ARTICLE

Open access in silico tools to predict the ADMET profiling and PASS (Prediction of Activity Spectra for Substances of Bioactive compounds of Garlic (Allium sativum L.)

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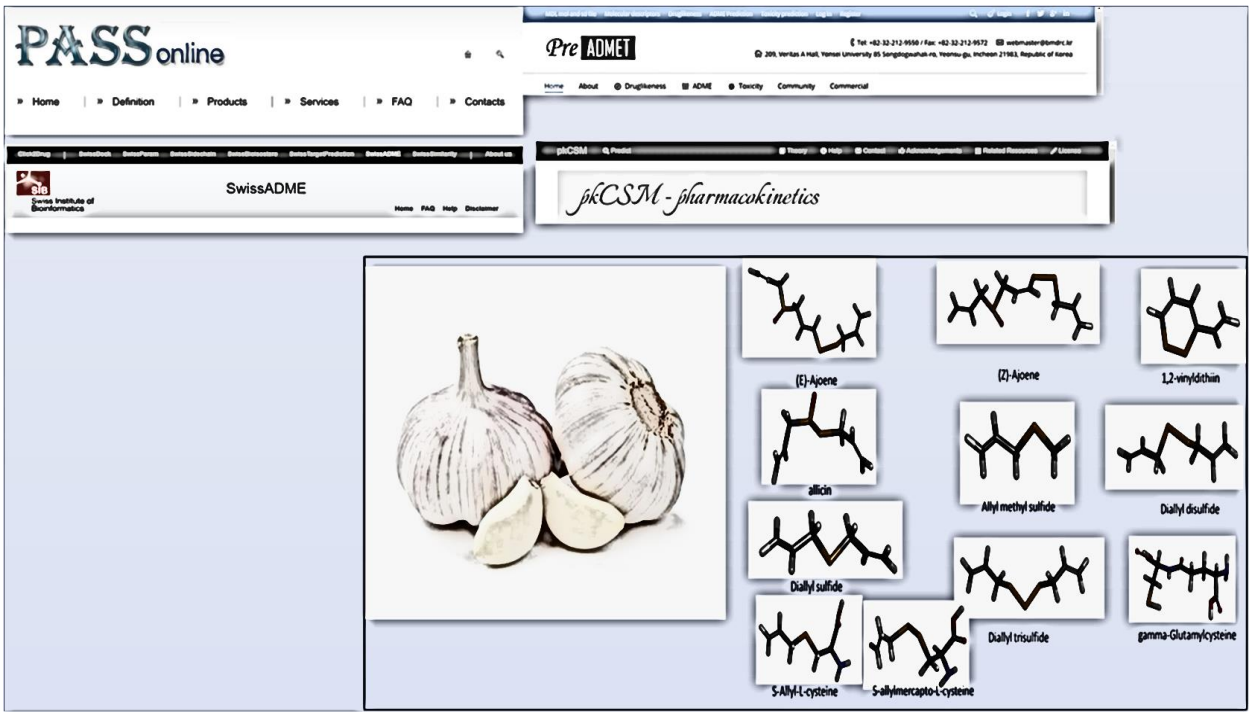
Graphical abstract



Keywords: in silico; ADMET; drug; open access; prediction; PASS Online ((Prediction of Activity Spectra for Substances) , Garlic Compounds; S-allylcysteine (SAC) and S-allylmercaptocysteine (SAMC)

Abstract

Background: Garlic (Allium sativum L.) is a common spice with many health benefits, mainly due to its diverse bioactive compounds, (see below) such as organic sulphides, saponins, phenolic compounds, and polysaccharides. Several studies have demonstrated its functions such as anti-inflammatory, antibacterial, and antiviral, antioxidant, cardiovascular protective and anticancer

property. In this work we have investigated the main bioactive components of garlic through a bioinformatics approach. Indeed, we are in an era of bioinformatics where we can predict data in the fields of medicine. Approaches with open access in silico tools have revolutionized disease management due to early prediction of the absorption, distribution, metabolism, excretion, and toxicity (ADMET) profiles of the chemically designed and eco-friendly next-generation drugs. **Methods:** This paper encompasses the fundamental functions of open access in silico prediction tools, as PASS database (Prediction of Activity Spectra for Substances) that it estimates the probable biological activity profiles for compounds. This paper also aims to help support new researchers in the field of drug design and to investigate best bioactive compounds in garlic. **Results:** screening through each of pharmacokinetic criteria resulted in identification of Garlic compounds that adhere to all the ADMET properties. **Conclusions:** It was established an open-access database (PASS database, available bioinformatics tool SwissADME, PreADMET pkCSM database) servers were employed to determine the ADMET (metabolism, distribution, excretion, absorption, and toxicity) attributes of garlic molecules and to enable identification of promising molecules that follow ADMET properties.

1. Introduction

Garlic (Allium sativum L.) is a species in the onion genus Allium. This is a common spice with many health benefits, mainly due to its diverse bioactive compounds, (see below fig.1) such as organic sulphides, saponins, phenolic compounds, and polysaccharides. Garlic has been demonstrated to exhibit potentially beneficial for cancer prevention. Several studies have demonstrated its functions such as anti-inflammatory, antibacterial, and antiviral, antioxidant, cardiovascular protective. anticancer property. [1-2] Observations over the past years have shown that the consumption of garlic in the diet provides strong protection against cancer risk. [2]. In literature we can find some papers, where it was demonstrated decreased rates stomach cancer associated with garlic intake. [3-5]

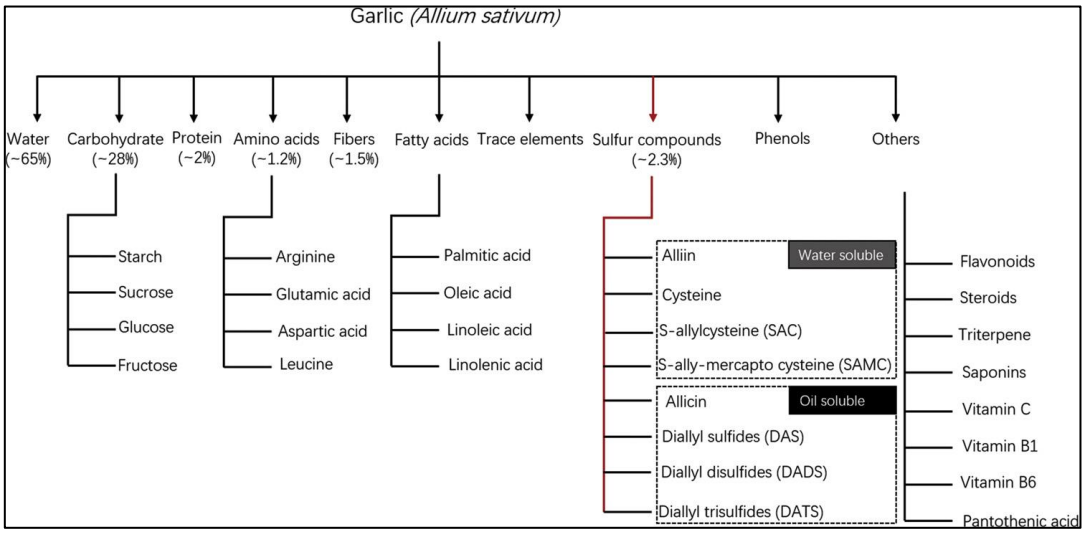


Fig. 1. Major classification of the bioactive constituents in garlic. Generally, garlic bulb contains approximately 65 % water, 28 % carbohydrates (mainly fructans), 2 % protein (mainly alliin), 1.2 % free amino acids (mainly arginine), 1.5 % fiber, and 2.3 %organosulfur compounds [2]

