Int J Pharm Bio Sci 2016 Oct; 7(4): (B) 702 - 706

Original Research Article **Bioinformatics**

International Journal of Pharma and Bio Sciences

ISSN
0975-6299

SEQUENCE ANALYSIS OF SERUM PARAOXONASE 1 OF BATHYERGIDAE FAMILY SPECIFIC RATS

SUBHAMAY PANDA*[1, 2] AND SUBHAJIT HAZRA[1]

[1]*Department of Pharmacy, Gupta College of Technological Sciences, Ashram More, G.T. Road, Asansol-713301, West Bengal.*
[2]*Indian Institute of Human and Social Sciences (IIHSS), Sitarampur, Asansol-713359, West Bengal.*

ABSTRACT

Initially characterized as an organophosphate hydrolase, paraoxonase (PON) was seen to catalyses the hydrolysis of paraoxon organophosphate insecticides and sarin nerve gases as well as other similar compounds. The paraoxonase gene cluster contains three adjacent gene members, PON1, PON2, and PON3. Emerging from the same lactonase precursor (fungus), all of the PON genes share profound level of sequence identity and a very similar β propeller protein structure. Formerly a study on the structural characterization of arylesterase (paraoxonase) of *Ancylostoma duodenale* was done to understand their structural improvements. The objective of this study was to determination of protein sequence level characteristics of PON1 of bathyergidae family specific rats (naked mole rat (*Heterocephalus glaber*) and Damaraland mole rat (*Fukomys damarensis*) with the help of different bioinformatical tools. The present study clearly demonstrates that protein evolution occurred within the bathyergidae family specific rats i.e., in naked mole rat and Damaraland mole-rat.

KEYWORDS: **Paraoxonase, Bathyergidae family, Sequence analysis, *Heterocephalus glaber, Fukomys damarensis,* Protein sequence**

SUBHAMAY PANDA
Department of Pharmacy, Gupta College of Technological Sciences, Ashram More, G.T. Road, Asansol-13301, West Bengal.
Indian Institute of Human and Social Sciences (IIHSS), Sitarampur, Asansol-713359, West Bengal.

***Corresponding Author**

INTRODUCTION

Elementary studies on Paraoxonase (PON) were directed to identify its very basic characteristics. The basic characteristic was of its potential to hydrolyse an organophosphorus hydrolase. In the experiment it was seen that paraoxonase catalysed the hydrolysis reaction of paraoxone – an organophosphorus insecticide, sarin – a chemical warfare agent categorized as a nerve agent and other such agents [1]. Immediately after this initial stage of findings many other experiment was conducted. These experiments threw light on other major characteristics of PONs. Observation from experiments showed that paraoxonase had a protective ability towards proteins modification as N-homocysteinylation. This was only possible because PON's had the ability to detoxify compounds as Hcy thiolactone, oxLDL etc [2]. Further investigation on the abilities of paraoxonase was made use of in 1960 for an investigation method to measure PON1 activity in various tissues and species of animals. Thus it soon became evident from the investigation on serum of humans that a considerable intra-specie difference existed in PON1 activity [3]. Mazur in 1946 explained the enzymatic hydrolysis of organophosphorus compounds carried out by animal tissues [4]. Aldridge in 1953 was the first to coin the term PON1. It was during a study of hydrolysis rates of different organophosphorus insecticides in various tissues of rabbits. It was found from the rate study that rabbit serum exhibited a high rate of paroxon degradation. It was also seen that the compound responsible for paroxon degradation was cleved by esterase [5, 6]. Thereafter esterase was separated in two categories B-esterases and A-esterases. This separation was based on whether they were inhibited or could hydrolyze substrates catalytically. On the basis of these points of differentiation Aldridge referred serum A-esterase as paraoxonase. This was thus named due to its ability to hydrolyze the toxic oxon metabolite of parathion. Furtherance of research resulted in the identification of two isomers of the original paraoxonase enzyme named as PON2 and PON3 respectively. The earlier researched one was given the name of PON1 [7]. The paraoxonase (PON's) is a family which comprises of 3 members PON1, PON2, and PON3 coded by adjacently located genes on chromosomes corresponding to7q21-22 [7-8] and on mouse on chromosome 6 [9]. Formerly a study on the structural characterization of arylesterase (paraoxonase) of *Ancylostoma duodenale* was done to understand their structural improvements [10]. Recently bioinformatics research methods make it feasible to address complex research questions in biological science with *in-silico* research analysis [11, 12]. With this starting point the objective of this study was to determination of protein sequence level characteristics of PON1 of bathyergidae family specific rats (naked mole rat (*Heterocephalus glaber*) and Damaraland mole rat (*Fukomys damarensis*)) with the help of different bioinformatical tools.

