International Journal of Chemical Studies

P-ISSN: 2349–8528
E-ISSN: 2321–4902
IJCS 2018; 6(2): 254-257

Received: 09-01-2018
Accepted: 10-02-2018

Pravas Ranjan Sahoo
Department of Veterinary Biochemistry, CVSc & A.H. Orissa University of Agriculture and Technology, Bhubaneswar, Odisha, India

Swagat Mohapatra
Department of Veterinary Physiology, CVSc & A.H. Orissa University of Agriculture and Technology, Bhubaneswar, Odisha, India

Gyanaranjan Sahoo
Department of Veterinary Biochemistry, CVSc & A.H. Orissa University of Agriculture and Technology, Bhubaneswar, Odisha, India

Prakash Chandra Behera
Department of Veterinary Biochemistry, CVSc & A.H. Orissa University of Agriculture and Technology, Bhubaneswar, Odisha, India

Correspondence
Pravas Ranjan Sahoo
Department of Veterinary Biochemistry, CVSc & A.H. Orissa University of Agriculture and Technology, Bhubaneswar, Odisha, India

Deciphering physiochemical and structural characterization of p53 tumor suppressor protein in domestic animals through *In silico* approaches

Pravas Ranjan Sahoo, Swagat Mohapatra, Gyanaranjan Sahoo and Prakash Chandra Behera

Abstract
The p53 is one important tumor suppressor protein involved in different cell processes such as DNA repair, apoptosis and cell cycle in domestic animals. Due to above importance, this protein needs to be characterized both in physiochemical and structurally for further exploration in modern biological research. In this study, the amino acid sequences of p53 protein of selected domestic animals were retrieved from NCBI site and various physiochemical parameters of this protein were compared among the selected domestic animals through different *in silico* programmes. The secondary and quaternary structure of this protein among the animals was also compared through homology modeling prediction method. It was found that this protein is an unstable, hydrophobic protein in all domestic animals with amino acids varied from 367 to 386 in number. The structural organization showed that this protein is a homo tetramer with number random coil (64%) followed by alpha helix (19%). So this study would provide a better platform for drug designer to develop new therapeutics against the cancer.

Keywords: p53 protein, physiochemical parameters, *In silico*, homology modeling

Introduction
The tumor suppressor p53 protein has important role in cancer prevention not only in human but also in domestic animals by regulating different signaling pathways in body [1]. This protein helps in various important processes such as programme cell death, senescence response, DNA repair and genomic stability in the living cells [2]. This protein is nuclear transcription factor which transactivates numerous target genes involved in the induction of cell cycle arrest [3]. Besides this, the activation of p53 leads to post-translational modifications in the protein itself, which subsequently activates p53-targeted genes [4]. This tumor suppressive function p53 protein is mainly attributed due the presence of DNA-binding domain in its structure [5]. Due to above importance, this protein needs to be characterized with respect to its structural and physiochemical properties at molecular level for better understanding during drug development against the cancers [6]. The physiochemical, secondary and quaternary structural organization of this protein is indeed for the functional study in the domestic animals under *in silico* platform. In sillico study is the method of analysis of protein structure, its physiochemical properties performed on computer simulation [7]. So this present study was carried out with the objective to develop a comparative statement regarding physiochemical and structural organization of p53 protein among domestic animals using various bioinformatics tools.

Materials and Method
This study was carried out in the Dept. of Vety. Biochemistry, CVSc & AH, Bhubaneswar, Odisha under different computational approaches. The following research works were majorly carried out in this study.

Retrival of amino acid sequence
The tumor suppressor p53 protein sequences of seven selected animals such as cattle, buffalo, goat, sheep, pig, dog and poultry along with their respective accession number CAA57348.1, NP-001277773.2, XP-005693587.1, NP-001009403.1, NP-998989.3, NP-001003210.1, and NP_990595.1 were retrieved from National Centre for Biotechnological Information (NCBI) under FASTA format

Estimation of physiochemical properties

The physiochemical parameters such as amino acids composition, Molecular weight, Theoretical pI, Extinction Coefficient, Absorbance, Instability index, aliphatic index and Grand average of hydropathicity (GRAVY) of the p53 protein among the selected animals were estimated under ProtParam characterization tools on the Expert Protein Analysis System (Expasy) server [8]. Hydropathy-plot analysis was performed using the ProtScale tool.

