

Investigating the *Panax ginseng* proteome using an *in silico* method to find bioactive peptides that can alter the dynamics of breast cancer carcinogenesis

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Breast cancer is a major public health concern throughout the world, particularly in Cameroon. Numerous modalities, including surgery, radiotherapy, and chemotherapy, have been used for clinical cancer therapy. Despite advances in anti-cancer therapies, the use of these methods to treat cancer is often associated with deleterious side effects and multi-drug resistance to conventional anti-cancer drugs, which has led to the development of new therapeutic methods, anti-cancer peptides (ACP). The aim of this study was to produce peptides that could be potential modulators of breast cancer, using *in silico* prediction as complementary tool. Several techniques were applied to achieve this goal: Protein hydrolysis, protein-peptide molecular docking, and target prediction have all been employed as techniques. It has been determined that 156 genes may play a role in the carcinogenesis of breast cancer. Ten (10) kinases (CDK1, NLK, CDK4, CDK2, CSNK2A1, GSK3B, HIPK2, MAPK14, CK2ALPHA, and MAPK1) and 10 transcription factors (TRIM28, CHD1, SOX2, NANOG, UBTF, TCF7L2, TCF3, CTCF, SUZ12, and NFE2L2) were identified as involved in the process of breast cancer carcinogenesis. Only nineteen of the 1324 peptides that were synthesised exhibited an affinity of greater than 20%, and approximately twenty-eight distinct proteins were found. The peptides AIF, AIN, APQVAVL, ESK, IAK, PSIPK, PVGF, and SDTEL are all intriguing candidates for additional research, according to the overall findings. The study's non-allergic peptides had binding energy values of less than or equal to -5.00 kcal.mol. Future studies will use both *in vitro* and *in vivo* methods to examine the positive effects of hydrolyzed peptides.

Keywords: *Panax ginseng*, Proteome analysis, In-silico methods, Bioactive peptides, Breast cancer carcinogenesis

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Introduction

Breast cancer is a varied condition with a range of molecular subtypes and biological features (1). It is the most prevalent cancer in women and the second most common cancer globally among newly diagnosed cancer cases (2). Three in ten of the malignancies diagnosed in both sexes were related to the cervix (110,300 instances) and breast (129,400 cases in females) (3). In sub-Saharan Africa (SSA), breast cancer is the leading cause of cancer-related deaths among women worldwide and a major public health concern. Compared to high-income nations, mortality rates in Sub-Saharan Africa (SSA) are disproportionately high, despite the region's disease frequency appearing to be lower. With SSA reporting the majority of cases, the global illness burden is increasing; yet, the lack of information leads to inadequate data that is hardly indicative of the true disease burden in this group (4). There are various types of breast cancer. Just 5 to 7% of breast tumors are hereditary malignancies caused by germline mutations in the high-penetrance BrCa1 and BrCa2 tumor suppressor genes (5). For 90% of breast cancers that are said to be sporadic, the initial

mechanisms are still very poorly understood. However, the long-suspected role of ovarian hormones in promoting breast cancer is now well established, opening up the possibility of prevention. Apart from risk factors, family history, and delay in first pregnancy are essentially linked to the time of natural or therapeutic exposure to ovarian hormones. For all these types of breast cancers, the main forms of treatment include surgery, radiation therapy, and chemotherapy; however, the outcomes of these treatments are not always desirable. Improved therapeutic agents for the treatment of breast cancer are needed because of the drawbacks of these traditional treatments, including non-selectivity, multidrug resistance, bioavailability, and the cost of clinical intervention (6). Numerous natural products have been investigated for the treatment of breast cancer. Nevertheless, many of these natural compounds had harmful side effects and were limited by their low water solubility (7). In order to get around these restrictions, it is possible to create a number of structural analogues of natural products that have strong anti-breast cancer properties and less adverse effects than their precursor molecules. In 2018–2019, patients receiving radiation consultations at the "anti-cancer centre, CAC" in

Annaba, Algeria, participated in an ethnobotanical study of medicinal plants historically used in breast cancer treatment. It was feasible to compile all the data regarding patients' use of medicinal herbs thanks to this study. 35 plant species, distributed across 27 botanical families, were identified through the analysis of the questionnaire data. Notably, 12 quotes were obtained for *Panax ginseng*., 8 for *Zingiber officinale* Roscoe, 9 for *Curcuma longa* L., 10 for *Allium sativum* L., 11 for *Berneris vulgaris* L., and 8 for *Annona muricata* L (8). More research has been done on ginseng's potential benefits in treating cancer-related fatigue. 290 cancer patients participated in a double-blind trial where they were randomly allocated to receive either a placebo or escalating doses of American ginseng (750–2000 mg daily) for eight weeks. When ginseng was administered to patients instead of a placebo, their fatigue scores improved, especially when the dosage was between 1000 and 2000 mg daily. Compared to the placebo arm, more patients in the ginseng arm self-reported a benefit and satisfaction with their therapy (9). However, it is not only total *Panax ginseng* extracts that can be used to treat breast cancer; some bioactive proteins/polypeptides from this plant can also inhibit breast cancer cell growth as presented in several studies (10–11). Because ginseng has been extensively studied for its potential therapeutic uses in modern medicine, in breast cancer (12–14) and can also suppress oxidative stress-induced (6). In order to identify and characterise bioactive peptides that may be used as prospective breast cancer treatments, we have decided to study the proteome of *Panax ginseng* for this manuscript.

