of the F-36 alkaloid and DKV-8 conjugate on papillary muscle contraction induced by rapid cooling of a myocardial preparation. (B) Under these conditions. Temperature gradient from $+37$ to $+2$ °C. The ordinate axis shows the force of contraction of the papillary muscle expressed as a percentage of the maximum value, taken as 100% ($t = +36 \pm 0.5$ °C). * $-p < 0.05$, ** $-p < 0.01$ ($n = 5$).

The participation of SR Ca^{2+} transport systems plays a key role in Ca^{2+} homeostasis in cardiac muscle cells. Based on the results of the above study, we can conclude that the positive inotropic effect of the F-36 and the DKV-8 on rat heart papillary muscle contractile activity is very low, indicating that RyR_2 is involved.

Effect of F-36 and DKV-8 on SERCA2a

In a series of experiments, the effects of biologically active compounds on SERCA2a activity were investigated. The sarcoplasmic reticulum SERCA2a plays a critical role in regulating intracellular Ca^{2+} concentration, maintaining Ca^{2+} homeostasis, and facilitating cardiac muscle relaxation by mediating Ca^{2+} uptake into the SR [41,42]. Based on the preceding

experiments, the positive inotropic effects of F-36 and DKV-8 may be related to modulation of Ca^{2+} accumulation processes in the SR. To test this hypothesis, the impact of these compounds on SERCA2a was examined.

Initially, experiments were conducted using CPA, a SERCA2a inhibitor that disrupts SR Ca^{2+} uptake [43,44]. The dose-dependent effects of CPA on papillary muscle contractility were evaluated at a stimulation frequency of 1 Hz. CPA exhibited a negative inotropic effect, reducing papillary muscle contraction force by $84.2 \pm 3.6\%$ at a concentration of $15 \mu\text{M}$. The half-maximal inhibitory concentration IC_{50} of CPA on papillary muscle contraction was determined to be $5.6 \mu\text{M}$ (Figure 9).

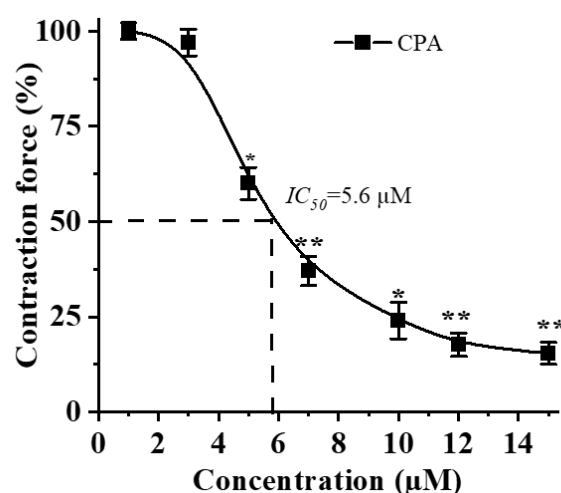


Figure 9 Dose-dependent effect of CPA on papillary muscle contractile activity. The ordinate axis shows the amplitude of the contraction force expressed as a percentage (%) of the maximum, and the abscissa axis shows the concentration of CPA (μM) (* $-p < 0.05$; ** $-p < 0.01$). Stimulation frequency 1 Hz $t = +36 \pm 0.5^\circ\text{C}$; $n = 5$.

In subsequent experiments, the effects of F-36 and DKV-8 on papillary muscle contractile force were evaluated in the presence of the SERCA2a inhibitor CPA at its half-maximal inhibitory concentration (IC_{50}

$= 5.6 \mu\text{M}$). Under these conditions, the amplitude of rat papillary muscle contraction in the presence of CPA ($5.6 \mu\text{M}$) increased to $49.8 \pm 3.4\%$, $76.4 \pm 3.3\%$, and $92.6 \pm 2.2\%$, respectively, relative to the control (Figure 10).

