Since January 2020 Elsevier has created a COVID-19 resource centre with free information in English and Mandarin on the novel coronavirus COVID-19. The COVID-19 resource centre is hosted on Elsevier Connect, the company's public news and information website.

Elsevier hereby grants permission to make all its COVID-19-related research that is available on the COVID-19 resource centre - including this research content - immediately available in PubMed Central and other publicly funded repositories, such as the WHO COVID database with rights for unrestricted research re-use and analyses in any form or by any means with acknowledgement of the original source. These permissions are granted for free by Elsevier for as long as the COVID-19 resource centre remains active.
Developmental and Comparative Immunology 50 (2015) 19–25

Short communication

Characterization of the duplicate L-SIGN and DC-SIGN genes in miiuy croaker and evolutionary analysis of L-SIGN in fishes

Chang Shu, Shanchen Wang, Tianjun Xu *

Laboratory of Fish Biogenetics & Immune Evolution, College of Marine Science, Zhejiang Ocean University, Zhoushan 316022, China

A R T I C L E   I N F O

Article history:
Received 8 November 2014
Revised 6 January 2015
Accepted 9 January 2015
Available online 13 January 2015

Keywords:
Miiuy croaker
DC-SIGN
L-SIGN
Gene synteny
Evolutionary analysis
Expression patterns

A B S T R A C T

Dendritic cell-specific ICAM-3-grabbing non-integrin (DC-SIGN/CD209) and liver/lymph node-specific ICAM-grabbing non-integrin (L-SIGN/CD299) which are homologues of DC-SIGN are important members in C-type lectin receptors family as key molecules to recognize and eliminate pathogens in the innate immune system. DC-SIGN and L-SIGN have become hot topics in recent studies which both served as cell adhesion and phagocytic pathogen recognition receptors in mammals. However, there have been almost no studies of DC-SIGN and L-SIGN structure and characters in fish, only DC-SIGN in the zebrafish had been studied. In our study, we identified and characterized the full-length miiuy croaker (Miiuythys miiuy) DC-SIGN (mmDC-SIGN) and L-SIGN (mmL-SIGN) genes. The sequence analysis results showed that mmDC-SIGN and mmL-SIGN have the same domains with other vertebrates except primates, and share some conserved motifs in CRD among all the vertebrates which play a crucial role in interacting with Ca2+ and for recognizing mannose-containing motifs. Gene synteny of DC-SIGN and L-SIGN were analyzed for the first time and gene synteny of L-SIGN was conserved among the five fishes. Interestingly, one gene next to L-SIGN from gene synteny had high similarity with L-SIGN gene that was described as L-SIGN-like in fish species. While only one L-SIGN gene existed in other vertebrates, two L-SIGN in fish may be in consequence of the fish-specific genome duplication to adapt the specific environment. The evolutionary analysis showed that the ancestral lineages of L-SIGN gene in fishes experienced purifying selection and the current lineages of L-SIGN gene in fishes underwent positive selection, indicating that the ancestral lineages and current lineages of L-SIGN gene in fishes underwent different evolutionary patterns. Both mmDC-SIGN and mmL-SIGN were expressed in all tested tissues and ubiquitously up-regulated in infected liver, spleen and kidney at different sampling time points, indicating that the mmDC-SIGN and mmL-SIGN participated in the immune response to defense against bacteria infection.

© 2015 Elsevier Ltd. All rights reserved.

