ALEXION

August 22, 2012

Ann Farrell, M.D.
Acting Director, Division of Hematology Products (HFD 160)
Food and Drug Administration
Center for Drug Evaluation and Research
5901-B Ammendale Road
Beltsville, MD 20705-1266

Re: Soliris® (eculizumab)
Response to Form FDA 483
BLA #125166/172
Sequence #0343

Dear Dr. Farrell:

Reference is made to Alexion's biennial inspection conducted at the Alexion Rhode Island Manufacturing Facility (ARIMF) of Smithfield, Rhode Island (FEI 3006568549) on the dates of July 12, 16-18, 20, 24-26 and August 6, and also to the Form FDA 483 issued on August 6, 2012.

Attached please find Alexion's responses to the observations listed on the Form FDA 483.

As requested by Megan A. Haggerty (Investigator) during the August 6th close-out meeting, a copy of the Alexion responses is also being sent to the District Director in Stoneham, Massachusetts.

If there are any questions regarding this submission, please contact Cornelius Dunn directly at (203) 271-8390 or me (the undersigned) at (203) 271-8334 or by email at lyonsm@alxn.com.

Sincerely,

Mary F. Lyons, RAC
Senior Manager, Regulatory Affairs
Alexion Pharmaceuticals, Inc.

cc: Capt. Mutahar Shamsi, 1 Montvale Avenue, 4th Floor, Stoneham, MA 02180

Observation 1:

The firm's investigations into Soliris (b) (4) *microbiological contamination events are inadequate. For example:*

a. *Risk assessments* (b) (4) *and* (b) (4) *for release of Soliris drug substance lot* (b) (4) *(drug product lots A75445C and A75445D), and the release for forward processing of Soliris drug substance lot* (b) (4) *are not sufficient justification alone for lot release. Soliris lot* (b) (4) *contained a count of TNTC (too numerous to count)/10 mL at the* (b) (4) *step in April 2011 and Soliris lot* (b) (4) *contained counts of TNTC/10 mL and 40 cfu/1 mL at the* (b) (4) (b) (4) *step in March 2012. The isolates were identified as Bacillus thuringiensis and Acinetobacter radioresistens for lo* (b) (4) *nd as Bacillus thuringiensis for lot* (b) (4) *The potential impurities generated, i.e. non-host cell by-products, were not quantified and the process impurity clearance was not calculated. Specific analytical testing beyond routine release and stability has not been performed for these Soliris drug substance lots to verify whether potential impurities, i.e. non-host cell by-products, generated at the* (b) (4) *step were removed during the purification process.*

Additionally, a BPDR has not been filed for Soliris drug substance lot (b) (4) *(drug product lots A75445C and A75445D), as it exceeded its filed specification of* (b) (4) (b) *at the* (b) (4) *with a result of TNTC/10 mL.*

Alexion Response:

Risk assessment SOP QC-0394, "Bioburden Microbial Risk Management & Assessment" (b) (4)
(b) (4)
Risk assessments include an evaluation of the known facts related to the processing events, in process/bulk drug substance bioburden and endotoxin results, the quality of the drug substance determined from release test results, and the mechanisms of the purification steps used in the eculizumab manufacturing process.

Risk assessments (b) (4) and (b) (4) concluded that the risk to the eculizumab product quality was low. The information assessed per SOP QC-0394 and additional testing performed include:

1. Neither *Bacillus thuringiensis* nor *Acinetobacter radioresistens* are considered common human pathogens.

2. No impact to in-process material or processing equipment based on the test results from the routine in-process samples (provided below). The values for bioburden and endotoxin were all lower than the action limits:

    a. The bioburden result of post-filtered (b) (4) (b) (4) was 0 cfu/10mL (reported as <1 cfu/10mL)

b. The bioburden results for the pre-(b) (4) (b) (4) were all 0 cfu/10mL (reported as <1 cfu/10mL).

c. The endotoxin results for the pre(b) (4) (b) (4) were all <1.25 EU/mL, which is the limit of quantitation for this sample.

d. The bioburden and endotoxin results for the(b) (4) were reported as <1 cfu/10mL and <0.0625 EU/mL, which is the limit of quantitation for this sample.

e. The final bulk drug substance bioburden and endotoxin results met specifications with results of 0 cfu/10mL and <0.1 EU/mg, which is the limit of quantitation for this sample.

3. Bulk drug substance release, stability, and non-routine characterization testing show no unexpected results:

a. The bulk drug substance for Soliris lots(b) (4) and(b) (4) met all release criteria. Additionally, both lots are on(b) (4) stabili(b) (4) have met all stability criteria, and have shown no adverse trends. Drug substance lot (b) (4) was placed on accelerated stability and showed no unexpected results. The(b) (4) (b) (4) and (b) (4) b) (4) test result data are provided in Attachment 1a-1.

b. Drug substance lot(b) (4) was subjected to intact(b) (4) (b) (4) nd showed no unexpected results. Drug Substance lot (b) (4) was subjected t(b) (4) on August 10, 2012, after approval of(b) (4). The major component (b) (4) of the (b) (4) The overall(b) (4) features are consistent with eculizumab bulk drug substance with no additional unknown features. These data can be made available upon request.

c. Drug substance from lot (b) (4) was subjected t(b) (4) (b) (4) (b) (4) No unexpected results were obtained and the drug substance was determined to be comparable to lots of drug substance that did not experience the bioburden event at the(b) (4) step. Alexion commits to completion of the following analytical tests on drug substance lo(b) (4) by **October 1, 2012**: (b) (4) (b) (4) (b) (4)

d. Lo(b) (4) in-process samples were subjected to(b) (4) (b) (4) testing. Lot(b) (4) was subjected t(b) testing.

