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Research Article

Analysis of Lactate Dehydrogenase like Unnamed Protein Product of *Mus musculus*

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Abstract: In order to know the evolutionary relationships of the LDH we have investigated and characterized (*in silico*) unnamed protein product of *Mus musculus* and described protein subunit structures, domain present as well as the phylogenetic relationships for mammalian unnamed protein product of *Mus musculus*. The unnamed protein product of *Mus musculus* show 99% similarity with L-lactate dehydrogenase A chain isoform 2 of *Mus musculus* at the amino acid level. The secondary structure of unnamed protein product of *Mus musculus* is single peptide & contains 43% helix, 19% beta sheet, 37%loop and contains lactate/malate dehydrogenase, NAD binding domain and lactate/malate dehydrogenase, alpha/beta C-terminal domain. Phylogenetic analysis of 22 subunits indicated that mammalian and rodents L-lactate dehydrogenase form distinct clades. These results indicate a relatively distant evolutionary relationship between mammals and rodents.
Keywords: Lactate dehydrogenase, mammals, *Mus musculus, in silico,* Phylogenetic relationships, Domain.

INTRODUCTION

Mammalian lactate dehydrogenase (LDH; E.C.1.1.1.27) comprises three major families of conserved enzymes that catalyse the reversible interconversion of pyruvate and lactate, a key metabolic step in glycolysis and other metabolic pathways [1, 2]. Five LDH tetrameric isozymes are reported in somatic mammalian tissues, comprising LDHA and LDHB subunits, whereas the homotetrameric LDHC isozyme is found only in mature testis and spermatozoa [3, 4]. Phylogenetic studies have indicated that the LDHC gene has arisen from independent gene duplication during vertebrate evolution, including separate LDHB gene duplications in fish & birds [5-7] and an LDHA gene duplication during mammalian evolution [8]. Transcription studies have reported two other human LDHA-like genes, designated as LDH6A and LDH6B, which are expressed in brain and testis respectively and located on chromosome 11 (LDH6A in tandem with human LDHA and LDHC genes) and chromosome 15 (LDH6B, an intronless gene) [9]. The LDH-A (muscle), -B (heart), and -C (testis) polypeptides are encoded by three different gene loci that originated from an ancestral gene during the course of evolution [10]. In this study, we use the *in silico* method for predicting functions of unnamed protein product of *Mus musculus* to score protein-protein functional interaction, pairs predicted from protein sequence similarity, secondary structure, domain present & phylogenetic analysis.

MATERIALS & METHODS

Sequence of unnamed protein product of *Mus musculus* (>gi|74204388|dbj|BAE39947.1|) was retrieved from NCBI database (http://www.ncbi.nlm.nih.gov). BLAST P (http://www.ebi.ac.uk) [16] is used to search homolog's [11]. Multiple sequence alignment is done for homologous protein obtained from EBI BLAST to predict the functions of uncharacterized genes. The amino acid sequences and conserved domain were analyzed using the NCBI Blast server 2.0; and the conserved domains databases (http://smart.embl-heidelberg.de). Sequence relationship to other amino acid sequences of *Mus musculus* unnamed protein was analyzed using ClustalW (http://www.ebi.ac.uk/clustal w). The protein structure was predicted by geno 3D (http://geno3d-pbil.ibcp.fr) and RaptorX server developed by Xu group (http://raptorx.uchicago.edu/), protein- protein interactions were predicted by KBDOCK (http://kbdock.loria.fr/), the phylogenetic tree was constructed using neighbor-joining method (*http://www.ebi.ac.uk*). Guide tree, showing how unnamed protein product of *Mus musculus* is more closely related to proteins of other organisms, which is then used to predict functions of unnamed protein.

RESULTS AND DISCUSSION

The data base searching reveals unnamed protein product of *Mus musculus* which was searched for conserved domain analysis followed by modeling of protein. Result of multiple sequence alignment of 22 subunits showed that unnamed protein product of *Mus musculus* is closely related to the L-lactate dehydrogenase.

