

**Agilent  
G2721AA/G2733AA  
Spectrum Mill MS  
Proteomics Workbench**

**Installation Guide**



**Agilent Technologies**

# Notices

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## Software Revision

This guide is valid for the B.06.00 revision or higher of the Agilent G2721AA/G2733AA Spectrum Mill MS Proteomics Workbench software, until superseded.

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### CAUTION

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## In This Guide...

This guide contains information to install the Spectrum Mill MS Proteomics Workbench on a server system.

Instructions for setting up the client system to connect to the Spectrum Mill MS Proteomics Workbench server is found in the *Spectrum Mill MS Proteomics Workbench Quick Start Guide*.

### **1 Preparation for Installation**

In this chapter, you prepare your system for installation. You set up the hardware, check the software requirements, and download the proteomics database.

### **2 Software Installation**

In this chapter, you install the Spectrum Mill MS Proteomics Workbench software.

### **3 Troubleshooting**

This chapter contains troubleshooting information for installation and basic operation.

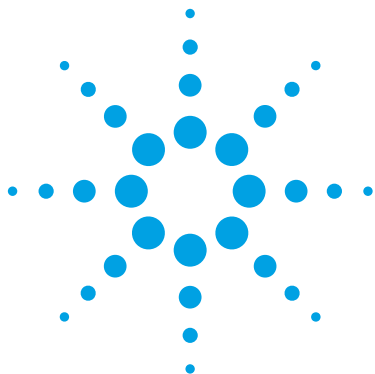


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# 1 Preparation for Installation

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This chapter describes the steps that you, the customer, need to prepare to install or upgrade the Spectrum Mill Workbench on the server. Instructions for setting up the client system can be found in the Spectrum Mill Workbench *Quick Start Guide*.

Spectrum Mill Workbench supports the analysis of Agilent LC/MS Trap data files (\*.d), and Agilent LC/MS Q-TOF data files (\*.d). You can also purchase additional Data Extractors to support other data types:

- *G2722AA Data Extractor for Thermo Finnigan LCQ* to support \*.RAW files
- *G2723AA Data Extractor for Generic Peak Lists* to support Waters Q-ToF \*.PKL files, written by ProteinLynx, and .MGF files that can be exported by other instrument software.



## 1 Preparation for Installation

### To upgrade your Spectrum Mill Workbench

In addition, if you purchased the G2721AA license, you can purchase a second license (G2729AA) to install on a PC server with more than two CPUs, or to install on additional server PCs.

## To upgrade your Spectrum Mill Workbench

- If your current operating system is not supported by Spectrum Mill Workbench, upgrade on a different computer that does.
  - Follow the instructions in this guide to do a new installation on the new computer.
  - Move your data and license to the new computer. See [“To move Spectrum Mill Workbench to a different computer”](#) on page 46.
- To update Spectrum Mill Workbench on a computer with supported operating system, see [“To upgrade or reinstall your Spectrum Mill MS Proteomics Workbench”](#) on page 43.

The supported operating system are Windows 7 Professional (64-bit), Windows 10 Professional (64-bit), Windows Server 2008 R2, Windows Server 2012 R2, and Windows Server 2016.



## Step 1. Unpack and check the software kit

If any items are missing, contact Agilent Technologies.

- 1 Check that you have all the materials listed below:
  - Spectrum Mill software disc
  - *Installation Guide*
  - *Quick Start Guide*
  - *Application Guide*
  - *Familiarization Guide*
  - *Software User License Certificate*
- 2 If you purchased an additional extractor, or an additional product license, check that you also have the appropriate *Software User License Certificate*.

The Spectrum Mill software disc contains:

- Spectrum Mill software
- Sample database NCBIInr.stdmix, and other databases, used in the *Familiarization Guide*
- Example data
- Manuals in PDF format

## Step 2. Check that you have the supported server hardware

- Check that you have the supported server hardware.

Check with Agilent Technologies for specifics. See the *MassHunter Spectrum Mill Site Preparation Checklist* (p/n G2721-90058).

## 1 Preparation for Installation

### Step 3. Set up the preconfigured Agilent Spectrum Mill Server

## Step 3. Set up the preconfigured Agilent Spectrum Mill Server

Do this step only if you are installing Spectrum Mill MS Proteomics Workbench on a preconfigured Agilent Spectrum Mill Server. Skip this step for upgrades.

This step has two sections: Windows 10 and Windows Server.

### Windows 10

While written for Windows 10, many of the steps in this section apply to Windows Server 2016 as well. The main differences between Windows 10 and Windows Server 2016 are:

- Server Manager runs only with Windows Server
- Windows Server 2016 predefines the Administrator user and you define a password, while for Windows 10, you create both a user that is added to the Administrators group and you add a password for that user.

Below are the steps for Windows 10:

- 1 Activate any licenses. (This step is automatic on preconfigured Windows 10 systems).
- 2 When asked who owns this computer, click **Work** (most likely) or **Personal**. (Follow the recommendation of your IT administrator.)
- 3 Choose how you will connect to this computer. Click **Join a local Active Directory domain**, or follow the recommendation of your IT administrator.
- 4 Create an account for this computer.
  - Type the user name, password, and a hint.
  - Record them for future reference.Observe that Windows starts.
- 5 Read the agreement that is displayed and click **I agree** to continue.
- 6 Rename the computer and/or add it to a domain. (This step is optional, depending on the recommendation of your IT administrator.)
  - a Right-click the Windows Start button.
  - b Click **System**.
  - c In the **Computer name, domain, and workgroup settings** section, click **Change Settings**.
  - d Type a valid DNS name.
  - e Reboot.

## Step 3. Set up the preconfigured Agilent Spectrum Mill Server

**Windows Server**

Depending on the version of Windows Server that is preconfigured (2008 or 2016), the steps might be slightly different than those below. For Windows Server 2016, refer to the Windows 10 instructions (just above) where applicable.

- 1 Set up and start the computer.
- 2 Follow the instructions in the Windows Setup Wizard.
- 3 If you are prompted for a license key, locate the Windows product key that came with your system and type the number in the screen. Click **Next**.
- 4 In the Licensing Modes screen, click **Next** to accept the default values.
- 5 In the Computer Name and Administrator Password screen, type a computer name and password. Click **Next**.

Write down the password and keep it in a safe place.

For Windows Server 2016, the user name is Administrator and cannot be changed.

- 6 In the Date and Time Setting screen, make any needed changes and click **Next**.
- 7 In the Network Settings screen, click **Next** to accept the default values.
- 8 In the Workgroup or Computer Domain screen, check with your Information Technology department to determine how to set the values.  
The computer reboots at this time.
- 9 After the computer reboots, log in with the administrator password that you set earlier in the set-up.
- 10 For Windows Server 2016, the Server Manager runs. Allow it to complete.
- 11 For Windows Server 2016, rename the computer and/or add it to a domain. (This step is optional, depending on the recommendation of your IT administrator.)
  - a Right-click the Windows Start button.
  - b Click **System**.
  - c In the **Computer name, domain, and workgroup settings** section, click **Change Settings**.
  - d Type a valid DNS name.
  - e Reboot.

## 1 Preparation for Installation

### Step 3. Set up the preconfigured Agilent Spectrum Mill Server

- 12** In the Windows Server Post-Setup Security Updates page, follow the instructions if the computer is connected to a LAN. Otherwise, click **Finish**.  
You can also do this step at a later time. If you selected to skip this step, you may be warned about computer risks.
- 13** Read the agreement that is displayed and click **I agree** to continue.
- 14** Close the Manage Your Server Window when it is displayed.
- 15** Continue at “[Step 9. Install the off-line data analysis program](#)” on page 18.

## Step 4. Configure the server

### CAUTION

Your computer and domain names must comply with Domain Name Services naming conventions. Use only letters, numbers and hyphens. Start each name with a letter, and do not end the name with a hyphen. Preconfigured servers are named "SpectrumMill."

Skip this step if you are installing on a preconfigured Agilent Spectrum Mill Server computer.

