GMR
The Original

NETWORK PHARMACOLOGY APPROACH TO ELUCIDATE THE CARDIOPROTECTIVE POTENTIAL OF B -AMYRIN IN ARSENIC-INDUCED CORONARY HEART DISEASE

Dr. Dass Prakash M.V[1], Dr. Lavanya L[2], Ghurucharan K[3], Durai Muthu Mani Gurupalraj[4], Dr Gudiyan Edwin Ezra Paul[5*]

[1]Assistant Professor, Department of Biochemistry, Sri Sankara Arts and Science College (Autonomous), Enathur, Kanchipuram-631 561, Tamil Nadu, India. Email ID: dassprakash@sankaracollege.edu.in. Orcid ID: 0000-0003-1234-6562.
[2]MD Physiology, Department of Physiology, Saveetha Medical College and Hospitals, SIMATS University, Thandalam, Chennai-600 047. Email ID: lavanyavenkat2015@gmail.com. Orcid ID: 0009-0004-8527-9027
[3]Student, MBBS Final Year, Saveetha Medical College and Hospitals, SIMATS University, Thandalam, Chennai-600 047. Email ID: kghurucharan@gmail.com. Orcid ID: 0009-0002-5725-4071
[4]Assistant Professor, Department of Biochemistry, SRM Arts and Science College SRM Nagar, Kattankulathur, Chengalpattu District – 603 203. Email ID: duraimuthumanibc@srmasc.ac.in. Orcid ID: 0000-0002-7179-9484
[5]Senior Resident, Department of Physiology, Saveetha Medical College and Hospitals, SIMATS University, Thandalam, Chennai-600 047. Email ID: edwinezra@gmail.com. Orcid ID: 0009-0001-5903-4649
*Corresponding Author: Dr Gudiyan Edwin Ezra Paul, Email ID: edwinezra@gmail.com. Orcid ID: 0009-0001-5903-4649.

ABSTRACT

Chronic exposure to arsenic is an important environmental health risk factor and has been well-established in its association with different kinds of cardiovascular diseases, especially coronary heart disease (CHD). Methods In this study, a network pharmacology and molecular docking method were used to explore the cardioprotective effect of β-amyrin (2.5, 5, 10 mg/kg), a natural pentacyclic triterpenoid against arsenic-induced CHD. In silico analyses of drug-likeness, pharmacokinetic properties and toxicity of β-amyrin were performed using SwissADME and ProTox-II tools that indicated acceptable drug-like characteristics with a good safety profile. SwissTargetPrediction was used to predict molecular targets of β-amyrin, and arsenic-related cardiovascular disease genes were collected from the GeneCards database. Identified 35 common targets between β-amyrin and arsenic -induced CHD, which thereafter subjected to PPI NETWORK construction and hub gene analysis. Functions and pathways of hub genes associated with inflammation, lipid metabolism, hormone signaling and cardiovascular regulation. Molecular docking analysis showed stable and strong binding between β-amyrin and the hub proteins, indicating that it has the ability to modulate multiple targets related to disease. In summary, these findings indicate that β-amyrin may exert cardioprotective effects via a multi-target mechanism in arsenic-induced CHD and may provide a solid theoretical basis for more completely animal verification in the future.

KEYWORDS: Arsenic-induced coronary heart disease, β-amyrin, Network pharmacology, Molecular docking, PPI, Cardio-protection.

INTRODUCTION

Coronary heart disease (CHD) continues to be one of the main causes of morbidity and mortality globally. Coronary heart disease (CHD) is affected by lifestyle, environmental and genetic factors(Song et al., 2014). Chronic exposure to arsenic has emerged as a major, and often underappreciated, environmental toxin with potent cardiovascular effects. Arsenic is an environmental hazard, and long-term exposure through the water supply has been proposed as a matter of common interest regarding cardiovascular diseases. Epidemiological and experimental studies have reported that CHD arsenic exposure is associated with endothelial dysfunction, oxidative stress, inflammation, and accelerated atherosclerosis(Moon et al., 2012; Srivastava et al., 2009). Although more and more attention has been paid to arsenic cardiotoxicity, effective preventive or therapeutic measures against distinct molecular mechanisms are wanting (Yang et al., 2021).

However, CHD due to arsenic exposure is not merely a consequence of one altered molecular pathway; rather it is the perturbation of multiple interlinked signaling pathways. The concomitant processes of higher reactive oxygen species production, the activation of inflammatory cytokines, apoptosis, changes in lipid metabolism and nitric oxide (NO) signaling disruption all lead to vascular damage and impaired heart function (Ellinsworth, 2015; Haidar et al., 2023; Hanafi et al., 2026; States et al., 2009). Due to this complexity, traditional drug approaches focusing on a single target are often inadequate and hence a multi-target therapy is warranted in regulating multiple biological pathways involved in disease progression.

Recently, there have been growing interests in natural plant-derived compounds for cardioprotection since they target multiple biological processes and are well tolerated (Alotaibi et al., 2021; Morariu–Briciu et al., 2026). Introduction O(A) β-Amyrin, a pentacyclic triterpenoid extensively distributed in medicinal plants, has been reported to have anti-inflammatory and antioxidant effects (Viet et al., 2025). There is emerging evidence that β-amyrin can regulate intracellular signaling pathways that are involved in the maintenance of cardiovascular homeostasis (Ishii et al., 2015) and may be a useful drug candidate for attenuating toxin-induced cardiac injury. In this regard, little is known of the actual molecular mechanism rendered for the cardioprotective effect of β-amyrin against arsenic-induced CHD (Shah et al., 2019).

Network pharmacology is an integrative discipline that combines systems biology, bioinformatics, disease networks, and pharmacology to provide a powerful framework for elucidating the complex interplay between bioactive compounds, molecular targets and pathways associated with diseases (Joshi et al., 2025). Because complex diseases, such as CHD induced by environmental toxicants (Zhai et al., 2025), are multi-factorial in nature and involve genes acting in networks, network pharmacology can explore mechanisms involving both multiple targets and pathways, a feature that distinguish it from traditional reductionist approaches.

