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Molecular Docking and ADMET-Based Screening of Cow Milk Derived Volatile Organic Compounds against Targets of Type-II Diabetes

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ABSTRACT

Nutraceuticals derived from milk and other sources play a significant role in the management of diabetes mellitus, particularly type 2 diabetes (T2DM). Milk contains a variety of volatile compounds, including ketones, esters, acids, aldehydes, and lactones, which contribute to its aroma and flavour. These compounds can be naturally present in milk or formed during processing, such as heat treatment or fermentation. These bioactive compounds, including proteins, lipids, and oligosaccharides found in milk, exhibit various biological activities that can aid in glycaemic control and reduce complications associated with diabetes. The objective of this study is to evaluate these compounds for pharmacophore studies and anti-diabetic properties. The reported volatile organic compounds from milk were pooled and analysed for their pharmacological bioactivities, like Lipinski's rule of five and ADMET, while maintaining an oral bioavailability. Further, these compounds are evaluated in silico for their potential anti-diabetic, by targeting α-amylase and DPP-4 proteins with the standard FDA-approved drug molecule Metformin. Among 41 volatile compounds, 13 compounds passed the drug likeness; these compounds were studied for their anti-diabetic activity by target proteins. Molecular Docking analysis revealed that α-Amylase interacted with (Benzoic acid -5.5 kcal/mol, 2-Ethyl-hexanoic acid -5.2 kcal/mol, 2-Amino-5-methylbenzoic acid -6 kcal/mol) and DPP-4 interacted with (Benzoic acid -5.5 kcal/mol, 2-Ethyl hexanoic acid -5.2 kcal/mol, 2-Amino-5-methylbenzoic acid -6.1 kcal/mol). Compounds in the milk had better interactions in comparison with the standard drug. Overall, the integration of nutraceuticals into diabetes management strategies offers a promising complementary approach to conventional therapies, potentially leading to improved patient outcomes and quality of life.

Keywords: *volatile organic compounds, Molecular Docking analysis, Lipinski's rule of five, ADMET, anti-diabetic therapeutics.*

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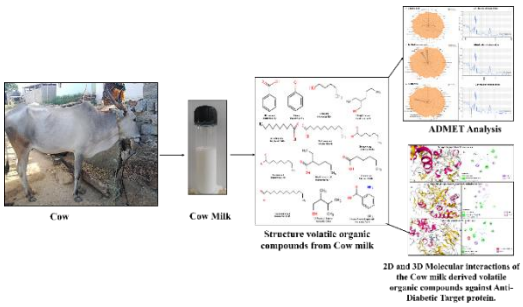
Graphical Abstract



1. INTRODUCTION

Milk contains a variety of volatile compounds, including ketones, esters, acids, aldehydes, and lactones, which contribute to its aroma and flavour. These compounds can be naturally present in milk or formed during processing, such as heat treatment or fermentation [1]. Raw milk from different geographical regions has different organoleptic qualities. Compounds that positively affect flavour can help turn regional differences into geographical advantages [2,3]

The feed also affects the volatile compounds, for instance, the bovines fed with mycorrhizal ensiled forage showed an increase of free fatty acids and ketones in the milk [4]. Apart from this, the microorganisms that ferment the milk also influence the final profile of the volatiles and flavour of dairy products [5]. The change in profile of VOCs is also found with the change in the season, with some bacterial species or groups occurring seasonally [6]. The volatile organic compounds in feed and forage can enter milk by two mechanisms, that is, through the digestive system or through the pulmonary system [7].

Diabetes mellitus is a collection of long-term metabolic disorders marked by persistently elevated blood sugar levels, or hyperglycemia. Insulin resistance (IR), reduced insulin production, or a combination of the two can be the result of dysregulation of insulin-mediated signalling pathways. After cancer and heart disease, it is proven to be a worldwide epidemic that is the leading cause of death. The primary kind of diabetes that makes up roughly 90% of cases worldwide is type 2 diabetes mellitus (T2DM), sometimes referred to as non-insulin-dependent diabetes mellitus. Insulin resistance or a progressive reduction in β-cell insulin production is its defining characteristic. Age, food preferences, sedentary lifestyle, genetic susceptibility, hormonal conditions like PCOS, environmental variables, and behavioural changes are some of the causes [8,9].

Despite the global availability of various anti-hyperglycaemic agents, insulin analogs and oral drugs like metformin for diabetes and its secondary complications continue to be a major concern. Moreover, these drugs have adverse side effects and so naturally derived plant based chemical compounds are becoming popular in managing T2DM [10]. The medicinal importance of milk has been overlooked, and little has been done to promote milk-based functional products [11]. This present research work attempts to screen milk-based functional products and in-silico assessment of their interactions with common anti-diabetic targets like α-amylase and DPP-4 is used for molecular docking and ADMET pharmacokinetic studies. Nevertheless, in vitro or in vivo experimental data continues to be the primary source of precision and accuracy in the anticipation of the illicit substances with anti-diabetic potential.

