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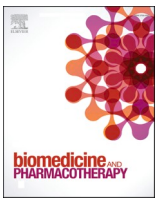

Genoprotective effect of cornelian cherry (*Cornus mas* L.) phytochemicals, electrochemical and *ab initio* interaction study

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ABSTRACT

Cornelian cherry (*Cornus mas* L.) has broad and multidimensional potential in preventing civilisational diseases. Part of these diseases results from DNA oxidative mutations. Thus, the paper aimed to predict how phenolics present in *C. mas* may interact with dsDNA in *ab initio* experiment and to check the effect of different cornelian cherry extracts on DNA structure and DNA oxidation. A special research model was designed using biosensor with a carbonpaste electrode. We resulted in various effects observed for phenolics and the extracts. Flavonoids, but of vitexin interacted with declining energy of the DNA models and liability of DNA oxidation. However, for 8-oxoguaniosine the trend was the opposite. Among the evaluated extracts, water-ethanolic extracts caused decline in adenine and guanine signals after dsDNA exposition on the extract. Principal component analysis showed that alcoholic extracts of cv. *Szafer* and *Słowianin*, which were rich in apigenin and kaempferol exhibit mild genoprotective effect.

1. Introduction

Cornelian cherry (*Cornus mas* L.) is plant which beneficial, pro-health properties have been rediscovered in the last few years [24]. Due to high amounts of anthocyanins, phenolics and vitamin C it has been evaluated in numerous papers dedicated to functional food and food chemistry aspects [3,18,21]. Despite the evaluated anti-radical, hypoglycaemic and hepatoprotective potential, no information may be found on its genoprotective potential. Although genoprotection is a rare aspect of functional food properties, it is vital, as results of such studies may help prepare novel dietary supplements and herbal medicines that can help people under chemotherapy [27,30]. Usually, genoprotection assay bases on analyses of reactive oxygen species (ROS) monitoring or observing fragmentation of DNA in electrophoresis. Meanwhile, the key result of genotoxicity is he transformation of nitrogen bases, which impairment with adjacent bases causes further mutation and damage to the whole DNA structure. One of the rare but precise method of determination of DNA interaction is square-wave voltammetry. Each purine base and its oxidated forms has unique potential of oxidation, which enables quantification of changes among them.

Phytochemicals may inhibit genotoxicity by reacting with ROS, but also they may act as a protection shield by creating a steric hindrance for ROS, interacting with DNA in major or minor grooves [12,14,16,19]. However, measurement of such interactions is extremely hard in in vitro and *in vivo* assays. Nonetheless, reliable information on the issue can be obtained with the help of molecular modeling tools.

The aim of this study was to validate complex genoprotective potential of cornelian cherry fruits, using electrochemical tool and *in silico* prediction of feasible interaction between DNA and phenolics present in the fruits.

Abbreviations: 8-oxoGC, 8-oxoguanosine-cytosine; A, adenine; ANOVA, Analysis of variance; AT, adenosine-thymidine; CPE, carbon paste electrode; G, guanine; GC, guanosine-cytosine; ROS, reactive oxygen species; SWV, Square-wave voltammetry; UHF, Unrestricted Hartree-Fock method.

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2. Material and methods

2.1. Cornelian cherry fruits and extracts preparation

The ripe fruits of Cornelian cherry were harvested in September 2018 in the orchard farm Szynsad in Dąbrówka Nowa, Błędów, Mazowieckie, Poland (51°47′01″N 20°43′04″E. The fruits of seven cultivars cultivated in Poland were taken under assessment: *Bolestraszycki*, *Wydubiecki*, *Szafer*, *Jolico*, *Florianka*, *Słowianin*, and *P5*.

Cornelian cherry extracts were composed of fresh fruits analogically as in our previous study [26]. Extraction was made by maceration of the fruits in distilled water or 40% (v/v) ethanol at a ratio fresh fruit: extractant 1:5 (w/v) for 30 min at 40 °C. Then, the residue of the matrices seeped through paper filters (Alfachem, Poznań, Poland). The prepared extracts were stored in 50-mL centrifugal tubes until examination at − 21 °C. Extracts used in the study were encoded as follows: BA – cultivar (cv.) *Bolestraszycki* alcoholic extract; BW - cv. *Bolestraszycki* aqueous extract; FA - cv. *Florianka* alcoholic extract; FW - cv. *Florianka* aqueous extract; SA - cv. *Szafer* alcoholic extract; SW - cv. *Szafer* aqueous; PA - cv. *P5* alcoholic extract; PW - cv. *P5* aqueous extract; XA - cv. *Wydubiecki* alcoholic extract; XW - cv. *Wydubiecki* aqueous extract; VA - cv. *Słowianin* alcoholic ex-tract; VW - cv. *Słowianin* aqueous extract; CA - cv. *Jolico* alcoholic extract; CW - cv. *Jolico* aqueous extract.