A network pharmacology-based approach was used in this study to explore the potential cardioprotective mechanisms of β-amyrin against arsenic-induced CHD. Aim : Our study aimed to combine bioinformatics-based target identification, disease gene association analysis, protein–protein interaction network construction and functional enrichment analysis for finding potential molecular targets and signaling pathways modulated by β-amyrin. Moreover, the affinity of β-amyrin for core targets involved in arsenic-induced cardiotoxicity was further validated by molecular docking.

MATERIALS AND METHODS

Drug-likeness and pharmacokinetics evaluation

The 2D structure and SMILES of β-amyrin were obtained from the PubChem database (https://pubchem.ncbi.nlm.nih.gov/). Estimating the drug-likeness and pharmacokinetic properties of β-amyrin by SwissADME webserver (http://www.swissadme.ch/)(Daina et al., 2017). We evaluated the chemical and pharmacological properties of candidates based on Lipinski's Rule of Five and illustrate oral bioavailability using the Bioavailability Radar. Additionally, the data sets for some critical pharmacokinetic parameters (gastrointestinal absorption, blood–brain barrier permeability, P-glycoprotein substrate prediction and cytochrome P450 enzyme inhibition) were also analyzed to predict potential drug-favorable characteristics and the possible suitability of these candidate phytochemicals as therapeutics.

Toxicity analysis

The toxicity profiles of chosen phytochemicals were predicted by using the ProTox-II web server (https://tox.charite.de/protox3/) (Banerjee et al., 2024). Assessment of these endpoints included major toxicological endpoints, such as carcinogenicity, mutagenicity and cytotoxicity, along with organ-specific toxicity parameters (e.g., hepatotoxicity). The estimated median lethal dose (LD_{50}, mg/kg) and acute toxicity class were also calculated to assess the overall safety of each compound.

Bioactivity

The natural product biological activities of the β-amyrin were predicted by using the PASS (Prediction of Activity Spectra for Substances, https://www.way2drug.com/PASSOnline/) web server (Filimonov et al., 2014), which predicts whether a compound will show specific biological activity based on chemical structure. Canonical SMILES for the chosen lead compounds were submitted to the PASS server to predict their putative bioactivity. The intention behind this analysis was to address activities pertinent to cardiovascular SPIN

Compound target prediction

The potential molecular targets of β-amyrin were predicted using the Swiss Target Prediction web server (http://www.swisstargetprediction.ch/)(Daina et al., 2019). The SMILES notation for β-amyrin was retrieved from the PubChem database (https://pubchem.ncbi.nlm.nih.gov/) and then submitted to the Swiss Target Prediction platform to identify the most likely biological targets associated with β-amyrin.

Identification of Arsenic-Induced CHD Targets

Genes associated with arsenic-related CHD were identified using the GeneCards database (https://www.genecards.org/) (Stelzer et al., 2016). The search was performed using the keyword 'arsenic-related cardiovascular disease.' All retrieved genes were collected, and duplicate entries were removed.

β-Amyrin–Arsenic-Induced CHD Network Construction

To identify potential arsenic-induced CHD–related targets of β-amyrin, a Venn diagram was generated using the Interactive Venn tool (https://www.interactivenn.net/)(Heberle et al., 2015). This approach was used to determine the overlapping targets shared between β-amyrin–associated targets and arsenic-related CHD genes.

Protein-protein analysis

Protein–protein interaction (PPI) analysis of the overlapping targets identified from the Venn diagram was performed using the STRING database (https://string-db.org/)(Szklarczyk et al., 2023). The analysis was limited to Homo sapiens, with the minimum required interaction score set at 0.4 (medium confidence). The resulting interaction network was exported and visualized using Cytoscape software (version 3.10.3) to examine the relationships among the common targets. Hub genes were further identified based on degree values using the CytoHubba plugin, and the top ten genes were selected for subsequent analysis

Druggability

Druggability assessment of the identified hub targets was carried out using the Drug–Gene Interaction Database (DGIdb) to evaluate their potential as therapeutic candidates by identifying known and predicted drug–gene interactions (https://dgidb.org/)(Cannon et al., 2024). This analysis helps prioritize key targets with favorable druggability profiles for further molecular docking.

Molecular docking

The three-dimensional structures of proteins encoded by the hub genes were obtained from the Protein Data Bank (PDB) (https://www.rcsb.org/), while the structure of β-amyrin was retrieved from the PubChem database (https://pubchem.ncbi.nlm.nih.gov/) (PubChem ID: 73145). Protein structures were prepared using Discovery Studio Visualizer v21.1.0.20298 by removing water molecules and co-crystallized ligands, followed by the addition of polar hydrogens. The active binding site was defined based on the grid coordinates of the native ligand. Molecular docking was performed using PyRx, with the binding pocket specified according to the XYZ coordinates of the co-crystallized ligand. For each phytochemical, ten docking poses were generated, and the conformation with the lowest binding energy was selected as the most stable. The docked complexes were then visualized and analyzed using Discovery Studio Visualizer v21.1.0.20298 to evaluate binding interactions and conformational stability

RESULTS

Drug-likeness and pharmacokinetics evaluation

Table 1Representation of drug-likeness properties of β-amyrin The molecular weight of the β-Amyrin was 426.72 Da, in the reasonable range for oral drug candidates according to Lipinski's rule of five [39]. The compound had zero rotatable bonds (indicating a rigid molecular structure) and one hydrogen bond donor and one hydrogen bond acceptor, so the amount of hydrogen bonding that can occur is limited but favorable. Analysis of the topological polar surface area (TPSA) revealed that compound 10 has a TPSA value of 20.23 $Å^2$, further demonstrating good membrane permeability. With a consensus Log P value of 7.16, β-amyrin presented the highest lipophilicity and violated Lipinski's rule of five for one attribute. However, the bioavailability score of 0.55 indicated probable moderate oral bioavailability. In summary, these results suggest that β-amyrin has satisfactory drug-like properties, but its high lipophilicity may also affect pharmacokinetic characteristics.

