Agilent Protein 80 Kit Guide
Notices


No part of this manual may be reproduced in any form or by any means (including electronic storage and retrieval or translation into a foreign language) without prior agreement and written consent from Agilent Technologies, Inc. as governed by United States and international copyright laws.

Manual Part Number
G2938-90062 Rev. C

Edition
12/2016

Printed in Germany
Agilent Technologies
Hewlett-Packard-Strasse 8
76337 Waldbronn

For Research Use Only
Not for use in diagnostic procedures

Warranty

The material contained in this document is provided “as is,” and is subject to being changed, without notice, in future editions. Further, to the maximum extent permitted by applicable law, Agilent disclaims all warranties, either express or implied, with regard to this manual and any information contained herein, including but not limited to the implied warranties of merchantability and fitness for a particular purpose. Agilent shall not be liable for errors or for incidental or consequential damages in connection with the furnishing, use, or performance of this document or of any information contained herein. Should Agilent and the user have a separate written agreement with warranty terms covering the material in this document that conflict with these terms, the warranty terms in the separate agreement shall control.

Safety Notices

CAUTION

A CAUTION notice denotes a hazard. It calls attention to an operating procedure, practice, or the like that, if not correctly performed or adhered to, could result in damage to the product or loss of important data. Do not proceed beyond a CAUTION notice until the indicated conditions are fully understood and met.

WARNING

A WARNING notice denotes a hazard. It calls attention to an operating procedure, practice, or the like that, if not correctly performed or adhered to, could result in personal injury or death. Do not proceed beyond a WARNING notice until the indicated conditions are fully understood and met.
Contents

1 Agilent Protein 80 Kit 4

2 Equipment Required for a Protein 80 Assay 6

3 Setting Up the Assay Equipment and Bioanalyzer 7
   Setting up the Chip Priming Station 8
   Setting up the Bioanalyzer 9
   Starting the 2100 Expert Software 10

4 Essential Measurement Practices 11

5 Agilent Protein 80 Assay Protocol 12
   Preparing the Gel-Dye Mix 12
   Preparing the Destaining Solution 14
   Preparing the Denaturing Solution 14
   Preparing the Samples and the Ladder 15
   Loading the Gel-Dye Mix and the Destaining Solution 16
   Loading the Ladder and Samples 18
   Inserting a Chip in the Agilent 2100 Bioanalyzer 19
   Starting the Chip Run 20
   Cleaning Electrodes after a Chip Run 22

6 Checking Your Agilent Protein 80 Assay Results 24
   Protein 80 Ladder Well Results 24
   Protein 80 Sample Well Results 27

7 List of Compatible Buffers and Buffer Compounds 29
**Agilent Protein 80 Kit**

<table>
<thead>
<tr>
<th>Protein 80 Kit (reorder number 5067-1515)</th>
<th>Protein 80 Reagents (reorder number 5067-1516) &amp; Supplies</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Protein Chips</strong></td>
<td><strong>Protein 80 Reagents (reorder number 5067-1516) &amp; Supplies</strong></td>
</tr>
<tr>
<td>25 Protein Chips</td>
<td>1 Electrode Cleaner</td>
</tr>
<tr>
<td></td>
<td>(red) Protein 80 Gel-Matrix (4 vials) in box labelled Part I. Store at 4 °C</td>
</tr>
<tr>
<td></td>
<td>1 Electrode Cleaner</td>
</tr>
<tr>
<td></td>
<td>(blue) Protein 80 Dye Concentrate(^1) in box labelled Part I. Store at 4 °C</td>
</tr>
<tr>
<td></td>
<td>(white) Protein 80 Sample Buffer (4 vials) in box labelled Part II. Store at -20 °C</td>
</tr>
<tr>
<td><strong>Syringe Kit</strong></td>
<td><strong>Syringe Kit</strong></td>
</tr>
<tr>
<td>1 Syringe</td>
<td>1 Syringe</td>
</tr>
<tr>
<td></td>
<td>4 Spin Filters</td>
</tr>
</tbody>
</table>