2.2. Chemicals

We used 0.05 M phosphate buffer with 0.01 M KCl (pH 7.0), and pure water in the study. The working electrode (CPE) was prepared using graphite powder (Sigma-Aldrich, Steinheim, Germany) and mineral oil (Sigma-Aldrich, Steinheim, Germany), mixed at the ratio of 7:3 (w/w) [17,26]. A platinum wire was used as an auxiliary electrode, and Ag/AgCl (3 M KCl) was applied as a reference.

In the study two complementary oligonucleotides at the following sequence 5′GCTCGGTACGGAAGTTGAC and 5′GTCAACTTCCGTACC GAGC (Tib Molbiol, Poznań, Poland) were used.

2.3. Electrochemical assessment

Based on the results of our previous research [26] that showed antioxidant properties of the tested extracts using square-wave voltammetry (SWV), we applied the same tool to observe the potential genoprotective effect of Cornelian cherry fruits extracts. The genoprotective effect was considered using two independent attempts:

a) Test of synergic interaction of the extract constituents with synthetic double-stranded DNA fragment (dsDNA);
b) Evaluation of extracts inhibitory potential against dsDNA oxidation by simulated Fenton reaction.

Double-stranded DNA fragments (dsDNA) were obtained by hybridising complementary oligonucleotides at a concentration of 10 μM, carried out in 0.05 M phosphate buffer with the addition of 0.5 M KCl for 15 min at the temperature of 40 °C.

2.3.1. Test of interaction between dsDNA fragments and the phenolics of the extract

Experiment was performed according to own protocol with slight modifications [17,23]. The SWV measurement started with the background measurement of the buffer. Electrodes placed in a well filled with 1 mL of phosphate buffer. During 60 s conditioning stage in the buffer, the electrode set was kept with potential of + 1.7 V and 5 s of equilibration time. Then, a calibration signal was recorded at range of 0.01–1.40 V with an amplitude of 0.04 V and a frequency of 50 Hz. The calibration curve was smoothed using the Savitzky-Golay algorithm. After the background measurement, the electrode set transferred to 4 μM dsDNA solution. The nucleic acid deposited on the surface of carbon paste working electrode (CPE) charged to a potential of + 0.5 V for 120 s. The electrode set was moved to a well filled with extract-buffer mixture in ratio 1:9 (v/v). Deposition of the extract phytochemicals occurred in no current conditions for only 60 s. Next, the electrode set was washed in a fresh buffer for 30 s, and the SWV measurement proceeded. The registered voltamperogram was smoothed, and then a background signal was subtracted. The corrected voltamperogram was saved, and the peak area was statistically analysed using Origin Pro 2022 (Origin, Germany). During all the stages, the solutions were stirred at 200 rpm. After each measurement, the layer of CPE was regenerated.

As a negative sample, a signal of dsDNA was measured, and instead of extract deposition, the electrodes were kept in 1:9 mixture of extractant (40%-ethanol/pure water) and buffer.

2.3.2. dsDNA oxidation study

Before the SWV measurement, the samples were prepared according to the following protocol. All amount of 400 μL of 10 μM dsDNA was mixed with 100 μL of tested extract, 2 μL of 0.5 M $FeSO_4$ aqueous solution, 3.34 μL of 30% H_2O_2 solution (POCH, Poland) and dissolved with phosphate buffer to a final volume of 2 mL. The final concentration was as follows: 2 μM dsDNA, approx. 0.05 g extract/L, 0.5 μM $FeSO_4$ and 0.21 μM H_2O_2. The whole sample was kept at temperature 37 °C for 1 h and then stored overnight at 4 °C until the SWV measurement.

As positive control was sample with 100 μL extractant instead of the extract, and the negative control was sample with 100 μL of extractant and 3.34 μL pure water instead of 30% H_2O_2 solution.

The SWV measurement started with the background measurement of the buffer, analogically as in the previous point. After the background measurement, the electrode set transferred to the sample. The double-stranded nucleic acid fragments deposited on the surface of carbon paste working electrode (CPE) working with a charge of + 0.5 V for 120 s. Next, the electrode set was washed in a fresh buffer for 30 s, and the SWV measurement proceeded. All other conditions, data preprocessing and analysis were done as mentioned above.