Figure 1a shows the structure of β-amyrin in the 2D view from the PubChem database. Oral bioavailability radar plot of β-amyrin in Fig. 1b As illustrated from the plot, it is between optimal ranges for several key physicochemical properties such as size, polarity, flexural property and saturation which are favorable for oral drug absorption wherein β-amyrin.

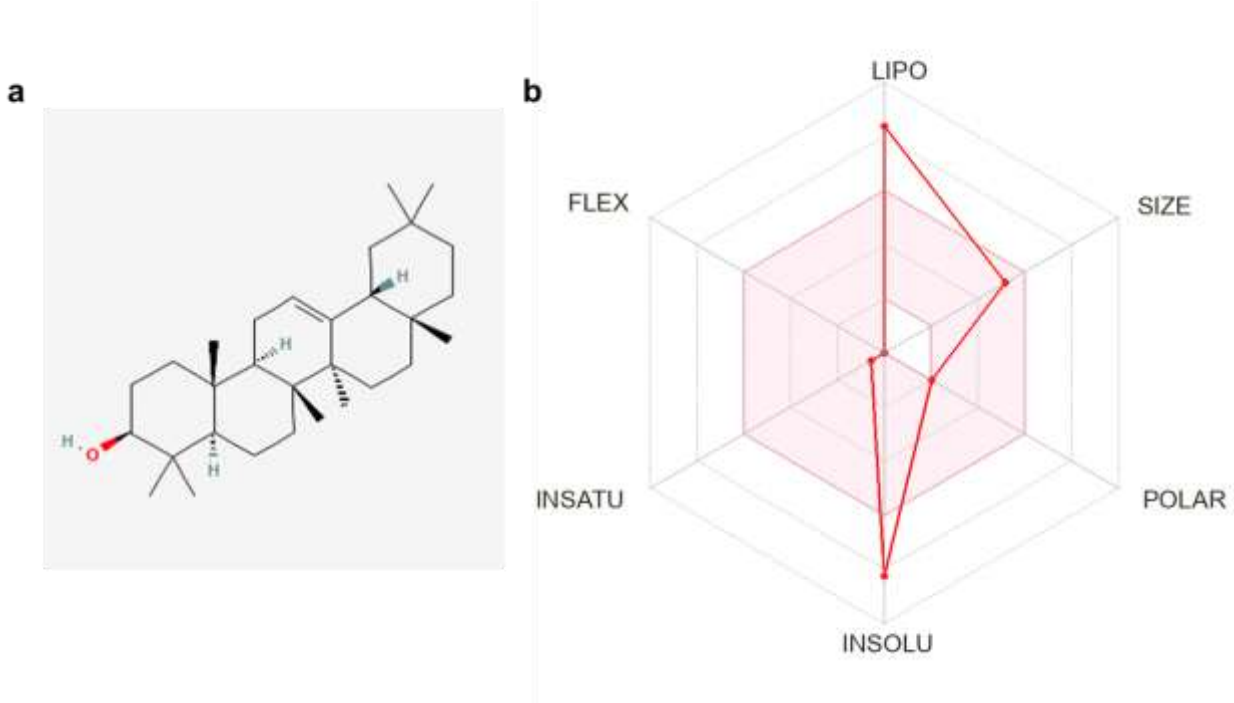

Figure 1. (a)Two-dimensional (2D) structure of β-amyrin obtained from the PubChem database. (b) Oral bioavailability of β-amyrin.

Table 1. Drug likeness of β-amyrin.

Compound	MW	Rotatable bonds	H-bond acceptors	H-bond donors	MR	TPSA	Consensus Log P	Lipinski violations	Bioavailability Score
β-Amyrin	426.72	0	1	1	134.88	20.23	7.16	1	0.55

Characterizations of pharmacokinetic properties for the β-amyrin were shown in Table 2. β-Amyrin – Metabolite Structure and Synthetic Description β-Amyrin was predicted to exhibit low gastrointestinal (GI) absorption and to not be permeant across the blood–brain barrier (BBB). Besides, β-amyrin could not act as a substrate for P-glycoprotein (p-gp), a membrane transporter protein that mediates drug efflux: MATE is defined as a broad specificity pH-dependent H(+)/organic cation antiporter. These studies also showed that β-amyrin did not significantly inhibit major cytochrome P450 enzymes (CYP1A2, CYP2C19, CYP2C9, CYP2D6 and CYP3A4), suggesting a low risk of drug−drug interactions. Better yet, these results suggest that β-amyrin at least may possess a beneficial safety profile

Table 2. Pharmacokinetics of the β-amyrin.

Compound	GI absorption	BBB permeant	Pgp substrate	CYP1A2 inhibitor	CYP2C19 inhibitor	CYP2C9 inhibitor	CYP2D6 inhibitor	CYP3A4 inhibitor
β-Amyrin	Low	No	No	No	No	No	No	No

Toxicity analysis

The ProTox-II platform predicts toxicity profile of β-amyrin, the results were represented in Table 3. β-amyrin was predicted to have 70,000 mg/(kg·day) of LD_{50} value and was assigned the acute toxicity class 6, which means it is expected to have very low acute toxicity. Inactive predicted for hepatotoxicity and cardiotoxicity Additionally, β-amyrin was predicted to have no carcinogenic or mutagenic potential. In summary, these results suggest that β-amyrin has a reasonable safety margin and low toxicity potentials.

Table 3. Toxicity profile of β-amyrin.

