BIOGRAPHICAL SKETCH

NAME: Andres Merits

POSITION TITLE: Professor of Applied Virology

EDUCATION/TRAINING

INSTITUTION AND LOCATION	DEGREE *(if applicable)*	Completion Date MM/YYYY	FIELD OF STUDY
Moscow State University (USSR)	M.S	06/1990	Biochemistry
Moscow State University (USSR/Russia)	Ph.D.	04/1994	Virology
University of Helsinki (Finland)	Postdoctoral	12/2001	Molecular virology/viral pathogenesis

A. Personal Statement

In 01.01.2003 I became research professor at University of Tartu and in 2007 I was elected to the position of Professor of Applied Virology in the Institute of Technology, University of Tartu. My laboratory is working on topics of the molecular biology of viruses with RNA genomes. The main focus is on genome replication of alphaviruses (Semliki Forest virus, Chikungunya virus, Ross River virus, O'nyong'nyong virus), flaviviruses (Zika virus, Dengue virus) and coronaviruses (SARS-CoV-2) and construction of virus-based gene expression vectors. Our research also includes construction of systems for antiviral drug screening (Chikungunya virus, Zika virus, SARS-CoV-2) and analysis of antiviral properties of low molecular mass compounds. We have been pioneers in the functional analysis of alphavirus replicase proteins, the development of enzymatic assays for them and the use of these tools to analyze the mechanisms of action of antiviral hit compounds. Our latest studies strongly support a potential new approach for detection of virus infection both *in vitro* and *in vivo*.
The research has lead to >170 peer-reviewed publications. According to Thomson Reuters Web of Science database (31.01.2022) they have been cited >5800 times (h-index = 46). My current research group addressing these topics consists from 8 researchers: three senior scientists (with Ph.D. or equivalent degree) and five Ph.D. students. This group is involved in intensive world-wide international collaborations. I have successfully supervised 13 PhD students.

Ongoing and recently completed projects:
The Wellcome Trust Collaboration Award 200171/Z/15/Z.
Alphey (PI), Role: co-PI
09/01/16-12/31/21
Genetic approaches to reducing vector competence of Aedes aegypti for chikungunya virus.

Estonian Ministry of Science and Education 2014-2020.4.01.15-013.
Tenson (PI), Role: co-PI
09/01/16-08/31/22
Centre of Excellence in in Molecular Cell Engineering, Estonia.

The Defense Advanced Research Projects Agency (DARPA)
Alphey (PI), Role: co-PI
10/01/18-12/31/21.
Predicting and preventing flavivirus spillover

PRG1154, Estonian Research Council
Merits (PI)
01/01/21-12/31/25
Role of arbovirus replicase proteins in RNA replication, virus-host interactions and vector transmission

B. Positions, Scientific Appointments, and Honors

Positions and Scientific Appointments

2013 - Present	Full Professor (permanent), Institute of Technology, University of Tartu, Estonia
2007-2012	Full Professor, Institute of Technology, University of Tartu, Estonia
2003-2006	Research Professor, Institute of Molecular and Cell Biology, University of Tartu, Estonia
2002-2003	Senior Research Scientist, Estonian Biocentre, Tartu, Estonia
2001-Present	Docent (Associated Professor), University of Helsinki, Finland

Honors

2019	Elected as Academy Professor by Estonian Academy of Sciences
2015	Estonian State Award in Molecular Biology and Chemistry

C. Contributions to Science

1. Functional studies of alphavirus replicase proteins. I have been working with alphaviruses since 1997 and for all this period my main research interest has been the alphavirus replicase, its components, their properties and interactions. The main research approaches have been development of reverse genetics for these viruses and tools for in depth studies of the viral replicase. I have been working with all four replicase proteins (nsP1, nsP2, nsP3 and nsP4), have developed methods of purification and enzymatic assays for all of them and participated in studies resulting in revealing 3D-structures of these enzymes. The main focus has been nsP2, the viral RNA helicase/protease and also the key enzyme involved in regulation of alphavirus replication.

