**In silico Identification and Analysis of Iron (Fe) Transporters in Various Plant Species**

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**Authors’ contributions**

This work was carried out in collaboration among all authors. Author AD designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors RR and MS mentored and monitored the work and reviewed the manuscript. All authors read and approved the final manuscript.

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**ABSTRACT**

In plants, Iron is an important micronutrient which is required for various processes like photosynthesis, respiration and for balanced redox potential. Iron has a significant role in human nutrition. Therefore, increasing the Fe content of economical parts through conventional, Molecular and/or Transgenic breeding will have dramatic impact on human health. Therefore, this study was aimed to identify and characterize homologues of OsIRT1, OsIRT2, OsVIT1, OsYSL2, OsYSL15 and OsYSL18 in 21 different plant species. The study revealed that, a total of 51 putative Fe transporter proteins homologues were identified which could be characterized with 555 to 770 amino acids sequence length with 61 to 84.8 kDa molecular weight and pI of 8.2 to 9.4 having basic nature with 9 to 15 TMD with an average of 13 TMDs. The sub-cellular localization of putative Fe transporters was predicted as the plasma membrane and all the identified Fe homologues contained oligopeptide transporter (OPT) domain proteins which play a significant role in Fe transport and homeostasis. Further, the analysis led to identification of highly conserved residues in the putative Fe sequences which could be used as potential motif signatures in identification of new Fe transporter. The interactome analysis for oryza sativa OsYSL15 transporter showed putative

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Interaction with NAS2, NAS1, DMAS1, OsJ_32857, IRT1, IRO2, IDEF1, IRT2, IDI2 which are found to be directly involved in Fe transport from roots to grain. This study, elucidates the valuable theoretical knowledge about the Fe genes, protein features and assist in molecular manipulation of Fe transporters in various plants for developing high Fe in economical part.

**Keywords:** OsYSL2; OsYSL15 and OsYSL18; transmembrane domains; iron (fe) transporter; oligopeptide transporter; motifs.

**1. INTRODUCTION**

Iron is an essential micronutrient for plants, which plays a significant role in important processes like photosynthesis, respiration and chlorophyll biosynthesis, nitrogen fixation and also a component in Fe-binding sites [1]. Despite of soil with large depositions of Fe, plants cannot utilize it because most of Fe is present in insoluble Fe3+ form. Therefore, iron uptake by the plants requires chelation of Fe3+ or rhizosphere acidification to reduce it to Fe2+ form easy uptaken [2]. On the other side, Fe readily accepts and donates electrons making it essential for plants at the same time, producing toxic reactive oxygen species [3]. Therefore, plants have developed efficient mechanisms for Fe uptake from root to shoot and transfer to grain [4,5,6].

The various mechanisms developed for Fe acquisition in plant species have been characterized into two categories: Non-graminaceous plants includes strategy I and graminaceous plants consist of Strategy II [7]. In non-graminaceous plants, ferric chelates are reduced at the root surface by Ferric Chelate Reductase (FRO) gene which cause the insoluble form of Fe3+ to Fe2+ on the surface of root cells, and reduced ions are uptaken by the plasma membrane of root [5]. In graminaceous plants, iron uptake involves biosynthesis and secretion of mugineic acid family phytosiderophores (MAs), which solubilize soil Fe. The phytosiderophores have high affinity for Fe3+ which bind efficiently and transport Fe to the interior of root cells. The MAs are synthesized from S-adenosyl-L-methionine (SAM) pathway by the involvement of three consecutive enzymatic reactions of nicotianamine aminotransferase (NAAT), nicotianamine synthase (NAS), and deoxymugineic acid synthase (DMAS) enzymes [8,9,10]. The synthesized MAs are released into the rhizosphere which solubilize Fe3+ and Fe2+-MA complexes. Then, iron is taken up into root cells through the Yellow Stripe 1 (YS1) and Yellow Stripe 1-like (YSL) transporters [11,12]. Interestingly, rice uptakes Fe by ferrous transporter (OsIRT1) as in non-graminaceous plants and Fe (III)-DMA uptake by OsYSL15 transporter as in graminaceous plants [11].