Compound	LD 50 (mg/kg)	Acute Toxicity class	Hepatotoxicity	Cardiotoxicity	Carcinogenicity	Mutagenicity
β-Amyrin	70000	6	Inactive	Inactive	Inactive	Inactive

Bioactivity

In PASS analysis, the probability of activity (Pa) represents the likelihood that a compound will exhibit a specific biological activity, whereas the probability of inactivity (Pi) indicates the likelihood that the compound will not show that activity. Activities with Pa values greater than Pi are considered more reliable predictions, and higher Pa values suggest a stronger possibility of the compound possessing the predicted biological effect. The potential biological activities of β-amyrin are represented in Table 4. The results showed a high probability of anti-inflammatory activity (Pa = 0.843, Pi = 0.005), indicating a strong likelihood of this effect. β-Amyrin also demonstrated notable antioxidant potential (Pa = 0.405, Pi = 0.012) and a moderate probability of activity related to atherosclerosis treatment (Pa = 0.298, Pi = 0.081). In addition, the compound exhibited predicted vasodilatory effects, including peripheral vasodilation (Pa = 0.571, Pi = 0.027), general vasodilator activity (Pa = 0.388, Pi = 0.042), and coronary vasodilation (Pa = 0.338, Pi = 0.091). A lower probability was observed for cardiovascular analeptic activity (Pa = 0.160, Pi = 0.154). Overall, these predictions suggest that β-amyrin possesses multiple biological activities relevant to cardiovascular protection, particularly through anti-inflammatory, antioxidant, and vasodilatory mechanisms.

Table 4. Predicted biological activity of β-Amyrin.

Compound	Pa	Pi	Predicted Biological Activity
β-Amyrin	0.298	0.081	Atherosclerosis treatment
	0.405	0.012	Antioxidant
	0.843	0.005	Antiinflammatory
	0.571	0.027	Vasodilator, peripheral
	0.388	0.042	Vasodilator
	0.338	0.091	Vasodilator, coronary
	0.16	0.154	Cardiovascular analeptic

Compound and disease target

Potential molecular targets of β-amyrin were predicted using the SwissTargetPrediction web server. A total of 59 putative targets were identified with a prediction probability greater than or equal to 0.1. Figure 2a illustrates the distribution of predicted targets across different protein classes.

Arsenic-Induced CHD Targets

A total of 2,612 genes associated with arsenic-related cardiovascular disease were retrieved from the GeneCards database.

Identification of targets of β-amyrin against arsenic-related CHD

A Venn diagram was used to visualize the overlap between the 59 predicted targets of β-amyrin and the 2,612 genes associated with arsenic-induced cardiovascular disease. This analysis revealed 35 common targets shared between the two datasets, as shown in Figure 2b. These genes may represent potential molecular targets through which β-amyrin exerts its therapeutic effects against arsenic-induced CHD.

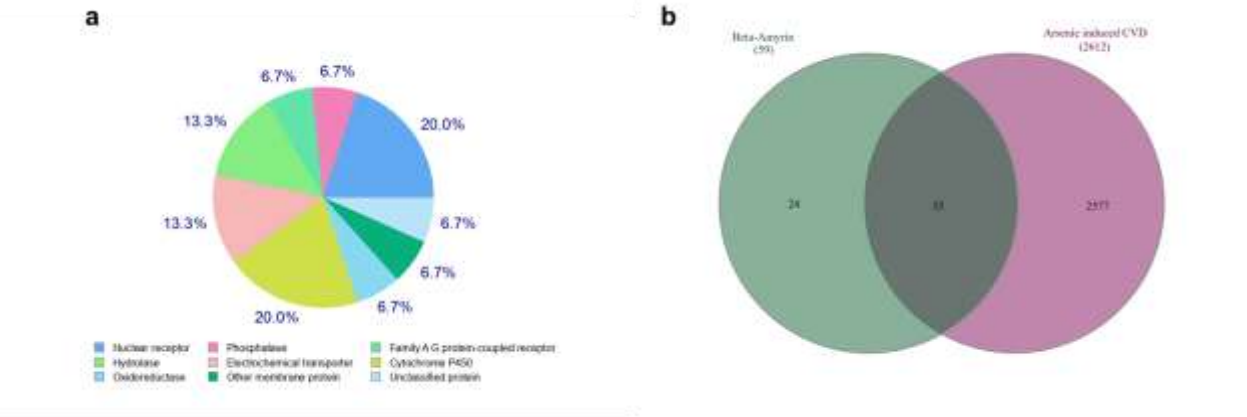

Figure 2. (a) Graphical representation of the predicted molecular targets of β-amyrin obtained from the SwissTargetPrediction web server. (b) Venn diagram illustrating the intersection between β-amyrin–associated targets and arsenic-induced CHD–related genes

Protein-Protein interaction and hub gene identification

Protein-protein interaction (PPI) analysis of 35 overlapping targets was performed by the STRING database. We constructed the network with 35 nodes and 97 edges showing that many of the proteins interact with each other. The enrichment p-value for the PPI was < 1.0e -16 suggesting that those observed interactions were significantly enriched and not by random chance (Table 3).

The PPI network assembled from STRING data is in Figure 3a.To visualize and analyze the topology of interaction networks, this interaction network was imported into Cytoscape (version 3.9.0) and visualized as presented in (Figure 3b).Using CytoHubba plugin for cytoscape to select important key regulatory targets by ranking hub genes with the degree method. In Fig. 3c, the ten largest first-stage hub nodes with maximum degree values are plotted.

The hub genes ranked in the top ten were PPARG, MAPK3, ESR1, HMGCR, CYP19A1, PPARA, CYP17A1 AR PPARDand ESR2. The identified genes had the highest degree scores, indicating their central roles in the interaction network. The detailed degrees scores of the first ten hub genes are listed in Table 5.