2.4. Phenolics chromatographical analysis

Phenolics in extracts were subjected to alkaline and acidic hydrolysis before the just analysis [22]. Acquity H class UPLC system equipped with the Waters Acquity PDA detector (Waters, USA) was used for the determination. A stationary phase Acquity UPLC® BEH C18 column (100 mm × 2.1 mm, particle size 1.7 μm) (Waters, Ireland) was used. The elution ran in a gradient mode using the following mobile phase composition: A: acetonitrile with 0.1% formic acid, B: 1% aqueous formic acid mixture (pH = 2). Concentrations of phenolic compounds were determined using an internal standard at wavelengths λ = 320 nm and 280 nm and finally given at mg/g extract. Compounds were identified based on a comparison of the retention time of the analysed peak with the retention time of standards, Selected compounds had to be analysed using the standard addition methods. The detection level for all tested compounds was 1 μg/g. Retention times for phenolic acids were: 9.55 min for 2,5-dihydroxybenzoic acid, 15.20 min for caffeic acid, 16.80 min for vanillic acid, 17.50 min for ferulic acid. Retention times for flavonoids were: 1.10 min for apigenin, 8.00 min for vitexin, 11.00 min for kaempferol, 16.90 min for luteolin, 17.00 min for quercetin and 17.50 min for naringenin.

2.5. Ab initio study of phenolics interaction with dsDNA

Performed was an interaction study between DNA models and phenolics detected in previous point of the study. For DNA models, we followed two independent approaches. First, a DNA model was part of dsDNA sequence applied in the SWV experiments (Fig. 1a), while the latter was a set of three base pairs: guanosine-cytosine (GC), 8-oxoguanosine-cytosine (8-oxoGC) and adenosine-thymidine (AT) (Fig. 1b).

For the first approach, phenolics docked to the A1 fragment, a model

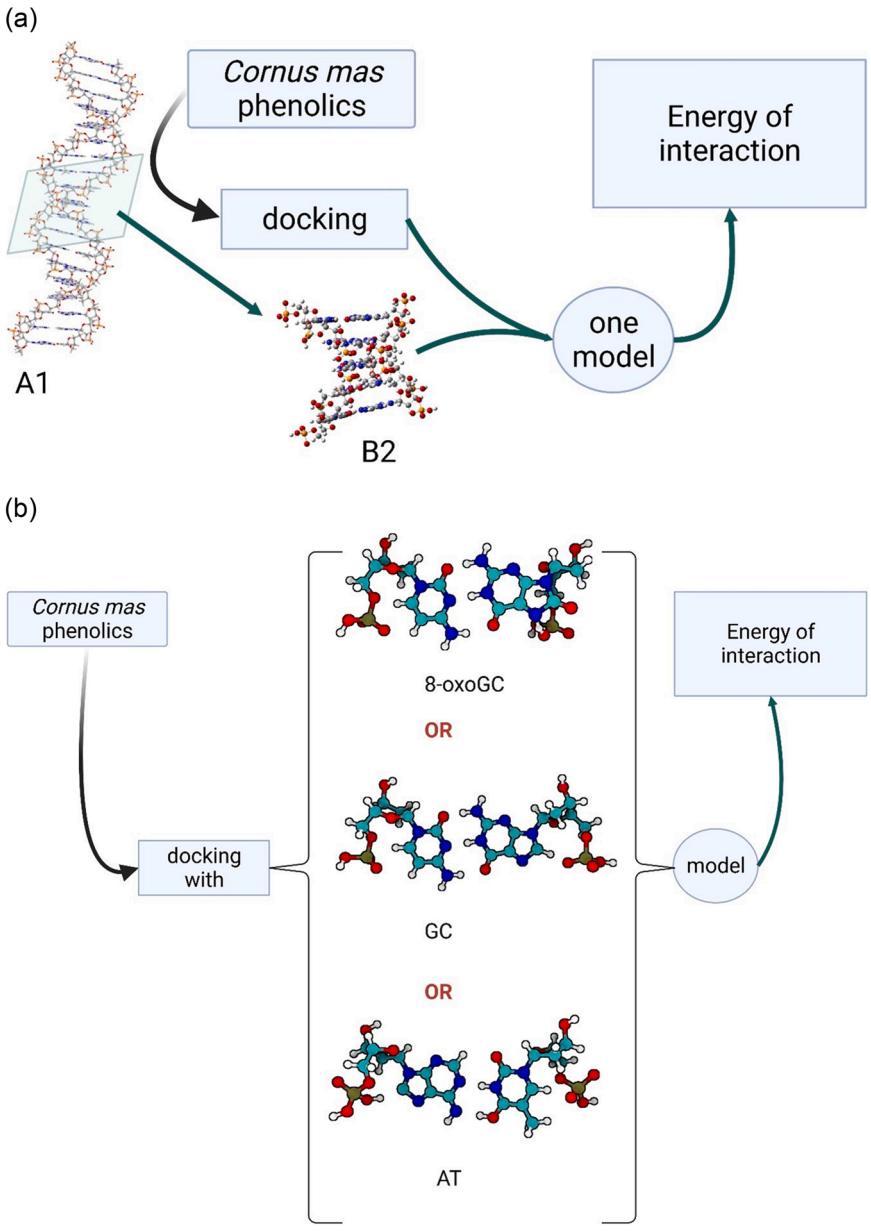

Fig. 1. Model of *in silico* study of *Cornus mas* phenolics interaction with dsDNA (a) and GC, 8-oxoGC and AT base pairs (b). Created with Biorender.