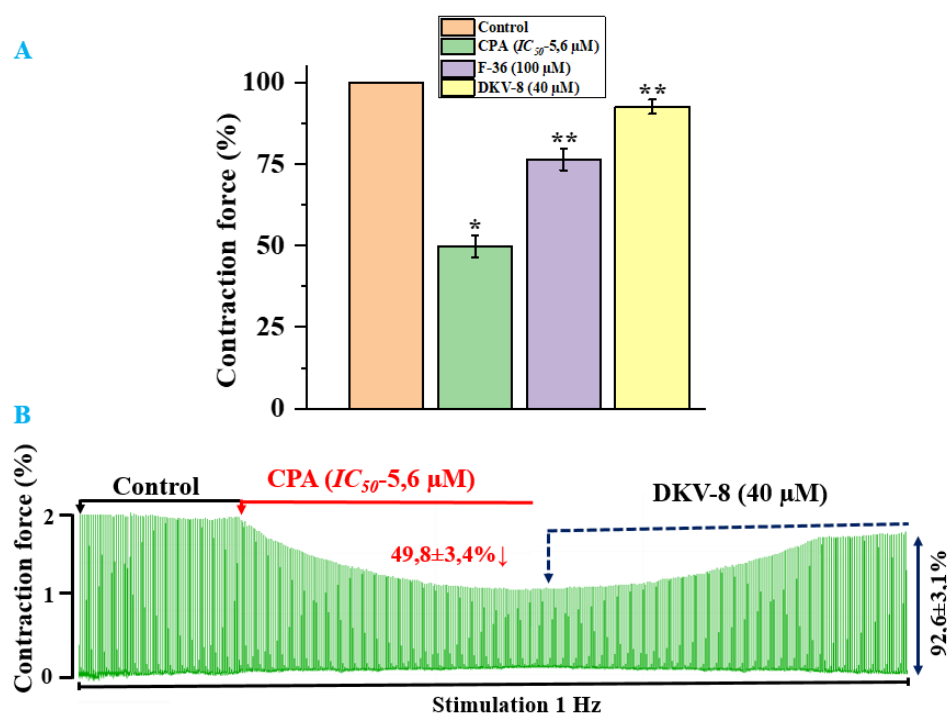


Figure 10 (A) Effect of the F-36 and the DKV-8 on the force of papillary muscle contraction in the presence of the Ca^{2+} -ATPase blocker CPA (IC_{50} 5.6 μ M) in the incubation medium. B. Under these conditions. On the ordinate axis - the force of contraction of the papillary muscle expressed as a percentage (%). Stimulation frequency 1 Hz ($t = +36 \pm 0.5$ °C; – * $-p < 0.05$; ** $-p < 0.01$; $n = 5$).

From the results of this experiment, it can be concluded that the positive inotropic effect of the F-36 and DKV-8 indicates that an increase in the force of muscle contraction is provided due to the additional accumulation of Ca^{2+} ions in the SR through SERCA2a and their release through RyR2.

***In-vitro* hypoxia experiments**

The development of hypoxia caused by impaired blood supply to the heart is accompanied by a significant reduction in ATP and creatine phosphate production in cardiomyocytes. These compounds serve as the primary energy sources that support the activity of several

membrane ion transport systems, including Na^+/K^+ -ATPase and Na^+, K^+ -ATP-dependent ion pumps [45,46]. As a consequence, the transport and homeostasis of Na^+ , K^+ , and Ca^{2+} ions in cardiomyocytes become disrupted, leading to disturbances in electrical activity and, ultimately, a decline in myocardial contractile function [47]

In subsequent experiments conducted under hypoxic conditions, a marked reduction in the contractile force amplitude of rat heart papillary muscles was observed. Specifically, the contraction force decreased by $69.4 \pm 3.8\%$ compared with the control (Figure 11).

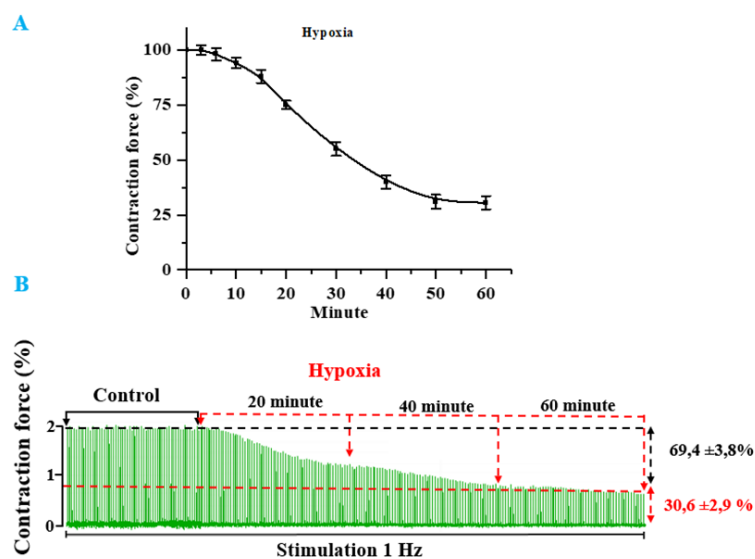


Figure 11 Effect of hypoxia on rat heart papillary muscle contractile activity. On the ordinate axis - muscle contraction force, expressed as a percentage of the control obtained under normal oxygenation of physiological solution and taken as 100%. The abscissa axis shows the deoxygenation time of the nitrogen-perfused solution. The drug was stimulated with a frequency of 1 Gs. In all cases, * $-p < 0.05$; ** $-p < 0.01$, (n = 5).

Since the force of papillary muscle contraction is largely mediated by Ca^{2+} ions, the results of these experiments suggest that the reduction in contractile force observed under hypoxic conditions is associated with a decrease in intracellular Ca^{2+} concentration ($[\text{Ca}^{2+}]_i$) [48,49].

In subsequent experiments, the effects of the studied compounds were evaluated under hypoxic conditions. It was found that F-36 and DKV-8 significantly increased the contractile force of papillary muscles under hypoxia by $59.9 \pm 4.9\%$ and $97.4 \pm 3.2\%$, respectively, compared with the control (**Figure 12**).