1. Introduction

Innate immunity acts in the first line of immune defense mechanisms, having an important function against foreign pathogens in vertebrates and invertebrates. In teleosts, the innate immune system plays a critical role in protecting organism against invading pathogens (Aoki et al., 2013). Pattern recognition receptors (PRRs) including Toll-like receptors (TLRs), C-type lectin receptors (CLRs), NOD-like receptors (NLRs) and RIG-I like receptors (RLRs) on antigen-presenting cells play a significant role in innate immunity which recognize pathogen associated molecular patterns (PAMPs) (Zhu et al., 2013a). C-type lectin receptors (CLRs) characterized by the presence of one or more homologous carbohydrate-recognition domain (CRD) are a large family of Ca2+-dependent carbohydrate binding proteins which can specifically recognize a number of PAMPs including bacteria, viruses, parasites and fungi (Lepenies et al., 2013). The CLRs including the collectins, selectins, lymphocyte lectins and proteoglycans were initially divided into two types based on the quantity of conserved CRDs: Type I CLRs such as the macrophage-mannose receptor, DEC205 and selectins have multiple CRDs at their NH2 terminus; and type II CLRs such as hepatic asialoglycoprotein receptors, macrophage lectin, DC-specific ICAM3-grabbing non-integrin (DC-SIGN), Langerin, DC-associated C-type lectin (Dectin-1) and DC immunoreceptor (DCIR) have a single CRD at the COOH-terminus (Redelinghuys and Brown, 2011). Dendritic cell-specific intercellular adhesion molecule 3 grabbing non-integrin, DC-SIGN (CD209) and its homolog, liver/lymphnode-specific intercellular adhesion molecule 3 grabbing non-integrin, L-SIGN also known as DC-SIGN-related receptor (DC-SIGNR), are two kinds of the type II CLRs sharing similar structure and both serve as cell adhesion and phagocytic pathogen recognition receptors (Khoo et al., 2008). Characters and functions of DC-SIGN and L-SIGN in human had been studied, that both can bind various viruses such as HIV-1, Ebola virus, cytomegalovirus, hepatitis C virus, Dengue virus, and SARS-coronavirus

* Corresponding author. Laboratory of Fish Biogenetics & Immune Evolution, College of Marine Science, Zhejiang Ocean University, Zhoushan 316022, China. Tel.: +86 580 2550826; fax: +86-580-2550826.
E-mail address: tianjunxu@163.com (T. Xu).

http://dx.doi.org/10.1016/j.dci.2015.01.004
0145-305X/© 2015 Elsevier Ltd. All rights reserved.
Differences are also seen in their tissue distribution that DC-SIGN is expressed on dendritic cells and some types of macrophages, while L-SIGN is expressed on sinusoidal endothelial cells in the liver, lymph node sinusoids, and in the placenta (Boilly-Larouche et al., 2007). Human DC-SIGN and L-SIGN structures are composed of 3 domains: a transmembrane domain, CRD and a neck-region which is highly conserved. The two genes were coded by 404 amino acids and 376 amino acids separately sharing 77% identical at the amino acid level (Khoo et al., 2008; Li et al., 2013). The dileucine motif, the tri-acidic cluster and an EPN sequence motif are conserved in both human DC-SIGN and L-SIGN (Khoo et al., 2008). But the structures, genomic organizations and functional motifs in teleost are not very clear. Until now, there have been almost no studies of DC-SIGN and L-SIGN structure and characters in teleost; only DC-SIGN in the zebrafish had been studied which showed that DC-SIGN in zebrafish has conserved domains with other vertebrates and plays a crucial role for the initiation and development of adaptive immunity in zebrafish (Lin et al., 2009).

In this study, we analyzed genomic organizations, gene structures, and synteny, evolutionary process and expression of miiuy croaker DC-SIGN (mmDC-SIGN) and L-SIGN (mmL-SIGN). We are the first to comprehensively analyze L-SIGN in fish and gene synteny on genomic level of DC-SIGN and L-SIGN in teleost.

2. Materials and methods

2.1. Database mining

To obtain the DC-SIGN and L-SIGN from miiuy croaker, the BLASTP and TBLASTN programs with a query set of the previously characterized DC-SIGN and L-SIGN were used to search the miiuy croaker transcriptome (Che et al., 2014) and whole genome database (unpublished data). Two corresponding scaffolds were identified which have a strong identity with sequences previously accepted as DC-SIGN and L-SIGN respectively. Miiuy croaker DC-SIGN and L-SIGN have been identified and the nucleotide sequences have been deposited in GenBank database under the accession numbers KM884829 and KM884828.