of whole sequence tested in SWV experiments. For the docking, we applied Autodock Vina software with Pyrx graphical interface [6,7,29]. Then, the ligands were added to B2 fragment which was a limited version of A1 (Fig. 1a). This limitation of the dsDNA macromolecule had to be done, as we had reached a problem with memory overload during the calculations. For such prepared models, the Hartree-Fock method was applied to calculate interaction energy using the STO-3 G basis set. Interaction energy was calculated using Gaussian 16 software [9].

After phenolic ligand to one of the base pair, a whole newly created model was stabilised by reoptimalization, and then interaction energy was calculated using Gaussian 16 software. To stabilise AT-, 8-oxoGC- and GC-based models, unrestricted Hartree-Fock method (UHF) and basis set 3–21 G* were applied. For interaction energy calculation, we applied 3–21 +G* , 3–21 G* and 3–21 G basis sets and UHF method.

2.6. Statistical analysis

Performed was statistical analysis to validate the effect of individual phytochemical on the genoprotective effect of each extract of *Corni fructus*. To achieve this goal, principal component analysis (PCA) was done involving qualitative (extract, compound) and quantitative variables (concentration of each compound in mg/kg, current of G/A peak [A] of dsDNA after interaction, energy of interaction between compound and B2/GC/8-oxoGC/AT model). For intensity of G peak after interaction energy of interaction in B2, GC, 8-oxoGC models were considered as variables, while for A peak variable were interaction with B2 and AT models.

Additionally, for HPLC determination of phenolics, analysis of variance (ANOVA) and Fisher post-hoc tests were done to illustrate significant differences between the concentration of individual components and between the extracts ($\alpha = 0.05$).

We used Origin Pro 2022 (Origin, Germany) software for statistical calculations.

3. Results and discussion

Interaction of dsDNA with extracts of *Corni fructus* resulted in higher area of peaks located at 1.0 and 1.2 V in voltammograms (Fig. 2). These

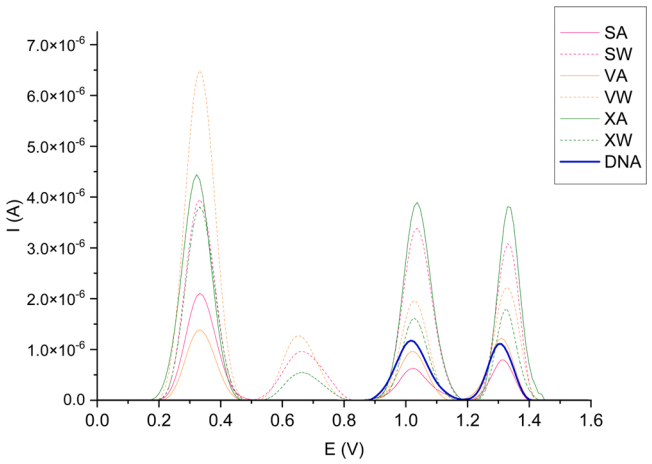

Fig. 2. Voltamperograms of dsDNA (blue line) and dsDNA after interaction with tested extracts (CA-XW).

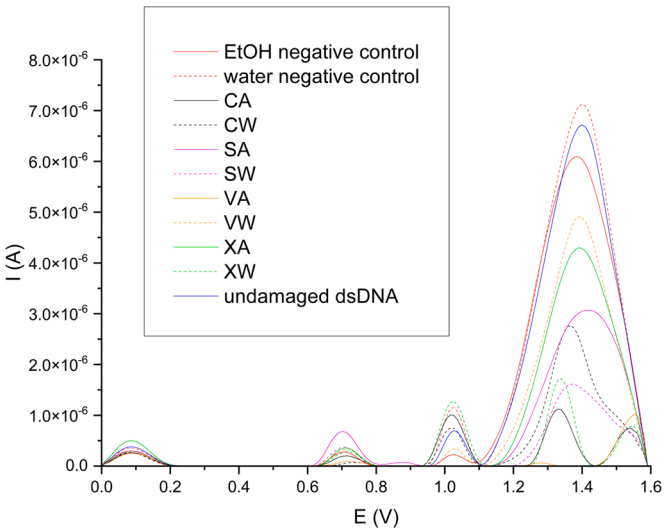

Fig. 3. Square-wave voltammograms of dsDNA samples oxidized with and without addition of *C. mas* extracts.