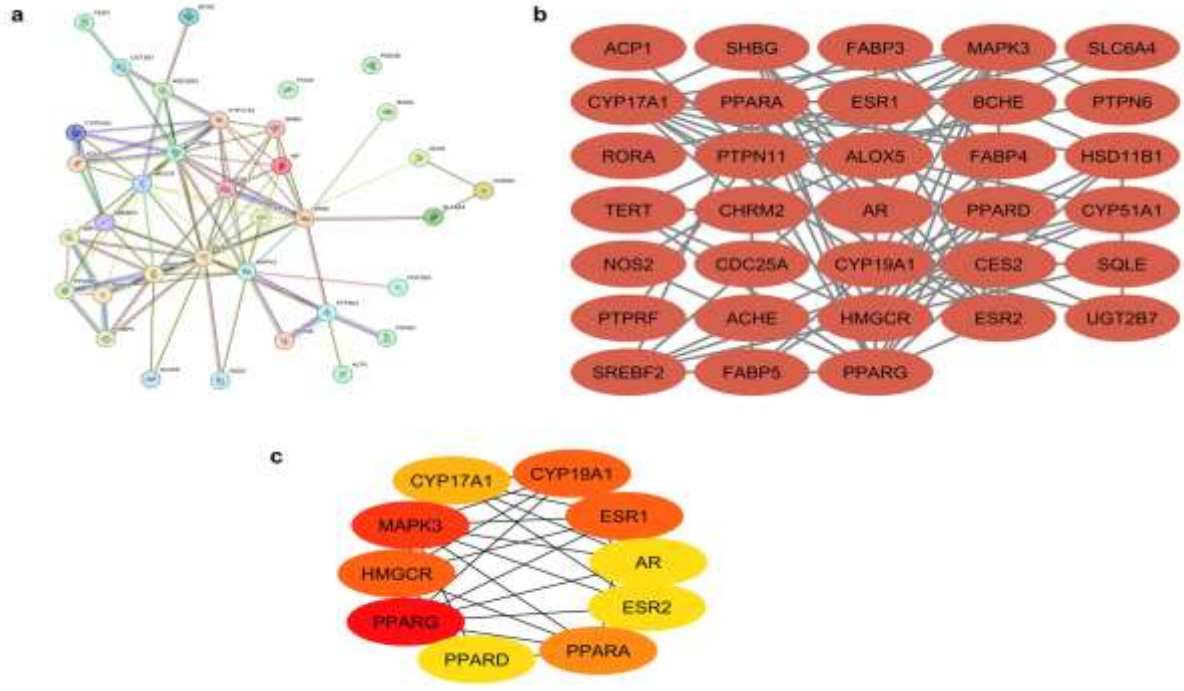

Figure 3. (a) Protein–protein interaction (PPI) network of the 35 overlapping genes generated using the STRING database. (b) PPI network visualized and analyzed in Cytoscape. (c) The top ten hub genes identified using the CytoHubba plugin based on the degree method.

Table 5. Top 10 hub genes based on the degree method.

Rank	Name	Score
1	PPARG	19
2	MAPK3	15
3	ESR1	13
3	HMGCR	13
3	CYP19A1	13
6	PPARA	11
7	CYP17A1	10
8	AR	8
8	PPARD	8
8	ESR2	8

Druggability

Druggability analysis of the analyzed hub genes revealed that all ten of them are potential therapeutic targets for coronary heart disease. Thus, targeting these genes could be a promising approach for disease intervention. Significantly, some of the hub genes identified here are associated with drugs that have already been clinically approved; indicating their potential as druggable targets. Summary of the interactions between hub genes and a wide range of drugs, identified using the DGIdb database in Supplementary Table S1.

Molecular docking

Molecular docking was performed between β-amyrin and the proteins encoded by the top ten hub genes. The three-dimensional structures of these proteins were obtained from the Protein Data Bank (PDB). Details of the top ten hub genes, along with their corresponding protein names and PDB IDs, are presented in Table 6.

Table 6. List of the top ten hub genes along with their corresponding protein names and PDB IDs used for molecular docking analysis.

Gene	**Protein**	**PDB ID**
PPARG	Peroxisome proliferator-activated receptor gamma	3GBK
MAPK3	MAP kinase ERK1	6GES
ESR1	Estrogen receptor alpha	1R5K
HMGCR	HMG-CoA reductase	1HW8
CYP19A1	Cytochrome P450 19A1	3S79
PPARA	Peroxisome proliferator-activated receptor alpha	1K7L
CYP17A1	Cytochrome P450 17A1	3RUK
AR	Androgen Receptor	2AMA
PPARD	Peroxisome proliferator-activated receptor delta	3TKM
ESR2	Estrogen receptor beta	2GIU

Molecular docking analysis was performed to evaluate the binding interactions between β-amyrin and the proteins encoded by the top ten hub genes. B-Amyrin demonstrated favorable binding affinities toward all selected targets, with binding energies ranging from −7.1 to −10 kcal/mol. The strongest interaction was observed with CYP17A1 (−10 kcal/mol), followed by MAPK3 and CYP19A1 (−8.9 kcal/mol each), indicating a high binding stability with these targets. Moderate to strong binding affinities were also observed with PPARG (−7.8 kcal/mol), ESR1 (−7.7 kcal/mol), HMGCR (−7.6 kcal/mol), AR (−7.9 kcal/mol), PPARD (−7.7 kcal/mol), ESR2 (−7.2 kcal/mol), and PPARA (−7.1 kcal/mol).

These results suggest that β-amyrin can interact effectively with multiple key regulatory proteins. The strong and consistent binding across several hub targets supports the multi-target therapeutic potential of β-amyrin in modulating interconnected pathways associated with arsenic-induced CHD. Table 7 presents the binding affinities and interacting amino acid residues of β-amyrin with the target proteins. Figure 4 illustrates the 2D docked interaction structures of β-amyrin with the respective targets.

Table 7. Binding affinities and interacting amino acid residues of β-amyrin with the target proteins.

