

Proving similarity between MRJP1 and Yellow Protein with using blast and NCBI.

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Abstract

The nature of honeybee social organization is being significantly influenced by the Yellow/MRJP protein family. In Yellow gene and MRJP1 gene, honeybees have 6 exons, with 5 introns, but their sizes are different from each other. *Apis mellifera* has 10 Yellow like gene, and 10 MRJP like gene. Amino acids of MRJP1 were collected from National Center for Biotechnology Information (NCBI). Amino Acid sequences were used in RaptorX to find protein binding prediction. And amino acids of Yellow gene were collected from National Center for Biotechnology Information (NCBI). Amino Acid sequences were used in RaptorX to find protein binding prediction. we have seen that in spite of their difference in their characteristics, but they have similar functional domain based on all researched that we had especially based on RaptorX that we could find their domain bindings. So, as we have found a single intellectual predecessor that encodes a member of the old existing Yellow family seems to have developed and called MRJP.

Keywords: MRJP, NCBI, Raptor, Biotechnology, *Apis mellifera*.

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Introduction

Cooperative brood care, overlapping generations within a colony of adults and a division of labor into reproductive and non-reproductive groups define Eusociality. In the *Apis* genus, many workers, sterile females, coordinate hive maintenance and tend to eggs laid by a single reproductive female, queen. The worker-queen differentiation has been very well studied and characterized. It results from nutritional differences during development where eggs exclusively feed "Royal Jelly", a high protein rich substance develop into queens. Royalactin, a 57-kDA protein in royal jelly; drives queen development through an EGfr-mediated signaling pathway [1]. However, it is often found in trace amounts while the Major Royal Jelly Protein (MRJP) family constitutes 90% of the total protein content found in royal jelly. *Apis mellifera* royal jelly contains 9 well-characterized mrjp genes, mrjp 1-9. However, the specifics of the MRJP's family role in queen development are no known. MRJP1 expression on Honeybee mushroom suggests a potential role in behavior [2] but expression pattern of the MRJPs genes across caste have not been systematically characterized [3]. Still, MRJP have been strongly linked with the evolution of eusociality.

The MRJP family appears to have evolved from a single progenitor, a member of the ancient, multifunctional Yellow Protein family. Expression of the yellow gene regulates body pigmentation and behavior. Its transmission is also independent of the fly's sex determination pathways. Mark David Drapeu and his team characterized the intron/exon structure of the 19 protein-encoding mrjp and yellow genes from mrjp and yellow genes from *Apis mellifera* through cDNAs sequencing and genomic comparison with their respective genes in *Drosophila melanogaster*. They concluded that the intron/exon structures of the mrjp genes are highly conserved, with each gene having

five introns in their coding sequences, all located in the exact position. This contrasted heavily with yellow's wide distribution leading them to conclude that MRJPs were a monophyletic family within only *Apis* genus. Since, then however multiple genomes have been sequences including a eusociality wasp species *Nasonia vitripennis*, and other closely related member of the bumblebee family.

MRJP protein was found and distinguished in the blood-sucking insects. It processes an agglutination activity which probably leads to intermediating in the evolution from Yellow like function towards the components of Royal Jelly. MRJP1 contains 60-70% of moisture, it's crude proteins are 12%-15%, total sugar 10%-16%, lipids 3%-6%, free amino acids, salts, and vitamins. MRJPs and Yellow are among proteins that have tendency to malfunctioning. They have diverse roles. Their roles are context dependent physiological [3]. Anyhow, a lot of the yellow/MRJP family members are facilitators of reproductive maturation. It seems that MRJP protein subfamily from the Yellow Protein family might have coincidence with the evolution of honey bee Eusociality. *A. Mellifera* caste calculation and determination happens when worker bees and living in the hives (nurse bees) produce, secrete, and feed a substance called Royal Jelly (RJ) in order to develop larvae. RJ is a natural source of essential amino acids, lipids, vitamins, acetylcholine, and a lot of other nutrients [4,5]. In the third stage of upbringing, all larvae are fed RJ; but after this stage the larvae who are appointed as future queens by workers receive RJ [3].

As the Major content of Royal Jelly (RJ) proteins, its proteins (MRJPs) are identified and consist of 80%-90% of the total RJ proteins which play significant and central role in honey bee development process. It is indicated in a recent report that MRJP/Yellow Protein families in *Apis mellifera* have at least 8

MRJP. Most MRJPs have shown characteristics that their segments are encoding the long homopeptides at the carboxyl terminal. It is thought that the structure is the accessible form of storing nutrition. There is a strong hydrophobic sequence functioning as putative signal peptide, in the N terminal of MRJP/Yellow Protein [6]. A single intellectual predecessor gene that encodes a member of the old existing Yellow Protein family seems to have gradually developed from a single intellectual predecessor gene [3]. Thus, we hypothesized that functional domain in Yellow gene and MRJP1 are same.

