Research Article CODEN: IJPRNK ISSN: 2277-8713
Mahalingam K, IJPRBS, 2013; Volume 2(6): 407-421 IJPRBS

INTERNATIONAL JOURNAL OF
PHARMACEUTICAL RESEARCH AND BIO-SCIENCE

A STUDY ON DNA BINDING AFFINITY OF CHEMICALS PRESENT IN COSMETIC PRODUCTS USING BIOINFORMATICS TOOLS

IMRAN KHAN, MOHD.FARHAN, PRATICHI SINGH, K. MAHALINGAM
School of Biosciences and Technology, VIT University, Vellore, Tamil Nadu, India

Accepted Date: 03/12/2013; Published Date: 27/12/2013

Abstract: This study focuses on the binding affinity of chemicals that are the components of widely used cosmetics, with human DNA and CYP1A2 protein, which is involved in there deactivation and excretion. Study was carried on the 21 selected possibly toxic chemicals which may be involved in DNA adduct formation and show possible proteins binding affinity, then a docking analysis has been performed by an automated docking server known as Patchdock. The five chemicals with highest Patch Dock scores with both DNA and CYP1A2 were mostly found to be important ingredient of many cosmetic products. Among these five chemicals four chemicals were found to be common namely oleic acid, polyethelene-glycol, alpha-tocopherol, and steric acid which show they have highest binding affinity towards both DNA and protein

Keywords: DNA adduct, binding affinity, docking analysis- Patchdock

PAPER-QR CODE

Corresponding Author: MR. K MAHALINGAM

Access Online On:

www.ijprbs.com

How to Cite This Article:

Mahalingam K, IJPRBS, 2013; Volume 2(6): 407-421

INTRODUCTION

The emerging science has raised a question of the risk of the chemicals present in daily used personal care products on human health. Use of chemicals such as Phenols, phytoestrogens and phthalates are found in wide-ranging of consumer products like cosmetics including shampoos, perfumes, lotions, nail polish and many more where they carry fragrance. Exposure from a cosmetic product might seem small, but daily use of cosmetic products applied directly on the skin may lead to significant levels of exposure (1).

It was also identified that 10,500 chemical ingredients that include carcinogens, degreasers, pesticides, endocrine disruptors, reproductive toxics, surfactants and plasticizers that might cause pollution. In the survey conducted by the David Suzuki Foundation in 2010 where there was a check on the ingredient lists for 12 sets of chemicals which were found to be the reason for the environmental and health concerns that forms cancer, asthma, allergies and reproductive disorders(2). The answers showed that most of the products reported many of the cosmetic ingredients and more than half of the products on which the survey was conducted proved that they contained more than one item. The chemicals that get rinsed off by humans go down the drain where a risk of contamination for the aquatic ecosystems can be traced. These components are found in the cosmetics in a very small proportion, but when the same chemicals are used regularly they add up form a large proportion and finally lead to defects. The chemicals are also found to have bioaccumulate nature and relentless (3).

DNA represents one of the most important molecular components of human cellular organization. DNA when covalently bonded to a chemical forms DNA adducts and these adducts prove to be useful as molecular markers in different scientific studies of harmful chemicals (4). These DNA binding chemicals can be conveniently categorized, depending on their mode of interaction, into two major classes (i) covalent binding, and (ii) non-covalent binding, including intercalative binding and DNA major- and minor groove binding. The interaction between small molecules and the DNA which is responsible for the particular protein is helpful for understanding the alteration in DNA, and its protein.

These chemical compounds are grouped under xenobiotic compounds, and are removed from the human body through xenobiotic metabolism pathways.A set of enzymes implicated in deactivation and excretion of these xenobiotic compounds are hepatic microsomal cytochrome P450 (5). In humans these cytochrome P450 plays a vital role as it metabolizes all the toxic compounds. There are 57 genes divided among 18 families of cytochrome P450 and 43 subfamilies in humans which include CYP1A1,CYP1A2,CYP1B1,CYP2A6,CYP3A4,CYP4A11,CYP5A1, etc. (6). Our study emphasized on CYP1A2 as it is the member of CYP family and also involved in xenobiotic metabolism.