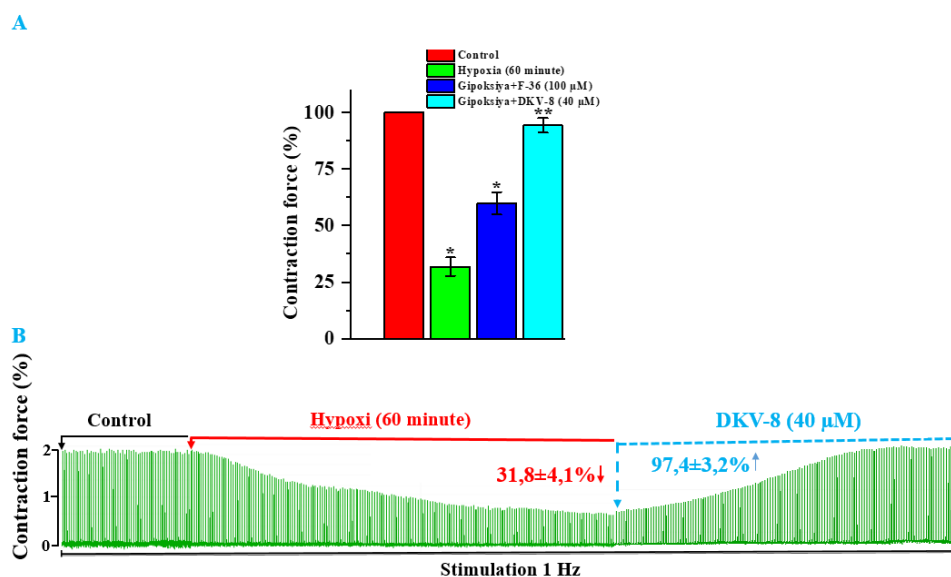


Figure 12 Effect of DKV-8 and F-36 on rat heart papillary muscle contractile activity under hypoxic conditions. On the ordinate axis - the muscle's contraction force is given in percentage. Stimulation frequency 1 Hz ($t = +36 \pm 0.5^\circ\text{C}$). * $-p < 0.05$, ** $-p < 0.01$, (n = 5).

The results of the study demonstrated that F-36 and DKV-8 effectively attenuate disturbances in papillary muscle contractile activity under hypoxic conditions. Additional evidence supporting the involvement of this mechanism in the positive inotropic effects of the studied compounds was obtained from experiments performed under conditions of L-type Ca^{2+} channel blockade with nifedipine. In the hypoxia model,

incubation with nifedipine ($\text{IC}_{50} = 0.01 \mu\text{M}$) resulted in a marked reduction in papillary muscle contractile force to $17.8 \pm 4.1\%$ compared with the control. Under these conditions, the administration of the isoquinoline alkaloid F-36 and the conjugate DKV-8 increased papillary muscle contraction force to $49.9 \pm 4.9\%$ and $74.4 \pm 3.2\%$, respectively, relative to the control (**Figure 13**).

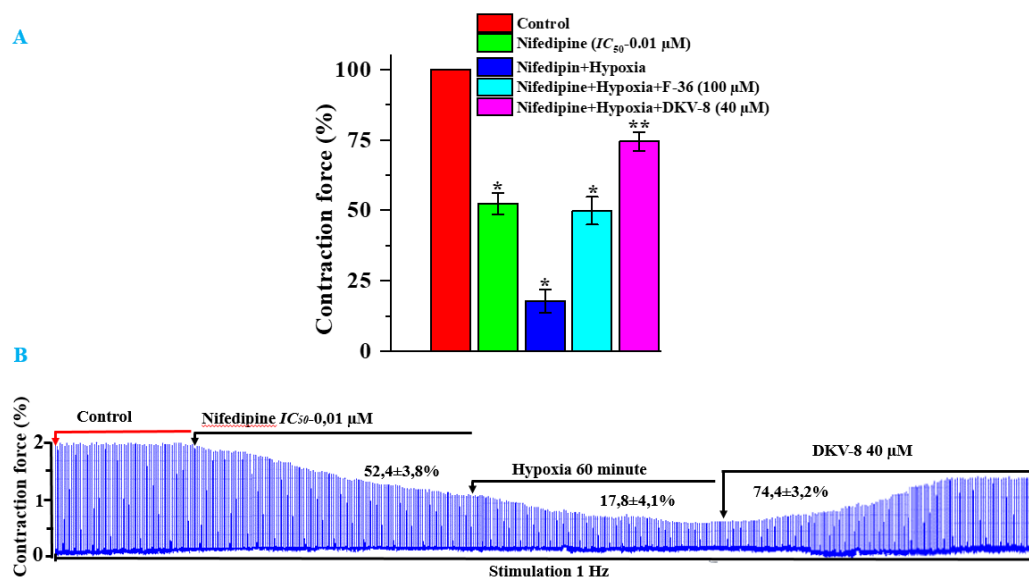


Figure 13 (A) Evaluation of the involvement of L-type Ca^{2+} channels in the contractile activity of rat heart papillary muscles under hypoxic conditions by F-36 and DKV-8. (B) The ordinate axis shows the amplitude value of the contraction force, expressed as a percentage (%) of the maximum. The frequency of drug excitation was 1 Hz. In all cases * $-p < 0.05$, ** $-p < 0.01$; $n = 5$.

