Summary of Changes Included in the Full Protocol Amendment of:

IMPAACT 2011
Phase I Placebo-Controlled Study of the Infectivity, Safety and Immunogenicity of a Single Dose of a Recombinant Live-Attenuated Respiratory Syncytial Virus Vaccine, LID ΔM2-2 1030s, Lot RSV#010A, Delivered as Nose Drops to RSV-Seronegative Infants 6 to 24 Months of Age

Version 1.0, dated May 3, 2016
DAIDS ES # 30072
IND # 16990

The Amended Protocol is identified as:

Version 2.0, dated November 9, 2016

Information/Instructions to Study Sites from the Division of AIDS

The information contained in this protocol amendment affects the IMPAACT 2011 study and must be submitted to site Institutional Review Boards (IRBs) as soon as possible for their review and approval. Approval must also be obtained from site regulatory entities if applicable per the policies and procedures of the regulatory entities. All IRB and regulatory entity requirements must be followed, and all required approvals of this amendment must be obtained before initiating this study.

Upon obtaining IRB approval and any other applicable regulatory entity approvals, each site should immediately begin implementing this amendment. Unless directed by site IRBs, re-consenting is not required for current study participants.

Sites are required to submit an amendment registration packet to the DAIDS Protocol Registration Office (DAIDS PRO) at the Regulatory Support Center (RSC). Sites will receive a registration notification for the amendment after the DAIDS PRO verifies that all required registration documents have been received and are complete. Sites should not await this notification before implementing this amendment.

This Summary of Changes, Version 2.0 of the protocol, and all associated IRB and regulatory entity correspondence should be retained in each site’s essential document files for IMPAACT 2011.
Summary of Modifications and Rationale

The primary purposes of this full version amendment are to:
1. Remove Table 11 from Appendix I due to intellectual property issues and renumber subsequent tables.
2. Include a provision in Section 9.3.1 to permit replacement of participants who discontinue early.
3. Change references to “PI” to “Protocol Chair” in Section 9.4.2 for consistency with the rest of the protocol.
4. Update hyperlinks to online resources available from DAIDS.
5. Include changes implemented via Clarification Memorandum #1, dated 10 August 2016.
6. Update contact information in the site roster.

Detailed Listing of Modifications

The modifications included in this summary of changes are listed below in order of appearance in the protocol. Additions to the text are indicated in bold; deletions are indicated by strikethrough. Unless otherwise stated, section and page numbers reflect the current version of the protocol. Changes are noted in the order of appearance.

1. Title page. The IND number was added, as in Clarification Memorandum #1.

2. Throughout the protocol, the version number was updated to Version 2.0, and the version date was updated to 9 November 2016.

3. The table of contents was updated to reflect modifications in the protocol.

4. Protocol Team Roster (page 8). Updated as per Clarification Memorandum #1 and to reflect current contact information for additional team members, as shown below.

   NICHD Medical Officer
   Jack Moye, Jr., MD
   Eunice Kennedy Shriver National Institute of Child Health & Human Development
   National Institutes of Health,
   DHHS Bldg. 6100 Rm. 3A01 MSC 7510 6710B Rm. 2146 MSC 7002
   6100 Executive Blvd Rockville 6100 Executive Blvd Rockville
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   Clinical Research Oversight Managers
   […]
   Susan Vogel, RN, BSN
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   Bethesda, MD 20817 20892
   Phone: 301-451-5138 240-669-5224
   Email: svogel@niaid.nih.gov
5. **Site Roster (page 11).** The site investigator at site 5030 was updated.

   Site 5030, Emory University School of Medicine  
   **Andres Camacho-Gonzalez, MD, MSc. Paul Spearman, MD**  
   Emory University  
   2015 Uppergate Drive  
   Atlanta, GA 30322  
   Phone: 404-727-5642  
   Email: acamac@emory.edu paul.spearman@emory.edu

6. **Section 1.2, Background (page 20), second paragraph under Vaccine Description.** Reference to Appendix I, Table 11 was removed; this table was deleted due to intellectual property issues.