Experimental section

Identification of genes involved in breast cancer

The genes used here have been identified in the literature, these genes were linked to breast cancer in both human and animal models. These genes were chosen to avoid duplication.

Enrichment of transcription factors and kinases

Network analysis was performed on all 156 BREAST CANCER-associated reported genes to ascertain the enrichment of transcription factors, kinases, and protein-protein interaction. The eXpression2Kinases (X2K) webserver, accessible at <https://maayanlab.cloud/X2K/>, was used for this, with humans (*Homo sapiens*) chosen as the target organism.

The Interaction Between Proteins

The gene network and gene fusions were revealed by the protein-protein interaction (PPI) profile of the 156 genes linked to breast cancer, which was retrieved from the STRING webserver (<https://string-db.org/>) on December 26, 2023.

Protein Hydrolysis and Evaluation of Bioactivity

The peptides to be isolated were obtained from the panax ginseng proteome. *Panax ginseng* is a perennial herb that is a member of the *Araliaceae* family and genus *Panax*.

For thousands of years, it has been a widely used and well-known medicinal herb in Eastern Asia. Based on chromosome number and genomic analysis, he has a chromosome complement of $2n = 48$, which designates an allotetraploid plant. While ginseng's medicinal properties and pharmacological effects have been extensively studied, genetic and genomic research on the plant has only just begun due to challenges in preserving individual plants and creating progenies (15). On the BIOPEP-UWM website, the sequences underwent simulated enzymatic hydrolysis utilising the digestive enzymes trypsin (EC 3.4.21.4), pepsin (EC 3.4.23.1), and chymotrypsin (EC 3.4.21.1). Peptides with at least three and a maximum of six amino acids and a bioactive affinity of greater than 20% were chosen after synthesis. The following are the genomic segments from which proteins have been extracted:

Squalene synthase 1 Gene: SS1(GSS, PSS, SQS, SS, SS4, SSA); 415 amino acids ([https:// www. uniprot. org/uniprotkb/ A0A068J840/entry](https://www.uniprot.org/uniprotkb/A0A068J840/entry))

Farnesyl pyrophosphate synthase Gene: FPS *Panax ginseng* (Korean ginseng) EC:2.5.1.10 342 amino acids

UDP-glycosyltransferase 1 Gene: UGT1 *Panax ginseng* (Korean ginseng) EC:2.4.1.363 475 amino acids

UDP-glucosyltransferase 29 Gene: UGT29 (UGT94B1, UGT94Q2) *Panax ginseng* (Korean ginseng) EC:2.4.1.365 442 amino acids

Pharmacokinetic characteristics of the hydrolyzed peptides predicted

Using the SMILES format and default parameters, an *in-silico* screening for functional hydrolysate peptides was conducted using the SwissADME server (www.swissadme.ch, accessed on December 30, 2023) for ADME (absorption, distribution, metabolism, and excretion). The AllerTOP v.2.0 webserver (<http://www.ddg-pharmfac.net/AllerTOP>, accessed on December 30, 2023) was used to estimate allergenicity.

Molecular docking

Peptides with no more than four amino acid residues, non-allergenicity, and a minimum 20% chance of hitting at least one molecular target protein are the requirements utilised for molecular docking investigations. Peptide-protein docking on the SERVER SwissDock (<http://www.swissdock.ch/>) was done based on the projected results from the preceding phase. ChemDraw Professionel 16 was used to draw the molecules, and Chem3D was used to display them.

Results and Discussion

Identification of genes involved in breast cancer

A total of 156 genes have been identified as being involved in breast cancer:

AXIN1, (FZD1, FZD10, FZD2, FZD8, FZD9), (LRP5, LRP6), TNFSF11, (WNT1, WNT10A, WNT10B, WNT16, WNT2, WNT2B, WNT3, WNT3A, WNT4, WNT5A, WNT5B, WNT6, WNT7A, WNT7), BATM, ATR, BAK1, BAX, (BRCA1, BRCA2), CDK4,

CDKN1A, DDB2, (GADD45A, GADD45B, GADD45G), JUN, MAPK1, MRE11, MYC, NBN, NCOA1, NFKB2, PARP1, POLK, PTEN, (RAD50, RAD51), SP1, TP53, CETN3, (AKT1, AKT2, AKT3), ARAF, EGFR, ESR1, FLT4, GRB2, HRAS, IGF1R, KIT, MTOR, PGR, (PIK3CA, PIK3CD, PIK3R1, PIK3R2, PIK3R3), CSNK2A2, CSNK2A3, CSNK2B, CSNK2A1, CSNK1A1, CSNK1A1L, (DVL1, DVL2, DVL3), (FRAT1, FRAT2), (FZD3, FZD5, FZD6, FZD7), GSK3B, (NOTCH1, NOTCH2, NOTCH3, NOTCH4), WNT11, (FGF1, FGF10, FGF16, FGF17, FGF18, FGF19, FGF2, FGF20, FGF21, FGF22, FGF23, FGF3, FGF4, FGF5, FGF6, FGF7, FGF8, FGF9), FGFR, (SHC1, SHC2, SHC3, SHC4), (SOS1, SOS2), (APC, APC2), AXIN2, CCND1, CTNNB1, ERBB2, SFRP4, SKP1, (TCF7, TCF7L1, TCF7L2), BRAF, ESR2, FOS, KRAS, (MAP2K1, MAP2K2, MAPK3), NRAS, RAF1, (RPS6KB1, RPS6KB2), CDK6, (E2F1, E2F2, E2F3), NCOA3, RB1, (DLL1, DLL3, DLL4), (HES1, HES5), (HEY1, HEY2, HEYL), JAG2.