These findings indicate that the cardioprotective effects of F-36 and DKV-8 under hypoxic conditions are associated, at least in part, with the involvement of L-type Ca^{2+} channels. Further experiments were conducted to evaluate the effects of the studied biologically active compounds on SERCA2a activity in myocardial cells under hypoxia. Under these conditions, the effects of the isoquinoline alkaloid F-36 (100 μM) and the conjugate

DKV-8 (40 μM) were examined in the presence of the SERCA2a inhibitor CPA; $\text{IC}_{50} = 5.6 \mu\text{M}$. It was found that the contraction force of papillary muscles increased by $28 \pm 4.6\%$ and $39 \pm 3.4\%$, respectively, compared with the control (where the amplitude of papillary muscle contraction under control hypoxia conditions was 19.2%) (**Figure 14**).

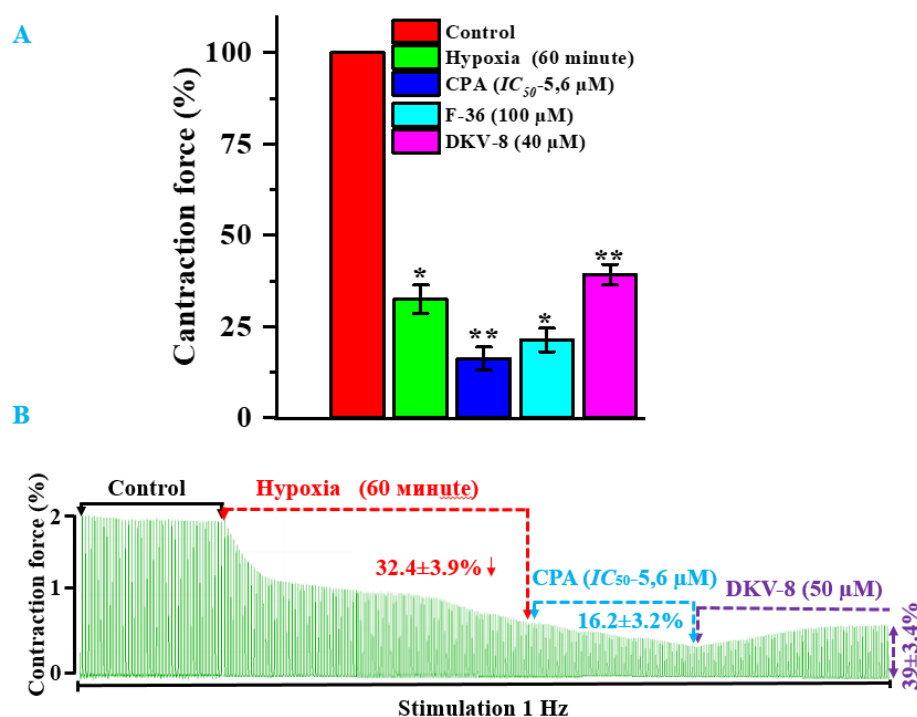


Figure 14 (A) Effect of F-36 and DKV-8 on the force of papillary muscle contraction in the presence of CPA (IC_{50} 5.6 μ M) under hypoxic conditions. (B) Under these conditions. On the ordinate axis - the force of contraction of the papillary muscle, expressed as a percentage (%). Stimulation frequency 1 Hz ($t = +36 \pm 0.5$ °C; * $-p < 0.05$; ** $-p < 0.01$; $n = 5$).

Analysis of the obtained results suggests that the positive inotropic effect on papillary muscle contractile activity, as well as the increase in Ca^{2+} content in the SR, is associated with modulation of the SERCA2a system in cardiomyocytes.

Discussion

The results of this study demonstrate that the conjugate of the isoquinoline alkaloid F-36 and DKV-8 produces a strong and dose-dependent positive inotropic effect on rat cardiac papillary muscle. The action of these compounds appears to be mediated by the simultaneous modulation of several systems involved in the regulation of Ca^{2+} homeostasis in cardiomyocytes. The findings indicate that both Na^+ channels and L-type Ca^{2+} channels participate in the positive inotropic effect of the studied compounds [50]. The direct interaction of F-36 and DKV-8 with plasma membrane ion channels may also contribute to a reduced risk of cardiac arrhythmias.

In the presence of the SERCA2a inhibitor CPA, the inotropic effect of these compounds was markedly

attenuated, suggesting a key role of the SERCA2a system in their mechanism of action. This property may be particularly relevant for improving pathological conditions associated with decreased SERCA2a activity in heart failure. At the same time, the experimental results indicate that the Na^+/Ca^{2+} exchanger (NCX) does not play a major role in this mechanism. The preservation of the effect in the presence of $NiCl_2$ suggests that the principal pathway of action of F-36 and DKV-8 is not mediated by NCX, but rather through Na^+ channels, L-type Ca^{2+} channels, and SERCA2a.