2.2. Sequence and phylogenetic analysis

The ORF of mmDC-SIGN and mmL-SIGN were predicted using the Open Reading Frame Finder (http://www.ncbi.nlm.nih.gov/gorf/gorf.html) and GENSCAN (Burge and Karlin, 1997). The potential protein domains were characterized by the SMART program (Letunic et al., 2006). All the other species DC-SIGN and L-SIGN cDNA sequences were obtained from the GenBank (http://www.ncbi.nlm.nih.gov/Genbank/) and Ensemble Genome Browser (http://www.ensembl.org/) databases (Supplementary Table S3). MEGA 5 software (Tamura et al., 2011) and DNAman were used for the multiple alignment and DNAstar was used to gain the divergence and percent identity values of all the known DC-SIGN and L-SIGN amino acid sequences (Thompson et al., 1994). A phylogenetic tree of several kinds CLR (CD23, CLEC4G, CD207, ASGR1, CD205, DC-SIGN, L-SIGN and L-SIGN-like) was constructed by the neighbor-joining (NJ) method with bootstrapped 1000 times using the Poisson model in the MEGAS5 program. While another phylogenetic tree of all the known L-SIGN sequences was established via the Bayesian approach using MrBayes v3.2 which is running 5,000,000 generation with 25% of trees burned (Ronquist and Huelsenbeck, 2003) and the resulting tree was visualized and edited by TreeView (Page, 1996). The neighboring genes of mmDC-SIGN and mmL-SIGN were confirmed in genomic regions by BLAST programs and the neighboring genes locations of L-SIGN in other fishes were confirmed by Genomics (Muffato et al., 2010).

2.3. Molecular evolutionary analysis

The nonsynonymous and synonymous rate ratio $\omega (d_{NS}/d_{SN})$ stands for the change of selective pressures. In short, $\omega = 1$, $< 1$, and $> 1$ were indicative of neutral evolution, purifying selection and positive selection. To test the evolution of L-SIGN gene in fishes, the CODEML of PAML software (Yang, 2007) and the Hyphy package of Data Monkey Web Server (Pond and Frost, 2005) were used to estimate the ratio of $\omega$. In both CODEML and the Data Monkey Web Server, several models were carried out as described by Zhu et al. (2013b).

2.4. Challenge experiments and RNA extraction

Healthy miiuy croakers individuals were obtained from Zoushan Fisheries Research Institute (Zhejiang, China), temporarily cultured in the seawater tank of 20 °C to adapt to the ambient and evaluate of the fish health under a natural photoperiod for two weeks. After acclimatizing, ten healthy tissues (liver, spleen, kidney, intestines, heart, muscle, gill, brain, eye, and fin) were obtained from the uninfected control fishes and kept at ~80 °C. The challenge experiments of miiuy croaker with a common bacterial pathogen, Vibrio anguillarum, were performed as previously described (Xu et al., 2014). Fish samples were randomly divided into two groups, injection and control groups. In injection group, fishes were injected with 1 ml suspension ($3.0 \times 10^7$ CFU/ml). At 6 h, 12 h, 24 h, 36 h, 48 h and 72 h post-injection, fish samples were killed and three tissues (liver, kidney and spleen) of infection were removed and stored at ~80 °C. Total RNA was extracted from the various tissues samples by Trizol reagent (Qiagen) following the manufacturer’s instructions and cDNA was synthesized using QuantScript RT Kit (TIANGEN) according to the manufacturer’s protocol which stored at ~20 °C for further experiment.

2.5. Expression analysis of DC-SIGN and L-SIGN

Three pairs of primers (DC-SIGN-RT-F/R, L-SIGN-RT-F/R,β-actin-RT-F/R) were designed to study the expression level of mmDC-SIGN and mmL-SIGN (Supplementary Table S1). Real time quantitative PCR(qRT-PCR) was used to test DC-SIGN and L-SIGN genes expression level in ten uninfected tissues (liver, spleen, kidney, intestines, heart, muscle, eye, brain, fin, gill) and in three infected tissues (liver, spleen and kidney) The RT-PCR was executed on a 7500 Real time PCR system (Applied Biosystems, USA) and every expression was performed in three independent replicates in the same condition. The conditions of the RT-PCR was carried out as described by Xu et al. (2014). Significance test of the expression level was analyzed by a Duncan test. Expression differences were considered at a significance level when $p < 0.05$ and all data were expressed as the mean ± SE (standard error).