\(^1\) This product is provided under a license by Life Technologies Corporation to Agilent Technologies. The purchase of this product conveys to the buyer the non-transferable right to use the purchased amount of the product and components of the product only as described in accompanying product literature. The sale of this product is expressly conditioned on the buyer not using the product or its components (1) in manufacturing; (2) to provide a service, information, or data to an unaffiliated third party for payment; (3) for therapeutic, diagnostic or prophylactic purposes; (4) to resell, sell or otherwise transfer this product or its components to any third party, or use for any use other than use in the subfields of research and development, quality control, forensics, environmental analysis, biodefense or food safety testing. For information on purchasing a license to this product for purposes other than described above contact Life Technologies Corporation, Cell Analysis Business Unit, Business Development, 29851 Willow Creek Road, Eugene, OR 97402, Tel: (541) 465-8300. Fax: (541) 335-0354.
### Table 1  Physical Specifications

<table>
<thead>
<tr>
<th>Type</th>
<th>Specification</th>
</tr>
</thead>
<tbody>
<tr>
<td>Analysis run time</td>
<td>30 min</td>
</tr>
<tr>
<td>Samples per chip</td>
<td>10</td>
</tr>
<tr>
<td>Sample volume</td>
<td>4 µL</td>
</tr>
<tr>
<td>Kit stability</td>
<td>4 months (for storage temperature see individual box)</td>
</tr>
</tbody>
</table>
| Kit size                    | 25 chips  
10 samples/chip  
= 250 samples/kit                                                  |
| Compatible buffers          | “List of Compatible Buffers and Buffer Compounds” on page 29       |

### Table 2  Analytical Specifications

<table>
<thead>
<tr>
<th>Type</th>
<th>Agilent Protein 80 Assay</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sizing range</td>
<td>5 – 80 kDa</td>
</tr>
<tr>
<td>Typical sizing resolution</td>
<td>10 %</td>
</tr>
<tr>
<td>Typical sizing accuracy</td>
<td>10 % (CAII(^1), BLG(^2))</td>
</tr>
<tr>
<td>Sizing reproducibility</td>
<td>3 % CV (CAII, BLG)</td>
</tr>
</tbody>
</table>
| Sensitivity (Signal/Noise>3) | 6 ng/µL CAII (15 ng/µL BSA\(^3\)) in PBS  
10 ng/µL (CAII) in 0.5 M NaCl (30 ng/µL BSA in 0.5 M NaCl) |
| Quantitative range          | 60 – 2000 ng/µL CAII in PBS                                                             |
| Qualitative range           | 6 – 4000 ng/µL CAII and BLG                                                            |
| Quantitation reproducibility | 20 % CV (CAII, BLG)                                                                   |

1  CAII = Carbonic Anhydrase  
2  BLG = beta-Lactoglobulin  
3  BSA = Bovine Serum Albumine
Equipment Required for a Protein 80 Assay

**Equipment supplied with the Agilent 2100 bioanalyzer**

- Chip Priming Station (reorder number 5065-4401)

**Additional material required (not supplied)**

- Pipettes (10 μl, 20 μl, 100 μl and 1000 μl) with compatible tips
- 0.5 ml microcentrifuge tubes
- Deionized water
- 1 M Dithiothreitol (DTT) solution or β-mercaptoethanol (BME)
- Microcentrifuge
- Heating block for 0.5 ml tubes or water bath

Check the Agilent Lab-on-a-Chip webpage for details on assays: [www.agilent.com/chem/labonachip](http://www.agilent.com/chem/labonachip).
Setting Up the Assay Equipment and Bioanalyzer

Before beginning the chip preparation protocol, ensure that the chip priming station and the bioanalyzer are set up and ready to use.

You have to

- replace the syringe at the chip priming station with each new protein kit
- adjust the base plate of the chip priming station
- adjust the syringe clip at the chip priming station
- finally, make sure that you start the software before you load the chip.

NOTE

The Agilent Protein 80 assay is a high sensitivity assay. Please read this guide carefully and follow all instructions to guarantee satisfactory results.
Setting up the Chip Priming Station

1 Replace the syringe:
   a Unscrew the old syringe from the lid of the Chip Priming Station.
   b Release the old syringe from the clip. Discard the old syringe.
   c Remove the plastic cap of the new syringe and insert it into the clip.
   d Slide it into the hole of the luer lock adapter and screw it tightly to the Chip Priming Station.

2 Adjust the base-plate:
   a Open the chip priming station by pulling the latch.
   b Using a screwdriver, open the screw at the underside of the base plate.
   c Lift the base plate and insert it again in position A. Retighten the screw.

3 Adjust the syringe clip:
   a Release the lever of the clip and lift it up or down to adjust it to the middle position.

NOTE Replace the syringe with each new Reagent Kit.
Setting up the Bioanalyzer

1. Open the lid of the bioanalyzer and make sure that the electrode cartridge is inserted in the instrument. If not, open the latch and insert the electrode cartridge.