Furthermore, the results obtained from caffeine-induced contraction and RCC experiments, which were used to evaluate the role of RyR2 in cardiomyocytes, indicate that these compounds do not cause excessive Ca^{2+} release from the SR. This finding is important because uncontrolled Ca^{2+} release can be harmful under pathological conditions. Therefore, the absence of excessive SR Ca^{2+} release suggests that F-36 and DKV-8 may have a lower arrhythmogenic potential.

The results of the present study demonstrated that the conjugate of the isoquinoline alkaloid F-36 and

DKV-8 significantly restored the acute decrease in papillary muscle contractile force observed under in vitro hypoxic conditions. Under hypoxia, the contraction force of papillary muscles decreased by $69.4 \pm 3.8\%$, whereas the administration of DKV-8 increased contractile activity by $97.4 \pm 3.2\%$. These findings clearly indicate that the studied compounds possess pronounced cardioprotective properties and can preserve myocardial functional activity under conditions of hypoxic injury.

Furthermore, even under hypoxic conditions, the effects of the L-type Ca^{2+} channel blocker nifedipine and the SERCA2a CPA remained significant in the incubation medium [51]. Under hypoxia, depletion of ATP leads to inhibition of Na^+/K^+ -ATPase activity, F-36 and DKV-8 appear to enhance SERCA2a activity, facilitating efficient reuptake of Ca^{2+} into the sarcoplasmic reticulum, thereby reducing excessive cytosolic Ca^{2+} accumulation and maintaining intracellular Ca^{2+} homeostasis.

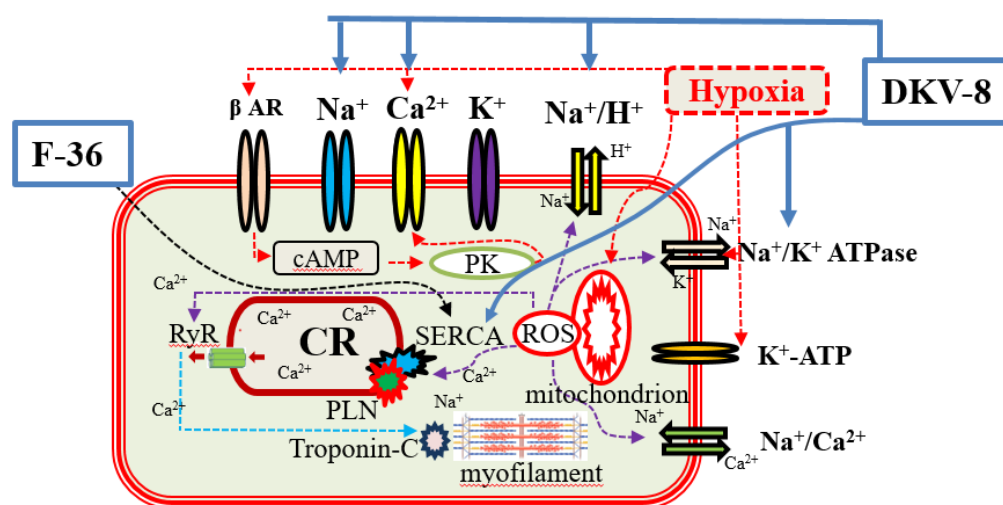


Figure 15 The effect of biologically active substances on membrane channels.

Conclusions

The combined mechanism of action of the isoquinoline alkaloid F-36 and the DKV-8 conjugate—characterized by the simultaneous activation of Na^+ and Ca^{2+} channels together with stimulation of SERCA2a activity—creates an optimal synergistic inotropic effect. This mechanism may not only enhance myocardial contractile force but also improve diastolic relaxation through more efficient Ca^{2+} reuptake into the SR, which represents an important pathological determinant in heart failure.

Furthermore, it was demonstrated that the conjugate of the F-36 alkaloid and DKV-8 exhibits a strong positive inotropic effect and pronounced cardioprotective activity under conditions of hypoxia/ischemia. The primary mechanism underlying these effects appears to involve the regulation of Ca^{2+} homeostasis in myocardial cells, particularly through modulation of L-type Ca^{2+} channels and SERCA2a activity in the sarcoplasmic reticulum.

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Ethics approval

The experimental protocols complied with the standards and requirements for the humane treatment of animals and the provisions of the Ethical Commission of the IBB at the National University of Uzbekistan. (Protocol No. 7 BEC/IBB-NUU of 04/07/2022) on the use of laboratory animals. Isolated cardiac papillary muscle preparations were obtained using a specific method.

Declaration of Generative AI in Scientific Writing

Minimal assistance from QuillBot was used only for paraphrasing selected phrases, while all scientific information, interpretations, and conclusions were independently developed by the authors.