3. Results and discussion

3.1. Structures and characteristics of mmDC-SIGN and mmL-SIGN genes

The full length mmDC-SIGN and mmL-SIGN genes were successfully obtained. The cDNA sequence of mmDC-SIGN which had 5 exons and 4 introns was 766 bp containing a 5'-UTR of 33 bp, a 3'-UTR of 94 bp and an ORF of 639 bp coding 212 amino acids (Fig. I A). The cDNA sequence of mmL-SIGN which had 6 exons and 5 introns was 1617 bp containing a 5'-UTR of 44 bp, a 3'-UTR of 628 bp with polyadenylation signal (AATAAA) at 172 bp
Fig. 1. Nucleotide sequence and deduced amino acid sequence of the miuy croaker DC-SIGN (A) and L-SIGN (B). The noncoding region was lowercase. Several motifs were in blue box and conserved cysteine-rich motifs were in green box. CRD was shown as underscore. The polyadenylation signal motif (AATAAA) was boxed in black (B). Schematic overview of domain structures comparison of DC-SIGN (C) and L-SIGN (D) from higher vertebrates to lower vertebrates. (E) Sequence alignment of L-SIGN and L-SIGN-like from several fishes was carried out by DNAman. Absolutely conserved amino acid is colored black; the comparatively conserved amino acid is colored pink and blue. CRD was shown as underscore and EPN, WND motifs were marked by black triangle. (F) Genomic synteny of DC-SIGN and (G) L-SIGN among several fishes. The same color represented the same genes and the arrows indicated the transcriptional direction. The CLR members were in black box.
downstream of the translation termination codon and an ORF of 945 bp coding 314 amino acids (Fig. 1B). Introns in both mmDC-SIGN and mmL-SIGN appeared the pattern of GT/AG known by comparison with other vertebrates DC-SIGN and L-SIGN sequences separately, while comparison of the deduced amino acid sequences showed that mmDC-SIGN shared 50.8% to 96.4% identity with DC-SIGN from other species and mmL-SIGN shared 46.9% to 77.3% identity with other species separately (Supplementary Table S2). Both mmDC-SIGN and mmL-SIGN amino acid sequences had two domains: a transmembrane region and a CRD differing from primates which had specific internal repeat domain (Fig. 1C, D). In the DC-SIGN CRD domain, EPD motif and WYD motif existed in miu croaker and large yellow croaker instead of EPN motif and WND motif in all other species for determining the carbohydrate binding specificity (Kong et al., 2011); EPN motif, WND motif and four amino acids (Glu269, Asn271, Glu276 and Asn292) existing in CRD domain of mmL-SIGN are well conserved from fish to human. All of the conserved motifs are essential for interacting with Ca²⁺ for recognizing carbohydrate ligand (Khoo et al., 2008). The tri-acidic cluster is conserved in human DC-SIGN (Svajger et al., 2010) and L-SIGN (Koppel et al., 2005), but not in miu croaker. Mm-DC-SIGN and mm-L-SIGN both have four conserved cysteine residues in positions Cys102, Cys196, Cys204 and positions Cys169, Cys180, Cys201, Cys278 to form the internal disulfide bridges revealed by multiple sequence alignment which also existed in other known DC-SIGN and L-SIGN sequences of vertebrate (Huang and Meng, 2009; Lin et al., 2009). Miu croaker DC-SIGN and L-SIGN had the same domains with other fish, amphibian and bird that showed both DC-SIGN and L-SIGN had conserved domains from fish to mammals except primates.