Meanwhile, the OsYSL2 gene encodes nicotianamine transporter which is responsible for the phloem transport of Fe and manganese in the Fe (II)-NA and Mn(II)-NA and also responsible for long-distance transport into sink tissues [13,14]. The constitutive expression of OsYSL2 gene in rice under the control of a sucrose transporter (OsSUT1) had yielded polished seed with higher concentrations of Fe. This shows the importance of OsYSL2 gene in biofortification of rice crop. Likewise, another Fe (III)-DMA transporter gene, OsYSL18 which is found to be expressed in pollen and pollen tubes suggesting a role in phloem Fe transport. It was found that the YSL family belongs to oligopeptide transporters (OPTs) family [15].

The Oligopeptide Transporter (OPT) family was highly conserved among both prokaryotes and eukaryotes. The OPT family has two clades, the peptide transporters and the yellow stripe-type iron-complex transporters (YS) [15,16]. Peptide transporter homologues primarily transport oligopeptides and glutathione derivatives. In A. thaliana, glutathione influence the expression of iron uptake and transport related genes when grown under iron deficiency conditions. This study also confirmed that over-expression of glutathione enhances the tolerance to Fe deficiency. These OPTs were mainly found in plasma membrane that import substrates from the apoplasm and the external environment. They also play a important role in plant growth and development [15]. OPTs were predicted to have several signature motifs with 12 transmembrane domains.

Other than OPTs, iron-regulated transporter (IRT) like protein (ZIP), zinc regulated transporter (ZRT) and natural resistance-associated macrophage protein (NRAMP) family were also involved in metal transportation. ZIP family transports Fe3+, Zn2+, Mn2+, Cd2+, Ni2+, and Co2+ [17]. Similarly, NRAMP localize in intracellular
vesicle or the plasma membrane which transports divalent metals. Likewise, IRT transporters generally localize to the plasma membrane [18,19].

From this, Fe transporters involve broad range of metal transporters with different family domains functioning in different ways to maintain the Fe homeostasis in plants. Thus, identification of new putative Fe transporters in different plant species and analyzing the molecular basis and topological information is carried out. To accomplish this, few functionally characterized Fe transporters Viz., OsIRT1 (Q75HB1), OsIRT2 (Q6L8G1), OsVIT1 (Q6MWE5), OsYSL2 (Q6H3Z6), OsYSL15 (Q6H3Z3) and OsYSL18 (Q941V3) were used for identification putative transporter in 21 plants species using sequence homology. The newly identified putative Fe transporters will be analyzed for conserved motif sequences, physicochemical properties, localization and transmembrane (TM) topologies and interaction partners. The results of this study will be a theoretical reference for understanding of structural and biological role of Fe transporters in various plants species, and this will pave the way for better understanding iron trafficking from source to sink in different plants.

2. MATERIALS AND METHODS

2.1 Retrieval of Fe Transporter Genes and Proteins

Seven Oryza sativa Fe transporter protein sequences such as OsIRT1 (Q75HB1), OsIRT2 (Q6L8G1), OsVIT1 (Q6MWE5), OsYSL2 (Q6H3Z6), OsYSL15 (Q6H3Z3) and OsYSL18 (Q941V3) were obtained from UniProtKB/Swiss-Prot database of NCBI (ncbi.nlm.nih.gov/). UniProtKB/Swiss-Prot database harbors the non-redundant, manually curated high-quality protein sequences [20]. These obtained sequences were used as queries in proteome datasets of 21 plant species including, Ananas comosus, Arabidopsis thaliana, Brassica oleracea, Capsella grandiflora, Citrus clementina, Eucalyptus grandis, Carica papaya, Brachypodium distachyon, Brasica rapa, Glycine max, Gossypium raimondii, Medicago truncatula, Manihot esculenta, Daucus carota, Linum usitatissimum, Phaseolus vulgaris, Populus trichocarpa, Setaria italica, Solanum lycopersicum, Sorghum bicolor and Zea mays in Phytozome database (phytozome.jgi.doe.gov/pz/portal.html) [21]. The homologous with E value equal to 0 was selected for further analysis. Later, redundant sequences were manually removed and a set 51 putative Fe transporter sequences were retrieved for subsequent analysis.