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Zhurakulov Sherzod Niyatkobulovich: Isoquinoline alkaloid 6,7-dimetoksi-1,2,3,4-tetragidroizoxinolin gidrokslorid (F-36) va digidrokversetin flavonoidi asosida sintezlangan 2-(3,4-Digidroksifenil)-6-((6,7-dimetoksi-1,2,3,4-tetragidroizo-xinolin-2(1N)-il)metil)-3,5,7-trigidroksixroman-4-on (DKV-8).
Adilbay Tlepovich Esimbetov: Visualization, Resources.
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References

- [1] VR Netala, SK Teertam, H Li and AZ Zhang. Comprehensive review of cardiovascular disease management: Cardiac biomarkers, imaging modalities, pharmacotherapy, surgical interventions, and herbal remedies. *Cells* 2024; **13(17)**, 1471.
- [2] ND Kolanu, ZR Syeda, N Joshi, P Singh and M Erukulla. The differential impact of medical therapy and lifestyle modification on cardiovascular health and risk of adverse cardiovascular events: A narrative review. *Cureus* 2024; **16(4)**, e57742.
- [3] IZ Jumaev, SN Boboev, PB Usmonov, SY Rustamov, AA Zaripov, SB Qurbonova, YT Mirzaeva, EB Ibragimov, AT Esimbetov and SM Adizov. Role of RyR2 and SERCA2a in the cardioprotective effects of vincanine and pyrazoline alkaloids. *Trends in Sciences* 2024; **22(1)**, 8626.
- [4] VV Uzbekov, BF Abdullaev, IZ Jumayev, YI Oshchepkova, PB Usmanov and SI Salikhov. Comparative study of the antiarrhythmic activity of liposomal forms of lappaconitine hydrobromide and its complex with glycyrrhizic acid monoammonium salt in the aconitine arrhythmia model. *Pharmaceutical Chemistry Journal* 2023; **56**, 1327-1332.
- [5] YI Oshchepkova, VV Uzbekov, IZ Jumayev, SY Rustamov, PB Usmanov and SI Salikhov. Antiarrhythmic activity of ethacizine hydrochloride and its supramolecular complexes with glycyrrhizic acid and its monoammonium salt. *Pharmaceutical Chemistry Journal* 2025; **59(6)**, 21-26.
- [6] YI Oshchepkova, VV Uzbekov, IZ Jumayev, SY Rustamov, PB Usmanov and SI Salikhov. Comparative study of antiarrhythmic and inotropic activity of amiodarone hydrochloride and its complexes with glycyrrhizic acid and monoammonium salt of glycyrrhizic acid. *Ekspierimental'Naya i Klinicheskaya Farmakologiya* 2023; **86(4)**, 15-22.
- [7] PB Usmonov, IZ Jumayev, SY Rustamov, AA Zaripov, AT Esimbetov, SN Juraqulov and VI Vinogradova. The combined inotropic and vasorelaxant effect of DHQ-11, a conjugate of flavonoid dihydroquercetin with isoquinoline alkaloid 1-aryl-6,7-dimethoxy-1,2,3,4-tetrahydroisoquinoline. *Biomedical and Pharmacology Journal* 2021; **14(2)**, 651-661.
- [8] SB Qurbonova, IZ Zhumaev, SN Boboev, PB Usmanov, SY Rustamov, AA Zaripov and SN Zhurakulov. Antiarrhythmic effect of 1-(3'-Bromophenyl)-6,7-methylenedioxy-1,2,3,4-tetrahydroisoquinoline hydrochloride. *Biomedical and Pharmacology Journal* 2025; **18(3)**, 2463-2473.