A chain isoform 2 of *Mus musculus*, it shows 99% similarity and more than 95% identity with L-lactate dehydrogenase of *Mus musculus* & *Rattus norvegicus*. The unnamed protein product of *Mus musculus* shows good conservation with L-lactate dehydrogenase protein at the amino acid level (Table 1). Immunochemical analyses and comparisons of amino acid composition were interpreted as evidence that unnamed protein product of *Mus musculus* was more closely related to the L-lactate dehydrogenase A chain isoform 2 of *Mus musculus*. The secondary structure of unnamed protein product of *Mus musculus* (Model generated by geno 3D) is single peptide & contains helix, sheet and loop (Fig. 1).

Table -1 Visual Blast of unnamed protein product of *Mus musculus* (http://www.ebi.ac.uk)

Sl. No.	Protein name/ Accession	Max score	Total score	Query cover	Ident
1	unnamed protein product [*Mus musculus*]/ BAE39947.1	679	679	100%	100%
2	L-lactate dehydrogenase A chain isoform 2 [*Mus musculus*] / NP_001129541.2	680	680	100%	99%
3	L-lactate dehydrogenase A chain isoform 1 [*Mus musculus*] / NP_034829.1	678	678	100%	99%
4.	unnamed protein product [*Mus musculus*]/ BAE41969.1	676	676	100%	99%
5.	unnamed protein product [*Mus musculus*] / BAE40283.1	676	676	100%	99%
6.	unnamed protein product [*Mus musculus*]/ BAE29167.1	670	670	99%	99%
7.	L-lactate dehydrogenase A chain [Rattus norvegicus]/ NP_058721.1	657	657	100%	96%
8.	Chain A, Rat Ldha In Complex With N-Bromophenoxy)/ 4AJ1_A	654	654	99%	96%
9	PREDICTED: L-lactate dehydrogenase A chain isoform X2 [Myotis lucifugus]/ XP_006093870.1	652	652	100%	95%
10	PREDICTED: L-lactate dehydrogenase A chain isoform X1 [Myotis lucifugus]/ XP_006093869.1	653	653	100%	95%
11.	PREDICTED: L-lactate dehydrogenase A chain isoform X1 [Jaculus jaculus]/ XP_004650857.1	653	653	100%	95%
12	PREDICTED: L-lactate dehydrogenase A chain isoform X3 [Ictidomys tridecemlineatus]/ XP_005326801.1	650	650	100%	95%
13	PREDICTED: L-lactate dehydrogenase A chain isoform 6 [Ceratotherium simum simum]/ XP_004418554.1	652	652	100%	95%
14	PREDICTED: L-lactate dehydrogenase A chain isoform X2 [Ictidomys tridecemlineatus]/ XP_005326800.1	652	652	100%	95%
15	L-lactate dehydrogenase A chain [Cricetulus griseus] / NP_001230979.1	650	650	100%	95%
16	lactate dehydrogenase A [Eospalax baileyi]/ AEI91098.1	650	650	100%	95%
17	PREDICTED: L-lactate dehydrogenase A chain isoform 2 [Felis catus]/ XP_003993090.1	650	650	100%	95%
18	PREDICTED: L-lactate dehydrogenase A chain isoform 2 [Ceratotherium simum simum]/ XP_004418550.1	649	649	100%	95%
19	L-lactate dehydrogenase A chain [Myotis davidii]/ELK24798.1	650	650	100%	95%
20	PREDICTED: L-lactate dehydrogenase A chain isoform X1 [Ictidomys tridecemlineatus]/ XP_005326799.1	651	651	100%	95%
21	PREDICTED: L-lactate dehydrogenase A chain isoform 1 [Ceratotherium simum simum]/ XP_004418549.1	650	650	100%	95%
22	Ldha protein, partial [*Mus musculus*]/ AAH05509.1	643	643	94%	99%