In this work we have investigated the main bioactive components of garlic through a bioinformatics approach. Indeed, we are in an era of bioinformatics where we can predict data in the fields of medicine. Approaches with open access in silico tools have revolutionized disease management due to early prediction of the absorption, distribution, metabolism, excretion, and toxicity (ADMET) profiles of the chemically designed and eco-friendly next-generation drugs. [6-7] This paper encompasses the fundamental functions of open access in silico prediction tools, as PASS database (Prediction of Activity Spectra for Substances) that it estimates the probable biological activity profiles for compounds. [8-9] This paper also aims to help support the researchers in the field of drug design and to investigate best bioactive compounds in garlic. As it has been before, Garlic contains 0.1-0.36% of a volatile oil these volatile compounds are generally considered to be responsible for most of the pharmacological properties of garlic. Garlic contains at least 33 sulfur compounds like aliin, allicin, ajoene, allylpropl, diallyl, trisulfide, s-allylcysteine, vinyldithiines, S-allylmercaptocystein, and others. Particular attention has been given to sulphide compounds of garlic for their anti-tumour properties, S-allylcysteine (SAC) and S-allylmercaptocysteine (SAMC) (See below fig.1-2) [10-20] It was established an open-access database (PASS database, available bioinformatics tool SwissADME, PreADMET pkCSM database) servers were employed to determine the ADMET (metabolism, distribution, excretion, absorption, and toxicity) attributes of garlic molecules and to enable identification of promising molecules that follow ADMET properties. [6-9]

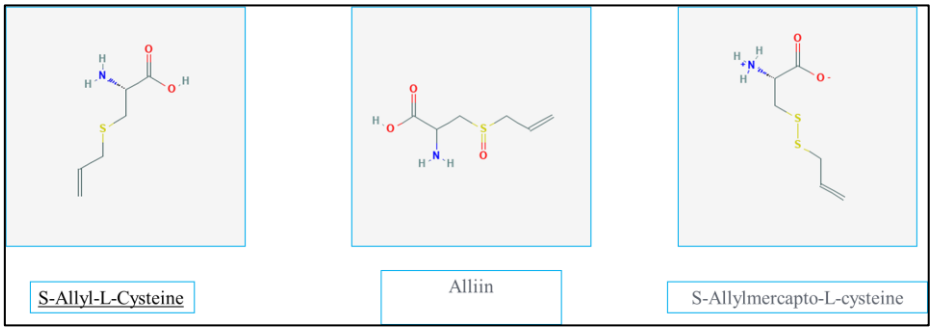


Fig 2 bioactive constituents in garlic sulphur compounds

2. Materials and methods

This paper encompasses the fundamental functions of open access in silico prediction tools, as PASS database, PreADMET and pkCSM (http://biosig.unimelb.edu.au/pkcsm/) servers were employed to determine the ADMET (metabolism, distribution, excretion, absorption, and toxicity) attributes of target molecules. PASS (Prediction of Activity Spectra for Substances) Online predicts over 4000 kinds of biological activity, including pharmacological effects, mechanisms of action, toxic and adverse effects, interaction with metabolic enzymes and transporters, influence on gene expression, etc. The first strategy is based on the suggestion that the more kinds of activity are predicted as probable for a compound, the more probable to find any useful pharmacological action in it. Prediction is based on the analysis of structure activity-relationships for more than 250,000 biologically active substances including drugs and drug-candidates. [8] The available bioinformatics tool SwissADME (http://www.swissadme.ch/index.php) [7] was used for finding drug-likeness attributes. Lipinski's rule of five [6] was used to analyze the properties such as; hydrogen bond donor (HBD), hydrogen bond acceptor (HBA), molecular weight (MW), and lipophilicity (log P).

PreADMET (https://preadmet.bmdrc.kr/) and pkCSM (http://biosig.unimelb.edu.au/pkcsm/) servers were employed to determine the ADMET (metabolism, distribution, excretion, absorption, and toxicity) attributes of target molecules.

2.1 Classification of the bioactive constituents in garlic

3D Conformer of several bioactive constituents in garlic were download from PubChem (https://pubchem.ncbi.nlm.nih.gov/) and create by Discovery Studio Biovia Visualizer Software [21] (see fig.3)

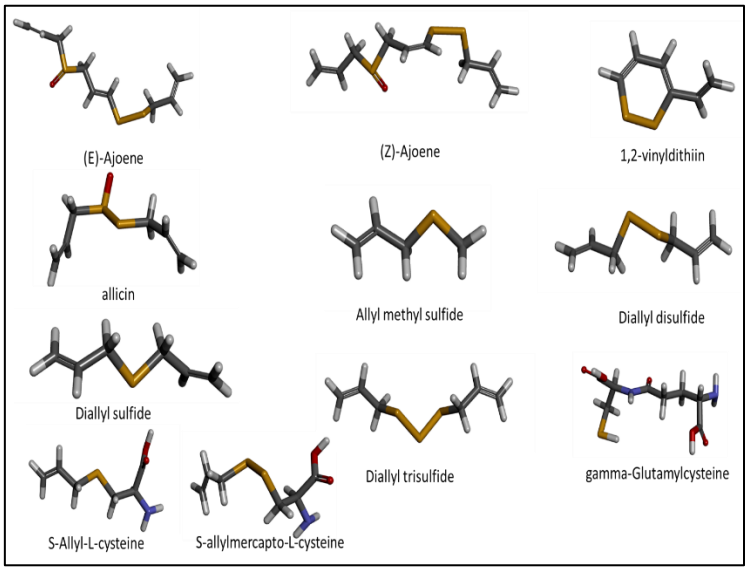


Fig 3 Principal Organosulfur Compounds From Garlic. Reproduced by Discovery Studio Biovia Visualizer Software [21]

3. Results and discussion

3.1 PASS database (Prediction of Activity Spectra for Substances)

PASS Online predicts over 3500 kinds of biological activity, including pharmacological effects, mechanisms of action, toxic and adverse effects, interaction with metabolic enzymes and transporters, influence on gene expression, etc. Prediction is based on the analysis of structure activity-relationships for more than 250,000 biologically active substances including drugs, drug-candidates, leads and toxic compounds. The concept of the biological activity spectrum was introduced to describe the properties of biologically active substances. The PASS (prediction of activity spectra for substances) software product, which predicts more than 300 pharmacological effects and biochemical mechanisms on the basis of the structural formula of a substance, may be efficiently used to find new targets (mechanisms) for some ligands and, conversely, to reveal new ligands for some biological targets. Average accuracy of prediction estimated in leave-one-out cross-validation procedure (each compound is excluded from the training set and its activity predicted based on SAR model obtained on the rest part of the training set) for the whole PASS training set is about 95% (Filimonov and Poroikov, 2008) [22] . Since PASS service is used by medicinal chemists, pharmacologists and toxicologists for several years (Lagunin et al., 2000) [23], there are many publications where PASS predictions were confirmed by subsequent synthesis and biological testing. To provide more accurate predictions for compounds belonging to new chemical classes and to extend the predictable area onto

new biological activities, we are permanently working on enlargement of PASS training set. Input data represents a structural formula of a compound in MOL file format. The output file represents a list of activities with two probabilities Pa (probability to be active) and Pi (probability to be inactive). Pa (probability "to be active") estimates the chance that the studied compound is belonging to the sub-class of active compounds (resembles the structures of molecules, which are the most typical in a sub-set of "actives" in PASS training set). Pi (probability "to be inactive") estimates the chance that the studied compound is belonging to the sub-class of inactive compounds (resembles the structures of molecules, which are the most typical in a sub-set of "inactives" in PASS training set). The first strategy is based on the suggestion that the more kinds of activity are predicted as probable for a compound, the more probable to find any useful pharmacological action in it. For each compound from available set of samples the following value can be calculated: [8-9] ; [22-23]

$$\mathbf{P} = \frac{\mathbf{1}}{\boldsymbol{n}} \sum \frac{\boldsymbol{P_a}}{\boldsymbol{P_a} + \boldsymbol{P_i}}$$

Fig 4 Prediction of Activity Spectra for Substances calculated : the output file represents a list of activities with two probabilities Pa (probability to be active) and Pi (probability to be inactive [8-9] ; [22-23]

In Tab 1 we report chemical-physical properties of Principal Organosulfur Compounds , S-allylcysteine (SAC) and S-allylmercaptocysteine (SAMC) in Garlic investigated by Pass Online Server (Prediction of Activity Spectra for Substances) that it estimates the probable biological activity profiles for compounds. As we can see from the table 1, SAMC and SAC demonstrated to a suppressive agent against several tumours and they have several functions such as anti-inflammatory, antibacterial, and antiviral, antioxidant, cardiovascular protective. [10-20] S-allylcysteine (SAC) and S-allylmercaptocysteine (SAMC) have a high value of 0.96-0.98 Pa (probability to be active) in human flavin-containing monooxygenase 3 (FMO3) and it has impact on enzyme activity, drug metabolism and disease. [28-29] Indeed, A flavin-containing monooxygenase (FMO) produced by A. sativum (AsFMO) was previously proposed to oxidize S-allyl-L-cysteine (SAC) to alliin, an allicin precursor. 30. Ferreira F, et all., (2013) have investigated the activity of the human flavin-containing monooxygenase (FMO) has been proposed to be impact on enzyme activity, drug metabolism and disease, like Trimethylaminuria (TMAu) or "fish odor syndrome" is a metabolic disorder characterized by the inability to convert malodorous dietarily-derived trimethylamine (TMA) to odourless TMA N-oxide by the flavin-containing monooxygenase 3 (FMO3). [30]