- 1 Verify that you have Windows Server 2008 R2, Windows Server 2012 R2, Windows Server 2016, Windows 7 Professional 64-bit, or Windows 10 Professional 64-bit.
  - a Click **Start > Run**. (For Windows 10 and Windows Server 2016, open the Start menu, click **All apps**, scroll down to and click the **Windows System** folder, then click **Run**.)
  - b In the **Open** text field, enter `winver`.

Windows Server requires Client Access Licenses (CALs) for authenticated server access by remote clients. Windows does not require CALs, but does have a fixed limit of remote connections. For more details, see the *Site Preparation Checklist* or contact your Agilent representative.
- 2 Upgrade your operating system with the latest service packs and security updates.
- 3 Configure the server hard drive. Refer to Windows online Help for instructions.
  - a Create an extended partition for the entire unformatted space.
  - b Do a quick format.
  - c Create logical partitions as listed in [Table 1](#).

[Table 1](#) lists the recommended configuration. You can also put the Data on the D: drive with the Database, or all files on the C: drive. For example, it is fine to have a C: volume for Windows and related programs, and a D: volume for Spectrum Mill and protein databases.

## 1 Preparation for Installation

### Step 4. Configure the server

**Table 1** Server hard disk configuration

Partition	Label	Size	Comments
C: (Primary)	Programs	100 to150 GB	
D: (Extended)	Database	100 GB	If you use NCBI FASTA database files, consider increasing the size of this volume. Because the drive stores sequence databases, it will be the least fragmented.
E: (Extended)	Data	rest of disk	This drive will be larger if the hard disk is greater than 220 GB. This volume should be the largest, because it will contain the data.

## Step 5. Set up the server software

Skip this step if you are installing on a preconfigured Agilent Spectrum Mill Server computer.

### CAUTION

Do not install the Spectrum Mill Workbench software on a server that runs the Matrix Science Mascot search engine or on any Apache-based web server.

---

- 1 Check that the name of the server is a valid domain name.

Domain names must use only letters, numbers and hyphens. They must start with a letter, and cannot end with a hyphen.

### CAUTION

Do not use an underscore in the server name or the Spectrum Mill software will not function properly.

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- 2 Check that you can connect to the internet.

- 3 Create the temporary folder **C:\temp**.

You will use this folder to run installation programs that you download.

- 4 If you have Windows 7 or Windows Server 2008, check that you have Internet Explorer 11.0:

- a Open Internet Explorer.
- b Click **Help > About Internet Explorer**.

Note that the Microsoft Edge browser cannot be used with Spectrum Mill.

- 5 If you need to upgrade your Microsoft Internet Explorer:

- a Go to <http://www.microsoft.com>.
- b Locate the download files for the latest version of Microsoft Internet Explorer.
- c Follow the instructions on the Web site to download and install the upgrade.

## 1 Preparation for Installation

### Step 6. Install IIS

## Step 6. Install IIS

Skip this step if you are installing on a preconfigured Agilent Spectrum Mill Server computer.

- 1 On the installation disc, open the folder **IIS Setup Scripts**.
- 2 Copy all of the files in that folder to a temporary folder on your local hard drive.
- 3 Install IIS:
  - a From the temporary folder on your local hard disk, right-click the file **DISM-IIS.bat**, and then select **Run as Administrator**.

This step can take several minutes.
  - b After installation, review the results, then close the window.
  - c From an Internet browser, go to <http://localhost> to verify that the IIS is properly installed. The IIS welcome screen appears if properly installed.




## Step 7. Install Java Runtime Environment for Windows

Skip this step if you are installing on a preconfigured Agilent Spectrum Mill Server computer.

- 1 If an older version of Java is installed, remove the program. If older 32-bit and 64-bit versions are installed, remove them both.
- 2 On the installation disc, in the **JAVA** folder, right-click the file in which the file name contains **i586**, and then click **Run as Administrator**.
- 3 Follow the instructions to install the Java Runtime Environment to the default location.

If you are warned that a newer version of Java is available, continue anyway.

Upon completion, the Java installer will open the browser to verify the Java version. Note that on Windows 10 this step runs the Microsoft Edge browser, which does not support add-ons. Simply close the browser and continue.

- 4 If you are updating from a different version:
  - a Click **Start > Control Panel > Programs > Java**. (For Windows 10, click the **Control Panel** icon , then click **Programs**, then click **Java**.)
  - b In the Java Control Panel, click the **Java** tab, then click **View**.
  - c In the Java Runtime Environment Settings dialog box, make sure that the version that you just installed is enabled.
- 5 Repeat all of these steps for the file in which the file name contains **x64**.

## Step 8. Install PERL

Skip this step if you are installing on a preconfigured Agilent Spectrum Mill Server computer.

- 1 On the installation disc, in the **PERL** folder, double-click the file appropriate for your operating system. For 64-bit operating system, use the file that contains **x64** in the name.
- 2 Follow the instructions to install PERL to the default location of **C:\PERL**. Continue to click **Next** or **Install** until the installation is done.
- 3 From the temporary folder on your local hard disk (into where you copied files from the folder **IIS Setup Scripts**), right-click this file, and then select **Run as Administrator**.
  - **Perl64IIS.bat**

## Step 9. Install the off-line data analysis program

- 1 Install the off-line version of data analysis as appropriate for your data type. See [Table 2](#).

Note that Spectrum Mill does not directly support MassHunter TOF .d data files. You can use MassHunter Qualitative Analysis program to first create a mass list, on which you can run a Manual PMF search. See the *Familiarization Guide* for more information.

### NOTE

You must have an additional license from the vendor to install an off-line copy of the data analysis software onto the Spectrum Mill server. Contact the vendor to purchase your license.

**NOTE**

To extract ThermoFisher Scientific (.raw) data, you must download and install the 64-bit version of the MSFileReader.

Spectrum Mill B.06.00 was tested with MSFileReader 2.2.62 and 3.0.29. Later versions may be incompatible.

Go to <https://thermo.flexnetoperations.com/control/thmo/product?plneID=632401> and follow the instructions to install the MSFileReader program. *Make sure to install the 64-bit version.*

MSFileReader can be installed before or after you install Spectrum Mill.

If Spectrum Mill is unable to create the rawfile2 object when you try to extract .raw files, then it is likely that MSVC Runtime needs to be installed, or the MSFileReader installation may not have completed properly. Contact ThermoFisher Scientific.

**Table 2** Required Data Analysis Software

Data Type	Data Analysis Software
Agilent 6300 Series Ion Trap LC/MS (*.d)	none needed
Agilent 6500 Series Quadrupole Time-of-Flight LC/MS (*.d)	none needed
Thermo Finnigan LCQ (*.RAW) LTQ (*.RAW) LTQ-Orbitrap (*.RAW) LTQ-FT (*.RAW) LTQ-ETD (*.RAW)	XCalibur (must be same or higher version than the one used to acquire data) or MSFileReader (64-bit version required).
Waters QTOF (*.pkl)	none needed

## Step 10. Run Windows Update

- 1 Run Windows Update, if you have not already done so.
- 2 Reboot the computer.

## Step 11. Download a database

- If you want to use the yeast or *e. coli* specific SwissProt database that is mentioned in the Spectrum Mill user guides, copy them from the UniProt Web site to the **C:\temp** folder. See [Table 3](#).
- If you want to use one or more supported databases that is available on the Internet, copy the database files to the **C:\temp** folder. See [Table 3](#).

At this time, just download the zipped data base files. Do not unzip them yet.

You can download the databases to another computer while you continue the installation of the Spectrum Mill MS Proteomics Workbench.

The SwissProt database is recommended over the NCBI because of the large size of the NCBI database.

### NOTE

If you download a commercial database, any subscription fee that is due to the database provider is the responsibility of your company or institution.