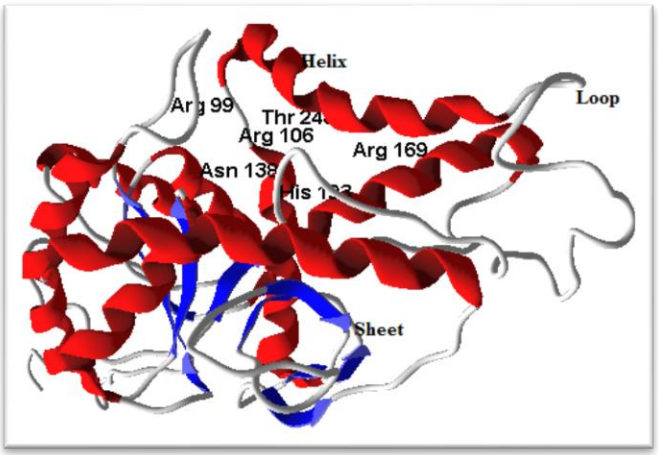

Fig. 1: Structure of unnamed protein product of *Mus musculus*

It contains lactate/malate dehydrogenase, NAD binding domain and lactate/malate dehydrogenase, alpha/beta C-terminal domain (table- 2). The L-lactate dehydrogenases are metabolic enzymes which catalyse the conversion of L-lactate to pyruvate, the last step in anaerobic glycolysis. Malate dehydrogenases catalyse the interconversion of malate to oxaloacetate [12, 13].

Table 2: Representing Pfam domain of unnamed protein product of *Mus musculus* (*http://kbdock.loria.fr/*)

Pfam ID	Pfam AC	Pfam description	Start	End	e-value
Ldh_1_N	PF00056	lactate/malate dehydrogenase, NAD binding domain	21	160	7.2e-51
Ldh_1_C	PF02866	lactate/malate dehydrogenase, alpha/beta C-terminal domain	164	324	3.4e-33

Their domains bind with inter-chain homo domain-domain interactions (Table-2 & Fig.-2) The result of Raptor X server indicates that unnamed protein product of *Mus musculus* contains 43% helix, 19% beta sheet, 37%loop (Fig- 3), protein with >100 residues, unnormalized global distance test (uGDT) >50 a good indicator, structure obtain from Raptor X server have overall uGDT (GDT): 330 (99) it also indicate that structure is good [14]. Phylogenetic analysis of 22 subunits indicated that mammalian and rodents L-lactate dehydrogenase form distinct clades. These results indicate a relatively distant evolutionary relationship between mammals and rodents (Fig. 4). The key LDH catalytic residues were present in all six human LDH subunitsincluding the active site proton acceptor (His193), as well as coenzyme (Arg99 and Asn138) and substrate (Arg106; Arg169; Thr248) binding residues [15], similarly unnamed protein product of *Mus musculus* also contain similar binding residues in same position (Fig. 1).

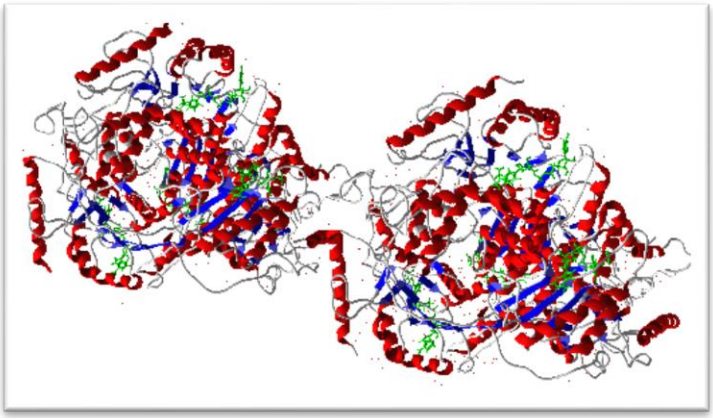

Fig. 2: *Hetro* biological interaction of unnamed protein product of *Mus musculus* (*http://kbdock.loria.fr/*)