![Electrode cartridge inserted in the instrument](image)

Figure 1  Electrode cartridge inserted in the instrument (graphic shows an example).

2. Remove any remaining chip.
Setting Up the Assay Equipment and Bioanalyzer

Starting the 2100 Expert Software

To start the software:

1. Go to your desktop and double-click the following icon.

The screen of the software appears in the Instrument context. The icon in the upper part of the screen represents the current instrument/PC communication status:

2. If more than one instrument is connected to your PC, select the instrument you want to use in the tree view.
Handle and store all reagents according to the instructions on the label of the individual box.

Avoid sources of dust or other contaminants. Foreign matter in reagents and samples or in the wells of the chip will interfere with assay results.

Upon arrival make aliquots for the sample buffer and the ladder with the typical amount required for daily use and store them at -20 °C. Keep the vial in use at 4 °C to avoid freeze-thaw cycles.

Allow all reagents and samples to equilibrate to room temperature for 30 minutes before use.

Protect all reagents from light. Remove light covers only when pipetting. The dye contained in the reagents decomposes when exposed to light and this reduces the signal intensity.

Always insert the pipette tip to the bottom of the well when dispensing the liquid. Placing the pipette at the edge of the well may lead to poor results.

Use a new syringe and electrode cleaners with each new kit.

Use loaded chips within 5 minutes. Reagents might evaporate, leading to poor results.

Do not touch the Agilent 2100 bioanalyzer during analysis and never place it on a vibrating surface.

Use 0.5 ml vials to denature samples. Using larger vials may lead to poor results, caused by evaporation.
After completing the initial steps in “Setting Up the Assay Equipment and Bioanalyzer” on page 7, you can prepare the reagents, load the chip, and run the assay, as described in the following procedures.

## Preparing the Gel-Dye Mix

### Handling DMSO

**WARNING**

Kit components contain DMSO. Because the dye binds to nucleic acids, it should be treated as a potential mutagen and used with appropriate care.

➔ Wear hand and eye protection and follow good laboratory practices when preparing and handling reagents and samples.

➔ Handle the DMSO stock solutions with particular caution as DMSO is known to facilitate the entry of organic molecules into tissues.

1. Allow the Protein 80 Dye Concentrate (blue) and the Protein 80 Gel-Matrix (red) to equilibrate to room temperature for 30 minutes.

**NOTE**

It is important that all the reagents have room temperature before starting the next step. Protect the Dye Concentrate from light.
2 Transfer the content (650 μl) of an Agilent Protein 80 Gel-Matrix vial (red) to a spin filter. Make sure the complete volume of 650 μl has been transferred.

3 Centrifuge at 2500 g ± 20 % for 15 min.

4 To the filtered and centrifuged Gel add 25 μl of the well vortexed Dye Concentrate (blue).

5 Mix thoroughly for 10-20 s (Vortexer) until a uniform color is obtained.

6 Label with the date and GD (Gel/Dye). Use within 4 weeks.

**NOTE**

Always use the volumes indicated. Using different volumes in the same ratio will produce inaccurate results.

**NOTE**

The gel-dye mix is sufficient for 9 chips. Use the gel-dye mix within four weeks of preparation, and protect it from light at all times. Store the gel-dye mix at 4 °C when not in use for more than one hour.
Preparing the Destaining Solution

1. Transfer the content (650 μl) of another Protein 80 Gel-Matrix vial (red ) to a spin filter. Make sure the complete volume of 650 μl has been transferred.
2. Centrifuge at 2500 g ± 20 % for 15 min.
3. Label with the date and DS (Destaining Solution). Use within kit life time.

**NOTE**
The prepared destaining solution is sufficient for 25 chips and is stable for the complete kit lifetime.

Preparing the Denaturing Solution

1. For reducing conditions, add 3.5 Vol-% of 1 M Dithiothreitol (DTT) or β-mercaptoethanol (BME) to an aliquot of sample buffer, white (e.g. 1.0 μl DTT or BME to an aliquot of 28.6 μl Sample Buffer).
   OR
   Alternatively, for non-reducing conditions add 3.5 Vol-% of water to your aliquoted sample buffer vial.
2. Vortex for 5 s.

