

***Borrelia burgdorferi*, Strain B31 (Clone 5A2)**

Catalog No. NR-13252

For research use only. Not for use in humans.

Contributor:

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Manufacturer:

BEI Resources

Product Description:

Bacteria Classification: *Spirochaetaceae*, *Borrelia*

Species: *Borrelia burgdorferi*

Strain: B31 (clone 5A2)

Original Source: *Borrelia burgdorferi* (*B. burgdorferi*), strain B31 (clone 5A2) 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.^{1,2}

Comments: Clone 5A2 lacks linear plasmids lp5, lp28-1 and lp56 and circular plasmid cp9 of the parent B31 strain.² The complete genome of *B. burgdorferi*, B31 has been sequenced (GenBank: [AE000783](https://www.ncbi.nlm.nih.gov/nuccore/AE000783)).³

B. burgdorferi is a Gram-negative, motile spirochete.⁴ 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.³ *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-13252 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 equivalent (Appendix I)

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

Note: Medium should be prepared fresh before each use.

Incubation:

Temperature: 32°C to 34°C

Note: Growth at 37°C may result in plasmid loss.²

Atmosphere: Microaerophilic

Note: 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 5 to 10 days.

Note: Subculturing should be minimized to avoid plasmid loss.^{2,5}

Citation:

Acknowledgment for publications should read "The following reagent was obtained through BEI Resources, NIAID, NIH: *Borrelia burgdorferi*, Strain B31 (Clone 5A2), NR-13252."

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. 6th ed. Washington, DC: U.S. Government Printing Office, 2020; see www.cdc.gov/biosafety/publications/bmbl5/index.htm.

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References:

1. Burgdorfer, W., et al. "Lyme Disease – A Tick-Borne Spirochetosis?" Science 216 (1982): 1317-1319. PubMed: 7043737.
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3. Fraser, C. M., et al. "Genomic Sequence of a Lyme Disease Spirochaete, *Borrelia burgdorferi*." Nature 390 (1997): 580-586. PubMed: 9403685.
4. Johnson, R. C., et al. "*Borrelia burgdorferi* sp. nov.: Etiologic Agent of Lyme Disease." Int. J. Syst. Bacteriol. 34 (1984): 496-497.
5. 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.

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6. Barbour, A. G. "Isolation and Cultivation of Lyme Disease Spirochetes." Yale J. Biol. Med. 57 (1984): 521-525. PubMed: 6393604.
7. 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.
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APPENDIX I: REVISED BARBOUR-STOENNER-KELLY 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.