

APEX2 User Manual

Version 1.22



M86-E00078 7/04 PRELIMINARY





APEX2 User Manual

Version 1.22

M86-E00078 7/04

This manual covers the APEX2 software package. To order additional copies of this publication, request the part number shown at the bottom of the page.

© 2004 Bruker AXS Inc. All world rights reserved.

Notice

The information in this publication is provided for reference only. All information contained in this publication is believed to be correct and complete. Bruker AXS Inc. shall not be liable for errors contained herein, nor for incidental or consequential damages in conjunction with the furnishing, performance, or use of this material. All product specifications, as well as the information contained in this publication, are subject to change without notice.

This publication may contain or reference information and products protected by copyrights or patents and does not convey any license under the patent rights of Bruker AXS Inc. nor the rights of others. Bruker AXS Inc. does not assume any liabilities arising out of any infringements of patents or other rights of third parties. Bruker AXS Inc. makes no warranty of any kind with regard to this material, including but not limited to the implied warranties of merchantability and fitness for a particular purpose.

No part of this publication may be stored in a retrieval system, transmitted, or reproduced in any way, including but not limited to photocopy, photography, magnetic, or other record without prior written permission of Bruker AXS Inc.

Address comments to:	Technical Publications Department Bruker AXS Inc. 5465 East Cheryl Parkway Madison, Wisconsin 53711-5373
	USA

All trademarks and registered trademarks are the sole property of their respective owners.

Printed in the U.S.A.

M86-E00078 7/04

ii

Table of Contents

Notice	ii
1. Introduction 1.1 APEX II Systems for Chemical Crystallography 1.2 User Manual Features	
1.3 X-ray Safety	1-3
2. Hardware Overview	2-1
2.1 System Components	2-1
2.1.1 APEX II Detector	
2.1.2 Goniometer	
2.1.4 K/60 X-ray Generator	
2.1.5 Radiation Salety Enclosure with Interfocks and Warning Lights	2-7 2 - 7
2 1 7 Refrigerated Recirculator for the Detector	2-8
2.1.8 Computers	
2.1.9 Accessories	
3. Software Overview	3-1
3.1 The Server Computer	
3.1.1 Bruker Instrument Service (BIS)	
3.1.2 Bruker Control Program (BCP)	3-2
3.1.3 APEX2 Server	3-3
3.2 The Client Computer	

3.2.1 Database and Database Connection 3.2.2 APEX2 GUI	3-4 3-5
. Program Start-Up and Shutdown	4-1
4.1 Server Computer Start-Up	4-1
4.1.1 Starting Bruker Instrument Service (BIS)	4-1
4.1.2 Starting the APEX2 Server	4-3
4.2 Client Computer Start-Up	4-4
4.2.1 Starting the Database	4-4
4.2.2 Starting APEX2	4-4
4.3 Client Computer Shutdown	4-6
4.3.1 Stopping APEX2	4-6
4.3.2 Stopping the Database	4-6
4.4 Server Computer Shutdown	4-7
4.4.1 Stopping the APEX2 Server	4-7
4.4.2 Stopping BIS	4-7
. Crystal Orientation	5-1
5.1 Mount the Goniometer Head on the Instrument	5-2
5.2 Center and Align the Sample	5-3
. Data Collection	6-1
6.1 Start a New Project and Describe the Sample	6-1
6.2 Determine the Unit Cell	. 6-3
6.2.1 Collect Images	. 6-3
6.2.2 Harvest the Reflections	6-6
6.2.3 Index the Reflections	6-9
6.2.4 Refine the Unit Cell	. 6-10
6.2.5 Determine the Bravais Lattice	. 6-12
6.3 Determine the Data Collection Strategy	6-13
	. 0 10

7. Data Integration and Scaling	7-1
7.1 Integration	7-2
7.1.1 Active Mask	7-8
7.1.2 Algorithm	7-8
7.1.3 Image Queue	7-8
7.2 Monitor the Integration Run with SaintChart	7-9
7.2.1 Overall Rsym and Coverage Statistics	7-15
7.2.2 Unconstrained Unit Cell Refinement	7-16
7.3 Scale	7-17
7.3.1 Setup Input Files	7-17
7.3.2 Parameter Refinement	7-18
7.3.3 Error Model Refinement	7-19
7.3.4 Display Diagnostics	7-20
7.3.5 Exit	7-25
8. Examine Data	8-1
8.1 XPREP	8-1
8.2 Space Group Determination	8-2
8.3 Reflection Statistics	8-4
8.4 Applying High Resolution Cutoff	8-5
8.5 Preparing an Output File	8-6
8.2 Simulated Precession Images	8-9
9. Structure Solution and Refinement	9-1
9.1 Overview	9-1
9.2 Solve the Structure	
9.3 XSHELL	9-4
9.3.1 Refine the Structure	9-4
9.3.2 Label the Atoms	9-9
9.3.3 Refine the Molecule	9-14
9.3.4 Look at Atomic Displacement Parameters (Thermal Ellipsoids)	9-19
9.3.5 Sort Atoms	9-21

9.3.6 Add Hydrogen Atoms	
Appendix A. SAINTPLUS	A-1
Appendix B. Crystal Screening	B-1
Appendix C. RLATT	C-1

Add Twinabs, Cell Now

Index

4

1. Introduction

1.1 APEX II Systems for Chemical Crystallography

Bruker AXS Kappa APEX II and SMART APEX II systems are the newest members in the Bruker Nonius product line of instrumentation for single crystal X-ray diffraction. The systems provide the tools for complete small molecule structure determination. The hardware and software are completely redesigned. The software features a new start to finish graphical user interface (GUI). The hardware features a new CCD detector based upon four port readout of a 4K chip and a choice of two goniometers. APEX II systems are enclosed in a fail-safe X-ray enclosure system.

The Kappa APEX II system features the Kappa 4-axis goniometer. Two computers are used for experiments. One computer, the server, controls the goniometer. The other computer, the client,

controls all other aspects of the experiment, from data collection through report generation.

The SMART APEX II system is an enhanced version of the SMART APEX fixed chi system. A single computer controls the data collection, and solution and refinement of the structure.

From a software and operational viewpoint, the APEX II systems use the GUI of the APEX2 software suite to control all operations, from crystal screening to report generation, for a typical crystallography study. This is a complete departure from the command driven, functionally separate modules of SMART, SAINTPLUS and SHELXTL. Enhanced versions of the proven and widely accepted programs used by these modules (e.g., SAINT, SADABS, XPREP, XS, XM, XL, etc.) underlie the GUI and provide powerful tools.

From a hardware viewpoint, APEX II systems share common hardware components with other

Bruker products. Other members of this new generation of instruments include the D8 ADVANCE and D8 DISCOVER, and the D8 GADDS systems for general diffraction. Documentation on some of these common hardware and software components is available in the user's manuals for the D8 family of instruments.

1.2 User Manual Features

This user manual and associated YLID test data are intended to provide you with a step-by-step guide to data collection and processing using the APEX2 software program.

The test data supplied was collected on an **X8** (or Kappa, per Ludger) diffractometer with an APEX II camera, and graphite monochromated molybdenum radiation from a sealed tube generator. The high quality data (resolution=0.75 Å) allows easy refinement of the hydrogen atom positions and determination of the absolute structure of the sample.

NOTE: Before using this manual, check that your system is in proper working order (e.g., the optics and goniometer are aligned) and that the APEX2 suite is properly installed.

1.3 X-ray Safety

WARNING

X-ray equipment produces potentially harmful radiation and can be dangerous to anyone in the immediate vicinity unless safety precautions are completely understood and implemented. All persons designated to operate or perform maintenance need to be fully trained on the nature of radiation, X-ray generating equipment and radiation safety. All users of the X-ray equipment are required to accurately monitor Xray exposure by proper use of X-ray dosimeters.

For safety issues related to the operation and maintenance of your particular X-ray generator, diffractometer and shield enclosure, please refer to the manufacturer operation manuals or your Radiation Protection Supervisor. The user is responsible for compliance with local safety regulations.

1 - 4

2. Hardware Overview

The two hardware platforms for the APEX II systems are the Kappa APEX II, the four-axis advanced research instrument, and the SMART APEX II, the three-axis laboratory instrument. Software functionality is essentially the same for both platforms.

2.1 System Components

The system (Figure 2.1 and Figure 2.2) consists of the following basic components.

- APEX II[™] CCD detector
- 4-axis Kappa or 3-axis goniometer
- K780 X-ray generator
- Radiation safety enclosure with interlocks
 and warning lights
- D8 controller
- Refrigerated recirculator for the detector
- Computer(s) (two for the Kappa APEX II and one for the SMART APEX II)
- Video microscope
- Accessories (temperature devices)





Figure 2.1 - Kappa APEX II system

Figure 2.2 - SMART APEX II system

2.1.1 APEX II Detector

The APEX II detector is specific to this system. Status lamps on the top of the detector housing indicate when the detector is on (green) and off (red).

On Kappa APEX II systems, the detector is mounted on a motorized 2θ track. The camera distance is computer controlled (a typical distance for the camera is 40 or 50 mm).

On SMART APEX II systems, the detector is mounted on a 2 θ dovetail track. The track has a scale that is calibrated in mm to indicate the distance from the crystal to the phosphor window (a typical distance for the camera is 40 or 50 mm).

2.1.2 Goniometer

The goniometer module and APEX II detector comprise the unique hardware of the system. This is the part of the instrument that actually performs the experiment.

Several components comprise the goniometer *module* with APEX II detector.

- Goniometer (4-axis or 3-axis)
- APEX II detector
- X-ray source (including shielded X-ray tube, X-ray safety shutter, and graphite crystal monochromator)
- K780 X-ray generator
- Timing shutter and incident beam collimator (with beam stop)
- Video camera

Kappa APEX II Goniometer

The Kappa goniometer uses a horizontally oriented Kappa goniometer with 2-theta, omega, kappa and phi drives and a motorized DX track for setting the detector distance. It includes mounting points for the video camera and for optional attachments such as the optional low temperature attachment.



Figure 2.3 - Kappa 4-axis goniometer components

With the kappa angle, the crystal can be oriented at chi from *-92 to 92* °?. This leaves the top of the instrument open for easy access.

SMART APEX II Goniometer

The SMART APEX II system uses a horizontally oriented D8 goniometer base with 2-theta, omega and phi drives, dovetail tracks for the Xray source and detector, and an additional mounting track for accessories such as the video camera and optional low-temperature attachment.

The 3-axis system incorporates a fixed chi stage with chi angle of approximately 54.74° and a phi drive with 360° rotation, which is so compact that it **swings into** the incident beam collimator, allowing free rotation in omega. All four axes $(2\theta, \omega, \phi, and \chi)$ intersect.



Figure 2.4 - SMART goniometer components

X-ray Source

Three components comprise the X-ray source: a shielded X-ray tube, an X-ray safety shutter, and a graphite crystal monochromator.

The sealed tube X-ray source, with a molybdenum (Mo) target, produces the X-ray beam used by the system.

The X-ray safety shutter is built into the X-ray tube shield. The shutter opens upon initiation of a set of exposures and closes upon the end of collection. Status lamps on the shutter housing indicate when the shutter is open (red) and closed (green). The shutter is also interfaced to the controller and to the safety interlocks.

A tunable graphite crystal monochromator selects only the K_{α} line (λ =0.71073 Å) emitted from the Mo X-ray source and passes it down the collimator system.

Timing Shutter and Collimator

On SMART APEX II systems, the monochromatic X-ray beam then passes through the labyrinth, the timing shutter, and the incident beam collimator before striking the specimen. On Kappa APEX II systems, the monochromatic Xrays pass through a small labyrinth, the timing shutter, a secondary labyrinth and the incident beam collimator before striking the sample.

• The labyrinth is a device that ensures that the collimator and shutter are tightly connected to prevent X-ray leakage.

- The timing shutter is a device which precisely controls the exposure time for each frame during data collection. Its status lamps indicate when the shutter is open (ON) and closed (OFF). For SMART APEX II systems, this assembly also houses an automatic attenuator. Kappa APEX II systems do not have an attenuator.
- The incident collimator is equipped with pinholes at both front (near crystal) and rear (near source). These pinholes help to define the size and shape of the incident X-ray beam that strikes the specimen. (Collimators are available in a variety of sizes, depending on your application.)
- The beam stop catches the remainder of the direct beam after it has passed the specimen. The beam stop has been aligned to minimize scattered X-rays and to prevent the direct beam from hitting the detector. The entire collimator assembly is supported by a collimator support assembly, which has been precisely aligned to guarantee that the X-ray beam passes through the center of the goniometer.



2.1.3 Video Camera/ microscope?--heading?

The video camera, an essential part of the system, allows you to visualize the crystal to optically align it in the X-ray beam and to measure the crystal dimensions and index crystal faces. The camera is interfaced to the computer and is operated through the VIDEO program. The video program includes several computer-generated reticules and scales to make centering and measuring the crystal easy.

2.1.4 K780 X-ray Generator

The K780 X-ray generator is a high-frequency, solid-state X-ray generator, which provides a stable source of power for operations up to 60 kilovolts (kV) and 50 milliamps (mA). For the APEX II system, power settings should never exceed the maximum power rating of the X-ray tube.

Typical maximum power settings for the APEX II system with a fine focus tube are 50 kV, 40 mA. For both types of tubes, the kV setting should not exceed 50 kV and the power (kV x mA) should not exceed the rating given on the tube cap.

This generator is interfaced to the controller and the power settings can be adjusted within the APEX2 software. This is usually not necessary though as the software automatically increases the power to the user defined values at the beginning of an experiment and lowers them when the instrument is inactive.

2.1.5 Radiation Safety Enclosure with Interlocks and Warning Lights

A common component of all systems in the D8 family is the radiation safety enclosure. This new design is fully leaded (leaded metal sides and panels, leaded windows) to protect you from stray radiation. The enclosure also includes warning lamps (a government requirement) that alert you when X-rays are being generated. And, as a special feature, the enclosure incorporates interlocks (for both hardware and software)—an automatic system-interruption device that senses when the doors and panels are open and prevents use of the shutter and data collection until you close the doors.

2.1.6 D8 Controller

The D8 controller is an electronic module enclosed in the rack behind the font panel of the instrument. It contains all of the electronics and firmware for controlling the generator, opening the X-ray shutters, and monitoring other instrument functions, such as safety interlocks, generator status, and detector statuses. For SMART APEX II systems, the goniometer is controlled by the D8. For Kappa APEX II systems, there is an additional module, the Kappa controller, for positioning the Kappa goniometer angles and driving the detector distance.

2.1.7 Refrigerated Recirculator for the Detector

The refrigerated recirculator uses an ethylene glycol/water mixture to absorb the heat from dual Peltier devices which are used to cool the CCD chip to approximately -58°F (-50°C). This minimizes dark current.

2.1.8 Computers

The Kappa APEX II system uses two high speed computers. The server controls the instrument and is used for crystal centering and screening. The client collects the data, stores the raw frames, processes the data, and solves and refines the structure. The two computers are linked via a hub and communicate with each other via TCP/IP protocols.

The SMART APEX II system uses a single high speed computer for control of the experiment, storage of raw frame data, integration of the data, and solution and refinement of the structure.

Often the computer(s) is (are) attached to a network of similarly configured computers with access to local and/or network printers.

NOTE: Connection to the external network must be done with care. Consult with local security experts.

2.1.9 Accessories

Various devices can be mounted on the goniometer base. These include optional low- and hightemperature attachments. Both instruments can be used with diamond-anvil cells.

3. Software Overview

This section presents an outline of the system software, including a brief description of the software layout, as well as the graphical user interface (GUI).

APEX2 runs on two computers: the server and the client. For SMART APEX II systems, the

server and the client execute on the same computer, but the functionality remains separate. The flowchart in Figure 3.1 shows the software layout. For both Kappa APEX II and SMART APEX II systems, the server and client communicate using TCP/IP protocol.



3.1 The Server Computer

The server computer communicates with the hardware, allowing the user to control the instrument. The software to align the system, as well as to align and screen samples, runs on the server computer.

3.1.1 Bruker Instrument Service (BIS)

BIS provides the link between the hardware and software. Once a connection is established, BIS executes hardware commands sent by the APEX2 software. The instrument service can also be used as a service tool, displaying diagnostic messages during operation.

3.1.2 Bruker Control Program (BCP)

BCP is used to configure BIS, as well as to provide instrument control and alignment tools.