The detail study of receptor ligand with various valuable tools also use in the field of drug development (4). For the better result and little time consumption many types of Informatics tools are used which are basically based on various mathematical algorithms. Docking is easily possible with the help of informatics tools which shows the 3D structure of the DNA and protein molecules, which display the binding with various chemical molecules. Docking is a method which predicts the preferred orientation of one molecule to a second when bound to each other to form a stable complex. Each molecule has a particular site for binding which can easily predict these sites with the help of docking. Docking also displays various alterations in the protein and its new altered structure. So for the protein chemical binding prediction there is various tools which are available free online for docking like Auto dock 4.0, Autodock Vena, Patch dock etc. Patch dock (http://bioinfo3d.cs.tau.ac.il/PatchDock/) is one of the most useful software easily accessibleonline. There is no need for installation and it gives better and accurate result in a few minutes.

The study includes chemicals which are exposed to humans in the daily routine in the form of cosmetics, these chemicals where docked using the online docking server PatchDock for identifying the binding affinity of these chemicals to the crystal structure of B-DNA and CYP1A2 proteinto check the binding affinity of these chemicals and the risk posed to the human health.

Materials and Methods

PatchDock:

It is a prediction server that plays a part in Critical Assessment of Prediction of Interactions (CAPRI). Its algorithm is motivated by image sequestration and object identification techniques that are employed in computer image (7). Here the surface of particular molecule is separated into two patches based on their surface shape. The patches like concave, convex or flat surface patches which are usually seen are perceived by means of segmentation algorithm then these patches are filtered and the one with hot spot residues are maintained. Once these patches are recognized they are superimposed via shape matching algorithm like Geometric Hashing and Pose-Clustering matching to match the above identified patches. The final step involves the filtering of all complexes with unacceptable penetration of receptor atom to the ligand atoms, finally the selected candidates are ranked on the basis of geometric shape complementarily (8)

DNA Molecule:

Crystalline structure of B-DNA with 16 base pairs is known as a receptor which was downloaded from protein data bank (PDB). Its PDB id is 3BSE. B-DNA is a regular form of DNA found in living organisms in which the double helix is right handed.

3D structure of CYP1A2 Protein:

3D structure of CYP1A2 in PDB (Protein Data Bank) format with pdb id-2HI4 was downloaded by Universal Protein Resource (UniProt), a freely accessible online server for protein sequence and functional information. It have two set of information, complete proteome and reference proteome. Complete proteome for the organism whose genome is completely sequenced and reference proteome for well-studied model organism. The structure is 95% of the natural sequence and contained 495 residues. Out of 495 residues 480 were observed and are deposited in the PDB. (9)

Ligands:

In this study 21 chemicals were used as ligands which are potential to human exposure (as shown in table 1). These chemicals are opted because they are xenobiotic chemicals and also play a role as DNA adducts and even their genotoxicity data and chemical structures are previously acknowledged, and are easily exposed to human population in numerous state of affairs (10).Their structures have been downloaded using PDB link.

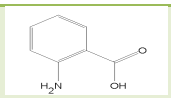
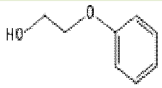
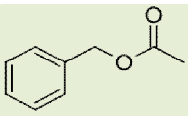
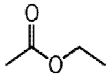
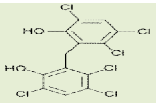
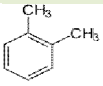
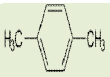
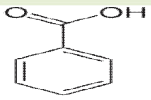
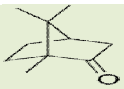
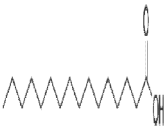
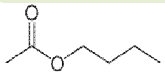
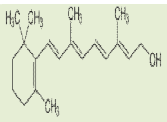
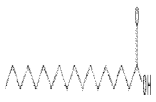
Docking procedure:

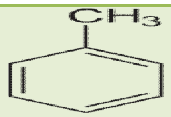
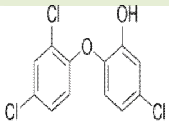
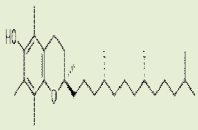
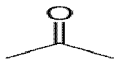
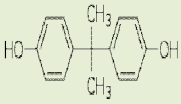
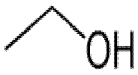
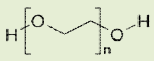
The selected receptor and ligands were docked by uploading these in Patch Dock server which is an automatic and online server for molecular docking. An email address to gain the result was specified keeping the clustering RMSD as 4.0 A°. The result was retrieved from the e-mail address given and downloaded.