MATERIALS AND METHODS

Primary sequences of proteins (PON1) were mined from National Centre for Biotechnology Information (http://ncbi/nlm/nih.gov) [13]. Signal P 4.1 server was used for detection of signal peptide within protein sequences (http://www.cbs.dtu.dk/services/SignalP/) [14]. Sequence level characterization of PON1 including number of amino acids, amino acid composition (%) profile, number of positively charged (Arg + Lys) and negatively charged (Asp + Glu) amino acid residues were calculated upon the FASTA sequences of mature protein molecules. Multiple sequence alignment was performed with the help of Clustal-X program, followed by manual inspection for errors [15]. Protein aligned sequence sets were represented in polarity coloring scheme using Jalview tool [16].

RESULTS AND DISCUSSION

Expression of different PONs enzyme has been seen to vary with their types. PON1 and PON3 have been seen to express in liver and to small degree in kidney [17]. PON2 was seen to have expressed in arterial cell wall, small intestine, spleen, stomach, testis, heart, kidney and liver [7, 18]. Previously it was demonstrated that with the help of bioinformatical research methods complex life science related query answered in a definite manner [19-20]. In this work protein multiple sequence alignment represented in polarity coloring scheme demonstrates conserved sites as well as variable sites of proteins (Fig 1) in naked mole rat (*Heterocephalus glaber*) and Damaraland mole-rat (*Fukomys damarensis*). The variable sites present in the following sequence position at 3, 6, 9, 10, 13, 21, 44, 71, 78, 105, 108, 150, 160, 165, 192, 196, 212, 214, 251, 258, 290, 293, 314, 316, 323, 326, 331, 336, 338, 341 within the multiple sequence alignment. Substitutions of amino acids at sequence level are slightly low in number i.e., 30 in the two sequences (8.4 %). At position 3 the residue replacement in between lysine and arginine. Both this amino acid is positive in nature. Same kind of substitution observed at position 21, 214, 290, and 338. In contrast, only in two sites i.e., 108 and 323 negatively charged amino acid residues (glutamic acid and aspartic acid) substitution occurs. The replacement with a same polarity residue dictates the conservation prevails at physicochemical characteristics level. Substitution of amino acids within a related protein chains by chemical equivalents often does not have any structural consequences when the 3D structure or protein function is concerned. Substitution of amino acids by chemical equivalents usually does not have any protein structural modifications when the molecular structure is concerned. On the other hand, polar to non-polar residue substitution and polar to hydrophobic residue substitution also observed within the multiple sequence alignment. The amino acid composition of PON1 present in naked mole rat (*Heterocephalus glaber*) was analysed (Fig 2). The amino acid composition (%) were found to be as: Alanine - 5.6%, Cysteine - 0.8%, Aspartate - 5.9%, Glutamate - 7.3%, Phenylalanine - 5.4%, Glycine - 5.9%, Histidine - 3.4%, Isoleucine - 5.6%, Lysine - 6.2%, Leucine - 12.4%, Methionine - 0.8%, Asparagine - 4.8%, Proline - 5.4%, Glutamine - 1.4%, Arginine - 2.0%, Serine - 6.2%, Threonine - 5.4%, Valine - 9.6%, Tryptophan - 1.1%, Tyrosine - 4.8%. The total number of amino acids that were identified was

355.The amino acid percentage within the protein sequence of PON1 in Damaraland mole rat (*Fukomys damarensis*) was analysed (Fig 2). The amino acid composition was found to be as: Alanine - 6.8%, Cysteine - 0.8%, Aspartate - 6.2%, Glutamate - 5.9%, Phenylalanine - 4.8%, Glycine - 6.2%, Histidine - 3.7%, Isoleucine - 6.2%, Lysine - 6.2%, Leucine - 12.7%, Methionine - 1.4%, Asparagine - 4.8%, Proline - 5.4%, Glutamine - 1.7%, Arginine - 2.3%, Serine - 5.9%, Threonine - 4.8%, Valine - 8.5%, Tryptophan - 1.1%, Tyrosine - 4.5%. The total number of amino acids that were identified was 354. Figure 3 and Figure 4 depicts the positively charged and negatively charged amino acid in % present within the protein sequence respectively. Very interestingly it was observed that charged amino acids were significantly different in the two sequences of PON1 of naked mole rat and Damaraland mole rat.

