Microbiology and Immunology

Microbiol Immunol 2017; 61: 185–189
doi: 10.1111/1348-0421.12484

NOTE

Predicting receptor functionality of signaling lymphocyte activation molecule for measles virus hemagglutinin by docking simulation

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ABSTRACT

Predicting susceptibility of various species to a virus assists assessment of risk of interspecies transmission. Evaluation of receptor functionality may be useful in screening for susceptibility. In this study, docking simulation was conducted for measles virus hemagglutinin (MV-H) and immunoglobulin-like variable domain of signaling lymphocyte activation molecule (SLAM-V). It was observed that the docking scores for MV-H and SLAM-V correlated with the activity of SLAM as an MV receptor. These results suggest that the receptor functionality may be predicted from the docking scores of virion surface proteins and cellular receptor molecules.

Key words docking simulation, hemagglutinin, measles virus, signaling lymphocyte activation molecule.

Recent environmental destruction on a global scale has facilitated contact between previously un-encountered species. If a virus is circulating in one species and the previously un-encountered species is naïve but susceptible to that virus, such contact may lead to interspecies transmission, which may cause diseases in the naïve species. This phenomenon has been observed for bat-to-human transmission of Ebola virus (1) and dog-to-lion transmission of canine distemper virus (2). Predicting susceptibility of various species to a virus would assist assessment of risk of such interspecies transmission.

Viral infection is initiated with interaction of virion surface proteins and cellular receptor molecules; this sometimes determines susceptibility (3). Susceptibility to murine norovirus is reportedly determined by the activity of CD300lf as a receptor for VP1 (4, 5). Similar phenomena have also been observed, for example, for MERS-CoV; the S protein of which binds to DPP4 (6); poliovirus, the VP1-VP3 of which bind to CD155 (7, 8); and so on. Because receptor functionality is a requirement for establishment of susceptibility, evaluation of the former may facilitate screening for the latter.

Receptor functionality may reflect the binding affinity of virion surface proteins and cellular receptor molecules. Binding affinity may be evaluated by using docking simulation analyzing 3-D structures, binding potential being measured as a docking score (9). Docking scores for DPP4 and MERS-CoV S protein reportedly correlate with the activity of DPP4 as a MERS-CoV receptor in mammals (6). It is therefore of interest to determine whether this relationship is also applicable to other viruses.

Measles virus is a member of the genus *Morbillivirus* in the family *Paramyxoviridae* in the order *Mononegavirales* (10). SLAM is known to be the principal cellular entry receptor for MV (11). The 3-D structure of the binding complex of MV-H and SLAM has been resolved (12). In addition, SLAM's activity as an MV receptor has been investigated by transfection experiments (11–17). The purpose of the present

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Received 3 January 2017; revised 11 March 2017; accepted 13 April 2017.

List of Abbreviations: DPP4, dipeptidyl peptidase 4; H, hemagglutinin; MERS-CoV, Middle East respiratory syndrome coronavirus; MV, measles virus; PDB, Protein Data Bank; S, spike; SLAM, signaling lymphocyte activation molecule; V, variable; VP, viral structural protein.

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study was to investigate the relationship between docking scores and activity of SLAM as an MV receptor by conducting docking simulation of MV-H and SLAM.

A type I transmembrane protein, SLAM belongs to the Ig superfamily (18). The Ig-like variable domain in the ectodomain of SLAM (SLAM-V) is responsible for binding to MV-H (14). The 3-D structure for the binding complex of MV-H and SLAM-V from cotton-top tamarin (*Saguinus oedipus*) has been resolved (PDB ID: 3ALZ, containing chain A for amino acid positions 188–606 of MV-H and chain B for positions 32–140 of SLAM-V) (12). The binding interface is composed of four sites (Sites 1–4); positions 505 and 507 of MV-H and positions 77 and 90 of SLAM-V are involved in Site 1; positions 530 and 533 of MV-H and position 123 of SLAM-V, as well as positions 552 and 554 of MV-H and positions 61 and 63 of SLAM-V, in Site 2; positions 191–195 of MV-H and positions 127–131 of SLAM-V in Site 3; and positions 524, 541, 543, and 552 of MV-H and positions 75, 119, and 130 of SLAM-V, as well as positions 483, 524, 543, and 545 of MV-H and positions 72 and 74 of SLAM-V, in Site 4 (12). In total, 55 pairs of amino acid positions between MV-H and SLAM-V appear to form the binding interface.