S-Allyl-L-cysteine	$C_6H_{11}NO_2S$	161.22	Pa	Pi	Acitivity
			0,965	0,002	Flavin-containing monooxygenase substrate
			0,963	0,003	CYP2E1 substrate
			0,962	0,003	CYP2E substrate
			0,942	0,001	S-alkylcysteine lyase inhibitor
			0,939	0,002	NADPH peroxidase inhibitor
			0,933	0,002	FMO3 substrate
			0,928	0,002	Lysine 2,3-aminomutase inhibitor
			0,924	0,003	Acylcarnitine hydrolase inhibitor
			0,905	0,004	Arylacetonitrilase inhibitor
S-allylmercapto-L-cysteine	$C_6H_{11}NO_2S_2$	193.3	Pa	Pi	Acitivity
			0,974	0,002	Flavin-containing monooxygenase substrate
			0,967	0,001	FMO3 substrate
			0,939	0,002	NADPH peroxidase inhibitor
			0,939	0,002	Protein-disulfide reductase (glutathione) inhibitor
			0,934	0,003	CYP2E1 substrate
			0,933	0,003	CYP2E substrate
			0,932	0,003	Antiseborrheic
			0,928	0,003	Lipid metabolism regulator
			0,925	0,004	Apoptosis agonist
			0,924	0,003	Acylcarnitine hydrolase inhibitor
			0,919	0,001	Cysteamine dioxygenase inhibitor
			0,916	0,003	Arylacetonitrilase inhibitor
			0,909	0,002	S-alkylcysteine lyase inhibitor
			0,903	0,005	TP53 expression enhancer

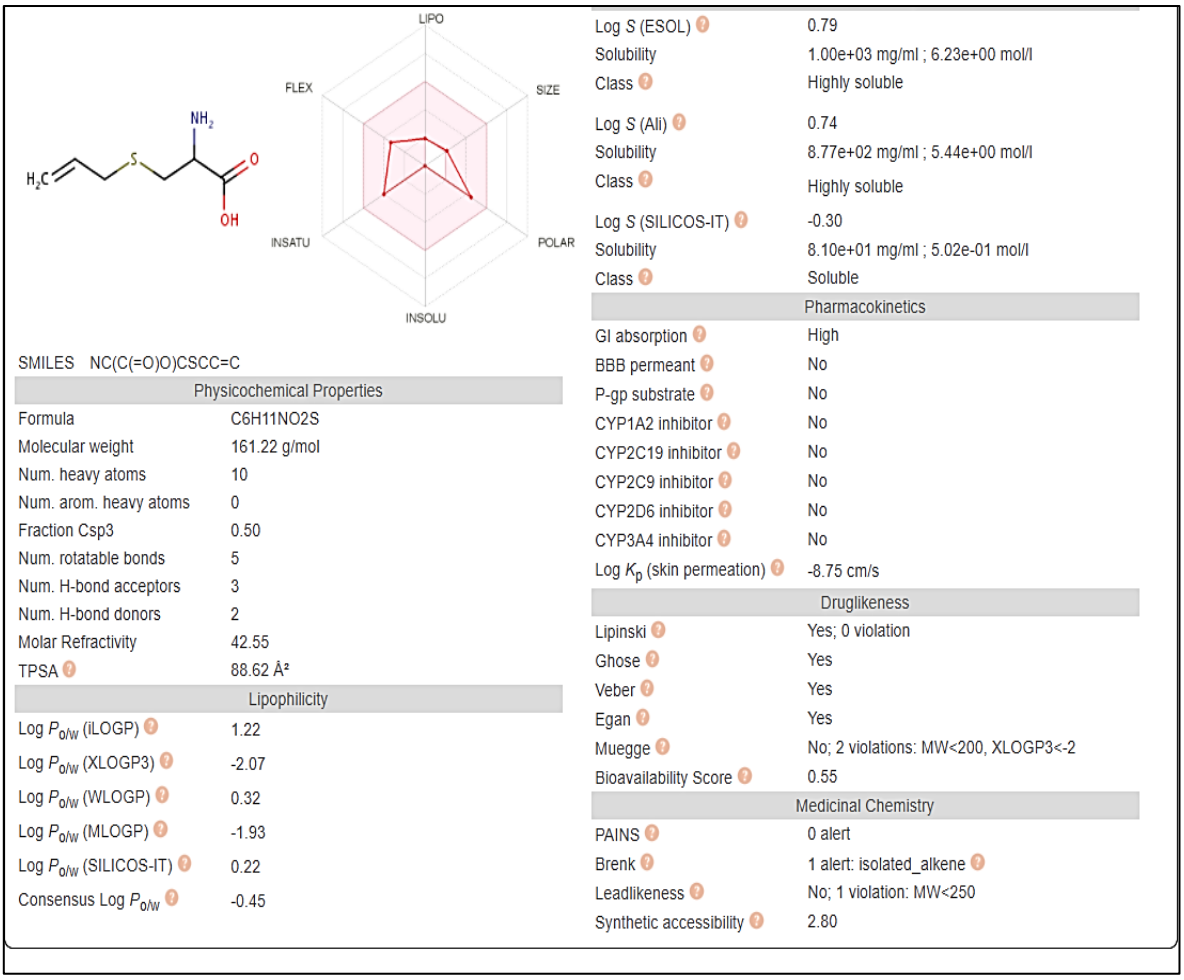
Tab 1 Prediction of Activity Spectra for Substances of Principal Organosulfur Compounds , as S-allylcysteine (SAC) and S-allylmercaptocysteine (SAMC)

3.2 In silico Organosulfur Compounds analysis and ADMET profiling

The available bioinformatics tool SwissADME (http://www.swissadme.ch/index.php) [7,24] was used for finding drug-likeness attributes. Lipinski's rule of five [6] was used to analyze the properties such as; hydrogen bond donor (HBD), hydrogen bond acceptor (HBA), molecular weight (MW), and lipophilicity (log P). PreADMET (https://preadmet.bmdrc.kr/) and pkCSM (http://biosig.unimelb.edu.au/pkcsm/) servers were employed to determine the ADMET (metabolism, distribution, excretion, absorption, and toxicity) attributes of target molecules.

3.2.1 SwissADME (http://www.swissadme.ch/index.php)

This website allows you to compute physicochemical descriptors as well as to predict ADME parameters, pharmacokinetic properties, druglike nature and medicinal chemistry friendliness of one or multiple small molecules to support drug discovery. This web service is a free web tool to evaluate pharmacokinetics, drug-likeness and medicinal chemistry friendliness of small molecules.[24]



SMILES NC(C(=O)O)CSCC=C

Physicochemical Properties	
Formula	C6H11NO2S
Molecular weight	161.22 g/mol
Num. heavy atoms	10
Num. arom. heavy atoms	0
Fraction Csp3	0.50
Num. rotatable bonds	5
Num. H-bond acceptors	3
Num. H-bond donors	2
Molar Refractivity	42.55
TPSA	88.62 Å²

Lipophilicity	
Log $P_{o/w}$ (iLOGP)	1.22
Log $P_{o/w}$ (XLOGP3)	-2.07
Log $P_{o/w}$ (WLOGP)	0.32
Log $P_{o/w}$ (MLOGP)	-1.93
Log $P_{o/w}$ (SILICOS-IT)	0.22
Consensus Log $P_{o/w}$	-0.45

