

***Borrelia burgdorferi*, Strain B31 (Clone 5A1)**

Catalog No. NR-13251

For research use only. Not for use in humans.

Contributor:

Steven J. Norris, Ph.D., Professor and Vice Chair for Research, Department of Pathology and Laboratory Medicine, University of Texas Health Science Center at Houston Medical School, Houston, Texas, USA

Manufacturer:

BEI Resources

Product Description:

Bacteria Classification: *Borreliaceae* (previously *Spirochaetaceae*)¹, *Borrelia*

Species: *Borrelia burgdorferi*

Strain: B31 (clone 5A1)

Original Source: *Borrelia burgdorferi* (*B. burgdorferi*), strain B31 (clone 5A1) was derived from the original B31 strain. The original B31 strain was isolated in 1981 from adult ticks (*Ixodes dammini/scapularis*) collected from lower vegetation on Shelter Island, New York, USA.^{2,3}

Comments: Clone 5A1 lacks linear plasmids lp5 and lp56 of the parent B31 strain but is known to retain the other nineteen plasmids found in strain B31.⁴ The complete genome of *B. burgdorferi*, Strain B31 has been sequenced (GenBank: [AE000783](#)).⁵

B. burgdorferi is a Gram-negative, motile spirochete.⁵ It is a zoonotic, vector-borne pathogen transmitted by ticks and the etiologic agent of Lyme disease, now the most common tick-transmitted disease in the United States.⁴ *B. burgdorferi* is predominant in North America, but also exists in Europe.

Material Provided:

Each vial contains approximately 0.5 mL of bacterial culture in Revised Barbour-Stoenner-Kelly broth supplemented with 10% glycerol.

Note: If homogeneity is required for your intended use, please purify prior to initiating work.

Packaging/Storage:

NR-13251 was packaged aseptically in cryovials. The product is provided frozen and should be stored at -60°C or colder immediately upon arrival. For long-term storage, the vapor phase of a liquid nitrogen freezer is recommended. Freeze-thaw cycles should be avoided.

Growth Conditions:

Media:

Revised Barbour-Stoenner-Kelly broth or agar or equivalent (Appendix I)

Note: Medium should be prepared fresh before each use.

Incubation:

Temperature: 32°C to 34°C (growth at 37°C may result in plasmid loss)²

Atmosphere: Microaerophilic (slower growth occurs under aerobic conditions)²

Propagation:

1. Keep vial in dry ice during inoculations.
2. Inoculate new cultures from scraping of frozen stock into a single tube of Revised Barbour-Stoenner-Kelly Medium.
3. Incubate the tube at 32°C to 34°C for 2 to 14 days. Do not shake culture during growth.

Note: Subculturing should be minimized to avoid plasmid loss.^{3,6}

Citation:

Acknowledgment for publications should read "The following reagent was obtained through BEI Resources, NIAID, NIH: *Borrelia burgdorferi*, Strain B31 (Clone 5A1), NR-13251."

Biosafety Level: 2

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 \(BMBL\)](#), 6th ed. Washington, DC: U.S. Government Printing Office, 2020.

Disclaimers:

You are authorized to use this product for research use only. It is not intended for human use.

Use of this product is subject to the terms and conditions of the BEI Resources Material Transfer Agreement (MTA). The MTA is available on our Web site at www.beiresources.org.

While BEI Resources uses reasonable efforts to include accurate and up-to-date information on this product sheet, neither ATCC® nor the U.S. Government makes any warranties or representations as to its accuracy. Citations from scientific literature and patents are provided for informational purposes only. Neither ATCC® nor the U.S. Government warrants that such information has been confirmed to be accurate.

This product is sent with the condition that you are responsible for its safe storage, handling, use and disposal. ATCC® and the U.S. Government are not liable for any damages or injuries arising from receipt and/or use of this product. While reasonable effort is made to ensure authenticity and reliability of materials on deposit, the U.S. Government, ATCC®, their suppliers and contributors to BEI Resources are not liable for damages arising from the misidentification or misrepresentation of products.