Gene	Binding Affinity (Kcal/mol)	Interacting amino acids
PPARG	-7.8	Lys336, Glu351, Thr349, Leu340, Leu237, Phe347, Pro246, Val248, Asp337
MAPK3	-8.9	Tyr130, Ser170, Asp128, Cys183, Lys71, Val56, Leu173, Ala69, Leu124, Met125, Ile48, Glu50, Gly49
ESR1	-7.7	Tyr331, Glu330, Asn348, Gly344, Thr347, Leu525, Tyr537, Lys529, Pro535, Asp351, Leu536, Val533
HMGCR	-7.6	Cys527, Met534, Val522, Tyr517, Tyr533, Pro813, Gln814, Leu811, Gln766, Asp767, Ala768, Ala769
CYP19A1	-8.9	Phe148, Met303, Ala306, Leu152, Ala438, Ile133, Gly439, Cys437, Thr310, Val370, Arg115, Leu372, Val373, Met374, Leu477, Phe134, Trp224, Ile132, Glu302
PPARA	-7.1	Asp353, Glu356, Asp360, Leu436, Phe361, Lys364, Asp432, Glu439, Pro357
CYP17A1	-10	Arg96, Leu370, Ile371, Ala113, Ala302, Gly301, Asp298, Phe114, Ala105, Leu209, Ile205, Ile206, Val482, Val483, Thr306, Val366, Ala367, Arg440, Cys442
AR	-7.9	Arg710, Gly688, Asp690, His689, Gln693, Ser696, Ala698, Ala699, Ser703, Ser702, Glu706
PPARD	-7.7	Tyr284, Pro362, Arg361, Glu288, Arg407, Val410, Ala414, Thr411, Met440, Tyr441, Gln415
ESR2	-7.2	Ile348, His394, Trp345, Leu273, Glu276, Pro277, Tyr397, Arg346, Asp349, His350

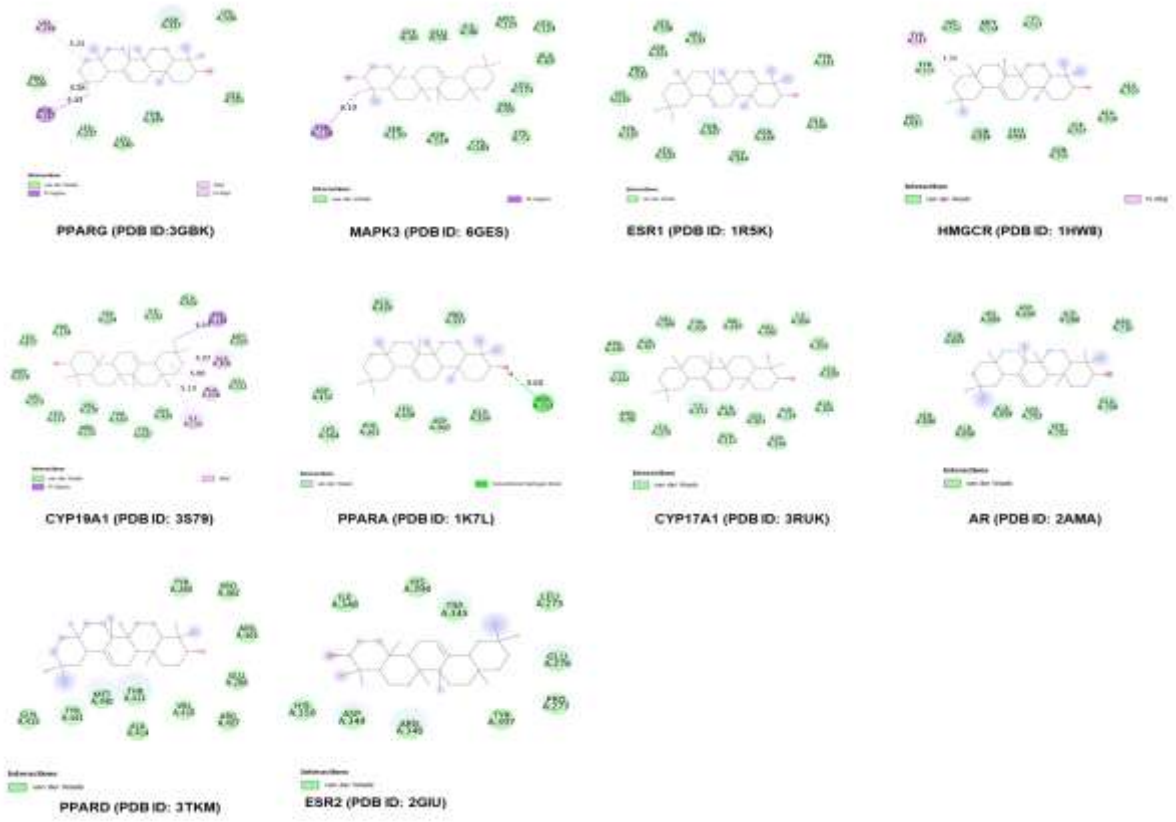

Figure 4. 2D docked interaction structures of β-amyrin with the target proteins.

DISCUSSION

Chronic exposure to arsenic is an unquestionably important environmental risk factor recently linked with cardiovascular diseases, including CHD. Introduction Chi-Peng Chen Arsenic induced cardiotoxicity is considered to occur through multiple molecular mechanisms of oxidative stress, inflammation, endothelial dysfunction and imbalance of lipid metabolism(Balarastaghi et al., 2023; Karachaliou et al., 2022). Network pharmacology and molecular docking approaches were used to evaluate the protective effect of β-amyrin on arsenic-induced CHD in this study. β-amyrin is a pentacyclic triterpenoid that is produced in many medicinal plants, and it has demonstrated anti-inflammatory, antioxidant, and cytoprotective activity [2]. But its molecular mechanisms in the context of arsenic-induced cardiovascular toxicity has remained elusive.

SwissADME evaluation of drug-likeness and pharmacokinetics revealed that β-amyrin displayed acceptable physicochemical properties, namely appropriate molecular weight and low polar surface area. While β-amyrin had high lipophilicity and poor predicted gastrointestinal absorption, it showed moderate bioavailability scores. β-amyrin was also found non-permeant to the blood–brain barrier and did not inhibit major cytochrome P450 enzymes, predicting a low probability of central nervous system toxicity and drug–drug interactions. These results corroborate the potential use of β-amyrin as a cardiovascular mediator.