- Das, P.K., **Merits, A.*** & Lulla A*. (2014). Functional crosstalk between distant domains of chikungunya virus non-structural protein 2 Is decisive for its RNA-modulating activity. Journal of Biological Chemistry. 289(9):5635-5653. PMCID: PMC3937639 **co-corresponding authors*.
- Abraham, R., Hauer, D., McPherson R.L., Utt, A., Kirby, I., Cohen, M.S., **Merits, A**, Leung, A.K.L. & Griffin, D.E. (2018) ADP ribosyl-binding and hydrolase activities of the alphavirus nsP3 macrodomain are critical for initiation of virus replication. Proceedings of the National Academy of Sciences of the U S A, 115(44):E10457-E10466. PMCID: PMC6217424
- Bakhache, W., Neyret, A., Bernard, E., **Merits, A.** & Briant, L. (2020). Palmitoylated cysteines in chikungunya virus nsP1 are critical for targeting to cholesterol-rich plasma membrane microdomains with functional consequences for viral genome replication. Journal of Virology, 94(10):e02183-19. PMCID: PMC7199415
- Law, Y.-S., Utt, A., Tan, Y.B., Zheng, J., Wang, S. Chen, M.W., Griffin, P.R., **Merits, A*.** & Luo, D*. (2019). Structural insights into RNA recognition by the chikungunya virus nsP2 helicase. Proceedings of the National Academy of Sciences of the U S A, 116(19):9558-9567. PMCID: PMC6511008. **co-corresponding authors*.
- Tan, Y.B., Lello, L.S., Liu, X., Law, Y.-S., Kang, C., Lescar, J., Zheng, J., **Merits, A*.** & Luo, D*. (2022). Crystal structures of alphavirus nonstructural protein 4 (nsP4) reveal an intrinsically dynamic RNA-dependent RNA polymerase fold. Nucleic Acids Research. doi: 10.1093/nar/gkab1302. **co-corresponding authors*.

2. Assembly of functional alphavirus replicase complexes. In order to form the functional replicase complex alphavirus replicase proteins should interact with each other and with the virus genome. I have been involved in studies that have revealed that in order to do so the proteins need to be produced in form of polyprotein precursors that are processed by nsP2 protease activity in a highly specific manner, and that disturbance of this processing pathway has major consequences for virus infectivity and virulence.

- Saul, S., Ferguson, M., Cordonin, C., Fragkoudis, R., Ool, M., Tamberg, N., Sherwood, K., Fazakerley, J.K. & **Merits, A.** (2015). Differences in processing determinants of non-structural polyprotein and in the sequence of non-structural protein 3 affect neurovirulence of Semliki Forest virus. Journal of Virology, 89(21):11030-45. PMCID: PMC4621116

- Hellström, K., Kallio, K., Utt, A., Quirin, T., Jokitalo, E., **Merits, A**. & Ahola, T. (2017). Partially uncleaved alphavirus replicase forms spherule structures in the presence and absence of RNA template. Journal of Virology, 91(18):e00787-17. PMCID: PMC5571266
- Lulla, V., Karo-Astover, L., Rausalu, K., Saul, S., **Merits, A*.** & Lulla, A*. (2018). Timeliness of proteolytic events is prerequisite for efficient functioning of the alphaviral replicase. Journal of Virology, 92(14):e00151-18. PMCID: PMC6026757. **co-corresponding authors.*
- Teppor, M., Žusinaite, E., Karo-Astover, L., Omler, A., Rausalu, K., Lulla, V., Lulla, A. & **Merits, A.** (2021). Semliki Forest virus chimeras with functional replicase modules from related alphaviruses survive by adaptive mutations in functionally important hotspots. Journal of Virology, doi: 10.1128/JVI.00973-21

3. Elucidating host-virus interactions that contribute to alphavirus replication and pathogenesis**.** My studies of the functions of replicase proteins and virus replication complexes lead to the analysis of the complicated interactions between alphavirus and the host. We have studied effects of mutations in alphavirus replicase proteins on the ability of virus to persist in infected host cells, and we have identified numerous host protein interactions with alphavirus replicases. Some of these interactions are essential for the virus and some (such as activation of interferon response) are generally regarded as harmful for the virus. However, our studies have clearly shown that this is not strictly true, some interactions (such as activation of interferon responses) can also represent a part of virus attack strategy and be related to viral pathogenesis.