Expression of kinase network related to identify gene

A set of 10 kinases (CDK1, NLK, CDK4, CDK2, CSNK2A1, GSK3B, HIPK2, MAPK14, CK2ALPHA, and MAPK1) and 10 transcription factors (TRIM28, CHD1, SOX2, NANOG, UBTf, TCF7L2, TCF3, CTCF, SUZ12, and NFE2L2) were identified as being among the proteins involved in the activity of the genes identified. Numerous intermediary proteins are also involved in the process of carcinogenesis, as shown in Figure 1 below.

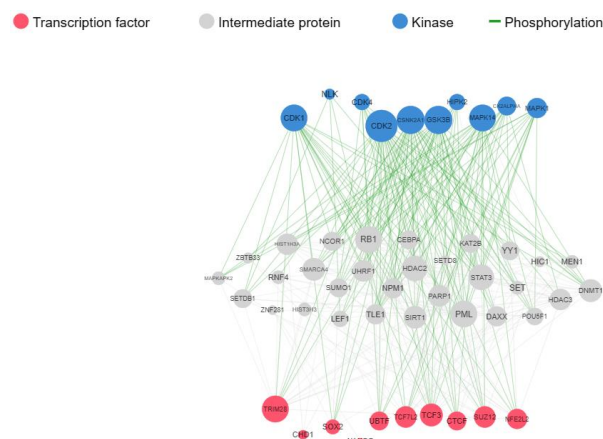


Figure 1. Transcription factors and kinases related to breast cancer.

The Interaction Between Proteins

Below you can see the enrichment analysis of ten different clusters as presented in Figure 2:

Cluster 1 in Red (13) (DLL1, DLL3, DLL4, HES1, HES5, HEY1, HEY2, HEYL, JAG2, NOTCH1, NOTCH2, NOTCH3, NOTCH4);

Cluster 2 in Brow (22) (ATR, BAK1, BAX, BRCA1, BRCA2, CDKN1A, CSNK2A1, DDB2, GADD45A, GADD45B, GADD45G, MAPK1, MAPK3, MRE11,

MYC, NBN, PARP1, POLK, PTEN, RAD50, RAD51, TP53);

Cluster 3 in Dark Golden Rod (CETN3);

Cluster 4 in Green Yellow (14) (ESR1, ESR2, FOS, IGF1, IGF1R, JUN, NCOA1, NCOA3, NFKB2, PGR, RPS6KB1, RPS6KB2, SHC1, SP1);

Cluster 5 in Green 2 (17) (APC, APC2, AXIN1, AXIN2, CSNK1A1, CSNK2A2, CSNK2A3, CSNK2B, CTNNB1, ERBB2, LEF1, RECK, SFRP4, SKP1, TCF7, TCF7L1, TCF7L2)

Cluster 6 in Green (10) (ARAF, CSNK1A1L, DVL1, DVL2, DVL3, FRAT1, FRAT2, GSK3B, MAP2K1, MAP2K2)

Cluster 7 in Blue (26) (EGF, FGF1, FGF10, FGF16, FGF17, FGF18, FGF19, FGF2, FGF20, FGF21, FGF22, FGF23, FGF3, FGF4, FGF5, FGF6, FGF7, FGF8, FGF9, FGFR1, GRB2, SHC2, SHC3, SHC4, SOS1, SOS2)

Cluster 8 in Light Sky Blue (7) (CCND1, CDK4, CDK6, E2F1, E2F2, E2F3, RB1)

Cluster 9 : Medium Blue (17) (AKT1, AKT2, AKT3, BRAF, EGFR, FLT4, HRAS, KIT, KRAS, MTOR, NRAS, PIK3CA, PIK3CD, PIK3R1, PIK3R2, PIK3R3, RAF1)

Cluster 10 in Purple (26) : (FZD1, FZD10, FZD2, FZD3, FZD5, FZD6, FZD7, FZD8, FZD9, LRP5, LRP6, TNFSF11, WNT1, WNT10A, WNT10B, WNT11, WNT16, WNT2, WNT2B, WNT3, WNT3A, WNT4, WNT5A, WNT5B, WNT6, WNT7A).