The activity of SLAM as MV receptor has been investigated by transfection experiments for human (*Homo sapiens*), dog (*Canis familaris*), cow (*Bos taurus*), cotton rat (*Sigmodon hispidus*), and mouse (*Mus musculus*). Human is the single natural host of MV; indeed, human SLAM has been shown to act as an MV receptor (11–17). Nevertheless, it has also been demonstrated that SLAMs from dog, cow, and cotton rat act as MV receptors, although less efficiently than human SLAM (13, 17). In contrast, mouse SLAM reportedly does not exhibit MV receptor functionality (14, 15). SLAM sequences of these species have been retrieved from the International Nucleotide Sequence Database; their accession numbers are listed in Table 1.

Transfection experiments have also been performed for chimeras of human and mouse SLAMs (15, 16). It was found that replacement of amino acid positions 58–67 of human SLAM-V with the corresponding positions of mouse SLAM-V abolishes MV-H binding ability of human SLAM. Conversely, replacement of the same positions of mouse SLAM-V with those of human SLAM-V confers MV-H binding ability to mouse SLAM. The amino acid sequences of positions 58–67 of human SLAM-V and the corresponding positions of mouse SLAM-V differ at positions 60, 61, and 63 (15). Since positions 61 and 63 of SLAM-V and positions 552 and 554 of MV-H constitute part of Site 2 in the binding interface as described above, these positions may be critical for binding of SLAM-V and MV-H.

For each of human, dog, cow, cotton rat, and mouse SLAM-Vs, and for human SLAM-V the amino acid positions 60, 61, and 63 of which were mutated to those of mouse SLAM-V (mutated human SLAM-V), and for mouse SLAM-V the corresponding positions of which were mutated to those of human SLAM-V (mutated mouse SLAM-V), pairwise alignment of amino acid sequences was achieved with cotton-top tamarin SLAM-V (International Nucleotide Sequence Database accession number: AF257239) using computer program MAFFT (version 7.305b) (19). Thence, the 3-D structure was constructed by homology modelling using MODELLER (version 9.17) (20) and using the 3-D structure of cotton-top tamarin SLAM-V as the template (PDB ID: 3ALZ, B chain).

The 3-D structure of SLAM-V obtained as described above and that of MV-H (PDB ID: 3ALZ, A chain) were employed for docking simulation with ClusPro (version 2.0), which is one of the best, fully automated web servers available for protein–protein docking (21). In ClusPro, missing atoms and polar hydrogens are automatically added to the proteins before docking simulation. Amino acid positions 61 and 63 of SLAM-V and positions 552 and 554 of MV-H were assumed to attract one another. Several candidate structures of

Table 1. Results from docking simulation of MV-H and immunoglobulin-like SLAM-V assuming that interacting amino acid positions are located within 10 Å

SLAM-V	Accession number†	Pairs within 10 Å‡	Docking score§
Human	AY040554	49	−963.9
Dog	AF325357	50	−872.6
Cow	AF329970	48	−802.0
Cotton rat	JX424845	50	−778.5
Mouse	AF160990	45	−683.9
Mutated human	N.A.	52	−773.7
Mutated mouse	N.A.	44	−785.8

†accession number in the International Nucleotide Sequence Database; ‡ number of pairs of amino acid positions located within 10 Å in the selected structure of the binding complex of MV-H and SLAM-V obtained from ClusPro (21) of the 55 pairs that form the binding interface in the original structure of MV-H and cotton-top tamarin SLAM-V (Protein Data Bank ID: 3ALZ) (12), §Docking score for the selected structure of the binding complex of MV-H and SLAM-V obtained from ClusPro (21).
N.A., not applicable.

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binding complexes together with their docking scores were generated for each SLAM-V. The candidate structures that appeared to be the most closely related to the original structure of the binding complex of MV-H and cotton-top tamarin SLAM-V (PDB ID: 3ALZ) (12) was selected as follows. In each candidate structure, physical distances were computed for 55 pairs of amino acid positions that are considered to form the binding interface in the original structure, as described above. On the assumption that interacting amino acid positions are located within 10 Å (22, 23), the structure in which the greatest number of the 55 pairs of amino acid positions are located within 10 Å was considered to be the most closely related to the original structure. In fact, more than 90% (50 of the 55 pairs) were located within 10 Å in the original structure.