No unexpected(b) (4) were observed that would suggest the presence of bacterial contaminants. The results were consistent with other results from lots that did not experience th(b) (4) step.

4. Based on the subsequent(b) (4) and(b) (4) in-process steps, the risk assessments also demonstrated that the likelihood of foreign proteins or low molecular weight molecules reported to be produced by *Bacillus thuringiensis* or *Acinetobacter radioresistens* being co-purified with eculizumab is low. See Attachment 1a-2 for a review of the purification process.

Further Assessment of Risk

In addition to the elements assessed in SOP QC-0394 and the additional testing discussed above, potential impurity quantity and clearance were calculated, toxicology assessment of calculated impurities completed, and adverse event information was evaluated for lot(b) (4) Lot (b) (4) is in Quarantine status and has not been filled into drug product.

Worst Case Calculations

Worst case calculations were performed to estimate the quantity of potential impurities generated and the clearance of the potential impurities. In order to perform an analysis of potential contaminant removal following a bioburden excursion in(b) (4) material, it is necessary to first establish(b) (4) The in-process bioburden testing o(b) (4) was too numerous to count (TNTC) for a 10 mL sample(b) (4) The(b) (4) unit operation for(b) (4) ook approximately(b) (4) Our working assumption is that a small number of bacteria were either present in the system or were introduced during the(b) (4) process, and this small inoculum grew during the(b) (4) of(b) (4) (b) (4) An assumption was made tha(b) (4) of the bacteria were introduced into the system. Using a(b) (4) for *Bacillus thuringiensis* results in a bioburden loa(b) (4) The reported(b) (4) for *Acenitobacter radioresistens*(b) (4) Therefore, use of the(b) (4) of *Bacillus thuringiensis* represents worse-case. The dry weight of a bacterial cell (*Escherichia coli*) has been reported to range fron(b) (4) as a worst case(b) (4) To provide a worst case scenario, it was assumed all of this mass was potential protein impurities. During the eculizumab process validation there was a(b) (4) Applying that clearance to the(b) (4) (b) (4) into the bulk drug substance. Drug substance lo(b) (4) esulting in a concentration of(b) pg protein/mL. A maximum 120 mL dose of Soliris would contain no more than 15 pg of the contaminant protein as a worst case estimation. (See Attachment 1a-3 for detailed calculation). While this represents a theoretical worst case calculation, the process removal data for the eculizumab process indicates that the actual removal is several orders of magnitude higher as indicated in the following paragraph.

The worst case assessment presented above (b) (4)
(b) (4)
by the amount (b) (4) The clearance of other
impurities (b) (4)
(b) (4) was
determined (b) (4)
amount (b) (4)
(b) (4)
(b) (4) (b) (4)
purification process (b) (4)
require (b) (4) which would be
expected t (b) (4)
step requires (b) (4) which would be
expected to (b) (4)
resulting in an overall (b) (4) from the (b) (4) steps.
These values suggest a potential impurity clearance across the eculizumab manufacturing
process o (b) (4)
(b) (4)

The risk assessment (b) (4) for lo (b) (4) will be amended to include all release, stability, and additional testing results as well as the potential impurities worst case calculation. The amendment will be completed by **October 19, 2012**.

Toxicology Assessment of Calculated Impurities

Based on a "worst case" calculation, eculizumab drug substance lot (b) (4) had a maximum of <15 pg/120mL standard human dose (<0.3 pg/kg assuming 50kg human) of *Bacillus thuringiensis* and *Acinetobacter radioresistens* associated proteins in the final drug product. The potential toxicity of *Bacillus thuringiensis* in mammals is associated with administration of 0.77 mg/kg or more of the purified *Bacillus thuringiensis* proteins.

The amount of *Bacillus thuringiensis* toxins and *Acinetobacter radioresistens* in the eculizumab lot (b) (4) were far below any concentrations reported to be associated with any signs or evidence of toxicity or other findings. *Bacillus thuringiensis* proteins have been associated with toxicity either as a component of a commercially-available insecticide preparation (purified and at high exposures), or when administered orally at very large doses. The purified native endotoxin isolated from this species of bacteria was associated with toxicity when administered parenterally at large doses. *Acinetobacter radioresistens* only was an opportunistic pathogen when an immune-compromised person was infected by whole, live organisms.

The measured eculizumab lot (b) (4) final bioburden of 0 CFU/10 mL and <0.1 EU/mg and negative endotoxin findings, estimated impurities from bioburden at ≤15 pg of *Bacillus thuringiensis* per 120 mL dose of eculizumab and total possible human dose level of a range of 3-8 total doses, was assessed in conjunction with the literature search on toxic effects of both bacteria and bacterial components or protein products.