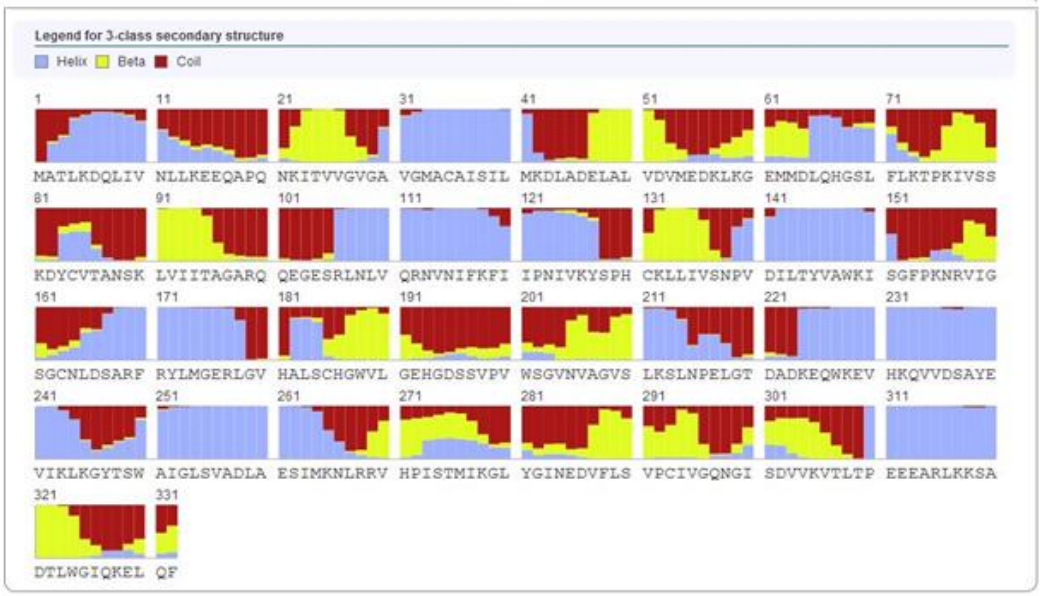

Fig. 3: Secondary structure of unnamed protein product of *Mus musculus* (http://raptorx.uchicago.edu/)

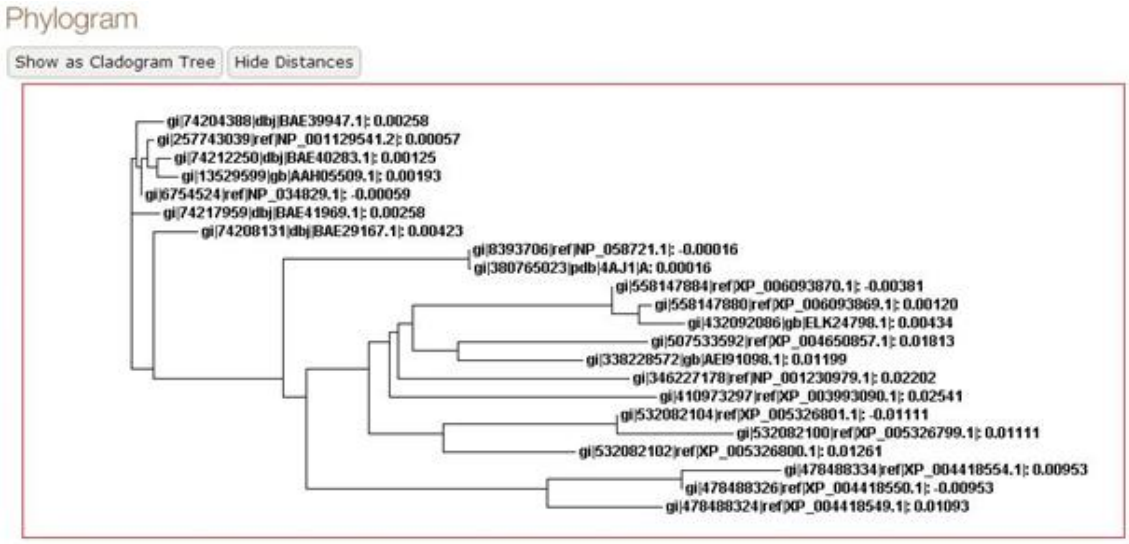

Fig. 4: Neighbor joining tree using PID percent of protein product of *Mus musculus* and homology protein present different organisms (http://www.ebi.ac.uk)

CONCLUSION

Lactate dehydrogenase like unnamed protein product of *Mus musculus* is important in medical significance because it is found extensively in body tissues, such as blood cells and heart muscle. Because it is released during tissue damage, it is a marker of common injuries and disease. It has been identified as a potential therapeutic target in the area of cancer metabolism.

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