- Nikonov, A., Mölder, T., Sikut, R., Kiiver, K., Männik, A., Toots, U., Lulla. A., Lulla, V., Utt, A., **Merits, A.** & Ustav M. (2013). RIG-I and MDA-5 detection of viral RNA-dependent RNA polymerase activity restricts positive-strand RNA virus replication. PLoS Pathogens, 9(9):e1003610. PMCID: PMC3764220
- Maillard, P.V., Van der Veen, A.G., Deddouche-Grass, S., Rogers, N.C., **Merits, A**. & Reis E Sousa, C. (2016). Inactivation of the type I interferon pathway reveals long dsRNA-mediated RNA interference. The EMBO Journal, 35(23):2505-2518. PMCID: PMC5167344
- Liu, X., Mutso, M., Utt. A., Lepland, A., Herrero, L., Taylor, A., Bettadapura, J., Rudd, P., **Merits A*** & Mahalingam, S*. (2018). Decreased virulence of Ross River virus harboring mutation in the first cleavage site of non-structural polyprotein is caused by a novel mechanism leading to increased production of interferon-inducing RNAs. mBio, 9(4):e00044-18. PMCID: PMC6106088. **co-corresponding authors*.
- Götte, B., Utt, A., Fragkoudis, R., **Merits, A*.** & McInerney, G*. (2020). Sensitivity of alphaviruses to G3BP deletion correlates with efficiency of replicase polyprotein processing. Journal of Virology*,* 94(7):e01681-19. PMCID: PMC7081891. **co-corresponding authors*

4. Analysis of the mechanisms used by host and transmission vectors to limit alphavirus infection and pathology. The increasing importance of alpaviruses, most notably chikungunya virus, promoted novel directions including analysis of host mechanisms counter-acting chikungunya virus infection. My research group has (mostly in international collaborations) worked on characterization of host defense mechanisms against alphaviruses. These studies have also revealed the complicated roles of host immunity that, as for many viruses, also impacts alphavirus-induced pathology.

- Teng, T.S., Foo, S.S., Simamarta, D., Lum, F.M., Teo, T.H., Lulla, A., Yeo, N.K., Koh, E.G., Chow, A., Leo, Y.S., **Merits, A.**, Chin, K.C. & Ng, L.F.P. (2012). Viperin restricts chikungunya virus replication and pathology. Journal of Clinical Investigations, 122(12): 4447-60. PMCID: PMC3533538
- Teo, T.H., Lum, F.M., Claser, C., Lulla, V., Lulla, A., **Merits, A.**, Rénia, L. & Ng, L.F.P. (2013). A pathogenic role for CD4+ T cells during chikungunya virus infection in mice. 2013. Journal of Immunology, 190(1):259-69. doi: 10.4049/jimmunol.1202177
- Pingen, M., Bryden, S.R., Pondeville, E., Schnettler, E., Kohl, A., **Merits, A.**, Fazakerley, J.K., Graham, G.J. & McKimmie, C.S. (2016). Host inflammatory response to mosquito bites defines severity of arbovirus infection. Immunity*,* 44(6):1455-69. PMCID: PMC4920956
- Bryden, S.R., Pingen, M., Lefteri, D.A., Major, J., Delang, L., Jacobs, S., Abdelnabi, R., Neyts, J., Miltenburg, J., Khalid, H., Tuplin, A., **Merits, A**., Pondeville, E., Edgar, J., Graham, G.J., Shams, K. & McKimmie, C.S. (2020). Pan-viral protection against arboviruses by targeting inoculation site-

based skin macrophages. Science Translational Medicine, 12(527):eaax2421. doi: 10.1126/scitranslmed.aax2421