   A comparison of the genomes of LID ΔM2-2 1030s and wt RSV is shown in Appendix I, Table 10, and a comparison with the previous RSV vaccine candidate RSV MEDI ΔM2-2 is shown in Appendix I, Table 11.

7. **Section 1.3.2, Preclinical Studies (page 22).** References to the former Table 11 were removed, and Tables 12, 13, and 14 were renumbered as Tables 11, 12, and 13 in the following sections.

   a) **Evaluation of the Attenuation Phenotype of LID ΔM2-2 1030s in Nonhuman Primates (page 24), second to last sentence:**

   The first NHP study was done in AGMs to evaluate an Experimental Lot of LID ΔM2-2 1030s in comparison to the previously tested RSV vaccine candidate LID ΔM2-2, and 2 additional further attenuated M2-2 deletion mutants (total dose administered: 2 x 10⁶ PFU per animal) (Appendix I, Table 11, **Table 11**, **Table 11**, and **Table 13**, **Table 12**).

   b) **Replication and Immunogenicity of an Experimental Lot of LID ΔM2-2 1030s in AGMs (Dose: 2 x 10⁶ PFU) (page 25), second paragraph, first sentence:**

   By plaque assay and by TaqMan assay, substantial shedding of the RSV LID ΔM2-2 control virus was detectable from the upper and lower respiratory tract over several days, with mean peak titers of 2.9 log₁₀ PFU per mL in the URT, and 4.2 log₁₀ PFU per mL in the LRT (Appendix I, **Table 11**, **Table 12**, and **Table 13**, **Table 12**; Figure 3).

   c) **Replication and Immunogenicity of an Experimental Lot of LID ΔM2-2 1030s in AGMs (Dose: 2 x 10⁶ PFU) (page 25), second paragraph, third sentence:**

   Remarkably, despite a lower level of virus replication, LID ΔM2-2 1030s induced moderate serum neutralizing antibody titers (Appendix I, **Table 13**, **Table 14**) in two of four AGMs.

8. **Section 2.1, Primary Objectives (page 29).** Objective reworded for consistency with Schema.
2.1.1 Safety: To assess the frequency and severity of study product-related solicited and unsolicited adverse events (AEs), from Study Day 0 through midnight of the 28th day following inoculation, in vaccinated participants.

9. Exclusion criterion 4.2.23 (page 35). Note added as per Clarification Memorandum #1.

10. Section 5, Study Product Considerations (page 36). Undiluted concentration of study product updated in the following sections as per Clarification Memorandum #1.
   
   a) Section 5.3.1, Vaccine (page 37), first sentence
   
   b) Section 5.5.3, Live Recombinant Respiratory Syncytial Virus (RSV) LID ΔM2-2 1030s (page 39), third paragraph, third sentence

11. Section 6.12, Additional Considerations for Laboratory Procedures (page 49), first sentence. Hyperlink to DAIDS Funded and/or Sponsored Laboratories in Clinical Trials Policy updated.

   Each study site and laboratory involved in this study will comply with the DAIDS policy on Requirements for DAIDS Funded and/or Sponsored Laboratories in Clinical Trials Policy, which is available at:
   
   https://www.niaid.nih.gov/research/daids-clinical-research-laboratory-specimens-management
   
   http://www.niaid.nih.gov/labsandresources/resources/daidsclinrsrch/Pages/Laboratories.aspx

12. Section 7.3.1, Adverse Event Reporting to DAIDS (page 54). Hyperlinks to DAIDS EAE Manual and DAIDS EAE Form updated in the following sections.
   
   a) First paragraph:

   Requirements, definitions, and methods for expedited reporting of adverse events (AEs) are outlined in Version 2.0 of the DAIDS EAE Manual, which is available on the RSC website at http://rsc.tech-res.com/clinical-research-sites/safety-reporting/manual
   
   b) Third paragraph, second sentence:

   This form is available on the RSC website: http://rsc.tech-res.com/clinical-research-sites/safety-reporting/daids/paper-eae-reporting

13. Section 7.3.3, Grading Severity of Events (page 54), first paragraph, last sentence. Hyperlink to DAIDS AE Grading Table updated.