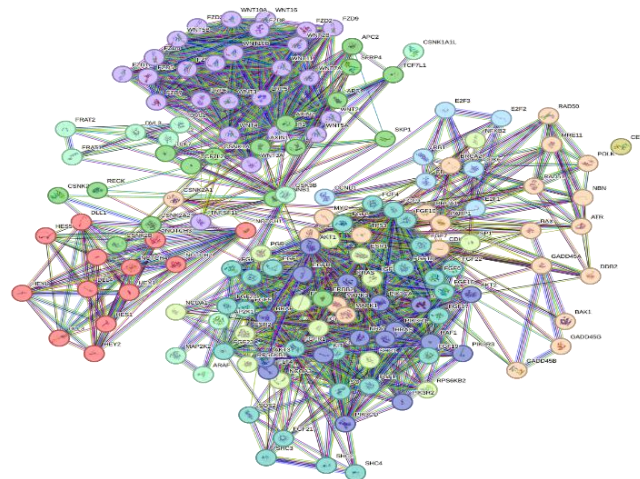


Figure 2. Proteomic interactome in breast cancer.

Protein Hydrolysis results

After hydrolysing 1324 peptides, only 19 peptides with a minimum of three and a maximum of six amino acids and a bioactive affinity of more than 20% were found to be of interest. The highest affinity between a peptide and a protein was observed between the inhibitor of apoptosis protein 3 and the IPPIY peptide, with an affinity of 85% as presented in Table 1.

Table 1. Predicted target probability between peptide and protein, obtained from an *in silico* model applied for searching active compounds in breast carcinogenesis.

	Selected Active Hydrolysate Peptide	% Probability of Predicted Targets name																											
		A	B	C	D	E	F	G	H	I	J	K	L	M	N	O	P	Q	R	S	T	U	V	W	X	Y	Z	Aa	Ab
1	AIF	53	36	26	25	24	22	20	20																				
2	AIN		20							24																			
3	APQVAVL	44		20					65																				
4	EIL			23						23																			
5	EPY		20							24																			
6	ESK										24																		
7	IAK			27					28			23																	
8	IPPIY	85	72	32		55		30	28				72	30															
9	ISR														22	21													
10	PSIPK			25													30												
11	PSVI	23																											
12	PTPR														25	21		25	21	20									
13	PVGF	74	27	20								23																	
14	QDVTEF																				52	20							
15	QEN																							20					
16	QTL																								20				
17	SDTEL																									20			
18	SEIR														29				29										
19	VIGW																									53	52	28	

A: Inhibitor of apoptosis protein 3; **B:** Mu opioid receptor (by homology); **C:** Angiotensin-converting enzyme; **D:** Cyclooxygenase-2; **E:** Delta opioid receptor; **F:** Neprilysin (by homology); **G:** Kappa Opioid receptor; **H:** HLA class I histocompatibility antigen A-3; **I:** HMG-CoA reductase; **J:** Disks large homolog 4; **K:** Protein kinase C epsilon; **L:** Cathepsin (B and K); **M:** Neurokinin 1 receptor (by homology); **N:** Neurotensin receptor 1 (by homology); **O:** Subtilisin/kexin type 6; **P:** Complement factor B; **Q:** Lipoxin A4 receptor (by homology); **R :** WD repeat-containing protein 5; **S:** Furin; **T:** Integrin alpha-V/beta-3; **U:** Beta-secretase 1; **V:** Beta-secretase 2; **W:** Cathepsin D; **X:** Sodium/glucose cotransporter 1; **X:** Sodium/glucose cotransporter 1; **Y:** Caspase-8; **Z:** Endothelin receptor ET-B; **Aa:** Endothelin receptor ET-B; **Ab:** Cholecystokinin B receptor.

Pharmacokinetic characteristics of the hydrolyzed peptides predicted

The Table 2 below shows the pharmacokinetic properties of proteins hydrolyzed into peptides. Various characteristics are observed, such as Physicochemical properties: molecular weight (MW), molar refractivity (MR), total polar surface area (TPSA). Lipophilicity: log P (consensus). Water solubility: ESOL log S, ESOL class. Pharmacokinetics: gastrointestinal absorption (GIA), blood-brain barrier (BBB), P-glycoprotein (P-gp) substrate, inhibition of cytochrome P450 (CYPs) type CYP1A2, CYP2C19, CYP2C9, CYP2D6, and CYP3A4, skin permeation (Log Kp). Drug-likeness: bioavailability score (BS). Medicinal chemistry: synthetic accessibility (SA). Of the nineteen peptides isolated, eight were non-allergenic.

Molecular Docking SCORE

This Table 3 below presents the binding affinity (in kcal/mol) of peptides to a specific target, as determined by CB-Dock Vina. Each row represents a different peptide, and each column represents a different binding site or variant of the target protein. The higher the negative value of the binding affinity, the stronger the binding interaction between the peptide and the target protein at that particular binding site. This is observed with peptide PVGF has binding affinities of -7.8, -8.9, and -9.1 kcal/mol for binding sites A, B, and C, respectively.