- [9] AA Zaripov, IZ Zhumaev, PB Usmanov, YT Mirzaeva, SY Rustamov, AT Esimbetov, GS Begdullaeva, SB Sobirov, SN Zhurakulov, DD Safarova and RI Yusupova. Mechanism of vasorelaxant action of isoquinoline alkaloid F-19 on rat aorta. *Biomedical and Pharmacology Journal* 2025; **18(2)**, 1580-1589.
- [10] AA Zaripov, IZ Jumayev, PB Usmanov, YT Mirzaeva, AT Esimbetov, SY Rustamov, SN Boboev, EB Ibragimov, SB Qurbonova and SN Zhurakulov. Protective effect of DHQ-11 against hypoxia-induced vasorelaxation. *Trends in Sciences* 2024; **21(11)**, 8192.
- [11] R Bagur and G Hajnoczky. Intracellular Ca^{2+} sensing: Its role in calcium homeostasis and signaling. *Molecular Cell* 2017; **66(6)**, 780-788.
- [12] L Fliegel. Molecular biology of the myocardial Na^+/H^+ exchanger. *Journal of Molecular and Cellular Cardiology* 2008; **44(2)**, 228-237.
- [13] S Wakabayashi, T Hisamitsu and TY Nakamura. Regulation of the cardiac Na^+/H^+ exchanger in health and disease. *Journal of Molecular and Cellular Cardiology* 2013; **61**, 68-76.
- [14] DS Escudero, NG Perez and RG Diaz. Myocardial impact of NHE1 regulation by sildenafil. *Frontiers in Cardiovascular Medicine* 2021; **8**, 617519.
- [15] H Sutanto, A Lyon, J Lumens, U Schotten, D Dobrev and J Heijman. Cardiomyocyte calcium handling in health and disease: Insights from in vitro and in silico studies. *Progress in Biophysics and Molecular Biology* 2020; **157**, 54-75.
- [16] AL Denniss, AM Dashwood, P Molenaar and NA Beard. Sarcoplasmic reticulum calcium mishandling: Central tenet in heart failure? *Biophysics Reviews* 2020; **12(4)**, 865-878.
- [17] A Agrawal, G Suryakumar and R Rathor. Role of defective Ca^{2+} signaling in skeletal muscle weakness: Pharmacological implications. *Journal of Cell Communication and Signaling* 2018; **12(4)**, 645-659.
- [18] EB Ibragimov, IZ Zhumaev, AA Zaripov, PB Usmanov, SY Rustamov, AT Esimbetov, SM Adizov and AK Kurbanov. Mechanism of vasorelaxant effect of the indole alkaloid 12-hydroxynorfluorocurarine hydrochloride on aortic smooth muscle contraction. *Journal of Applied Pharmaceutical Science* 2026; **16(3)**, 283-291.
- [19] SN Zhurakulov, VA Babkin, EI Chernyak, SV Morozov, IA Grigoryev, MG Levkovich and VI Vinogradova. Aminomethylation of 1-aryl-6,7-dimethoxy-1,2,3,4-tetrahydroisoquinolines by dihydroquercetin. *Chemistry of Natural Compounds* 2015; **51(1)**, 57-61.
- [20] S Despa and DM Bers. Na^+ transport in the normal and failing heart - remember the balance. *Journal of Molecular and Cellular Cardiology* 2013; **61**, 2-10.
- [21] ZF Ziyavitdinov, UZ Ishimov, NS Berdiev, IZ Zhumaev, YI Oshchepkova, PB Usmanov, and SI Salikhov. Supramolecular complex of lappaconitine hydrobromide and the monoammonium salt of glycyrrhizic acid: Synthesis, physicochemical characteristics, and antiarrhythmic activity. *Pharmaceutical Chemistry Journal* 2022; **56(2)**, 167.
- [22] KR DeMarco and CE Clancy. Cardiac Na channels: Structure to function. *Current Topics in Membranes* 2016; **78**, 287-311.
- [23] M Abdelsayed and C Antzelevitch. From beat to beat: How electrolytes shape the heart's rhythmic symphony and structure. *Journal of Cardiology and Cardiovascular Medicine* 2025; **10(3)**, 070-088.
- [24] IZ Zhumaev, SN Boboev, PB Usmanov, SB Qurbonova, SY Rustamov, AT Esimbetov, GS Begdullaeva, AA Zaripov, SM Adizov. Mechanism of positive inotropic effect of vincanine on cardiac muscle contraction activity. *Biomed Pharmacol J* 2022; **15(4)**, 2309-2316.
- [25] DM Bers and S Despa. Na^+ transport in cardiac myocytes; Implications for excitation-contraction coupling. *IUBMB Life* 2009; **61(3)**, 215-221.
- [26] DA Eisner. Ups and downs of calcium in the heart. *Journal of Physiology* 2018; **596(1)**, 19-30.
- [27] I Jumayev, P Usmanov, S Rustamov and S Zhurakulov. Comparative inotropic effects of the some isoquinoline alkaloids. *Biomedical and Pharmacology Journal* 2020; **13(1)**, 325-333.
- [28] J Satin and EA Schroder. Autoregulation of cardiac L-type calcium channels. *Trends in Cardiovascular Medicine* 2009; **19(8)**, 268-271.
- [29] MB Bhat, J Zhao, W Zang, CW Balke, H Takeshima, WG Wier and J Ma. Caffeine-induced release of intracellular Ca^{2+} from Chinese hamster