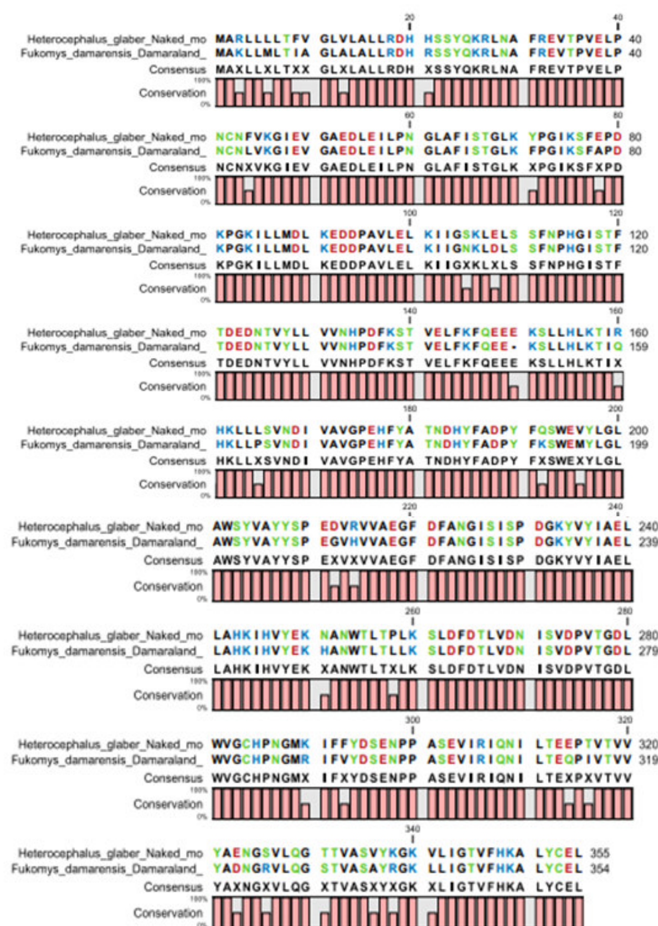

Figure 1
Multiple sequence analysis of Paraoxonase 1 (PON1) sequences of bathyergidae family specific rats: naked mole rat (Heterocephalus glaber) and Damaraland mole-rat (Fukomys damarensis).

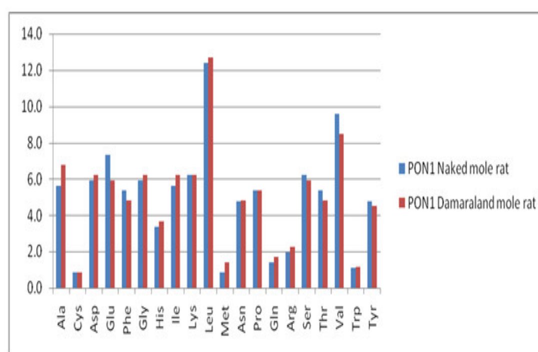

Figure 2
Amino acid characterization (%) of Paraoxonase 1 (PON1) of bathyergidae family specific rats: naked mole rat (Heterocephalus glaber) and Damaraland mole-rat (Fukomys damarensis).

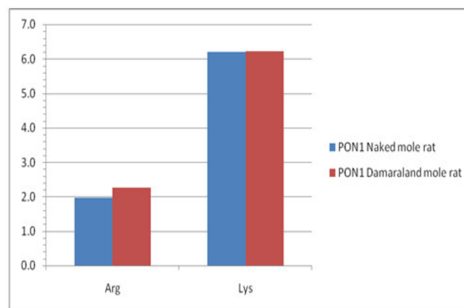

Figure 3

Positively charged amino acid (%) of Paraoxonase 1 (PON1) of bathyergidae family specific rats: naked mole rat (Heterocephalus glaber) and Damaraland mole-rat (Fukomys damarensis).

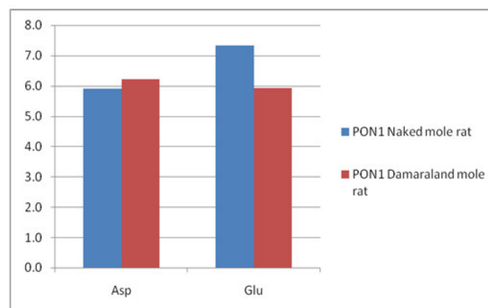

Figure 4

Negatively charged amino acid (%) of Paraoxonase 1 (PON1) of bathyergidae family specific rats: naked mole rat (Heterocephalus glaber) and Damaraland mole-rat (Fukomys damarensis)

CONCLUSION

Protein sequence specific analysis at the sequence level dictates how amino acids are mutated and generated new additional functions of bio-molecules especially in proteins. The present study clearly demonstrates that protein evolution occurred within the bathyergidae family specific rats i.e., in naked mole rat and Damaraland mole-rat. In future a detailed structural level analysis will be conducted and shaded additional knowledge to this important bio-molecule in naked mole rat and Damaraland mole-rat.

ACKNOWLEDGEMENT

We are very thankful to Prof. Debesh Chandra Majumder, Chairman, Trinity Trust, Asansol, West Bengal, Prof. Kalyan Kumar Sen, Principal, Gupta College of Technological Sciences, Aasnsol, West Bengal for providing infrastructure facilities for carrying out the research work. The authors are grateful to Late Swami Prameyananda (Ramakrishna Mission, Belur Math), Sri Shibaram Panda, Smt. Shibani Panda and Prof. Rakhi Chowdhury for the motivation and encouragement towards this research work.