3.2. Gene duplication of L-SIGN gene in fish

Interestingly, when we analyzed genomic synteny of L-SIGN in teleost, one gene next to L-SIGN which has high similarity with L-SIGN gene and clustered with L-SIGN in the phylogenetic tree of CLRs (Fig. 2A) was named as L-SIGN-like gene tentatively. L-SIGN-like gene was obtained from miu croaker, tilapia, medaka,

Fig. 2. (A) A neighbor-joining tree of several kinds CLRs. Numbers at the nodes indicated the bootstrap values. (B) Schematic model proposed for the evolution of L-SIGN members in 2R and 3R. The green arrows represented genome duplication. (C) A phylogenetic tree of L-SIGN genes was constructed using MrBayes with Bayesian method. GenBank accession numbers and Ensembl number of these genes are listed in the Supplementary Table S3.
The phylogenetic tree of members belong to CLR (Fig. 2A) showed that each kind CLR gathered together separately except DC-SIGN, L-SIGN and L-SIGN-like which gathered together that perhaps because of the genes with similarity among species, branches of CD23, CLEC4G, CD207, DC-SIGN, L-SIGN and L-SIGN-like grouped together showing these genes had a closer evolution position and had a relatively further evolution distance to ASGR1 and CD205. In branch of DC-SIGN and L-SIGN, L-SIGN-like sequences grouped with L-SIGN sequences closely which showed L-SIGN-like share high similarity with L-SIGN; all fishes and mammal sequences grouped separately that mmDC-SIGN grouped with large yellow croaker and mmL-SIGN grouped with Zebra Mbuna and tilapia L-SIGN-like.

Another phylogenetic tree of L-SIGN genes constructed by Bayesian approach (Fig. 2C) was used to test the positive selection in ancestral lineages of fishes. Firstly, the value of \( \omega \) was 0.26 in the one-ratio model which assumed that one unique \( \omega \) in all branches of the tree \( p = 0.00 \), Supplementary Table S4), showing that the entire L-SIGN genes underwent the purifying selection. Secondly, the free-ratio model fitted the data significantly better than the one-ratio model \( p = 0.00 \), Supplementary Table S4). Finally, the branch-site model was used to detect whether positive selection sites existed in ancestral lineages of fishes. No positive selection sites were found in ancestral lineages of fishes (Supplementary Table S4). Meanwhile, multiple ML methods were carried out to explore the selective pressures on L-SIGN genes of fishes. For fishes L-SIGN genes, no positive selection sites were detected in M2a and M8 model (Table 1), but other ML methods detected positive selection sites in this group (Table 2). To improve the accuracy of positive selection sites, we considered that the site under positive selection should be detected in at least three of the ML methods. So we found one positive selection site (287, Table 2) on fishes L-SIGN genes. The result showed that the ancestral lineages of fishes underwent purifying selection, but the current lineages of fishes underwent positive selection. The fishes had lived in aquatic environment which was filled with countless types and numerous amounts of pathogens. As the years went by, the aquatic environment had changed greatly and the pathogen significantly differs from before. Due to the change of aquatic environment, the fishes must further evolve to adapt to the environment. So we detected positive selection sites in current fishes of L-SIGN genes.

### 3.3. Genome evolution and phylogenetic analyses

Information from genome was used to analyze genomic synteny of mmDC-SIGN (Fig. 1F) and L-SIGN among several fishes (Fig. 1G) to extend their evolutionary analysis. We confirmed mmDC-SIGN located in scaffold 2 and mmL-SIGN located in scaffold 59 of miyi croaker genome, and also confirmed some genes surrounding mmDC-SIGN and mmL-SIGN to propose a linear framework of DC-SIGN and L-SIGN in teleost (Fig. 1F, G) which showed mmDC-SIGN located between CLEC4E and CD4 and no ZNF557, ZNF358 and LRRK8E genes surrounding the mmDC-SIGN which surrounded DC-SIGN in a chromosome of human and zebrfish separately (Lin et al., 2009). Miyi croaker L-SIGN was located between L-SIGN-like and TTC8, and also differences happened among the linear organization of the five fishes in basic chromosome number and the amount of surrounding genes. Both miyi croaker and tilapia formed a cluster containing five C-type lectin receptors, but only two C-type lectin receptors were clustered together in other three fishes. There was one L-SIGN and one L-SIGN-like in four fishes except platyfish (Fig. 1G). Among these fish genomes, genes from TTC8 to ENTPD5 showed that each kind CLR gathered together separately except DC-SIGN, L-SIGN and L-SIGN-like which gathered together that perhaps because of the genes with similarity among species, branches of CD23, CLEC4G, CD207, DC-SIGN, L-SIGN and L-SIGN-like grouped together showing that these genes had a closer evolution position and had a relatively further evolution distance to ASGR1 and CD205. In branch of DC-SIGN and L-SIGN, L-SIGN-like sequences grouped with L-SIGN sequences closely which showed L-SIGN-like share high similarity with L-SIGN; all fishes and mammal sequences grouped separately that mmDC-SIGN grouped with large yellow croaker and mmL-SIGN grouped with Zebra Mbuna and tilapia L-SIGN-like.