**Table 3** Supported data bases

Database	URL	File to download and unzip	Extract and rename with prefix
NCBI non-redundant	<a href="ftp://ftp.ncbi.nlm.nih.gov/blast/db/FASTA">ftp://ftp.ncbi.nlm.nih.gov/blast/db/FASTA</a>	nr.gz*	NCBIgb
SwissProt	<a href="http://www.uniprot.org/downloads">http://www.uniprot.org/downloads</a>	UniProtKB/Swiss-Prot > fasta	SwissProt
SwissProt (species-specific)	<a href="http://www.uniprot.org/downloads">http://www.uniprot.org/downloads</a>	Proteomes > fasta	SwissProt or UniProt
TrEMBL	<a href="http://www.uniprot.org/downloads">http://www.uniprot.org/downloads</a>	UniProtKB/TrEMBL > fasta	TrEMBL

\* This database can be excessively large. To improve performance, use the Spectrum Mill Protein Databases tools to create a species-specific database. The NCBI FASTA header format changed as of September 2016 and now uses GeneBank (gb) identifiers instead of gi. The older format is still supported.

## Step 12. Configure Internet Explorer

### NOTE

On Windows 10 and Windows Server 2016, to access Internet Explorer (and not the Edge browser):

- a) Type “Internet Explorer” in the search area on the Task Bar.
  - b) Select Internet Explorer (Desktop) to run. You may also right-click and add it to the Task Bar for easier access.
- 

#### 1 Disable Compatibility View.

- a Click **Tools > Compatibility View settings**.
- b Remove the server if it is listed in as a Compatibility View site.
- c Clear the check box for **Display intranet sites in Compatibility View**.
- d Click **Close**.

#### 2 Click **Tools > Internet options**.

If you do not see the menu bar, press the **Alt** key.

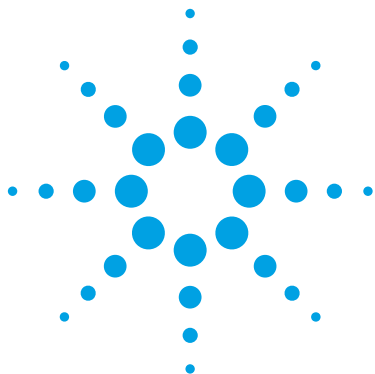
#### 3 Enable cookies so you can save form settings. (Parts a and b of this step are not required for Windows 10 or Windows Server 2016.)

- a Click the **Privacy** tab
- b Set the slider to **Low**, or click the **Advanced** button and enable cookies.  
For more detailed instructions to enable cookies, search Internet Explorer help for cookies.
- c If the drop-down menus in the Spectrum Mill workbench appear empty, you may need to add the Spectrum Mill server as a trusted site. Search Internet Explorer help for trusted site.

#### 4 Enable JavaScript (Active Scripting). (This step is not required for Windows 10 or Windows Server 2016.)

- d Click the **Security** tab.
- e Select **Local intranet**.
- f Click the **Custom level...** button.
- g In the Security Settings dialog, scroll down until you see the **Scripting** section.
- h Under **Active scripting**, click the **Enable** option.

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## 2 Software Installation

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This chapter describes the steps to install Spectrum Mill Workbench software on a server system.

To upgrade your Spectrum Mill MS Proteomics Workbench, see [“To upgrade or reinstall your Spectrum Mill MS Proteomics Workbench”](#) on page 43.

The client system requires no specific software installation. To set up the client system to access Spectrum Mill Workbench through Internet Explorer, refer to the Spectrum Mill Workbench *Quick Start Guide*.



## Installation

This section contains instructions for installing the Spectrum Mill Workbench software onto the server system.

If you are upgrading a previous installation of the Spectrum Mill Workbench software, go to [“To upgrade or reinstall your Spectrum Mill MS Proteomics Workbench”](#) on page 43.

### Step 1. Install Spectrum Mill Workbench on the server

Do this step on only the server system.

- 1 Insert the installation disc into the disc drive.
- 2 From the disc drive, run **setup.exe**.
- 3 If you are asked to install .NET 2.0, click **No**.
- 4 If the .NET 4.51 Installation panel appears, click **Install**, accept the agreement, and click **Next** in each screen to accept default values until .NET 4.51 is installed.

After the system reboots, the installation continues.

- 5 In the welcome screen, click **Next**.
- 6 If you get a message that says you have an unsupported version of PERL, then remove PERL from your system and install it according to the instructions in [“Step 8. Install PERL”](#) on page 18.
- 7 If your server does not have the recommended disk volume configuration, then you will be warned. Click **Yes** to continue, or click **No** to partition your drive volumes to the recommended configuration. See the *MassHunter Spectrum Mill Site Preparation Checklist* (p/n G2721-90058) for details.
- 8 Carefully read the License Agreement, particularly the “Confidentiality Terms and Conditions” section.

If you have purchased the software, read the on-line agreement carefully as this agreement may differ from the one in your purchase agreement. If you are installing an evaluation copy of this software, refer to the terms that you received separately from the software.

- 9 If you accept the terms, click **Yes** to continue.



## Step 1. Install Spectrum Mill Workbench on the server

- 10 On the Spectrum Mill Software and MS Data Files Location page, click the drive with the most amount of disk space, then click **Next**.

The Spectrum Mill Workbench software and data must reside on the same partition. The installation program defaults to the largest partition because data requires the most space for storage. You do not see this page of installation if you have only one partition.

- 11 On the Proteomics Database Location page, click the drive with the second most amount of space available, then click **Next**. If the disk has three partitions, avoid the use of C: for SeqDB.

If possible, do not choose the same drive as the one used to store the Spectrum Mill program and data files. The folder **\SeqDB** is created, and the example databases are installed into this folder. You will not see this page of installation if you have less than three partitions on the server.

Depending on your requirements, the disk space required for your database can grow to a range of 50 to more than 100 GB.

## Step 2. Request a License Key

Spectrum Mill is installed with a 30 day Temporary License. You must get a valid License Key to use the software after this period.

After you install the Spectrum Mill Workbench software, the Spectrum Mill License Activation program automatically runs. If you want to run this program outside of the installation process, click **Start > Programs > Agilent Spectrum Mill Software > Spectrum Mill License Activation**. (For Windows 10 and Windows Server 2016, click **Start > All Apps > Agilent Spectrum Mill Software > Spectrum Mill License Activation**.)

- 1 In the **Activate Spectrum Mill Modules** page:
  - a Mark the check box for each module that you purchased.
  - b Click **Activate**.
- 2 For each module that you purchased:
  - a Click **Delay**.

### NOTE

You must click **Delay**. If you do not, the License Key that you receive from the Session Key that is generated will not work.

- b Click **Next** if you have more modules, or click **OK** when you are done.
- 3 **IMPORTANT:** In the Notepad window that appears, add to the content the **Sales Order number** that was used to purchase your Spectrum Mill licenses.
- 4 Save the Notepad file and send it to [smlicense@agilent.com](mailto:smlicense@agilent.com).
- 5 Store your **Sales Order number** with your Spectrum Mill installation discs in a safe place.

When you receive your License Key, activate the license. See “[To activate the Software User License](#)” on page 45. Note that multicore and hyperthreaded CPUs do not count towards the CPU count. Only the physical CPU count is used for core licensing.

## Step 3. Configure security for Spectrum Mill Web Access

By default, Spectrum Mill configures IIS for Anonymous access. The Internet guest account (IUSR) has all the appropriate permissions to access Spectrum Mill. *Do this step only if Integrated Windows Authentication is required by your organization.*

- 1 Open an elevated command window.
- 2 Go to the `\SpectrumMill\milladmin` folder.
- 3 For each user who needs access:
  - Run `smSetPermissions.pl`.
  - Run `smSetmsdataSMPermissions.pl`.

Read the comments inside of these script files for details. Contact your IT department to assist in the proper configuration of security.

- 4 Close the command window.

These scripts can be customized. See the `readme.txt` file in the `\SpectrumMill\milladmin` folder for additional information.