2.2 Sequence Analysis of Fe Transporter Proteins

Physico-chemical features of putative Fe transporter sequences were analyzed by ProtParam tool (http://web.expasy.org/prot-param/) [22]. Sub-cellular localizations were predicted by CELLO server (http://cello.life.nctu.edu.tw/) [23]. Protein domain families were characterised in NCBI-CD search [24]. Transmembrane domains (TMDs) were predicted by TMHMM Server v. 2.0 (http://www.cbs.dtu.dk/services/TMHMM/) [25]. Motif analysis was performed using MEMEtool (http://meme-suite.org/tools/meme) [26] with following parameters; maximum number of motifs to find, 5; minimum width of motif, 6 and maximum width of motif, 50. The interactome network was generated by Cytoscape [27] using STRING data (http://string-db.org/) [28].

3. RESULTS AND DISCUSSION

3.1 Retrieval of Fe Transporter Genes and Proteins

The functionally characterized Oryza sativa Fe transporter protein such as OsIRT1 (Q75HB1), OsIRT2 (Q6L8G1), OsVIT1 (Q6MWE5), OsYSL2 (Q6H3Z6), OsYSL15 (Q6H3Z3) and OsYSL18 (Q941V3) were used as query sequence in protein dataset of Phytozome database to obtain the corresponding homologues in selected 21 plant species. In order to retrieve the very close homologues, the BLAST-p function with the E value equal to zero was used as selection criteria. None of the homologues for Fe transporters OsIRT1 and OsIRT2 and OsVIT1 had E-value equal to 0, which were discarded from further analysis. The homology search for OsYSL2, OsYSL 15 and OsYSL 18 showed presence of 54 unique putative Fe transporter among the 21 species. These included 3 genes from Ananas comosus, 1 gene from Arabidopsis thaliana, 1 gene from Brassica oleracea, 1 gene from Capsella grandiflora, 1 gene from Citrus clementina, 1 gene from Daucus carota, 3 genes from Eucalyptus grandis, 2 genes from Carica papaya, 10 genes from Brachypodium distachyon, 2 genes from Brasica rapa, 2 genes from Glycine max, 2 genes from Gossypium raimondii, 2 genes from Medicago truncatula, 3 genes from Manihot esculenta, 3 genes from
Fe transporter homologues were predicted as pI < 7 are acidic. The subcellular localizations of compounds with pI > 7 are basic, and those with basic characteristic with a pI > 8. The All the new identified protein demonstrated a reliability of the newly identified Fe transporters. also OPT g reported features of YSL family members and results were in accordance with the previously mentioned topological features and physicochemical properties of the newly identified putative Fe transporter are in accordance with the general features of Fe transporters mentioned in literatures. Overall, we may report that in feature studies regarding the identification of Fe transporters in more plant species, OPT transporter family (cl14607) domain, 9 to 15 TMDs and basic nature of Fe transporters could be used in their characterization. Hence, overall the bioinformatic tools can be used to find the Fe transporter homologues in different plant genomes. Even then, wet-lab studies are needed to confirm the Fe transport activities of these identified proteins.

3.2 Sequence Analysis of Fe Transporter Genes and Proteins

A total of 54 Fe transporter homologues were identified in proteome datasets of 21 plant species by homology search. Based on the physio-chemical analysis of 54 proteins, 3 proteins viz., Aco007552.1, Bradi3g01497.1.p and Brara.C03653.1.p were found to be unstable based on instability index which more than 40 which were excluded from further analysis. All identified Fe homologues contained oligopeptide transporter (OPT) domain proteins (Fig.1). Plant Oligopeptide Transporters (OPTs) comprise a small gene family (PF03169) which plays an important role in the mobilization of small peptides and secondary amino acids that can compound with metals. They were also reported to transport Fe in arabidopsis and oryza. In the present study, the putative Fe transporters contains 555 to 770 amino acids with 61 to 84.8 kDa molecular weight and pl of 8.2 to 9.4 with 9-15 transmembrane domain with an average of 13 TMDs (Table.1). The topological variations in TMDs may have arisen due to broad range activities of these family members. Earlier studies found that, OsYSL15 gene encodes protein of 672 amino acids and 16 transmembrane domains (TMs) with the N terminus located in the cytoplasm [11]. In another study, the analysis of the nine YSL family members using TMAP showed presence of 15 transmembrane domains [29,30]. The bioinformatic analysis of OsOPT proteins showed 12-16 transmembrane helices with molecular weights that ranging from 50 to 90 KDa. In another study, OPT gene analogues in Vitis and Populus ranged from 372 to 760 amino acids in length, with predicted pl ranging from 5.45 to 9.44 and polypeptides had 8-16 transmembrane helices and localized in plasma membrane. Our results were in accordance with the previously reported features of YSL family members and also OPT gene family which predicts the reliability of the newly identified Fe transporters.