two peaks are characteristic of purine bases of dsDNA (Fig. 2, blue line): the first signal originating from guanine and the other from adenine, which confirms the resulst of our previously published research ([17, 23]). The higher areas of dsDNA signals may indicate that DNA double-helical structure changed and untwisted in place of interaction, which caused higher oxidation registered in voltammograms. Meaningful growth of the peaks area was found after interaction with samples SW and XA. Nonetheless, after interaction with VA sample, unnoticeable changes were observed, while sample SA interacting with dsDNA resulted in a decrease of purine peaks, which may illustrate the genoprotective effect. This result is surprising as we had assumed that the presence of ethanol in the samples might affect the electrochemical analyses negatively. Meanwhile, the observed peak growths for the aqueous extract are more clearly visible. The observed growth in dsDNA area signals might have resulted from overdose of phytochemicals in the reacting samples. However, in the preliminary stage of our study [24], the usage of lower concentrations of the cornelian cherry extracts ended with inaccurate CPE response. What is more, if the unspecific interaction of phenolics with dsDNA promoted oxidation, we could observe signals of 8-oxoguanine (8-oxoG), the soxidised form of guanine. For the tested samples, we resulted in no meaningful trace of 8-oxoG, as no peak at the potential range 0.8–0.9 V was registered [28]. Our manuscript dedicated to the interaction of dsDNA with different concentrations of naringin [23] showed that concentration of naringin higher than 10 μM could promote oxidation of DNA instead of its protection. In the case of higher doses of phytocompounds, voltammograms obtained after its interaction with dsDNA can have higher signals than before the interaction as the layer of CPE binds free compounds interacting with DNA but not being chelated by it. This causes an overlay of signals and finally may bias the analysis. However, this error can be limited by washing CPE after the interaction stage (point 2.3.1). Additional peaks observed at 0.3 V and 0.7 V are characteristic of *Cornus mas* phenolic compounds, which had been proven in our previous research [24]. Additionally, the observed dsDNA peaks shifted towards higher potential values after interaction with the extracts. As the higher potential indicates lower oxidation feasibility, the voltamperograms may show that the overall effect of the extracts was positive, despite unravelling the double-helical structure. *Ab initio* determination of interaction with selected phenolics and HPLC quantitative determination should help deliver clearer conclusions.

Simulated Fenton-induced oxidation of dsDNA resulted in severe damage (Fig. 3). Compared to sunoxidised dsDNA, oxidation caused drop of peaks area in voltammograms. Meaningful is a broadened signal of adenosine at 1.4 V. This shift towards higher potential along with asymmetric layout may suggest that the registered peak represents oxidated guanine and pyrimidine bases (cytosine, thymine), as their signals could occasionally be registered for potential higher than 1.4 V. Compared to A, signals of G in Fig. 3 had significantly lower currents. This might result from higher liability of G to oxidation and indicate that G was oxidised in major. However, similar results were observed for the unoxidized DNA (i.e. sample without H_2O_2 addition), which may indicate that dosage of ferrous ions could induce oxidation even without presence of H_2O_2. In work of Lafave et al. [15] simulated Fenton reaction resulted in growth of G signal. However, in the referred study analysed was only oxidation of guanine.

Effect of the extracts on dsDNA oxidation varied very meaningfully. Sample of XA caused the highest drops in A signal and increased G peak to a level similar to A. Interaction study (Fig. 2) showed that dsDNA before the oxidation had G and A signals similar in their intensities. Moreover, XA interacting with dsDNA caused completely different growths in SWV peaks. This shows that the interaction of dsDNA with xenobiotics may change double-helical conformation, but a degree of this change does not relatelinearly with oxidation liability.

Paradoxically, SW that inhibited at most signal of dsDNA in interaction study, here affected the oxidation of dsDNA positively. In Fig. 3 can be observed no G peak, but noticeable is the presence of peak at 0.9 V, which may be considered as 8-oxoG. Control samples with no genoprotective agent had visible growths in A peak. Moreover, ethanolic control resulted with G area decrease for both A and G signals. Moreover, ethanolic control had the lowest G peak area. This may show that ethanol may boost H_2O_2 oxidation effect. A similar effect was observed in Ahmed et al. [1] study, where was a noticeable correlation between ethanol concentration and H_2O_2 reduction.

The peak location in the potential range 0.6–0.8 V is similar to that in Fig. 2. This may represent the extracts residues that stayed in the sample and were not oxidised. The registered signal may also be characteristic for ethanol, as it was also observed in ethanolic negative control. The last peak near the potential of 0.1 V origin from the non-subtracted part of the baseline and may also show a residue of ferrous ions, which did not react during the Fenton reaction [32]. The generated ferric ions cannot be registered during the experiment, as Fe^{3+} signal is visible in potential − 0.65 V, beyond the range used in this experiment [11,32].

