

**Vector pHAGE2 Containing the Angiotensin-Converting Enzyme 2 Gene**

**Catalog No. NR-52512**

**For research use only. Not for human use.**

**Contributor:**

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**Manufacturer:**

BEI Resources

**Product Description:**

The vector for the human angiotensin-converting enzyme 2 (ACE2) gene (GenBank: [NM\\_021804](#)) was subcloned into the pHAGE2 lentiviral backbone vector under the human EF1 $\alpha$  promoter with an intron to increase expression.<sup>1,2,3</sup> In addition, pHAGE2 includes the Woodchuck hepatitis virus post-transcriptional regulatory element to enhance levels of transcription and gene expression.<sup>3</sup> NR-52512 contains the beta-lactamase gene, *bla*, to provide transformant selection through ampicillin resistance in *Escherichia coli* (*E. coli*). The resulting size of the plasmid is approximately 9390 base pairs. The complete plasmid sequence and map (Figure 1) are provided on the BEI Resources webpage. The plasmid was produced in *E. coli* and extracted.

NR-52512 is part of a lentiviral expression system, and additional BEI Resources items are required for successful ACE2 expression. Lentiviral expression of ACE2 requires lentiviral helper plasmids (BEI Resources NR-52517, NR-52518 and NR-52519, which are provided in the kit NR-52948). Protocols for the use of these items are published.<sup>2</sup>

Note: NR-52512 does not include an antibiotic selection cassette for mammalian expression. Immunocytometry is required to identify cells expressing ACE2.

ACE2 is a human receptor expressed widely, including in the heart, kidney, small intestine and lung, and is involved in the regulation of hypertension and other cardiovascular diseases.<sup>4</sup> The SARS-related coronavirus 2 spike glycoprotein mediates viral binding to the host ACE2 receptor.<sup>5</sup> This protein forms a trimer, and when bound to ACE2 allows fusion of the viral and cellular membranes, allowing viral entry and replication. The interaction of ACE2 and the spike glycoprotein during viral infection is currently under study.<sup>4,5</sup>

**Material Provided:**

Each vial contains plasmid DNA in TE buffer (10 mM Tris HCl, 1 mM EDTA, pH 8.0). The DNA concentration and volume provided are shown on the Certificate of Analysis. The vial should be centrifuged prior to opening. Note: The contents of

the vial should be used to replicate the plasmid in *E. coli* prior to expression studies.

**Packaging/Storage:**

NR-52512 was packaged aseptically in screw-capped plastic cryovials. The product is provided frozen on dry ice and should be stored at -20°C or colder immediately upon arrival. Freeze-thaw cycles should be minimized.

**Citation:**

Acknowledgment for publications should read “The following reagent was obtained through BEI Resources, NIAID, NIH: Vector pHAGE2 Containing the Angiotensin-Converting Enzyme 2 Gene, NR-52512.”

**Biosafety Level: 1**

Appropriate safety procedures should always be used with this material. Laboratory safety is discussed in the following publication: U.S. Department of Health and Human Services, Public Health Service, Centers for Disease Control and Prevention, and National Institutes of Health. [Biosafety in Microbiological and Biomedical Laboratories](#). 5th ed. Washington, DC: U.S. Government Printing Office, 2009; see [www.cdc.gov/biosafety/publications/bmbl5/index.htm](http://www.cdc.gov/biosafety/publications/bmbl5/index.htm).

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**References:**

1. Bloom, J., Personal Communication.
2. Crawford, K. H. D., et al. "Protocol and Reagents for Pseudotyping Lentiviral Particles with SARS-CoV-2 Spike Protein for Neutralization Assays." *Viruses* 12 (2020): E513. PubMed: 32384820.
3. Murphy, G. J., et al. "Exogenous Control of Mammalian Gene Expression via Modulation of Translational Termination." *Nat. Med.* 12 (2006): 1093-1099. PubMed: 16892063.

4. Kai, H. and M. Kai. "Interactions of Coronaviruses with ACE2, Angiotensin II, and RAS Inhibitors- Lessons from Available Evidence and Insights into COVID-19." *Hypertens. Res.* 2020 Apr 27. PubMed: 32341442.
5. Hulswit, R. J. G., C. A. M. de Haan and B.-J. Bosch. "Coronavirus Spike Protein and Tropism Changes." *Adv. Virus Res.* 96 (2016): 29-57. PubMed: 27712627.

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**Figure 1: Plasmid Map of NR-52512**