BrukerInstrument.ini - BCP: Bruker Configuration						
File Edit View	Tools Help					
🗋 🗀 🚅 🔚 🖓	D8CONFIG					
Instrument	D8TOOLS					
	Generator RA	Ctrl+Shft+G				
	Detector Bias	Ctrl+Shft+B				
General Info	Ratemeter	Ctrl+Shft+R				
achorannic	Half-beam	Ctrl+Shft+K				
<u> 1967</u>	HI-STAR grid	Ctrl+Shft+H				
	Test Frame	Ctrl+Shft+T				
Configuration	Dark Current	Ctrl+Shft+D				
	Bad Pixels	Ctrl+Shft+P				
A	Spatial	Ctrl+Shft+S				
Gonio Heads	Flood Field	Ctrl+Shft+F				
00100110000	Attenuator	Ctrl+Shft+A				
Ω	Imagemeter	Ctrl+Shft+I				
Bobot & Dewars	Manual	Ctrl+Shft+M				
110001 0 0 011010	Limits	Ctrl+Shft+L				
	Collision recovery	Ctrl+Shft+C				
	Update & home	Ctrl+Shft+U				
	Encoder Offsets	Ctrl+Shft+E				
	Log/Test Security	Ctrl+L				
	Exercise test					
	Debug BIS					

Figure 3.3 - BCP main window

Figure 3.2 - Image of start/stop in BIS

3.1.3 APEX2 Server

The APEX2 Server provides tools for aligning and screening samples. There are two main items: Align Crystal and Simple Scans (see Figure 3.4).



Figure 3.4 - Simple Scans window

3.2 The Client Computer

The client can be any computer which is on the same network as the server. For SMART APEX II systems, it is usually the same computer as the server. The main portion of the APEX2 suite, the APEX2 client, runs on the client computer. The client is a GUI with multiple plug-ins or modules for different aspects of an experiment. The client includes a database which stores relevant data from each step in the experiment. Details of the functions available in the GUI will be explained in more detail later in the manual.

3.2.1 Database and Database Connection

As currently configured, the database is used internally by the APEX2 Suite and is not available for user customization or manipulation. It must be running before the APEX2 Suite is started and it should be stopped before the computer is shutdown (see Section 4.3.2).

The purpose of the database is for storage of data generated by the Bruker APEX2 software.

3.2.2 APEX2 GUI

The APEX2 GUI has one main window (see Figure 3.5). This window is divided into four sections:

- Window Tool Bar
- Tool Icon Bar
- Task Bar
- Task Display Area



Figure 3.5 - APEX2 GUI

Window Tool Bar

The tool bar provides pull-down menus for a variety of file operations, image tools, and help files.

Option	Description			
[Symbol]	Use this menu to select the following: Restore, Move, Size, Minimize, Maxi- mize, and Close.			
File	Use this menu to select the following: Login, Logout, New, Open, Save, Close, Import (Spatial), Export (P4P File) and Exit.			
Instrument	Use this menu to select the following: Connection, Status, Toggle Shutter and Abort.			
Windows	Use this menu to select the following: Cascade and Tile.			
RLATT (available when you select Reciprocal Lattice Viewer)	Use this menu to select the following: Rotate, Edit, Orientation, Unit Cell Tool, Measure Distance, Measure Angle and Visualization.			
View (available when you select Data Collection Strategy)	Use this menu to select the following: Detailed Strategy.			
Table 3.1 – Window tool bar options				

Tool Icon Bar

The tool icon bar provides shortcuts to the options available through the window tool bar.

Icon	Description
B	Create a new file.
2	Open a file.
	Save a file.
N?	"What's this?" context-sensitive help.
• *	Open an image.
4	Select previous image. This icon is visible only when an image is open.
D	Select next image. This icon is visible only when an image is open.
0	Draw a circle. This icon is visible only when an image is open.
Table 3.2 – To	ool icon bar options

3 - 6

lcon	Description
\	Draw a plotting line. This icon is visible only when an image is open.
2 ¹⁰	Change the part of the image displayed while zoomed in. This icon is visible only when an image is open.
	Select a region of the image. This icon is visible only when an image is open.

Table 3.2 – Tool icon bar options

Task Bar

The task bar provides menus for all of the options in the APEX2 Suite: crystal evaluation and indexing (Evaluate Crystal), data collection (Collect), data processing (Integrate and Scale), and instrument setup (Instrument).





Reciprocal Lattice Viewer - 3D visualization of lattice projected in reciprocal space.



Collect



Integrate

Scale

Data Collection Strategy - Simulated data collection and determination strategy.

Experiment - Sequence editor for data col-

Oriented Scans - Measure different images with the crystal aligned along the axes.

Integrate Images - Integration of different

lection experiments.



XPREP (Space Group Determination) -Run XPREP.



Precession Images - Create synthesized precession images based on measured frames.

Solve Structure

Structure solution.



Refine Structure



Run XCIF to generate a report.



Instrument

Table 3.3 - Task bar options

Examine Data

Table 3.3 – Task bar options

data.

absorption correction.

M86-E00078

Report



Scale - Scale intensities and perform

Task Display Area

The task display area is the main area for tasks, user input and selected output. This area displays images, the reflections used in indexing, and the observed and predicted diffraction patterns. It also displays the runs for data collection and solution and refinement. (For version 1.22, space group determination, SaintChart output, XShell refinement, and Xcif report generation do not use the Task Display Area; they open in a new window). All other plug-ins open in the Task Display Area of the GUI.



Figure 3.6 - The Task Display area showing COSMO

4. Program Start-Up and Shutdown

As mentioned previously, the APEX2 Suite is composed of several programs. All of the programs are started in a similar fashion. For ease of use there is usually a desktop icon for the folder containing these programs and desktop icons directly to these programs. However the Click on Start > Programs > Bruker ... path is always available. This more complicated method is what will be used in this discussion.

4.1 Server Computer Start-Up

Two programs must be running: Bruker Instrument Service and APEX2 Server.

NOTE: For Kappa APEX II systems, the programs will be on the server computer in the goniometer cabinet. For SMART APEX II systems, there is typically only one computer.

4.1.1 Starting Bruker Instrument Service (BIS)

1. Click on Start > Programs > Bruker AXS Programs > Bruker Instrument Service.

After a brief initialization period, a window will appear (see Figure 4.1). On Kappa APEX II sys-

tems, the goniometer will move to reference positions.

BIS: Bruker I	nstrument Serv	tice - BIS	5 1.2.04/15 Ap	n-2004				×
😑 Com	nand		😑 File		😑 Status			Windows Service Manager
	Command	File	Status		Command	File	Status	Install Service Remove Service
Client 1	•	•	۲	Client 6	•			
Client 2		•	۲	Client 7		۲		Debugging Tools
Client 3		•	۲	Client 8				Debug Level 2 Filer "NOOPERATION"
Client 4		•	•	Client 9				Goniometer 0 V
Client 5		•	۲	Client 10	۲			Monitoring Clear Messages
					Marrison	er Derei	ad bur DIC	
ICHEOKINSTRU ICHEOKDETECT ICHEOKINSTRU ICHEOKINSTRU ICHEOKINSTRU ICHEOKINSTRU	MENTSTATUS] DRSTATUS] MENTSTATUS] MENTSTATUS] TORSTATUS] MENTSTATUS]							*
ICHECKINSTRU ICHECKDETECT ICHECKINSTRU	MENTSTATUS] ORSTATUS] MENTSTATUS] MENTSTATUS]							
					Defective Messa	ges Recei	red by BIS	
					Message	s Sent by I	915	
								-
Command								2
								Process
Help		H	6	StatEIS	5	Stop 81S		Exit Send to

Figure 4.1 - The BIS window

If a small pop-up window appears that says "This second instance of BIS is exiting" (see Figure 4.2), BIS was already running. Click on ok to clear this informational message.



Figure 4.2 - BIS warning message

NOTE: With a SMART APEX II or a Kappa APEX II, the kappa goniometer will home and the kappa server will activate when BIS is started. This may take a minute or two. The server is a service tool and should not be used to control the instrument.

4.1.2 Starting the APEX2 Server

1. Click on Start > Programs > Bruker Nonius Programs > APEX2 Server.

A window will appear (see Figure 4.3).



Figure 4.3 - Initial APEX2 Server window

2. In the top left corner, click on the pull-down menu Instrument (see Figure 4.4).

<mark>۴</mark> ۸	The Apex2Server v1.0-19					
<u>F</u> ile	Instrument	<u>W</u> indows				
IN	<u>C</u> onne	ction				
F	<u>S</u> tatus					
E	Toggle					
	Abort					
	Center Crystal					

Figure 4.4 - Connecting to the instrument

- 3. Click on Connection and a new window will appear (see Figure 4.5). The name of your server should already be filled in.
- 4. Click on connect.

Tinstrument Connection					
Host Name:	bruker-server				
	Connect	Cancel			

Figure 4.5 - Connection window

NOTE: If the name is wrong then the instrument is not properly configured and you should consult the system manager. (It is possible to configure the instrument to automatically connect and then this window will not appear). **tell how to do this.**

4.2 Client Computer Start-Up

On the client computer, two programs are also required: the database and APEX2. It is best to start the database before starting APEX2.

NOTE: For the SMART APEX II, there is typically only one computer for the client and server software.

4.2.1 Starting the Database

 For Windows systems, click on Start > Programs > Bruker AXS Programs > Start Database or click on the Start Database icon on the desktop.



1b. For Linux systems, open a terminal window and type "bnrun startdb" or click on the Start Database icon.

A window should appear that says the database system is ready.



Figure 4.6 - The database is ready

You can minimize this window. If the database has not previously been closed properly (e.g.,

after a power failure), a window will appear (see Figure 4.7) that states that another postmaster is running. If this happens, stop the database and then start it again.

🧯 Start Database

start Lock fil, "/data/postmaster.pid" already exists. Is another postmaster (pid 1788) running in "/data"?

Figure 4.7 - Database failure message

4.2.2 Starting APEX2

 For Windows systems, on the client computer click on Start > Programs > Bruker Nonius Programs > APEX2 or click on the APEX2 icon on the desktop.



1b. For Linux systems, open a terminal window and type bnrun apex2 or click on the APEX2 icon.

- 2. A window will appear to login to the database by entering a user and password (see Figure 4.8).
 - 2.1 If the system manager has set up the system to automatically enter the user name and password, step 2 is skipped.

T Login	<u>? ×</u>
User:	
Password:	
<u>D</u> K.	<u>C</u> ancel

Figure 4.9 - APEX2 start-up window

Figure 4.8 - Login request

3. An empty start-up window will appear (see Figure 4.9).				٩		
🖬 Apex2 v1	.0-20 - User:	(guest)				
Eile Instrument	Windows			 		Figur
	R					

4.	Click o	n File.
🕈 Apex2 v1.0-		
Eile	Instrument	<u>w</u>
ĥ	Login	?
	Logout	
B	<u>N</u> ew	
2	<u>O</u> pen	
U F	Couo	

Figure 4.10 - clickfile2.bmp

- 5. Use the options in this menu to create a new project or to open an existing project.
- 6. If the window in Figure 4.11 appears then APEX2 thinks the database is already in use. Answer "Yes" to close the window.

📅 Sar	nple locked 🔀
i	This sample is locked by another session. Do you want to unlock the sample?



4.3 Client Computer Shutdown

NOTE: The order of stopping these programs is important. If you attempt to close the database before APEX2 is stopped, the database will remain open until APEX2 is stopped.

4.3.1 Stopping APEX2

 For Windows or Linux systems, click on the X in the upper right corner of the window or click on File > Exit in the upper left. It is not necessary to disconnect from the instrument.

4.3.2 Stopping the Database

 For Windows systems, click on Start > Programs > Bruker Nonius Programs > Stop Database or click on the Stop Database icon.



1b. For Linux systems, in a terminal window enter bnrun stopdb or click on the Stop Database icon. A window will appear and quickly disappear, and the Start Database window will close.



Figure 4.12 - Stop database screen

NOTE: Occasionally the windows won't disappear and the Start Database window will display a "smart shutdown request" (see Figure 4.12). This message means that the database is waiting to close until applications that it might write to are closed. Exit APEX2 to solve this problem. If the message still appears, check for other processes that may still be running (e.g., COSMO).
4.4 Server Computer Shutdown

Stop APEX2 Server before BIS. It is ok if the order is inverted. Generally, BIS is never stopped.

4.4.1 Stopping the APEX2 Server

 Click on the X in the upper right corner of the window or click on File > Exit in the upper left. It is not necessary to disconnect from the instrument.

4.4.2 Stopping BIS

It is almost never necessary to stop and exit BIS. If necessary, click on the stop BIS button on the bottom of the BIS window and then click on the exit button at the bottom of the window.

4 - 8

5. Crystal Orientation

We are now ready to begin data collection with the instrument using the YLID test crystal. We assume that your system manager has set up the system properly and that all system default parameters have been set appropriately.

The data collection process is divided into five steps:

See Section 5:

1. Centering/aligning on the diffractometer (from the APEX2 Server—the center module) 2. Crystal quality check (from the APEX2 Server—the simple scans module)

See Section 6:

- 3. Cell determination (from APEX2—the cell determination module)
- 4. Data collection setup (from APEX2—the strategy module)
- 5. Data collection (from APEX2—the experiment module)

The first steps, mounting, aligning and screening a crystal, are performed on the server computer.

5.1 Mount the Goniometer Head on the Instrument

Use extreme care when handling the goniometer head to prevent damage to your sample on the end of the small glass fiber.

- Open the enclosure doors. Push either of the rectangular green Open Door buttons on the side posts. This will release the door locks for approximately five seconds. During this time, pull out on one or both of the handles to physically open the doors.
- 2. In the APEX2 Server GUI, under Setup click on Center Crystal.

The centering buttons will appear and the video window will open.



Figure 5.1 -

The bottom five buttons will drive the goniometer to various predefined positions that are designed to simplify crystal centering. The top two buttons will drive phi by either 90 or 180 degrees.

3. Click on Mount to mount an empty goniometer head and determine the position.

NOTE: On Kappa APEX II systems, in the mount position, the key or pin on the mounting base will be toward the back of the instrument. On SMART APEX II systems, at $\phi=0^{\circ}$, the key on the mounting base of the goniometer head will be at the 12:00 position.



Use extreme care when handling the goniometer head to prevent damage to the sample on the end of the small glass fiber.

- 4. Carefully remove from its case the goniometer head containing the YLID test crystal.
- 5. Place the goniometer head onto its base on the phi drive, aligning the head's key slot with the key (pin) in the base.
- Snugly screw the head's collar to the base so the head does not move. Do not overtighten it.

5.2 Center and Align the Sample

To obtain accurate unit cell dimensions and to collect good quality data, align the center of the sample with the center of the X-ray beam and maintain the alignment for the entire experiment. Your video camera should be aligned so the crosshairs of the video camera coincide with the center of the goniometer and the center of the X-ray beam (see manual M86-Exx024 for instructions on aligning the microscope to the center of the instrument). Although, if the microscope is not centered, you can still align the sample—the key here is that the crystal stay in the same place in the microscope in all orientations.

NOTE: On SMART APEX II systems, adjust the height while in the mount position.

NOTE: Use the thin end on the goniometer wrench to unlock the adjustment slides at the beginning of the centering process and to lock them at the end—locking needs only a very slight touch. The other end of the wrench is used to make adjustments.

NOTE: If the image of the crystal is difficult to see, illuminate the sample with a high-intensity

lamp and/or temporarily place a light-colored piece of paper on the front of the detector.

NOTE: Centering is often easier if the crystal is rotated to give a good view before the actual centering process is started (e.g., down an edge for a plate). To do this, drive to one of the centering positions, loosen the screw that locks the goniometer head, rotate the crystal to a suitable orientation and then tighten the screw again.

 Click on the Center button—the crystal and goniometer head will be positioned perpendicular to the microscope. To center the sample, make adjustments in the height and with the translation screw that faces the front of the diffractometer.



Figure 5.2 - Crystal initially mounted

- 2. Use the z-axis (translation) screw and the xor y-axis (translation) adjustment screw that is perpendicular to the microscope axis to move the crystal so it is centered in the microscope (see Figure 5.8 and Figure 5.9).
 - 2.1 (For the SMART APEX II) Make adjustments in the x and y translations, but not to the height (z).



Figure 5.3 - Crystal centered

- 3. Adjust the height with the z-axis screw.
 - 3.1 (SMART APEX II) Click on the Right button. Adjust the height and the translation screw that is facing up.
- 4. Adjust the translation with the x- or y-axis screw, whichever is pointed toward you.
- 5. Adjust the height again with the z-axis screw.
- 6. Adjust the crystal position using the x- or yaxis screw.

7. Click on Drive Phi 180 and adjust (adjust what? x/y) again.



Figure 5.4 - Drive Phi 180

8. Check the other translation. (how? which one?)

Repeat steps 3 and 4 as necessary to keep the crystal in-line with the microscope.

9. Click on Drive Phi 90.



Figure 5.5 - Drive Phi 90

Repeat steps 6 and 7 as many times as necessary to keep the crystal in the same place in the microscope.