The very low concentrations of bacterial protein component contamination that have been calculated as possibly present in the lot, are much lower than the large doses of *Bacillus thuringiensis* proteins associated with any adverse effects in any *in vivo* or *in vitro* studies. Risk assessments of crops that express far higher concentrations of *Bacillus thuringiensis* toxin than any total human exposure from eculizumab lot (b) (4) concluded that crops expressing *Bacillus thuringiensis* toxin posed no risk to workers exposed to the crops or to consumers of the crops (Carstens 2012, Alink 2008). Therefore, the risk of clinical adverse events associated with administration of this lot of eculizumab is considered to be very low to negligible.

Adverse Event Evaluation

The assessment of the postmarketing safety database for 01 October 2011 to 26 July 2012, which coincides with the distribution of drug product lot A75445, and the previous two periods, 01 April 2010 to 31 December 2010 and 01 January 2011 to 30 September 2011, does not identify any safety concern for drug product Lot A75445 (from bulk drug substance lot (b) (4) as regards lack of efficacy, infusion related reactions and bacillus infection. Additionally, a review of the identified cases does not support a safety finding for either the lot A75445 or Soliris overall in the US, as the cases are few in each category, often not well documented, do not follow a temporal association with the release of the lot A75445 and are confounded by complex medical course, concurrent illnesses, diagnostic difficulties, numerous treatments, time of Soliris treatment initiation, length of Soliris treatment, and individual patient drug clearance.

The risk assessment conducted for lot (b) (4) per SOP QC-0394 concluded that the risk to the eculizumab product quality was low. Further assessment of risk through an estimation of potential impurity quantity and clearance, toxicology assessment and a review of adverse event information support the initial conclusion of low risk.

Alexion recognizes that FDA expects, in instances when in-process bioburden action limits are exceeded, the corresponding risk assessment should quantify the potential impurities generated, i.e. non-host cell by-products, and the process impurity clearance calculated. In addition, specific analytical testing beyond routine release and stability should be performed for the corresponding Soliris drug substance lots to verify whether potential impurities generated were removed during the purification process.

Alexion will assess analytical methodologies and procedures to ensure expectations for risk assessments are met. Future risk assessments utilized for favorable disposition of lots that experience an in-process bioburden action limit excursion will include information to address FDA expectations.

Alexion will complete a revision of SOP QC-0394, "Bioburden Microbial Risk Management & Assessment" by **October 31, 2012** that will include the following requirements:

- Completion of risk assessment for lot discarded due to bioreactor contamination

- Assessment, and attachment of raw data, (b) (4) results for any action level excursion from (b) (4)
- Place lots intended for release with any action level excursion from (b) (4) on stability if not already designated for stability testing
- Assessment of defined process impurity removal against historical data for any action level excursion from (b) (4)
- Assessment of testing required, beyond current in-process and release tests, to adequately complete risk assessment. Completion of the identified testing is required to support favorable lot disposition.
- For action level excursion events from (b) (4) document a worst-case carry through of potential bioburden-related impurities

BPDR

A BPDR has been filed concurrent with the submission of these 483 observation responses (as BLA Sequence 342). The BPDR is for Soliris drug product lots A75445C and A75445D produced from eculizumab drug substance lo (b) (4) vials of drug product lots A75445C and A75445D have been consumed. The remaining inventory has been placed in blocked status. Follow-up to the BPDR submission will include the additional (b) (4) bulk drug substance testing to be completed **October 19, 2012**.

Observation 1 (continued):

b. *In the past 15 months, the Soliris drug substance (b) (4) process has experienced 7 contamination events. Five contamination events occurred in 3 bioreactors and (b) (4) (b) (4) contamination events occurred at the (b) (4) The firm has not adequately investigated to determine whether these contamination events are linked and has not adequately prevented reoccurrence of microbial contamination in the Soliris (b) (4) manufacturing process.*

Three bioreactor microbial contamination events occurred between October 2011- January 2012 for lots (b) (4) Two (b) (4) contamination events occurred in April 2011 for lot (b) (4) and in March 2012 for lot (b) (4) Each aforementioned contamination event isolated Bacillus thuringiensis as the contaminating microorganism, however the QC Microbiology Department has not confirmed whether the bioreactor contamination events and the (b) (4) contamination events were the same strain; only two of the bioreactor contamination events were analyzed for, and confirmed as the same strain.

Two additional bioreactor contamination events occurred in July 2012 for eculizumab lots (b) (4) and (b) (4) lot (b) (4) was contaminated with Lysinibacillus boronitolerans and lot (b) (4) was contaminated with Bacillus thuringiensis. Investigations are ongoing for these events.

In all 5 of the bioreactor microbial contamination events, bioreactors (b) (4) (b) (4) and/or (b) (4) were in use when the contamination occurred. Although the firm believes the cause of the three October 2011- January 2012 bioreactor microbial contamination events were due to faulty high-high limit switches, the firm has not ruled out whether there is another root cause for the contamination events. In the (b) (4) (b) (4) microbial contamination events, three potential root causes were identified but a definitive root cause was not confirmed.