Ligand-DNA Adduct Formation

To investigate the interaction of the ligands to the important DNA architecture including the DNA minor groove and major groove. The Docking analysis were performed to detect the preferable orientation and binding to the DNA structure.

Table 1:- Names of Ligands used in docking:

Serial Number	Names of Ligand	Chemical structure	Molecular formula	Molecular weight (g/mol)	PDB id
1	2-aminobenzoic acid		$C_7H_7NO_2$	137.138	BE2
2	2-phenoxyethanol		$C_8H_{10}O_2$	138.16	268
3	Benzyl Acetate		$C_6H_5CH_2OCOCH_3$	150.18	J0Z
4	Ethyl acetate		$C_4H_8O_2$	88.11	EEE
5	Hexachlorophene		$C_{13}H_6Cl_6O_2$	406.902	H3P
6	o-xylene		C_8H_{10}	106.17	OXE
7	p-xylene		C_8H_{10}	106.17	PXY
8	Benzoic Acid		$C_7H_6O_2$	122.12	BEZ
9	Camphor		$C_{10}H_{16}O$	152.23	CAM
10	Palmitate		$C_{16}H_{32}O_2$	256.42	PLM
11	Oleic Acid		$C_{18}H_{34}O_2$	282.46	OLA
12	Butyl Acetate		$C_6H_{12}O_2$	116.16	8JZ
13	Retinol		$C_{20}H_{30}O$	286.45	RTL
14	Steric Acid		$C_{18}H_{36}O_2$	284.48	STE

15	Toluene		C_7H_8	92.14	MBN
16	Triclosan		$C_{12}H_7Cl_3O_2$	289.54	TCL
17	Alpha-Tocopherol		$C_{29}H_{50}O_2$	430.71	VIV
18	Acetone		C_3H_6O	58.08	ACN
19	Bisphenol A		$C_{15}H_{16}O_2$	228.29	2OH
20	Ethanol		C_2H_6O	46.07	EOH
21	Polyethyleneglycol		$C_{2n}H_{4n+2}O_{n+1}$	variable	PE3

Results:

The list of top 21 complexes between receptor and ligand were sent to the user e-mail address. The results obtained from the patch dock server were in form of tables for each receptor and ligand docking, containing solution numbers which represent the number of solutions, geometric shape complementarity depicted by score values, according to which the solutions were sorted (7).

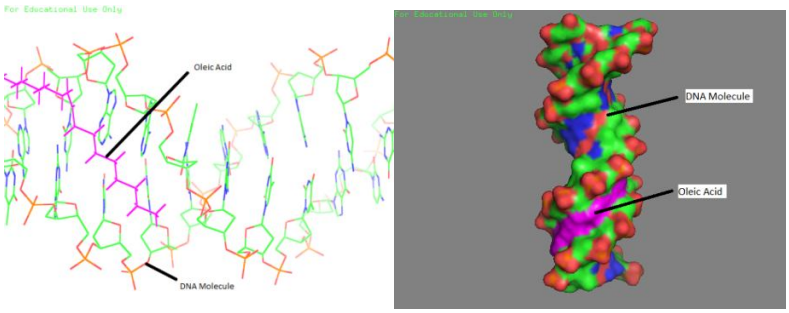
The area represents the approximate area of interface in the complex. ACE values stands for Atomic contact energy (11). Transformations were the 3D transformations including the 3 translational parameters and 3 rotational angles which are applied on the ligand molecules (12). PDB file of the complex with the highest score values were downloaded to view the structure of the complex.