5. Development of tools and assays applicable for screening of alphavirus replicase inhibitors and characterization of their mechanisms of action. The increasing importance of alpaviruses particularly chikungunya virus also promoted novel directions aimed to counter-act chikungunya virus infection and to develop antiviral strategies such as antiviral compounds. Advantage was taken from our basic studies on the structure and function of alphavirus replicase proteins. In addition, based on our insights into the pathways of virus replicase complex formation we have developed multiple tools and assays applicable to the screening of antiviral compounds, analysis of their mechanism of action, and characterization of resistance against these compounds.

- Pohjala, L., Utt, A., Varjak, M., Lulla, A., **Merits, A.**, Ahola, T. & Tammela, P. (2011). Inhibitors of alphavirus entry and replication identified with a stable chikungunya replicon cell line and virus-based assays. PLoS One, 6(12):e28923. PMCID: PMC3242765
- Utt, A., Das, P.K., Varjak, M., Lulla, V., Lulla, A. & **Merits A**. (2015). Mutations conferring a non-cytotoxic phenotype on chikungunya virus replicons compromise enzymatic properties of non-structural protein 2. Journal of Virology, 89(6):e3145-62. PMCID: PMC4337533
- Das, P.K., Puusepp, L., Varghese, F.S., Utt, A., Ahola, T., Kananovich, D.G., Lopp, M., **Merits, A*.** & Karelson, M*. (2016). Design and validation of novel chikungunya virus protease inhibitors. Antimicrobial Agents and Chemotherapy, 60(12):7382-7395. PMCID: PMC5119020. **co-corresponding authors*
- Teo, T.H., Chan, Y.H., Lee, W.W., Lum, F.M., Amrun, S.N., Her, Z., Rajarethinam, R., **Merits, A**., Rötzschke, O., Rénia, L. & Ng, L.F.P.(2017). Fingolimod treatment abrogates chikungunya virus-induced arthralgia. Science Translational Medicine, 9(375):eaal1333. doi: 10.1126/scitranslmed.aal1333

6. Outbreak of Zika virus (ZIKV) has boosted our interest to this pathogen and flaviviruses in general. In a very short time we have been able to contribute to the filed by developing a new ZIKV reverse genetics system (currently most commonly used system in world, at least 50 different laboratories have requested and obtained it form us), applying it for studies of anti-ZIKV compounds; developing of novel assay for diagnostics of ZIKV infection and by analyzing ZIKV/mosquito vector interactions:

- Mutso, M., Saul, S., Rausalu, K., Susova, O., Žusinaite, E., Mahalingam, S. & **Merits, A**. (2017). Reverse genetic system, genetically stable reporter viruses and packaged subgenomic replicon based on Brazilian Zika virus isolate. Journal of General Virology, 98, 2712-2724. doi: 10.1099/jgv.0.000938.
- Varghese, F.S., Rausalu, K., Hakanen, M., Saul, S., Kümmerer, B., Susi, P., **Merits, A**. & Ahola, T. (2017). Obatoclax inhibits alphavirus membrane fusion by neutralizing the acidic environment of endocytic compartments. Antimicrobial Agents and Chemotherapy, pii: AAC.02227-16. 61. pii: e02227-16. doi: 10.1128/AAC.02227-16
- Lum, F.-M., Lin, C., Susova, O.Y, Teo, T.-H., Fong, S.-W., Mak, T.-M., Lee, L.K., Chong, C.-Y., Lye, D.C.B., Lin, R.T.P., **Merits, A.,** Leo, Y.-S. & Ng, L.F.P. (2017). Sensitive detection of Zika virus antigen in patients' whole blood as an alternative diagnostic approach. The Journal of Infectious Diseases, 216, 182-190. doi: 10.1093/infdis/jix276
- Varjak, M., Donald, C.L., Mottram, T., Sreenu, V.B., **Merits, A.,** Maringer, K., Schnettler, E. & Kohl, A. (2017). Characterization of the Zika virus induced small RNA response in Aedes aegypti cells. PLOS Neglected Tropical Diseases, **11**:e0006010. doi: 10.1371/journal.pntd.000601
- Gestuveo, R.J., Royle, J., Donald, C.L., Lamont, D.J., Hutchinson, E.C., **Merits, A**., Kohl, A. & Varjak, M. (2021). Analysis of Zika virus capsid-*Aedes aegypti* mosquito interactome reveals pro-viral host factors critical for establishing infection. Nature Communications, 12(1):2766. doi: 10.1038/s41467-021-22966-8.