### 3.4. Expression analysis of mmDC-SIGN and mmL-SIGN

In order to explore expression patterns of DC-SIGN and L-SIGN genes, the expression level of mmDC-SIGN and mmL-SIGN genes in different tissues were confirmed by qRT-PCR. We set gill as calibrator to normalize DC-SIGN and L-SIGN expression level in ten
uninfected tissues. The highest expression level of mmDC-SIGN was in liver, muscle and intestines, while the expression in gill and heart was weak. Miiuy croaker L-SIGN have a very high level expression in intestines and spleen and moderate expression in kidney and liver, while the expression level in heart, muscle, eye, gill, fin and brain was much lower than other tested tissues (Fig. 3A). In Vibrio anguillarum challenged groups, the tissue distribution and temporal response levels of mmDC-SIGN and mmL-SIGN at different hours were detected in the infected liver, kidney and spleen. In infected liver group, both DC-SIGN and L-SIGN have a sharp up-regulation in 6 h and floating up and down from 12 h to 36 h, then reaching the peak at 48 h and sharply decreasing in 72 h (Fig. 3B). In spleen, the expression of DC-SIGN was volatile from 6 h to 72 h; the highest expression level and lowest expression level were at 24 h and 12 h. The expression of L-SIGN floats up and down from 6 h to 36 h, then increased gradually and reached its peak at 72 h (Fig. 3C). In the kidney, the expression of DC-SIGN increased and reached its peak at 24 h and the lowest expression level was at 36 h, while the highest expression level and lowest expression level of L-SIGN occurred in 48 h and 72 h, respectively (Fig. 3D). Both the expressions of

| Gene   | PAML-M8 | SLAC | FEL | REL | FUBAR | MEME | Total |
|--------|---------|------|-----|-----|-------|------|-------|
| L-SIGN | None    | None | 12,40,50,51,96,287,405 | 303,341 | 287 | 5,12,40,51,68,72,86,210,277,284,287,303,326,330,333,346,372,403,405 | 287 |

Table 2
Different methods test positive selection on L-SIGN gene in teleosts.

- Codons identified by more than one ML method are underlined.
- Codons with p values <0.1.
- Codons with Bayes factor >50.
- Codons with posterior probability >0.9.

**Fig. 3.** Expression analysis of miiuy croaker DC-SIGN (blue) and L-SIGN (red). Expression profiles in ten tissues (A) and expression in three infected tissues at six time points: liver (B), spleen (C) and kidney (D). Above these data, lowercase letters (a, b, c, d, e, f) implemented by SPSS software (p < 0.05) stand for significant differences at different tissues of DC-SIGN and L-SIGN separately.
mmDC-SIGN and mmL-SIGN were up-regulated in infected liver, spleen and kidney at different sampling time points, and mmL-SIGN had the same trend with mmDC-SIGN in floating up and down in liver and kidney. These results indicated that the mmDC-SIGN and mmL-SIGN participated in the immune response to defend against bacterial infection.