Prediction of secondary structure

The secondary structural organization of this protein was done to find out the alpha helix, extended strand, and Random coil composition through GORIV secondary structure prediction [9] method among the domestic animals.

Tertiary (3D) structure prediction

The 3D structure of this protein was predicted through Swiss model upon ProMod3 Version 1.1.0 programme [10] and protein sequence was identified for homology modelling. Energy levels and protein stabilization was taken into account to optimize model structure [11]. QMEAN Z 6 score was found to estimate the degree of nativeness of the predicted structure.

Result and Discussion

In this study, the results of the physiochemical properties of the tumor suppressor p53 protein in domestic animals were shown in Table No 1. It was found that the amino acids composition varied significantly from cattle (386) to poultry (367) numbers of amino acids with approximately theoretical pI between 6.34 to 8.21. This theoretical pI will help in the elucidation of other properties such as solubility and mobility of this protein [12].

Table 1: Showing physiochemical parameters of tumor suppressor p53 protein in domestic animals

Paramaters	cattle	Buffalo	Goat	Sheep	pig	dog	chicken
Number of amino acids	386	386	382	382	386	381	367
Molecular weight	43255.87	43355.94	42852.35	42809.26	42862.32	42485.92	40169.05
Theoretical pI	6.34	6.24	6.24	6.24	7.10	6.80	8.21
Instability index	80.27	78.70	77.09	80.22	70.32	71.09	73.99
Aliphatic index	62.93	63.42	62.04	62.04	64.46	63.96	67.71
Grand average of hydropathicity (GRAVY)	-0.727	-0.730	-0.711	-0.712	-0.652	-0.690	-0.555
Ext. coefficient	39140	39140	37650	37525	37525	44515	26650
Abs 0.1% (=1 g/l)	0.905	0.903	0.879	0.877	0.875	1.048	0.663

In this study, the instability index which is a measure of the stability of a protein, was found above 70, can be inferred that this protein is unstable in all domestic animals [13]. The aliphatic index of this protein was found above 60 can be inferred that, the relative volume of this protein is not occupied by aliphatic side chains amino acids which makes it more thermo unstable [14]. However GRAVY values of selected domestic animals are found less than zero, which implies that this protein more hydrophilic in nature due to presence of more serine (9.8%), and glutamic acid (8%) similar to the finding of [15]. Further the hydrobphobicity of this was checked through drawing hydropathy plot shown in fig 1., It was found that this protein has no big peaks in the N terminal but contain a large negative peak towards carboxyl terminal indicates this protein is more hydrophilic In accordance with the result of [16].

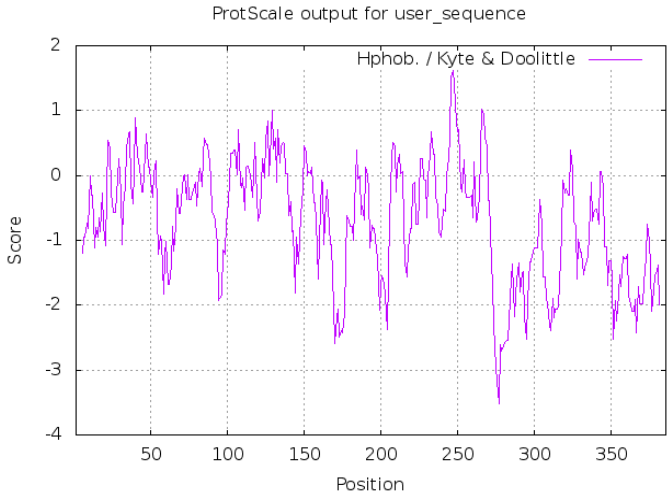


Fig 1: The hydrophobicity plot of p53 tumor suppressor protein of cattle

The secondary structure analysis of this protein showed that there is more aboundance of random coil around 64.25% in cattle but there is no such variation in the domestic animals. On other hand alpha helix composition varied more significantly among the domestic animals. It may due to more variation of proline residue among the domestic animals as stated by [17]. The result of secondary structure analysis was given in the Table No 2 and represented in Fig no 2