7. Since 2020 my group has also started to work with SARS-CoV-2. We have constructed single plasmid based infectious cDNA clone for SARS-CoV-2 and use it to create tools for our studies of SARS-CoV-2 adaptations, resistance to antiviral compounds, functions of viral RNA structures and mechanism of virus-host interactions. We have generated >200 mutant versions of SARS-CoV-2 genome including version resistant to remdesivir (with mutation in nsP12). We have analyzed the secondary structures preset in viral RNA genome, and effects of combinations of drugs against SARS-CoV-2. We have completed construction and pre-clinical evaluation of candidates for live attenuated SARS-CoV-2 vaccine. Currently we are working on development of permanent replicon cell lines for SARS-CoV-2 and on construction of virus-free RNA replication system for SARS-CoV-2.

a. Rihn, S. J*., **Merits, A*.,** Bakshi, S., Matthew L. Turnbull, M.L., Wickenhagen, A. et al. Zaid, A., Wilson S. J. & Mahalingam, S. (2021). A plasmid DNA-launched SARS-CoV-2 reverse genetics system and coronavirus toolkit for COVID-19 research. PloS Biology, 19(2):e3001091. doi: 10.1371/journal.pbio.3001091. **co-first authors*
b. Yang, S.L., DeFalco, L., Anderson, D., Zhang, Y., Aw, A., Lim, X.N., Tan, A.K.Y., Zhang, T., Chawla, T., Su, Y., Lezhava, A., **Merits, A.,** Wang, L.-F., Huber, R. & Wan, Y. (2021). Comprehensive mapping of SARS-CoV-2 interactions in vivo reveals functional virus-host interactions. Nature communications, 12(1):5113. doi: 10.1038/s41467-021-25357-1.
c. Szemiel, A.M., **Merits, A.,** Orton, R.J., MacLean, O.A., Pinto, R.M., Wickenhagen, A., Lieber, G., Turnbull, M.L., Wang, S., Furnon, W., Suarez, N., Mair. D., Filipe, A.S., Willett, B.J., Wilson, J.S., Patel, A.H., Thomson, E.C., Palmarini, M., Kohl, A., & Stewart, M.E. In vitro selection of Remdesivir resistance suggests evolutionary predictability of SARS-CoV-2. PLoS Pathogens, https://doi.org/10.1371/journal.ppat.1009929
d. Ianevski, A., Yao, R., Zusinaite, E., Lello, L.S., Wang, S., Jo, E., Yang, J., Ravlo, E., Wan, W., Lysvand, H., Løseth, K., Oksenych, V., Tenson, T., Windisch, M.P., Poranen, M., Nieminen, A.I., Nordbø, S.A., Fenstad, M.H., Grødeland, G., Aukrust, P., Trøseid, M., Kantele, A., Lastauskienė, E., Vitkauskiene, A., Legrand, N., **Merits, A.,** Bjørås, M. & Kainov, D.E. (2021) Synergistic interferon alpha-based combinations for treatment of SARS-CoV-2 and other viral infections. Viruses, 2021;13(12):2489. doi: 10.3390/v13122489