2. METHODOLOGY

Accession of Cow Milk Volatile Organic Compounds And Anti-Diabetic Target Proteins: The three-dimensional structure of antidiabetic target proteins such as α-amylase (PDB_ID:2QV4) and dipeptidyl peptidase IV (PDB_ID: 2ONC) was downloaded from the RCSB protein database (https://www.rcsb.org/). Chemical structures of cow milk volatile organic compounds (Table SMT1: Types and Chemical components of VOCs in milk) used in the study were identified and reported by Yuan *et al*., 2023 and the PubChem database (https://pubchem.ncbi.nlm.nih.gov/) was used for accession of each chemical structure into SDF format from to study the possible molecular binding mechanism of the chosen organic volatile chemical compounds as potential inhibitors of target proteins as anti-diabetic therapeutics [12].

Molecular Docking: Molecular docking was performed to study the binding interactions between the cow milk volatile organic chemical compounds against α-amylase and DPP-4 proteins as potential anti-diabetic target proteins. 14 cow milk volatile compounds' ligands were subjected to molecular docking analysis with target proteins under control with standard FDA-approved drug inhibitors using cavity detection-guided blind docking (CB-Dock) [13]. The docked complex results were visualized using PyMol Viewer software and Accelrys Discovery Studio Client (version 21.1). Docking provides information about binding affinity and possible interactions that contribute to the inhibition and act as a potential anti-diabetic activity [14].

ADMET and Pharmacokinetic Analysis: The ADMET (absorption, distribution, metabolism, elimination, and toxicity) properties of the high binding energy volatile organic compounds against anti-diabetic targets compared with Metformin (Pub chem id: 4091) a FDA Approved oral anti-diabetic drug used for screening and prediction of the important pharmacokinetic properties by using *pKCSM* an online server (https://biosig.lab.uq.edu.au/pkcsm/prediction) [15]. The simplified molecular-input line-entry system (SMILES) formats each high binding energy milk volatile organic compounds which were retrieved from the PubChem server (https://pubchem.ncbi.nlm.nih.gov/) and ProTox 3.0 (https://comptox.charite.de/protox3/) a webserver was used for pharmacokinetic properties toxicological endpoints (Mutagenicity, Carcinogenicity, Immunotoxicity, Hepatotoxicity) and the level of toxicity (LD50, mg/Kg) was studied [16]. These predictions provide valuable insights into the drug-like properties of the compounds, aiding in the selection of candidates for further *in vitro* and *in vivo* evaluation.

3. RESULTS AND DISCUSSION

Diabetes is a metabolic condition defined by abnormal blood sugar levels. Targeting starch-hydrolyzing enzymes (α-amylase) and dipeptidyl peptidase 4 (DPP-4) expressed on the surface of numerous cells is one of the key strategies to lower the risk of Type-2 diabetes mellitus (T2DM). Molecular docking predicts the binding affinity and orientation of a small molecule (ligand) with a protein target (receptor). This helps identify potential drug candidates that can interact with specific proteins involved in diabetes, like DPP-IV and α-amylase. Out of 41 compounds, 13 compounds (Benzoic acid, Phenol, n-Decanoic acid, Dodecanoic acid, 1-Pentanol, 2-Ethyl-1-hexanol, Heptanoic acid, Nonanoic acid, 2-Ethyl-hexanoic acid, Hexanoic acid, Tetradecanoic acid, 2,3-Dimethyl-1-butanol, 2-Amino-5-methylbenzoic acid) passed the drug likeness (Lipinski rule) of milk volatile compounds. The detailed chemical structure and drug likeness are shown in Figure 1 and Table 1.

Table 1: Screening and list of drug likeness (Lipinski rule) passed Milk volatile compounds.

Sl. No	Compound name	PubChem ID	HBA	HBD	logP	MR	MW	TPSA
1	Benzoic acid	243	2	1	1.39	33.40	122.1	37.3
2	Phenol	996	1	1	1.39	28.47	94.11	20.2
3	n-Decanoic acid	2969	2	1	3.21	51.96	172.3	37.3
4	Dodecanoic acid	3893	2	1	3.99	61.57	200.3	37.3
5	1-Pentanol	6276	1	1	1.17	27.31	88.15	20.2
6	2-Ethyl-1-hexanol	7720	1	1	2.19	41.73	130.2	20.2
7	Heptanoic acid	8094	2	1	2.04	37.54	130.2	37.3
8	Nonanoic acid	8158	2	1	2.82	47.15	158.2	37.3
9	2-Ethyl-hexanoic acid	8697	2	1	2.29	42.34	144.2	37.3
10	Hexanoic acid	8892	2	1	1.65	32.73	116.2	37.3
11	Tetradecanoic acid	11005	2	1	4.77	71.18	228.4	37.3
12	2,3-Dimethyl-1-butanol	29656	1	1	1.27	32.12	102.2	20.2
13	2-Amino-5-methylbenzoic acid	76255	3	2	1.86	42.77	151.2	63.3
14	Metformin	4091	5	3	0.26	36.9	129.2	91.5