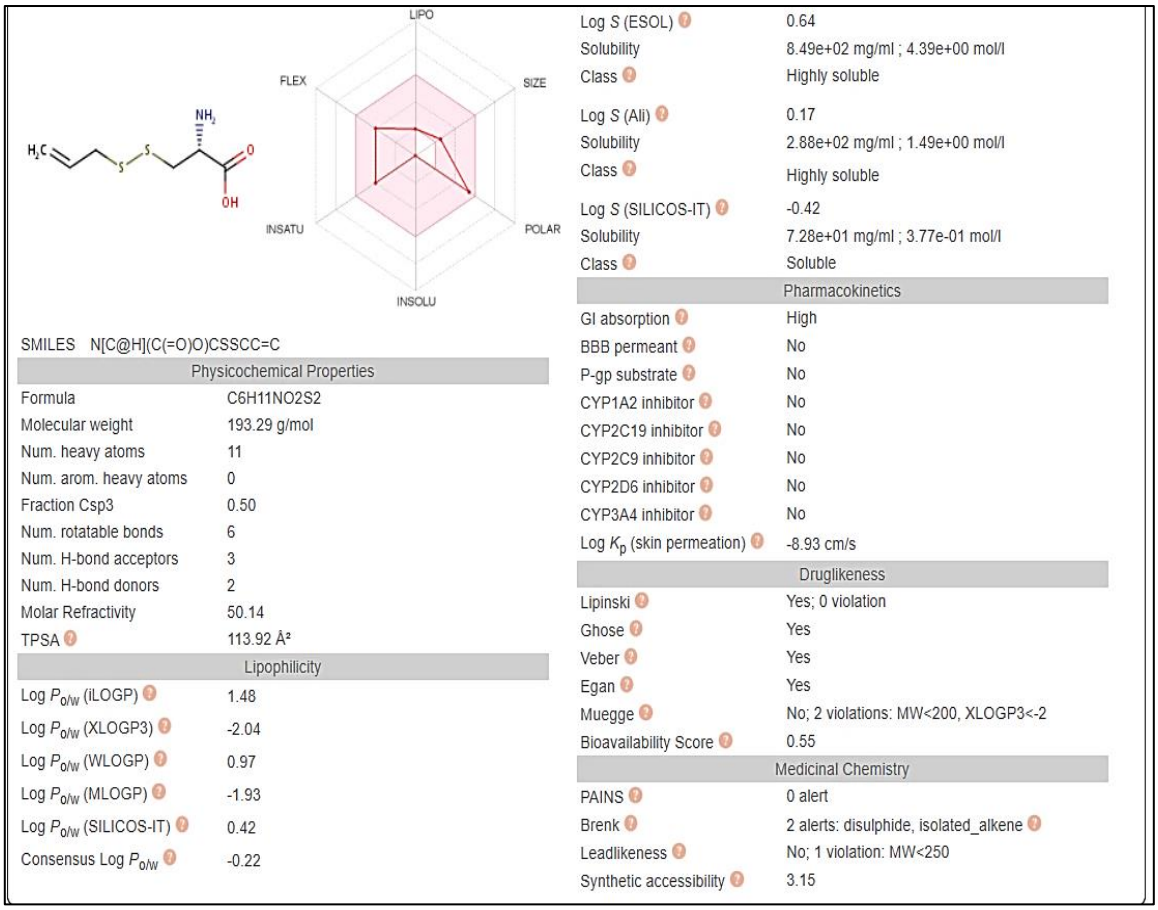
Log *S* (ESOL)	0.79
Solubility	1.00e+03 mg/ml ; 6.23e+00 mol/l
Class	Highly soluble
Log *S* (Ali)	0.74
Solubility	8.77e+02 mg/ml ; 5.44e+00 mol/l
Class	Highly soluble
Log *S* (SILICOS-IT)	-0.30
Solubility	8.10e+01 mg/ml ; 5.02e-01 mol/l
Class	Soluble

Pharmacokinetics	
GI absorption	High
BBB permeant	No
P-gp substrate	No
CYP1A2 inhibitor	No
CYP2C19 inhibitor	No
CYP2C9 inhibitor	No
CYP2D6 inhibitor	No
CYP3A4 inhibitor	No
Log K_p (skin permeation)	-8.75 cm/s

Druglikeness	
Lipinski	Yes; 0 violation
Ghose	Yes
Veber	Yes
Egan	Yes
Muegge	No; 2 violations: MW<200, XLOGP3<-2
Bioavailability Score	0.55

Medicinal Chemistry	
PAINS	0 alert
Brenk	1 alert: isolated_alkene
Leadlikeness	No; 1 violation: MW<250
Synthetic accessibility	2.80

Fig 5 physicochemical descriptor , ADME parameters, pharmacokinetic properties, druglike nature and medicinal chemistry friendliness of S-allylcysteine (SAC) predicted by SwissADME Database SwissADME (http://www.swissadme.ch/index.php)



SMILES N[C@H](C(=O)O)CSSCC=C

Physicochemical Properties	
Formula	C6H11NO2S2
Molecular weight	193.29 g/mol
Num. heavy atoms	11
Num. arom. heavy atoms	0
Fraction Csp3	0.50
Num. rotatable bonds	6
Num. H-bond acceptors	3
Num. H-bond donors	2
Molar Refractivity	50.14
TPSA	113.92 Å²
Lipophilicity	
Log $P_{o/w}$ (iLOGP)	1.48
Log $P_{o/w}$ (XLOGP3)	-2.04
Log $P_{o/w}$ (WLOGP)	0.97
Log $P_{o/w}$ (MLOGP)	-1.93
Log $P_{o/w}$ (SILICOS-IT)	0.42
Consensus Log $P_{o/w}$	-0.22

Log *S* (ESOL)	0.64
Solubility	8.49e+02 mg/ml ; 4.39e+00 mol/l
Class	Highly soluble
Log *S* (Ali)	0.17
Solubility	2.88e+02 mg/ml ; 1.49e+00 mol/l
Class	Highly soluble
Log *S* (SILICOS-IT)	-0.42
Solubility	7.28e+01 mg/ml ; 3.77e-01 mol/l
Class	Soluble
Pharmacokinetics	
GI absorption	High
BBB permeant	No
P-gp substrate	No
CYP1A2 inhibitor	No
CYP2C19 inhibitor	No
CYP2C9 inhibitor	No
CYP2D6 inhibitor	No
CYP3A4 inhibitor	No
Log K_p (skin permeation)	-8.93 cm/s
Druglikeness	
Lipinski	Yes; 0 violation
Ghose	Yes
Veber	Yes
Egan	Yes
Muegge	No; 2 violations: MW<200, XLOGP3<-2
Bioavailability Score	0.55
Medicinal Chemistry	
PAINS	0 alert
Brenk	2 alerts: disulphide, isolated_alkene
Leadlikeness	No; 1 violation: MW<250
Synthetic accessibility	3.15

Fig 6 physicochemical descriptors) , ADME parameters, pharmacokinetic properties, druglike nature and medicinal chemistry friendliness of S-allylmercaptocysteine (SAMC) predicted by SwissADME Database SwissADME (http://www.swissadme.ch/index.php)

3.2.2. PreADMET (https://preadmet.bmdrc.kr/)

PreADMET is a web-based application for predicting ADME data and building drug-like library using in silico method. It was describe a new web-based application called PreADMET, which has been developed in response to a need for rapid prediction of drug-likeness and ADME/Tox data. [25-26] In figure 7 we report ADMET (metabolism, distribution, excretion, absorption, and toxicity) attributes of S-allylcysteine (SAC)) predicted by PreADMET (https://preadmet.bmdrc.kr/):

- BBB (in vivo blood-brain barrier penetration (C.brain/C.blood) >> 0.229899 Value
- Calculated water solubility value in buffer system by SK atomic types (mg/L) >> 92985.4 Value
- in vitro Caco2 cell permeability (Human colorectal carcinoma; nm/sec) >> 6.65342 Value
- in vitro Cytochrome P450 2C19 inhibition >> Inhibitor
- in vitro Cytochrome P450 2C9 inhibition >> Inhibitor
- in vitro Cytochrome P450 2D6 inhibition >> Inhibitor
- in vitro Cytochrome P450 2D6 substrate >> No value
- in vitro Cytochrome P450 3A4 inhibition >> No value
- Human intestinal absorption (HIA, %) >> 81.972219 Value
- in vitro MDCK cell permeability (Mandin Darby Canine Kidney) >> 209.694
- in vitro P-glecoprotein inhibition>> No value
- in vitro plasma protein binding (%) >> 11.674627
- in vitro skin permeability (transdermal delivery) >> -3.10116 Value