Alexion Response:

Alexion completed investigation of the (b) bioreactor and (b) (4) contamination events per SOP TMS-0027, "Bioreactor Contamination Investigation" and SOP TMS-0028, (b) (4) and Purification Equipment Investigation." The procedures require investigations to be completed by a cross-functional team of subject matter experts from Technical Services, Manufacturing, Facilities, Quality Control and Quality Assurance. Investigations conducted per this SOP are systematic inquiries and examinations of factors to determine root cause(s) or the most probable root cause(s). The procedure requires a review of manpower, material, method, equipment and environmental factors. Activities that are reviewed include equipment (b) (4) (b) (4) (b) (4) and (b) (4)

(b) (4) Events

The (b) (4) deviation investigations in April 2011 and in March 2012 identified most probable root causes related (b) (4) operation procedures. Specifically, the potential cause for the April 2011 event was an insufficient procedure that did not identify the potential impact of residual WFI in a (b) (4) (b) (4) and a lack of (b) (4) prior to use. Preventative actions included procedure modification. The potential causes identified for the March 2012 event were contamination of the (b) (4) system from system drain during repeated (b) (4) integrity testing and lack of (b) (4) (b) integrity during (b) (4) (b) (4) (b) (4). The potential loss of (b) (4) integrity was not associated with a (b) (4) prior to use as occurred in April 2011. Preventative actions included procedural changes associated with (b) (4) integrity testing and venting of the (b) (4) to prevent pressure increase. The processing step (b) (4) associated with the (b) (4) operations (b) (4) operation. Also, the processing equipment used for the (b) (4) process is not equipped with the (b) (4) identified in the bioreactor investigation events discussed below.

Bioreactor Events, October 2011 and January 2012

Investigations of each bioreactor microbial contamination events between October 2011 and January 2012 (lots (b) (4)) were conducted. A root cause was not identified for lot (b) (4) and a sterile boundary breach associated with a (b) (4) (b) was identified for lo (b) (4). Investigation of lo (b) (4) identified a faulty high-high limit switch which would allow compromise of the sterile boundary. The commonality of these switches on all bioreactors was investigated and linked as a potential cause to the previous contaminations (Investigation summary report (b) (4) for lot (b) (4) Attachment 1b-1). Removal of the limit switches prevented contamination reoccurrence as (b) (4) consecutive production bioreactor runs were successful after removal.

Bioreactor Events, July and August 2012

The investigation of the bioreactor contamination events that occurred during the 2012 GMP inspection is ongoing. Alexion has engaged (b) (4) consulting firms to support the investigation. Production in the bioreactors has been suspended to support completion of investigational and corrective actions. Initial findings suggest ineffective routine CIP of non-routine soils contributed to ineffective SIP. Non-routine soils were produced due to a prolonged delay in manufacturing for preventative maintenance. Each bioreactor contamination experienced in July and August 2012 was initiated after the prolonged delay. All evidence uncovered to date indicates that the causes of the most recent contaminations are not related to the root cause previously identified for the October 2011 to January 2012 events. Alexion will provide a report of the investigation to FDA once it is complete. The reports (or an update if the reports are not completed) will be sent to FDA by **October 1, 2012**.

The data collected during all investigations do not indicate a link between the three bioreactor microbial contamination events between October 2011-January 2012 and two (b) (4) (b) (4) contamination events in April 2011 and in March 2012. Alexion will document an

assessment of the root causes identified for the five previous contamination events (three bioreactor and two (b) (4) against the findings of the investigations of the July and August 2012 events to confirm the adequacy of previous root cause determinations. The assessment will also determine if commonalities exist between the previous and recent events that necessitate additional preventative actions. In addition, available strains from the five previous contamination events will be compared to the organisms isolated from the recent events including environmental isolates. The results will be used to help substantiate and exclude potential root causes for the recent events.

Observation 1 (continued):

c. *The adequacy and effectiveness of the routine CIP cycle has not been evaluated as part of the bioreactor contamination events, for example where Bacillus thuringiensis has been isolated. The cycle was validated with the intent for use in routine cleaning; it is not validated or effective for when bioreactors are subjected to bioburden exceeding upper in-process specification limits or for post decontamination events. For example:*

1. *Following a confirmed Bacillus thuringiensis contamination in bioreactors* (b) (4) *and* (b) (4) *for Soliris lo*(b) (4) *on January 14, 2012, a routine CIP cycle was executed for* (b) (4) *on January 18, 2012. Although the post-CIP visual inspection indicated a passing result on January 18, 2012, a bioreactor entry confirmed the CIP cycle was not effective as a soil was observed at the liquid level of the bioreactor. The firm has not evaluated the effectiveness of their routine CIP cycle with bioburden counts that exceed upper in-process specification limits.*

2. *Following a confirmed Bacillus thuringiensis contamination in bioreactor* (b) (4) *for Soliris lo*(b) (4) *on July 18, 2012, the decontamination of bioreactor* (b) (4) *was executed, followed by a routine CIP cycle. Although the post-CIP visual inspection indicated a passing result on July 20, 2012, a bioreactor entry occurred on July 21, 2012, where confirmation of the CIP cycle failure occurred when visual residue was observed. The firm has not evaluated the effectiveness of the post decontamination CIP cycle, which is the same cycle run for routine operations, with preceding decontamination procedures executed a*(b) (4)

   Additionally, post-CIP visual inspection procedures are not sufficient in determining whether CIP cycles are effective. For example, the post-CIP visual inspection following production of Soliris lo(b) (4) *in bioreactor* (b) (4) *on January 18, 2012 passed although upon subsequent vessel entry, soil was observed at the normal operating liquid level of the bioreactor.*

Alexion Response:

Alexion follows its Clean in Place (CIP) processes to remove manufacturing soils generated by the routine eculizumab process. The CIP is validated and re-qualifie(b) (4) During planned cleaning qualification activities, an in-depth visual inspection is performed by Validation as part of sample collection to verify cleaning effectiveness.