With Integrated Window Authentication, a logon is required to access the Spectrum Mill site. You can set Windows file and directory access permissions for directories under `\SpectrumMill\msdataSM` so that access to data is restricted for only those users and groups that have read and write permissions. Set the permission on a directory and all of its subdirectories, with the `msdataSM` directory accessible by all users.

### CAUTION

If you use Integrated Windows Authentication, then change the system to allow Anonymous Access. Users will possibly no longer be able to access files on the server until the ownership is changed to that of the Anonymous user as the user IUSR. Go to the Microsoft web site for more information.

---

For more information on configuring security on the Microsoft Internet Information Services (IIS), refer to <http://www.iis.net>.

## 2 Software Installation

### Step 4. Finish the installation on the server

## Step 4. Finish the installation on the server

- 1 Click the Spectrum Mill icon on the desktop. You can instead use Internet Explorer to go to <http://yourcomputer.yourdomain/millhome.htm> to verify that you can connect to the Spectrum Mill home page. Do not use “localhost” or a loopback IP address.

The **Spectrum Mill** icon on the Desktop also connects to the home page.

### CAUTION

The Spectrum Mill Workbench program will only work properly if you access it through its http address, and not as a shared drive.

- 2 If you get a message to disable Compatibility View, click **OK**, and then turn off Compatibility View for the millhome.htm page.

See [step 1](#) of “[Step 12. Configure Internet Explorer](#)” on page 21 for details.

- 3 Check that the server, PERL, and CGI.pm are working:

- a Open the site <http://yourcomputer.yourdomain/millhtml/hello.htm> with your browser.

- b Click **Print PERL is working**.

If PERL is working correctly, you will see these statements:

**Hello World.**

**If you can see this PERL is working.**

**If you can see this CGI.pm is working.**

If you get a message that PERL is not working, see “[If PERL does not work](#)” on page 51.

- 4 If you plan to use a default instrument to generate MS/MS data other than the Agilent Q-TOF LC/MS:

- a Open the file **E:\SpectrumMill\ millhtml\SM\_js\SMglobals.js** with a text editor, such as Notepad.

- b Replace **ESI-QTOF-AGILENT** as the value for **defaultInstrument** with the appropriate instrument variable, such as **ESI-ION-TRAP-AGILENT**.

**5** Install the Example Data:

- a** Insert the Example Data DVD into the media drive and run **setup.exe**.
- b** Mark the check box for each of the example data files that you want to install.
- c** Click **Next**.

The data files are copied to **SpectrumMill\msdataSM\ExampleData** folder. You can also access the data files on the installation disc, in the **ExampleData** folder.

Note that the example data files for Thermo Finnigan are already extracted. You can extract these files again if you have the appropriate extractor license, and have installed and configured the data analysis software for that data.

- d** In the Start Copying Files page of installation, review the summary information and click **Next**.

If you have errors while you run or install the software, refer to [“Troubleshooting”](#) on page 49.

You can open the **readme.txt** file after installation in the **SpectrumMill\msdataSM\ExampleData** folder. This file describes the contents of the Example Data and what parameters to use when you process the data.

## 2 Software Installation

### Step 5. Install the databases

## Step 5. Install the databases

- 1 Install the databases that you downloaded into the **C:\temp** directory.
  - a Unzip the files into the **\SeqDB** folder. By default, this folder is on the **D:** drive.
  - b Rename the database files according to the names in [Table 3](#) on page 20.
  - c Delete the zipped files in the **C:\temp** directory to optimize the recovery image process.
- 2 Create the indices for each database in the **\SeqDB** folder, including the **NCBInr.stdmix** database, which is installed with the software:
  - a In the Spectrum Mill home page, under **Utilities**, click **Protein Databases** page.
  - b Select **Create indices for new database**.
  - c Check that you see the form shown [Figure 1](#).

Agilent Spectrum Mill - Protein Database Utilities

Spectrum Mill MS/MS Search PMF Search Tool Belt Help

Create indices for new database  Concatenate FASTA files  Compare two databases

Re-index existing database  Create subset with indices from saved hits  Calculate statistics

Create species subset database  Make subset FASTA file from Accession Numbers  Make Non-redundant

Database summary report  Create or append user database  Create category file

Update Database List (After a re-install or indexing a database, if you don't see the database listed, click the "Update Database List" button)

Create Indices

Newly downloaded database: NCBInr.stdmix

Existing databases: SwissProt

**Figure 1** Protein Database Utilities form.

- d Enter the *exact* name of the new database. Be sure to use capital and lower-case letters *exactly* as in the database name.
- e Click the **Create Indices** button.
- f As the indices are created, check that you see a display like the one shown in [Figure 2](#) on page 31.

Protein Database
Counting entries in database and creating index file (.idx). Processing entries to find species, accession number, and unknown sequence characters. Calculating Protein MW and pI and creating unknown sequence characters file (.unk). 18 entries in the database. 18 entries processed. 0 entries contained unreadable species. Creating mole_wt file (.mw). Creating pI file (.pI). Creating species file (.sp). Creating species list file (.sl). Creating accession_number file (.acn).  <b>Indexed Database:</b> NCBIInr.stdmix

**Figure 2** Screen to monitor database indices creation.

- g** Click **Back** to return to the Protein Database page.
- h** Click **Update Database List** and verify that the newly indexed database is in the list of databases.

## 2 Software Installation

### Step 6. Share the data folder (msdataSM) and database folder (SeqDB)

## Step 6. Share the data folder (msdataSM) and database folder (SeqDB)

You must share the folder **msdataSM** so that acquisition systems can copy files to the server. You can share the folder **SeqDB** if you want users to be able to update the databases on the server from a different computer.

### CAUTION

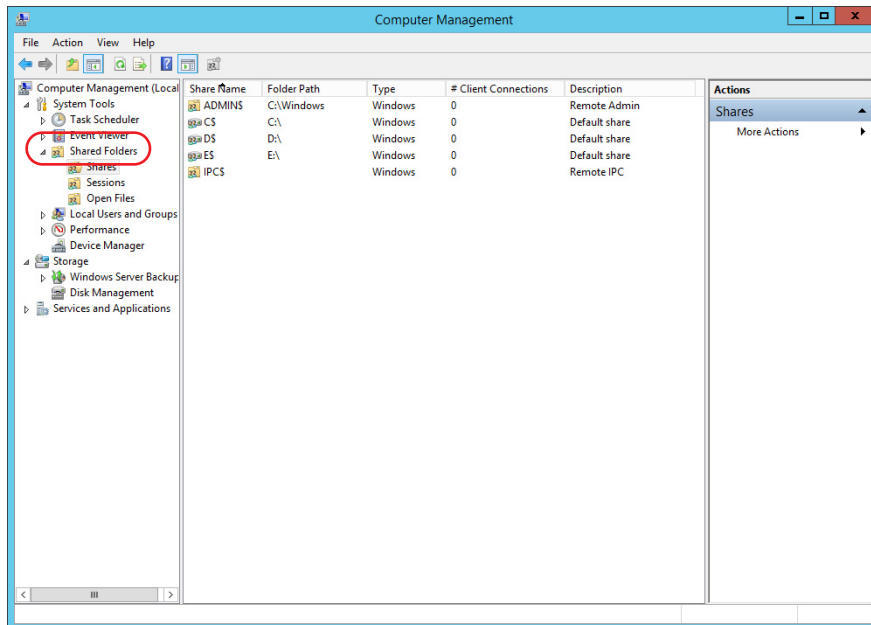
You must use the Computer Management console (Windows 7) or Server Manager (Windows Server) to share the **SeqDB** and **msdataSM** folders. If you share them in any other way, permission settings for Spectrum Mill files will be set incorrectly.

### Windows 10 and Windows Server 2012 and Windows Server 2016

- 1 Log in as an administrator.
- 2 Open the **Server Manager**.

Right-click the **Windows** button, and then click **Computer Management**.