Molecular docking results

To further explore the inhibition mechanism of the three hit molecules, we predicted the 2D interactions of each, with the key amino acid residues playing a major role in **A**, **B** and **Y** activities. 2D interaction diagram in Figure 3 showed that **AIF** can form four conventional hydrogen bonds acceptor between oxobutanaminium group with Thr308A, Asp309A, and Glu314A, as well as hydrophobic interactions of diphenylmethanamide group with Lys297A, Gly306A, Leu307A and the azepin group with Trp323A. On the other hand, Figure 4 revealed that **AIN** forms one conventional hydrogen bond acceptor with Asp147R and hydrophobic contacts between its phenol and phenyl groups with Gln124R, Val143R, Ile144R, Met151R, and Ile296R residues. SDTEL formed two conventional hydrogen bonds; donor with Thr337A and acceptor with Glu396B. A π - π interaction of chlorobenzene group with Phe399B, and finally hydrophobic interactions of pyridazine group with residues Tyr334A, Ser338A and chlorobenzene group with Phe399B (Figure 5). Figures 6 and 7 present 3D view for AIN et Mu opioid receptor and AIF et inhibitor of apoptosis protein.

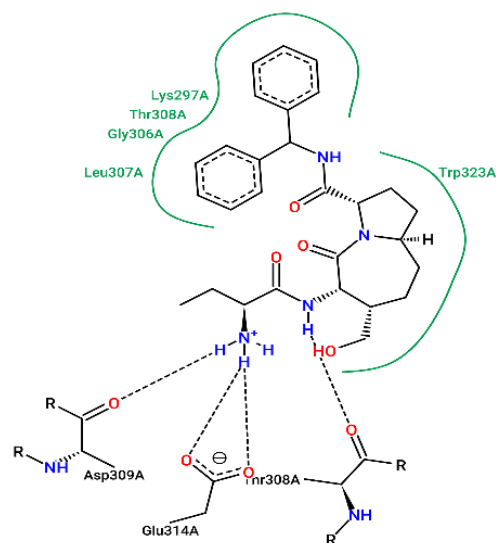


Figure 3. 2D interaction diagram between AIF (oxobutanaminium group) and Thr308A, Asp309A, and Glu314A.

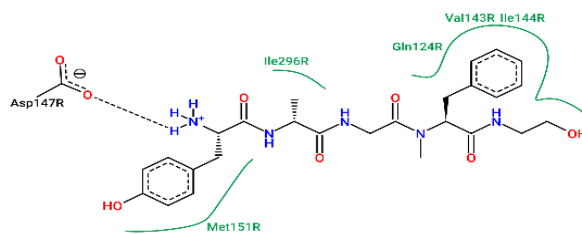


Figure 4: Interaction between AIN and Asp147R by conventional hydrogen bond acceptor, and hydrophobic contacts between its phenol and phenyl groups with Gln124R, Val143R, Ile144R, Met151R, and Ile296R residue.

Table 2. Predicted pharmacokinetic parameter applied to proteins and peptides, in a search for active compounds for breast cancer.

SN	Selected Hydrolysate Peptide	Active	Predicted ADME Parameter										Log Kp (cm/s)	BS	SA	Allergenicity	
			MW (g/mol)	MR	TPSA (Å2)	Log P	ESOL S	Log	ESOL Class	GIA	BB B	P-gp					CYPs Inhibitor
1	AIF		349.2	94.7	121.5	0.56	-0.40		Very soluble	High	No	No	No	-9.62	0.55	3.49	Non-Allergen
2	AIN		316.35	77.99	164.61	-1.52	1.16		Highly soluble	Low	No	No	No	-10.75	0.55	3.62	Non Allergen
3	APQVAVL		696.84	183.23	272.22	-0.98	-0.10		Very soluble	Low	No	No	No	-13.27	0.17	6.17	Non Allergen
4	EIL		373.44	96.09	158.82	-0.12	0.69		Soluble	Low	No	No	No	-10.74	0.11	3.89	Allergen
5	EPY		407.42	105.26	170.26	-0.71	0.40		Soluble	Low	No	No	No	-11.26	0.11	3.54	Allergen
6	ESK		362.38	85.54	205.07	-2.92	3.96		Soluble	Low	No	Yes	No	-14.20	0.17	3.68	Non allergen
7	IAK		330.42	87.41	147.54	-0.42	0.73		Soluble	Low	No	No	No	-10.30	0.55	3.67	Non allergen
8	IPPIY		601.73	169.08	182.37	1.40	-2.85		Soluble	Low	No	No	Yes	-9.64	0.17	5.34	Allergen
9	ISR		374.44	94.96	203.65	-1.61	1.61		Soluble	Low	No	Yes	No	-11.72	0.17	4.08	Allergen
10	PSIPK		540.65	146.43	203.19	-0.72	-0.02		Very soluble	Low	No	No	No	-11.76	0.17	5.16	Non allergen
11	PSVI		414.50	109.99	156.86	2.00	-0.62		Soluble	Low	No	Yes	No	-11.06	0.55	4.24	Allergen
12	PTPR		469.54	125.79	209.97	-1.96	0.92		Highly soluble		No	No	No	-12.19	0.17	4.67	Allergen
13	PVGF		418.49	114.09	136.63	0.21	-0.46		Very soluble	Low	No	Yes	No	-10.28	0.55	3.65	Non allergen
14	QDVTEF		737.75	178.68	346.74	-2.42	0.93		Highly soluble	Low	No	Yes	No	-14.84	0.11	6.06	Allergen
15	QEN		389.36	87.48	245.00	-3.35	2.96		Highly soluble	Low	No	Yes	No	-13.43	0.11	3.75	Allergen
16	QTL		360.41	88.77	184.84	-1.85	1.79		Highly soluble	Low	No	No	No	-11.89	0.55	3.81	Allergen
17	SDTEL		563.56	130.22	294.78	-3.33	1.73		Highly soluble	Low	No	Yes	No	-13.81	0.1	5.16	Non allergen
18	SEIR		503.55	123.77	270.05	-2.27	1.51		Highly soluble	Low	No	Yes	No	-12.85	0.17	4.92	Allergen
19	VIGW		473.57	128.85	166.41	0.81	-1.64		Very soluble	Low	No	Yes	No	-9.58	0.55	4.35	Allergen