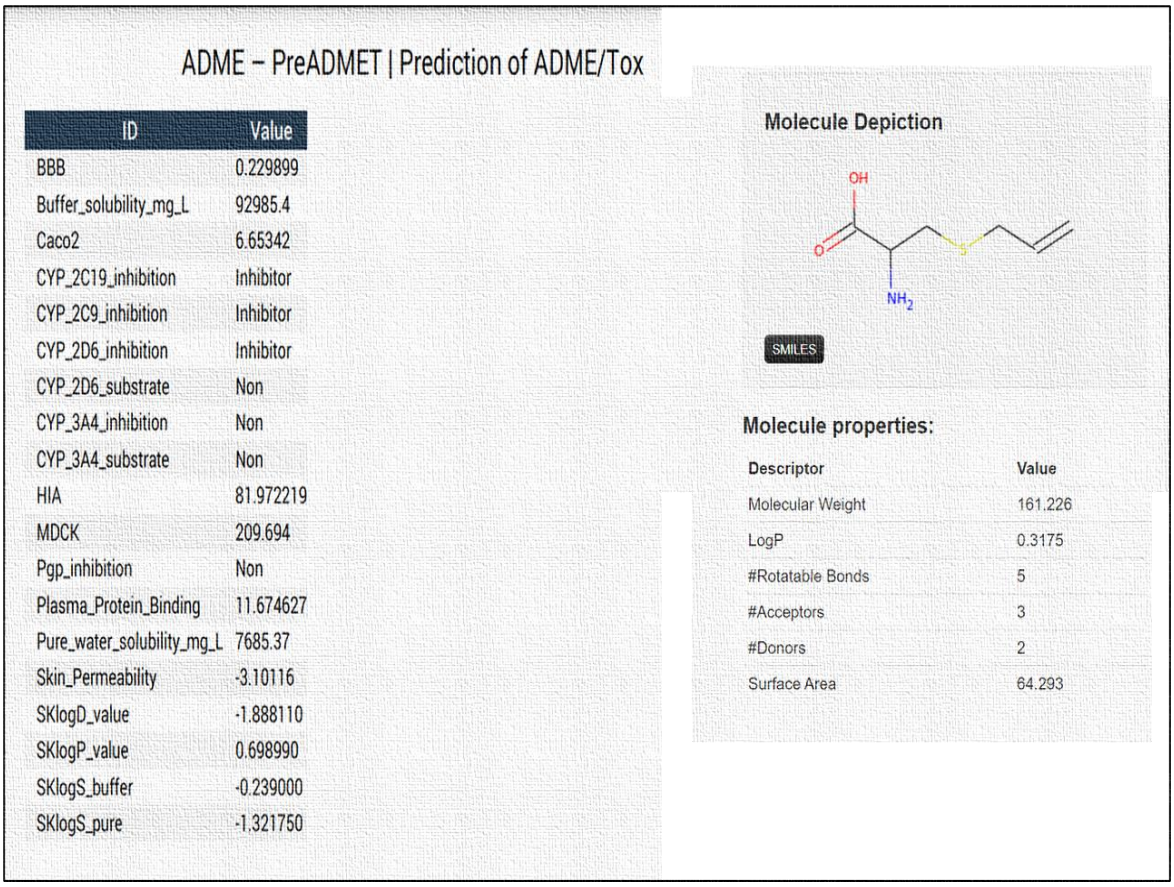


Fig 7 ADMET (metabolism, distribution, excretion, absorption, and toxicity) attributes of S-allylcysteine (SAC) predicted by PreADMET (https://preadmet.bmdrc.kr/)

In figure 8 we report ADMET (metabolism, distribution, excretion, absorption, and toxicity) attributes of S-allylmercaptocysteine (SAMC) predicted by PreADMET (https://preadmet.bmdrc.kr/):

- BBB (in vivo blood-brain barrier penetration (C.brain/C.blood) >> 0.190643 Value
- Calculated water solubility value in buffer system by SK atomic types (mg/L) >> 2537.48 Value
- in vitro Caco2 cell permeability (Human colorectal carcinoma; nm/sec) >> 1.60099 Value
- in vitro Cytochrome P450 2C19 inhibition >> Inhibitor
- in vitro Cytochrome P450 2C9 inhibition >> Inhibitor
- in vitro Cytochrome P450 2D6 inhibition >> Weakly
- in vitro Cytochrome P450 2D6 substrate >> No value
- in vitro Cytochrome P450 3A4 inhibition >> No value
- Human intestinal absorption (HIA, %) >> 82.576618 Value
- in vitro MDCK cell permeability (Mandin Darby Canine Kidney) >> 201.239
- in vitro P-glecoprotein inhibition>> No value
- in vitro plasma protein binding (%) >> 0.000000
- in vitro skin permeability (transdermal delivery) >> -2.8397 Value

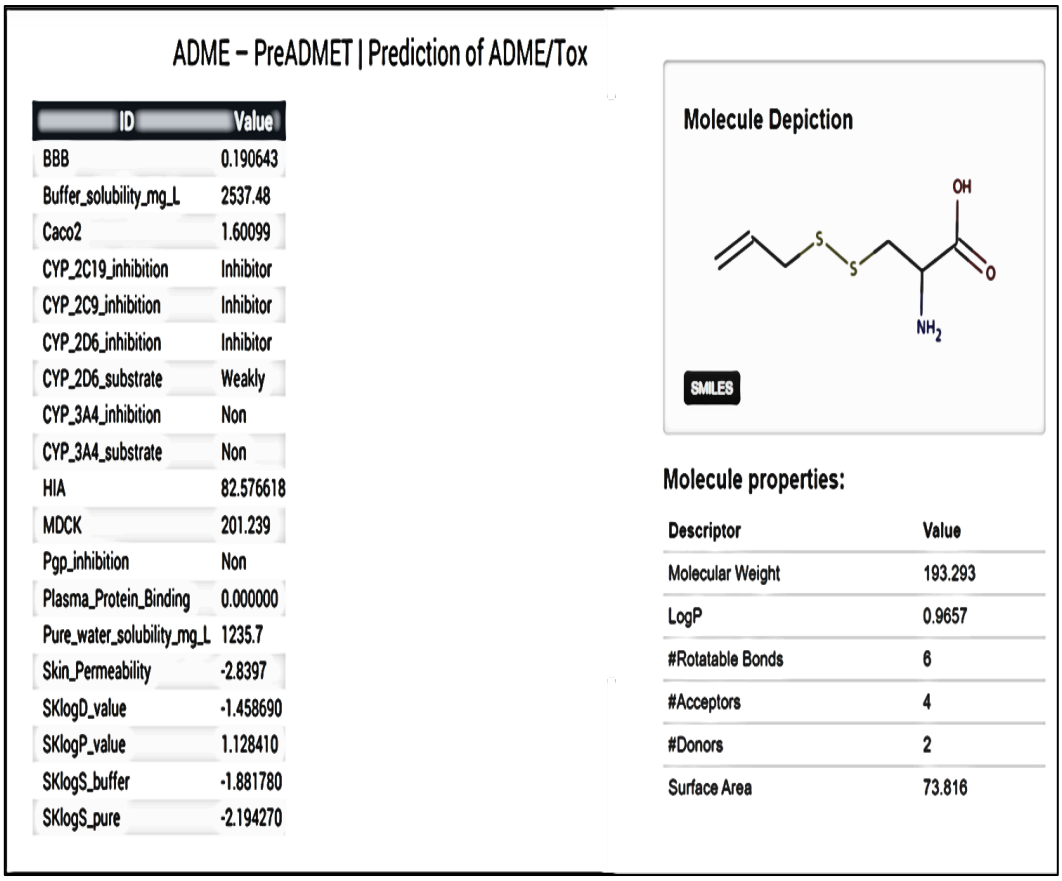
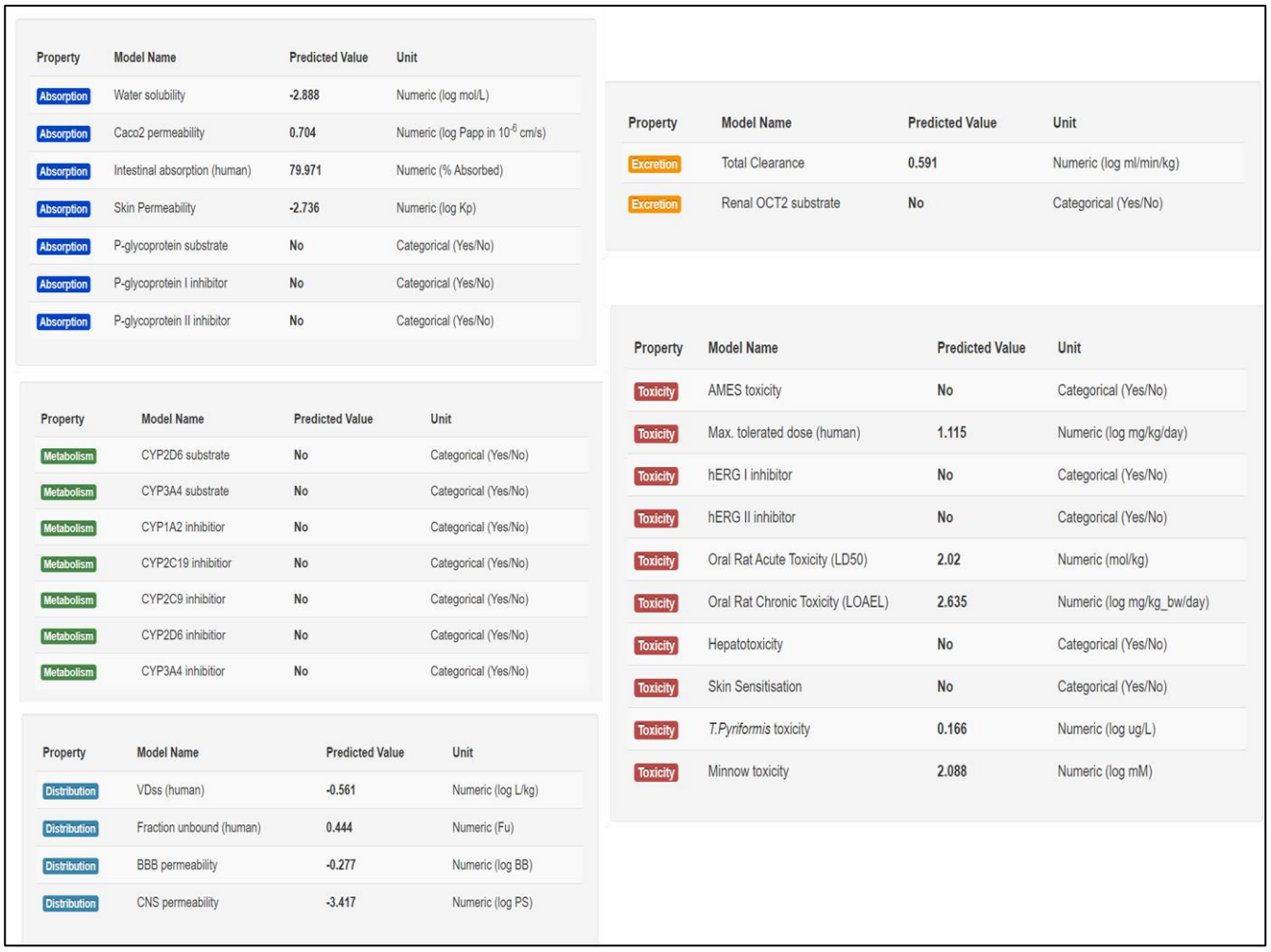