Table 2: Showing the composition of different secondary structure of tumor suppressor p53 protein in domestic animals

	cattle	Buffalo	Goat	Sheep	pig	dog	chicken
Alpha helix (%)	19.69	22.80	20.68	20.94	25.13	22.05	15.53
Extended strand (%)	16.06	16.06	16.49	16.49	15.80	16.80	17.71
Random coil (%)	64.25	61.14	62.83	62.57	59.07	61.15	66.76

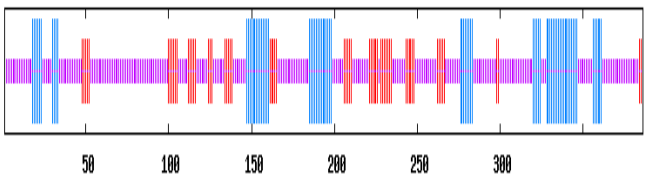


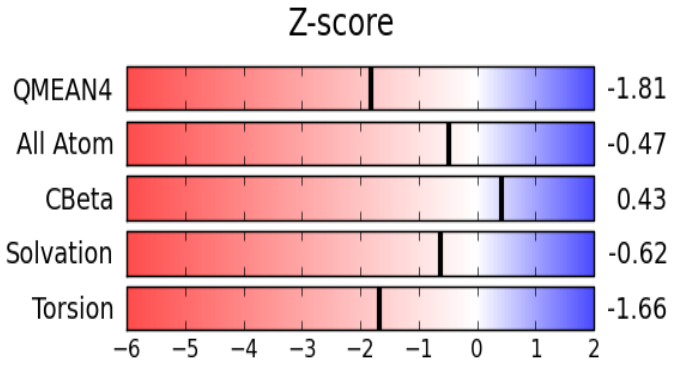
Fig 2: Secondary structure of tumor suppressor p53 Protein of bos taurus

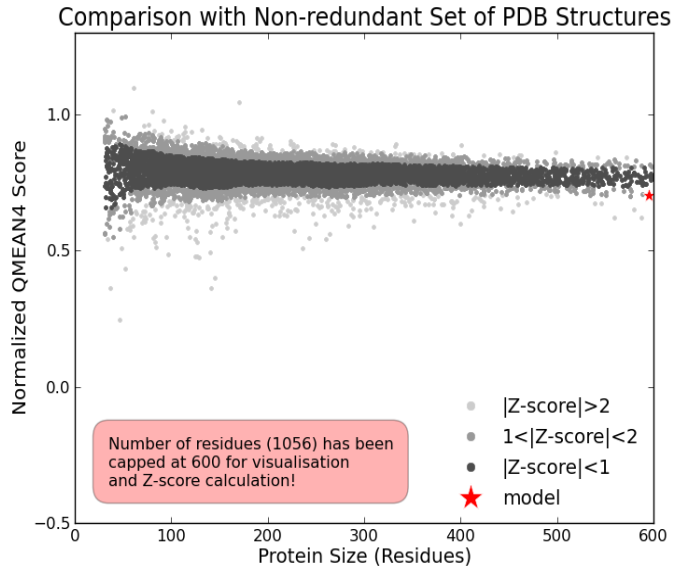
Prediction of 3 Dstructures of protein is of great importantance for the researchers to undertake further functional studies [18]. In this study the swiss modeling result revealed that this protein is a homotetramer similar to the finding of [19]. The template having maximum identity and the lowest QMEAN Z score were considered for further study. This study has given three most suitable predicted 3D modes with binding site to ligand Zinc ion. The Model II having

84.62% identity with QMEAN Z score -1.81 was considered as the best model viewed in Rasmol Graphics, shown in fig 3 of all selected animals. The QMEAN Z score which is indicator of the degree of nativeness of the predicted 3D model revealed that this model is most appropriate one for further functional study. The Z score and energy distribution curve analysis of *bos taurus* was given only in figure 4.

Fig 3: Showing 3D structure of p53 protein of domestic animals in cartoon model as displayed in Rasmol

Fig 4: Showing Z score and energy distribution curve of the best model of *bos taurus* p53 protein through homology Modeling





Conclusion

This study can be concluded that there is no much variation in physiochemical and structural organization of the p53 tumor protein among the domestic animal. This protein is a unstable, homotrimer, hydrophilic protein with large amount of random coils in its structure. The 3 dimensional structural organization of this protein give the best protein model with little variation among animals. So this study would be a better platform for the researchers for further functional studies in the domestic animals**.**

Acknowledgment

The authors are thankful to the Dean, College of Veterinary Science and Animal Husbandry, OUAT, Odisha, India, for providing the necessary facilities to undertake this study.