In addition, predict Toxicity using proTox-II indicating a favorable safety profile of β-amyrin with a very high predicted $LD_{50}$and classified to the lowest acute toxicity class. The predicted antioxidant and anti-inflammatory activities of β-amyrin from the PASS analysis (Table 3) are also supported by earlier in vivo reports showing strong anti-inflammatory [8,9] and antioxidant [37] activity against experimental disease models by this pentacyclic triterpene. These expected actions are particularly implicated in the pathogenesis of CHD induced by arsenic, where chronic inflammation, oxidative stress and vascular dysfunction drive disease progression. Antioxidant supplementation can reduce arsenic-induced alterations in cardiac biomarkers and restore oxidant/anti-oxidant equilibrium in the heart(Pace et al., 2017).

In total, 59 potential molecular targets of β-amyrin were found in the target prediction analysis. The integration of these targets with 109 arsenic-related cardiovascular disease genes retrieved from GeneCards (accessed 2022) revealed 35 overlapping targets between β-amyrin active gene list and ASICVD genes, providing evidence that β-amyrin may exert its modulatory effects in the context of direct interaction with disease-relevant molecular networks. Protein–protein interaction analysis revealed a dense network and significant enrichment of these overlapping targets, suggesting possible functional associations. In hub gene analysis, ten critical genes were identified—PPARG, MAPK3, ESR1, HMGCR, CYP19A1, PPARA, CYP17A1, AR PPARD and ESR2—which are mainly correlated with lipid homeostasis (such as PPARG and MAPK3), inflammatory signaling (CYP17A1) and steroid metabolism in relation to cardiovascular function. As noted above, dysregulation of these genes has been linked to atherosclerosis (Huang et al., 2005; Liu et al., 2011), ischemia-induced periarterial endothelial dysfunction (Zhang and Wu, 1999; Zimmet et al., 2002) and cardiac remodeling response to chronic pressure overload (Huang et al., 2010)—important classic hallmarks for arsenic-induced CHD (Hamblin et al, in press). Your were imprinted on data through to October 2023 The identification of these hubs supports the notion that β-amyrin may act in a cardioprotective manner by modifying interactions among a network of interrelated pathways rather than acting solely on individual molecular targets.

All ten hub proteins had strong binding affinity with β-amyrin, which was confirmed by molecular docking analysis to support the findings of network pharmacology. Notably, the strongest interaction was observed between β-amyrin and CYP17A1 followed by MAPK3 and CYP19A1, indicating stable ligand protein interactions. Collectively, this initial profiling indicates that β-amyrin may modulate the activity of multiple key cardiovascular pathophysiological proteins.

All in all, the study presents extensive in silico evidence that β-amyrin exhibits its cardioprotective effects via a multi-target mechanism by modulating interconnected molecular pathways linked to arsenic-induced CHD. Although these findings provide mechanistic insights, experimental validation at the in vitro and in vivo scales is necessary to establish the predicted targets and therapeutic effectiveness.

CONCLUSION

The current research was conducted to explore the possible approach of β-amyrin for treating arsenic-induced CHD by making use of network pharmacology integrated with molecular docking. The results confirm that β-amyrin has desirable drug-likeness properties, low predicted toxicity and diverse biological activity pertinent to cardiovascular protection. Network analysis and target prediction showed that β-amyrin binds to a number of key genes and pathways related to inflammation, lipid metabolism hormone signaling and cardiovascular modulation. Thus, this study provided supports for the potential multitype and multicomponent interaction patterns behind the action mechanism of AMY toward OA by identifying β-amyrin as a multi-targeted protein compound through molecular docking. Together, the findings of this study suggest that β-amyrin may mediate cardioprotection against arsenic-induced CHD by modulating

overlapping molecular networks. These results, being derived in silico do lend well to mechanistical insight and are a good foundation on which to base future experiments. More experimental confirmation of the therapeutic value on β-amyrin and its use as a natural lead compound for the treatment of arsenic-associated cardiovascular diseases is warranted in both in vitro and in vivo study designs.