SN: Serial number. Molecular weight (MW), molar refractivity (MR), total polar surface area (TPSA). Lipophilicity: log P (consensus). Water solubility: ESOL log S, ESOL class. Pharmacokinetics: gastrointestinal absorption (GIA), blood-brain barrier (BBB), P-glycoprotein (P-gp) substrate, inhibition of cytochrome P450 (CYPs) type CYP1A2, CYP2C19, CYP2C9, CYP2D6, and CYP3A4, skin permeation (Log Kp). Drug-likeness: bioavailability score (BS). Medicinal chemistry: synthetic accessibility (SA).

Table 3. Affinity of peptides and proteins to a specific target through *in silico* model, in a search for active compounds for breast cancer.

	Selected Hydrolysate Peptide	Active	CB-Dock Vina Binding Affinity (kcal.mol−1)															
			A	B	C	D	E	F	G	H	I	J	L	M	Q	Y		
1	AIF		-7,2	-6,6	-7,6	-7,5	-5,4	-6,8	-7,1	-6,3								
2	AIN			-9							-8,6							
3	APQVAVL		-7.6		-7.9					-6.5								
4	EIL				-8.2						-8.4							
5	IAK				-7.8					-8.2			-10					
6	PSIPK				-7.1										-8.2			
7	PVGF		-7.8	-8.9	-9.1									-8.8				
8	SDTEL															-9		

A: Inhibitor of apoptosis protein 3; B: Mu opioid receptor; C: Angiotensin-converting enzyme; D: Cyclooxygenase-2; E: Delta opioid receptor; F: Neprilysin (by homology); G: Kappa Opioid receptor; H: HLA class I histocompatibility antigen A-3; I: HMG-CoA reductase; J: Disks large homolog 4; L: Cathepsin (B and K); M: Neurokinin 1 receptor y Q: Lipoxin A4 receptor; Y: Caspase-8.

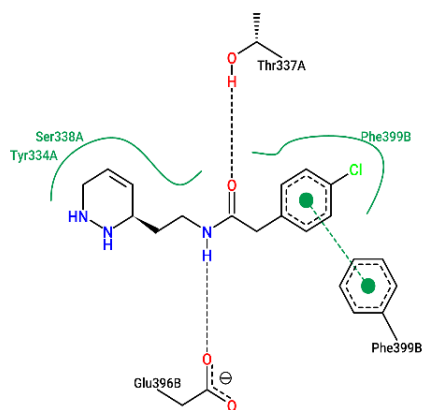


Figure 5. SDTEL with conventional hydrogen bonds (donor with Thr337A and acceptor with Glu396B). π - π interaction of chlorobenzene group with Phe399B, and hydrophobic interactions of pyridazine group with residues Tyr334A and Ser338A.

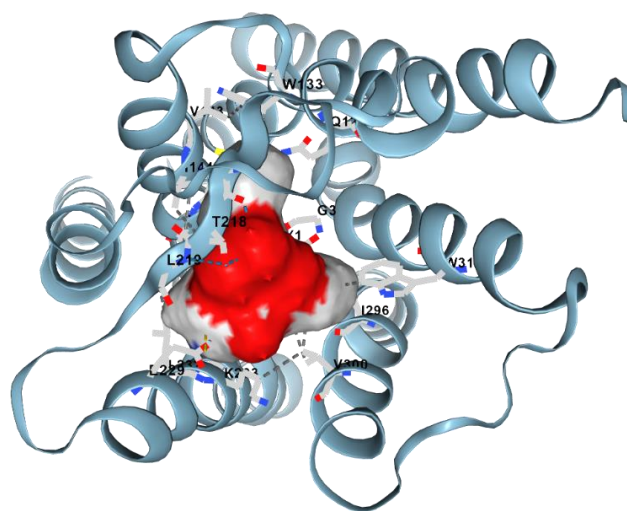


Figure 6. 3D view for AIN et Mu opioid receptor and AIF et inhibitor of apoptosis protein.