From 44 to 52 of the 55 pairs of amino acid positions were found to be located within 10 Å in the selected structures; these were largely comparable to the original structure (Table 1). Transfection experiments have shown that SLAM is functional as an MV receptor in human, functional but with less efficiency in dog, cow and cotton rat, and non-functional in mouse (11–17). The docking score for MV-H and SLAM-V is consistently lowest (indicating the strongest interaction) in human, intermediate in dog, cow and cotton rat, and highest (indicating the weakest interaction) in mouse (Table 1). The probability of obtaining this order of docking scores ([human] < [dog, cow and cotton rat] < [mouse]) by chance is 0.05, which is of marginal significance.

In additional transfection experiments, recombinant human SLAM containing the amino acid sequence of mutated human SLAM-V did not act as an MV receptor, whereas recombinant mouse SLAM containing the sequence of mutated mouse SLAM-V did (15, 16). Accordingly, the docking scores for mutated human SLAM-V were found to be higher than those for SLAM-Vs functional as MV receptor, whereas the docking scores for mutated mouse SLAM-V were lower than those for SLAM-Vs non-functional as MV receptor (Table 1). The probability of obtaining this order of docking scores ([human, dog, cow, cotton rat, and mutated mouse] < [mouse and mutated human]) by chance is 0.0476, which is statistically significant. Thus, there is a correlation between docking scores for MV-H and SLAM-V and the activity of SLAM as an MV receptor.

In the above analysis, interacting amino acid positions were assumed to be located within 10 Å (22, 23). To examine the robustness of the results against violation of this assumption, the above analysis was repeated assuming that interacting amino acid positions were located within 5 Å or 15 Å. In the original structure of the binding complex of MV-H and cotton-top tamarin SLAM-V (PDB ID: 3ALZ) (12), 27 and 55 pairs of the 55 pairs of interacting amino acid positions are located within 5 Å and 15 Å, respectively. When 5 Å was used as the threshold value, the selected structures sometimes differed from those in Table 1 (dog, mouse and mutated human SLAM-Vs) (Table S1). However, the correlation between the docking score of MV-H and SLAM-V and the activity of SLAM as an MV receptor was unaffected. Similarly, different structures were sometimes selected when 15 Å was used as the threshold value (dog, cow and mutated human SLAM-Vs) (Table S2). In this case, however, multiple structures were selected for two SLAM-Vs (cow and mutated human), and in one case (cow) the docking score of the additional structure did not support the correlation. This result was apparently attributable to the threshold value being too large to distinguish different structures. Indeed, all 55 pairs of interacting amino acid positions were found to be located within 15 Å in multiple selected structures. These observations suggest that the results obtained in the present study are robust provided the threshold value assumed for the physical distance between interacting amino acid positions is appropriate (22, 23).

Previous docking simulation studies of MV-H and SLAM-V have examined the relationship between docking scores and susceptibility to MV in mammals (24, 25). However, the relationship has remained unclear, probably because susceptibility is determined by multiple infection-related processes. In fact, although dog, cow and cotton rat SLAMs have been shown to act as MV receptors in transfection experiments (13, 17), these species are not considered to be susceptible to MV (24, 25). It is possible that infection processes of MV that are irrelevant to receptor functionality are inhibited in these species. Cross-immunity to MV elicited by infection of other morbilliviruses, such as canine distemper virus and peste-des-petits-ruminants virus, may also suppress infection of MV in these species (26).

In the present study, it was observed that the docking scores of MV-H and SLAM-V correlated with the activity of SLAM as an MV receptor, suggesting that receptor functionality may be predicted from the docking scores of virion surface proteins and cellular receptor molecules. This approach may be useful for screening for susceptibility of various species to a virus when assessing the risk of interspecies transmission (1, 2). It will be of interest to investigate whether this relationship is further applicable to other viruses (6).

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ACKNOWLEDGEMENTS

The author thanks two anonymous reviewers for valuable comments. This study was supported by a Grant-in-Aid for Research in Nagoya City University to Y.S.