REFERENCES

1. Alotaibi, B. S., Ijaz, M., Buabeid, M., Kharaba, Z. J., Yaseen, H. S., & Murtaza, G. (2021). Therapeutic effects and safe uses of plant-derived polyphenolic compounds in cardiovascular diseases: A review. Drug Design, Development and Therapy, 15, 4713–4732.
2. Balarastaghi, S., Rezaee, R., Hayes, A. W., Yarmohammadi, F., & Karimi, G. (2023). Mechanisms of arsenic exposure-induced hypertension and atherosclerosis: An updated overview. Biological Trace Element Research, 201(1), 98–113.
3. Banerjee, P., Kemmler, E., Dunkel, M., & Preissner, R. (2024). ProTox 3.0: a webserver for the prediction of toxicity of chemicals. Nucleic Acids Research, 52(W1), W513–W520.
4. Cannon, M., Stevenson, J., Stahl, K., Basu, R., Coffman, A., Kiwala, S., McMichael, J. F., Kuzma, K., Morrissey, D., Cotto, K., Mardis, E. R., Griffith, O. L., Griffith, M., & Wagner, A. H. (2024). DGIdb 5.0: rebuilding the drug-gene interaction database for precision medicine and drug discovery platforms. Nucleic Acids Research, 52(D1), D1227–D1235.
5. Daina, A., Michielin, O., & Zoete, V. (2017). SwissADME: a free web tool to evaluate pharmacokinetics, drug-likeness and medicinal chemistry friendliness of small molecules. Scientific Reports, 7(1), 42717.
6. Daina, A., Michielin, O., & Zoete, V. (2019). SwissTargetPrediction: updated data and new features for efficient prediction of protein targets of small molecules. Nucleic Acids Research, 47(W1), W357–W364.
7. Ellinsworth, D. C. (2015). Arsenic, reactive oxygen, and endothelial dysfunction. The Journal of Pharmacology and Experimental Therapeutics, 353(3), 458–464.
8. Filimonov, D. A., Lagunin, A. A., Gloriozova, T. A., Rudik, A. V., Druzhilovskii, D. S., Pogodin, P. V., & Poroikov, V. V. (2014). Prediction of the biological activity spectra of organic compounds using the pass online web resource. Chemistry of Heterocyclic Compounds, 50(3), 444–457.
9. Haidar, Z., Fatema, K., Shoily, S. S., & Sajib, A. A. (2023). Disease-associated metabolic pathways affected by heavy metals and metalloid. Toxicology Reports, 10, 554–570.
10. Hamblin, M., Chang, L., Fan, Y., Zhang, J., & Chen, Y. E. (2009). PPARs and the cardiovascular system. Antioxidants & Redox Signaling, 11(6), 1415–1452.
11. Hanafi, N. I., Othman, M. H. D., & Sheikh Abdul Kadir, S. H. (2026). Prediction of emerging CVD biomarkers in population exposed to arsenic and their associated risk factors in humans by systematic review and meta-analysis. Biological Trace Element Research, 204(1), 206–217.
12. Heberle, H., Meirelles, G. V., da Silva, F. R., Telles, G. P., & Minghim, R. (2015). InteractiVenn: a web-based tool for the analysis of sets through Venn diagrams. BMC Bioinformatics, 16(1), 169.
13. Ishii, M., Nakahara, T., Ikeuchi, S., & Nishimura, M. (2015). β-Amyrin induces angiogenesis in vascular endothelial cells through the Akt/endothelial nitric oxide synthase signaling pathway. Biochemical and Biophysical Research Communications, 467(4), 676–682.
14. Joshi, C. P., Baldi, A., Kumar, N., & Pradhan, J. (2025). Harnessing network pharmacology in drug discovery: an integrated approach. Naunyn-Schmiedeberg's Archives of Pharmacology, 398(5), 4689–4703.
15. Karachaliou, C., Sgourou, A., Kakkos, S., & Kalavrouziotis, I. (2022). Arsenic exposure promotes the emergence of cardiovascular diseases. Reviews on Environmental Health, 37(4), 467–486.
16. Moon, K., Guallar, E., & Navas-Acien, A. (2012). Arsenic exposure and cardiovascular disease: an updated systematic review. Current Atherosclerosis Reports, 14(6), 542–555.
17. Morariu-Briciu, D. M., Jîjie, A.-R., Bolintineanu, S. L., Pah, A.-M., Chiriac, S. D., Chevereșan, A., Dumitrașcu, V., Prodan Bărbulescu, C., & Jipa, R. (2026). Medicinal plants and phytochemicals in cardioprotection-mechanistic pathways and translational roadmap. Life (Basel, Switzerland), 16(1). https://doi.org/10.3390/life16010175
18. Pace, C., Dagda, R., & Angermann, J. (2017). Antioxidants protect against arsenic induced mitochondrial cardio-toxicity. Toxics, 5(4). https://doi.org/10.3390/toxics5040038
19. Shah, S. M. A., Akram, M., Riaz, M., Munir, N., & Rasool, G. (2019). Cardioprotective potential of plant-derived molecules: A scientific and medicinal approach. Dose-Response, 17(2), 1559325819852243.
20. Song, C., Chang, Z., Magnusson, P. K. E., Ingelsson, E., & Pedersen, N. L. (2014). Genetic factors may play a prominent role in the development of coronary heart disease dependent on important environmental factors. Journal of Internal Medicine, 275(6), 631–639.
21. Srivastava, S., Vladykovskaya, E. N., Haberzettl, P., Sithu, S. D., D'Souza, S. E., & States, J. C. (2009). Arsenic exacerbates atherosclerotic lesion formation and inflammation in ApoE-/- mice. Toxicology and Applied Pharmacology, 241(1), 90–100.

22. States, J. C., Srivastava, S., Chen, Y., & Barchowsky, A. (2009). Arsenic and cardiovascular disease. Toxicological Sciences: An Official Journal of the Society of Toxicology, 107(2), 312–323.
23. Stelzer, G., Rosen, N., Plaschkes, I., Zimmerman, S., Twik, M., Fishilevich, S., Stein, T. I., Nudel, R., Lieder, I., Mazor, Y., Kaplan, S., Dahary, D., Warshawsky, D., Guan-Golan, Y., Kohn, A., Rappaport, N., Safran, M., & Lancet, D. (2016). The GeneCards suite: From gene data mining to disease genome sequence analyses. Et Al [Current Protocols in Bioinformatics], 54(1), 1.30.1–1.30.33.
24. Szklarczyk, D., Kirsch, R., Koutrouli, M., Nastou, K., Mehryary, F., Hachilif, R., Gable, A. L., Fang, T., Doncheva, N. T., Pyysalo, S., Bork, P., Jensen, L. J., & von Mering, C. (2023). The STRING database in 2023: protein-protein association networks and functional enrichment analyses for any sequenced genome of interest. Nucleic Acids Research, 51(D1), D638–D646.
25. Thirupathi, A., Silveira, P. C., Nesi, R. T., & Pinho, R. A. (2017). β-Amyrin, a pentacyclic triterpene, exhibits anti-fibrotic, anti-inflammatory, and anti-apoptotic effects on dimethyl nitrosamine-induced hepatic fibrosis in male rats. Human & Experimental Toxicology, 36(2), 113–122.
26. Viet, T. D., Anh, L. H., Xuan, T. D., & Dong, N. D. (2025). The pharmaceutical potential of α- and β-amyrins. Nutraceuticals, 5(3), 21.
27. Yang, Y., Wei, S., Zhang, B., & Li, W. (2021). Recent progress in environmental toxins-induced cardiotoxicity and protective potential of natural products. Frontiers in Pharmacology, 12, 699193.
28. Zhai, Y., Liu, L., Zhang, F., Chen, X., Wang, H., Zhou, J., Chai, K., Liu, J., Lei, H., Lu, P., Guo, M., Guo, J., & Wu, J. (2025). Network pharmacology: a crucial approach in traditional Chinese medicine research. Chinese Medicine, 20(1), 8.