Fig 8 ADMET (metabolism, distribution, excretion, absorption, and toxicity) attributes of S-allylmercaptocysteine (SAMC) predicted by PreADMET (https://preadmet.bmdrc.kr/)[27]

3.2.3 pkCSM (http://biosig.unimelb.edu.au/pkcsm/)

Drug development has a high attrition rate, with poor pharmacokinetic and safety properties a significant hurdle. Computational approaches may help minimize these risks It was seen that pkCSM performs as well or better across different pharmacokinetic properties than other freely available methods. This server is useful for Small-molecule pharmacokinetics prediction. In figure 9- 10 we report Prediction of pharmacokinetic properties: ADMET (metabolism, distribution, excretion, absorption, and toxicity) attributes of S-allylcysteine (SAC) and S-allylcysteine (SAMC) predicted by pkCSM, respectively. (http://biosig.unimelb.edu.au/pkcsm/ [27]



Property	Model Name	Predicted Value	Unit
Absorption	Water solubility	-2.888	Numeric (log mol/L)
Absorption	Caco2 permeability	0.704	Numeric (log Papp in 10^{-6} cm/s)
Absorption	Intestinal absorption (human)	79.971	Numeric (% Absorbed)
Absorption	Skin Permeability	-2.736	Numeric (log Kp)
Absorption	P-glycoprotein substrate	No	Categorical (Yes/No)
Absorption	P-glycoprotein I inhibitor	No	Categorical (Yes/No)
Absorption	P-glycoprotein II inhibitor	No	Categorical (Yes/No)

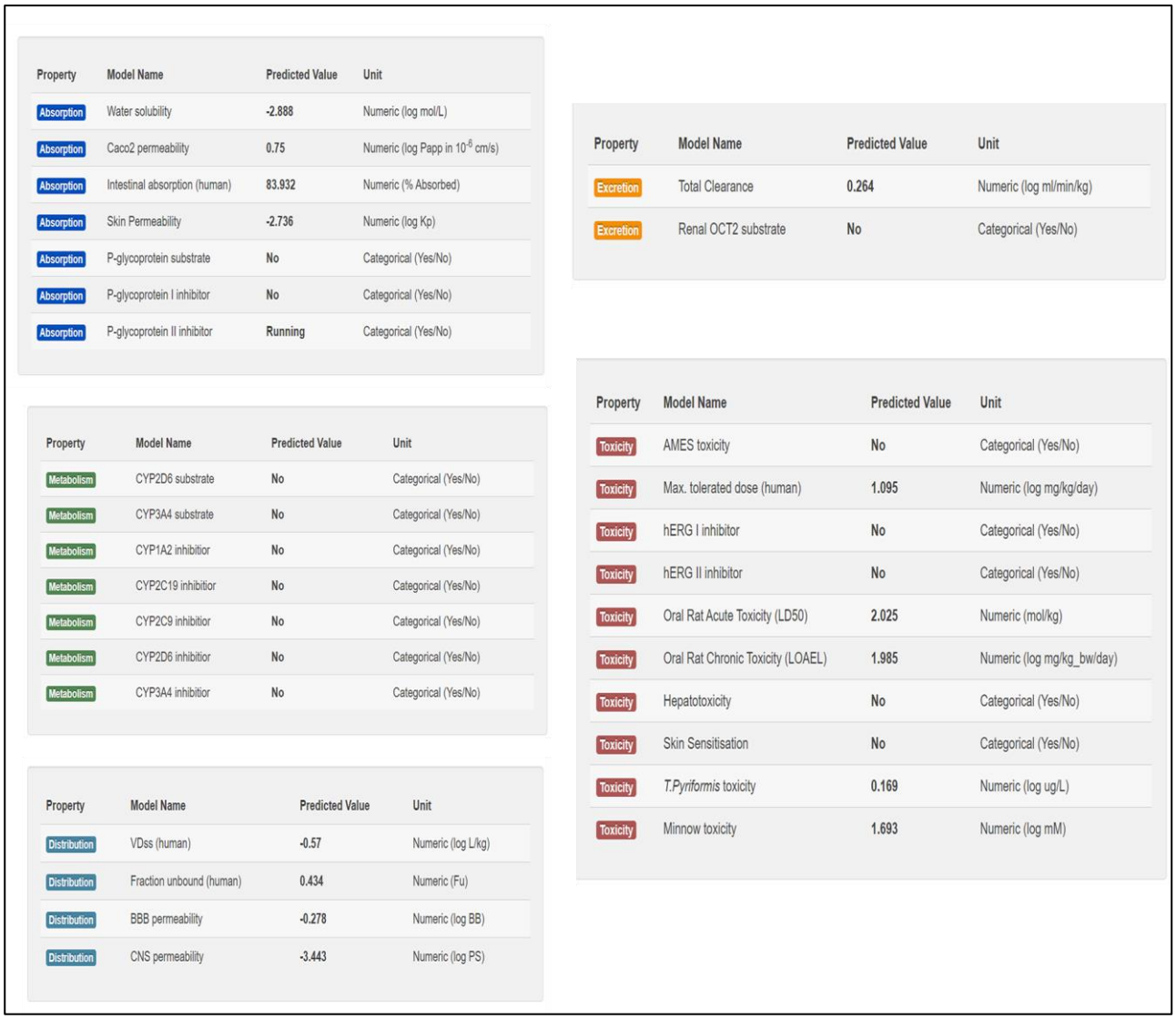
Property	Model Name	Predicted Value	Unit
Excretion	Total Clearance	0.591	Numeric (log ml/min/kg)
Excretion	Renal OCT2 substrate	No	Categorical (Yes/No)