Table 1- Binding of DNA molecule (3BSE) with selected chemicals

Serial Number	Ligand PDB id	Molecule	Patch dock scores	ACE values
1	OLA		5676	-336.54
2	VIV		5614	-381.31
3	STE		4772	-255.78
4	PLM		4670	-240.06
5	PE3		4650	-166.69
6	RTL		4178	-298.95
7	H3P		3822	-195.78
8	TCL		3610	-170.48
9	2OH		3416	-194.88
10	J0Z		3044	-191.37
11	268		2774	-191.16
12	8JZ		2764	-161.9
13	PXY		2732	-183.29
14	CAM		2538	-149.28
15	OXE		2510	-165.49
16	BE2		2426	-173.12
17	MBN		2420	-139.84
18	BEZ		2372	154.57
19	EEE		2352	-111.97
20	ACN		1710	-85.28
21	EOH		1458	-66.5

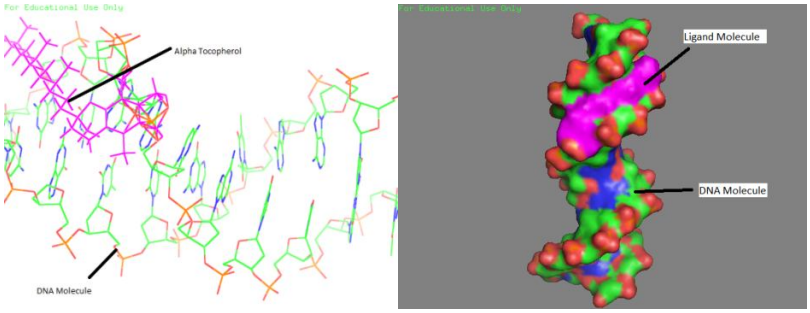
The Patch dock results of crystalline structure of B-DNA and chemicals interaction were found to show highest values for- 3BSE-OLA, 3BSE-VIV, 3BSE-STE, 3BSE-PLM, 3BSE-PE3, with patchdockscores 5676, 5614, 4772,4670, and 4650 respectively.

Binding of DNA molecule (3BSE) with selected chemicals



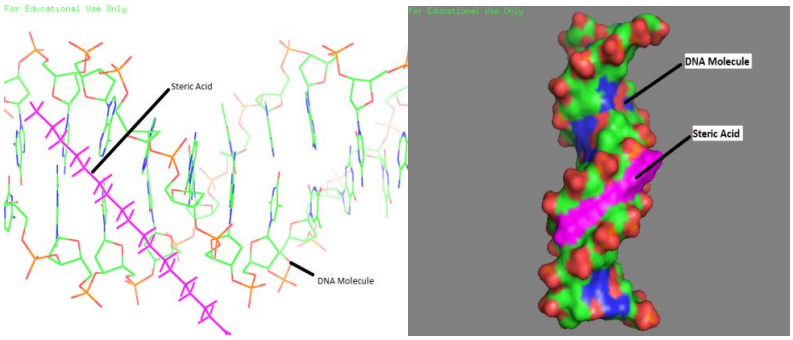
(a) (b)

Figure 1-Binding of Oleic acid to DNA molecule with score 5676, Stick Model (a), Surface Model (b)



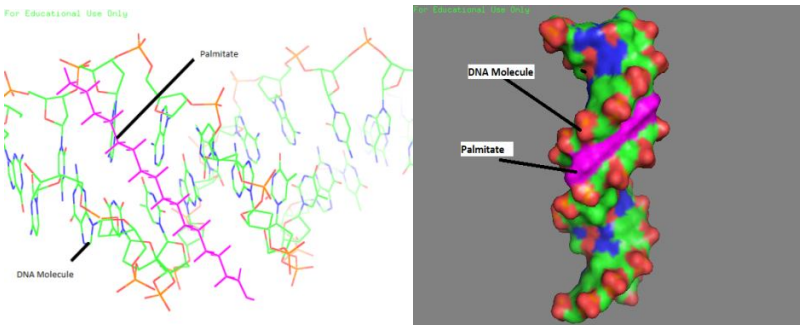
(a) (b)

Figure 2-Binding of Alpha Tocopherol to DNA molecule with score 5614, Stick Model (a), Surface Model (b)