DISCLOSURE

The author declares no conflict of interest.

REFERENCES

1. Hoffmann M., Hernandez M.G., Berger E., Marzi A., Pohlmann S. (2016) The glycoproteins of all filovirus species use the same host factors for entry into bat and human cells but entry efficiency is species dependent. *PLoS ONE* **11**: e0149651.
2. Viana M., Cleaveland S., Matthiopoulos J., Halliday J., Packer C., Craft M.E., Hampson K., Czupryna A., Dobson A.P., Dubovi E.J., Ernest E., Fyumagwa R., Hoare R., Hopcraft J.G., Horton D.L., Kaare M.T., Kanellos T., Lankester F., Mentzel C., Mlengeya T., Mzimbiri I., Takahashi E., Willett B., Haydon D.T., Lembo T. (2015) Dynamics of a morbillivirus at the domestic-wildlife interface: Canine distemper virus in domestic dogs and lions. *Proc Natl Acad Sci USA* **112**: 1464–9.
3. Helenius A. (2013) Virus entry and uncoating. In: Knipe D.M., Howley P.M., Cohen J.I., Griffin D.E., Lamb R.A., Martin M.A., Racaniello V.R., Roizman B., eds. *Fields Virology, Sixth Edition*. Philadelphia: Lippincott Williams & Wilkins, pp. 87–104.
4. Haga K., Fujimoto A., Takai-Todaka R., Miki M., Doan Y.H., Murakami K., Yokoyama M., Murata K., Nakanishi A., Katayama K. (2016) Functional receptor molecules CD300lf and CD300ld within the CD300 family enable murine noroviruses to infect cells. *Proc Natl Acad Sci USA* **113**: E6248–55.
5. Orchard R.C., Wilen C.B., Doench J.G., Baldridge M.T., McCune B.T., Lee Y.-C.J., Lee S., Pruett-Miller S.M., Nelson C.A., Fremont D.H., Virgin H.W. (2016) Discovery of a proteinaceous cellular receptor for a norovirus. *Science* **353**: 933–6.
6. van Doremalen N., Miazgowicz K.L., Milne-Price S., Bushmaker T., Robertson S., Scott D., Kinne J., McLellan J.S., Zhu J., Munster V.J. (2014) Host species restriction of Middle East respiratory syndrome coronavirus through its receptor, dipeptidyl peptidase 4. *J Virol* **88**: 9220–32.
7. Ida-Hosonuma M., Sasaki Y., Toyoda H., Nomoto A., Gotoh O., Yonekawa H., Koike S. (2003) Host range of poliovirus is restricted to simians because of a rapid sequence change of the poliovirus receptor gene during evolution. *Arch Virol* **148**: 29–44.
8. Suzuki Y. (2006) Ancient positive selection on CD155 as a possible cause for susceptibility to poliovirus infection in simians. *Gene* **373**: 16–22.
9. Kozakov D., Beglov D., Bohnuud T., Mottarella S., Xia B., Hall D.R., Vajda S. (2013) How good is automated protein docking? *Bioinformatics* **81**: 2159–66.
10. Griffin D.E. (2013) Measles virus. In: Knipe D.M., Howley P.M., Cohen J.I., Griffin D.E., Lamb R.A., Martin M.A., Racaniello V.R., Roizman B., eds. *Fields Virology, Sixth Edition*. Philadelphia: Lippincott Williams & Wilkins, pp. 1042–69.
11. Tatsuo H., Ono N., Tanaka K., Yanagi Y. (2000) SLAM (CDw150) is a cellular receptor for measles virus. *Nature* **406**: 893–7.
12. Hashiguchi T., Ose, T., Kubota M., Maita N., Kamishikiryo J., Maenaka K., Yanagi Y. (2011) Structure of the measles virus hemagglutinin bound to its cellular receptor SLAM. *Nat Struct Mol Biol* **18**: 135–41.
13. Tatsuo H., Ono N., Yanagi Y. (2001) Morbilliviruses use signaling lymphocyte activation molecules (CD150) as cellular receptors. *J Virol* **75**: 5842–50.
14. Ono N., Tatsuo H., Tanaka K., Minagawa H., Yanagi Y. (2001) V domain of human SLAM (CDw150) is essential for its function as a measles virus receptor. *J Virol* **75**: 1594–600.
15. Ohno S., Seki F., Ono N., Yanagi Y. (2003) Histidine at position 61 and its adjacent amino acid residues are critical for the ability of SLAM (CD150) to act as a cellular receptor for measles virus. *J Gen Virol* **84**: 2381–8.