- ovary cells expressing skeletal muscle ryanodine receptor: Effects on full-length and carboxyl-terminal portion of Ca^{2+} release channels. *Journal of General Physiology* 1997; **110(6)**, 749-762.
- [30] A Belevych, Z Kubalova, D Terentyev, RL Hamlin, CA Carnes and S Gyorke. Enhanced ryanodine receptor-mediated calcium leak determines reduced sarcoplasmic reticulum calcium content in chronic canine heart failure. *Biophysical Journal* 2007; **93(11)**, 4083-4092.
- [31] KA Rakhmanova, SN Zhurakulov, FM Tursunkhodjayeva, AA Azamatov, DM Saidkhodjayeva, IZ Jumayev. Analgesic and Anti-Inflammatory Effects of 1-(4'-Dimethylaminophenyl)-6, 7-Dimethoxy-1,2,3,4-Tetrahydroisoquinoline Hydrochloride. *Biomed Pharmacol J* 2022; **15(2)**, 891-895
- [32] M Karmazyn, XT Gan, RA Humphreys, H Yoshida and K Kusumoto. The myocardial Na^+ - H^+ exchange. *Circulation Research* 1999; **85**, 777-786.
- [33] KD Dwyer, CA Snyder and KKK Coulombe. Cardiomyocytes in hypoxia: Cellular responses and implications for cell-based cardiac regenerative therapies. *Bioengineering* 2025; **12(2)**, 154.
- [34] C Leoty, C Huchet-Cadiou, S Talon, S Choisy and W Hleihel. Caffeine stimulates the reverse mode of $\text{Na}^+/\text{Ca}^{2+}$ exchanger in ferret ventricular muscle. *Acta Physiologica Scandinavica* 2001; **172(1)**, 27-37.
- [35] KN Bradley, ER Flynn, TC Muir and JG McCarron. Ca^{2+} regulation in guinea-pig colonic smooth muscle: The role of the $\text{Na}^+/\text{Ca}^{2+}$ exchanger and the sarcoplasmic reticulum. *Journal of Physiology* 2002; **538(2)**, 465-482.
- [36] K Scranton, S John, M Angelini, F Steccanella, S Umar, R Zhang, JI Goldhaber, R Olcese and M Ottolia. Cardiac function is regulated by the sodium-dependent inhibition of the sodium-calcium exchanger NCX1. *Nature Communications* 2024; **15(1)**, 3831.
- [37] M Shigekawa and T Iwamoto. Cardiac $\text{Na}^+/\text{Ca}^{2+}$ exchange molecular and pharmacological aspects. *Circulation Research* 2001; **88(9)**, 864-876.
- [38] S Kurihara and T Sakai. Effects of rapid cooling on mechanical and electrical responses in ventricular muscle of guinea-pig. *Journal of Physiology* 1985; **361**, 361-378.
- [39] DM Bers, JH Bridge and KW Spitzer. Intracellular Ca^{2+} transients during rapid cooling contractures in guinea-pig ventricular myocytes. *Journal of Physiology* 1989; **417**, 537-553.
- [40] JA Lahorra, DF Torchiana, GT Jr, CA Bashour, C Hahn, JS Titus, GA Geffin and WM Daggett. Rapid cooling contracture with cold cardioplegia. *The Annals of Thoracic Surgery* 1997; **63(5)**, 1353-1360.
- [41] S Huke and BC Knollmann. Increased myofilament Ca^{2+} -sensitivity and arrhythmia susceptibility. *Journal of Molecular and Cellular Cardiology* 2010; **48(5)**, 824-833.
- [42] M Periasamy, P Bhupathy and GJ Babu. Regulation of sarcoplasmic reticulum Ca^{2+} ATPase pump expression and its relevance to cardiac muscle physiology and pathology. *Cardiovascular Research* 2008; **77(2)**, 265-273.
- [43] KF Frank, B Bolck, E Erdmann and RH Schwinger. Sarcoplasmic reticulum Ca^{2+} -ATPase modulates cardiac contraction and relaxation. *Cardiovascular Research* 2003; **57(1)**, 20-27.
- [44] Y Uyama, Y Imaizumi and M Watanabe. Cyclopiazonic acid, an inhibitor of Ca^{2+} -ATPase in sarcoplasmic reticulum, increases excitability in ileal smooth muscle. *British Journal of Pharmacology* 1993; **110(2)**, 565-572.
- [45] SN Boboev, IZ Zhumaev, PB Usmanov, AA Zaripov, SY Rustamov, SB Qurbonova, EB Ibragimov, SB Sobirov, SN Zhurakulov. Role of F-4 Isoquinoline Alkaloid, Dihydroquercetin Flavonoid and Conjugate DKV-6 in Myocardial RyR2 and SERCA2a under Normal and Hypoxic Conditions. *Biomed Pharmacol J* 2026; **19(1)**, 578-591
- [46] MB Sikkell, C Hayward, KT MacLeod, SE Harding and AR Lyon. SERCA2a gene therapy in heart failure: An anti-arrhythmic positive inotrope. *British Journal of Pharmacology* 2014; **171(1)**, 38-54.
- [47] MJ Taggart and S Wray. Hypoxia and smooth muscle function: Key regulatory events during metabolic stress. *Journal of Physiology* 1998; **509(2)**, 315-325.

- [48] ShS Khushmatov, IZ Zhumaev, SN Zhurakulov, ASh Saidov, and VI Vinogradova. Synthesis And Comparative Inotropic Effects Of Several Isoquinoline Alkaloids. *Pharmaceutical Chemistry Journal* 2020 **54(1)**, 9-13.
- [49] L Santacruz, AJL Arciniegas, M Darrabie, JG Mantilla, RM Baron, DE Bowles, R Mishra and DO Jacobs. Hypoxia decreases creatine uptake in cardiomyocytes, while creatine supplementation enhances HIF activation. *Physiological Reports* 2017; **5(16)**, e13382.
- [50] G Scholtysik. Cardiac Na⁺ channel activation as a positive inotropic principle. *Journal of Cardiovascular Pharmacology* 1989; **14(S3)**, 24-29.
- [51] YT Mirzayeva, AA Zaripov, IZ Zhumaev, PB Usmanov, SY Rustamov, SN Boboev, SB Qurbonova, EB Ibragimov, MK Musaeva, SB Sobirov, SM Adizov. The Protective Effect of Indole Alkaloid Vincanine Against Hypoxia-Induced Vasorelaxation Model of Rat Aorta. *Biomed Pharmacol J* 2024; **17(1)**, 483-491.