- 10. Adjust the crystal position using the goniometer screws. Why adjusting now--what are we adjusting?
- 11. Click on Drive Phi 180 and adjust again. (adjust what? x/y)

Repeat steps 3–11 as necessary until x is centered. Then adjust the height using the following steps.

- 1. Click the Left button and note the height. The goniometer drives to place the fiber horizontal and to the left.
- 2. Click the Right button and check that the crystal height stays in the same place in the microscope.
 - 2.a If the height is in the same place, you are done.

2.b If the height is not in the same place, adjust the height and repeat steps 1 and 2.



Figure 5.6 - Check the crystal height

3. Click the Top button.



Figure 5.7 - The crystal is centered

The crystal is now centered on the instrument. All of the next steps (except removing the crystal from the diffractometer) are performed with APEX2 on the client computer.





Figure 5.9 - Standard goniometer head in detail

6. Data Collection

The data collection process is carried out on the client computer using APEX2. Once data collection is started, exit APEX2. Data collection will continue.

6.1 Start a New Project and Describe the Sample

- 1. Left-click on File > New.
- 2. In the window that appears enter the sample name.

New Sa	nple	? ×
Name:	ylid_manual	
Template:	<u></u>	<u>_</u>
Folder:	C:\frames\guest\ylid_manual	
	ОК	Cancel

Figure 6.1 - The New Sample window

- 3. Click OK.
- 4. The task bar will appear with the Setup section open. Left-click on Describe.

5. Enter the requested information into the Describe window.

Setup	Name:	ylid_manual				
Describe	Compound:	Test ylid for manual			1	
	Formula:	C11 H10 02 S1			_	
	Crystal Color:	Appearance translucent	Intensity	-	Primary Color	Secondary Color
Center Crystal	Crystal Dimension:	0.22 × 0.2	25	× 0.23	nm]	
	Crystal Shape:	Oblated sphere				

Figure 6.2 - Describe window

6. Close this module. The data will automatically save to the database.

6.2 Determine the Unit Cell

6.2.1 Collect Images

1. In the task bar, left-click on Collect and then on Experiment

Since there was no connection to the instrument when APEX2 was started, the program recognizes that it needs a connection in order to collect images. It will ask to connect (see Figure 6.3).

Tinstrument Connection							
Host Name:	bruker-server						
	Connect	Cancel					

Figure 6.3 - Instrument Connection window

- 2. Click on Connect.
- 3. Click on Append Matrix Strategy at the bottom left of the window.

Setup Experiment Monitor Experiment											
Ima	ge location:	C:\frames\c	uest\ylid_manual		2	🗸 Attenuate	if topped		Default time:	5.000 [se	ec/frame] 💌
Filer	name or prefix:	matrix				Correlate I	rames		Default width:	0.500 10 [d	egrees] 💌
First	Run:	1 🚔				🗖 Generate	new Dark Ima	ges	Detector Forma	it: 51	2x512 <u>-</u>
	Operation	Active	Distance [mm]	2Theta [deg]	Omega (deg)	Phi [deg]	Chi [deg]	Time [sec]	Width [deg]	Sweep [deg]	Direction
1	Phi Scan	Yes	40.000	30.000	-45.000	0.000	54.740	default	default	6.000	positive
2	Phi Scan	Yes	40.000	30.000	-45.000	120.000	54.740	default	default	6.000	positive
3	Phi Scan	Yes	40.000	30.000	-45.000	240.000	54.740	default	default	6.000	positive
4	No Operation	Yes									
5	No Operation	Yes									
6	No Operation	Yes									
7	No Operation	Yes									
8	No Operation	Yes									
9	No Operation	Yes									
10	No Operation	Yes									
11	No Operation	Yes									
12	No Operation	Yes									
13	No Operation	Yes									
14	No Operation	Yes									
15	No Operation	Yes									
16	No Operation	Yes									
17	No Operation	Yes									
Appe	nd Strategy	Append Matri	x Strategy						Ē	Validate E:	kecute / Resum

Figure 6.4 - Append matrix runs

- Adjust the scan time and scan width if desired. The default values are usually good. The default time of 10 seconds is acceptable for most samples, but shorter times will not adversely affect most experiments.
- 2. Click on Execute/Resume. The view will shift to the Monitor Experiment view (see Figure 6.5). The program will collect a series of three runs with twelve frames per run. This typically takes less than ten minutes. The images will stop changing when the experiment is done.

NOTE: Adjust the time (upper right) to match the scattering ability of the crystal (e.g., shorter times for strong diffractors and longer exposure times for weak diffractors). If the exposure times are five seconds or less, click on the check mark by Correlate Frames to turn off this feature. Frame correlation measures each frame twice at typically 1/2 the time and then combines the two together. This is usually not necessary with shorter exposure times.

NOTE: The format for frame names is shown in Figure 6.5. Every frame is assigned a name. For this figure the name is

ylid_manual_01_005.sfrm. This means that the

frame is for the project ylid_manual and that this is the fifth image of the first run.

📅 Apex2 v1.0-20 - Us	er: (guest) - File: ylid_manual - [Experiment]	
🍯 Eile Instrument Win	dows	🔊 Leix
🗅 🧀 🔲 📢		
C:\frames\guest\ylid_m	anual/ylid_manual_01_0005.sfm 📃 🖬 🎓 🔛 🗸 🕨 🔛	
Setup Evaluate Crystal Collect	Setup Experiment Monitor Experiment	
ß	arrows move arrows move	280
Data Collection	between frames between runs	260
¥		240
Experiment		220
····		200
	· · · · ·	180
Unented Scans		160
		140
	•	100
		80
		60
	1 10 100 10000 100000 1000000	:
Integrate	Description Value	<u> </u>
Scale	1 Image type PhiScan Image	
Examine Data	2 Start position Distance=35.00 2Theta=338.00 Omega=286.99 Phi=3.61 Chi=3.83	
Solve Structure	3 End position Distance=35.00 2Theta=338.00 Omega=286.99 Phi=4.11 Chi=3.83	
Refine Structure	4 Scan range 0.5000	-
Report	Image Header <u>A Tool Editor</u> <u>A Cursor Position</u>	
Instrument		

Figure 6.5 - Monitor Experiment view

To change the color of the image display (e.g., Black on White), right-click in the intensity bar to the right of the image display (see Figure 6.6).



Figure 6.6 - Color tool

After the first run is completed, there **usually** is sufficient information to start the indexing step.

If state "usually" then need to give information about what to do if there isn't enough information.

NOTE: APEX2 assigns to every collected frame a name. **Per Ludger, basename.run#.frame#.sfrm**

6.2.2 Harvest the Reflections

1. Left-click on Evaluate Crystal > Determine Unit cell.



Reciprocal Lattice Viewer

Figure 6.7 - The Determine Unit Cell (Indexing) icon

This will open the image viewer, but with a tool bar to the right for indexing (see Figure 6.8). The plug-in initializes with the first run (e.g., matrix_01).



Figure 6.8 - Image viewer with indexing tool

2. The name of the first image is already entered. Click on Harvest Spots.

NOTE: All other options are gray at this point because there are no reflections available.

A blue progress bar will appear as the software determines the best background level to use for harvesting. Then a window with two sliders will appear.



Figure 6.9 - Indexing sliders

The right slider selects which image is displayed. The left slider increases or decreases the l/s(I), the cut off criteria for accepting reflections. Generally, the defaults are fine.

 Change the run number in the "First Image" box to matrix_02_0001 and click on Harvest Spots. The run number is 02. The image or frame number is 0001.

At this point you should have 100 to 300 reflections harvested.

4. Change the run number in the "First Image" box to matrix_03_0001 and click on Harvest Spots.

NOTE: If run three is not completed, a window will pop-up that says "Do you want to continue with the images that could be read?" If this happens, skip step 3 and go to Section 6.2.3. Then return to Section 6.2.2 and harvest the spots before refining.

6.2.3 Index the Reflections

1. Click on Index.

A window will open.



Figure 6.10 - Indexing tool

The defaults are usually OK. Use the slider to omit reflections with lower I/sigma from the calculations. If indexing is difficult, try reducing the number of reflections used.

If indexing is difficult, use the RLATT tool. This tool is described in **Appendix ?**.

2. Click on Index.

After approximately 30 seconds the index window will display a possible cell and the OK button will no longer be gray. The values shown in Figure 6.11 are reasonable for the ylid.

-Reduced U	nit Cell
A Axis:	5.929 Å
B Axis:	8.981 Å
C Axis:	18.348 Å
Alpha:	89.955 °
Beta:	89.811 *
Gamma:	89.953 °
Volume:	976.9 Å^3

Figure 6.11 - The unit cell

The spot statistics are also acceptable with 98% (i.e., (238/244)x100) of the selected spots indexed.

Total Spots:	259
Selected Spots:	244
Indexed Spots:	238
	L.

Figure 6.12 - Focus on the spot results

There are often a few reflections that are not indexed. The reciprocal lattice viewer can be used to look at the spots used in the indexing, but first refine this cell.

3. Click on OK to accept the indexing results.

6.2.4 Refine the Unit Cell

There is not a correct order for the following steps. Use this as a guideline with the main goal of creating a stable converged refinement.

1. Click Refine in the Indexing Tools menu. The refinement window will open.



Figure 6.13 - The Refine menu with histograms displayed

The YLID test crystal should have an orthorhombic primitive cell with approximate cell dimensions of a=5.95Å, b=9.03Å, c=18.38Å, and α = β = γ =90°.

- 2. Click View Histograms. The histograms show how observed data compares to the data calculated using the current unit cell. The HKL values should be close to integers and the rotation angle differences should not be significantly larger than the step size used to collect the matrix frames.
- 3. Click Refine several times.
- 4. Check the Constrain Distance and Constrain Beam Center boxes.

NOTE: Check the constraints to fix the parameters listed. Uncheck the constraints to allow the parameters to refine.

- 5. Uncheck Constrain Pitch, Roll, and Yaw.
- 6. Click Refine several times.
- 7. Uncheck Constrain Distance and Constrain Beam Center, and check Constrain Pitch, Roll, and Yaw.
- 8. Click Refine several times.

NOTE: In most cases, the angle zeroes are close to zero and should not be refined. The

M86-E00078

crystal should now be aligned, so refinement of the crystal center is not necessary.

6.2.5 Determine the Bravais Lattice

1. After refining, click Bravais Lattice and look for other unit cell choices (i.e., look at fit values).

	Fit	A Axis [Å]	B Axis [Å]	C Axis [Å]	Alpha [°]	Beta [*]	Gamma [*]	Volume [Å^3]
Cubic F	379.54	21.36	21.36	21.37	129.93	60.85	147.60	3970.7
Cubic I	436.63	19.37	10.82	20.51	68.44	31.37	80.30	1985.3
Cubic P	491.52	5.96	9.04	18.42	89.94	89.97	89.90	992.7
Hexagonal P	215.27	5.96	9.04	18.42	89.94	90.03	90.10	992.7
Rhombohedral R	236.36	5.96	10.82	56.31	94.32	96.06	123.32	2978.0
Tetragonal I	174.92	5.96	9.04	38.40	76.42	81.08	89.90	1985.3
Tetragonal P	154.90	5.96	9.04	18.42	89.94	89.97	89.90	992.7
Orthorhombic F	102.67	5.96	19.03	37.32	87.11	99.16	108.15	3970.7
Orthorhombic I	81.26	5.96	9.04	38.40	103.58	98.92	89.90	1985.3
Orthorhombic C	90.74	5.96	19.03	18.42	90.04	90.03	108.15	1985.3
Orthorhombic P	0.59	5.96	9.04	18.42	89.94	89.97	89.90	992.7
Monoclinic C	90.74	19.03	5.96	18.42	89.97	90.04	71.85	1985.3
Monoclinic P	0.52	9.04	5.96	18.42	90.03	90.06	89.90	992.7
Triclinic P	0.00	5.96	9.04	18.42	89.94	89.97	89.90	992.7

Figure 6.14 - Bravais lattice display

Note that even though monoclinic has a slightly better fit, the software makes the correct choice of the higher symmetry cell.

- 2. Click on the appropriate Bravais Lattice, in this case orthorhombic.
- 3. Press OK to accept the suggested lattice settings.
- 4. Refine again.
- 5. Refine for several more cycles, changing the constraints one or two at a time.

Now you have a unit cell ready for determining a data collection strategy.

6.3 Determine the Data Collection Strategy

APEX2 includes a powerful algorithm, COSMO, for determining an efficient strategy that fully utilizes the flexibility of your instrument.

 Left-click on Collect > Data Collection Strategy.



Figure 6.15 - The strategy display

NOTE: COSMO will use information from cell determination to set defaults. You can modify the suggested values. **How?**

- 2. Check the inputs for defining the data collection.
 - 2.1 Set the data collection distance. For SMART APEX II systems this should be set to the actual detector distance. For Kappa APEX II systems there is a variable (DX) and the distance will default to the shortest reasonable distance. Generally for the APEX II detector, the distance should be about the same as the longest cell dimension. Typically distances ranging from 35 to 45 are reasonable.
 - 2.2 Set the exposure time and press return. For normal crystals on an APEX II, five seconds is a reasonable time.
 - 2.3 Click Same to set all of the times to be the same.

NOTE: If the "Same" feature is not chosen, **the times for shells can be set** to collect high angle data more slowly than inner shell data.

2.4 Set the desired redundancy (**0.50** is a reasonable value).

- 2.5 Check the other values (Laue class, Lattice, etc.).
- 2.6 Each time a value is changed, COSMO recalculates the statistics for the runs. The results are displayed in the column labeled Current.
- 2.7 Below the Target and Priority columns is a pull-down menu with several different strategies. Choose the one that best meets the needs of the experiment (for the example, "Best in 2 hours").--cut this text, we don't need to give a step by step analysis of the YLID.

At this point, if all of the runs available were collected it would take 183.98 hours and get a redundancy of 452.86. Clearly this is not desirable.

- 3. Click on Refine Strategy.
- 4. A list of options will appear. Again click Refine Strategy.

Execute:	Refine Strategy
	Refine Strategy
	Sort Runkgor Completeness

Figure 6.16 - UserManual017.bmp

NOTE: The objective in refine strategy (COSMO) is to get good completion (98% or better) with high redundancy in a reasonable amount of time. When COSMO is first started it will tell you the completion, redundancy, and time for all of the available runs. It is almost

never necessary to let COSMO run to completion. Typically it should be stopped when completion is greater than 99% and the time is close to what is desired.



Figure 6.17 - Completeness and redundancy chart

In this example, as shown in Figure 6.18, the completion is 99.76% and the time is approximately 2.33 hours.

NOTE: Time estimates are approximate. They depend on the number of rescans, general instrument overhead, backlash compensation, etc. If estimated times are consistently longer or shorter, modify the COSMO hardwire profile.

 Click Stop when the completeness nears 100% and the time and redundancy approach the desired values. It is not necessary to wait until the refinement reaches 100%.

Current	Target	Pri	ority
99.76	100.00	100	1 T
7.48	50.00	5	Ť
2.33	2.00	10	Ť
3	Best in 2 hours		•

Figure 6.18 - Strategy status and priority control

- 6. Click Refine Strategy.
- 7. A list of options will appear. Click on sort runs for completion.



Figure 6.19 - Choose "Sort Runs for Completeness"



Figure 6.20 - UserManual022.bmp

8. Go to View > Detailed Strategy.

The next window shows the runs to be collected.

ŧ	Curi	rent Strateg	У								
Г		Scan Type	CCD	2Theta	Omega	Phi	Chi Axis	Width	#Images	Sweep	Time
ľ	1	Strategy	35.00	-22.00	+286.99	-358.39	+3.83 Phi	0.50	739	369.50	7.5
	2	Strategy	35.00	-32.00	-24.79	+27.75	-99.47 Omega	permanent	153	76.50	7.5
ſ	3	Strategy	35.00	-7.00	-4.01	-26.26	-55.59 Omega	as-is	74	37.00	7.5
ſ	4	Strategy	35.00	+28.00	+48.51	-14.91	-34.09 Phi	partial	162	81.00	7.5
ľ	5	Strategy	35.00	-32.00	+317.27	+63.87	+93.86 Phi	disabled	127	63.50	7.5
ľ	6	Strategy	35.00	-32.00	-170.70	+23.43	+98.83 Omega	+0.50	96	48.00	7.5

Figure 6.21 -

NOTE: If for some reason it is necessary to start over, change the distance slightly (by 0.02 for example) and press Return. COSMO will reload all of the possible runs.

You are now ready to collect data.

6.4 Data Collection/Run Experiment

- 1. Click Collect > Experiment.
- 2. Go back into the experiment window and delete the three matrix runs if they are still there.