Alexion acknowledges that the validated CIP cycles are not always sufficient to remove non-routine soils following contamination events. The CIP cycles post contamination events for lots (b) (4) and (b) (4) were followed by additional cleaning to restore the vessels to a clean state. The bioreactors were then sampled for cleaning verification and an in-depth inspection, including tank entry, was performed prior to use in the manufacturing process.

The existing decontamination procedure SOP MFG-0147 "Decontamination of Bioreactors" is currently being evaluated in order to assess its adequacy in mitigating heavy bioreactor soiling as a result of the contamination events. This includes an evaluation of the decontamination method and sequence of operations. The procedure for decontamination will be updated to reflect the modified methodology upon confirmation of its effectiveness by **October 10, 2012.**

Additionally, an alternative CIP cycle is being developed and evaluated to adequately and effectively remove the soils created by the decontamination process under evaluation. This includes evaluation of cleaning reagent concentration and contact times. Verification of the modified CIP cycle will be accomplished through visual inspection, rinse and swab samples. Data collected as part of this verification will be housed in the appropriate quality system. The cycle parameters for the alternative CIP cycle post-decontamination will be updated upon confirmation of its effectiveness by **December 10, 2012.**

SOP TMS-0017 "Visual Inspection of Cleaned Equipment and Sampling of Equipment that Exceed Validated Dirty-Hold Times" will be updated to provide specific instructions on how and where to inspect the equipment, how to document visible residues or anomalies, and how to assess any visible residues or anomalies. Each time non-routine soiling of equipment occurs from decontamination events, CIP visual inspections will be enhanced to require a more formal inspection which will include tank entry. Alexion will complete a revision of this procedure by **September 20, 2012.**

Observation 1 (continued):

d. *The firm has not adequately assessed necessitation for an increased frequency of a sporicidal agent throughout the clean rooms, particularly to address environmental monitoring isolate Bacillus thuringiensis. Bacillus thuringiensis has been isolated in 4 Soliris bioreactor contamination events and 2* (b) (4) *events between April 2011-July 2012.*

Alexion Response:

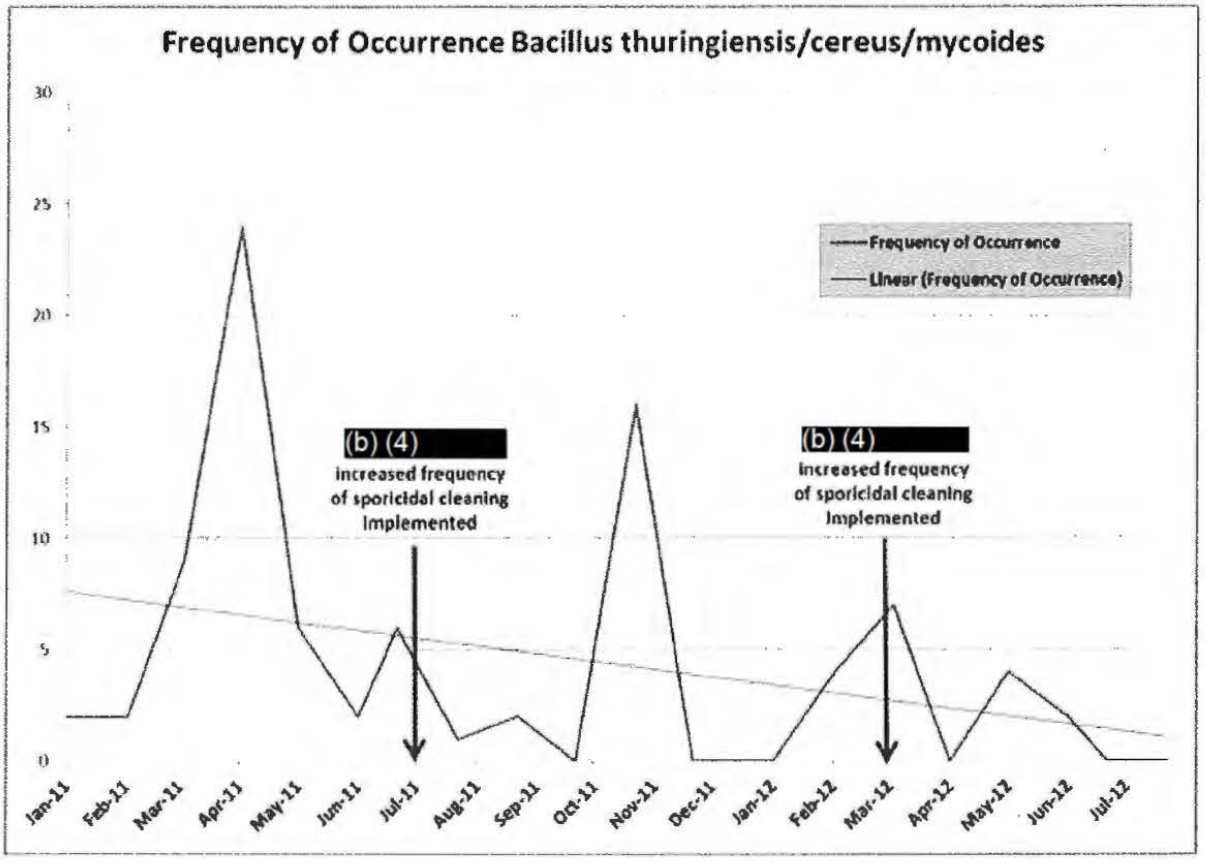
Since January 2011, Alexion has modified its use of sporicidal agents on two occasions as a result of a comprehensive review of environmental monitoring (EM) data.