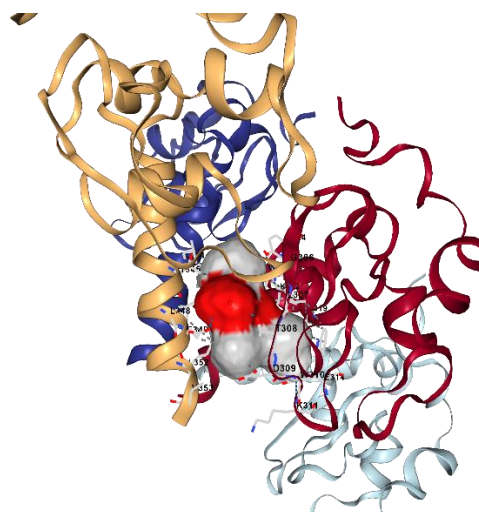


Figure 7. 3D view for AIN et Mu opioid receptor and AIF et inhibitor of apoptosis protein.

Discussion

Breast cancer is an illness that poses a worldwide risk to public health in sub-Saharan Africa (SSA). The key factors influencing the available treatments hormonal, biological, radiation, or chemotherapy are the kind and stage of the disease. Most women in SSA undergo mastectomy since there are limited options for advanced stages and few facilities for radiation and chemotherapy (4). There are several well-documented side effects of these drugs that severely reduce quality of life, and patients either relapse or become resistant to treatment. This calls for the creation of cutting-edge treatment strategies. Many peptides were synthesised specifically for this investigation, and they have shown increased affinities for certain genes implicated in the molecular carcinogenesis of breast cancer. the affinity for the protein known as the inhibitor of apoptosis protein 3 (XIAP), which is detected by the AIF peptide and inhibits apoptotic cell death. Deregulation of XIAP can result in cancer, neurodegenerative disorders, and auto immunity and high proportions of XIAP may function as tumor marker (16). AIF's capacity to attach to XIAP indicates that, in the event that XIAP is overexpressed, this peptide may interact with it and trigger apoptosis once more. The fact that XIAP exhibits affinity for AIF suggests that its primary anti-apoptotic activity may be modulated, permitting wounded cells to proceed through normal apoptosis.

Prescription opioids act through the Mu opioid receptor, which the AIF, AIN and PVGF peptides can also interact with. This receptor may be involved in perineural invasion, a crucial process in the spread of cancer (17). These peptides may function as opioids, which are the primary choice for treating moderate to severe pain brought on by cancer. Opioids, either by themselves or in conjunction with non-opioid analgesics and adjuvant drugs, are well-known to be crucial for managing pain in cancer patients during the perioperative phase. The angiotensin-converting enzyme shown affinity with a number of peptides (AIF, APQVAVL, EIL, IAK, PSIPK, PVGF). ACE2 has been reported to be tumour suppressive in multiple cancer subtypes. According to a study on breast cancer, the capacity of cells to migrate is adversely connected with ACE2 protein levels, and ACE2 downregulation aids in the invasion and metastasis of breast cancer cells (18). AIF peptide can also inter-act with cyclooxygenase-2. The overexpression of this enzyme is responsible for the elevated prostaglandin production and characteristic feature of breast cancer and his inhibition facilitates anti-inflammatory, analgesic, and antipyretic effects of non-steroid anti-inflammation drugs (19). AIF peptide could also have an effect on epigenetic regulation as they have an affinity with neprilysin. Neprilysin is a cell-surface enzyme that cleaves and inactivates a number of substrates including endothelin-1 (ET1); neprilysin negatively regulates the ET axis in breast cancer, and epigenetic suppression of neprilysin in invasive breast cancer cells enables invasion (20). This AIF peptide appears to be very important because of its

affinity with neprilysin and helps us understand that the management of breast cancer is not only genetic but should also be epigenic. For animals to develop normally and maintain tissue-specific gene expression patterns, epigenetic mechanisms are necessary. Mutations in gene function and cellular cancer can result from disruptions in epigenetic mechanisms. One characteristic of cancer is the global alteration of the epigenetic landscape. It is now understood that epigenetic anomalies, in addition to genetic modifications, have a role in the initiation and course of cancer, a disease that was previously thought to be hereditary in nature. Three peptides (AIF, APQVAVL, IAK) can inter-act with HLA class I histocompatibility antigen A-3 who is a complex of genes on chromosome 6 in humans which encode cell surface proteins responsible for regulation of the immune system. Malignant tumours frequently exhibit loss of surface expression of class I major histocompatibility antigens, which has been postulated as a means of evading cytotoxic T lymphocytes (21); these peptides could therefore have the capacity to prevent this loss of expression during carcinogenesis. AIN peptides can inter-act with HMG-CoA reductase. By decreasing intracellular cholesterol, HMG-CoA reductase inhibition may lessen the production of intratumoural autocrine hormones. It may also induce the expression of nitric oxide synthase, which will lead to the death of breast cancer cells (22). this action is possible may be because, HMG-CoA reductase is the rate-limiting enzyme for cholesterol synthesis and is regulated via a negative feedback mechanism mediated by sterols and non-sterol metabolites derived from mevalonate, the product of the reaction catalyzed by reductase. EIL peptides can inter-act with Disks large homolog 4 (DLG) who encodes the protein PSD-95, or postsynaptic density protein 95, often referred to as SAP-90, or synapte-associated protein 90. The membrane-associated guanylate kinase (MAGUK) family includes PSD-95. It is drawn into the same potassium channel and NMDA receptor clusters as PSD-93. It is possible that these two MAGUK proteins will interact at postsynaptic locations to create a multimeric scaffold that will allow receptors, ion channels, and related signalling proteins to cluster together (23). The advancement of the tumour in breast cancer may be aided by aberrant expression or activity of proteins that control cell polarity. Discs large homolog (DLG) proteins are essential for tissue morphogenesis and cell polarity maintenance. Prior research on individuals with breast cancer revealed that DLG3 was more expressed in cancerous than non-cancerous tissues; however, it is unclear how DLG3 expression relates to the course and prognosis of breast cancer. IAK peptides inter-act with Cathepsin (B and K) who is a crucial component of intracellular proteolysis, cathepsin B is a member of the cysteine cathepsins, a family of lysosomal cysteine proteases. The CTSB gene encodes cathepsin B in humans. Pre-malignant lesions, some malignancies, and a number of other clinical diseases all have elevated levels of cathepsin B. in breast cancer, Cathepsin B has a wide range of cellular origins at the original and metastatic site