First	Run:	1	×.	
	Operation		Active	Distance [mm]
1	Phi Scan		Yes	40.0
2	Phi Sean		Yes	40.0
<u>_</u>	Cut	•	Yes	40.0
2	Delete	pn	Yes	
: 🖻	Copy 1/5	pn	Yes	
E 🛍	Paste	pn	Yes	
7	Clear	pn	Yes	

Figure 6.22 - UserManual014.bmp

3. Click Append Strategy.

4. The program changes the name to the name of the current project (in this example, ylid_manual).

sup		nitoi Experim	ent									
Ima	ge location: 🛛 🗍	:\struc\gues	t\ylid_manual		2	Attenuate if topped Correlate Frames			Default time: 10.000 [sec/frame] Default width: 0.500 [degrees]			
File	name or prefix:	(id manual										Ì
<u> </u>	. P					E c						
FIRS	rinun: Ji	T				i ue	nerate new Dark	Images	Detector Fo	rmac j	5128512	
	Operation	Active	Distance [mm]	2Theta [deg]	Omega [deg]	Phi [deg]	Chi [deg]	Time [sec]	Width [deg]	Sweep [deg]	Direction	Ĩ
1	Phi Scan	Yes	35.000	-22.000	286.99	-358.39	3.83	5.00	0.50	369.50	automatic	
2	Omega Scan	Yes	35.000	-32.000	27.75	27.75	-99.47	5.00	0.50	76.50	automatic	
3	Omega Scan	Yes	35.000	-7.000	26.26	-26.26	-55.59	5.00	0.50	37.00	automatic	
4	Phi Scan	Yes	40.000	28.000	-14.91	-14.91	-34.09	5.00	0.50	81	automatic	
5	No Operation	Yes										ſ
6	No Operation	Yes										
7	No Operation	Yes										Ī
8	No Operation	Yes										Ī
9	No Operation	Yes										
10	No Operation	Yes										
11	No Operation	Yes										
12	No Operation	Yes										
13	No Operation	Yes										Ī
14	No Operation	Yes										
15	No Operation	Yes										
16	No Operation	Yes										
17	No Operation	Yes										



NOTE: At the top of the experiment window are controls for data collection. Usually the default values are correct. For data collection times of less than five seconds, correlation can usually be turned off. If new dark frames are required, APEX2 will automatically collect them. Checking "Generate New Darks" will force the collection of darks before every run. In Figure 6.23, the time and width are explicitly set for each run, so changing the default width and time will have no affect. If the explicit time or width for a run is deleted so the box is empty, the default values will appear and be used.

 Click Execute/Resume. The focus will shift to Monitor Experiment and images will start to appear. This may take a minute or two if new darks are being collected. Images will start to appear.

NOTE: When resuming after a power failure, APEX2 will automatically skip images that were previously collected with matching angles and generator settings. Otherwise it will ask if you want to overwrite the images.

NOTE: After data collection is started, the experiment window can be closed and APEX2 can be stopped. The server computer must be left on. If communications are lost between the



Figure 6.24 - Monitor experiment view

7. Data Integration and Scaling

Before the data can be used to solve and refine the crystal structure, convert the information recorded on the frames to a set of integrated intensities and scale all of the data.

M86-E000787 - 1

7.1 Integration

- 1. Click on Integrate in the Task Bar.
- 2. Click on the Integrate Images icon. The following window will open.

Space	Group: P	222 -			Resolution Limit (Å): 0.651
	Starting Image Filename	N	Images	Output Filename	▲
1					
2					
3					
4					
5					
6					
4					
8					
10					
11					
12					
12					
13					
15					
10					
17					
18					
19					
20					
21					
22					
23					-1
<u> </u>	1				
Refin	ement Options	Options		Find Runs Import Runs from Experimen	Start Integration



3. Check the default values.

There are two items of interest on the top of the window. The space group tool and the resolution limit value.

Space Group:	P222	▶ ▼
- p p		

Figure 7.2 - The space group tool

M86-

The space group tool allows the user to set the symmetry for integration. Typically this value is correct when the integration window opens.



Figure 7.3 - Choosing $\mathsf{P2}(1)\mathsf{2}(1)\mathsf{2}(1)$ with the space group tool

The resolution window on the upper right sets the upper resolution for integration.

Resolution Limit [Å]:	0.750

Figure 7.4 - The resolution tool

The APEX2 Suite will suggest a resolution cutoff. In this case the suggested value changed to 0.75. In the center of the bottom are two buttons for defining the data collection runs to be integrated.

Find Runs Import Runs from Experiment

Figure 7.5 - The Find and Import Runs buttons

The import runs button determines the runs to be integrated from the experiment that has just been submitted. Using this button allows the user to start integration while the data is still being collected.

NOTE: SAINT will integrate all of the data currently measured and then will wait for the next image, integrate that, and wait for the next, etc.

The Find Runs button is used to browse to the set of runs to be integrated. The button is normally used when the data collection is finished.

M86-E000787 -

1. Click on Find Runs.

The following window will open, usually it will have the correct directory and basename as the defaults. If these are not correct, then use the browse button to find the runs for the data collection of interest.

📲 Find Runs	? ×
Find Runs in Directory:	c:\struc\guest\data_manual\ylid_manual
Image Base Name:	ylid_manual
Browse	OK Cancel

Figure 7.6 - Find Runs tool view

Both the Import Runs from Experiment and the Find Runs buttons will generate a list of runs as shown in Figure 7.7.

Spa	ace l	Group:	P222	•	
		Starting	Image Filename	Images	Output Filename
1	1	C:\struc	\guest\data_manual\ylid_manual\ylid_manua	739	\structures\data_for_manual\ylid_manual\work\ylid_n
2		C:\struc	\guest\data_manual\ylid_manual\ylid_manua	153	\structures\data_for_manual\ylid_manual\work\ylid_n
3	1	C:\struc	\guest\data_manual\ylid_manual\ylid_manua	74	\structures\data_for_manual\ylid_manual\work\ylid_n
4	1	C:\struc	\guest\data_manual\ylid_manual\ylid_manua	161	\structures\data_for_manual\ylid_manual\work\ylid_n

Figure 7.7 - Completed runs list

Figure 7.7 shows a run list entry being changed. The original run is shown first and then the run with the starting image number modified. In the last strip, the number of frames to process has been changed. Double-click on any of the fields in this list to open up that value for editing.

C:\struc\guest\data_manual\ylid_manual\ylid_manua	74
manual/ylid_manual/ylid_manual_04_0001.sfm 😅	161
 C:\struc\guest\data_manual\ylid_manual\ylid_manua	74
manual\ylid_manual\ylid_manual_04_0051.sfrm 🔀	161
	74
C:\struc\guest\data_manual\ylid_manual\ylid_manua	111

Figure 7.8 - Example of manually editing a run list

2. Check the refinement defaults.

The two buttons on the bottom far left are used to change the default options for the refinement of the data. The default values are generally very good. There is seldom any need to change those in the refine window.

Per-Image Refinement	
Enable Orientation Refinement	Damping Factor: 1.000
F Enable Box Size Refinement	Initial XVZ Box Size [1] 0.994 0.994 0.994
Periodic Refinement	Global Refinement
Enable Periodic Refinement	F Enable Global Refinement
Enable Initial Passes	
Frequency (Images): 50 👲	Max. Number of Reflections: 9939
Constrain Metric Symmetry of Unit Cell to:	Constrain Metric Symmetry of Unit Cell to:
 Space Group P222 	Group P222 Space Group P22 Space G
Cuystal System Orthorhombic 🕑	Crystal System: Onhonhombic 💌
Refinement Parameters	Refinement Parameters
Detector	Detector
Horizontal Beam Center	Horizontal Beam Center
Vertical Beam Center Distance	Vertical Beam Center Distance
- Pitch	- Pitch
- P Rol	- P Rol
I DI Auss	
	OK Cancel

Figure 7.9 - Refine Window

3. Change the default options for the integration in the "Integration Options" window.

The integration window has a few values that are changed more frequently.

Thegratics Options	<u>? x</u>
Model Profiles	Background Update Background Update Scaling Factor: 1.000
Intensity/Sigma Lower Limit for Model Profile Update: 10.000 Fraction of Model Profile Maximum for Simple Sum Mask: 0.050	Image Queue All-Width (Images): 7
Intensity/Sigma Upper Limit for LS Model Profile Fit. 8.000 Lower Resolution Limit for LS Model Profile Fit [Å]: 9999.000 Profile 20/2 Half-Wriths: 4 4 4	Beam Monitor
More Options	OK. Cancel

Figure 7.10 - Integration Options

M86-E000787 -

The default values in the Model Profiles window are usually good.

Model Profiles]
🗖 Blend Profiles from All D	etector Regions
Intensity/Sigma Lower Limit for Model Profile Update:	10.000
Fraction of Model Profile Maximum for Simple Sum Mask:	0.050
Intensity/Sigma Upper Limit for LS Model Profile Fit:	8.000
Lower Resolution Limit for LS Model Profile Fit [Å]:	9999.000
Profile XYZ Half-Widths:	4 4 4

Figure 7.11 - Model Profiles

For weakly diffracting crystals it is sometimes helpful to change some options in the Model Profiles window. Reducing the Intensity/Sigma ratios from 10 and 9 to, for example, 5 and 4 or 3 gives the profile fitting algorithm more reflections to model. Clicking on Blend Profiles from All Detector Regions to increase the profile blending can also help with weakly diffracting crystals.

 Click on More Options. This results in an expanded window (see Figure) and gives access to several other useful features, in particular Active Mask (see Figure 7.13), Algorithm (see Figure 7.14), and Image Queue (see Figure 7.15).
Marintegration Options	<u>? x</u>
Model Profiles	Background Update
Enable LS Profile Fitting	Background Update Scaling Factor: 1.000
Blend Profiles from All Detector Regions	
Intensity/Sigma Lower Limit for Model Profile Update: 10.000	Image Queue
Fraction of Model Profile Maximum for Simple Sum Mask: 0.050	Active Image Queue Half-Width [Images]: 7
Intensity/Sigma Upper Limit for LS Model Profile Fit: 8.000	- Poom Monitor
Lower Resolution Limit for LS Model Profile Fit [Å]: 9999.000	Enable Beam Monitor Normalization
Profile XYZ Half-Widths: 4 4 4	Normalize each Run Separately
Active Mask	Twin Overlap Determination
 Generate Mask: 	Minimum Common Volume [%]: 4.000
Fractional Lower Limit of Average Intensity: 0.000	Separation Factor: 1.000
C. Has De Frider Marke	Maximum Range: 1.300
C Use Pre-Existing Mask.	Output / Diagnostic Files
Active Mask File (*_am):	Generate Diagnostic Plot Files
Algorithm	📕 Keep Temporary Files
Use Narrow Frame Algorithm	C Append Listing Files
C Us Wide Frame Algorithm	📕 Hide Log Window
Reference Correction (Multiwire Detector only)	Verbosity of Listing File: 2
Multiwire Reference Correction	Snapshot Output Frequency [Images]: 100 🚔
Fewer Options	OK Cancel

Figure 7.12 - The Integration Options window with "More Options" selected

7.1.1 Active Mask

Active Mask No Generate Mask:	
Fractional Lower Limit of Average Intensity:	0.000
C Use Pre-Existing Mask:	
Active Mask File (*am):	B

Figure 7.13 - The Active Mask tool

The Active Mask is used to mask out areas that are covered by the beam stop, low temp nozzle or other obstructions. Typically a value of 0.7 is good here. SAINT will determine an active mask for each run.

NOTE: If this option is set, the user should examine the active mask images that are written into the work directory. These images can be opened with the image viewer. They should be all red except for a black area showing the shape of the obstruction, typically the beam stop.

7.1.2 Algorithm

Algorithm	C Use Narrow Frame Algorithm
	Use Wide Frame Algorithm



The algorithm buttons are used to change from narrow to wide frame integration. Frames collected with narrow scans width (less than one) should be integrated as narrow frames. Those greater than one should be integrated as wide frames.

7.1.3 Image Queue



Figure 7.15 - The Image Queue tool

The Image Queue sets the number of frames being used to determine profiles. For crystals with high mosaic spread (big wide reflections) increasing the queue size can improve integration.

- 5. Click on the button on the far right of the bottom row, Start Integration. This will start the integration and will open the SaintChart window for monitoring the integration.
- Examine the output. Double-click on any of the output entries for one additional tool the ls button at the far right of the top. This will open the SAINT listing file for that integration run (see Figure 7.16).



Figure 7.16 - Using the runs list Is tool

7.2 Monitor the Integration Run with SaintChart

SaintChart is a powerful tool for monitoring the progress of the calculation and for graphically presenting the results of the integration process.



Figure 7.17 - SaintChart view

As shown in Figure 7.17, when the SaintChart window opens the graphs are small, and under Linux the text area is large.

- 1. Change the view.
 - 1.1 Change the text area by clicking and dragging on the line between the text and graphical areas.



Figure 7.18 - SaintChart view with graphical area expanded

1.2 Expand the windows to fill the available graphical area by clicking on the four-square icon in the upper right.

To the left of the graphical area are a series of check boxes that select the displays.

- 1. Add other displays.
 - 1.1 Click on integration progress to add the new display seen in Figure 7.19. When the integration is complete, the blue progress bars will be full and the remaining time will be zero.

🗄 Integration Pr	ogress			- <u>–</u> –
Run	Images	Progress	ETA (Remaining)	
ylid_manual_01_0001.sfrm	684 (739)		22:25:28 (00:00:24)	
ylid_manual_02_0001.sfrm	0 (153)		22:28:08 (00:03:05)	
ylid_manual_03_0001.sfrm	0 (74)		22:30:13 (00:05:09)	
ylid_manual_04_0001.sfrm	0 (161)		22:32:56 (00:07:53)	
All	684 (1127)		22:32:56 (00:07:53)	
I				

Figure 7.19 - Integration progress display

One of the most powerful windows for monitoring the progress of the integration is Spot Position Overlay (in Per-Image Statistics). Pull up on the Window Tool Bar for Integrated Spots (the cursor arrow is pointing to this tool bar in Figure 7.20) to display the entire window (see Figure 7.21).



Figure 7.20 - Spot Position Overlay

In this full view, the Integrated Spots window looks much like the image viewer used previously, but there are no tools. At the bottom of the window is an entry box that can be used to change the display frequency of the Integrated Spots images. The default is shown every fifth image.



Figure 7.21 - Full view of the Spot Position Overlay

1. Examine SAINT text output.

At the bottom of the SaintChart window is a scrolling text window that displays the text output from Saint. (This window can be expanded by dragging the horizontal bar above the text as indicated by the arrow in Figure 7.22).

2. Examine the final results.

At the end of the integration, examine this text critically. In particular, look at the Overall Rsym, the coverage statistics and the unconstrained unit cell refinement.

For a typical small molecule structure, the overall Rsym is usually less than 8%. Higher values may indicate problems with absorption, twinning or poor crystal quality.

	Spot <u>S</u> h	nape_C	Correla	ation_				_		👪 In <u>te</u>	egrated	Spo
coefficient		U, İ, J		N. C.	a Maria	India	لم بالملا	•				
	25.000	I	12									
	0verall 0.000 1.000	# 12956	Pairs 1442	Uniq 1451	Merg 12948	%≺2s 17.3	<i 4707.54</i 	:≻ ≺#Si(40 27.(g> ⊲] 07 2	Bg≻ Rs .89 0.03	ym. dI/I 38 0.000	: dI/ 0.
	Centric 0.000 1.000	# 2413	Pairs 447	Uniq 454	Merg 2407	%≺2s 21.5	≺I 7064.44	<pre>> <#Si(2 39.3</pre>	g≻ <1 39 3	Bg≻ Rs .08 0.0;	ym. dI/I 33 0.000	: dI/ 0.
Co.	verage St	atistic	s for	ylid_mau	nual_Om	.raw						
										Shell.		
1	Angstrms	#0bs	Theory	%Compl	Redund	Rsym	Pairs	*Pairs	Rshell	#Sigma	*<2s	
L t	0 1.615	178	178	100.00	11.78	0.032	178	100.00	0.032	82.97	4.7	
L C	0 1.282	327	328	99.70	11.38	0.033	327	99.70	0.038	31.34	2.6	
	o 1.120	473	4/5	99.58	10.95	0.034	- 473 - 614	99.58	0.039	12 70	11.0	
L L	0 1.010	760	763	99 61	10.71	0.034	759	99 49	0.043	9 00	12.5	
L t	0 0.240	897	903	99 34	10.25	0.035	896	99 22	0.063	7 32	18.9	
L.	0 0.845	1033	1036	99 71	9 77	0.036	1031	99 52	0.000	4 84	28 1	
L.	0.808	1183	1189	99.50	9.44	0.037	1181	99.33	0.083	4.45	30.9	
l t	0 0.777	1317	1320	99.77	9.18	0.037	1313	99.47	0.098	3.34	40.3	
l t	o 0.750	1451	1452	99.93	8.93	0.038	1442	99.31	0.116	2.65	50.8	
Av Lo	eraged "l cal refin	ocal" o ement r	cell le: results	ast squa for com	ares re mponent	finemen 1 21	t =====		=== 05/0	06/04 23	2:32:31	

7.2.1 Overall Rsym and Coverage Statistics

Figure 7.22 - Final Saint Integration Statistics

Add text.

