



## Study of a Novel Pattern of MDR1 protein and its Significance as Drug target against Malaria

\*<sup>1</sup> Susanta Kumar Panda, <sup>2</sup> Amit Kumar

\*<sup>1</sup> CMJ University- Shilong, <sup>2</sup> BioAxis DNA Research Centre Pvt. Ltd, Hyderabad

<sup>1</sup>Phone: 9437111758, Email ID: susantapanda.pkd@gmail.com, sushant@dnaresearch.in

<sup>2</sup>Phone: 9246338983, Email ID: amit.kumar@dnaresearch.in

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### Abstract:

The increased incident and rapid spread of drug resistance in *Plasmodium falciparum* has become a major public health obstacle in India. Multi-Drug Resistance1 (MDR1) Protein has been one of the major protein to resist various types of antimalarial drugs. There is a need to develop new drugs against malaria combating the drug resistance ability of the plasmodium. The current research is based on the Comparative study of MDR1 protein in different Model organisms using Insilco approach to determine the potential drug target for malaria treatment. MDR1 protein from different plasmodium species and few higher vertebrates were taken for comparative study. Multiple sequence alignment (MSA), domain and disorder prediction were performed to study the patterns which are biologically significant and conserved in the protein. Through our analysis we found a specific pattern which is repeated twice in MDR1 protein of plasmodium species and the same pattern was even found in higher vertebrates. The study of this pattern signifies a specific variation in the plasmodium species that infects rodents which may illustrates uniqueness of this plasmodium species exhibiting different pathogenicity style. The conservation among the plasmodium species and higher vertebrates were studied. The mutation probability at this region might be the possible reason for drug resistance. Any mutation at this conserved region can be a potent target site. This knowledge therefore, will facilitate the rationale to design a new effective drug as well as check the emergence of multi-drug resistance.

### Introduction:

Malaria caused due to *Plasmodium Falciparum* is dangerous malignant malaria that takes millions of lives every year on a world wide scale. This Parasite has managed to develop resistance to many antimalarial drugs. The search for the molecular basis of drug resistance in the parasite has lead to the identification of the *PFMDR1* and *PFCRT* genes, which have been associated with resistance to the Chloroquin and other antimalarial Drugs in plasmodium falciparum malaria[1-2]. Resistance to

anti-malarial drugs is one of the major obstacles to effective malaria treatment and control. For the better understanding of the mechanism by which the plasmodium operate and develop resistance are necessary to implement new strategies to defeat this deadly disease. Although multilateral malaria research programmes are currently in progress to study the molecular biology of the parasites, a definitive explanation for the drug resistance remains elusive. New approaches like the use of rodent plasmodia represent interesting in vivo models that could help to understand better the molecular aspect of drug resistance in the plasmodia. All over world particularly in endemic areas plasmodium falciparum resistant to chloroquin and sulphadoxine-pyrimethamine is common. In India as per the World Health Organisation (WHO) guideline National Vector Disease Control programme has undertaken [3]. At present ACT (AS+SP) combination Drug Therapy is being implemented in 117 districts of high endemic districts of States [4-5]. Protein interactions and specific Amino Acid position changes and Domain analysis of Membrane transporters, such as the *Plasmodium falciparum* multi drug resistance 1(*pfmdr1*) and the *Plasmodium* chloroquine resistant transporter (*pfcrt*), which is a member of the ABC superfamily, have been identified as key contributors in decreasing susceptibility to several anti-malarial drugs[6]. Research to identify additional potential contributors to *Plasmodium* drug resistance has lead to the identification of new candidate transporter Proteins, some of which belong to the ABC transporter super family. Alteration in the parasitic membrane protein *PFMSF*, *PFCRT* and *PFMDR1* are believed to be major contributors to resistance through decreasing intracellular drug accumulation. The *PFMDR1* Protein was classified under the ABC transporter super family based on the sequence and organization of their conserved Amino Acid binding domains [7]. Characteristic motifs within these Amino Acid binding sites are found in the majority of AAA Domains. This review highlights the patterns of MDR1 protein

associated with the heterologous expression of this protein and its association with functional and pathological effect.

#### Material and Methods:

In the current study, Multi drug resistance protein is taken from 2 Plasmodium species which infects Humans (*Plasmodium Falciparum* and *Plasmodium vivax*), 4 plasmodium species which infects Rodents (plasmodium species are

*Plasmodium Yoelii Yoelii*, *Plasmodium Chabudi*, *Plasmodium Berghei*, *Plasmodium Knowlesi*) and from Higher vertebrates which includes Homo sapiens, Mus musculus, Rattus norvegicus. . Sequence analysis was done using Blast to find the sequence similarity between different MDR1 proteins. For performing comparative study Multiple sequence alignment is performed using CLUSTAL W. To study the patterns in the sequences TEXSHADE tool is used. Specific patterns in the conserved region were analyzed and the biological significance of the patterns was

studied. The probable disorder prone sites in the patterns were analyzed using the disorder prediction tools RONN and DISEMBL. To identify the role of patterns in protein function Domain analysis is performed using SMART. Finally A comparative homology studies of the *MDR1* protein in different level of Model organisms were done.