In this study, DC-SIGN and L-SIGN genes were identified from the miiyu croaker genome and also analyzed the characters of both genes. Miiyu croaker DC-SIGN and L-SIGN had similar domains with other fishes. We proposed a linear framework of DC-SIGN and L-SIGN in teleost and confirmed that mmDC-SIGN had no conserved genomic synteny with human and zebrafish, and L-SIGN genes in teleost had conserved synteny and underwent positive selection to adapt to the aquatic environment. We also confirmed that the two genes participated in the immune response in teleost. Most important of all, it is the first time to comprehensively analyze L-SIGN in fish and comparative genomics of gene synteny in DC-SIGN and L-SIGN to provide more information on the study of DC-SIGN family genes. L-SIGN-like gene which has high similarity with L-SIGN gene was obtained in miiyu croaker, tilapia, medaka, stickleback from gene synteny and clustered with L-SIGN in phylogenetic tree. But only one L-SIGN gene was found in mammals and frog. Two L-SIGN genes existed in some fishes maybe because of FSGD. From gene synteny, the L-SIGN and L-SIGN-like clustered together maybe to perform more functions to recognize and defend pathogens. The information can reveal the adaptive evolution and selection in the changing environment of L-SIGN genes. Our study will provide more information in studying the evolutionary process and expression patterns of immune genes.

Acknowledgments

This study was supported by National Natural Science Foundation of China (31370049) and Zhejiang Province Natural Science Foundation of Distinguished Young Scientists (LR14C040001).

Appendix: Supplementary material

Supplementary data to this article can be found online at doi:10.1016/j.dci.2015.01.004.