Property	Model Name	Predicted Value	Unit
Metabolism	CYP2D6 substrate	No	Categorical (Yes/No)
Metabolism	CYP3A4 substrate	No	Categorical (Yes/No)
Metabolism	CYP1A2 inhibitior	No	Categorical (Yes/No)
Metabolism	CYP2C19 inhibitior	No	Categorical (Yes/No)
Metabolism	CYP2C9 inhibitior	No	Categorical (Yes/No)
Metabolism	CYP2D6 inhibitior	No	Categorical (Yes/No)
Metabolism	CYP3A4 inhibitior	No	Categorical (Yes/No)

Property	Model Name	Predicted Value	Unit
Toxicity	AMES toxicity	No	Categorical (Yes/No)
Toxicity	Max. tolerated dose (human)	1.115	Numeric (log mg/kg/day)
Toxicity	hERG I inhibitor	No	Categorical (Yes/No)
Toxicity	hERG II inhibitor	No	Categorical (Yes/No)
Toxicity	Oral Rat Acute Toxicity (LD50)	2.02	Numeric (mol/kg)
Toxicity	Oral Rat Chronic Toxicity (LOAEL)	2.635	Numeric (log mg/kg_bw/day)
Toxicity	Hepatotoxicity	No	Categorical (Yes/No)
Toxicity	Skin Sensitisation	No	Categorical (Yes/No)
Toxicity	*T.Pyriformis* toxicity	0.166	Numeric (log ug/L)
Toxicity	Minnow toxicity	2.088	Numeric (log mM)

Property	Model Name	Predicted Value	Unit
Distribution	VDss (human)	-0.561	Numeric (log L/kg)
Distribution	Fraction unbound (human)	0.444	Numeric (Fu)
Distribution	BBB permeability	-0.277	Numeric (log BB)
Distribution	CNS permeability	-3.417	Numeric (log PS)

Fig 9 Prediction of pharmacokinetic properties: ADMET (metabolism, distribution, excretion, absorption, and toxicity) attributes of S-allylcysteine (SAC) SAMC) predicted by pkCSM (http://biosig.unimelb.edu.au/pkcsm/) [27]



Property	Model Name	Predicted Value	Unit
Absorption	Water solubility	-2.888	Numeric (log mol/L)
Absorption	Caco2 permeability	0.75	Numeric (log Papp in 10^{-6} cm/s)
Absorption	Intestinal absorption (human)	83.932	Numeric (% Absorbed)
Absorption	Skin Permeability	-2.736	Numeric (log Kp)
Absorption	P-glycoprotein substrate	No	Categorical (Yes/No)
Absorption	P-glycoprotein I inhibitor	No	Categorical (Yes/No)
Absorption	P-glycoprotein II inhibitor	Running	Categorical (Yes/No)

Property	Model Name	Predicted Value	Unit
Excretion	Total Clearance	0.264	Numeric (log ml/min/kg)
Excretion	Renal OCT2 substrate	No	Categorical (Yes/No)

Property	Model Name	Predicted Value	Unit
Metabolism	CYP2D6 substrate	No	Categorical (Yes/No)
Metabolism	CYP3A4 substrate	No	Categorical (Yes/No)
Metabolism	CYP1A2 inhibitior	No	Categorical (Yes/No)
Metabolism	CYP2C19 inhibitior	No	Categorical (Yes/No)
Metabolism	CYP2C9 inhibitior	No	Categorical (Yes/No)
Metabolism	CYP2D6 inhibitior	No	Categorical (Yes/No)
Metabolism	CYP3A4 inhibitior	No	Categorical (Yes/No)

Property	Model Name	Predicted Value	Unit
Toxicity	AMES toxicity	No	Categorical (Yes/No)
Toxicity	Max. tolerated dose (human)	1.095	Numeric (log mg/kg/day)
Toxicity	hERG I inhibitor	No	Categorical (Yes/No)
Toxicity	hERG II inhibitor	No	Categorical (Yes/No)
Toxicity	Oral Rat Acute Toxicity (LD50)	2.025	Numeric (mol/kg)
Toxicity	Oral Rat Chronic Toxicity (LOAEL)	1.985	Numeric (log mg/kg_bw/day)
Toxicity	Hepatotoxicity	No	Categorical (Yes/No)
Toxicity	Skin Sensitisation	No	Categorical (Yes/No)
Toxicity	*T.Pyriformis* toxicity	0.169	Numeric (log ug/L)
Toxicity	Minnow toxicity	1.693	Numeric (log mM)

Property	Model Name	Predicted Value	Unit
Distribution	VDss (human)	-0.57	Numeric (log L/kg)
Distribution	Fraction unbound (human)	0.434	Numeric (Fu)
Distribution	BBB permeability	-0.278	Numeric (log BB)
Distribution	CNS permeability	-3.443	Numeric (log PS)

Fig 9 Prediction of pharmacokinetic properties: ADMET (metabolism, distribution, excretion, absorption, and toxicity) attributes of S-allylcysteine (SAMC) predicted by pkCSM (http://biosig.unimelb.edu.au/pkcsm/)[27]

Conclusions

This paper encompasses the fundamental functions of open access in silico prediction tools, as PASS database (Prediction of Activity Spectra for Substances) that it estimates the probable biological activity profiles for compounds. This paper also aims to help support new researchers in the field of drug design and to investigate some of bioactive compounds in garlic. Particular attention we investigated Pharmacokinetic properties by several server, of S-allylcysteine (SAC) and S-allylmercaptocysteine (SAMC) for their as anti-inflammatory, antibacterial, and antiviral, antioxidant, cardiovascular protective and anticancer property. Screening through each of pharmacokinetic criteria resulted in identification of Garlic compounds that adhere to all the ADMET properties. It was established an open-access database (PASS database, available bioinformatics tool SwissADME, PreADMET pkCSM database) servers were employed to determine the ADMET (metabolism, distribution, excretion, absorption, and toxicity) attributes of garlic molecules and to enable identification of promising molecules that follow ADMET properties. Further investigations will be conducted in Vitro and In vito by the SAMC, using Layered double hydroxides (LDH) ,which are one type of layered materials and are also known as anionic clays, are promising layered materials due to some of their interesting properties, such as to facile tunability of their composition, structure morphology and biocompatibility vehicle, against several cancer cells.

Author contributions

I.V.F. conceived, designed and wrote the paper and performed the calculations and analyzed the data.

Declaration of Competing Interest

The authors declare they have no potential conflicts of interest to disclose.

