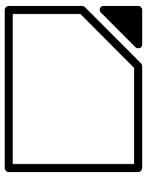

LibreOffice
The Document Foundation

Gromacs [Ligand – Complex System Setting Up]

Copyright

Contributors

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Feedback

Please direct any comments or suggestions about this document to:
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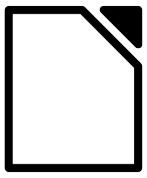
Contents



LibreOffice
The Document Foundation

在 *GROMACS* 中使用 *GAFF* 力场处理配体并建立任务

Type Optional Subtitle Here

背景和软件准备

这个教程是基于 John E. Kerrigan[1] 和 Justin Lemkul[2] 的教程编写的。用过 Amber 的朋友应该都知道，GAFF 力场在处理小分子配体（非标准残基）时效果不错。幸运的是，我们能够在 GROMACS 中使用 GAFF 力场，但是需要通过一定的脚本来实现。

在开始之前，我们需要以下软件：

1) GROMACS;
2) AmberTools 包括 Antechamber, tleap 等;
3) PyMol 等可视化软件;
4) Swiss-Pdbviewer 用于补全 PDB 文件;
5) Acpype 脚本;
6) Python 和 Linux 系统;
7) Chimera，用于处理小分子（加氢，加电荷）

坐标文件的获得

我们以 Justin Lemkul 的教程中的 T4 溶菌酶体系（PDB：3HTB）为例开始我们的教程。从 PDB 数据库中得到原始 PDB 文件后我们首先应该检查文件中是否有缺失的原子、残基，如果有，可以用 Swiss-Pdbviewer 补全要从中提取配体和蛋白质。

```
grep 'ATOM ' 3HTB > protein2.pdb
grep 'JZ4 A' 3HTB > ligand.pdb
```

之后，我们使用 AMBER99SB 力场对蛋白质本身进行处理，并生成坐标和拓扑文件。同时我们在此指定使用 TIP3P 的水分子模型。

```
pdb2gmx -ff amber99sb -f protein2.pdb -o trp.pdb -p topol.top -water tip3p
-ignh
```

用 Chimera 预处理小分子配体

上一步中，我们得到了小分子配体的 PDB 文件，我们用 Chimera 打开，得到如下界面。

1 http://cinjweb.umdnj.edu/~kerrigje/pdf_files/trp_drug_tutor.pdf

2 http://www.bevanlab.biochem.vt.edu/Pages/Personal/justin/gmx-tutorials/complex/