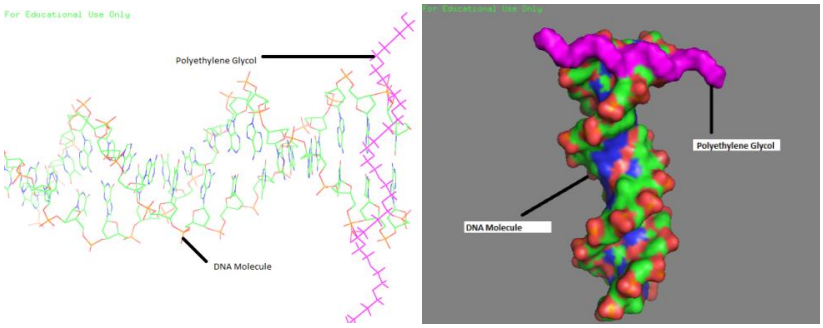
(a) (b)

Figure 3-Binding of Steric acid to DNA molecule with score 4772, Stick Model (a), Surface Model (b)



(a) (b)

Figure 4 Binding of Palmitate to DNA molecule with score 4670, Stick Model (a), Surface Model (b)



(a) (b)

Figure 5-Binding of Polyethylene Glycol to DNA molecule with score 4650, Stick Model (a),Surface Model (b)

Ligand-Protein Binding

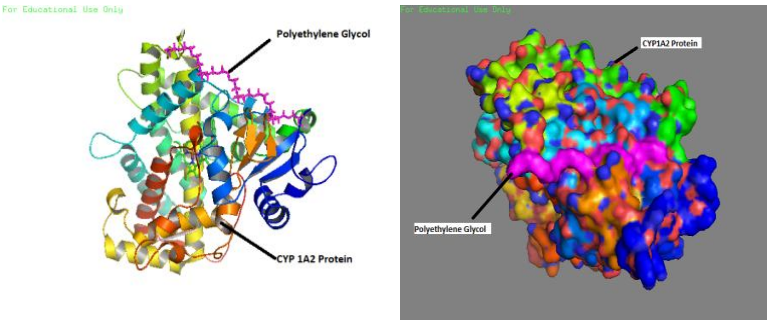
To further predict the protein binding ability of the ligand, CYP1A1 protein crystalline structure was selected.

Table 2 - Cyp1A2 (2HI4) Patch Dock score with selected chemicals.

Serial Number	Ligand Molecule PDB id	Patch dock scores	ACE values
1	PE3	7068	-79.05
2	VIV	6070	-136.09
3	OLA	5146	-84.46
4	RTL	4812	-31.12
5	STE	4742	-17.35

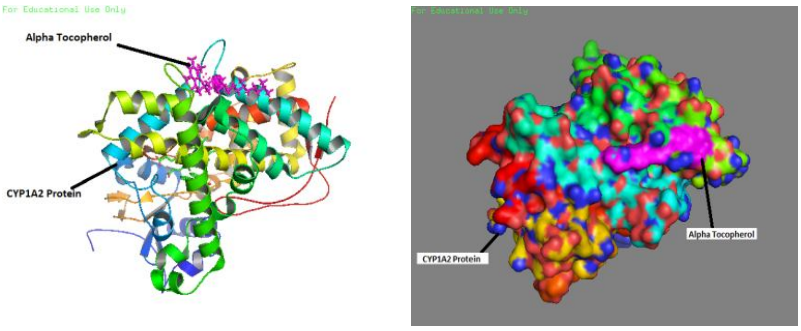
6	PLM	4644	-135.66
7	H3P	4254	-17.73
8	TCL	3926	-59.27
9	2OH	3662	-98.94
10	J0Z	3090	-114.14
11	268	2802	-51.61
12	PXY	2744	3.75
13	CAM	2666	8.85
14	8JZ	2648	-9.83
15	OXE	2642	3.47
16	BEZ	2566	-14.12
17	BE2	2468	10.82
18	MBN	2320	-48.57
19	EEE	2168	-48.91
20	ACN	1696	-6.38
21	EOH	1492	-15.6

The Patch dock results of CYP1A2 and chemicals interaction were found to show highest values for-2HI4-PE3, 2HI4-VIV, 2HI4-OLA, 2HI4-RTL, 2HI4-STE with patchdock scores 6460, 6070, 5146,4812, and 4742 respectively.