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Supporting Information

Compound	Molecular Formula	Molecular Weight (g/mol)	Pa	Pi	Acitivity
(E)-Ajoene and Z)-Ajoene	$C_9H_{14}OS_3$	234.4	0,972	0,004	Antineoplastic
			0,949	0,002	CYP2E1 inhibitor
			0,94	0,004	Apoptosis agonist
			0,881	0,004	CYP2E1 substrate
			0,875	0,004	CYP2E substrate
			0,869	0,001	Chemoprotective
Vinylthiiran	C_4H_6S	86.16	Pa	Pi	Acitivity
			0,932	0,001	Growth hormone agonist
			0,926	0,003	Gluconate 2-dehydrogenase (acceptor) inhibitor
			0,865	0,003	Chloride peroxidase inhibitor
			0,875	0,014	Aspulvinone dimethylallyltransferase inhibitor
			0,822	0,013	Mucomembranous protector
			0,81	0,004	Fatty-acyl-CoA synthase inhibitor
			0,806	0,004	Thioredoxin inhibitor
			0,802	0,006	Cl--transporting ATPase inhibitor
Allicin	$C_6H_{10}OS_2$	162.3	Pa	Pi	Acitivity
			0,959	0,003	Apoptosis agonist
			0,884	0,001	Chemoprotective
			0,852	0,02	Aspulvinone dimethylallyltransferase inhibitor
gamma-Glutamylcysteine	$C_8H_{14}N_2O_5S$	250.27	Pa	Pi	Acitivity
			0,984	0,001	Mucositis treatment
			0,983	0,001	Protein-disulfide reductase (glutathione) inhibitor
			0,982	0	Hydrogen dehydrogenase inhibitor
			0,979	0	Yeast ribonuclease inhibitor
			0,976	0,001	Fucosterol-epoxide lyase inhibitor
			0,974	0,001	Levanase inhibitor
			0,97	0,002	Monodehydroascorbate reductase (NADH) inhibito
S-Allyl-L-cysteine	$C_6H_{11}NO_2S$	161.22	Pa	Pi	Acitivity
			0,965	0,002	Flavin-containing monooxygenase substrate
			0,963	0,003	CYP2E1 substrate
			0,962	0,003	CYP2E substrate
			0,942	0,001	S-alkylcysteine lyase inhibitor
			0,939	0,002	NADPH peroxidase inhibitor
			0,933	0,002	FMO3 substrate
			0,928	0,002	Lysine 2,3-aminomutase inhibitor
			0,924	0,003	Acylcarnitine hydrolase inhibitor
			0,905	0,004	Arylacetonitrilase inhibitor
S-allylmercapto-L-cysteine	$C_6H_{11}NO_2S_2$	193.3	Pa	Pi	Acitivity
			0,974	0,002	Flavin-containing monooxygenase substrate
			0,967	0,001	FMO3 substrate
			0,939	0,002	NADPH peroxidase inhibitor
			0,939	0,002	Protein-disulfide reductase (glutathione) inhibitor
			0,934	0,003	CYP2E1 substrate
			0,933	0,003	CYP2E substrate
			0,932	0,003	Antiseborrheic
			0,928	0,003	Lipid metabolism regulator
			0,925	0,004	Apoptosis agonist
			0,924	0,003	Acylcarnitine hydrolase inhibitor
			0,919	0,001	Cysteamine dioxygenase inhibitor
			0,916	0,003	Arylacetonitrilase inhibitor
			0,909	0,002	S-alkylcysteine lyase inhibitor
			0,903	0,005	TP53 expression enhancer
Allyl methyl sulfide	C_4H_8S	88.17	Pa	Pi	Acitivity
			0,965	0,002	Flavin-containing monooxygenase substrate
			0,941	0,004	Apoptosis agonist
			0,935	0,002	FMO3 substrate
			0,914	0,004	Mucomembranous protector
			0,9	0,003	CYP2E1 inhibitor
			0,876	0,002	S-alkylcysteine lyase inhibitor
Diallyl sulfide	$C_6H_{10}S$	114.21	Pa	Pi	Acitivity
			0,926	0,002	Flavin-containing monooxygenase substrate
			0,889	0,01	Aspulvinone dimethylallyltransferase inhibitor
			0,876	0,006	Mucomembranous protector
			0,872	0,002	FMO3 substrate
			0,869	0,003	Fatty-acyl-CoA synthase inhibitor
Diallyl disulfide	$C_6H_{10}S_2$	146.3	Pa	Pi	Acitivity
			0,987	0,002	Apoptosis agonist
			0,944	0,001	Cysteamine dioxygenase inhibitor
			0,935	0,003	CYP2E1 inhibitor
			0,931	0,003	Inflammatory Bowel disease treatment
			0,92	0,001	Bcl2 antagonist
			0,917	0,001	Chemoprotective
			0,904	0,003	Antioxidant
			0,896	0,003	Atherosclerosis treatment
Diallyl trisulfide	C_6H_{10S3}	178.3	Pa	Pi	Acitivity
			0,981	0,001	Antineoplastic (cervical cancer)
			0,979	0,002	Antineoplastic (ovarian cancer)
			0,977	0,004	Antineoplastic
			0,97	0,002	Antineoplastic (lymphocytic leukemia)
			0,964	0	Antineoplastic (sarcoma)
			0,955	0,003	Apoptosis agonist
			0,94	0,002	Antineoplastic (non-small cell lung cancer)
			0,934	0,003	Antineoplastic (breast cancer)

Tab 2 PASS Online Server (*Prediction of Activity Spectra for Substances) of Principal Organosulfur Compounds in Garlic . The output file represents a list of activities with two probabilities Pa (probability to be active) and Pi (probability to be inactive). Pa (probability "to be active") estimates the chance that the studied compound is belonging to the sub-class of active compounds (resembles the structures of molecules, which are the most typical in a sub-set of "actives" in PASS training set). Pi (probability "to be inactive") estimates the chance that the studied compound is belonging to the sub-class of inactive compounds (resembles the structures of molecules, which are the most typical in a sub-set of "inactives" in PASS training set). [8]*

Compound	Molecular Formula	Molecular Weight (g/mol	Pa	Pi	Possible adverse & toxic effects
(E)-Ajoene and Z)-Ajoene	$C_9H_{14}OS_3$	234.4	0,449	0,036	Thrombocytopoiesis inhibitor
			0,371	0,093	Adrenal cortex hypoplasia
			0,36	0,103	Weight gain
			0,311	0,123	Hypomagnesemia
Vinylthiiran	C_4H_6S	86.16	Pa	Pi	Possible adverse & toxic effects
			0,907	0,005	Weight loss
			0,801	0,019	Weakness
			0,771	0,017	Nephrotoxic
			0,765	0,017	Muscle weakness
Allicin	$C_6H_{10}OS_2$	162.3	Pa	Pi	Possible adverse & toxic effects
			0,682	0,011	Thrombocytopoiesis inhibitor
			0,616	0,046	Withdrawal
			0,639	0,073	ophilic dermatosis (Sweet's syndrome)
gamma-Glutamylcysteine	$C_8H_{14}N_2O_5S$	250.27	Pa	Pi	Possible adverse & toxic effects
			0,924	0,001	Bullous pemphigoid
			0,891	0,008	Pure red cell aplasia
			0,858	0,007	Ulcer, aphthous
			0,852	0,003	Anemia, sideroblastic
			0,856	0,011	Neurotoxic
			0,857	0,016	Diarrhea
			0,809	0,012	Nephrotoxic
S-Allyl-L-cysteine	$C_6H_{11}NO_2S$	161.22	Pa	Pi	Possible adverse & toxic effects
			0,835	0,017	Pure red cell aplasia
			0,832	0,02	Diarrhea
			0,801	0,017	Neurotoxic
			0,786	0,004	Anemia, sideroblastic
			0,784	0,022	Ulcer, aphthous
S-allylmercapto-L-cysteine	$C_6H_{11}NO_2S_2$	193.3	Pa	Pi	Possible adverse & toxic effects
			0,786	0,004	Anemia, sideroblastic
			0,804	0,023	Pure red cell aplasia
			0,783	0,015	Nephrotoxic
			0,777	0,02	Neurotoxic
			0,764	0,014	Ataxia
			0,758	0,028	Ulcer, aphthous
			0,716	0,021	Weight gain
			0,728	0,039	Toxic
Allyl methyl sulfide	C_4H_8S	88.17	Pa	Pi	Possible adverse & toxic effects
			0,819	0,006	Thrombocytopoiesis inhibitor
			0,725	0,012	Hypothermic
			0,708	0,005	Lacrimal secretion stimulant
			0,711	0,027	Hyperglycemic
Diallyl sulfide	$C_6H_{10}S$	114.21	Pa	Pi	Possible adverse & toxic effects
			0,801	0,006	Thrombocytopoiesis inhibitor
			0,769	0,008	Withdrawal
			0,77	0,011	Weight gain
			0,781	0,028	ophilic dermatosis (Sweet's syndrome)
			0,795	0,044	Twitching
Diallyl disulfide	$C_6H_{10}S_2$	146.3	Pa	Pi	Possible adverse & toxic effects
			0,888	0,004	Thrombocytopoiesis inhibitor
			0,787	0,043	Shivering
			0,741	0,011	Withdrawal
			0,734	0,017	Weight gain
			0,719	0,02	Ataxia
			0,738	0,04	ophilic dermatosis (Sweet's syndrome)
			0,705	0,016	Hypomagnesemia
			0,755	0,067	Twitching
Diallyl trisulfide	C_6H_{10S3}	178.3	Pa	Pi	Possible adverse & toxic effects
			0,734	0,009	Thrombocytopoiesis inhibitor
			0,711	0,073	Shivering

Tab 3 PASS Online Server (*Prediction of Activity Spectra for Substances) calculated possible adverse and toxic effects of Principal Organosulfur Compounds in Garlic [8]*