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Phylogenetic analysis of p53 tumor suppressor gene of *bos taurus* through *in sillico* platform

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Abstract
p53, an important tumor suppressor gene that not only regulates the cell cycle but also helps in apoptosis, genomic stability, and inhibition of angiogenesis which leads a better platform for invention of new therapeutics and diagnostics in human as well as in animals by exploiting this gene. So phylogenetic analysis of this gene is essential to know the evolutionary relationship among the animals for better understanding in modern biological research. In this study, the p53 tumor suppressor mRNA gene of *bos Taurus* was retrieved and percentage identity and similarity was estimated with other selected animals through basic alignment search tool (BLAST). Multiple sequence alignment and phylogenetic analysis of this gene was performed through *in sillico* platform. In this study, the identity and similarity percentage was found more than 70% in all selected animals which inferred that this gene is conserved among all mammalian species. So this study would be helpful for the better development of research strategies about of p53 gene for the animal as well as human benefit in nearest future.

Keywords: Phylogenetic analysis, *in sillico,* p53 tumor suppressor gene basic alignment search tool (BLAST), Multiple sequence alignment

Introduction
Tumor protein p53, a homologus gene in animals, has not only important role in tumor suppression [1] but also helps in conserving genomic stability by preventing mutation [2]. It is involved in different processes like apoptosis, DNA repair, and cell cycle and anti-aging mechanism [3]. The present cancer research is being extensively going on by investigating the p53 tumour suppressor pathway, in particular in human as well as in animals [4]. Due to important role in the maintenance of stem cells throughout development, this gene has tremendous role in cell cycle as a differentiation regulator [5]. Due to above importance of this gene, the new discoveries about the function and control of p53 are still needed for development of better therapeutics and diagnostics. The phylogenetic analysis of this gene is important for investigating the patterns and mechanism of evolution [6]. The analysis of identity and similarity percentage among animals not only provides the evolutionary history but also provide the basis of comparative genomics among animals [7]. So this study was mainly focused on the comparative genomics of the p53 gene which will provide a better platform for the researcher to explore the genetic variation in p53 found in different organisms.

Materials and Methods
Retrieval of nucleotide and amino acid sequences of p53 gene
Fourteen nucleotide and aminoacid sequences of p53 gene of selected animals (cattle, water buffalo, goat, sheep, pig, Sumatran Orangutan, human being, killer whale, Small-eared galago, Domestic Cat, black snub-nosed monkey, leopard, Rhesus monkey and Wild Bactrian camel) were retrieved from National Centre for Biotechnological Information (NCBI) (http://www.ncbi.nlm.nih.gov) with accession number of gene bank under FASTA format.

Determination of identity and similarity (%) of selected animals as compared to bovine
The identity and similarity (%) of the selected animal as compared to bovine was estimated through Basic Local Alignment Search Tool (BLAST) algorithm [8] under BLASTP 2.8.0 application programme. The E value was also estimated between the animals.

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Phylogenetic analysis

Multiple and pairwise alignment of all fourteen aminoacid sequences was done by CLUSTALW programme [9]. Phylogenetic analysis of *bos Taurus* tumor protein p53 aminoacid sequences was carried out by using JTT+G model of maximum likelihood statistical method under MEGA 7 application platform [10]. The evolutionary tree was reconstructed with bootstrap method of 1000 replications.

Results and Discussions

All fourteen retrieved nucleotide and amino acid sequence of p53 gene, along with accession number and nucleotide amino acid length are shown in Table No 1. It was found that the length of nucleotide was varied much significantly among the animals i.e. from pig (1147) to black snub-nosed monkey (2595) in base pair, but the amino acids sequences were varied less significantly among the selected animals which is in accordance with the finding of [11]. It can be inferred that the amino acid sequences are more conserved among the animals which can give a better way for evolutionary analysis of this gene [12].