All the new identified protein demonstrated a basic characteristic with a pl > 8. The compounds with pl > 7 are basic, and those with pl < 7 are acidic. The subcellular localizations of Fe transporter homologues were predicted as plasma membrane. The results were in accordance with the previous functionally characterized OsYSL15 transporters proteins which were found to be localized in Plasma membrane [31,32]. Therefore, we state that topological features and physicochemical properties of the newly identified putative Fe transporter are in accordance with the general features of Fe transporters mentioned in literatures. Overall, we may report that in feature studies regarding the identification of Fe transporters in more plant species, OPT transporter family (cl14607) domain, 9 to 15 TMDs and basic nature of Fe transporters could be used in their characterization. Hence, overall the bioinformatic tools can be used to find the Fe transporter homologues in different plant genomes. Even then, wet-lab studies are needed to confirm the Fe transport activities of these identified proteins.

3.3 Conserved Motif Analysis

We have searched the most conserved five motif sequences in Fe transporter proteins using MEME tool (Fig.2). Motif 1(VSFVGLFSVPLRKIMIIDYKLYPSGTATAHLI NSFHTPQGAKLKKQ) motifs 2 (FWFYTAGDGCGFSQPFPTFGLKAYENRFYFD FSATYYVGVMICPHVINIS) and Motif 3 (CGVMNNIVSTASDLQMDKTFGYLTILTSPRM FVSQVGTAMGCVISPVCV) were 50 amino acid residues long and shared by all the plant species. While, motif 4 (AYIFAPVLANAYCGLTDWMASNYGKLAFTIG) and Motif 5 (YDDQRTTEVFLKDOITWFAAYQVYVAIAAISTI TIPHMFHQ) were 36 and 41 amino acid sequences respectively. These motifs were also shared by all the plant species. The conserved domain analysis using NCBI CD search showed that all the consensus sequences belongs to OPT protein family (Fig.2). Overall, these sequences are highly conserved in all analyzed species; therefore, they could be used in characterization of Fe transporters in different plant species.

3.4 Predicted Interaction Partner Analysis

The protein functions in a can be known by Protein protein interactions (PPIs). The function of newly identified proteins can be predicted on the evidence of their interaction with a protein, whose function is already revealed. It was proved that more than 80% of proteins form complex to yield biological function to the proteins [33].
Moreover, detailed study of PPIs will help in modeling of functional pathways to understand the molecular mechanisms of cellular processes. With this view, to further characterize the potential interaction partners of the putative Fe transporters, we did interactome analysis using STRING server. Protein sequence from *A. thaliana* (AT1G65730.1) (Fig. 3A) and *O. sativa* (OsYSL15) (Fig. 3B) was used as representatives of dicots and monocots, respectively for interactome study. The analysis showed that ten putative interactors viz., OPT6, OPT1, OPT3, TOC34, TOC33, OPT8, OPT5, OPT9, OPT4 and NRMP3 proteins with medium to high confidence score were predicted for *A. thaliana* AT1G65730.1 protein. The AtOPT3, a member of the OPT family was found to be involved in Fe transport in arbidopsis and it is mainly up regulated under Fe deficiency conditions [34]. Further, opt3 knock down mutant exhibit a defect in Fe loading in seeds, showing it as a candidate for Fe biofortification [34]. With this, AtOPT3 plays a critical role in iron metabolism and accumulation in seeds [34]. Another gene, OsOPT7 whose expression is 60-fold higher in shoots and roots of plants when grown under iron deficiency [35]. These results indicate that OsOPT7 also plays an important role in Fe transport in rice. Similarly, OsOPT3, OsOPT4, OsOPT5, and OsOPT7 were found to transport ferrous and/or ferric iron chelated to nicotianamine which helps in Fe transport and homeostasis [36]. The functionally characterized gene, AtNRAMP3 expression is up regulated by iron deficiency and mediates in long distance transport The loss of function of AtNRAMP3 gene led to increased accumulation of manganese and zinc in the roots, indicating its role in multiple ion transport. AtNRAMP4 and AtNRAMP5 transporters are responsible accumulation of Fe in cytosol during germination [18]. OPTs are most likely involved in Glutathione (GSH) transport and play direct as well as indirect roles in many GSH mediated processes, metal homeostasis [15]. Glutathione plays key role in mitigating biotic and abiotic stress by mediating redox potential of the cell. This GSH preserve cell redox homeostasis and improve internal iron availability. The present analysis revealed that all the proteins were predicted to interact with AT1G65730.1 protein directly or indirectly for iron transport and accumulation.