7.2.2 Unconstrained Unit Cell Refinement

Performing final unit cell least squares on file d:\structures\data_for_manual\ylic Input file contains 5552 reflections

Global Refinement -- single-component data... Maximum allowed reflections = 9999 One reflection will be stored in memory per l reflection(s) read

Orientation least squares, component 1 (5552 input reflections)... Wavelength, relative uncertainty: 0.7107300, 0.0000089

Reflection Su	mmary:						
'RLV.Excl' ar	e reflect	ions exclud	ded after	cycle 1 b	ecause RLV	error exces	eded 0.0250
Component	Input	RLV.Excl	Used	WorstRes	BestRes	Min.2Th	Max.2Th
1	5552	0	5552	8.1065	0.7884	5.025	53.580
Orientation ('UB') mat	rix:					
-0.0584509	0.03334	84 -0.048	3062				
0.0005328	0.10494	0.017	5150				
0.1573586	0.01210	09 -0.0180	0036				
A	в	с	Alpha	Beta	Gamma	Ve	ol
5.9572	9.0273	18.3668	90.088	89.999	90.033	987.'	72
0.0001	0.0002	0.0004	0.001	0.001	0.001	0.0	05
Corrected for	goodness	; of fit:					
0.0005	0.0008	0.0019	0.005	0.005	0.005	0.2	22

Figure 7.23 - Unconstrained unit cell refinement

Check the angles in the unconstrained unit cell refinement to get a quick confirmation that the initial lattice determination was correct. In the unconstrained refinement, angles that are required to be 90 or 120 are allowed to refine. If they differ considerably from the expected value then the data should be checked carefully.

M86-

7.3 Scale

The scaling process uses the SADABS program to put all of the measured data on the same scale. This process involves five steps:

- 1. Setup input files.
- 2. Parameter refinement.
- 3. Error model refinement.
- 4. Display diagnostics.
- 5. Exit.

7.3.1 Setup Input Files

 Click on Scale > Scale in the task bar. Figure 7.24 shows a portion of the initial Scale work area. The defaults and file names are typically correct.

Base	ylid_manual 😂							
Output .hkl file	ylid_manual_0m.hkl							
Diagnostic Plots File Name	ylid_manualm.eps							
Title of Diagnostic Plots	ylid_manual							
Log File	ylid_manualm.abs							
Use only centrosymmetric	Use only centrosymmetric point groups							
Point Group	mmm							
Additional Spherical Abs	orption Correction							
Mu [*] r of Equivilant Sphere								
☑ Lambda/2 Correction								
Correction Factor	0.0015							
Allow for crystal decomposit by B-value refinement	ion None 💌							
Extra Linear Correction to b to Each Reflection:	Applied None							

Figure 7.24 - FIG7.28a

M86-E000787 -

2. Click Next and check that the defaults are correct. Generally the Absorber Type is all that changes.

7.3.2 Parameter Refinement

 Click on Refine. Figure 7.25 shows a typical refinement result. The R-values are reduced and the Mean weight increases. Typical final values are 3–4% for the R-values and 0.95–0.98 for the Mean Weight.



Figure 7.25 - Xscale window after refinement

7 - 18

7.3.3 Error Model Refinement

Figure 7.26 shows typical Error Model results.



Figure 7.26 - Error model results

NOTE: If the R-values are significantly higher than this step, repeat the entire scaling calculation. Click the check box by the "bad" run and then click Repeat Parameter Refinement.

7.3.4 Display Diagnostics

- 1. Click Finish to produce a series of diagnostic plots. Some of these are recaps of the plots that have already been displayed.
- 2. Click on the "folder" tab at the bottom of the screen to select the individual plots as shown in Figure 7.27. **Why? To do what?**

Figure 7.27 - Tabs for selecting diagnostic plots

The Scale Variations plot shows the overall variation in Scale and R(int) for the individual frames. The Scale plot should be flat (for highly absorbing, irregularly shaped crystals it will probably be sinusoidal) and the R(int) plot should not show large variations (more than 2%).



Figure 7.28 - Overall scale and R(int) variations

The Intensity Statistics plots look at R(int) and IE2-1I as a function of resolution. Typical plots of R(int) versus Resolution increase to the right as seen in Figure 7.29. An IE2-1I plot should have a constant resolution. IE2-1I is a strong indicator of centric and acentric space groups. The two horizontal lines indicate the expected values for centric (top) and acentric (bottom) space groups. This plot clearly suggests an acentric space group.



Figure 7.29 - Intensity Statistics

The plots Chi-Squared values for the data as a function of resolution and intensity should be mostly flat. Values below one are not unusual for weaker high resolution data. The plots shown in Figure 7.30 are typical.



Figure 7.30 - Chi-squared

M86-

The special distribution plots are generated for each data collection run. They indicate, using red and blue dots, points of average intensity for a particular reflection after scaling. The actual intensity measured at that spot on the detector vary by more than three standard deviations. Figure 7.31 show two spatial distribution plots side by side. This is the way they are typically output by Scale.



Figure 7.31 - Spatial distribution

Figure 7.32 shows a single plot so that the dots are easier to see. Note that the points of disagreement are spread fairly evenly over the entire detector face.



Figure 7.32 - A single Spatial Distribution

If the spots are clustered in an area or if there are significantly more spots of one color than there are of another, then the data should be examined critically. Figure 7.33 is from another data set where the active mask was not used during integration and the reflections collected in that area are consistently smaller than

expected. This kind of problem may affect the final results.

Scan 1, detector 2-theta -12.00°, II-<I>I >3.00su (red+, blue-), errors (black)



Figure 7.33 - A Spatial Distribution plot showing a problem area

7.3.5 Exit

1. Click on Exit XScale to close the scaling GUI.

```
Exit AXScale
```

Figure 7.34 - Exit XScale

NOTE: Scale is one of the few GUIs where it is necessary to exit using a special exit button. This avoids an error message that SADABS has exited improperly.

M86-

8. Examine Data

In this step, the space group for the sample is determined and optional simulated precession photographs are calculated to further evaluate the overall quality of the data. APEX2 provides two tools for this: XPREP for space group determination and Precession Images for looking at undistorted slices of reciprocal space. This is the final step before beginning the structure solution and refinement process.

8.1 XPREP

- 1. Click on XPREP (Space Group Determination) under Examine in the task bar.
- 2. Check that the two files in the pop-up window are correct (see Figure 8.1) and click OK.

Select Files For XPrep	<u>?</u> ×
P4P file: guest\data_manual\ylid_manual\work\ylid_manual_0m.p4p	B
HKL file: guest\data_manual\ylid_manual\work\ylid_manual_0m.hk	1
0K Can	cel

NOTE: The scaling process has created two files: ylid_manual_0m.p4p containing the final unit cell parameters from integration and ylid_manual_0m.hkl containing the corrected intensities. Typically these are the files to use for

Figure 8.1 - Select files for XPREP input

space group, but you can browse to choose other files.

8.2 Space Group Determination

 XPREP evaluates the data and looks at the mean intensities and the mean int/sigma. Since these are large for all groups except P, XPREP suggests that the lattice is P (see Figure 8.2). Press Enter to accept.

Figure 8.2 - Lattice statistics

2. XPREP determines the reduced cell based on the lattice entered above. Since the lattice was primitive and the magnitudes of the cell dimensions were proper (a<b<c), the original and reduced cells are the same (see Figure 8.3). Press Enter to search for a higher symmetry cell.

C XPREP	Ver 6.	.14 W95	/98/NT	/2000/№	1E Copyr	ight Bru	uker Nor	nius 2	_ 🗆 🗙
Current c	lataset	: ylid_m	anual_Om			Wavele	ngth: 0.	71073 <mark>Ch</mark>	iral: ?
Original	cell: Esds:	5.957 0.001	9.027 0.001	18.366 0.002	90.00 0.00	90.00 0.00	90.00 0.00	Vol Lat	987.7 tice: P
Current o	cell:							Vol	987.7
Matrix: 1									1.0000
[D] Read, modify or merge DATASETS [C] Define unit-cell CONTENTS [P] Contour PATTERSON sections [F] Set up shelxt1 FILES [H] Search for HIGHER metric symmetry [R] RECIPROCAL space displays [S] Determine or input SPACE GROUP [U] UNIT-CELL transformations [A] Masorption, powder, SIR, SAD, MAD etc. [T] Change TOLERANCES [M] Test for MEROHEDRAL TWINNING [O] Self-rotation function [L] Reset LATTICE type of original cell [Q] UUT program Select option [H]: []								TS NYS NS	

Figure 8.3 - Reduced cell

For the YLID, there is no higher symmetry cell found. The program has determined that the YLID crystal has an orthorhombic primitive lattice (see Figure 8.4). Press Enter to accept.

Determination	of reduce	d (Niggli) cell					
Transformation 1.0000 0.0	from ori 0000 0.00	ginal cel 00 0.0	1 (HKLF-	matrix) 000 O.	: 0000	0.0000	0.0000	1.0000
Unitcell:	5.957	9.027	18.366	90.00	90.00	90.00		
Niggli form:	a.a = b.c =	35.49 0.00	b.b a.c		81.49 0.00	c.c = a.b =		.32
Search for hig Identical indi	ther METRI ces and F	C symmetr riedel op	y posites	combine	d before	e calcula	ting R(:	sym)
Option A: FOM	= 0.000 d	eg. ORT	HORHOMBI	C P-lat	tice]	R(sym) =	0.019 [4105]
Cell: 5.957 Matrix: 1.0000	9.027	18.366	90.00	90.00 1.0000	90.00	Volum 0.0000	e: 0.0000	987.68 1.0000
Option B retai	ns origin	al cell						

Figure 8.4 - Higher symmetry cells

3. The next logical step is to determine the space group. XPREP suggests this (see Figure 8.5). Press Enter to determine the space group.

🗖 XPREP Ver 6	.14 W95	/98/NT	/2000/N	1E Copyr	ight B <mark>r</mark> ւ	uke <mark>r</mark> Nor		_ 🗆 🗙
Current dataset	;; ylid_m	anual_Om		Wavele	ength: 0.	71073 <mark>Ch</mark>	iral: ?	
Original cell: Esds:	5.957 0.001	9.027 0.001	18.366 0.002	90.00 0.00	90.00 0.00	90.00 0.00	Vol Lat	987.7 tice: P
Current cell:							Vo 1	
Matrix: 1.0000								
Crystal system:	Orthorh	ombic		Lattice				
[D] Read, modify or merge DATASETS [C] Define unit-cell CONTENTS [P] Contour PATTERSON sections [F] Set up shelxtl FILES [H] Search for HIGHER metric symmetry [R] RECIPROCAL space displays [S] Determine or input SPACE GROUP [U] UNIT-CELL transformations [A] Absorption, powder, SIR, SAD, MAD etc. [T] Change TOLERANCES [M] Test for MEROHEDRAL TWINNING [O] Self-rotation function [L] Reset LATTICE type of original cell [Q] QUIT program								

Figure 8.5 - Determine the space group

4. If the space group is known or if the compound is known to be chiral, enter that information (see Figure 8.6). Generally, it is sufficient to press Enter to start the space group determination.

[S]	Determine SPACE GROUP
[C]	Must be CHIRAL (sample is optically active)
[N]	NOT NECESSARILY chiral (eg. may be racemate)
[1]	INPUT known space group
[E]	EXIT to main menu or [Q] QUIT program
Sel	ect option [S]:

Figure 8.6 - Space group options

5. XPREP has chosen the crystal system [O] (see Figure 8.7). Press Enter to accept.

[A]		[M] Mon	ic, [0]	Or	thor	nombic,	[T]	Tetragonal,
[H]	Trigonal/He	xagonal,	Cubic		[E]	EXIT		
		0] :						

Figure 8.7 - Choose the crystal system

6. XPREP has chosen the crystal lattice [P] (see Figure 8.8). Press Enter to accept.

Lattice exceptions							Obv	Rev	A11
N (total) =		8071	8097	8092	8088	12130	10773	10764	16154
N (int>3sigma) =		6377	6375	6480		9616	8529	8517	12777
Mean intensity =	0.0	205.0	196.9	192.1		198.0	209.4	206.8	205.8
Mean int/sigma =	0.0								
Lattice type [P, A Select option [P]:	, в,	C, I, F	, O(obv	.), R(r	ev. rho	mb. on	hex. ax	es)]	

Figure 8.8 - Choose the lattice

7. XPREP evaluates the data and looks at the systematic absences for all possible glide planes and screw axes (see Figure 8.3). These are displayed across the middle of the figure. By examining the number of reflections with I>3 sigma(I), the mean intensities, and the mean int/sigma, which should all be very small for a systematic absence, XPREP derives a suggested space group, P2(1)2(1)2(1). Press Enter to accept.

8.3 Reflection Statistics.

Mean $ E^*E-1 = 0.694$ [expected .968 centrosym and .736 non-centrosym]												
Systems	atic :	absenc	e excep	tions								
N	761	768	759	18	437	434	443	28	308	314	306	39
N I>3s	572	557	549		319	325	280		239	2.53	216	
<1> 2	25.1	238.3	274.2		457.1	405.7	201.4		277.4	337.8	291.0	0.8
<i s=""></i>					14.2	14.0	10.9	0.5		14.9		
Identio Option	al i: Spa	ndices ce Gro	and Fr up No.	iedel Type	opposi Axes	ites co s CSD	ombined R(sym)	befor) N(ed	te calo A) Sys	ulatin st. Ab:	ng R(sy s. CF	m) OM
[A] P2	(1)2(1)2(1)	# 19	chira	al 1	5917	0.019	410	05 0.0		.9 1.	
Select	opti	on [A]	:									

Figure 8.9 - Systematic absences and a suggested space group

- 1. XPREP returns to the general menu seen previously (see Figure 8.5). This time D is chosen to evaluate the dataset. Press Enter to accept.
- 2. There are multiple choices for data manipulation (see Figure 8.10). "S" chooses a display of statistics.



Figure 8.10 - The data manipulation menu



Figure 8.11 - The merge data menu

4. A table of statistics appears (see Figure 8.12). Examine the data presented. Is the completion near 100%? Is the redundancy good? Are Rint and Rsigma small and increasing smoothly from top to bottom? In particular look at the last two lines which compares all of the data with the high resolution data. The completion should be near 100% for both the high resolution shell and the complete data set. The redundancy and Rint should be similar for the two. The Mean Intensity and the Mean I/sigma(I) will usually be quite different. The Mean I/sigma(I) for the high resolution data should be greater than 3.0.

For this data set the difference between the Rint for the high resolution data and the complete data set is slightly larger than expected. Press Enter to continue.

🔁 XPREP Vei	r 6.14 W	195/98	/NT/2000)/ME Copyr	ight Bru	iker Nor	nius 2	_ 🗆 🗙
Resolution	#Data #	Theory	<pre>%Complete</pre>	Redundancy	Mean I	Mean I/	s Rint	Rsigma
Tof = 1.95			100.0	12 07	1140.8	71 33	0 0295	0.0118
1.95 - 1.50	105	105	100.0	10.94	495.4	66.41	0.0256	0.0151
1.50 - 1.30	108	108	100.0	11.38	276.9	65.26	0.0329	0.0145
1.30 - 1.15	126	126	100.0		211.2		0.0327	0.0157
1.15 - 1.05	134	134	100.0	9.61	119.8	46.09	0.0361	0.0174
1.05 - 0.95	176	176	100.0	8.94			0.0467	0.0221
0.95 - 0.90			100.0	8.56		32.54	0.0580	0.0262
0.90 - 0.85	148	148	100.0	8.14	43.2	24.60	0.0743	0.0352
0.85 - 0.80	199	199	100.0	7.23	37.0	20.31	0.0822	0.0441
0.80 - 0.75	236	236	100.0	6.67	27.7	14.66	0.1113	0.0638
0.75 - 0.70	327	327	100.0	5.92	18.4	8.81	0.1506	0.1083
0.70 - 0.67	248	256	96.9	4.50	10.8	4.51	0.2104	0.2361
0.00.0.07	011	010			10.7	0.00	0 1400	0.1116
0.00 - 0.07	2027	2045	99.0	3.09	146.0	20.02	0.1400	0.0206
1111 - 0.07	2037	2043	33.0		140.0			0.0200
Merged [A],	lowest	resolut		37 Angstrom	as, 65	0 outlie	rs downw	eighted
Enter <cr> t</cr>		ue						

Figure 8.12 - Intensity statistics

5. Since the difference between the Rint for the high resolution data and the complete data set is slightly larger than expected, it is reasonable to apply a high resolution cutoff using the H option in the data manipulation menu. (see Figure 8.13). Type H and press Enter.