and is increased in different subtypes of breast cancer. It is involved in numerous mechanisms that promote tumour growth and appears to be particularly connected to the metastasis of breast cancer to the lung and bone (24), this peptide may have a positive effect on cancer cachexia, a frequent occurrence in cancer patients. PVGF inter-act with Neurokinin 1 receptor γ who is a receptor widely expressed in the neurological and immunological systems and are members of the GPCR family. Smooth muscle contraction, secretion, proliferation, and nociception are just a few of the physiological processes that neurokinin receptors and neurokinins work together to regulate. The overexpression of SP in breast cancer is linked to a poor prognostic value, a greater tumour grade, and an expression of NK-1R with a high Ki-67 index (the nuclear Ki-67 protein is linked to proliferative pathways). The information points to NK-1R and SP as potential cancer predictors (25). Peptide PSIPK can inter-act with lipoxin A4 receptor who is an endogenous lipoxygenase-derived eicosanoid mediator, is produced from arachidonic acid. LXA4 exerts potent dual anti-inflammatory and pro-resolving effects on various cell types. LXA4 has been found to suppress leukocyte-mediated injury, promote chemotaxis of monocytes and phagocytosis of apoptotic neutrophils. In fact, lipoxin A4, a novel therapeutic drug for the treatment of breast cancer metastases, dramatically decreased the expression of CD11b adhesion receptor on human neutrophils produced by incubation with tumour-conditioned media (26). SDTEL can inter act with Caspase-8 a member of the cysteine-aspartic acid protease (caspase) family and is encoded by the CASP8 gene. A crucial part of cell apoptosis's execution phase is the sequential activation of caspases. The prodomain, big protease subunit, and small protease subunit that make up caspases are inactive proenzymes. Most breast tumours overexpress the protein caspase-8 (27).

Abbreviations

A: Inhibitor of apoptosis protein 3; **B:** Mu opioid receptor; **C:** Angiotensin-converting enzyme; **D:** Cyclooxygenase-2; **E:** Delta opioid receptor; **F:** Neprilysin (by homology); **G:** Kappa Opioid receptor; **H:** HLA class I histocompatibility antigen A-3; **I:** HMG-CoA reductase; **J:** Disks large homolog 4; **L:** Cathepsin (B and K); **M:** Neurokinin 1 receptor γ **Q:** Lipoxin A4 receptor; **Y:** Caspase-8.

Legend: SN: Serial number. Legend: Physicochemical properties: molecular weight (MW), molar refractivity (MR), total polar surface area (TPSA). Lipophilicity: log P (consensus). Water solubility: ESOL log S, ESOL class. Pharmacokinetics: gastrointestinal absorption (GIA), blood-brain barrier (BBB), P-glycoprotein (P-gp) substrate, inhibition of cytochrome P450 (CYPs) type CYP1A2, CYP2C19, CYP2C9, CYP2D6, and CYP3A4, skin permeation (Log Kp). Drug-likeness: bioavailability score (BS). Medicinal chemistry: synthetic accessibility (SA).

Conclusions

Breast cancer is a disease in which abnormal breast cells grow out of control and form tumours. If left unchecked, the tumours can spread throughout the body and become fatal. In the clinic, numerous modalities have been used for cancer therapy, such as radiotherapy, chemotherapy, and surgery. Anti-cancer peptides (APCs), a new class of therapeutic agents, have been developed in response to the fact that, despite advancements in anti-cancer medicines, their usage in the treatment of cancer is frequently linked to harmful side effects and multi-drug resistance to traditional anti-cancer drugs. These peptides (AIF, AIN, APQVAVL, EIL, AIK, PSIPK, PVGF, SDTEL) are viable possibilities for modifying breast cancer carcinogenesis, according to the findings of this study, which sought to create peptides that might interact with molecules implicated in breast cancer. in vivo and in vitro research are running, for further information concerning these peptides.

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Conflict of interest

The authors declare no conflicts of interest.

Supplementary sections

The datasets presented in this study can be found in online repositories. The names of the repository/repositories and accession number(s) can be obtained by requesting to the corresponding author.

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