   The DAIDS AE Grading Table is available on the RSC website at http://rsc.tech-res.com/docs/default-source/safety/daids_ae_grading_table_v2_nov2014.pdf?sfvrsn=8
   

14. Section 9.3.1, Sample Size and Randomization (page 61), end of first paragraph. New sentence added to permit replacement of participants who discontinue early.
In the event that a participant is discontinued early from the study and the team decides that additional data would be needed to answer the study objectives, an additional participant may be enrolled in the same treatment arm as the discontinued participant.

15. Section 9, Statistical Considerations (page 61). Updates to one-sided Fisher’s Exact test made in the following sections as per Clarification Memorandum #1.
   a) Section 9.3.1, Sample Size and Randomization, sixth paragraph, second sentence
   b) Section 9.3.1, Table 9: Magnitude of Difference in Responses Detectable with 80% Power
   c) Section 9.3.1, Figure 6: Power curves for comparisons between vaccine and placebo responses
   d) Section 9.5.1, Assessment of Primary Objectives, fifth paragraph, second sentence

16. Section 9.4.2, Monitoring by the NIAID Intramural Data and Safety Monitoring Board (page 65), second to last and last sentences. References to “PI” were changed to “Protocol Chair” for consistency with the rest of the protocol.

The Protocol Chair will notify the DSMB of any cases of intentional or unintentional unblinding as soon as possible. The Protocol Chair will notify the Board at the time pausing or halting criteria are met and obtain a recommendation concerning continuation, modification, or termination of the study.


All DAIDS policies referenced in this section are available at:
https://www.niaid.nih.gov/research/daids-clinical-research-policies-standard-procedures
www.niaid.nih.gov/labsandresources/resources/daidsclinrsrch/Pages/ClinicalSite.aspx


All DAIDS policies referenced in this section are available at:
www.niaid.nih.gov/labsandresources/resources/daidsclinrsrch/Pages/ClinicalSite.aspx

19. Section 10.4, Quality Control and Quality Assurance (page 68), first paragraph. Hyperlink to DAIDS policy on Requirements for Clinical Quality Management Plans updated.

Study sites must ensure that essential documents and participant research records are subject to continuous quality control and quality assurance procedures consistent with the DAIDS policy on Requirements for Clinical Quality Management Plans, which is available at:
www.niaid.nih.gov/labsandresources/resources/daidsclinrsrch/Pages/ClinicalSite.aspx

Study sites must comply with the requirements of the DAIDS policy on Enrolling Children (including Adolescents) in Clinical Research, which is available at:


For additional information on the protocol registration process and specific documents required for initial and amendment registrations, refer to the current version of the DAIDS Protocol Registration Manual, which is available on the RSC website:
http://rsc.tech-res.com/clinical-research-sites/protocol-registration/

22. Section 13.4, Protocol Deviation Reporting (page 73), second paragraph. Hyperlink to DAIDS policies updated.

All DAIDS policies referenced in this section are available at:
https://www.niaid.nih.gov/research/daids-clinical-research-policies-standard-procedures

23. Section 15, References (page 75), number 16. Reference updated for consistency with standard format.

24. Appendix I, Table 11 (page 79). Original Table 11 removed from protocol due to intellectual property issues.

Table 11: Comparison of Genomic Sequences of wt rRSV A2 D46 (GenBank Accession Number KT992094), RSV MEDI ΔM2-2 (Lot RSV#002A), and LID ΔM2-1030s (Lot RSV#010A)