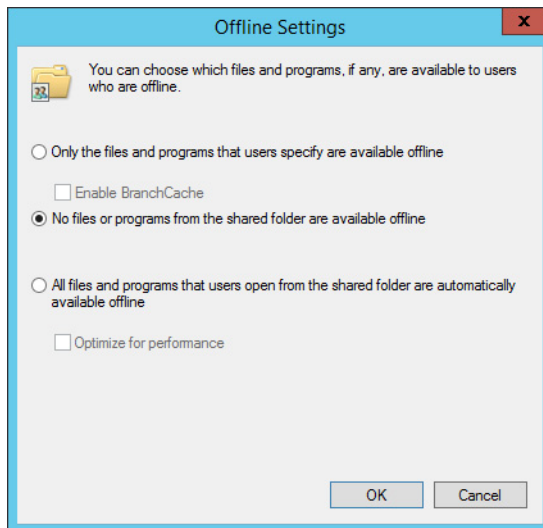
- 3 In the navigation pane, expand **Shared Folders**.





## Step 6. Share the data folder (msdataSM) and database folder (SeqDB)

- 4 Right-click **Shares**, and then click **New Shares**.
- 5 In the **Welcome to the Create a Shared Folder Wizard** page, click **Next**.
- 6 In the **Folder Path** page, click **Browse**, locate and select **SpectrumMill\msdataSM**, and then click **Next**.
- 7 In the **Name, Descriptions, and Settings** page, click **Change**.
- 8 In the **Offline Settings** page, select **No files or programs from the shared folder are available offline**.



- 9 Click **OK**, then click **Next**.

## 2 Software Installation

### Step 6. Share the data folder (msdataSM) and database folder (SeqDB)

**10** In the **Shared Folder Permissions** page, select the kind of permissions that you want for the shared folder.

To customize settings for your organization, click **Customize permissions**. See your system administrator or the Windows Help for more information.



**11** Click **Finish**.

## Windows 7

**1** Log in as an administrator.

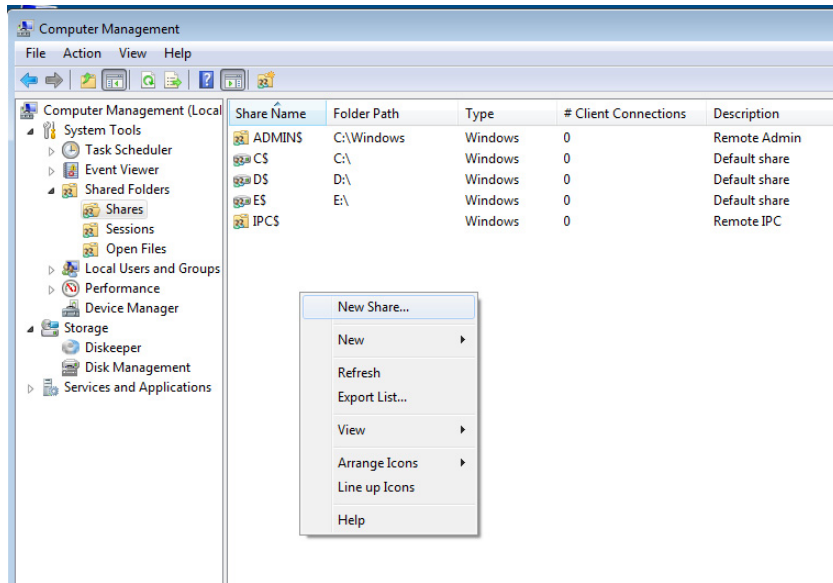
**2** Open the Computer Management center.

Click the **Start** button, right-click **Computer**, and then select **Manage**.

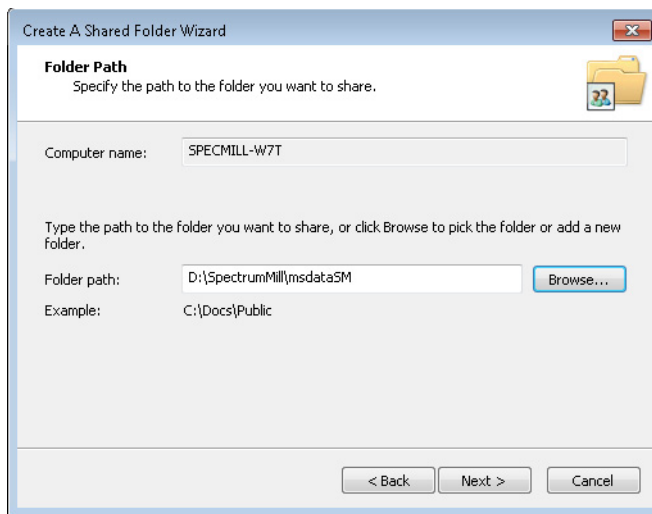
**3** In the navigation pane, click **System Tools > Shared Folders > Shares**.

**4** Right-click the pane that lists the shared folders and select **New Share**.

## Step 6. Share the data folder (msdataSM) and database folder (SeqDB)



- 5 In the **Create a Shared Folder Wizard** welcome page, click **Next**.
- 6 In the **Folder Path** page, locate and select **SpectrumMill\msdataSM**, click **OK**, and then click **Next**.

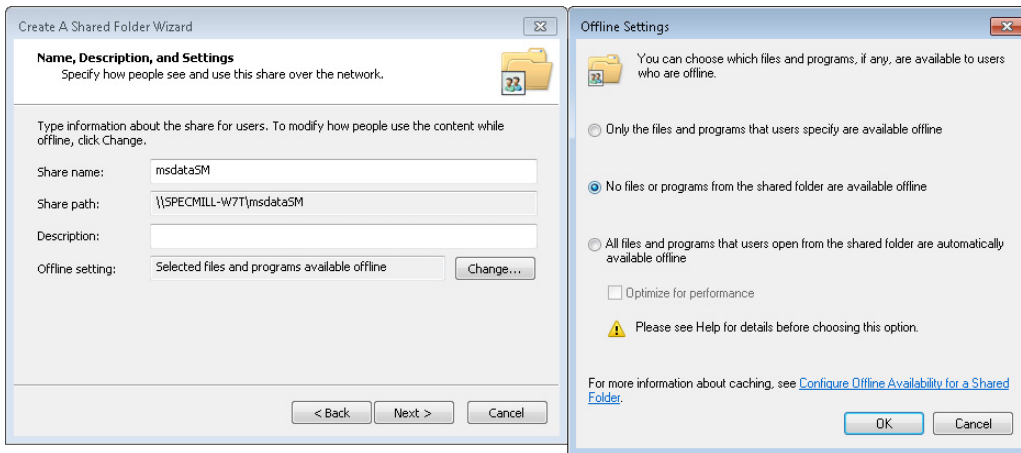


## 2 Software Installation

### Step 6. Share the data folder (msdataSM) and database folder (SeqDB)

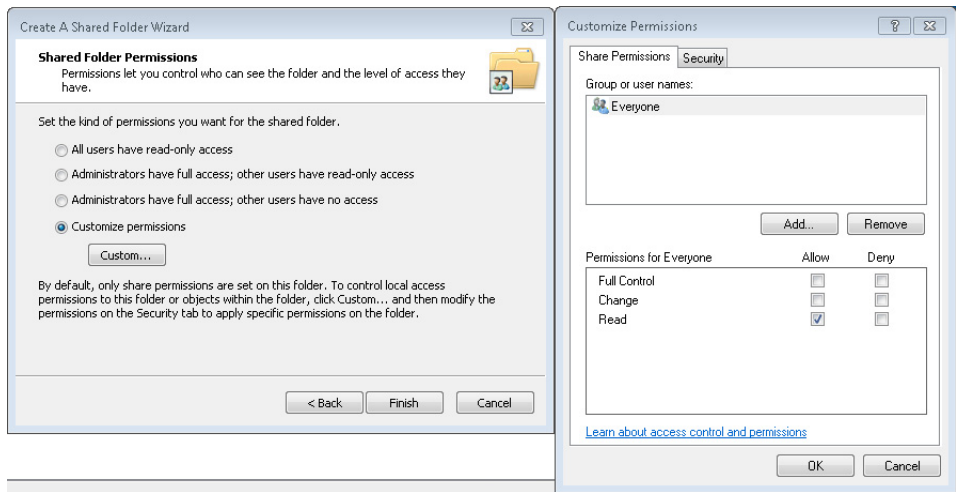
#### 7 In the **Name, Descriptions and Settings** page:

- a Click **Change**.
- b Select **No files or programs from the shared folder are available offline**.
- c Click **OK** to return to the **Name, Descriptions and Settings** page.
- d Click **Next**.