Table 1: Showing retrieved nucleotide and amino acid sequence of p53 gene with accession number of selected animals from NCBI

	Gene bank accession no	length	Name of gene	Gene bank accession no of protein	length	Name of protein
Bos taurus (cattle)	X81704.1	1161	p53 mRNA	CAA57348.1	386	p53 protein
Bubalus bubalis (water buffalo)	NM_001290844.2	1161	p53 mRNA	NP_001277773.2	386	p53 protein
Capra hircus (goat)	XM_005693530.3	2218	p53 mRNA	XP_005693587.1	382	p53 protein
Ovis aries (sheep)	NM_001009403.1	2155	p53 mRNA	NP_001009403.1	382	p53 protein
Sus scrofa (pig)	AF124298.2	1143	p53 mRNA	AAF28891.2	381	p53 protein
Pongo abelii (Sumatran Orangutan)	XM_002826974.4	2570	p53 mRNA	XP_002827020.2	393	p53 protein
Homo sapiens (Human being)	JQ694051.1	1270	p53 mRNA	AFN61606.1	393	p53 protein
Orcinus orca (killer whale)	XM_004266896.2	2181	p53 mRNA	XP_004266944.1	387	p53 protein
Otolemur garnettii (Small-eared galago)	XM_012806041.2	2138	p53 mRNA	XP_012661495.1	390	p53 protein
Felis catus (Domestic Cat)	NM_001009294.1	2143	p53 mRNA	NP_001009294.1	386	p53 protein
Rhinopithecus bieti (black snub-nosed monkey)	XM_017850544.1	2595	p53 mRNA	XP_017706033.1	412	p53 protein
Panthera pardus (leopard)	XM_019413568.1	2186	p53 mRNA	XP_019269113.1	386	p53 protein
Macaca mulatta (Rhesus monkey)	NM_001047151.2	2184	p53 mRNA	NP_001040616.1	393	p53 protein
Camelus ferus (Wild Bactrian camel)	XM_006175816.2	2165	p53 mRNA	XP_006175878.1	381	p53 protein

The percentage of identity and similarity of *bos Taurus* p53 protein to other selected animals is shown in Table No 2. The identity (%) was found 98% which is highest in buffalo but found 76 % i.e. in lowest in Small-eared galago. The Similarity (%) of aminoacid sequence was found more than 85% in all

Table 2: Showing percentage of identity and similarity of *bos Taurus* p53 gene to other selected animals

	Identity (%)	Similarity (%)
Bubalus bubalis	98	97
Capra hircus	93	94
Ovis aries	92	95
Sus scrofa	92	95
Pongo abelii	79	86
Homo sapiens	79	86
Orcinus orca	88	90
Otolemur garnettii	76	85
Felis catus (domestic cat)	82	87
Rhinopithecus bieti	79	86
Panthera pardus (leopard)	81	87
Macaca mulatta	79	86
Camelus ferus (Wild Bactrian camel)	85	90

Selected animals similar to the finding of [13]. It can be inferred that the p53 amino acid sequences are closely related in the animals according to their feeding habitant [14].

In modern biological research, the directional selection in molecular evolution is mainly performed through the phylogenetic analysis [15]. The result of evolutionary relationship of the retrieved fourteen amino acid sequences in this study was shown in fig 1. It was found that the *bos taurus* and *bubalis bubalis* are more clustered together and *Macaca mulatta* is more distantly related with the bovine which is

similar with [16]. It may be due the adaptive changes of p53 gene in the genetic regulation of ontogeny as stated by [17].
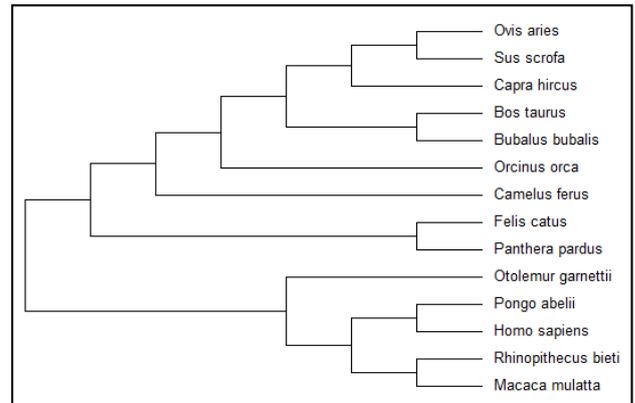

Fig 1: Showing the phylogenetic analysis of *bos taurus* p53 protein

Conclusion

It can be concluded that the p53 protein is an important tumor suppressor protein not only in bovine but also in other animals including human being. The amino acid sequences are more conserved among the animals with high percentage of identity and similarity. So this study may provide a platform for the researchers to establish the comparative genomics in animals.

Acknowledgement

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