Use Restrictions:

This material is distributed for internal research, non-commercial purposes only. This material, its product or its derivatives may not be distributed to third parties. Except

as performed under a U.S. Government contract, individuals contemplating commercial use of the material, its products or its derivatives must contact the contributor to determine if a license is required. U.S. Government contractors may need a license before first commercial sale. This material may be subject to third party patent rights.

References:

1. Gupta, R. S., S. Mahmood and M. Adeolu. "A Phylogenomic and Molecular Signature Based Approach for Characterization of the Phylum *Spirochaetes* and Its Major Clades: Proposal for a Taxonomic Revision of the Phylum." *Front. Microbiol.* 4 (2013): 217. Erratum in: *Front. Microbiol.* 4 (2013): 322. PubMed: 23908650.
2. Burgdorfer, W., et al. "Lyme Disease – A Tick-Borne Spirochetosis?" *Science* 216 (1982): 1317-1319. PubMed: 7043737.
3. Norris, S. J., Personal Communication.
4. Fraser, C. M., et al. "Genomic Sequence of a Lyme Disease Spirochaete, *Borrelia burgdorferi*." *Nature* 390 (1997): 580-586. PubMed: 9403685.
5. Johnson, R. C., et al. "*Borrelia burgdorferi* sp. nov.: Etiologic Agent of Lyme Disease." *Int. J. Syst. Bacteriol.* 34 (1984): 496-497.
6. Purser, J. E. and S. J. Norris. "Correlation between Plasmid Content and Infectivity in *Borrelia burgdorferi*." *Proc. Natl. Acad. Sci. USA* 97 (2000): 13865-13870. PubMed: 11106398.
7. Barbour, A. G. "Isolation and Cultivation of Lyme Disease Spirochetes." *Yale J. Biol. Med.* 57 (1984): 521-525. PubMed: 6393604.
8. Kawabata, H., S. J. Norris and H. Watanabe. "BBE02 Disruption Mutants of *Borrelia burgdorferi* B31 Have a Highly Transformable, Infectious Phenotype." *Infect. Immun.* 72 (2004): 7147-7154. PubMed: 15557639.
9. Casjens, S., et al. "A Bacterial Genome in Flux: The Twelve Linear and Nine Circular Extrachromosomal DNAs in an Infectious Isolate of the Lyme Disease Spirochete *Borrelia burgdorferi*." *Mol. Microbiol.* 35 (2000): 490-516. PubMed: 10672174.

ATCC® is a trademark of the American Type Culture Collection.



APPENDIX I: REVISED BARBOUR-STOENNER-KELLY (BSK) MEDIUM

1. Prepare the Revised BSK Medium directly before each use following the recipe below by dissolving each component one at a time in distilled water:

HEPES	5.64 g
Neopeptone	4.7 g
Sodium citrate	0.7 g
Glucose	5.64 g
NaHCO ₃	2.0 g
TC-Yeastolate	2.0 g
Sodium pyruvate	0.75 g
N-acetylglucosamine	0.37 g
Bovine serum albumin, fraction V	47.0 g
Distilled water	840 mL

2. Adjust the pH of the base medium to 7.5 using 1 N HCl or 1 N NaOH and filter-sterilize using a 0.22 µm filter.
 3. Aseptically add the next two components to the base medium:

CMRL 1066 Medium, 10× (w/o Glutamine and NaHCO ₃)	100.0 mL
Heat-inactivated rabbit serum	60.0 mL

4. Mix well and aseptically dispense into appropriate vessels. The medium may be stored in aliquots of 50 mL in freezer-safe vessels and stored frozen at -20°C until use. Once thawed, each aliquot should be kept at 2°C to 8°C and used within one month.
 5. Adjust the pH of the complete medium to 7.5 to 7.6, as needed, using sterile solutions of 1 N HCl or 1 N NaOH, before use.

Note: Medium should be prepared fresh directly before each use or immediately aliquoted and frozen at -20°C until needed. Once thawed, each aliquot should be kept at 2°C to 8°C and used within one month.