Accepted Manuscript

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PII: S0022-3549(17)30131-4

DOI: 10.1016/j.xphs.2017.02.022

Reference: XPHS 664

To appear in: *Journal of Pharmaceutical Sciences*

Received Date: 12 January 2017

Revised Date: 4 February 2017

Accepted Date: 13 February 2017

Please cite this article as: Hsiao K, Alves J, Patel R, Adams M, Nashine V, Goueli S, A High-Throughput Bioluminescent Assay to Monitor the Deamidation of Asparagine and Isomerization of Aspartate Residues in Therapeutic Proteins and Antibodies, *Journal of Pharmaceutical Sciences* (2017), doi: 10.1016/j.xphs.2017.02.022.

A high-throughput bioluminescent **assay to monitor the deamidation of asparagine and isomerization of aspartate residues in therapeutic proteins and antibodies**

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Abstract:

Since the introduction of Herceptin and Rituximab in 1986 therapeutic antibodies have gained tremendous momentum in treatment of broad range of several diseases such as cancer and inflammation. Selection of the clinical candidate mAb usually starts with large-scale *in vitro* screening and profiling of multiple mAbs to identify candidates that show high *in vitro or in vivo* activity, and thus it is necessarily to identify and eliminate potentially unstable mAbs during the lead selection process. Antibodies undergo a variety of degradation reactions which may result in compromised bioactivity and safety profile. The non-enzymatic post-translational modification of both deamidation of asparagine (Asn) and isomerization of aspartate (Asp) residues is one of the major chemical reactions occurring in proteins during production and storage resulting in formation of protein variants that may affect the quality, safety, and functionality of the therapeutic proteins. Current Methods (HPLC and LC/MS) for monitoring isoaspartate (isoAsp) formation are time consuming, require specialized equipment and trained personnel and are not amenable to high throughput scale (HTS). We have developed a robust, homogenous, high throughput formatted, and sensitive assay to accurately monitor the formation of isoAsp under several conditions such as new formulations, storage periods, and temperature.