[M]	Sort-MERGE current data (no scaling)	[0]	Change CURRENT dataset
[L]	LEAST-SQUARES scale and merge datasets	[W]	WRITE dataset to file
[1]	INCLUDE Rfree flags from another file	[R]	READ in another dataset
[S]	Display intensity STATISTICS	[D]	DELETE stored dataset
[F]	FACE-indexed absorption corrections	[P]	PSI-scan absorption corr.
[T]	Copy file, TRANSFORM hkl and cosines	[A]	MAD, SAS, SIR or SIRAS
[H]	Apply HIGH/low resolution cutoffs	[N]	NORMALIZE/scale sigmas
[G]	Generate simulated powder diagrams	[E]	EXIT to main menu
[Q]	QUIT program		
Sel	ect option [E]: <mark>h</mark>		

Figure 8.13 - Requesting a high resolution cutoff

M86-E00078

8.4 Applying High Resolution Cutoff

1. Enter a high resolution limit of 0.75 (see Figure 8.14) and press Enter to accept the low resolution cutoff of infinity.

High resolution limit in Angstroms [1.0]: 0.75

Figure 8.14 - The resolution cutoff menu

2. Applying the cutoff removes approximately 3000 reflections from the data set (see Figure 8.15). The next step is to see if the cutoff has improved the statistics. Type S and press Enter.

XPREP Ver 6.14 W95/98/NT/2000/ME Copyright Bruker Nonius 2 💶 🗙							
Index # Data Filename or Source of Data							
1 16154 ylid_manual_Om.hkl 2 13067 ylid_manual_Om.hkl <- curr	rent dataset						
 [M] Sort-MERGE current data (no scaling) [L] LEAST-SQUARES scale and merge datasets [I] INCLUBE Firee flags from another file (S) Display intensity STATISTICS [F] FACE-indexed absorption corrections [T] Copy file, TRANSFORM hki and cosines (H) Apply HIGH/low resolution cutoffs [G] Generate simulated powder diagrams [O] QUIT program 	 [C] Change CURRENT dataset [W] WRITE dataset to file [R] ERAD in another dataset [D] DELETE stored dataset [P] PSI-scan absorption corr. [A] MAD, SAS, SIR or SIRAS [N] NORMALIZE/scale signas [E] EXIT to main menu 						
Select option [E]: <mark>s</mark>							

Figure 8.15 - Requesting statistics after applying a high resolution cutoff

3. As before in step 3, enter [A] to merge all equivalents.

4. After the cutoff, all of the statistics look better (see Figure 8.16). Press Enter to continue.

Resolution	#Data	#Theory	<pre>%Complete</pre>	Redundancy	Mean I	Mean I/s	Rint	Rsigma
Inf - 2.25	74	74	100.0	12.59	1369.5	73.60	0.0292	0.0116
2.25 - 1.70			100.0	11.18	584.2	66.45	0.0284	0.0145
1.70 - 1.45			100.0	10.84	414.1	67.42	0.0285	0.0142
1.45 - 1.30			100.0	11.34		63.58	0.0342	0.0152
1.30 - 1.15	126	126	100.0	10.11	211.2	56.76	0.0327	0.0157
1.15 - 1.05	134	134	100.0	9.61	119.8	46.09	0.0361	0.0174
1.05 - 0.95	176	176	100.0	8.94	92.5	40.47	0.0467	0.0221
0.95 - 0.90			100.0	8.56		32.54	0.0580	0.0262
0.90 - 0.85	148	148	100.0			24.60	0.0743	0.0352
0.85 - 0.80	199	199	100.0				0.0822	0.0441
0.80 - 0.75			100.0		27.7	14.66		0.0638
0.85 - 0.75	435	435	100.0	6.92	31.9	17.25	0.0949	0.0534
Inf = 0.75	1462	1462	100.0	8.94	197.5	39.10	0.0339	0.0168
Merged [A],	lowest	resolut		37 Angstrom	s, 610) outlie:	rs downw	eighted
Enter <cr> t</cr>		nue						

Figure 8.16 - Statistics after high resolution cutoff

5. This response will return to the data manipulation menu (see Figure 8.10, Figure 8.13 or Figure 8.15). The default answer should be E for exit. Press Enter to accept this default.

8.5 Preparing an Output File

- 1. Exit the data manipulation menu to return to the general menu. This time chose C to define the unit cell contents.
- A window will open displaying the current formula, Z, the density and the atomic volume (see Figure 8.17). In this example, the formula is incorrect and Z has been set to six to try to achieve a reasonable density and atomic volume. Since this formula is incorrect it must be modified now. The correct chemical formula for the YLID crystal is C11 H10 O2 S.
 - 2.1 At Select Option do not accept the default answer of E. Type in F to enter a new formula.
 - 2.2 In response to the question "Enter Formula," type the correct formula C11 H12 O2 S1 and press Enter.
 - 2.3 Check that the information is correct. Check that Z seems reasonable for the space group, that the density is as expected (1.1 to 1.4 for organic molecules, higher for inorganic compounds), and that the atomic volume is around 17 or 18. Significant variation from the expected values may indicate an incorrect molecular formula or missing counter ions or solvates. The values at the bottom of the window look fine for the YLID. Press Enter to accept.

M86-E00078

■ XPREP Ver 6.14 W95/98/NT/2000/ME Copyright Bruker Nonius 2 Current formula is: Cil Tentative Z (number of formula units/cell) = 6.0 giving rho = 1.333,	3.	The n write o solutio Enter	ext de out the on pro to ace	efault e files ocess cept.	action nece (see	n for 2 ssary Figur	XPRE / for th re 8.1	EP is t ne stru 8). Pr	o ucture ess)
non-H atomic volume = 15.0 and following cell contents and analysis:	Curr	ent dataset	t: ylid_m	anual_On	.hkl		Wavele:	ngth: 0.7	71073 <mark>Chi</mark>	iral: ?
C 66.00 100.00 *	Orig	inal cell:	5.957	9.027	18,366	90.00	90.00	90.00	Vol	987.7
[Z] change Z, [F] new FORMULA, [R] change RADIATION,		Esds:							Latt	ice: P
[2] EXIL CO main menu Or [0] QUIT program	Curr	ent cell:							Vol	
Select option [E]: f	Matr	ix: 1.0000	0.0000	0.0000	0.0000	1.0000	0.0000	0.0000	0.0000	1.0000
Enter formula; numbers follow elements or brackets, 2nd character of element name must be lower case, may include: Me, Et, Pp, Bu, Ph, or Cp: C11 H10 or S1	Crys	tal system:	: Orthorh	ombic <mark>S</mark>	pace gro	up: P2(1) <mark>#</mark> 19	[chi] <mark>[</mark>	aue: 3
	Form Z:	ula: C11 H: 4.00 Dens:	12 02 S1 ity: 1.4	01 At.v	ol: 17.	6 F (000): 44	Formul 0.00 Mu	la wt: [mm-1]:	208.27
Tentative Z (number of formula units/cell) = 4.0 giving rho = 1.387, non-H atomic volume = 17.6 and following cell contents and analysis:		Read. modi:	fy or mer	ce DATAS	ETS		Define	unit-cel	1 CONTENT	TS
C 44.00 64.05 % H 40.00 4.89 %	[P]	Contour PA	TTERSON =	ections		[F]	Set up	shelxtl 1	FILES	
0 8.00 15.52 % S 4.00 15.54 %	[H] [S]	Search for Determine (HIGHER n or input	etric sy SPACE GE	mmetry IOUP	[R] [V]	RECIPRO UNIT-CE	CAL space LL transi	e display formatior	7S NS
[2] change Z, [F] new FORMULA, [R] change RADIATION,	[A]	Absorption	, powder,	SIR, SA	LD, MAD e	tc. [T]	Change	TOLERANCI	ES	
[E] EXIT to main menu or [Q] QUIT program	[L]	Reset LATT	ICE type	of origi	nal cell	[0]	QUIT pr	ogram	unction	
Select option [E]:	Sele	ct option	[F]:							

Figure 8.17 - Defining and checking the unit cell contents

Figure 8.18 - Requesting output files

 The program asks for an output file name. Since this data has been cutoff at a resolution of 0.75A, it makes sense to change the output file name to ylid_res75 to reflect the cutoff (see Figure 8.19). Press Enter to accept.

Current dataset:	ylid_m	anual_Om			Wavele:	ngth: O.	71073 C	hiral:
Original cell: Esds:	5.957 0001	9.027 0.001	18.366 0.002	90.00 0.00	90.00 0.00	90.00 0.00	Vol La	987.7 ttice: F
Current cell:							Vol	
Matrix: 1.0000								
Crystal system:		ombic <mark>S</mark>	pace gro	up: P2(1) <mark>#</mark> 19		Laue: 3
Formula: C11 H12 Z: 4.00 Densit	02 S1	D1 At.v		6 <mark>F (000</mark>		Form 0.00 M	ila wt: i[mm-1]:	208.27
 [D] Read, modify [P] Contour PAT [H] Search for 1 [S] Determine or [A] Absorption, [M] Test for MED [L] Reset LATTIC 	y or mer TERSON so HIGHER mu c input : powder, ROHEDRAL CE type o	ge DATAS ections etric sy SPACE GF SIR, SA TWINNIN of origi	SETS Numetry ROUP ND, MAD e IG .nal cell	[C] [F] [R] [U] etc. [T] [O] . [Q]	Define Set up RECIPRO UNIT-CE Change Self-ro QUIT pr	unit-ce. shelxtl CAL spac LL trans TOLERANG tation : ogram	ll CONTE FILES ce displ sformati CES function	NTS ays ons
Select option []								
Output file name			nsion) [y	lid_manu	a1_0m]:	ylid_res	s75 <mark>.</mark>	

Figure 8.19 - Changing the file name

5. After entering the file name (or hitting return to accept the default file name) an input file for the structure solution module is created and displayed on the screen. And the program asks "Do you wish to (over)write the intensity data file ylid_res75.hkl." Since the file name has been changed, this question must be answered with a y (see Figure 8.20).



Figure 8.20 - The input file for structure solution and a final question

6. Exit XPREP (see Figure 8.21). Press Enter to exit the program.



Figure 8.21 - Exit XPREP from the general menu

NOTE: There are many other features in XPREP that can be accessed from the general menu; reciprocal space plots, simulated powder patterns and a test for merohedral twinning are very useful tools.

8.6 Simulated Precession Images

 Click on Precession Images under Examine in the task bar. Precession Images provide an undistorted view of layers of the reciprocal lattice. APEX2 generates simulated precession images by finding the appropriate pixels in a series of ".sfrm" images. You must specify the images to examine and the zones to calculate.

T python		? ×
Cook in: Cook in:	nual/ylid_manuak matrix_01_ matrix_01_ matrix_01_ matrix_01_ matrix_01_ matrix_01_ matrix_02	
a matic_01_0004.stm a matic_01_0004.stm a matic_01_0005.stm a matic_01_0006.stm ↓ Directory: ylid_manual	matrix_02_ matrix_02_ matrix_02_ matrix_02_	ОК
File type: Directories		Cancel

Figure 8.22 - The simulated precession work area

2. To open the file selection window, left-click on the folder beside the file name. Choose the sets of images that you want to use for the calculation by clicking on the check boxes (see Figure 8.23).

📅 python	? ×
Look jn: C/struc/guest/data_ma work communication log-20040502 gz matrix_01_0001.stm matrix_01_0002.stm matrix_01_0003.stm matrix_01_0006.stm matrix_01_0006.stm	nucl/ylid_manucl/
Directory: viid_manual File type: Directories	OK Cancel

Figure 8.23 - File Selection window with matrix runs deselected

3. Check the default input value. Modify as needed and press Calculate.

Calculating more zones does not significantly increase the calculation time. In the example, 1kl, h1l and hk1 have been added to the defaults of 0kl, h0l and hk0. Fractional values (e.g., 0.5kl) are allowed. Reducing the resolution may speed the calculations slightly. The thickness defines the range of pixels above and below the requested range. For example, if the 0kl zone is requested with a thickness of 0.1 then the simulation is looking for all pixels that have -0.1<h<0.1, and any value, including fractional values, for k and l.

Planes	0kl h0l hk0 hk1 1kl h1l	
Images from	ruc/guest/data_manual/ylid_manual	2
	(1127 image files in 4 runs)	
best resolution	0.77	
Thickness	0.10	
	Calculate	
Created files		

Figure 8.24 - Detail of the input section

4. A progress bar will appear and after approximately 5–10 minutes (depending on the number of "sfrm" files read) an image will appear on the screen. Display other planes by clicking on the calculated images to the bottom right of the work area or by browsing as usual with the view plugin.

Synthesizing precession images	? ×
Processing 1127 image files	Ŀ\$
	2%
	Cancel



NOTE: This is a relatively slow calculation since every sfrm file is examined to determine if it has pixels that would contribute to a requested zone. For example, if the 0kl zone is requested with a thickness of 0.1 then the simulation is looking for all pixels to have -0.1<h<0.1 and any value, including fractional values, for k and l.

 User the simulated patterns to check space group symmetry (see Figure 8.25, Figure 8.26 and Figure 8.27) and to find signs of twinning (see Figure 8.28).

I am having trouble generating acceptable images for 8.26 to 8.29, but they will get done eventually.

Include SIMPREC5.bmp here

Figure 8.26 - The 0kl plane for the test YLID

Include SIMPREC6.bmp here

Figure 8.27 - The h0l plane for the test YLID

Include SIMPREC7.bmp here

Figure 8.28 - The hk0 plane for the test YLID

Include SIMPREC7.bmp here

Figure 8.29 - A plane from a rotationally twinned crystal showing the two lattices

Curt will add images of a good and bad example.

9. Structure Solution and Refinement

9.1 Overview

You are now ready to solve and refine the crystal structure for the YLID test crystal. The various steps in solving and refining the structure are carried out within the APEX2 GUI. These tools will prepare input files for the solution and refinement programs of the SHELXTL package. The integration process (SAINT) has produced two important files—YLID1M.p4p, containing the final unit cell parameters and other important information on how the experiment was carried out, and YLID1M.raw, containing the actual intensity data. These files are all that is required to begin the structure solution and refinement process. The various steps in solving and refining the structure are carried out using the programs of the **SHELXTL package**.

9.2 Solve the Structure

The process of obtaining an initial model of the compound is started by clicking in the Task Bar on Solve Structure and then on Structure Solution.

- 1. This will open the Structure Solution GUI. Check that there is a reasonable formula in the formula window.
- 1.1 If you entered a correct formula in the display GUI, then all is ready to start a normal direct method run.
- 1.2 If the formula is incorrect, change it in the formula window and press Return. The input file will update.



Figure 9.1 - Default Solve Structure view

2. Click on Solve Structure to start the calculations and open an output window. This window can be used to monitor the progress of the direct methods calculations.

Tructure Solution Log Window

- 11	
	172 Reflections and 3346. unique TPR for R(alpha) 465 Unique negative quartets found, 465 used for phase refinement
	110 Unique NQR employed in phase annealing 128 Parallel refinements, highest memory = 3822 / 44055
	Try Ralpha Nqual Sigma-1 M(abs) CFOM Seminvariants
	1890781. 0.064-0.770 0.946 1.154 0.064* ++ ++ +-+- Freq: 0 0 0 97 13 0 0 0 0 0 0 0 1 0 0 1 0 1 0 0 1 0 0 1 2 1 1 1 1
	917713. 0.060 -0.799 0.946 1.151 0.060° ++ + Freq: 0 0 1 201 25 0 0 0 0 0 0 0 0 1 0 0 1 0 1 2 2 1 0 0 1 2 2 2 1 1 / 256
	256. Phase sets refined - best is code 917713. with CFOM = 0.0597
	Fourier and peaksearch
	RE = 0.154 for 14 atoms and 467 E-values Fourier and peaksearch RE = 0.140 for 14 atoms and 467 E-values Fourier and peaksearch
	+ ylid_reso75 finished at 07:46:22 Total elapsed time: 0.9 secs +

Figure 9.2 - Structure Solution output

 Click on the arrow to the right of Direct 1 in the Method box (see Figure 9.3). This gives three choices for structure solution using direct methods.

-Method-		
e	Direct 1	
c	Dual Space 1	
0	Patterson 1 🔍	🗹 Structure Expansion
-	Computing Effort (log scale)	+
	ŀ	

Figure 9.3 - Method box

M86-E00078

In most cases the default values will give a good initial model, but there are several other options available for more difficult problems.