**NOTE**
The total volume of the accordingly prepared denaturing solution (200 μl sample buffer plus 7 μl 1 M DTT or water) is sufficient for 10 chips. Use the prepared denaturing solution within 1 week. To avoid freeze thaw cycles, store the denaturing solution as well as smaller aliquots of sample buffer at 4 °C when not in use for more than 1 hour.
Preparing the Samples and the Ladder

**NOTE** For a list of compatible buffers, please refer to the chapter “List of Compatible Buffers and Buffer Compounds” on page 29.

1. Combine 4 μl protein sample and 2 μl denaturing solution in a 0.5 ml vial.
2. Place the sample vials and a vial containing a 6 μl aliquot of Protein 80 Ladder (yellow) in the heating block or the water bath at 95 °C for 5 min. Cool down afterwards.
3. Spin vials for 15 s.
4. Add 84 μl deionized water to samples and ladder and vortex.

**NOTE** The diluted samples and ladder are stable for one day. Store samples at 4 °C when not in use for more than 1 hour.

Always make aliquotes of the reagents to maintain a constant quality! E.g. you might want to prepare twenty five 6 μl aliquots of ladder (amount needed for one chip) and store them at -20 °C.
Agilent Protein 80 Assay Protocol

Loading the Gel-Dye Mix and the Destaining Solution

Before loading the gel-dye mix, make sure that the base-plate of the Chip Priming Station is in position (A) and the adjustable clip is set to the middle position. Refer to “Setting up the Chip Priming Station” on page 8 for details.

1. Allow the gel-dye mix to equilibrate to room temperature for 30 minutes before use. Always protect the gel-dye mix from light during this time.

2. Take a new Protein chip out of its sealed bag and put it on the Chip Priming Station.

3. Pipette 12 μl of gel-dye mix at the bottom of the well marked 6.

When pipetting the gel-dye mix, make sure not to draw up particles that may sit at the bottom of the gel-dye mix vial. Insert the tip of the pipette to the bottom of the chip well when dispensing. This prevents a large air bubble forming under the gel-dye mix. Placing the pipette at the edge of the well may lead to poor results.

4. Set the timer to 60 seconds, make sure that the plunger is positioned at 1 ml and then close the Chip Priming Station. The lock of the latch will click when the Priming Station is closed correctly.
5 Press the plunger of the syringe down until it is held by the clip.

6 Wait for exactly 60 seconds and then release the plunger with the clip release mechanism.

7 Visually inspect that the plunger moves back at least to the 0.3 ml mark.

8 Wait for 5 s, then slowly pull back the plunger to the 1 mL position.

9 Open the Chip Priming Station.

10 Pipette 12 μl of gel-dye mix in all wells labeled with “G”

11 Pipette 12 μl of the destaining solution in the well marked Os.

NOTE Protect the gel-dye mix from light. Store the gel-dye mix at 4 °C when not in use for more than 1 hour.
Loading the Ladder and Samples

1. Pipette 6 μl of sample (prepared as described in “Preparing the Samples and the Ladder” on page 15) in all 10 sample wells (Note: all 10 sample wells must be filled either with ladder or sample).

2. Pipette 6 μl of the diluted ladder into the well marked with the ladder symbol 🛠.

**NOTE**
Do not leave any wells empty or the chip will not run properly. Pipette a sample or ladder replicate in any empty sample well.

3. Place the chip in the Agilent 2100 Bioanalyzer and start the assay immediately.
Inserting a Chip in the Agilent 2100 Bioanalyzer

1. Open the lid of the Agilent 2100 bioanalyzer.
2. Check that the electrode cartridge is inserted properly. Refer to “Setting up the Bioanalyzer” on page 9 for details.
3. Place the chip carefully into the receptacle. The chip fits only one way.

**CAUTION**
Sensitive electrodes and liquid spills
Forced closing of the lid may damage the electrodes and dropping the lid may cause liquid spills resulting in bad results.

⇒ Do not use force to close the lid and do not drop the lid onto the inserted chip.

4. Carefully close the lid. The electrodes in the cartridge fit into the wells of the chip.
5. The 2100 expert software screen shows that you have inserted a chip and closed the lid by displaying the chip icon at the top left of the Instrument context.
Starting the Chip Run

NOTE Please note that the order of executing the chip run may change if the Agilent Security Pack software (only applicable for Agilent 2100 expert software Revision B.02.02 and higher) is installed. For more details please read the 'User’s Guide' which is part of the Online Help of your 2100 expert software.