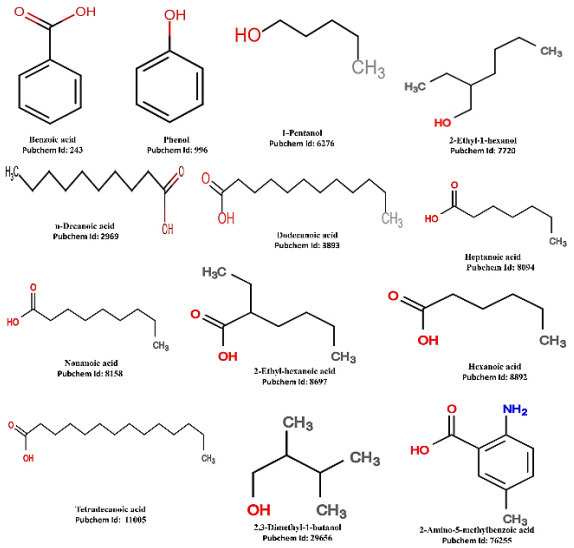


Figure 1: Structures of Bioactive volatile organic compounds from Cow milk.

Molecular docking analysis:

Molecular interactions of α-Amylase with Cow milk volatile organic compounds:

To aid in digestion, the calcium metalloenzyme α-amylase breaks down polysaccharide specifically starch and glycogen molecules into smaller ones, such as glucose and maltose [17]. The enzyme also raises postprandial

hyperglycemia and blood glucose levels. For the control and maintenance of postprandial blood glucose rise, α-amylase is also known as a therapeutic target. It has been shown that some enzymatic inhibitors, such as acarbose, miglitol, and voglibose, are effective in targeting **α** -amylase, which has prompted researchers to develop potent alpha-amylase inhibitor drugs [18, 19]. The role of alpha-amylase in diabetes is still centred on developing safer and more efficient inhibitors to regulate blood glucose levels in the wake of postprandial hyperglycemia. The ability of both natural and synthetic drugs to inhibit alpha-amylase slows down the breakdown of carbohydrates and reduces the amount of glucose absorbed into the bloodstream [20].

The Molecular interactions of cow milk volatile compounds with α-amylase were observed. The compounds with interactive amino acid of target protein and energy values are Benzoic acid (AspA:196;GluA:233;TyrA:62) (-5.3 kcal/mol), Phenol (HisA:299, TyrA:62) (-4.6 kcal/mol), n-Decanoic acid (HisA:215, AsnA:5) (-4.7 kcal/mol), Dodecanoic acid (HisA:305, GlyA:306, TyrA:62, TrpA:58, LeuA:165, TrpA:59) (-4.9 kcal/mol), 1-Pentanol (AsnA:250, LysA:227, LeuA:214, PreA:38) (-3.5 kcal/mol), 2-Ethyl-1-hexanol (AsnA:250, TyrA:2, ProA:228, LysA:227, LeuA:211, IleA:230) (-4.1 kcal/mol), Heptanoic acid (ArgA:421, AspA:402, ProA:332, PheA:335) (-4.4 kcal/mol), Nonanoic acid (ArgA:3146, AspA:317, IleA:312) (-4.6 kcal/mol), 2-Ethyl-hexanoic acid (AspA:300, TyrA:62) (-4.9 kcal/mol), Hexanoic acid (ProA:228, TyrA:2, LeuA:211, LysA:227, GlyA:251) (-4.3 kcal/mol) Tetradecanoic acid (GlyA:306, HisA:305, TyrA:62, TrpA:58, LeuA:165, TrpA:59) (-4.8 kcal/mol), 2,3-Dimethyl-1-butanol (AspA:197, AlaA:198, yrA:62) (-4.3 kcal/mol), 2-Amino-5-methylbenzoic acid (AspA:300, AspA:197, HisA:299, TyrA:62, TrpA:58) (-6 kcal/mol) and FDA approved antidiabetic drug molecule Metformin (AsnA:301, GlyA:309, AlaA:310, IleA:312, AspA:317) (-5.3 kcal/mol). The interaction of α-amylase molecular interactions and Binding Energy (kcal/mol) of screened milk volatile organic compounds were shown in Table 2 and Figure 2.

Table 2: α-Amylase molecular interactions and Binding Energy (kcal/mol) of screened milk volatile organic compounds.