CONFLICT OF INTEREST

Conflict of interest declared none.

REFERENCES

1. Costa LG, Cole TB, Jarvik GP, Furlong CE. Functional genomic of the paraoxonase (PON1) polymorphisms: effects on pesticide sensitivity, cardiovascular disease, and drug metabolism. Annu Rev Med. 2003; 54:371-92.
2. She ZG, Chen HZ, Yan Y, Li H, Liu DP. The human paraoxonase gene cluster as a target in the treatment of atherosclerosis. Antioxid Redox Signal. 2012; 16:597-632.
3. Camps J, Pujol I, Ballester F, Joven J, Simo JM. Paraoxonases as potential antibiofilm agents: their relationship with quorum-sensing signals in Gram-negative bacteria. Antimicrob Agents Chemother. 2011; 55:1325-31.
4. Mazur A. An enzyme in animal tissues capable of hydrolysing the phosphorus-fluorine bond of alkyl fluorophosphates. J Biol Chem. 1946; 164:271-89.
5. Aldridge WN. Serum esterases. I. Two types of esterase (A and B) hydrolysing p-nitrophenyl acetate, propionate and butyrate, and a method for their determination. Biochem J. 1953; 53:110-7.
6. Aldridge WN. Serum esterases. II. An enzyme hydrolysing diethyl p-nitrophenyl phosphate (E600) and its identity with the A-esterase of mammalian sera. Biochem J. 1953; 53:117-24.
7. Primo-Parmo SL, Sorenson RC, Teiber J, La Du BN. The human serum paraoxonase/arylesterase gene (PON1) is one member of a multigene family. Genomics. 1996; 33:498-507.

8. Sorenson RC, Primo-Parmo SL, Camper SA, La Du BN. The genetic mapping and gene structure of mouse paraoxonase/arylesterase. Genomics. 1995; 30:431-8.
9. Litvinov D, Mahini H, Garelnabi M. Antioxidant and anti-inflammatory role of paraoxonase 1: implication in arteriosclerosis diseases. N Am J Med Sci. 2012; 4:523-32.
10. Panda S, Kumari L. Molecular modeling and structural analysis of arylesterase of Ancylostoma duodenale. International Journal of Pharma and Bio Sciences. 2016; 7:611-616.
11. Panda S, Chandra G. Sequence analysis and phylogenetic study of some toxin proteins of snakes and related non-toxin proteins of chordates. Bioinformation. 2013; 9:259-66.
12. Panda S, Chandra G. Physicochemical characterization and functional analysis of some snake venom toxin proteins and related non-toxin proteins of other chordates. Bioinformation. 2012; 8:891-6.
13. Coordinators NR. Database resources of the National Center for Biotechnology Information. Nucleic Acids Res. 2015; 43(Database issue): D6-17.
14. Petersen TN, Brunak S, von Heijne G, Nielsen H. SignalP 4.0: discriminating signal peptides from transmembrane regions. Nat Methods. 2011; 8:785-6.
15. Larkin MA, Blackshields G, Brown NP, Chenna R, McGettigan PA, McWilliam H, et al. Clustal W and Clustal X version 2.0. Bioinformatics. 2007; 23:2947-8.
16. Waterhouse AM, Procter JB, Martin DM, Clamp M, Barton GJ. Jalview Version 2--a multiple sequence alignment editor and analysis workbench. Bioinformatics. 2009; 25:1189-91.
17. Reddy ST, Wadleigh DJ, Grijalva V, Ng C, Hama S, Gangopadhyay A, et al. Human paraoxonase-3 is an HDL-associated enzyme with biological activity similar to paraoxonase-1 protein but is not regulated by oxidized lipids. Arterioscler Thromb Vasc Biol. 2001; 21:542-7.
18. Ng CJ, Wadleigh DJ, Gangopadhyay A, Hama S, Grijalva VR, Navab M, et al. Paraoxonase-2 is a ubiquitously expressed protein with antioxidant properties and is capable of preventing cell-mediated oxidative modification of low density lipoprotein. J Biol Chem. 2001; 276:44444-9.
19. Panda S, Kumari L, Hui S, Panda S. Structural insight of homeobox DNA binding domain of Hox-B7a protein of Esox lucius. Journal of PharmaSciTech. 2016; 6:1-4.
20. Panda S, Kumari L, Panda S. Structural understanding of cytotoxin 1 of Naja sputatrix: a potential anticancer agent. Journal of Drug Delivery and Therapeutics. 2016; 6:59-63.

This article can be downloaded from www.ijpbs.net