1. In the Instrument context, select the appropriate assay from the Assay menu.

2. Accept the current File Prefix or modify it.

Data will be saved automatically to a file with a name using the prefix you have just entered. At this time you can also customize the file storage location and the number of samples that will be analyzed.
3 Click the Start button in the upper right of the window to start the chip run. The incoming raw signals are displayed in the Instrument context.

![Start](image)

4 To enter sample information like sample names and comments, select the Data File link that is highlighted in blue or go to the Data context and select the Chip Summary tab. Complete the sample name table.

**NOTE**

If absolute quantitation is required with a standard protein, mark the check box Use For Calibration and enter standard concentration.
To review the raw signal trace, return to the Instrument context.

**CAUTION**

Contamination of electrodes

Leaving the chip for a period longer than 1 hour (e.g. over night) in the Bioanalyzer may cause contamination of the electrodes.

➔ Immediately remove the chip after a run.

After the chip run is finished, remove the chip from the receptacle of the bioanalyzer and dispose of it according to good laboratory practices.

**Cleaning Electrodes after a Chip Run**

When the assay is complete, *immediately* remove the used chip from the Agilent 2100 bioanalyzer and dispose of it according to good laboratory practice. Then perform the following procedure to ensure that the electrodes are clean (no residues are left over from the previous assay).

**NOTE**

Use a new electrode cleaner with each new kit.

**CAUTION**

Leak currents between electrodes

Liquid spill may cause leak currents between the electrodes.

➔ Never fill too much water in the electrode cleaner.

1. Slowly fill one of the wells of the electrode cleaner with 350 μl deionized analysis-grade water.
2. Open the lid and place the electrode cleaner in the Agilent 2100 bioanalyzer.
3 Close the lid and leave it closed for about 10 seconds.
4 Open the lid and remove the electrode cleaner.
5 Wait another 10 seconds to allow the water on the electrodes to evaporate before closing the lid.

**NOTE**
After 5 chip runs, empty and refill the electrode cleaner.
After 25 chip runs, replace the used electrode cleaner by a new one.

**NOTE**
When switching between different assays, a more thorough cleaning may be required. Refer to the Agilent 2100 Bioanalyzer Maintenance and Troubleshooting Help for details. This is part of the Online Help of the 2100 Expert Software.
Protein 80 Ladder Well Results

To check the results of your run, select the Gel or Electropherogram tab in the Data context. The electropherogram of the ladder well window should resemble the one shown below.

![Protein 80 ladder](image)

**Figure 2**  Protein 80 ladder
Major features of a successful ladder run are:

- 6 ladder peaks and all ladder peaks are well resolved
- Flat baseline
- Readings at least 15 fluorescence units higher than baseline readings

If the electropherogram of the ladder well window does not resemble the one shown above, for assistance, please refer to the *2100 Expert Maintenance and Troubleshooting Guide* within the online Help of the 2100 Expert software.

In some of your runs, you might see a double system peak, as shown below. Usually this can be handled by the software and does not cause a problem.

**Figure 3**  Protein 80 ladder with double system peak
Checking Your Agilent Protein 80 Assay Results
Protein 80 Ladder Well Results

In case both system peaks are identified as ladder peaks, exclude the first peak of the two system peaks by doing the following:

1. Move the cursor over the second peak in the peak table and click the right mouse button.

2. Select *Exclude Peak* to make the change come into effect.
Protein 80 Sample Well Results

To review the results of a specific sample, select the sample name in the tree view and highlight the Results sub-tab. The electropherogram of the sample well window should resemble the one shown here for the Protein 80 assay.

![Protein peaks of a successful sample run](image)

**Figure 4** Protein peaks of a successful sample run

Major features of a successful protein sample run are:

- All sample peaks between the lower and upper marker peaks
- Two marker peaks, system peak(s)
- Lower marker peak between 10.5 and 15.5 seconds (raw migration time: analysis turned off by clicking the 'Don't Analyze' button)
- Upper marker peak between 38 and 48 seconds (raw migration time)
- Baseline readings between 20 and 250 fluorescence units in the ladder well (Turn analysis off)
- Both marker peaks well resolved from sample peaks (depending on sample)
Checking Your Agilent Protein 80 Assay Results

Protein 80 Sample Well Results

**NOTE** Baseline correction can affect quantitation when analyzing broad peaks (e.g. non-reduced IgG or cell lysates) and may then be turned off for accurate quantitation.

For easier identification of the correct lower and upper marker, turn off the alignments to identify and manually assign markers. To turn the alignment off, select *Don’t analyze* and compare markers in samples to markers in ladder by following the drift in the gel-like image.