Methods

Amino Acids of MRJP1 were collected from National Center for Biotechnology Information (NCBI). Amino Acid sequences were used in RaptorX to find protein binding prediction. Figure 1 shows a picture of prediction binding prediction.

Amino Acids of Yellow gene were collected from National Center for Biotechnology Information (NCBI). Amino Acid sequences were used in RaptorX to find protein binding prediction. Figure 2 shows a picture of prediction binding prediction.

They used HPLC techniques to purify MRJP1 oligomer, and they had the following analysis: The molecular of MRJP1 oligomer was 290 KDa. They used blue native page. By proteome analysis, they found out that the 55 KDa proteins were identified as Apisimin by sequencing terminal amino acid. Within MRJP1 oligomer, this protein might act as a subunit joining protein [7].

Results

We hypothesized that functional domain in Yellow gene and MRJP1 are same:

Major Royal Jelly Proteins (MRJPs) contains 90% of total RJ protein. MRJPs have a share of a common evolutionary origin with the Yellow Protein family. This is a representative from insects and some bacteria. 10 MRJP genes are dually arrayed in a 60 kb cluster. They are flanked by five Yellow genes. An analysis of the intron/exon structures of these genes and the sequences of their protein products suggests that the MRJP gene array evolved *via* multiple, rapid duplications of a specific yellow precursor, Yellow-e3 transcriptional profile of MRJP and Yellow genes in the honey bee shows that despite some similarities, the proteins which are encoded by these genes have evolved diverse physiological and developmental functions. Anyhow, the data given here, in combination with all the information from flies and ants, indicates a common theme. The nature of honey bee social organization is being significantly influenced by the Yellow/MRJP protein family. In Yellow gene and MRJP1 gene, honey bees have 6 exons, with 5 introns, but their sizes is different from each other. *Apis mellifera* has 10 Yellow like gene, and 10 MRJP like gene. No Yellow like genes is found in *Apis cerana*, and *Apis flora*. The structure of Intron/Exon of MRJP genes are highly stored up. They are exactly located in the same position with each gene having five introns within their coding sequences. MRJP is

being found in head and brain tissue *via* Northern Blot, ESTS, PCR, Microarray, and Proteomics [3]. Some of the MRJP genes with having well characterized nutritive role; they are highly expressed in worker bee hypo pharyngeal glands that secrete RJ because all nine MRJPs have been found in the RJ proteome. For instance, Yellow gene shows queen specific expression. It is being shown and expressed in very young embryos, with a maternal expression matter. The Yellow-h gene is predominant and expressed at larval stage, in developing queens. It is suggesting a special role for this gene's product. Implying about nutritional role of MRJP, it is going to have phenotypic implications in the gland as compared to brain functions or developmental processes. The biological significantly importance of a given MRJP product will depend on where and when its message is expressed [3]. When it comes to pigmentation (melanization), Yellow is required in cuticle cells for the presence of Do PA melanin, and wrongly expressed Yellow is enough and sufficient for the deposition and formation of melanin [8,9]. The properties of nearby cells may influence Yellow by dopamine-like receptors and a hormone-like mechanism [10]. When we activate different signal transduction pathways in different cell types (Cuticle *vs.* neural), it would allow the different downstream, genes relevant to particular phenotypes (pigmentation *vs.* behavior) to be activated [3]. Similar to Yellow, the MRJP's have N terminal signal peptides directing their secretion from cells, and post -secretion. These signals are chopped and broken apart [11]. Furthermore, like Yellow [12], MRJPs are glycoproteins with N linked sugar chains [13].

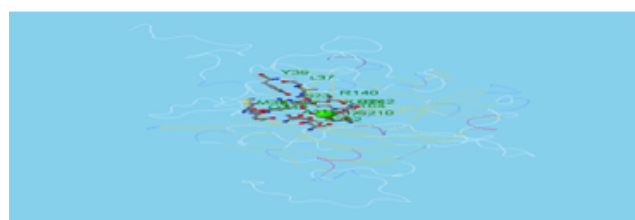
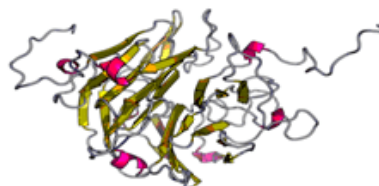


Figure 1. This is the binding site prediction of MRJP1 protein. This was made by RaptorX program.

- Binding residues: S23 L37 Y39 R140 S142 E164 K165 D207 S210 S262 A311 M341 Q342 K34, Corresponding ligands: CA, Binding residues: F33 V48 Y63 P64 D65 L91 F113 L115, Corresponding ligands: GIV, GXL, FUC, FUL, GAL
- Binding residues: W92 L110 K122 Q123 V124 E125 T157 V159 Y172 N174, Corresponding ligands: FUC, GXL, FUL, SFU, NAG

- Binding residues: N220 Y222 L231 Y233 H250 E252 L273W28E288, Corresponding ligands: PG4, FUC, MFU, SFU, AS
- Binding residues: D30 I32 K51 G57 L335 I358 N360, Corresponding ligands: GXL, GIV, FUC, GAL, SFU.