Sl.No	Compound name	α-Amylase (PDB_ID:2QV4)		
		No of H bonds	H.Bonding interaction	Energy
1	Benzoic acid	2	AspA:196;GluA:233;TyrA:62	-5.3
2	Phenol	1	HisA:299;TyrA:62	-4.6
3	n-Decanoic acid	2	HisA:215;AsnA:5	-4.7
4	Dodecanoic acid	2	HisA:305;GlyA:306;TyrA:62;TrpA:58;LeuA:165;TrpA:59	-4.9
5	1-Pentanol	0	AsnA:250;LysA:227;LeuA:214;PreA:38	-3.5
6	2-Ethyl-1-hexanol	1	AsnA:250;TyrA:2;ProA:228;LysA:227;LeuA:211;IleA:230	-4.1
7	Heptanoic acid	3	ArgA:421;AspA:402;ProA:332;PheA:335;	-4.4
8	Nonanoic acid	2	ArgA:3146;AspA:317;IleA:312	-4.6
9	2-Ethyl-hexanoic acid	1	AspA:300;TyrA:62	-4.9
10	Hexanoic acid	2	ProA:228;TyrA:2;LeuA:211;LysA:227;GlyA:251	-4.3
11	Tetra decanoic acid	2	GlyA:306;HisA:305;TyrA:62;TrpA:58;LeuA:165;TrpA:59	-4.8
12	2,3-Dimethyl-1-butanol	1	AspA:197;AlaA:198;TyrA:62	-4.3
13	2-Amino-5-methylbenzoic acid	2	AspA:300;AspA:197;HisA:299;TyrA:62;TrpA:58	-6

14	Metformin	6	AsnA:301;GlyA:309;AlaA:310;IleA:312;AspA:317	-5.3

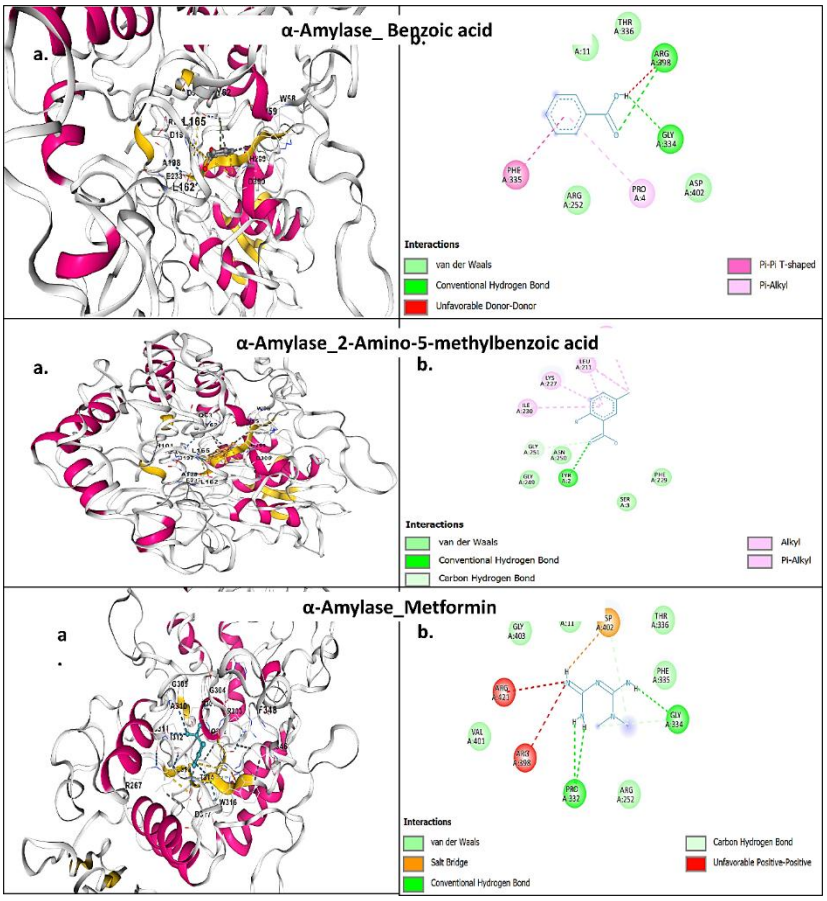


Figure 2: 2D and 3D molecular interactions of α-Amylase with high Binding Energy (kcal/mol) of screened milk volatile organic compounds.

Molecular interactions of DPP-4 with cow milk volatile organic compounds

Dipeptidyl peptidase 4 (DPP-4) is a protein that plays a variety of roles in inflammation, glucose metabolism, and other biological processes. This enzyme degrades incretin hormones, such as GLP-1, which increases the release of insulin and decreases the blood sugar level [21,22] DPP4 affects immune cells and other organs in a way that promotes inflammation. Due to its function in controlling GLP-1, DPP4 inhibition (DPP4i) has been investigated as a potential treatment target for type 2 diabetes [23,24].