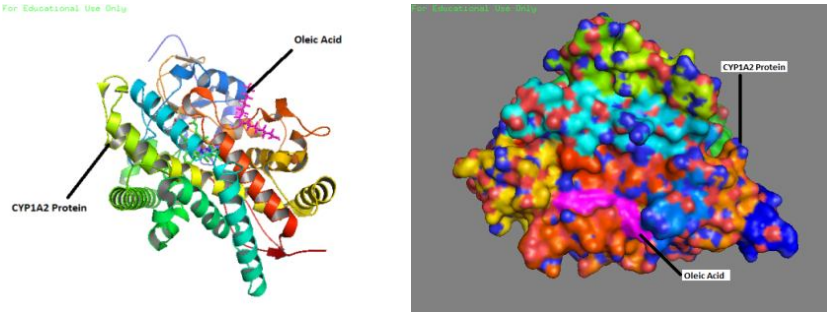
(a) (b)

Figure 6-Binding of Polyethylene Glycol to CYP1A2 Protein with score 7068, Publication Model (a), Surface Model (b)



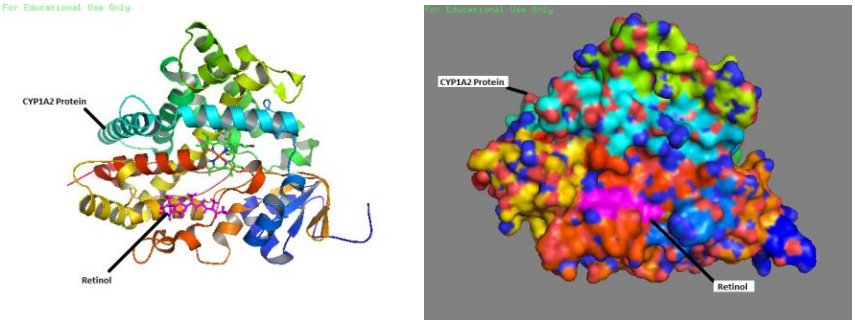
(a) (b)

Figure 7-Binding of Alpha Tocopherol to CYP1A2 Protein with score 6070, Publication Model (a), Surface Model (b)



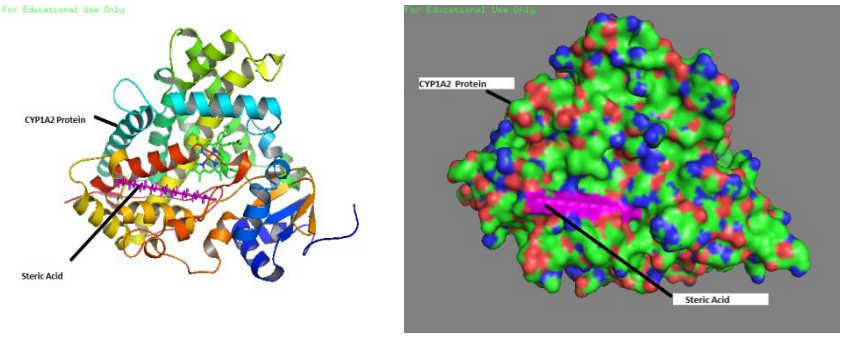
(a) (b)

Figure 8-Binding of Oleic Acid to CYP1A2 Protein with score 5146, Publication Model (a),Surface Model (b)



(a) (b)

Figure 9-Binding of Retinol to CYP1A2 Protein with score 4812, Publication Model (a),Surface Model (b)



(a) (b)

Figure 10-Binding of Steric Acid to CYP1A2 Protein with score 4742, Publication Model (a),Surface Model (b)

DISCUSSION:

The chemicals showing the highest Patch Dock score with DNA molecule and CYP1A2 protein were almost found to be in a similar trend shown in Table-1 and Table-2 respectively. Five chemicals showing highest scores with both DNA molecule and CYP1A2 protein were Oleic Acid, Polyethylene Glycol, Alpha Tocopherol, Retinol, Steric Acid, and Palmitate. This displays there higher binding affinity for both the molecules.