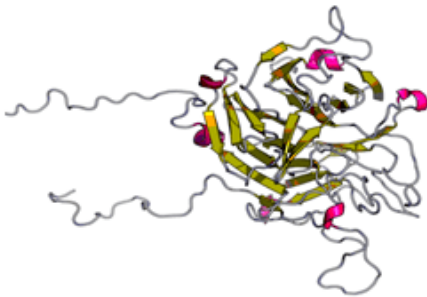


Figure 2. This is binding site prediction of Yellow Gene.

This was made through RaptorX program. 1) Binding residues: V59 Y120 R121 A180 F248 T307 F353 V374, Corresponding ligands: CA, 2) Binding residues: V310 V357 K358, Corresponding ligands: CA, EDO, 3) Binding residues: P73 W75 A119 Y120 T177 F178 E200 L201 E245 I247 F248 P373 V374 F375, Corresponding ligands: AHR, GOL, DAN, EDG, ACP, 4) Binding residues: N362 N364 P393 A395 T396, Corresponding ligands: CA, MN, 5) Binding residues: P254 Y260 R261 T262, Corresponding ligands: CA, MN.

In Yellow gene based on RaptorX and (Figure 2), V and K are query residues predicted binding sites. But in MRJP1 based on Raptor X and figure1; S, L, Y, K, M, Q, E, A are query residues predicted binding sites.



Figure 3. The NHL (NCL-1, HT2A and LIN-41)-repeat is found in multiple tandem copies, typically as 6 instances.

It is about 40 residues long and resembles the WD repeat and other beta-propeller structures. The repeats have a catalytic activity in Peptidyl-glycine alpha-amidating monooxygenase; proteolysis has shown that the Peptidyl-alpha-hydroxyglycine Alpha-amidating Lyase (PAL) activity is localized to the repeats. Tripartite motif-containing protein 32 interacts with

the activation domain of Tat (Figure 3). This interaction is mediated by the NHL repeats [14].

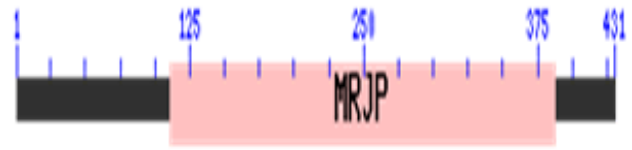


Figure 4. Figure of Yellow protein.

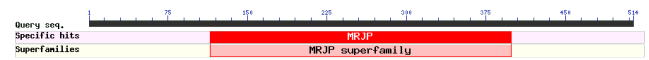


Figure 5. Royal jelly is the food of queen bee larvae, and is responsible for the high reproductive ability of the queen.

Major royal jelly proteins make up around 9 of larval jelly proteins (Figure 5). This family also the sequence-related Yellow Protein of drosophila which controls pigmentation of the adult cuticle and larval mouth parts [14].

Ligand name in MRJP1 and Yellow gene are CA, so they have same ligand names. As we know, MRJP1 locates on chromosome 11, CDNA length: 1430, number of introns: 5+1. This family also the sequence-related Yellow Protein of drosophila which controls pigmentation of the adult cuticle and larval mouth parts, based on Figures 4 and 5 [14]. But, Yellow gene is on chromosome 10, CDNA length: 1436 Number of introns: 5+1 [3]. But Yellow genes have and shares intron/exon structure of MRJP genes characteristics [10]. As we hypothesized that MRJP1 and Yellow gene are same based on fictional, we have seen that in spite of their difference in their characteristics, but they have similar functional domain based on all researched that we had especially based on RaptorX that we could find their domain bindings. So, as we have found a single intellectual predecessor that encodes a member of the old existing Yellow family seems to have developed and called MRJP [10].

Discussion

As we see that most MRJPs have shown characteristics that their segments are encoding the long homopeptides at the carboxyl terminal. It is thought that the structure is the accessible form of storing nutrition [6]. There is a strong hydrophobic sequence functioning as putative signal peptide, in the N terminal of MRJP/ Yellow Protein [6]. Also data were analyzed in blast and NCBI; we got the result because MRJP1 and Yellow Protein have similar protein domain, they have similar set of sequence, so they function similar. We hypothesized that functional domain in Yellow gene and MRJP1 are same, and we got results from NCBI and Blast which proved our hypotheses. Data were helped us to recognize that MRJP1 and Yellow Protein are similar to each other from aspect of functionality.

Conclusion

We demonstrated that MRJP1 which has important role in honeybees, and Yellow Protein are similar functionality by using RaptorX and Blast. In future, they can search which

genes are most responsible for MRJP1 and Yellow proteins, and what will their benefits be?

Acknowledgments

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