## Step 6. Share the data folder (msdataSM) and database folder (SeqDB)

- 8** In the **Shared Folder Permissions** page:
- a** Click **Custom permissions**, then click **Custom**.
  - b** Set permissions for groups or individual users.  
*Do not give Change permission to the group Everyone.*
  - c** Click **OK** to return to the **Shared Folder Permissions** page.
  - d** Click **Finish**.



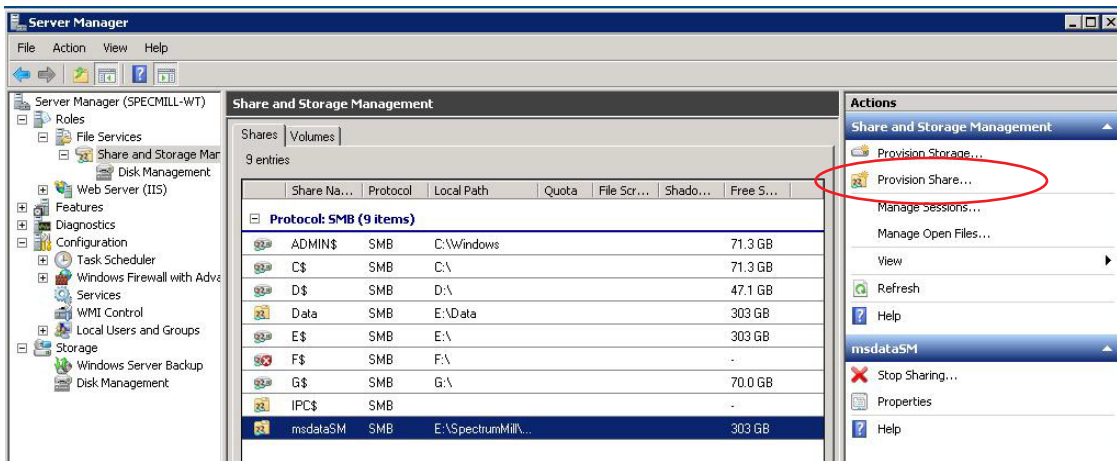
- 9** *Optional.* Repeat these steps to share the folder **SeqDB**.

## 2 Software Installation

### Step 6. Share the data folder (msdataSM) and database folder (SeqDB)

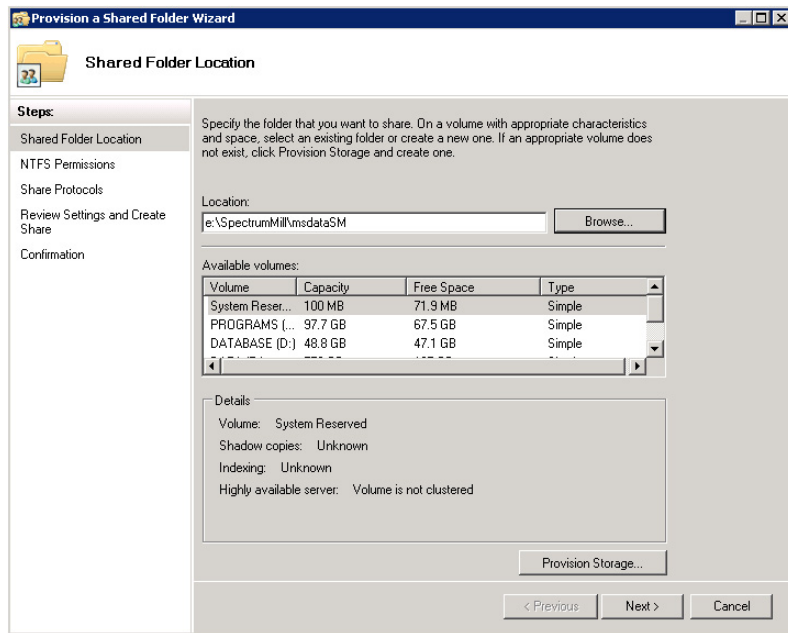
#### Windows Server 2008 R2

- 1 Log in as an administrator.
- 2 Open the Server Manager.  
Click the **Start** button, right-click **Computer**, and then select **Manage**.
- 3 In the navigation pane, click **Roles > File Services > Share and Storage Management**.
- 4 Under **Actions**, click **Provision Share**.



## Step 6. Share the data folder (msdataSM) and database folder (SeqDB)

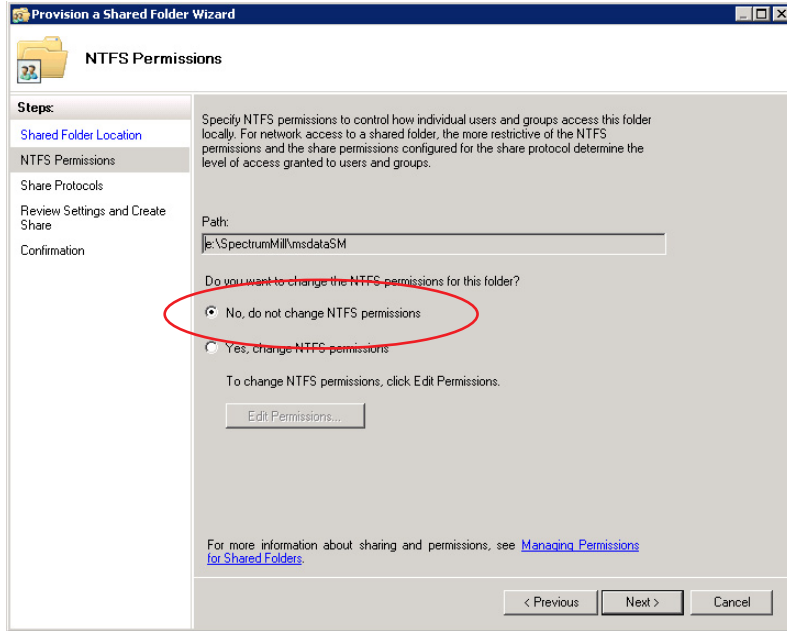
- 5 In the **Shared Folder Location** page, click **Browse**, locate and select **SpectrumMill\msdataSM**, and then click **Next**.



## 2 Software Installation

### Step 6. Share the data folder (msdataSM) and database folder (SeqDB)

- 6 In the **NTFS Permissions** folder, click **No, do not change NTFS permissions**, and then click **Next**.



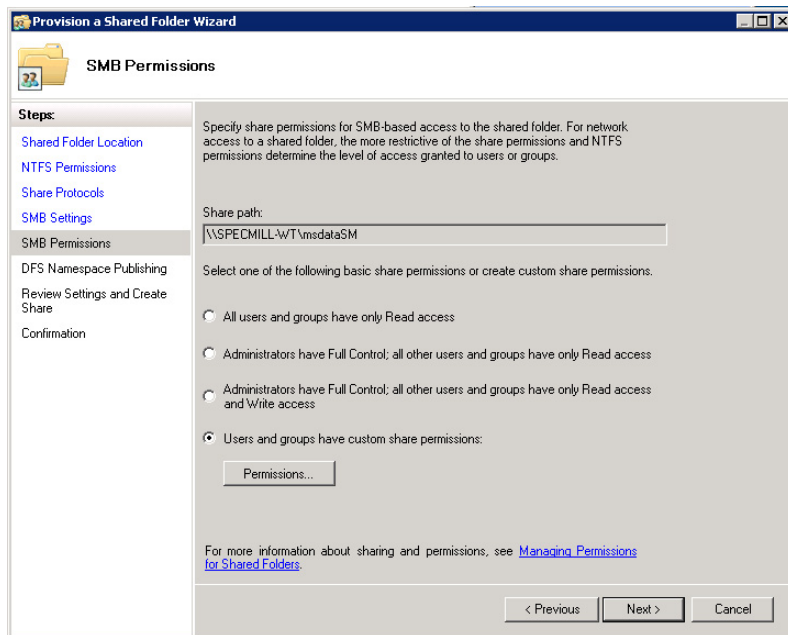
- 7 In the **Share Protocols** page, verify the **Share name**, then click **Next**.