The Molecular interactions of cowmilk volatile compounds with DPP4 was observed the compounds with interactive amino acid of target protein and energy values are Benzoic acid (GlnB:489, LysB:516, ArgB:522, AlaB,526, SerB:539, IleB:491) (-5.5kcal/mol), Phenol (IleB:491, SerB:539, ValB:537, AlaB:526) (-4.5 kcal/mol), n-Decanoic acid (TrpB:591, LysB:516, AspB:507, TrpB:589) (-5 kcal/mol), Dodecanoic acid (ArgB:631, ValB:618, TyrB:628, TyrB:593, PheB:319, TyrB:624) (-5.3 kcal/mol), 1-Pentanol (LysB:474, ArgB:522, GlnB:489, IleB:491) (-3.8 kcal/mol), 2-Ethyl-1-hexanol (TrpB:591, TrpB:589) (-4.4 kcal/mol), Heptanoic acid (TrpB:591, LysB:516, TyrB:714, TrpB:589) (-4.7 kcal/mol), Nonanoic acid (TrpB:591, LysB:516, ValB:508, TrpB:589) (-5.2 kcal/mol), 2-Ethyl-hexanoic acid (GlnB:489, ArgB:522, LysB:474, IleB:491, LysB:516) (-4.9 kcal/mol), Hexanoic acid (ValB:508, LySB:516, TrpB:591, TrpB:589, TyrB:714) (-4.6 kcal/mol), Tetra decanoic acid (TrpB:591, TrpB:589 (-5.2 kcal/mol), 2,3-Dimethyl-1-butanol (AspB:518, ArgB:522 (-4.2 kcal/mol), 2-Amino-5-methylbenzoic acid, (AspB:507, LysB:516, ValB:508, TrpB:591) (-5.8 kcal/mol), Metformin (ValB:537, LysB:516 -5.4 kcal/mol). The interaction of DPP-4 molecular interactions and Binding Energy (kcal/mol) of screened milk volatile organic compounds were shown in Table 3 and Figure 3.

Table 3: Dipeptidyl Peptidase 4 (DPP-4) molecular interactions and Binding Energy (kcal/mol) of screened milk volatile compounds

Sl.No	Compound name	No of H bonds	H.Bonding interaction	Energy
1	Benzoic acid	3	GlnB:489;LysB:516;ArgB:522;AlaB;526;SerB:539;IleB:491	-5.5
2	Phenol	0	IleB:491;SerB:539;ValB:537;AlaB:526	-4.5
3	n-Decanoic acid	2	TrpB:591;LysB:516;AspB:507;TrpB:589	-5
4	Dodecanoic acid	1	ArgB:631;ValB:618;TyrB:628;TyrB:593;PheB:319;TyrB:624	-5.3
5	1-Pentanol	2	LysB:474;ArgB:522;GlnB:489;IleB:491	-3.8
6	2-Ethyl-1-hexanol	1	TrpB:591;TrpB:589;	-4.4
7	Heptanoic acid	3	TrpB:591;LysB:516;TyrB:714;TrpB:589	-4.7
8	Nonanoic acid	3	TrpB:591;LysB:516;ValB:508;TrpB:589	-5.2
9	2-Ethyl-hexanoic acid	3	GlnB:489;ArgB:522;LysB:474;IleB:491;LysB:516	-4.9
10	Hexanoic acid	3	ValB:508;LySB:516;TrpB:591;TrpB:589;TyrB:714	-4.6
11	Tetra decanoic acid	1	TrpB:591;TrpB:589	-5.2
12	2,3-Dimethyl-1-butanol	1	AspB:518;ArgB:522	-4.2
13	2-Amino-5-methylbenzoic acid	3	AspB:507;LysB:516;ValB:508;TrpB:591	-5.8
14	Metformin	2	ValB:537;LysB:516	-5.4

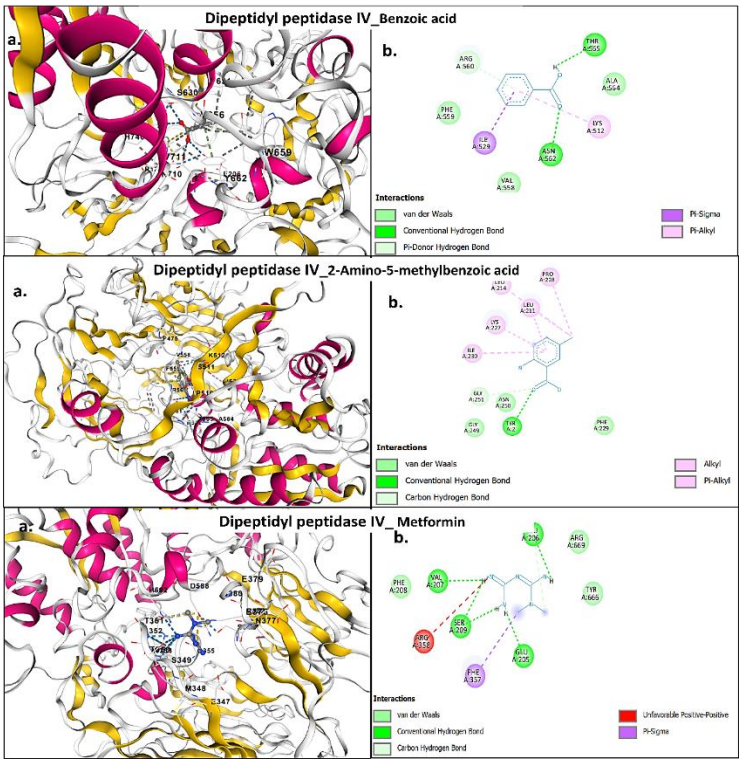


Figure 3: 2D and 3D molecular interactions of DPP-4 with high Binding Energy (kcal/mol) of screened milk volatile organic compounds