For troubleshooting, please refer to the *2100 Expert Maintenance & Troubleshooting Guide*. 
The following tables list protein sample buffers and buffer components which are known to be compatible with the Protein 80 kit.

For an updated list please refer to the web-site www.agilent.com/chem/labonachip.

### Salts and Buffers (Composition Measured before Sample Preparation)

<table>
<thead>
<tr>
<th>Buffer Composition</th>
<th>pH</th>
</tr>
</thead>
<tbody>
<tr>
<td>50 mM Tris / 500 mM NaCl / 500 mM imidazole</td>
<td>7.5</td>
</tr>
<tr>
<td>250 mM imidazole in PBS</td>
<td>7.4</td>
</tr>
<tr>
<td>50 mM Tris / 10 mM gluthathione</td>
<td>8.0</td>
</tr>
<tr>
<td>20 mM Tris / 100 mM NaCl / 30 mM reduced glutathion</td>
<td>7.4</td>
</tr>
<tr>
<td>50 mM MgCl$_2$ in PBS</td>
<td></td>
</tr>
<tr>
<td>6 M urea in PBS</td>
<td></td>
</tr>
<tr>
<td>25 mM HEPES / 150 mM NaCl</td>
<td>7.5</td>
</tr>
<tr>
<td>20 mM NaAc</td>
<td></td>
</tr>
<tr>
<td>25 mM NaF</td>
<td></td>
</tr>
<tr>
<td>200 mM (NH$_4$)$_2$SO$_4$</td>
<td></td>
</tr>
<tr>
<td>25 mM PIPES pH 7.0</td>
<td></td>
</tr>
<tr>
<td>100 mM Tris/150 mM sodium citrate pH 7.5</td>
<td></td>
</tr>
<tr>
<td>1 M NaCl (it might happen that the upper marker decreases)</td>
<td></td>
</tr>
<tr>
<td>PBS pH 7.4</td>
<td></td>
</tr>
<tr>
<td>10 mM HCl</td>
<td></td>
</tr>
<tr>
<td>10 mM NaOH</td>
<td></td>
</tr>
<tr>
<td>10 mM EDTA</td>
<td></td>
</tr>
</tbody>
</table>
## List of Compatible Buffers and Buffer Compounds

### Protein 80 Sample Well Results

<table>
<thead>
<tr>
<th>Detergents</th>
<th>Possible Effects</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 % Triton X-100 in PBS pH 7.4</td>
<td>broad system peak, decreases sizing range for smaller proteins, reproducibility of quantitation might be affected, effect is less pronounced when a protein is present</td>
</tr>
<tr>
<td>0.25 % Tween 20 in PBS pH 7.4</td>
<td>broad system peak, decreases sizing range for smaller proteins, reproducibility of quantitation might be affected</td>
</tr>
<tr>
<td>0.375 % zwittergent E3-14 in PBS pH 7.4</td>
<td>large system peak, impairs sizing range</td>
</tr>
<tr>
<td>0.5 % sarcosyl in PBS pH 7.4</td>
<td>baseline artefact, slight hump appears around 15 kDa, less problematic if a protein is added</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Other additives</th>
<th>Possible Effects</th>
</tr>
</thead>
<tbody>
<tr>
<td>40 % acetonitril + 0.05 % formic acid</td>
<td>precipitates SDS, upper marker decreased, quantitation might be affected, maybe more spike in the baseline</td>
</tr>
<tr>
<td>1 % SDS</td>
<td>more diverse system peak, dip after system peak might be larger</td>
</tr>
<tr>
<td>10 % DMSO</td>
<td>no observations</td>
</tr>
<tr>
<td>30 % glycerol in PBS</td>
<td>no observations</td>
</tr>
<tr>
<td>50 mM guanidine</td>
<td>compatible at low concentrations, at higher concentrations than 50 mM guanidine precipitates SDS and quantitation is affected</td>
</tr>
<tr>
<td>20 % methanol + 0.25 % formic acid</td>
<td>precipitates SDS, upper marker decreased, quantitation might be affected</td>
</tr>
<tr>
<td>1 % PEG 2000 (polyethylene glycol)</td>
<td>leads to three baseline artefacts of approximately 25, 45, and 58 kDa size, reproducibility of quantitation might be affected</td>
</tr>
</tbody>
</table>
In This Book

you find the procedures to analyze protein samples with the Agilent Protein 80 reagent kit and the Agilent 2100 Bioanalyzer instrument.