References

Alvarez, C.P., Lasala, F., Carrillo, J., Muniz, O., Corbi, A.L., Delgado, R., 2002. C-type lectins DC-SIGN and L-SIGN mediate cellular entry by Ebola virus in cis and in trans. J. Virol. 76, 6841–6844.
Aoki, T., Hikima, J., Hwang, S.D., Jung, T.S., 2013. Innate immunity of finfish: primordial conservation and function of viral RNA sensors in teleosts. Fish Shellfish Immunol. 35, 1689–1702.
Boily-Larouche, G., Zijenah, L.S., Mbfivo, M., Ward, B.J., Roger, M., 2007. DC-SIGN and DC-SIGNR genetic diversity among different ethnic populations: potential implications for pathogen recognition and disease susceptibility. Hum. Immunol. 68, 523–530.
Burke, C., Karlin, S., 1997. Prediction of complete gene structures in human genomic DNA. J. Mol. Biol. 268, 78–94.
Che, R.B., Sun, Y.Y., Sun, D.Q., Xu, T.J., 2014. Characterization of the miiyu croaker (miichthys miiyu) transcriptome and development of immune-relevant genes and molecular markers. Plos ONE 9, e94046.
Geijtenbeek, T.B., Torensla, R., van Vliet, S.J., van Duijnhoven, G.C., Adema, G.J., van Kooyk, Y., et al., 2000. Identification of DC-SIGN, a novel dendritic cell–specific ICAM-3 receptor that supports primary immune responses. Cell 100, 575–585.
Huang, Y.W., Meng, X.J., 2009. Identification of a porcine DC-SIGN-related C-type lectin, porcine CLEC4G (LSECtin), and its order of intron removal during splicing: comparative genomic analyses of the cluster of genes CD23/CLEC4G/DC-SIGN among mammalian species. Dev. Comp. Immunol. 33, 747–760.
Khoo, U.S., Chan, K.Y., Chan, V.S., Lin, C.L., 2008. DC-SIGN and L-SIGN: the SIGNs for infection. J. Mol. Med. 86, 861–874.
Kong, P., Wang, L., Zhang, H., Song, X., Zhou, Z., Yang, J., et al., 2011. A novel C-type lectin from bay scallop Argopecten irradians (AICTL-7) agglutinating fungi with manmace specificity. Fish Shellfish Immunol. 30, 836–844.
Koppel, E.A., van Gisbergen, K.P., Geijtenbeek, T.B., van Kooyk, Y., 2005. Distinct functions of DC-SIGN and its homologues L-SIGN (DCSIGNR) and mSIGNR1 in pathogen recognition and immune regulation. Cell. Microbio. 7, 157–165.
Lepenies, B., Lee, J., Sonkaria, S., 2013. Targeting C-type lectin receptors with multivalent carbohydrate ligands. Adv. Drug Deliv. Rev. 65, 1271–1281.
Letunca, I., Copley, R.R., Pilb, B., Pinkert, S., Schultz, J., Bork, P., 2006. SMART 5: domains in the context of genomes and networks. Nucleic Acids Res. 34, 257–260.
Li, H., Fu, W.P., Hong, Z.H., 2013. The VNTR polymorphism of the L-SIGN gene and susceptibility to HIV-1 infection in Han Chinese population. Infect. Genet. Evol. 17, 137–141.
Lin, A.F., Xiang, L.X., Wang, Q.L., Dong, W.R., Gong, Y.F., Shao, J.Z., 2009. The DC-SIGN of zebrafish: insights into the existence of a CD209 homolog in a lower vertebrate and its involvement in adaptive immunity. J. Immunol. 183, 7398–7410.
Lozach, P.Y., Lortat-Jacob, H., de Lacroix de Lavallette, A., Staropoli, I., Foug, S., Amara, A., et al., 2003. DC-SIGN and L-SIGN are high affinity binding receptors for hepatitis C virus glycoprotein E2. J. Biol. Chem. 278, 20358–20366.
Muffato, M., Louis, A., Poisnel, C.E., Crollius, H.R., 2010. Genomicus: a database and a browser to study gene synteny in modern and ancestral genomes. Bioinformatics 26, 1119–1121.
Page, R.D., 1996. TreeView: an application to display phylogenetic trees on personal computers. Comput. Appl. Biosci. 12, 357–358.
Pond, S.L., Frost, S.D., 2005. Datamonkey: rapid detection of selective pressure on individual sites of codon alignments. Bioinformatics 21, 2531–2533.
Redelinghuys, P., Brown, G.D., 2011. Inhibitory C-type lectin receptors in myeloid cells. Immunol. Lett. 136, 1–12.
Ronquist, F., Huelsenbeck, J.P., 2003. MrBayes 3: Bayesian phylogenetic inference under mixed models. Bioinformatics 19, 1572–1574.
Svajger, U., Anderluh, M., Jerais, M., Obermaier, N., 2010. C-type lectin DC-SIGN: an adhesion, signalling and antigen-uptake molecule that guides dendritic cells in immunity. Cell. Signal. 22, 1397–1405.
Tamura, K., Peterson, D., Peterson, N., Stecher, G., Nei, M., Kumar, S., 2011. MEGAS: molecular evolutionary genetics analysis using maximum likelihood, evolutionary distance, and maximum parsimony methods. Mol. Biol. Evol. 28, 2731–2739.
Thompson, J.D., Higgins, D.G., Gibson, T.J., 1994. CLUSTALW: improved sensitivity of progressive multiple sequence alignment through sequence weighting, positions-specific gap penalties and weight matrix choice. Nucleic Acids Res. 22, 4673–4680.
Xu, T.J., Zhi, Z.H., Sun, Y.N., Ren, L.P., Wang, R.X., 2014. Characterization and expression of the CXCR1 and CXCR4 in miiyu croaker and evolutionary analysis shows the strong positive selection pressures imposed in mammal CXCR1. Dev. Comp. Immunol. 44, 133–144.
Yang, Z., 2007. PAML 4: phylogenetic analysis by maximum likelihood. Mol. Biol. Evol. 24, 1586–1591.
Zhu, L.Y., Nie, L., Zhu, G., Xiang, L.X., Shao, J.Z., 2013a. Advances in research of fish immune-relevant genes: a comparative overview of innate and adaptive immunity in teleosts. Dev. Comp. Immunol. 39, 39–62.
Zhu, Z.H., Wang, R.X., Ren, L.P., Xu, T.J., 2013b. Characterization of the CCR3 and CCR9 genes in miiyu croaker and different selection pressures imposed on different domains between mammals and teleosts. Dev. Comp. Immunol. 41, 631–643.