- 8 In the **SMB Settings** page, add a description for the share, then click **Next**.



## Step 6. Share the data folder (msdataSM) and database folder (SeqDB)

- 9 In the **SMB Permissions** page:
- Click **Users and groups have custom share permissions**.
  - Click **Permissions**.
  - Set permissions for groups or individual users, and click **OK**.  
*Do not give Change permission to the group Everyone.*



10 In the **DFS Namespace Publishing** page, click **Next**.

11 In the **Review Settings and Create Share** page:

- Click **Create**.
- In the **Confirmation** message, click **Close**.

For more information, refer to the Windows Server online Help.

12 *Optional*. Repeat these steps to share the folder **SeqDB**.

## Step 7. Get started

- 1 Make sure your Agilent representative reviews these topics with you:
  - Directory structure
  - Restoring recovery images
- 2 Read the *Quick Start Guide*. Use the *Quick Start Guide* to:
  - Set up each client system.
  - Learn how to access the server from client systems.
  - Transfer data from the client to the server.
  - Get an overview of the Spectrum Mill software.
- 3 Review these exercises in the Familiarization Guide:
  - Chapter 1, “Basic Workflow - Interactive Processing of MS/MS Data,” up to and including Task 2, Exercise 1
  - Chapter 4, “Workflow Automation”
- 4 Sign and FAX the “[Customer Acceptance](#)” form on [page 53](#) to Agilent Technologies. Keep a copy for your records.
- 5 When you get your License Key from Agilent, activate your Software User License. See “[To activate the Software User License](#)” on page 45.

## Other Tasks

This section contains instructions related to the installation of the Spectrum Mill Workbench software and the activation and deactivation of the Software User License.

### To upgrade or reinstall your Spectrum Mill MS Proteomics Workbench

Do these steps if you are upgrading or reinstalling your Spectrum Mill MS Proteomics Workbench.

#### CAUTION

Do not upgrade your Windows operating system, unless the operating system is not supported by Spectrum Mill, or unless required by your organization. Contact your Agilent representative for more information.

---


**1** If needed, back up the folders that contain data and customized files:

- **\SpectrumMill\millauto** (parameter files)
- **\SpectrumMill\msparams\_mill** custom modifications)
- **\SpectrumMill\msdataSM** (data files)
- **\SpectrumMill\millhome.htm**
- **\SpectrumMill\millbin**
- **\SpectrumMill\millhtml**
- **\SpectrumMill\millscripts**
- **\SpectrumMill\milladmin**

The protein database, data and result files, and custom files are preserved when you remove Spectrum Mill Workbench from the computer. But make a backup copy to make sure that you do not lose anything important.

## 2 Software Installation

### To upgrade or reinstall your Spectrum Mill MS Proteomics Workbench

- 2 Remove Spectrum Mill MS Proteomics Workbench.
  - a Click **Control Panel > Programs and Features**. (For Windows 10, click the **Control Panel** icon , then under **Programs**, click **Uninstall a program**.)
  - b Right-click **Agilent SM** and click **Uninstall**. Continue as instructed to complete the removal of Spectrum Mill.

The protein database, data and result files, and custom files are preserved when you remove Spectrum Mill MS Proteomics Workbench from the computer. To make sure that you do not lose anything important, make a backup of these files.

- 3 *Optional*. Update Java Runtime Environment to the latest supported version:
  - a Uninstall Java Runtime Environment from your system.
  - b Follow the instructions in “[Step 7. Install Java Runtime Environment for Windows](#)” on page 17 to install the latest supported version.
- 4 If your current version of Spectrum Mill is lower than B.05.00, update PERL to the latest supported version:
  - a Uninstall PERL from your system.
  - b Follow the instructions in “[Step 8. Install PERL](#)” on page 18 to install PERL.
  - c Update your browser to Internet Explorer 11.0 or higher.
  - d Disable Compatibility View. See [step 1](#) of “[Step 12. Configure Internet Explorer](#)” on page 21 for details.
- 5 Install the Spectrum Mill MS Proteomics Workbench. See “[Step 1. Install Spectrum Mill Workbench on the server](#)” on page 24.
- 6 Update the Protein Database List.
  - a Start the Spectrum Mill Workbench software.
  - b Open the Protein Databases page.
  - c Click **Update Database List**.
- 7 Check your custom modifications:
  - a In the MS/MS Search page, click **Choose**.
  - b Make sure that the custom modifications are listed.If the modifications are not listed:
  - 1 Close the dialog box.

- 2 Delete `\SpectrumMill\msparams_mill\smconfig.xml`.
- 3 Click **Choose** to generate a new `smconfig.xml` file.

## To activate the Software User License

You have 30 days from the day that you installed the software to activate the license with the License Key.

To request a License Key, see “[Step 2. Request a License Key](#)” on page 26.

- 1 Click **Start > Programs > Agilent Spectrum Mill Software > Spectrum Mill Software License Activation**. (For Windows 10 and Windows Server 2016, click the **Start** icon, then click **All Apps > Agilent Spectrum Mill Software > Spectrum Mill License Activation**.)
- 2 In the Activate Spectrum Mill Modules page:
  - a Mark the check box for each module that you purchased.
  - b Click **Activate**.
- 3 For each module:
  - a Enter the **License Key** number that Agilent Technologies provided.
  - b If you are given a License Data number, then enter that number into the **License Data** field.
  - c Click **Next** if you have more licenses to activate, or click **OK** when you are done.

The core license includes the license to extract Agilent 6200 Series Time-of-Flight LC/MS data, Agilent 6300 Series Trap LC/MS and Agilent 6500 Series Quadrupole Time-of-Flight LC/MS data.

## To deactivate the Software User License

The license you activate is good only on the PC for which you requested a License Key. If you want to transfer the license to another PC, you need to first deactivate it from the original system.

- 1 Send an e-mail message to [smlicense@agilent.com](mailto:smlicense@agilent.com) to request a deactivation key.
- 2 Run the steps in “[To activate the Software User License](#)” on page 45, but use the deactivation key as the **License Key**.

The license on the system reverts to a 30 day Temporary License.

## To move Spectrum Mill Workbench to a different computer

### CAUTION

If you upgrade from a single processor computer to a dual processor computer, you must obtain the G2721AA Dual CPU License. Contact Agilent if you had only purchased the G2733AA license.

- 1 On the computer on which Spectrum Mill Workbench is currently installed, deactivate the license. See “[To deactivate the Software User License](#)”.
- 2 If you want to move data files and customized settings to the new computer, back up these files:
  - `\SpectrumMill\millauto` (parameter files)
  - `\SpectrumMill\msparams_mill` (custom modifications)
  - `\SpectrumMill\msdataSM` (data files)
  - `\SpectrumMill\millhome.htm`
  - `\SpectrumMill\millbin`
  - `\SpectrumMill\millhtml`
  - `\SpectrumMill\millscripts`
  - `\SpectrumMill\milladmin`

- 3 Install the Spectrum Mill Workbench software on the new system. Complete the steps in [“Installation”](#) on page 24.

Make sure you use the Computer Key and Session Key of the new system to request a new License Key. The original License Key works on only the original system.