For *in silico* study, we selected 10 compounds, i.e. 4 phenolic acids and 6 flavonoids (Table 1). Compounds were selected based on their different qualitative and quantitative content in the tested extracts in our previous study [25]. For these compounds were developed models, which has been docked to B1, GC, 8-oxoGC and AT fragments.

The prevalent compound in all extracts was naringenin, a compound

Table 1
Phenolics (mg/kg extract) present in *Corni fructus* samples.

Samples	2,5-DHBA	caffeic acid	ferulic acid	vanillic acid	naringenin	vitexin	quercetin	apigenin	kaempferol	luteolin
SA	$198.8^{bC} \pm 1.1$	$3.8^{aBC} \pm 0.2$	$7.3^{aC} \pm 0.2$	$0.5^{aA} \pm 0.2$	$205.9^{bBC} \pm 3.3$	$13.5^{aA} \pm 0.3$	$291.4^{bC} \pm 10.8$	$45.7^{aC} \pm 1.0$	$1.5^{aAB} \pm 0.2$	$0.3^{aA} \pm 0.0$
SW	$51.6^{bA} \pm 39.2$	$2.8^{aB} \pm 0.1$	$4.3^{aA} \pm 0.2$	Nd	$128.7^{cA} \pm 2.0$	$17.6^{aAB} \pm 0.9$	$3.6^{aA} \pm 0.5$	$0.1^{aA} \pm 0.0$	$0.3^{aA} \pm 0.1$	$0.3^{aA} \pm 0.0$
VA	$113.3^{bB} \pm 1.7$	$3.0^{aB} \pm 0.1$	$6.7^{aCB} \pm 0.3$	$3.7^{aB} \pm 0.2$	$438.3^{cC} \pm 15.2$	$46.9^{aC} \pm 2.1$	$172.4^{bB} \pm 5.9$	$42.6^{aC} \pm 2.2$	$2.7^{aB} \pm 0.1$	$2.2^{aB} \pm 0.1$
VW	$65.5^{bAB} \pm 0.7$	$0.6^{aA} \pm 0.2$	$4.7^{aAB} \pm 0.3$	Nd	$140.7^{cAB} \pm 4.6$	$12.0^{aA} \pm 0.5$	$3.3^{aA} \pm 0.2$	$0.1^{aA} \pm 0.0$	$0.2^{aA} \pm 0.0$	$0.5^{aA} \pm 0.0$
XA	$182.0^{bC} \pm 1.7$	$4.8^{aC} \pm 0.1$	$7.7^{aC} \pm 0.2$	$3.6^{aB} \pm 0.3$	$434.2^{dC} \pm 2.0$	$26.5^{aB} \pm 1.4$	$265.2^{cC} \pm 3.8$	$21.1^{aB} \pm 0.5$	$2.1^{a} \pm 0.1$	$2.5^{aB} \pm 0.2$
XW	$43.6^{bA} \pm 0.9$	$1.3^{aA} \pm 0.1$	$2.8^{aA} \pm 0.1$	Nd	$98.7^{cA} \pm 0.9$	$10.8^{aA} \pm 0.8$	$3.6^{aA} \pm 0.4$	$0.3^{aA} \pm 0.1$	$0.2^{aA} \pm 0.0$	Nd

2,5-DHBA – 2,5-dihydroxybenzoic acid; Nd – not detected; N = 3; results given as mean ± sd; lowercase letters indicate significant differences between compounds ($P < 0.05$); capital letters indicate significant differences between samples ($P < 0.05$).

which genoprotective role was confirmed in our previous study [23]. Moreover, naringin and apigenin exhibit genoprotective potential as they may absorb UVB radiation in human cell lines [10]. Significant difference was found between ethanolic and aqueous extracts, especially in quercetin, apigenin and vanillic acid (Table 1). Hydrocinnamic acids and flavonols are the two most prevalent groups in fruits. They can be found in different kernel fruits, including cornelian cherry, bird cherry, and black cherry [4,5,20]. Therefore, these properties may show anti-radical and genoprotective properties. Acidic methanolic extract of *Citrullus colocynthus* containing kaempferol, luteolin, apigenin, ferulic and vanillic acids protected plasmid DNA against H_2O_2 and UV induced oxidation [31]. Alcaraz et al. [2] observed similar effects. They noted that apigenin administrated after X-ray exposition decreased by 42% number of micronuclei in erythrocytes, while quercetin resulted in 26% growth. However, the authors concluded that different is the effect observed whether genoprotective agents administrated before or after the exposition on oxidation.

Interaction between *Cornus mas* phenolics and the oligonucleotide and singular base pairs showed noticeable variability (Table 2). For B2 fragments highest interaction energy had flavonoids, especially luteolin and kaempferol. However, for GC luteolin, roughly no interaction could be predicted. This shows that interaction xenobiotic and oligonucleotide is not strictly limited to Hoogsteen-type interaction with singular base-pair but is also affected by interactions between xenobiotics and adjacent base-pairs in dsDNA. Steric hindrance may affect the strength of interaction, as well.