Pharmacophore ADMET Analysis:

Pharmacophore modelling, a key aspect of computational drug design, involves identifying the essential chemical features of a molecule that are crucial for its interaction with a biological target [2,6]. These features, represented in 2D or 3D, can be used to find new drug candidates with similar properties or to optimize existing ones. Pharmacophore models are also valuable for predicting off-target effects, absorption, distribution, metabolism, and toxicity (ADMET) properties, as well as for virtual screening and improving drug discovery processes.

Therapeutic development time and resources can be improved by using this integrated strategy to effectively screen and optimize therapeutic candidates [27].

ADMET analysis of highly interacting diabetic target chemical compounds with a standard drug molecule. Table 4 shows ADMET results of the Benzoic acid, Methyl benzoic acid from cow volatile organic compounds, and FDA-approved antidiabetic drug Metformin as the reference drug, whereas Figure 4 shows the bioavailability radar for the relevant metabolites and the reference drug. Figure 4 particularly illustrates 12 key points. The toxicity radar chart is intended to quickly illustrate the confidence of positive toxicity results. The ADMET results provide valuable insights into the drug-likeness and pharmacokinetic properties of the selected metabolites from cow milk volatile organic compounds compared to metformin.

Absorption is an important factor that influences a drug's efficacy and pharmacokinetics, or how it functions in the body. Water solubility, Caco-2 permeability, and intestinal absorption (in humans) are all associated with drug absorption into the bloodstream after oral administration. Skin permeability affects how topical drugs are distributed. A transporter protein called P-glycoprotein (P-gp) can affect how well medications are transported and absorbed. P-gp inhibitors can be used to make medications more bioavailable [28]. Benzoic acid shown Absorption Water solubility (-1.738 log mol/L), Caco2 permeability (1.707 log Papp in 10-6 cm/s), Intestinal absorption (human) (100%), Skin Permeability (-2.728 log Kp), Methylbenzoic acid shown Absorption Water solubility (-2.22 log mol/L), Caco2 permeability (0.662 log Papp in 10-6 cm/s), Intestinal absorption (human) (85.917%), Skin Permeability (-2.735 log Kp) and Metformin Water solubility (-2.707 log mol/L), Caco2 permeability (-0.339 log Papp in 10-6 cm/s), Intestinal absorption (human) (59.401%), Skin Permeability (-2.735log Kp).

Distribution is the potential for highly interacting cow milk volatile organic compounds which was assessed using the parameters of VDss (human), Fraction unbound (human), BBB permeability, and CNS permeability predicted by using the pkCSM platform. The results obtained are summarized in the table 4 . A pharmacokinetic measure that indicates the volume into which a medication dosage would need to be delivered in order to produce the same concentration observed in blood plasma, the volume of distribution (VDss) gives an idea of how the drug is distributed throughout the body. A high VDss implies a substantial concentration in tissues, for instance, because of tissue binding or high lipid solubility; a low VDss shows high water solubility or high plasma protein binding since more of the medication stays in the plasma [29].

Fu (unbound fraction) is also an important pharmacokinetic metric as it affects several elements of drug effectiveness and side effects (including glomerular filtration in the kidneys, total clearance, and hepatic metabolism). Therefore, it is essential to accurately predict Fu throughout drug research. Since unbound medications in plasma may interact with targets, including proteins, enzymes, receptors, and channels to exhibit pharmacological activity, the plasma unbound fraction (Fu) of a drug is an essential part of evaluating its effectiveness (Bowman *et al*., 2018; Chibuye *et al*., 2025). Benzoic acid distribution in VDss (human) (-1.64 log L/kg), Fraction unbound (human) (0.523 Fu), BBB permeability (-0.22 log BB), CNS permeability (-2.002 log PS), Methylbenzoic acid VDss (human) (-1.633 log L/kg), Fraction unbound (human) (0.552 Fu), BBB permeability (-0.37 log BB), CNS permeability (-2.3log PS) and Metformin (-0.232 log L/kg), Fraction unbound (human) (0.811 Fu), BBB permeability (-0.946 log BB), CNS permeability (-4.238 log PS).