- 4 Activate the license on the new system. See [“To activate the Software User License”](#) on page 45.
- 5 If needed, copy the backed-up files onto the new computer, in the same location as on the old computer.
- 6 If needed, remove Spectrum Mill Workbench from the old computer. See [“To remove the Spectrum Mill Workbench software”](#) on page 47.

## To remove the Spectrum Mill Workbench software

Do this procedure only if you want to completely remove Spectrum Mill Workbench from your computer.

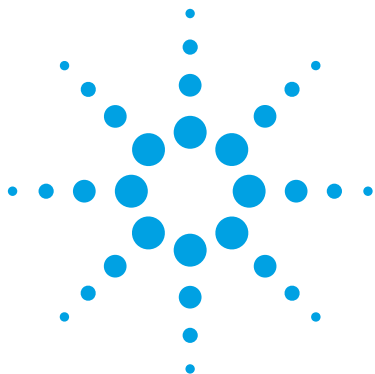
- 1 If you do not plan to reinstall the Spectrum Mill Workbench software on this system, deactivate the license. See [“To deactivate the Software User License”](#).
- 2 If needed, back up the folders that contain data and customized files:
  - `\SpectrumMill\millauto` (parameter files)
  - `\SpectrumMill\msparams_mill` custom modifications)
  - `\SpectrumMill\msdataSM` (data files)
  - `\SpectrumMill\millhome.htm`
  - `\SpectrumMill\millbin`
  - `\SpectrumMill\millhtml`
  - `\SpectrumMill\millscripts`
  - `\SpectrumMill\milladmin`

The protein database, data and result files, and custom files are preserved when you remove Spectrum Mill Workbench from the computer. But make a backup copy to make sure that you do not lose anything important.

- 3 Remove Spectrum Mill MS Proteomics Workbench.
  - a Click **Control Panel > Programs and Features**. (For Windows 10, click the **Control Panel** icon , then under **Programs**, click **Uninstall a program**.)
  - b Right-click **Agilent SM** and click **Uninstall**. Continue as instructed to complete the removal of Spectrum Mill.
- 4 If needed, disable IIS services:
  - a Click **Control Panel > Programs and Features**. (For Windows 10, click the **Control Panel** icon, then click **Programs**.)
  - b In the navigation pane, click **Turn Windows features on or off**.
  - c Clear the check box for **Internet Information Services** or for the individual IIS components that you want to disable.
- 5 Delete the files and folders listed in [step 2](#).

Uninstallation does not remove files that were added or modified after Spectrum Mill Workbench was installed. These files must be manually removed if you want to completely remove Spectrum Mill Workbench from the computer.





## 3 Troubleshooting

- If the Select Data Directory doesn't work 50
- If the Instrument and other drop-down menus are empty 50
- If PERL does not work 51
- If you have performance problems 51
- If you are prompted to download "faindex.cgi" 52
- If you are denied permission to update the database list 52

This chapter describes the problems that you might encounter when you set up your computer or install and use the Spectrum Mill Workbench.

Refer also to the online help for additional troubleshooting information. See "Tips and Tricks" at [http://your\\_server/millhtml/SM\\_instruct/tips.htm#IIS](http://your_server/millhtml/SM_instruct/tips.htm#IIS)

A list of known problems or issues for the Spectrum Mill Workbench software, along with possible solutions, is found on the Agilent web site at [http://www.chem.agilent.com/cs/Library/Support/Patches/SSBs/Spectrum\\_Mill.html](http://www.chem.agilent.com/cs/Library/Support/Patches/SSBs/Spectrum_Mill.html).



## If the Select Data Directory doesn't work

- ✓ Make sure that you are running Internet Explorer and not the Microsoft Edge browser.
- ✓ Check that you haven't exceeded the number of connections for which you have a license.
  - Close one or more connections to the server until the problem goes away.
- ✓ Check that PERL is working correctly.
  - See [“If PERL does not work”](#) on page 51.
- ✓ Check that all data files are under subfolders of **msdataSM**.
  - If Spectrum Mill finds data files in the msdataSM folder or in folders above that level, it will stop looking for data files and not find the ones that are under **msdataSM**.
- ✓ Check that security has been set up.
  - See [“Step 3. Configure security for Spectrum Mill Web Access”](#) on page 27.
- ✓ Check that cookies are enabled.

## If the Instrument and other drop-down menus are empty

- ✓ Check that Compatibility View is disabled. See [step 1](#) of [“Step 12. Configure Internet Explorer”](#) on page 21 for details.
- ✓ Cookies are not enabled in the browser, or the Spectrum Mill server is not set up as a trusted site.
  - See [step 3](#) of [“Step 12. Configure Internet Explorer”](#) on page 21 for more information.

## If PERL does not work

- ✓ Make sure that the IIS Perl mappings are correctly set:  
Run **Perl64IIS.bat** as administrator. See [step 3](#) under “[Step 8. Install PERL](#)” on page 18 for details.
- ✓ Make sure permissions are properly set for **millscripts** and **millbin**.  
From the **SpectrumMill\milladmin** folder, *as administrator*, run these PERL scripts:
  - smConfigureIIS.bat
  - smSetPermissions.pl

## If you have performance problems

Data directories contain very small files and over time, become heavily fragmented. This causes the speed of the extractor to decline.

- ✓ Check your memory usage. Use Task Manager to monitor memory usage.
- ✓ Check the RAID disks for any problems.
- ✓ Unless you have a solid state drive (SSD), defragment the data drive.
  - 1 Archive older data that is no longer active and remove it from the server.
  - 2 Right click **My Computer** on the Desktop, then click **Manage**. (If you have Windows 10, click the Windows button, then click **All Apps > Windows Administrative Tools > Defragment and Optimize Drives**.)
  - 3 Click **Disk Defragmenter** under **Storage**. (If you have Windows 10, click the name of the drive, then click **Optimize**.)

Unless you have an SSD, you should also defragment the drive after you install and index a new database.

### 3 Troubleshooting

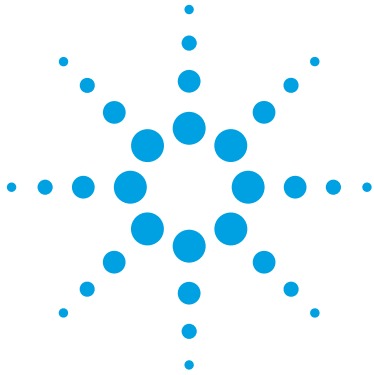
If you are prompted to download “faindex.cgi”

#### If you are prompted to download “faindex.cgi”

- ✓ See the second bullet under “If PERL does not work” on page 51.

#### If you are denied permission to update the database list

- ✓ Check that the file `spectrumMill\millhtm\sm_js\dbnames.js` has **Modify** permissions for IUSR. See “[Step 3. Configure security for Spectrum Mill Web Access](#)” on page 27.



## Customer Acceptance

- The installation is satisfactorily completed and the warranty period will commence. All specifications due to the *Agilent G2721AA/G2733AA Spectrum Mill MS Proteomics Workbench* installation have been met.

To help ensure the best possible support for your Spectrum Mill MS Proteomics Workbench, please FAX a copy of this completed form to:

Agilent Spectrum Mill Product Support  
**(408) 553-3165.**

\_\_\_\_\_  
Sales Order Number

\_\_\_\_\_  
Company Name

\_\_\_\_\_  
Engineer Signature/Date

\_\_\_\_\_  
Customer Signature/Date

\_\_\_\_\_  
Version Number

\_\_\_\_\_  
Customer Name (print)

- G2721AA/G2733AA    G2722AA    G2723AA    G2729AA

- List all materials (if any) that are damaged or not accompanied with this shipment:

\_\_\_\_\_  
\_\_\_\_\_  
\_\_\_\_\_



**Agilent Technologies**

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[www.agilent.com](http://www.agilent.com)

## In This Book

This book contains preparation, installation and troubleshooting instructions to set up your Spectrum Mill Workbench software on a server system.

Instructions for setting up the client system to access the Spectrum Mill Workbench software is found in the Spectrum Mill Workbench *Quick Start Guide*.

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