Interaction with AT resulted with the highest interaction energy for vitexin and the lowest for kaempferol. The negative charge of interaction between AT and the latter means that this interaction is favourable [8]. For GC favourable was an interaction between quercetin, apigenin and naringin. These differences related with different locations of nucleo- and electrophile places in both compared base pairs. Also, this clearly shows preferred binding places for tested phenolics. For B2 all compounds had positive binding energy, which relates to the feasible destabilization of the dsDNA structure. The double helix unravelling was confirmed in the previous part of the work (Fig. 2). As simple groove binding cannot affect the destabilization of the DNA structure, clear is that the steric effect and long-range interaction might induce the unravelling in SWV study. This can be especially specific for vitexin, which presence in vicinity of B2 resulted with ultra-high interaction energy (Table 2).

Oxidation of guanine lead to formation of 8-oxoG, which hardly paired with cytosine via hydrogen bonding and destabilised the entire dsDNA structure. We checked how this mutated analogue of guanine may interact in a triangular set of phenolic-8-oxoGC and if the phenolics may stabilize this unfavourable base pair. *In silico* analysis showed that caffeic acid was the one that interacted with 8-oxoGC stabilized the whole molecule set. Conversely, flavonoids were found to interact with GC and AT base-pairs favourably with high interaction energies. This may show that cornelian cherry phenolics protect dsDNA against oxidation not only *a priori* but also after the oxidation occurs. The destabilization of double helix at singular locus may help NER and BER mechanisms delete the oxidated form of guanosine [13,33].

Results of the interaction study, content of individual phenolics in tested cultivars of *C. mas* and interaction energies between them and B2 and the base-pairs helped us construct PCA model of the potential genoprotective effect (Fig. 4).

PCA showed that aqueous extracts of *C. mas* had similar genoprotective properties (Fig. 4). Especially, meaningful vicinity was found between aqueous extracts, while this VW and XW were mostly characterized by interaction of phenolics with 8-oxoGC and GC. A strong, positive relation was found in interactions with B2 and AT models, which shows that interaction with AT base-pairs was prevalent in evaluated B2 fragment and, therefore in dsDNA fragment in the electrochemical study. Conversely, SA and VA were in similar statistical relation with content of individual phenolics, while they showed opposite relation with B2 and AT interactions. Surprisingly, energy of interaction with GC had little statistical force in the evaluated model of the research. The opposite relation between interaction and content of phenolics may show highest contents of phenolics leads to unspecific interactions with DNA and negative changes in the double helix.

Electrochemical study showed that changes in the signal of guanine were in line with changes for adenine (Fig. 4). Moreover, PCA showed that the presence of SA and VA affected currents of adenine and guanine negatively after the interaction thus, they exhibit a genoprotective effect. Conversely, all other extracts affected growth in- the electrochemical signals, showing no genoprotective effect. XA sample was characterized mainly through raising the electrochemical signals, which may be considered as an oxidation-promoting effect.

Nonetheless, these results should be evaluated in a more advanced model e.g., human cell model, as the evaluated parameters represent only nearly 50% of the observed effects.

4. Conclusions

Our study showed that ethanolic extracts had stronger power to interact with dsDNA. The presented study also showed that different cultivars of cornelian cherry may have various or no genoprotective effect. The differences based on different quantitative content of individual phenolics in the prepared extracts and different energy of interaction between them and modeled nucleotides. Apigenin and kaempferol were the most potent genoprotective agents, while vitexin was considered a compound causing interactions with dsDNA, which maydestabilize the double-helical structure. Despite noticeable insight of this work on phenolics and cornelian cherry genoprotective potential, the observed effects still need verification in more advanced models, including human cell lines or other *in vivo* experiments.

Table 2
Energy of interaction (kcal/mol) between DNA fragments and *Cornus mas* phenolics.