- 1. Chose the solution method by clicking the appropriate radio button on the left in the Method box.
- 2. Highlight and click on Direct 1 to set up a default run with a simple TREF instruction.
- Highlight and click on Direct 2 to set up an extended run with TREF 10000. By adding 10000 to the TREF command, more attempts are made to determine a solution. This will take more time, but if you see a good solution in the output window it is possible to stop at that point by clicking on the STOP button. Good solutions typically have a CFOM of 0.06 or less.
- 4. Highlight and click on Direct 3 to set up an extreme run with two instructions, ESEL 1.0 and TREF 100000. By adding ESEL 1.0, more reflections are used in the solution process. This is particularly useful for acentric triclinic structures and for pseudo symmetric structures. Increasing the number of tries in the TREF command to 100000 runs until ended by clicking on the STOP button. There is only one predefined option for the dual space calculations and also only one for the heavy atom Patterson methods.

At the end of the direct methods runs, the output display will look like the figure. Good figures of

merit are near 1.0 for Sigma-1 and M(abs) and less than 0.06 for Ralpha and the CFOM. Structures can be solved with figures of merit that deviate from these numbers, but they may require more effort.

If all looks reasonable, click OK in the output display and Exit in the solutions plug-in. The next step is structure refinement using XSHELL.

9.3 XSHELL

9.3.1 Refine the Structure

The structure solution step produced statistics that indicate a solution had been found. The real proof is in the initial model that is produced. XSHELL provides the tools to view the model and determine if it makes chemical sense.

- 1. Start XShell, the molecular viewing and refinement plug-in, by clicking on Refine Structure in the Task Bar and then on the Structure Refinement icon.
- This will open a window to select files for XShell. In usual operations, the file name will be preselected. If a different file is desired, select Browse.

🕈 Sele	ct Files For XShell	<u>? x</u>
RES file:	C:\struc\guest\ylid_manual\work\ylid_reso75.res	
	OK	Cancel

Figure 9.4 - The Select Files tool

3. Click on OK to open a molecular display window.



Figure 9.5 - Initial view of the YLID molecule

M86-E00078

4. Right-click in the background to open a popup window with options to change the

	image.	
¥	Wireframe	
	Ball And Stick	
	<u>T</u> hermal Ellipsoid	
	Pipes	
	Select atoms	
	Hi <u>d</u> e Part(s)	
	Show <u>a</u> ll Atoms	
	Disassogiate Atoms	
	Unit Cell	
•	Hydrogen Labels	
•	Labels	
	Sort	
	Grow	
	Pac <u>k</u>	
	Tri <u>m</u>	
	Zoom Out	
	Toggle Pan And Rotate	
	Restore Deleted Atoms	
	Eind Duplicate Labels	
	Information on All Atoms	
	Add Hydrogen atoms	

Figure 9.6 - XSHELL menu
The Information On All Atoms option shows a list of the peaks and their heights. The drop in height between Q13 (the last real peak) and Q14 (the first noise peak) is typical of a correct solution.

	LABEL	PEAK HEIGHT	TYPE	1NI OKHA	v Non On P	7	HVBRID	CHARGE	LIEO
1	51	0.0000	s	0.8105	0.1820	0.2401	sp3	0	0.05
2	01	161.0300	0	0.6925	0.10	0.1253	sp3	0	0.05
3	Q2	158.7400	Q.	0.3333	0.3057	0.1756	sp3	0	0.05
4	Q3	155.9800	Q	0.4332	0.1961	0.1488	sp3	0	0.05
5	Q4	141.7900	Q	0.5055	0.3	0.685	sp3	0	0.05
6	Q5	141.6800	Q	0.8344	0.898	0.1307	sp3	0	0.05
7	Q6	140.8200	Q	0.6372	0.1262	0.1726	sp3	0	0.05
8	Q7	140.6700	Q	0.3537	0.1161	0.834	sp3	0	0.05
9	Q8	127.0800	Q	0.1821	0.1416	0.360	sp3	0	0.05
10	Q9	109.9200	Q	0.8324	0.3761	0.2281	sp3	0	0.05
11	Q10	109.4100	Q	0.1499	0.486	0.244	sp3	0	0.05
12	Q11	104.8200	Q	0.2908	0.615	0.380	sp3	0	0.05
13	Q12	98.8900	Q	0.4789	0.856	0.93	sp3	0	0.05
14	Q13	85.8200	Q	0.6561	0.1836	0.3232	sp3	0	0.09
15	Q14	66.1600	Q	0.5674	0.1814	0.2395	sp3	0	0.05
16	Q15	64.7300	Q	0.8062	0.4621	0.2409	sp3	0	0.05
17	Q16	54.7300	Q	0.9493	0.1856	0.1563	sp3	0	0.05
18	Q17	40.2200	Q	0.52	0.989	0.237	sp3	0	0.05

Figure 9.7 - Atom information screen

At the bottom left of this window is a slide bar which is used for deselecting peaks. The arrow can be moved by left-clicking and dragging the arrow or by left-clicking on either side of the arrow. Left-clicking to the left of the arrow (i.e., where the cursor is pointed in Figure 9.8) will remove peaks.

-Residual	(Difference Fourier Q Peaks)	
0	No. of QPeaks : 22	· · · · 22

Figure 9.8 - The Q-peak slider

5. Slide the pointer down while watching the molecular display. Change the number of Q peaks to 15.

At 15 the molecule is much cleaner, but there are still peaks that do not make sense.



Figure 9.9 - Image after the number of Q peaks was changed to 15 $\,$

Two more clicks left on the Q-peak slider gives a reasonable model.



Figure 9.10 - Image with all noise peaks removed

 Put the cursor over the bonds to check distance. The distance will be displayed at the bottom right. The distance of 1.178 is typical for C=0 in aromatized ring systems.

9.3.2 Label the Atoms

1. Label the atoms and set the atom types. Left-click on atoms to select them. Left-click on the peaks for the two oxygen atoms (Q2 and Q5 in this example).



Figure 9.11 - View of model with the probable oxygen peaks selected

2. If it is difficult to see the color and labels, change the color scheme by left-clicking on Preferences and then right-clicking on Background Color. Choose colors and click Apply. Click Cancel to exit the background color mode. Note that the selected peaks are blue in Figure 9.13.

Preferences Help		
Atom Preferences	Ctrl+F	Q Q
Open <u>G</u> L Preferences	Ctrl+G	
🥦 XShell GUI St <u>y</u> le	•	SELECT BACKGROUND COLOR
Background Color	Ctrl+B	C Solid
bs		Gradient Foreground Background
		<u>Apply</u>
Jon.		

Figure 9.12 - Selecting the background color



Figure 9.13 - New background color

3. Right-click to view a pop-up menu and select Labelling.



Figure 9.14 - Select Labelling

M86-E00078

The Atom Labelling box will open.

ATOM LABELLING IN ORDER OF SELECTION
First Sequence # 1 Ascending Suffix Descending
Relabel Cancel

Figure 9.15 - Atom Labelling box

- 4. The two selected atoms need to be changed to oxygen atoms. Do this one of two ways:
- Click on the Element field and type in the element symbol (case does not matter).
- Click on the El button to the right of the Element field. This will open a periodic table. Left-click on the appropriate element symbol to select it (the periodic table will automatically close).

F	Perio	odic	Tal	ble														1	! ×
	IA							PE	RIO		ΓABL	.E						VIII	A
	н	IIA											III1	IVA	VA	VIA	VIIA	He	
	Li	Ве											В	с	N	0	F	Ne	
	Na	Mg	IIIB	IVB	VB	VIB	VIIB		VIII	в	IB	IIB	Al	Si	Ρ	s	а	Ar	
	к	Ca	Sc	Ti	۷	Cr	Mn	Fe	Co	Ni	Cu	Zn	Ga	Ge	As	Se	Br	Kr	
	Rb	Sr	Y	Zr	Nb	Mo	Tc	Ru	Rh	Pd	Ag	Cd	In	Sn	Sb	Te	Ι	Xe	
	Cs	Ba	La	Hf	Ta	w	Re	Os	Ir	Pt	Au	Hg	Tİ	Рb	Bi	Po	At	Rn	
	Fr	Ra	Ac		_											_			
			_		Се	Pr	Nd	Pm	Sm	Eu	Gd	ть	Dy	Но	Er	Tm	Yb	Lu	
	G	ancel			Th	Pa	U	Np	Pu	Am	Cm	Bk	Cf	Es	Fm	Md	No	Lr	
																		Q	

Figure 9.16 - Refine014

5. Once the element type is set correctly, click on Relabel to number the peaks sequentially in the order they were selected starting with the number in the First Sequence # field.

 Seeing the color of atoms can be difficult in the Wireframe view used so far. Right-click in the background with no atoms selected to get a slightly different action menu. In this menu, highlight and left-click on Pipes. This makes the bonds thicker and makes it obvious that there is an incorrectly labeled atom. That will be corrected when the carbon atoms are labeled.



7. Left-click on the remaining peaks in the order that you want them labeled.

NOTE: If working on the YLID test data, look at Figure 9.18 and click on the peaks to give the same order as used here. Right-click and choose Labelling. Select Carbon and apply the labels.

- In the labelling tool, change the el type to C (carbon). The starting atom number will change to 1—the first available number for carbon atoms.
- 9. Click Relabel.
- 10. Click Cancel to close the window.

9.3.3 Refine the Molecule

The labeled ylid molecule is now ready for refinement.



Figure 9.18 - Correctly labeled model

 Left-click on the Refine icon in the tool bar (the cursor is pointing to it in Figure 9.19) or Type Control+R.



Figure 9.19 - The Refine icon

This will open the Refine Box. Click on Refine to launch the least-squares refinement program, XL using the default parameters as given.

REFINE
Least squares options
🔽 Full Matrix
🗖 Conjugate Gradient
Refinement Cycles 4
Number of residual (Q)peaks
Sigma Cutoff 🗌 n Sigma 📃
Resolution Cutoff 🔲 2 Theta
Invert structure
Late-stage refinement options
🗖 Refine all non-H atoms anisotropically
Use suggested weights
Squared Term 0.000000
Linear Term
Refine extinction parameter
Generate ACTA[CIF] information file
<u>R</u> efine <u>E</u> dit File <u>C</u> ancel

Figure 9.20 - The Refine menu

After a few seconds, a Refine window will open with output results for the calculation.

Max.sl wR2 = GooF = Mean s	hift = 0.0 0.2818 = S = shift/esd	32 A for C before cyc 2.496; F = 0.686	11 Ma e 3 for estrainec Maximur	x. dU =-0 2445 da I GooF = n = 2.4	1.008 for C ta and 5 2.496 56 for y	2 7 / for C11	57 param 0 restrai at 11:	eters nts 50:43		1
Max. sl wR2 = GooF = Mean s Max. sl wR2 = GooF = R1 = (wR2 =	hift = 0.0 0.2787 = S = shift/esd hift = 0.0 0.2783 = S = 0.0879 fc 0.2783,	15 A for C before cyc 2.473; F = 0.180 04 A for O before cyc 2.470; F r 2315 F GooF = S	11 Ma e 4 for estrained Maximul 2 Max e 5 for estrained > 4 sig(F = 2.470	x. dU =-(2445 da I GooF = m = -0.8 . dU = 0. 2445 da I GooF = o) and (), Restra	1.001 for C 2.473 54 for y 000 for C4 ta and : 2.470 0.0918 for ined GooF	2 for 02 for all 2	57 paramı 0 restrai at 11: 57 parame 0 restrai 2445 data 2.470 for	eters nts 50:44 ters nts all data		
R1 = (Highes Deepe	0.0932 fo t peak st hole	r 1440 u 1.18 at 0 -0.75 at (ique refl .2500 0.3 .1606 0.	ong - inv ections af 3358 0.2 2830 0.2	ert and rep ter mergin 419 [0.5 403 [0.5	ig for 1 A fi 51 A f	Fourier rom S1] rom S1]	**		
+++++ + ylid	+++++ _res75	++++++ finishe	+++++ at 11:50	+++++ :44 Tot	+++++ al CPU tim	++++ e:	++++++ 0.3 secs	++++++ +	+++++	+++++
+++++ •	+++++	++++++	+++++	+++++	+++++	++++	++++++	++++++	+++++	+++++
	04	1						Γ	Duinh	1

Figure 9.21 - Isotropic refinement output

In the output displayed above, note that the R1 value is 0.08. This is typical for a preliminary isotropic refinement for an organic molecule with data to a resolution of 0.75 and no hydrogen atoms included. The refinement program also indicates that the model needs to be inverted to get the correct absolute structure.

NOTE: This indication of the absolute configuration is usually reliable, but all assignments of absolute structure should be confirmed later in the refinement process by including TWIN and BASF cards in the instruction input.

2. Click on OK to return to XShell. The molecule display will refresh with the results of the least-squares calculations.



Figure 9.22 - The model after isotropic refinement

The peaks in the diagram represent difference in the electron density between the refinement model and the experimental electron density as defined by the measured data. Many of these difference peaks are near the sulfur and oxygen atoms. Refinement of all atoms anisotropically should correct the model. In the refinement box click on Invert Structure and Refine All Non-H Atoms Anisotropically as in Figure 9.23.

Number of residual (Q)peaks	20
Sigma Cutoff 🛛 🗖 n Sigma	
Resolution Cutoff 🦵 2 Theta	
Invert struct	ure.
Late-stage refinements options	
Refine all non-H atoms anisotr	opically

Figure 9.23 - Inverting and choosing anisotropic refinement

3. Click on the Refine button to launch the least-squares refinement program. The output window will open and a summary of the results of individual cycles of refinement will appear.

GooF = S = 1.6 Mean shift/esd = Max. shift = 0.010	27; Restrained Goo 1.801 Maximum = A for C5 Max. dU	F = 1.627 for -15.000 for U13 S1 = 0.004 for C5	0 restraints at 11:52:34	
wR2 = 0.1427 be GooF = S = 1.2 Mean shift/esd = Max. shift = 0.005	fore cycle 3 for 244 (57) Restrained Goo 0.503 Maximum = A for C5 Max.dU	5 data and 127 / F = 1.257 for -2.462 for U13 S1 = 0.002 for C6	127 parameters 0 restraints at 11:52:34	
wR2 = 0.1408 be GooF = S = 1.2 Mean shift/esd = Max. shift = 0.001 wR2 = 0.1407 be GooF = S = 1.2 R1 = 0.0468 for wR2 = 0.1407, G R1 = 0.0505 for Highest peak 0. Deepest hole -0.	fore cycle 4 for 244 (36; Restrained Goc 0.095 Maximum = A for C7 Max. dU fore cycle 5 for 244 (33; Restrained Goc 2315 Fo > 4siq(Fo) a iooF = S = 1.233, R 1440 unique reflection 49 at 0.8141 0.0816 24 at 0.6815 0.3980	5 data and 127 / F = 1.236 for 0.561 for U33 C6 0.000 for C5 5 data and 2 / F = 1.233 for nd 0.0489 for all estrained GooF = 10.2288 [0.95 A f 0.2288 [0.95 A f 0.1149 [1.21 A	127 parameters 0 restraints at 11:52:34 127 parameters 0 restraints 2445 data 1.233 for all data -Fourier rom C10] from C3]	
++++++++++++++++++++++++++++++++++++++	+++++++++++++ finished at 11:52:34	++++++++++++++++++++++++++++++++++++++	++++++++++++++ 0.5 secs +	+++++++++
+++++++++++++++++++++++++++++++++++++++	+++++++++++++++++++++++++++++++++++++++	+++++++++++++++++++++++++++++++++++++++	+++++++++++++++++++++++++++++++++++++++	.+++++++++

Figure 9.24 - Refine023

The value for R1, 0.047, is typical for an initial anisotropic refinement with no hydrogen atoms included. Since the mean shift/esd is greater than 0.1, the model is still changing. Since the highest peak in the difference map has a height of 0.49 and is 0.5A from C10, there are certainly some hydrogen atoms to be included.

Examination of the difference peaks in the resulting difference electron density map shows that the top difference peaks all correspond to hydrogen atoms.



Figure 9.25 - Difference peaks correspond to expected hydrogen atom positions

9.3.4 Look at Atomic Displacement Parameters (Thermal Ellipsoids)

It is generally good practice to examine the displacement parameters at this point in structure determination.

1. Remove difference peaks with the slider tool.

Resid	ual (Diff	ference Fou	rier Q Peak:	s) —	
12	1.1	1.1	1.1	1.0	
0		No. of QF	eaks: O		5

Figure 9.26 - Removing all Q-peaks with the slider bar

2. Right-click on the background and select Thermal Ellipsoids.



Figure 9.27 - Selecting Thermal Ellipsoids



Figure 9.28 - Refine2015

Alternatively, a list of Ueq's (equivalent isotropic displacement parameters) can be generated by requesting information on all of the atoms. Examining these values for outliers is good practice.