- A review of all EM excursions from January – June 2011, in which *Bacillus thuringiensis/cereus/ mycoides*[1] were isolated, showed that 69.8% were contributed by room (b) (4) In response to the excursion rate and taking into account the potential impact of certain production operations, the frequency of sporicidal cleaning in room (b) (4) and (b) (4) was increased from (b) (4) to after (b) (4) (not exceeding (b) (4)) in July 2011.

- Additionally, ongoing review of all EM excursions from July 2011 – March 2012 in which *Bacillus thuringiensis/cereus/mycoides*[1] were isolated showed that 18.4% were contributed by rooms (b) (4) and (b) (4) . As a result, the increased frequency of sporicidal cleaning in rooms (b) (4) and (b) (4) was extended to include rooms (b) (4) and (b) (4) in March 2012.

Cleaning of rooms (b) (4) and (b) (4) with a sporicidal agent following (b) (4) operations has reduced the number of excursions contributed by *Bacillus thuringiensis/cereus/ mycoides*[1] in the facility as shown in Figure 1 below. This cleaning regimen will be continued, with the maximum interval between cleanings not exceeding (b) (4)

Figure 1: Frequency of Occurrence of Excursions Contributed by *Bacillus thuringiensis/cereus/mycoides*[1]



Additionally, Alexion will take the following actions:

- The (b) (4) EM of (b) (4) rooms (b) (4) will be augmented to increase the routine sampling site frequency from (b) (4) to (b) (4) (b) (4) Additional surface (b) (4) sites will be included for investigational purposes to further increase the ability to detect potential sources of *Bacillus thuringiensis* and other spore-formers. The additional sampling locations were chosen taking into consideration factors such as proximity to the bioreactor and (b) (4) equipment, and potential for high personnel traffic. This additional sampling and monitoring will be performed for a (b) (4) period starting on August 9, 2012. This could be extended based on the course of the investigation into the bioreactor contamination events which occurred in July and August 2012. A status update with data collected as a result of the augmented sampling will be

provided as part of the bioreactor contamination investigation reports or update on **October 1, 2012** (reference response to observation #1b).

- An evaluation of the frequency of cleaning with sporicidal agents will be conducted upon conclusion of the 2012 manufacturing campaign and documented via a risk assessment justifying any follow-up actions. The risk assessment will be completed by **January 31, 2013**.

[1] *Bacillus thuringiensis/cereus/mycoides*

The primary source of microbial identifications for the Alexion QC Microbiology department is the (b) (4) system by (b) (4) This system is used for environmental monitoring (EM) isolates, water, and in-process product contaminant identification. For isolates related to critical investigations and/or in which a species level identification is required and not achieved via th(b) (4)

The(b) (4) system is an automated microbiology system(b) (4) The system utilizes (b) (4) (b) (4) Supplemental tests may be used t(b) (4) are often due to the fact that these (b) (4) he supplemental reactions/test to differentiate the 3 species of *Bacillus cereus/thuringiensis/mycoides* are as follows:

Bacillus thuringiensis: (b) (4)
Bacillus cereus:(b) (4)
Bacillus mycoide(b) (4)

All of the above differentiating tests are made via microscopic or macroscopic observation(b) (4) These tests are not routinely performed in lieu of(b) (4) methods when speciation is needed and for purposes of EM and water isolate trending. It should be noted that during the generation of all lists, percentages, and graphs requested during the inspection and also in this response, the most conservative approach was taken to account for all *Bacillus thuringiensis* isolated at ARIMF since January 1, 2011 to July 15, 2012. All strains of *Bacillus* reported (b) (4) *Bacillus thuringiensis/cereus/mycoides* were included in the total count for the occurrence of *B. thuringiensis* at ARIMF in both EM and product contaminations so that no potential *B. thuringiensis* would go unaccounted for. On those occasions where a species was required and (b) (4) *B. thuringiensis/cereus/mycoides* was sent out for(b) (4) the vast majority were confirmed to be *Bacillus thuringiensis*.