In *O. sativa*, NAS2, NAS1, DMAS1, OsJ_32857, IRT1, IRO2, IDEF1, IRT2, IDI2, OS04To444800-02 were found to be putative interaction partners. Under Fe deficiency situations, mediates uptake of Co, Cd, Mn and Zn. The IRT2 transporter which is highly similar to IRT1 and mainly capable of Fe and Zn transport in roots of *A. thaliana* under Fe-limited conditions. Similar to IRT1, the IRT2 also involves in transportation of Zn, Mn and Cd. Phytosiderophores are formed by the trimerization of S-adenosylmethionine to form one molecule of nicotianamine (NA) by Nicotianamine synthase (NAS). NA is a biosynthetic precursor of phytosiderophores and is thus a crucial component for iron (Fe) acquisition. Overexpression of HvNAS1 has shown to increase Fe and Zn up to three and two fold, respectively, in rice grains [37]. Likewise, OsNAS1, OsNAS2, and OsNAS3 were differentially regulated by Fe level and found to be involved in long distance transport of Fe. The knock-down of OsDMAS1 significantly up-regulates the genes involved in Fe uptake and homeostasis.

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**Fig. 1.** The picture showing identification of OPT domain in GRMZM2G026391 protein sequence
Fig. 2. Sequence logo of the five most conserved motifs of Fe transporter proteins in 21 plant species

Fig. 3. Interaction partner analysis of Fe transporters of A. thaliana (AT1G65730.1) (A) and O. sativa (OsYSL15) (B) which were selected as representatives of dicots and monocots respectively. 10 potential interactors of both species were visualized. Stronger associations are represented by thicker lines.

The gene OsIRO2, performs a crucial function of Fe uptake from soil and Fe translocation to grain during seed maturation [38]. OsIRO2 gene is induced under Fe starvation and is positively regulated by IDE1. OsIRO2 also positively regulates various genes related to Strategy II, IDE1 and IDE2 [39]. The two rice transcription factors IDE1 (IDE-binding factor 1) and IDE2, which bind specifically to IDE1 and IDE2, respectively which regulate iron homeostasis related genes in rice during Fe deficiency.
## Table 1. List of putative Fe transporters in 21 plant species and their gene and protein features