Oleic Acid is a type of naturally occurring fatty acid present in animals and some vegetables oils, widely used as surfactants in manufacturing of soaps (13), also as an thickening agent in most of the cosmetic products like lotion and creams.. Alpha tocopherol is a type of vitamin E that is absorbed in the intestine (14) it is used as an antioxidant to provide stability to fats in products like oils, creams, soup powders and chewing gums (15). But the high consumption of Alpha tocopherol leads to bind with DNA minor groove forming large adducts, that may be alter the DNA structure and stability, also the protein binding may result in denaturation of protein structure and also in loss of a previously available binding site. Steric acid is a natural occurring fatty acid in vegetable fats mainly used in the production of detergents, soaps, and cosmetics such as shampoos and shaving cream products.

Palmitate is a salt of Palmitic acid which is the most common type of fatty acid found in microorganisms, plants and animals (16). It is widely used in cosmetic products like conditioners and in soap manufacturing. A number of studies suggest that to UV radiation induces palmitate which generates free radicals. (17,18). Free radicals are chemically reactive substances whose interactions with DNA may cause mutations leading to cancer. And according to our study Palmitate shows fourth highest binding affinity, directing towards its possible toxic competences.

Polyethylene glycol which is widely used in cosmetics as solvents, thickners, softners, and moisture carriers (3).It is the basis of many skin creams and sexual lubricants. Continuous use of theses product may result in harmful effects like cancer and allergies in skin by increasing the skin permeability thus also known as permeability enhancer (19). Retinol is a vitamin A used in cosmetics and its antioxidant ability stands for its wide use in dermatology therapy, but major side effects of Retinol treatment is the development of hypervitaminosis-A syndrome in some cases. All these chemicals having reports of being highly used in cosmetics and the higher Patch Dock scores with DNA denoting there adduct formation ability as well as higher binding with protein predicting their protein denaturing ability.

Human skin is largest and most important passive physical barrier that protects the human body from external toxic chemicals from entering the body and defends the body in active form.Many chemicals can cross the skin barriers and enter the blood stream. These chemicals are reported to effect the XME xenobiotic metabolizing enzymes are located in keratinocytes present on epidermis layer of skin(20, 21). Thus the accurate detection of the XME levels in skin describes the consequences of the chemicals exposed to the skin (22). Our study also supports the hypothesis by showing high binding affinity of the chemicals present in the cosmetics with the CYP1A1 protein. The study is limited with the in-Silico prediction here by further in-Vivo validation is required.

CONCLUSION

The chemicals opted for this study was the xenobiotic compounds which are widely used in cosmetics and are highly exposed to humans. The five chemicals with highest Patch Dock scores with both DNA and CYP1A2 were mostly found to be important ingredient of many cosmetic products. Among these five chemicals four chemicals were found to be common namely oleic acid, polyetheleneglycol, alpha-tocopherol, and steric acid which show they have highest binding affinity towards both DNA and protein. This may predict the potential of these chemicals to bind to DNA molecules and protein, which may pose a hazardous effect of these chemicals if continued in use. Hence, the cosmetics in which these chemicals are present should be avoided and natural supplements should be preferred rather than these probably toxic compounds. Thus this research paves a way to researchers to prioritizing the harmful chemical used in large scale in cosmetic products.

REFERENCES:

1. Gunstone, FD, Harwood JL, and Dijkstra AJ: The Lipid Handbook with Cd-Rom. CRC Press 2007; 3: 24- 36.

2. Main KM, Mortensen GK, Kaleva MM, Boisen KA, Damgaard IN, Chellakooty M, Schmidt IM, Suomi AM, Virtanen HE, Petersen JH, Andersson AM, Toppari J and Skakkebaek NE: Human breast milk contamination with phthalates and alterations of endogenous reproductive hormones in infants three months of age. Environmental Health Perspectives 2006; 114: 270-276.

3. Rahman, MF, Yanful EK and Jasim SY, "Endocrine disrupting compounds (EDC s) and pharmaceuticals and personal care products (PPCPs) in the aquatic environment: implications for the drinking water industry and global environmental health," Journal of Water and Health 2009; 2: 224–243.