Observation 2:

Deviation 3324 was initiated due to a leak on the interior of the drywall in a column in the cell culture area (b) (4) A tent was constructed prior to cutting into the drywall to investigate the leak which was observed to have caused mold on the column drywall interior. This deviation was not adequately investigated for the following:

a. *The deviation did not list in-process batches in (b) (4) during the work or assess whether there was any impact to the batches manufactured in the cell culture area (b) (b) (4) The deviation assumed since the cell culture operations are closed that there was no impact to batch quality.*

b. *Increased environmental monitoring sampling, e.g. augmented site sampling and/or frequency, did not occur during the construction of the tent in the controlled cell culture suite, during the Facilities Department's activities within the tent, and during the deconstruction of the tent to verify the environment was not compromised due to these non-routine activities.*

c. *Prior to execution, the Quality Unit did not approve the cleaning plan performed by Manufacturing following deconstruction of the tent.*

d. *The Facilities Department lacks specific instructions on cleaning procedures prior to deconstruction of the tent.*

Alexion Response:

a. On December 27, 2011, while performing work to repair sheetrock on a column in (b) (b) (4) due to a leak from (b) (4) (b) (4) n the mezzanine, what appeared to be mold was observed around the interior surfaces of the drywall surrounding the column. The area was tented in accordance with SOP FAC-0049 "Temporary Gray Spaces at ARIMF" while all work was being performed and the area was cleaned with (b) (4) prior to the tent being removed.

The lots being manufactured in (b) (4) while the maintenance work was being performed were lo (b) (4) which was being transferred from bioreactor (b) (4) (b) (4) o bioreactor (b) (4) and lot (b) (4) which was on (b) (4) n bioreactor (b) (4) The drywall repair maintenance activities were performed December 27-28, 2011. Environmental monitoring performed i (b) (4) on December 29, 2011 and January 6, 2012 showed no excursions for room (b) (4) Lots (b) (4) and (b) (4) performed as expected for the duration of the manufacturing process; all operational data points were within expected results. In process Bioburden results were as follows:

- Lot (b) (4) –
  - (b) (4) Bioburden: < 1CFU/10mL (action limit (b) CFU/10mL)
  - (b) (4) ioburden: < 1CFU/10mL (action limi (b) (4) CFU/10mL)
- Lo (b) (4)
  - (b) (4) Bioburden: < 1CFU/10mL (action limit (b) CFU/10mL)
  - (b) (4) Bioburden: < 1CFU/10mL (action limit (b) CFU/10mL)
  - (b) (4) Bioburden: < 1CFU/10mL (action limit (b) CFU/10mL)
  - (b) (4) Bioburden: < 1CFU/10mL (action limit (b) (4) CFU/10mL)

Lot (b) (4) was successfully (b) (4) from (b) (4) on January 11, 2012. Lot (b) (4) was successfully (b) (4) from (b) (4) on January 1, 2012.

There was no impact to the cell culture operational performance of lots (b) (4) and (b) (4) as a result of the deviation event. There were no deviations associated with in-process controls or bioburden and endotoxin results for lots (b) (4) and (b) (4) throughout the manufacturing process. A review of the release data for lot (b) (4) (CoA approved February 15, 2012) and lo (b) (4) (CoA approved February 17, 2012) confirmed no impact to batch quality as a result of deviatio (b) (4)

Deviation (b) (4) was reopened and updated on August 15, 2012 to add an assessment by QA of cell culture operation performance and release data for the in-process batches manufactured in (b) (4) at the time the work was performed; the updated deviation assessment documents confirmation of no impact to batch quality.

b. Alexion will revise SOP FAC-0049 "Temporary Gray Spaces at ARIMF" to require pre-defined augmented environmental monitoring and cleaning activities to be performed during the Facilities Department's activities within the tent, and during the deconstruction of the tent to verify the environment within the Manufacturing areas is not compromised. The revision of SOP FAC-0049 will be complete by **September 28, 2012**.

c. The revision to SOP FAC-0049, "Temporary Gray Spaces at ARIMF" will include a reference to SOP FAC-0028, "Facilities Maintenance Program" to ensure QA oversight and pre-approval and post approval of all corrective work performed within

the Manufacturing areas. The revision of SOP FAC-0049 will be complete by **September 28, 2012.**

d. The revision of SOP FAC-0049 will also include specific instructions for the cleaning to be performed by Facilities within the tented area prior to deconstruction of the tent and cleaning to be performed by Manufacturing following deconstruction of the tent. The revision of SOP FAC-0049 will be complete by **September 28, 2012.**

Observation 3:

The procedure "Quality Control of Microbiological Media" SOP QC-020l, version 5.0 is used for growth promotion of (b) *used in the environmental monitoring program. The procedure does not specifically describe how to select environmental isolates to be used in growth promotion. The firm has selected* (b) (4)

Alexion Response:

Each lot of media used for Alexion's QC environmental monitoring (EM) program is qualified for growth promoting properties prior to release per SOP QC-0201, "Quality Control of Microbiological Media". The ATCC microorganisms specified in this procedure were chosen in accordance with appropriate compendia requirements (USP <61> and USP <71>) and also the media manufacturer's recommendations. In addition to employing ATCC isolates, EM media is also growth promoted employing in-house facility isolates including the Gram positive spore forming rod (b) (4) and Gram positive coccus (b) (4) These organisms were chosen due to their high prevalence in recovery from the EM program and thus making them readily available. To improve our analyst's ability to achieve proper working concentrations of these microbes when performing growth promotion, in house strains have been (b) (4) by (b) (4) at specified concentrations and are now employed during growth promotion (b) (4)
to include (b) (4)
(b) (4)

Alexion will take the following actions:

1. SOP QC-0402, "Environmental Monitoring and Water (b) (4) Reviews", will be revised to require an (b) (4) review of all environmental isolates from the previous (b) (b) (4) of environmental monitoring. This review will determine the most frequently occurring species of Gram positive rods, Gram negative rods, Gram positive cocci and Fungi. This review will be documented in the (b) Environmental Monitoring trend report each year. SOP QC-0402 will be revised by **September 28, 2012**.