| Sl. no | Gene ID (Phytozome) | Conserved protein domain family | Sub cellular localization | Exon number | Protein length | MW (kDa) | pI | Instability index | TMD |
|--------|---------------------|---------------------------------|---------------------------|-------------|----------------|---------|----|------------------|-----|
| 1      | Aco010337.1         | OPT                             | Plasma Membrane           | 6           | 711            | 77.5    | 8.49 | 39.05            | 14  |
| 2      | Aco010643.1         | OPT                             | Plasma Membrane           | 7           | 682            | 74.6    | 8.75 | 36.06            | 15  |
| 3      | AT1G65730.1         | OPT                             | Plasma Membrane           | 4           | 688            | 75.7    | 9.17 | 33.7             | 14  |
| 4      | Bol022433           | OPT                             | Plasma Membrane           | 6           | 666            | 73.3    | 9.13 | 29.8             | 14  |
| 5      | Bradi2g53950.2.p    | OPT                             | Plasma Membrane           | 13          | 679            | 73.7    | 9    | 31               | 12  |
| 6      | Bradi3g01520.1.p    | OPT                             | Plasma Membrane           | 2           | 676            | 74.3    | 9    | 35               | 12  |
| 7      | Bradi3g50260.1.p    | OPT                             | Plasma Membrane           | 7           | 676            | 73.7    | 9.26 | 31.9             | 12  |
| 8      | Bradi3g50263.1.p    | OPT                             | Plasma Membrane           | 7           | 682            | 74.9    | 9    | 35.8             | 14  |
| 9      | Bradi3g50267.2.p    | OPT                             | Plasma Membrane           | 7           | 770            | 84.8    | 9.4  | 35               | 14  |
| 10     | Bradi5g16190.1.p    | OPT                             | Plasma Membrane           | 6           | 707            | 76.7    | 8.5  | 34.5             | 12  |
| 11     | Bradi5g17220.2.p    | OPT                             | Plasma Membrane           | 9           | 664            | 72.0    | 8.2  | 31               | 13  |
| 12     | Bradi5g17230.2.p    | OPT                             | Plasma Membrane           | 23          | 704            | 76.3    | 9.2  | 31.8             | 14  |
| 13     | Bradi5g25987.1.p    | OPT                             | Plasma Membrane           | 6           | 687            | 74.9    | 8.77 | 33.9             | 14  |
| 14     | Brara.F02656.1.p    | OPT                             | Plasma Membrane           | 6           | 666            | 73.2    | 9    | 29.4             | 14  |
| 15     | Cagra.2460s0005.1.p | OPT                             | Plasma Membrane           | 8           | 672            | 73.6    | 9    | 33               | 14  |
| 16     | Ciclev10030844m     | OPT                             | Plasma Membrane           | 6           | 704            | 77.5    | 8.89 | 38.9             | 14  |
| 17     | DCAR_030390         | OPT                             | Plasma Membrane           | 7           | 714            | 78.6    | 8.3  | 32.8             | 14  |
| 18     | Eucgr.K02316.2.m    | OPT                             | Plasma Membrane           | 6           | 707            | 78.4    | 9    | 32.5             | 14  |
| 19     | Eucgr.K02318.1.p    | OPT                             | Plasma Membrane           | 8           | 704            | 76.3    | 9.2  | 31.8             | 14  |
| 20     | Eucgr.K02319.1.p    | OPT                             | Plasma Membrane           | 13          | 714            | 78.2    | 9    | 33.6             | 12  |
| 21     | evm.model.supercontig_108.9 | OPT                               | Plasma Membrane           | 6           | 693            | 76.0    | 8.9  | 31               | 14  |
| 22     | evm.model.supercontig_3.127 | OPT                               | Plasma Membrane           | 7           | 678            | 74.4    | 9.2  | 30.1             | 12  |
| 23     | Glyma.09G164500.1.p | OPT                             | Plasma Membrane           | 6           | 703            | 77.6    | 9    | 29.5             | 14  |
| 24     | Glyma.16G212900.1.p | OPT                             | Plasma Membrane           | 6           | 702            | 77.6    | 8.9  | 29.4             | 14  |
| 25     | Gorai.004G262100.1 | OPT                             | Plasma Membrane           | 6           | 697            | 76.8    | 8.7  | 30.3             | 14  |