#### Results and Discussion:

From the sequence analysis we found that the MDR1 protein was showing maximum similarity among Plasmodium species infecting Humans and Rodents. The Plasmodium MDR1 protein when compared with the higher vertebrates showed less similarity of 30%. From the multiple sequence alignment various conserved patterns were analyzed. From the analysis we found that the specific pattern of conserved region

**LSGGQKQRISIRAIMRNP KILILDEATSSL**  
D is repeated in the protein sequence at the position 463-491 and 1321-1343.

**Table 1: Results of Conserved patterns along with its position and features**

Conserved Regions/Residues- Amino Acids	Position	Domain	Disordered Regions
SGCGKST	415-421	AAA-Domain	
IGVVSA	457-462	AAA-Domain	
SLP	547-549	AAA-Domain	
TLVG	554-557	AAA-Domain	
<b>LSGGAKQRI</b>	563-571	AAA-Domain	Disorder
<b>I</b>	573	AAA-Domain	
<b>RNP KIL</b>	579-584	AAA-Domain	
<b>LDEATS</b>	586-591	AAA-Domain	
IAHRLST	619-625	AAA-Domain	
RFY	1133-1135	AAA-Domain	
GKST	1166-1169	AAA-Domain	
IVSQEP	1253-1258	AAA-Domain	
ENI	1267-1269	AAA-Domain	
<b>LSGGGQKQRIAIARAL</b>	1312-1326	AAA-Domain	Disorder
<b>ILLLDEATS</b>	1332-1340	AAA-Domain	
<b>LD</b>	1342-1343	AAA-Domain	Disorder
SEK	1346-1348	AAA-Domain	Disorder
IAHR	1368-1370	AAA-Domain	
IVV	1381-1383	AAA-Domain	
LP	71-72	ABC-Membrance	
FG	78-79	ABC-Membrance	
GIG	165-167	ABC-Membrance	
F	179	ABC-Membrance	
LTL	194-196	ABC-Membrance	
FWYGT	294-298	ABC-Membrance	

Table 1 shows the conserved patterns highlighting the major repeated conserved pattern

**“LSGGQKQRISIRAIMRNP KILILDEATSSLD”**

Even though MDR1 protein of higher vertebrates was showing very less similarity when compared with the plasmodium, this specific pattern is even conserved in MDR1 of Higher vertebrates also, showing the significance of the pattern. The further

analysis of this pattern through domain, disorder prediction found that these conserved regions are lying under AAA domain and these patterns are found to be disordered.

**Figure 1:** Fig 1 shows the pattern **LSGGQKQRISIAIRNPNKILILDEATSSLD** from 463 to 491 position of MDR1 protein.



**Figure 2:** Fig 2 shows the pattern **LSGGQKQRIAIARALLREPKILLLDEATSSLD** from 1312 to 1343 position of MDR1 protein

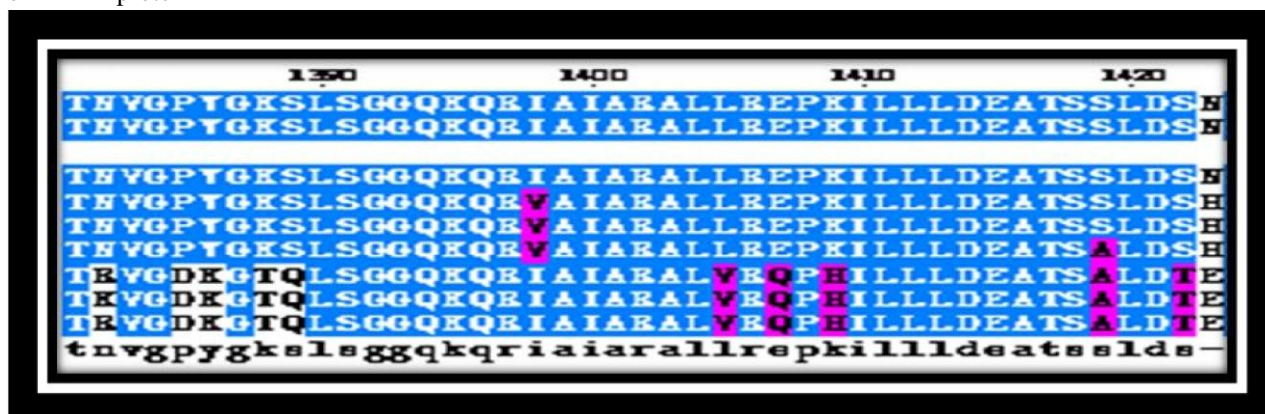


Fig 1 and 2 shows the specific pattern **LSGGQKQRISIAIRNPNKILILDEATSSLD** of MDR1 protein in Different Model organisms. First 2 rows shows the protein pattern of Plasmodium species infecting Homo sapiens, Next 4 rows shows the protein pattern of Plasmodium species infecting Rodents, Last 3 rows shows the protein pattern in Homo sapiens, Mus musculus and Rattus Novergicus respectively.

From the overall analysis we traced a common pattern which is present twice at the Amino Acid positions 463-491 and 1312-1343 in the same protein sequence. In this pattern we have observed a common variation in Plasmodium species infecting Rodents where in place of I, V is present and G in place of A. This variation is only found in Plasmodium species infecting rodents suggesting

the unique pathological style of the plasmodium species indicating that they are rodent specific. This gives a scope for the comparative analysis of rodent and human infected plasmodium species. TEXSHADE analysis also gives the information about the conservation within the MDR1 pattern in Plasmodium species and higher vertebrates. There is a minor variation in the conserved pattern. The

mutation in these positions imparts drug resistance within the plasmodium species. The mutational study in these positions helps to suggest a potential target for anti malarial drug therapy. Additional research is required to test this hypothesis and targeting such sites of mutation for drug designing and docking. The current study may be proved to be a favorable approach against malarial drug therapy.

#### Conclusion:

Drug resistance is an important problem in Falciparum Malarial diseases, which is very difficult to tackle by the limited number of drugs available. A good knowledge on drug resistance can help the researcher to find strategies to increase the efficacy. From the overall analysis we traced a novel pattern that was conserved among the species and was in two repeats in a single protein sequence. The pattern gave significant information about the uniqueness of Rodent infected plasmodium falciparum and predicting the possible potent drug target for malarial treatment.

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