Fragment	Basis set	Compound	Energy of interaction
B2	STO-3 G	2,5-DHBA	0.19
		caffeic acid	708.92
		ferulic acid	148.09
		vanillic acid	190.75
		naringenin	372.35
		vitexin	9320.11
		quercetin	217.66
		apigenin	6.74
		kaempferol	672.80
		luteolin	884.78
GC	3–21 G	2,5-DHBA	149.40
		caffeic acid	17.30
		ferulic acid	20.97
		vanillic acid	-0.33
		naringenin	-8.61
		vitexin	22.35
		quercetin	-4.19
		apigenin	-4.85
		kaempferol	27.00
		luteolin	0.01
	3–21 G*	2,5-DHBA	149.40
		caffeic acid	17.17
		ferulic acid	21.15
		vanillic acid	-0.33
		naringenin	-8.38
		vitexin	22.40
		quercetin	-4.96
		apigenin	-4.78
		kaempferol	27.04
		luteolin	0.60
	3–21 +G*	2,5-DHBA	151.51
		caffeic acid	19.35
		ferulic acid	20.50
		vanillic acid	0.36
		naringenin	-7.24
		vitexin	25.92
		quercetin	-2.68
		apigenin	-21.72
		kaempferol	26.38
		luteolin	2.23
8-oxoGC	3–21 G	2,5-DHBA	0.56
		caffeic acid	-10.55
		ferulic acid	13.07
		vanillic acid	31.54
		naringenin	5.49
		vitexin	11.95
		quercetin	12.21
		apigenin	-0.14
		kaempferol	13.62
		luteolin	12.79
	3–21 G*	2,5-DHBA	0.08
		caffeic acid	-8.17
		ferulic acid	13.12
		vanillic acid	31.54
		naringenin	5.85
		vitexin	12.66
		quercetin	14.15
		apigenin	-0.14
		kaempferol	14.0
		luteolin	14.14
	3–21 +G*	2,5-DHBA	2.39
		caffeic acid	-11.38
		ferulic acid	15.10
		vanillic acid	32.31
		naringenin	6.30
		vitexin	11.69
		quercetin	9.52
		apigenin	0.73
		kaempferol	14.15
		luteolin	12.38
AT	3–21 G	2,5-DHBA	-1.50
		caffeic acid	5.86
		ferulic acid	6.10
		vanillic acid	8.10
		naringenin	23.35
		vitexin	194.30
		quercetin	-0.74
		apigenin	3.51
		kaempferol	-4.44
		luteolin	10.02
	3–21 G*	2,5-DHBA	-1.80
		caffeic acid	5.84
		ferulic acid	6.07
		vanillic acid	8.12
		naringenin	23.29
		vitexin	194.13
		quercetin	-0.84
		apigenin	3.53
		kaempferol	-4.55
		luteolin	9.88
	3–21 +G*	2,5-DHBA	-0.18
		caffeic acid	7.62
		ferulic acid	7.98
		vanillic acid	9.25
		naringenin	24.85
		vitexin	194.53
		quercetin	0.63
		apigenin	4.75
		kaempferol	-4.00
		luteolin	12.11

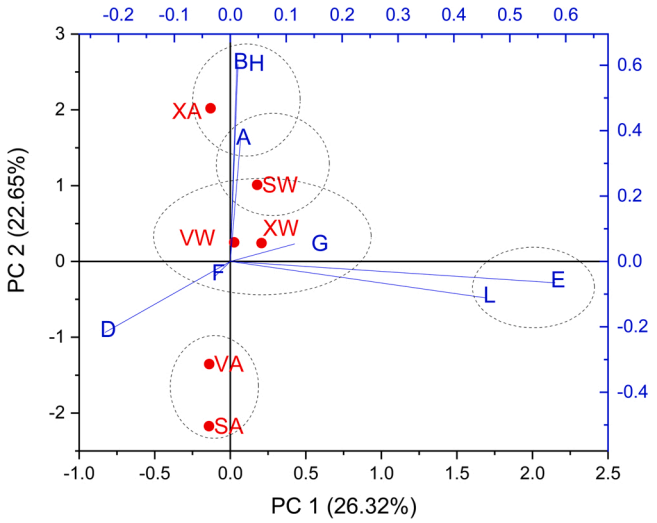

Fig. 4. Principal component analysis of tested cornelian cherry extracts (red points SA-XW) and evaluated electrochemical and *in silico* parameters (blue): A – extract; B – Guanine current after interaction; D – content (mg/kg) of individual phenolics; E – energy of interaction (kcal/mol) between phenolics and B2; F – energy of interaction between phenolics and GC; G – energy of interaction between 8-oxoGC and individual phenolics; H – Adenine current after interaction; L – energy of interaction between individual phenolics and AT.

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CRediT authorship contribution statement

Conceptualization: O.S.; Data curation: O.S.; Formal analysis: O.S.; Funding acquisition: O.S., M.L. and J.K.; Investigation: O.S. and K.S.; Methodology: O.S., M.L. and K.S.; Project administration: O.S.; Resources: J.K.; Software: O.S. and M.L.; Supervision: M.L. and J.K.; Validation: M.L.; Visualization: O.S.; Writing – original draft: O.S.; Writing - review & editing: M.L.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Input, control and output files of simulations performed in *ab initio* study, as well as structures of docked phenolics and nucleotides will be available from Open Data Repository of Poznan University of Life Sciences at link: https://doi.org/10.18150/5VVM8J. All other data will be available from the corresponding author on reasonable request.

Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.biopha.2022.113216.

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