16. Xu Q., Zhang P., Hu C., Liu X., Qi Y., Liu Y. (2006) Identification of amino acid residues involved in the interaction between measles virus haemagglutin (MVH) and its human cell receptor (signaling lymphocyte activation molecule, SLAM). *J Biochem Mol Biol* **39**: 406–11.
17. Carsillo T., Huey D., Levinsky A., Obojes K., Schneider-Schaulies J., Niewiesk S. (2014) Cotton rat (*Sigmodon hispidus*) signaling lymphocyte activation molecule (CD150) is an entry receptor for measles virus. *PLoS ONE* **9**: e110120.
18. Tatsuo H., Yanagi Y. (2002) The morbillivirus receptor SLAM (CD150). *Microbiol Immunol* **46**: 135–42.
19. Katoh K., Misawa K., Kuma K., Miyata T. (2002) MAFFT: A novel method for rapid multiple sequence alignment based on fast Fourier transform. *Nucleic Acids Res* **30**: 3059–66.
20. Sali A., Blundell T.L. (1993) Comparative protein modelling by satisfaction of spatial restraints. *J Mol Biol* **234**: 779–815.
21. Kozakov D., Hall D.R., Xia B., Porter K.A., Padhorny D., Yueh C., Beglov D., Vajda S. (2017) The ClusPro web server for protein-protein docking. *Nat Protoc* **12**: 255–78.
22. Cohen M., Potapov V., Schreiber G. (2009) Four distances between pairs of amino acids provide a precise description of their interaction. *PLoS Comput Biol* **5**: e1000470.
23. Tek A., Chavent M., Baaden M., Delalande O., Bourdot P., Ferey N. (2012) Advances in human-protein interaction—interactive and immersive molecular simulations. In: Cai W. ed. *Protein-Protein Interactions—Computational and Experimental Tools*. Shanghai: InTech, Rijeka, pp. 27–64.
24. Maruyama T., Shimane Y., Ohishi K., Iwasawa M., Hatada Y., Usui K., Takaki Y., Yoshida T., Tanaka S., Anzaki S., Komeiji Y., Watanabe C., Okiyama Y., Mochizuki Y., Fukuzawa K. (2014) Analysis of global ecosystem ecology by fragment molecular orbital (FMO) method - analyses of the interactions between virus hemagglutinins and their receptors -. In *Annual Report of the Earth Simulator April 2013–March 2014*. Yokohama: Center for Earth Information Science and Technology, Japan Agency for Marine–Earth Science and Technology, pp. 155–60.
25. Maruyama T., Shimane Y., Ohishi K., Iwasawa M., Hatada Y., Takaki Y., Yoshida T., Tanaka S., Anzaki S., Komeiji Y., Mochizuki Y., Fukuzawa K. (2015) Analysis of global ecosystem ecology by fragment molecular orbital (FMO) method - analyses of the interactions between virus hemagglutinins and their receptors -. In *Annual Report of the Earth Simulator April 2014–March 2015*. Yokohama: Center for Earth Information Science and Technology, Japan Agency for Marine-Earth Science and Technology, pp. 131–5.
26. de Vries R.D., Ludlow M., Verburgh R.J., van Amerongen G., Yuksel S., Nguyen D.T., McQuaid S., Osterhaus A.D.M.E., Duprex W.P., de Swart R.L. (2014) Measles vaccination of

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nonhuman primates provides partial protection against infection with canine distemper virus. *J Virol* **88**: 4423–33.

SUPPORTING INFORMATION

Additional supporting information may be found in the online version of this article at the publisher's web-site.

Table S1. Results from docking simulation of MV-H and immunoglobulin-like SLAM-V assuming that interacting amino acid positions are located within 5 Å

Table S2. Results from docking simulation of MV-H and immunoglobulin-like SLAM-V assuming that interacting amino acid positions are located within 15 Å

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