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<th>Gene Region</th>
<th>RSV Nucleotide (cDNA)</th>
<th>Encoded Amino Acid Residue</th>
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<td>Wild Type D46 rRSV</td>
<td>Wild Type D46 cDNA</td>
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<tr>
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Summary of Changes to IMPAACT 2011 Page 7 of 9 From Protocol Version 1.0 to Version 2.0
### Summary of Changes to IMPAACT 2011 Page 8 of 9
From Protocol Version 1.0 to Version 2.0

**November 2016**

From Protocol Version 1.0 to Version 2.0

<table>
<thead>
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**Note:**

- Genomic position in reference to wt RSV A2 D46 (19). GenBank Accession Number KT992094). Relative to this standard, the RSV MEDI AM2-2 cDNA contains a single nucleotide deletion at position 1099 (which is incidental to its phenotype and occurs because the D46 cDNA contains an engineered 1 nt insertion at this site, (19)). The RSV MEDI AM2-2 cDNA also contains a 234 nt-deletion of the M2-2 ORF (KT992094 nt 8202-8435); The LID AM2-2 1030s cDNA contains a 112 nt deletion of the SH noncoding region (KT992094 nt 4109-4610) and a 241 nt deletion of the M2-2 ORF (KT992094 nt 8189-8429), but for simplicity the numbering of all sequence positions is based on the complete sequence of wt recombinant strain A2 D46 (19). All sequences are positive sense.

- **2.** nucleotide dimorphism: ~25% of the total population has A residue, ~75% of the total population has G residue

- **3.** ~25% of the total population has serine assignment; ~75% of the total population has glycine assignment.

- **4.** On the amino acid level, apart from differences in the ∆M2-2 deletion, LID ∆M2-2 1030s differs from RSV MEDI ∆M2-2 at three codons as indicated: codon 51 in NS2; codon 24 in N; and codon 1321 in L, the latter being the "1030s" mutation.

- **5.** nucleotide dimorphism: ~50% of the total population has C residue, ~50% of the total population has T residue.

- **6.** nucleotide dimorphism: ~50% of the total population has T residue, ~50% of the total population has G residue

- **7.** nucleotide dimorphism: ~50% of the total population has T residue, ~50% of the total population has G residue

- **8.** ~50% of the total population has asparagine assignment, ~50% of the total population has lysine assignment.

- **9.** Three possible translation initiation codons of the M2-2 ORF were removed by mutagenesis, silent on the amino acid level of the overlapping reading frame of M2-1.

- **10.** This sequence is missing in LID AM2-2 1030s because that virus has a longer M2-2 deletion that includes these positions.

- **11.** Differences between LID AM2-2 1030s and MEDI AM2-2 that are due to incidental mutations in the two backbones (arising from differences in the wt viruses and wt cDNAs from which both were derived as well as several restriction sites that were engineered in the original D46 cDNA, (19)).

- **12.** Mutations added to stabilize the LID AM2-2 1030s cDNA in E. coli.

Light shading indicates sequence positions involved in the AM2-2 mutation and the 1030s mutation in LID AM2-2 1030s.
25. Appendix I, Tables 12, 13, and 14 (page 79), Tables renumbered to account for removal of Table 11.

Table 11-12: Viral titers of nasopharyngeal swab samples from AGMs inoculated with LID ΔM2-2, RSV ΔSHΔM2-2, LID ΔM2-2 1030s, or RSV cp ΔSHΔM2-2a

Table 12-13: Viral titers of tracheal lavage samples from AGMs inoculated with LID ΔM2-2, RSV ΔSHΔM2-2, LID ΔM2-2 1030s, or RSV cp ΔSHΔM2-2a

Table 13-14: Neutralizing antibody titers of AGMs inoculated with LID ΔM2-2, RSV ΔSHΔM2-2, LID ΔM2-2 1030s, or RSV cp ΔSHΔM2-2a