2. Upon approval of the (b) (4) reports, a gap analysis of the of the environmental organisms listed in SOP QC-0201, "Quality Control of Microbiological Media", will be conducted and SOP QC-0201 revised, if necessary, to ensure appropriate species of highest occurrence identified in the EM trend reports are included in the media growth promotion program.

3. Additionally, SOP QC-0201 will be revised to include the rationale utilized for inclusion of environmental isolates in the (b) (4) media growth promotion program. SOP QC-0201 will be revised by **December 14, 2012**.

Observation 4:

Deviations are not required to be initiated for all visual inspection failures, for example post CIP visual inspection failures. For example, pooling was observed on July 26, 2011 following a CIP cycle for (b) (4) *A deviation was not initiated; subsequently a second CIP cycle was performed. Additionally, the Quality Unit does not review the bioreactor use and cleaning logbooks, therefore they are not required to be notified of such failures.*

Alexion Response:

The Manufacturing procedures for performing Clean in Place (CIP) of equipment instruct the operators to notify a Supervisor if there are any visual inspection failures and to repeat the CIP. See Table 1 below. Use of the equipment for forward processing only occurs upon completion of a successful CIP, which includes a passing visual inspection.

Alexion will revise all manufacturing specific procedures, identified in Table 1 below, that include instruction for CIP or Clean out of Place (COP) of equipment to include the requirement to generate a deviation upon failed visual inspection, post CIP. Generation of a deviation will assure notification to the Quality Unit of such failures. Revision of these procedures will include instruction to reference SOP TMS-0017 "Visual Inspection of Cleaned Equipment and Sampling of Equipment that Exceed Validated Dirty-Hold Times" for visual inspection criteria and to document in the equipment logbook the reason for failure. Revision of these procedures will be completed by **September 28, 2012.**

Table 1: Manufacturing Procedures for Equipment CIP Operations

Doc. #	Title
MFG-0115	Operation and Maintenance of the (b) (4)
MFG-0116	Clean Out of Place
MFG-0121	CIP / Transfer o(b) (4) , and (b) (4)
MFG-0122	CIP / Transfer of (b) (4) , and (b) (4)
MFG-0126	Operation and CIP of (b) (4) and the (b) (4) and (b) (4) Lines
MFG-0185	Operation and CIP of (b) (4))
MFG-0186	Operation and CIP of (b) (4)
MFG-0187	Operation and CIP of (b) (4)
MFG-0148	CIP / SIP / Transfer Operation o (b) (4)

Doc. #	Title
MFG-0149	Operation of th(b) (4)
MFG-0150	Operation of the(b) (4)
MFG-0157	CIP / SIP / Transfer Operation (b) (4)
MFG-0161	Operation of (b) (4)
MFG-0169	Operation of(b) (4)
MFG-0170	Operation of (b) (4)
MFG-0193	Operation of(b) (4)
MFG-0194	Operation of (b) (4)

Observation 5:

The Quality Unit has not ensured verification of whether current sampling procedures for larger drummed raw materials could potentially contaminate the room in which they are sampled or other raw materials. Raw materials sampled in larger drummed containers are taken in an unclassified room which is not a controlled, qualified, or certified area. Additionally, the cleaning procedure following raw material sampling is not specific on how to clean the area where sampling occurs on the floor.

Alexion Response:

Alexion currently has the following controls in place to minimize risk of cross contamination between materials sampled:

- Only one material lot is sampled at a time within the respective sampling area.
- Each raw material container is cleaned with (b) (4) before being introduced to the sampling area.
- The sampling area is cleaned pre and post sampling in between material lots with (b) (4) (b) (4)
- Disposable sampling apparatus (e.g. scoops, pipets) and sampling thieves are used.
- Personal Protective Equipment for sampling to include lab coat, sterile sleeves, sterile gloves, bouffant, beard cover (if applicable), is required.

Alexion will revise SOP QC-0346, "Sampling and Retains of Incoming Raw Materials" as described below. The revision will be completed by **September 28, 2012**.

- Detailed diagram(s) of approved sampling areas located in (b) (4).
- Increase detailed cleaning procedures of sampling area pre and post sampling.
- Details on the cleaning frequency of the sampling room
    - Cleaning of floors, bench tops and dedicated sampling areas will increase from (b) (4)
    - Cleaning will include alternating the cleaners (b) (4) (b) (4)

Currently (b) (4) raw materials sampled at Alexion are sampled in an (b) (4) (b) (4) (b) (4) The remaining raw materials are larger drums that are sampled in an unclassified room which is not a controlled, qualified, or certified area. As a temporary solution, Alexion will purchase and use a (b) (4) (b) while sampling from larger drummed containers. This temporary solution will enhance control for the large drum dispensing

operation by providing additional environmental isolation and will be implemented by **September 28, 2012**.

Alexion also commits to initiating and completing a project to provide and qualify a classified area within the facility where raw materials in larger drummed containers will be sampled. This project will be completed and qualified by **April 1, 2013**.