References

1. Vousden KH, Prives C. Blinded by the light: The growing complexity of Cell. 2009; 53(137):413-431
2. Deepthi NC, Kumar VP, Babu AR, Priyadarshini IU. Role of Tumor Suppressor Protein p53 in Apoptosis and Cancer Therapy. Journal of Cancer Science and Therapy. 2011; S17:001.
3. Lacroix M, Toillon RA, Leclercq G. p53 and breast cancer, an update. Endocrine-Related Cancer 2006; 13:293–325.
4. Bai L, Zhu W. p53: structure, function and therapeutic application. Journal of cancer molecules 2006; 2(4):141-153.
5. Ozaki T, Nakagawara A. Role of p53 in Cell Death and Human Cancers. 2, Cancers. 2011; 3:994-1013.
6. Almazov VP, Kochetkov DV, Chumakov PM. Use of p53 for Therapy of Human Cancer. Molekuliarnaia biologiia 2007; 41(6):947-963.

7. Dantas G, Kuhlman B, Callender D, Wong M, Baker D. A Large Scale Test of Computational Protein Design: Folding and Stability of Nine Completely Redesigned Globular Proteins. Journal of Molecular Biology. 2003; 332 (2): 449.
8. Gasteiger E, Hoogland C, Gattiker A, Duvaud S, Wilkins MR, Appel RD *et al*, Protein Identification and Analysis Tools on the ExPASy Server; (In) John M. Walker (ed): The Proteomics Protocols Handbook, Humana Press 2005; 571-607.
9. Garnier J, Gibrat JF. Robson B GOR secondary structure prediction method version IV. Methods in Enzymology, R.F. Doolittle Ed. 1996; 266:540-553.
10. Biasini M, Bienert S, Waterhouse A, Arnold K, Studer G, and Schmidt T *et al*. SWISS-MODEL: modeling protein tertiary and quaternary structure using evolutionary information. Nucleic Acids Research. 2014: 42:W252-W258.
11. Bhavani B, Bindu V, Srinath M, Shailaja A, Giri CC. Comparative protein profile studies and *in silico* structural/functional analysis of HMGR (ApHMGR) in Andrographis paniculata (Burm.f.) Wall. Ex Nees. Annals of Phytomedicine. 2017; 6(1):30-44.
12. Dewald I, Isakin O, Schubert J, Kraus T, Chanana M. Protein identity and envir onmental parameters determine the final physicochemical properties of protein-coated metal nanoparticles. Journal of Physical Chemistry C 2015; 119:25482-25492.
13. Guruprasad K, Reddy BVB, Pandit MW. Correlation between stability of a protein and its dipeptide composition: a novel approach for predicting in vivo stability of a protein from its primary sequence. Protein Engineering. 1990; 4:155-161.
14. Ikai AJ. Thermostability and aliphatic index of globular proteins. Journal of biochemistry 1980; 88:1895-1898.
15. Kyte J, Doolittle RE. A simple method for displaying the hydropathic character of a protein. Journal of Molecular Biology. 1982: 105-132.
16. Fung PK, Krushkal J, Weathers PJ. Computational analysis of the evolution of 1 deoxy d xylulose 5 phosphate reductoisomerase, an important enzyme in plant terpene biosynthesis. Chemistry & Biodiversity 2010; 7:1098-1110.
17. Woolfson DN, Williams DH. The influence of proline residues on alpha-helical structure. FEBS Letter. 1990; 277(1-2):185-8.
18. Bhagavathi S, Prakash A, Gulshan W. An insight to virtual ligand screening methods for structure-based drug design and methods to predict protein structure and function in lung cancer: approaches and progress. Journal of Critical Reviews. 2014; 1:10-24.
19. Zhu J, Zhou W, Jiang J, Chen X. Identification of a novel p53 functional domain that is necessary for mediating apoptosis. Journal of Biological Chemistry. 1998; 273:13030-13036.