

Protein Analysis with the Agilent 2100 Bioanalyzer System

An overview of the protein assay portfolio

Technical Overview



Introduction

The Agilent 2100 Bioanalyzer system is a microfluidic system for the electrophoresis-based analysis of biomolecules. It has developed into a key platform for routine QA/QC of protein samples by supporting three distinguished protein analysis kits:

- Protein 80 kit – for protein analysis in the low molecular weight range
- Protein 230 kit – for general protein analysis up to 230 kDa
- High Sensitivity Protein 250 kit – for picogram sensitivity and lowest level of impurity detection

Besides the ease-of-use of prevalidated reagents, this protein assay portfolio offers a number of advantages over techniques like slab-gel electrophoresis or standard CE-SDS methods:

- Wide molecular weight range, from 5 kDa to 250 kDa
- Broad protein loading capacity from pg/ μ L to μ g/ μ L



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Conclusion

The Agilent 2100 Bioanalyzer system in conjunction with its protein assay portfolio is a versatile tool for the validation of protein preparations. The results of the presented set of experiments are summarized in Table 2 to help with assay selection.

References

1. Agilent Protein 80 Kit Guide, Agilent reference number G2938-90062
2. Agilent Protein 230 Kit Guide, Agilent reference number G2938-90054
3. Agilent High Sensitivity Protein 250 Kit Guide, Agilent reference number G2938-90310.
4. Reviewer Guidance, Validation of Chromatographic Methods, Center for Drug Evaluation and Research, Food and Drug Administration, 1994.

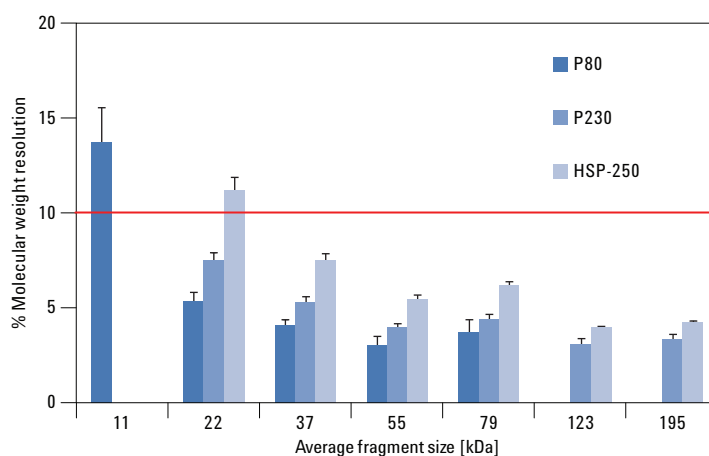


Figure 4
Molecular weight resolution across the size range of the three protein assays: The protein size standards of the corresponding assays were analyzed and the % molecular weight resolution was calculated for pairs of adjacent ladder peaks.

	P80 Assay	P230 Assay	HSP-250 Assay
Molecular weight range	5-80 kDa	14-230 kDa	10-250 kDa
Application area	Routine assessment of low MW impurities	Routine assessment of protein impurity and size in different molecular weight range	Highly sensitive detection of impurities
Precision⁺ for: Sizing	< 3%		< 3%
Relative concentration % Purity	< 20% < 6%		< 20% < 5%
Impurity detection*	≤ 1.5%		0.03%
Protein staining/labeling	On-chip		Off-chip
Sample preparation time	~ 10 min/10 samples		~ 45 min/10 samples
Run time	~ 30 min		
MW resolution (Rs = 0.8)	< 10% for higher MW proteins		

⁺ %CV (n = 5 chips)

* with IgG2

Table 2
Summary of the present study: Listed are the key characteristics of the three 2100 Bioanalyzer protein assays.

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© Agilent Technologies, Inc., 2019
February 1, 2019
Publication Number 5990-5283EN



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