| 26     | Gorai.013G070800.2 | OPT                             | Plasma Membrane           | 12          | 559            | 61.4    | 9    | 36.4             | 11  |
| 27     | GRMZM2G004440_P01   | OPT                             | Plasma Membrane           | 6           | 679            | 73.8    | 9    | 33.9             | 13  |
| 28     | GRMZM2G018148_P01   | OPT                             | Plasma Membrane           | 3           | 684            | 75.2    | 8.9  | 37.3             | 12  |
| 29     | GRMZM2G026391_P01   | OPT                             | Plasma Membrane           | 15          | 672            | 72.9    | 9    | 29.6             | 14  |
| No | Accession    | Description | Rank | Size (bp) | GC (%) | Complex | Start (bp) | End (bp) |
|----|--------------|-------------|------|-----------|--------|---------|------------|---------|
| 30 | Lus10013323  | OPT         | Plasma Membrane | 7     | 725     | 79.5   | 8.6       | 39.43   | 12       |
| 31 | Lus10034177  | OPT         | Plasma Membrane | 6     | 717     | 78.8   | 8.72      | 34.5    | 12       |
| 32 | Lus10043408  | OPT         | Plasma Membrane | 6     | 717     | 78.8   | 8.8       | 33.7    | 14       |
| 33 | Manes.04G061100.1.p | OPT     | Plasma Membrane | 6     | 687     | 75.4   | 8.8       | 32.4    | 12       |
| 34 | Manes.05G182000.1.p | OPT     | Plasma Membrane | 6     | 714     | 79.3   | 8.5       | 32.4    | 12       |
| 35 | Manes.05G192800.1.p | OPT     | Plasma Membrane | 8     | 664     | 72.7   | 9         | 30.3    | 14       |
| 36 | Medtr3g092090.2 | OPT     | Plasma Membrane | 16    | 664     | 72.8   | 8.7       | 32.1    | 14       |
| 37 | Medtr5g091600.1 | OPT     | Plasma Membrane | 5     | 682     | 74.4   | 8.9       | 31.6    | 12       |
| 38 | Phvul.004G138900.1.p | OPT     | Plasma Membrane | 6     | 701     | 77.2   | 8.84      | 28.5    | 12       |
| 39 | Potri.017G150500.2 | OPT     | Plasma Membrane | 17    | 680     | 74.8   | 8.59      | 30      | 12       |
| 40 | Potri.017G151200.1 | OPT     | Plasma Membrane | 6     | 698     | 76.9   | 8.9       | 30.39   | 12       |
| 41 | Potri.017G151800.1 | OPT     | Plasma Membrane | 6     | 698     | 76.8   | 8.8       | 28.5    | 12       |
| 42 | Seita.1G118000.1.p | OPT     | Plasma Membrane | 6     | 678     | 74.4   | 9.25      | 38.8    | 14       |
| 43 | Seita.1G118200.1.p | OPT     | Plasma Membrane | 2     | 686     | 75.3   | 8.95      | 36.5    | 12       |
| 44 | Seita.1G254100.1.p | OPT     | Plasma Membrane | 7     | 687     | 74.4   | 8.9       | 31.6    | 14       |
| 45 | Seita.1G254200.1.p | OPT     | Plasma Membrane | 7     | 623     | 67.3   | 8.8       | 36.4    | 13       |
| 46 | Sobic.003G343300.1.p | OPT   | Plasma Membrane | 6     | 680     | 74.1   | 9.27      | 33      | 12       |
| 47 | Sobic.004G011800.1.p | OPT   | Plasma Membrane | 4     | 685     | 75.7   | 8.7       | 38.7    | 15       |
| 48 | Sobic.004G011900.1.p | OPT   | Plasma Membrane | 4     | 684     | 75.0   | 8.9       | 37.6    | 11       |
| 49 | Sobic.006G164300.2.p | OPT   | Plasma Membrane | 15    | 677     | 73.6   | 9.1       | 27.76   | 14       |
| 50 | Solyc03g031920.2.1 | OPT    | Plasma Membrane | 6     | 696     | 76.8   | 9.25      | 32.2    | 14       |
| 51 | Solyc02g081570.2.1 | OPT    | Plasma Membrane | 6     | 694     | 76.2   | 8.9       | 36.1    | 12       |
4. CONCLUSION

The present study is focused on identification of homologues of OsYSL2, OsYSL15 and OsYSL18 Fe transporters in 21 different plant species. The study found a total of 51 putative Fe transporter proteins which could be characterized with 555 to 770 amino acids sequence length with 61 to 84.8 kDa molecular weight and pI of 8.2 to 9.4 having basic nature with 9 to 15 TMD with an average of 13 TMDs having oligopeptide transporter (OPT) domain and localized in plasma membrane. Therefore, Identification of putative Fe transporter homologues in various plant species and their analysis at different structural levels has led us to better understand the Fe transporters. Hence, the results of this study will become essential for further elucidation of Fe transporters in different plants, as well as they will make significant contributions to the future studies for developing genetically engineered and biofortified plants.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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