Several drugs are substrates for CYP2D6 and CYP3A4, while others inhibit CYP1A2, CYP2C19, CYP2C9, CYP2D6, and CYP3A4. Substrates are metabolized by the enzyme, while inhibitors reduce the enzyme's activity. This information is crucial for understanding potential drug-drug interactions, as inhibitors can increase the levels of substrates in the body by slowing down their metabolism. The entire rate at which a medication is eliminated from the body, taking into account contributions from all elimination routes (renal, hepatic), is referred to as total clearance in ADMET. One of the kidneys' transporter proteins, OCT2, helps specific substances (substrates) enter kidney cells from the circulation. An important stage in renal excretion occurs when a medication is an OCT2 substrate, which indicates that it may be carried into the kidney cells by this transporter. It is possible to anticipate a drug's renal clearance, potential for drug-drug interactions, and general pharmacokinetic behaviour by looking at its ADMET qualities and determining if it is an OCT2 substrate [30,31]. Total Clearance for Benzoic acid (0.707 log ml/min/kg). Methylbenzoic acid (0.583 log ml/min/kg) and Metformin (0.1 log ml/min/kg) are observed.

The toxicity study of AMES toxicity, Max. tolerated dose (human), Oral Rat Acute Toxicity (LD50), Oral Rat Chronic Toxicity (LOAEL), T.Pyriformis toxicity, Minnow toxicity are observed for Benzoic acid is (0.612 log

mg/kg/day, 2.17 mol/kg, 2.637 log mg/kg bw/day, 0.087 log ug/L, 1.838 log mM), Methylbenzoic acid is (0.844 log mg/kg/day, 1.734 mol/kg, 1.875 log mg/kg_bw/day, 0.282 log ug/L, 1.911 log mM) and Metformin (0.902 log mg/kg/day, 2.453 mol/kg, 2.158 log mg/kg_bw/day, 0.25 log ug/L, 3.972 log mM). The toxicity predicted LD50 values of Benzoic acid - LD50:290mg/kg, Methylbenzoic acid - LD50: 1600mg/kg and Metformin - LD50: 680mg/kg.

Table 4: ADMET Analysis of highly interacted diabetic target chemical compounds with a standard drug molecule.

Model Name	Benzoic acid	Methyl benzoic acid	Metformin	Units
Absorption				
Water solubility	-1.738	-2.22	-2.707	Numeric (log mol/L)
Caco2 permeability	1.707	0.662	-0.339	Numeric (log Papp in 10-6 cm/s)
Intestinal absorption (human)	100	85.917	59.401	Numeric (% Absorbed)
Skin Permeability	-2.728	-2.735	-2.735	Numeric (log Kp)
P-glycoprotein substrate	No	No	Yes	Categorical (Yes/No)
P-glycoprotein I inhibitor	No	No	No	Categorical (Yes/No)
P-glycoprotein II inhibitor	No	No	No	Categorical (Yes/No)
Distribution				
VDss (human)	-1.64	-1.633	-0.232	Numeric (log L/kg)
Fraction unbound (human)	0.523	0.552	0.811	Numeric (Fu)
BBB permeability	-0.22	-0.37	-0.946	Numeric (log BB)
CNS permeability	-2.002	-2.3	-4.238	Numeric (log PS)
Metabolism				
CYP2D6 substrate	No	No	No	Categorical (Yes/No)
CYP3A4 substrate	No	No	No	Categorical (Yes/No)
CYP1A2 inhibitor	No	No	No	Categorical (Yes/No)
CYP2C19 inhibitor	No	No	No	Categorical (Yes/No)
CYP2C9 inhibitor	No	No	No	Categorical (Yes/No)
CYP2D6 inhibitor	No	No	No	Categorical (Yes/No)
CYP3A4 inhibitor	No	No	No	Categorical (Yes/No)
Excretion				
Total Clearance	0.707	0.583	0.1	Numeric (log ml/min/kg)
Renal OCT2 substrate	No	No	No	Categorical (Yes/No)
Toxicity				
AMES toxicity	No	Yes	Yes	Categorical (Yes/No)
Max. tolerated dose (human)	0.612	0.844	0.902	Numeric (log mg/kg/day)
hERG I inhibitor	No	No	No	Categorical (Yes/No)
hERG II inhibitor	No	No	No	Categorical (Yes/No)

Oral Rat Acute Toxicity (LD50)	2.17	1.734	2.453	Numeric (mol/kg)
Oral Rat Chronic Toxicity (LOAEL)	2.637	1.875	2.158	Numeric (log mg/kg_bw/day)
Hepatotoxicity	No	No	No	Categorical (Yes/No)
Skin Sensitisation	No	No	Yes	Categorical (Yes/No)
T.Pyriformis toxicity	0.087	0.282	0.25	Numeric (log ug/L)
Minnow toxicity	1.838	1.911	3.972	Numeric (log mM)