4. Sobhani AM, Amini SR, Tyndall JDA, Azizi E, Daneshtalab M, Khalaj A: A theory of mode of action of azolylalkylquinolines as DNA binding agents using automated flexible ligand docking. Journal of Molecular Graphics and Modelling 2006; 25: 459–469.

5. Guengerich FP: Cytochrome p450 and chemical toxicology. Chem. Res. Toxicol2008; 21(1): 70–83.

6. Nelson D: Cytochromes P450 in humans and zebrafish: CYP26B1. Arch BiochemBiophys 2005; 371(2): 345-7

7. Duhovny D, Nussinov R, Wolfson HJ. Efficient Unbound Docking of Rigid Molecules. In Gusfield et al., Ed. Proceedings of the 2'nd Workshop on Algorithms in Bioinformatics(WABI) Rome, Italy, Lecture Notes in Computer Science. Springer Verlag 2002; 2452:185-200.

8. Duhovny SD, Inbar Y, Nussinov R and Wolfson HJ: PatchDock and SymmDock: servers for rigid and symmetric docking. Nucl Acids Res 2005; 33: W363-367.

9. Sansen S, Yano JK, Reynald RL, Griffin KJ, Stout CD and Johnson EF: Adaptations for the oxidation of polycyclic aromatic hydrocarbons exhibited by the structure of human P450 1A2. J BiolChem 2007; 282: 14348-14355.

10. Pandya P, Islam MM, Kumar GS, Jayaram B and Kumar S: DNA Minor Groove Binding of Small Molecules: Experimental and Computational Evidence. J ChemSci 2010; 122(2): 247-257.

11. Zhang C, Vasmatzis G, Cornette JL and Delici C: Determination of atomic desolvation energies from the structure of crystallized protein. J. Mol. Biol 1997; 267(3): 707-726.

12. Venugopal S and Mohan R: In silico docking analysis of Staphylococcus Aureus virulent protein with antimicrobial peptides. IJPRD 2011; 3(12): 79-86.

13. Smolinske SC: Handbook of Food, Drug, and Cosmetic Excipients 1992; 1: 247–248.

14. Rigotti A: Absorption, transport, and tissue delivery of vitamin E". Molecular Aspects of Medicine 2007; 28 (5–6): 423–36.

15. Palmieri B, Gozzi G and Palmieri G: Vitamin e added silicone gel sheets for treatment of hypertrophic scars and keloids. International Journal of Dermatology 1995; 34 (7): 506–9.

16. Gunstone, FD, Harwood JL, and Dijkstra AJ: The Lipid Handbook with Cd-Rom. CRC Press 2007; 3: 40-48.

17. Yin JJ, Xia Q and Fu PP: UVA photoirradiation of anhydroretinol—formation of singlet oxygen and superoxide. ToxicolInd Health 2007; 23(10):625-31.

18. Xia Q, Yin JJ, Wamer WG, Cherng SH, Boudreau MD, Howard PC, Yu H and Fu PP: Photoirradiation of retinylpalmitate in ethanol with ultraviolet light—formation of photodecomposition products, reactive oxygen species, and lipid peroxides. Int J Environ Res Public Health 2006; 3(2):185-90.

19. Epstein SS: Toxic Beauty: how cosmetic and personal care product endangered your health…and what you can do about it. Ben Bella Books 2009; 1:1-214.

20. Oesch F, Fabian E, Oesch-Bartlomowicz B, Werner C and Landsiedel R: Drug-metabolizing enzymes in the skin of man, rat, and pig. Drug Metab Rev 2007; 39: 659–698.

21. Baron JM, Holler D, Schiffer R, Frankenberg S, Neis M, Merk HF and Jugert FK: Expression of multiple cytochrome P450 enzymes and multidrug resistance-associated transport proteins in human skin keratinocytes. J Invest Dermatol 2001; 116: 541–548.

22. Gibbs S, van de Sandt JJ, Merk HF, Lockley DJ, Pendlington RU and Pease CK. Xenobiotic metabolism in human skin and 3D human skin reconstructs: a review. Curr Drug Metab 2007; 8: 758–772.