3. Right-click on the background and click on Information on All Atoms.

Eind Duplicate Labels	
Information on All Atom	าร
Add Hydrogen atoms	•

Figure 9.29 - refine 2005

This produces the table in Figure 9.30.

	LABEL	PEAK HEIGHT	TYPE	х	Y	Z	HYBRID	CHARGE	UEQ
1	51	0.0000	s	0.8097	0.1815	0.2405	sp3	0	0.0353467
2	01	0.0000	0	0.3345	0.3026	0.1765	sp3	0	0.04722
3	02	0.0000	0	0.8432	0.890	0.1287	sp3	0	0.0488033
4	C8	0.0000	c	0.3591	0.1141	0.827	sp3	0	0.03482
5	C6	0.0000	с	0.1496	0.502	0.228	sp3	0	0.0549167
6	C3	0.0000	С	0.6896	0.8	0.1251	sp3	0	0.03449
7	⊂1	0.0000	С	0.4329	0.1949	0.1500	sp3	0	0.0335967
8	C9	0.0000	с	0.5093	0.24	0.679	sp3	0	0.0371967
9	C2	0.0000	C	0.6354	0.1245	0.1726	sp3	0	0.0332767
10	C7	0.0000	с	0.1756	0.1410	0.377	sp3	0	0.0462133
11	C10	0.0000	С	0.8345	0.3779	0.2289	sp3	0	0.04692
12	C5	0.0000	c	0.2978	0.620	0.378	sp3	0	0.0566333
13	C4	0.0000	c	0.4862	0.891	0.76	sp3	0	0.0486733
14	⊂11	0.0000	C	0.6496	0.1778	0.3230	sp3	0	0.0572667

Figure 9.30 - List of parameters with Ueq to the right

Examining the values here is difficult because the atoms are not in order.

9.3.5 Sort Atoms

1. Right-click on the background and click on "Sort..." to sort the atoms into a sensible order.



Figure 9.31 - Choose the Sort option

A box will appear to the right of the molecule display. Since S1, O1 and O2 are in a reasonable order, only the carbon atoms need to be ordered.

SORT ATOMS						
Sort Bin	Atom List					
Sort(Alpha-Numeric) Sort(Numeric-Alpha) Insert Selected After- Insert <u>A</u> ll After>	<top list="" of=""> 51 01 02 C8 C6 C3 C1 C2 C9 C7 C10 C5 C4 C11 C1 C4 C11</top>					
<move s<="" td=""><td colspan="6"><move atoms="" bin<="" selected="" sort="" td="" to=""></move></td></move>	<move atoms="" bin<="" selected="" sort="" td="" to=""></move>					
	e Ali Atoms To Sort Billy					
<u>o</u> k	⊆ancel					

Figure 9.32 - refine2016

NOTE: Generally, it is easier to sort and number atoms before adding hydrogen atoms.

2. Highlight the atoms to sorted (i.e., left-click and drag over the atoms) and move them to the sort bin (i.e., left-click on Move Selected Atoms to Sort Bin).



Figure 9.33 - refine2017

3. After the atoms are in the sort bin, click on Sort(Alpha-Numeric) to get them into a normal order. 4. Highlight O2 and click on Insert All After --- > as in Figure 9.34.

Sort Bin	Atom List
C1 C2 C3 C5 C6 C7 C7 C8 C9 C10 C11	<top list="" of=""> S1 O1 O2</top>
<u>Sort(Alpha-Numeric</u> Sort(<u>N</u> umeric-Alpha	:) a)
Insert Selected After	

Figure 9.34 - refine 2019

5. Click on OK to accept the sorting.



Figure 9.35 - refine2011

The resulting list of atom information is easier to examine.

	LABEL	PEAK HEIGHT	TYPE	X	¥	2	HYBR3D	CHARGE	UEQ
1	51	0.0000	5	0.0097	0.1015	0.2405	sp3	0	0.0353467
2	01	0.0000	0	0.3345	0.3026	0.1765	sp3	0	0.04722
э	02	0.0000	0	0.0432	0.090	0.1287	sp3	0	0.0400033
4	CI	0.0000	<	0.4329	0.1949	0.1500	sp3	0	0.0335967
5	C2	0.0000	c	0.6354	0.1245	0.1726	sp3	0	0.0332767
6	C3	0.0000	c	0.6096	0.0	0.1251	sp3	0	0.03449
7	C4	0.0000	c	0.4062	0.091	0.76	sp3	0	0.0406733
0	CS	0.0000	c	0.2978	0.620	0.378	sp3	0	0.0566333
9	C6	0.0000	<	0.1496	0.502	0.228	sp3	0	0.0549167
10	07	0.0000	<	0.1756	0.1410	0.377	sp3	0	0.0462133
11	CB	0.0000	c	0.3991	0.1141	0.827	sp3	0	0.03402
12	C9	0.0000	c	0.5093	0.24	0.679	sp3	0	0.0371967
10	C10	0.0000	c	0.0345	0.3779	0.2209	sp3	0	0.04692
14	C11	0.0000	C	0.6496	0.1778	0.3230	sp3	0	0.0572667

Figure 9.36 - Sorted atoms

The Ueq are reasonable if there are:

- lower values for the sulfur atom and the five member ring
- higher values for the terminal atoms—methyls and carbonyls
- sensible numbers for the six member ring lower for C8 and C9 higher for C5 and C6 and in between for C4 and C7

9.3.6 Add Hydrogen Atoms

At this point, refine the hydrogen atoms either by renaming them using the labelling tool as before and then including them in the refinement.

or

Include the hydrogen atoms in fixed idealized positions riding on the atom to which they are attached.

In this manual we will illustrate the later fixed idealized method.

NOTE: In some cases (e.g., hydrogen atoms on hetero atoms or on geometrically strained carbon atoms), it is better to refine the hydrogen atom positions. A general rule is to refine if there is some doubt about the position of the hydrogen atom and there is different electron density present.

1. Using the slider tool, remove all of the hydrogen atoms.



Figure 9.37 - Retread model with difference peaks removed

2. Right-click in the background and the click on Hybridize All.



Figure 9.38 - Refine025

The colors of the atom labels will change to indicate the atom hybridization.

NOTE: If the atom hybridizations are hard to see, right-click on the background and open the Information on All Atoms window.





9 - 24



Figure 9.40 - Model with added Hydrogen atoms

3. The following warning will appear indicating that the hydrogen atoms have been provisionally added and that the final addition with naming will take place when the least-squares calculations are started. Click OK.



Figure 9.41 - Hydrogen atom warning

 Click on the Refine button to launch the least-squares refinement program. The output window will open and a summary of the results of individual cycles of refinement will appear.

GooF = S = 0.482; Mean shift/esd = 0.50	Restrained GooF = 6 Maximum = -2.50	0.482 for 2 for y C11	0 restraints at 11:53:17	
Max. shift = 0.011 A to mP2 = 0.0781 before	H11B Max.dU = 0	0.000 for C1	120 parameters	
GooF = S = 0.475;	Restrained GooF =	0.475 for	0 restraints	
Mean shift/esd = 0.03 Max. shift = 0.001 A fo	4 Maximum = 0.24 1 H11B Max.dU = 0	3 for U13 C4).000 for C5	at 11:53:17	
wR2 = 0.0781 before (ycle 4 for 2445 dati	a and 129/	129 parameters	
GooF = S = 0.475; Mean shift/and = 0.00	Restrained GooF =	0.475 for	0 restraints	
Max. shift = 0.001 A fo	H11B Max. dU = 0	0.000 for C5	at 11:55:17	
wR2 = 0.0781 before (ycle 5 for 2445 data	a and 2/1	29 parameters	
R1 = 0.0284 for 2315	Fo > $4\sin(E_0)$ and 0.	.0303 for all 19	445 data	
wR2 = 0.0781, GooF	= S = 0.475, Restrain	ned GooF = h	.475 for all data	
R1 = 0.0291 for 1440	unique reflections after	er merging for	Fourier	
Highest peak 0.25 at Deepest hole -0.16 a	0.7757 0.3500 0.20	34 [U.81 A fr 36 [1 29 A fr	om C2 J rom C6 J	
Deepest Hole 0.10 a	. 0.1720 0.4703 0.02	.50 [1.27 H II	011 00]	
+++++++++++++++++++++++++++++++++++++++	+++++++++++++++++++++++++++++++++++++++	+++++++++	+++++++++++++++++++++++++++++++++++++++	+++++++++++++++++++++++++++++++++++++++
+ yiid_res/5 tinis	ned at 11:53:17 Tota	i CPU time:	U.5 secs +	
+++++++++++++++++++++++++++++++++++++++	+++++++++++++++++++++++++++++++++++++++	++++++++++	+++++++++++++++++++++++++++++++++++++++	++++++++++

Figure 9.42 - Refinement results

The value for R1, 0.029, is typical for an anisotropic refinement with riding hydrogen atoms and a data set measured to 0.75Å. Since the mean shift/esd is less than 0.01, the model is not changing significantly. Since the highest peak and the deepest hole in the difference map are similar there are probably no missing atoms.

The resulting difference electron density map appears rather ugly. This is because the number of difference peaks to be displayed has been left at 20.

M86-E00078



Figure 9.43 - Difference peaks in a well refined model

9.3.7 Final Refinement for Publication

 For the final cycles of refinement reduce the number of peaks to be displayed to five and turn on Use Suggested Weights and Generate ACTA(CIF) Information File as shown in the next two figures.

NOTE: If the suggested weights vary significantly from 0.06 and 0.00, then change the values to 0.06 and 0.00.



Figure 9.44 - Refine032

ſ	Late-stage refinement options							
	Refine all non-H atoms anisotropically							
	Vise suggested weights							
2	Squared Term 0.058500							
	Linear Term 0.000000							
	Refine extinction parameter							
	Generate ACTA[CIF] information file							
į	<u>R</u> efine <u>E</u> dit File <u>C</u> ancel							

Figure 9.45 - Select weights and request ACTA output

 Click on Edit File, choose INS and click OK to open the instruction file for the XL. This allows you to add instructions that are not directly available in the GUI.



Figure 9.46 - Open the .ins file for editing

- 3. Check that the temperature card is correct.
 - 3.1 If torsion angles are of interest, add a CONF (confirmation).

TEMP 23 conf mpla 12 c1 c2 c3 c4 c5 c6 c7 c8 c9 s1 o1 o2 c10 c11 mpla 6 c4 c5 c6 c7 c8 c9 c1 c2 c3 o1 o2 s1 c10 c11 mpla 5 c8 c9 c1 c2 c3 c4 c5 c6 c7 o1 o2 s1 c10 c11 mpla 10 c1 c2 c3 c4 c5 c6 c7 c8 c9 s1 o1 o2 c10 c11

Figure 9.47 - Add instructions in the .ins file



Figure 9.48 - Difference peaks

- 4. Display the anisotropic atoms displacement parameters for the YLID.
- 5. If planarity and angles between planes are of interest, add mpla cards.

9.3.8 Generate an Atomic Displacement (Thermal Ellipsoid) Plot

- 1. Remove difference peaks with the slider tool.
- 2. Right-click on the background and select Thermal Ellipsoids.



Figure 9.49 - Thermal ellipsoids with large hydrogen atoms

- 3. If the hydrogen atoms in Figure 9.49 are large, change their size.
 - 3.1 Click on preferences in the Window Tool Bar and choose Atom Preferences.



Figure 9.50 - Atom preferences tool

At the top of the box that is opened there are three values that can be changed to change the size of atoms and the bonds to them.

ATOM PREFERENCES						
t Atom Radius	Bond Radius					
	1 PREFERENCES It Atom Radius					

Figure 9.51 - Default atom preferences

4. Change the hydrogen radius to 0.08 and the bond radius to 0.15 to make the hydrogen atoms smaller.

ATOM PREFERENCES							
Element	Atom Radio	us Bond Radius					
PeriodicTable H	0.08	0.15					

Figure 9.52 - Modified atom preferences



Figure 9.53 - Thermal ellipsoid plot with smaller hydrogen atoms

5. Position atom labels by right-clicking on the atom and choosing Position Label.

Delete Atom
Hide Atom
Labelling
<u>S</u> et Pivot Point
Hide <u>L</u> abel
Position Lobel
Change Part

Figure 9.54 - Position Label menu

NOTE: A message may appear that the font file needs to be defined "Please set your font directory for the first time for opengl rendering." If this happens, click on Preferences > OpenGL Preferences. Then click on the true type font For.OpenGL. In Windows browse to C:\bn\xshell\fonts\ or in Linux go to /usr/local/ bin/xshell/fonts. Choose a font.



Figure 9.55 - Positioned labels

 Convert this image into a high quality jpeg for inclusion into reports by clicking on render in the Windows Tool Bar and choosing Black and White to High Quality JPEG file.

Constrain Model <u>R</u> ender Preferences <u>H</u> e	ab '
	To Scree <u>n</u> Ctrl+N
Black and White	To Prin <u>t</u> er Ctrl+T
	JPG tighQuality JPEG File Ctrl+U

Figure 9.56 - Generate JPEG



Figure 9.57 - The final rendered thermal ellipsoid/atomic displacement parameter plot

Appendix A. SAINTPLUS

Occasionally it is necessary to integrate images outside of the APEX2 GUI. Usually this is necessary in order to import a matrix (p4p) file. SaintPlus executed from a command window is designed for this purpose.

- 1. Open a Command Prompt window.
- 2. Change directories to the folder of interest.
- 3. For Windows, enter the command bnrun c:\bn\src\plugins\saint\saintplus.pyc

(see Figure A.1). For Linux, enter the command bnrun /usr/local/bn/src/plugins/saint/ saintplus.pyc.

Command Prompt - bnrun c:\bn\src\plugins\saint\saintplus.pyc C:\struc\guest\ephe=lt>cd \ C:\>cd \struc\guest\ephe=lt\ C:\struc\guest\ephe=lt>bnrun c:\bn\src\plugins\saint\saintplus.pyc

Figure A.1 - Starting SAINTPLUS

After the command is executed, the SaintPlus window will open (see Figure A.2).

SaintPl	us				. 🗆 🗙
Eile Options	Utilities		~		
Space Group:	P1	-		Resolution Limit (Å): 1.700	_
Starting	1 Image Filename	Images	Matrix Filename	Output Filename	<u> </u>
2					
3					-
4					
5					
6					
7					
Refinement 0	ptions Integration Options		Find Runs Replace Matrix Files	Start Integration	

Figure A.2 - Initial integration window

This window is like the Saint window discussed previously, but with the following differences:

- Defaults are not determined by previous knowledge of the sample.
- There is a button to replace the matrix files.
- 4. Click Find Runs.
- 5. Browse to find the images to integrate.

🎬 Find Runs				? >	<
Find Runs in Directory:					
Image Base Name:					
	_		_		
Browse		OK		Cancel	

Figure A.3 - Find Runs tool view

6. Click on Replace Matrix Files and browse to find a p4p or spin file to use.



Figure A.4 - Find Matrix tool view

7. This defines the information on the runs, but the defaults for space group and resolution have not been updated. Click on space group and select the correct space group, in this case Pmmm.



				1 2(1)2(1)2(1)	
P1				Pmm2	
Triclinic	۲			Pmc2(1)	
Monoclinic	►	E_01_0	002	Pcc2	
Orthorhombic	•	P	►	Pma2	
Tetragonal	+	Ι	►	Pca2(1)	
Rhombohedral	۲	F	►	Pnc2	
Hexagonal	۲	С	►	Pmn2(1)	
Cubic	+			Pba2	
tions I Integration Or					
	Pnn2				
				Pmmm*	
				NE	

Figure A.5 - Space group tool

8. Double-click on the Resolution Limit and enter an appropriate value.

NOTE: If the starting values for SAINT need to be changed frequently, change the default by modifying the SAINT.ini file. Consult the SAINT manual 269-0175xx for further information **on Windows or Linux**.

9. Use the Refinement Options and Integration Options buttons to set up the Saint run. In

this case Generate Active Mask has been turned on and set to 0.7.

Calify Generate Mask:	
Fractional Lower Limit of Average Intensity: 0.700	
C Use Pre-Existing Mask: Active Mask File (*., am):	
	,

Figure A.6 - Active Mask tool

10. Click on Start Integration to start the calculations and open the SaintChart window for monitoring the results.

A - 4

Appendix B. Crystal Screening

crystal selection and mounting

3 - 2

Appendix C. RLATT

1. Use the Reciprocal Lattice tool to look at the reflections.



Figure C.1 - UserManual013.bmp

Index

DPS	SMART
HKL	Sparks indexing
Kaercher Strategy	XM
ProScale	XPREP
SADABS	
SAINT+	CurtI'll add to this list of keywords. If there are any you specifically want added, write them in.

A - 2