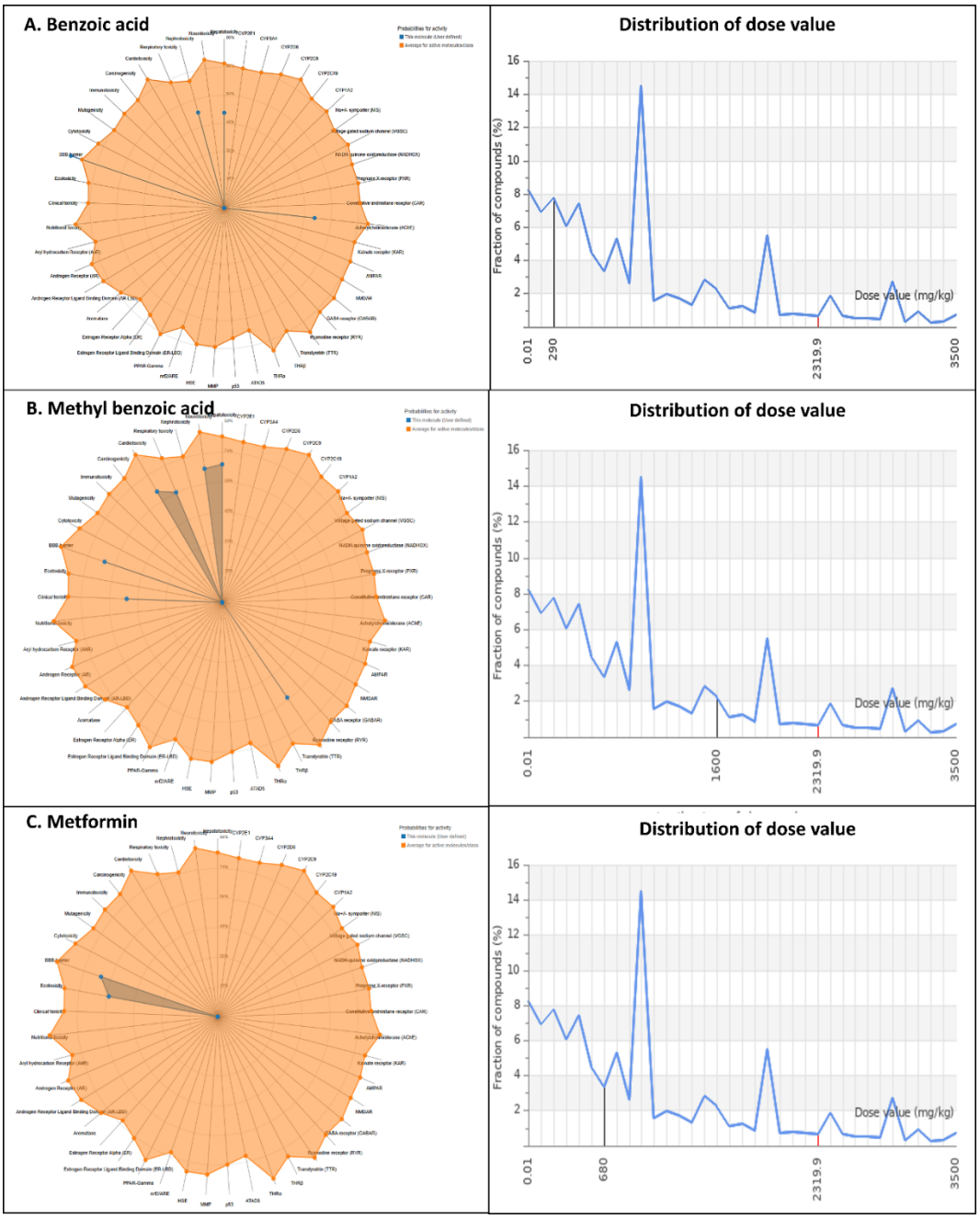


Figure 4: Toxicity radar chart and Distribution of dose value of A. Benzoic acid, B. Methylbenzoic acid and C. Metformin.

4. CONCLUSION

In conclusion we screened by identifying 13 volatile organic compounds in cow's milk and their anti-diabetic effects by targeting enzymes (α-amylase) and dipeptidyl peptidase 4 (DPP-4) proteins involved in starch digestion and glucose regulation. Over all computational predictions along with the reported pharmacological properties postulated that the Benzoic acid, 2-Ethyl-hexanoic acid, 2-Amino-5-methylbenzoic acid are found to attach at the active site of anti-diabetic target proteins. We believe the medicinal advantages of cow milk derived volatile organic compounds beyond its traditional nutritional value and develop functional products with therapeutic applications against type-II diabetes.

Conflict of Interest and Declarations

Authors Contribution

VSM, **MR** and **SSM** designed the research concept, executed the experiments and data analysis; and prepared the manuscript; **KKB** helped in Docking studies and proof correction. All authors read and approved the final manuscript.

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Compliance with Ethical Standards

Conflict of Interest: The authors state that they don't have any conflict of interest.

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