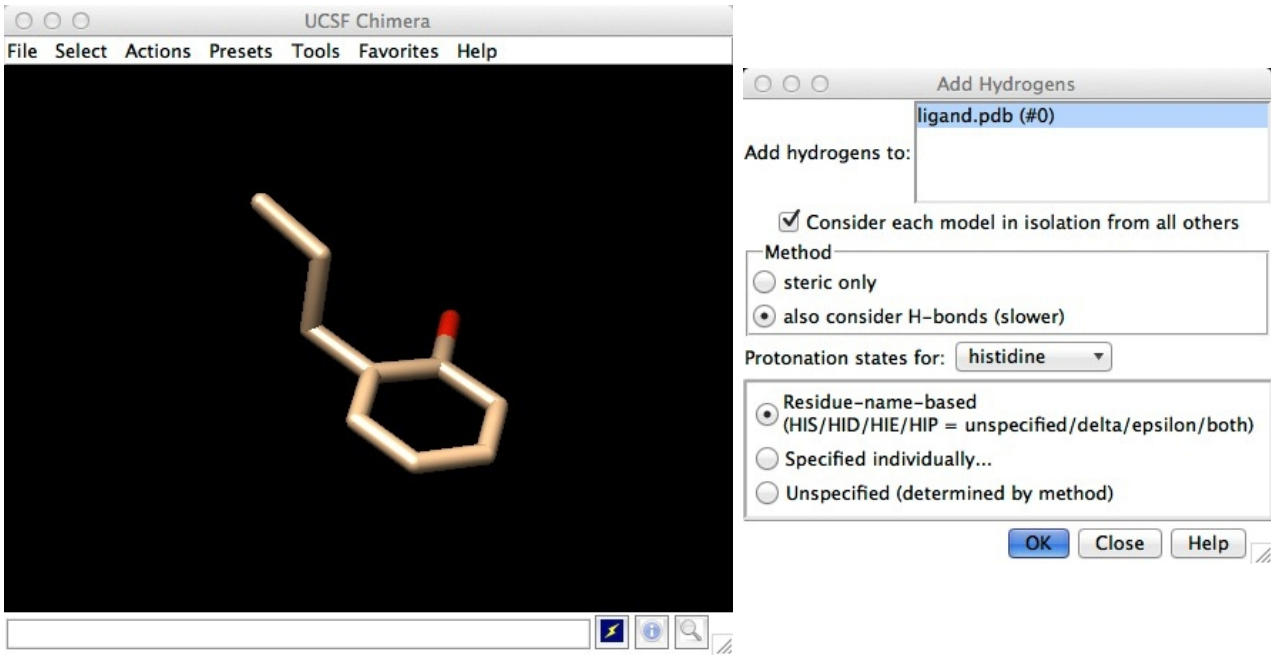


Figure 1: Chimera 基本界面

点击 Tools->Structural Editing->AddH 进行加氢，按照图 1 默认的选项即可，点击 OK，可以发现多了几个氢原子。
接着给配体分子加电荷，选择 AM1-BCC 的电荷计算方法，同时事先应该计算好配体的静电荷，然后在这一部指派给它，在此我们选择+0。

最后，我们把配体保存成 mol2 格式的文件。

ACPYPE 处理配体文件

ACPYPE 是 Alan Wilter SOUSA DA SILVA 等人编写的可以调用 Antechamber 对配体进行 GAFF 力场处理并转换成 GROMACS 格式的 python 脚本，可以在 https://code.google.com/p/acpype/下载得到，使用方法也非常的简单。下载后，可以使用

```
chmod +x acpype.py
```

确保脚本可以直接执行。下面我们用该脚本处理上一步得到的配体的 mol2 文件。

```
./acpype -i ligand.mol2
```

如果正常执行结束，会生成一个名为 ligand.acpype 的文件夹，其中有一系列生成的文件。如脚本执行果未正常结束，应该确保 antechamber,tleap 等命令在系统上可以使用。

接着，我们把处理得到的配体的坐标文件与蛋白质的坐标文件合并，得到复合物的 PDB 文件

```
grep -h ATOM protein2.pdb ligand.acpype/ligand_NEW.pdb > complex.pdb
```

同时把生成的 ITP 拓扑文件复制出来到当前目录

```
cp ligand.acpype/ligand_GMX.itp ligand.itp
```

此外，需要注意的是，当前 ligand.itp 中 meloculetype 部分配体的名字还要改成配体的名称 JZ4，这个名称需要和在稍后在 topol.top 中添加的名称一致。

因此，下面需要在 topol.top 文件中添加关于配体的信息。

首先是在开头几行的

```
#include "amber99sb.ff/forcefield.itp"
```

下方添加：

```
#include “ligand.itp”
```

然后在最后几行的[molecules]里面添加：

```
JZ4   1
```

至此，我们将配体的拓扑和结构信息与蛋白质的进行了整合。

定义元胞、加溶剂

首先定义一个十二面体的元胞

cubic

```
editconf -f complex.pdb -o newbox.gro -bt dodecahedron -d 1.0
```

-c

然后在元胞内加入水溶剂，并得到坐标文件

```
genbox -cp newbox.gro -cs spc216.gro -p topol.top -o solv.gro
```

加离子

在加离子之前，我们需要知道当前体系的带电情况，使用如下命令

```
grep qtot topol.top
```

返回结果的最后一行即为当前的静电荷数，得到 qtot 6,即带 6 个正电荷，在加离子之前，我们需要使用 grompp 程序得到 Run Input File，其后缀为 tpr。但此外，还需要一个 MD Parameters 参数文件，这里我们使用进行能量最小化的参数文件 em.mdp。

em.mdp

```
; LINES STARTING WITH ';' ARE COMMENTS
title    = Minimization ; Title of run

; Parameters describing what to do, when to stop and what to save
integrator  = steep     ; Algorithm (steep = steepest descent
minimization)
emtol    = 1000.0    ; Stop minimization when the maximum force <
10.0 kJ/mol
emstep      = 0.01      ; Energy step size
nsteps      = 50000     ; Maximum number of (minimization) steps to
perform
energygrps  = system ; Which energy group(s) to write to disk

; Parameters describing how to find the neighbors of each atom and
how to calculate the interactions
nstlist     = 1          ; Frequency to update the neighbor list
and long range forces
ns_type     = grid      ; Method to determine neighbor list
(simple, grid)
rlist    = 1.0    ; Cut-off for making neighbor list (short range
forces)
coulombtype = PME    ; Treatment of long range electrostatic
interactions
rcoulomb = 1.0    ; long range electrostatic cut-off
rvdw     = 1.0    ; long range Van der Waals cut-off
pbc      = xyz    ; Periodic Boundary Conditions (yes/no)
```

执行

```
grompp -f em.mdp -c solv.gro -p topol.top -o ions.tpr
```

接着进行加离子 -neutral

```
genion -s ions.tpr -o solv_ions.gro -p topol.top -pname NA -nnname CL -nn 6
```

期间程序会询问你将什么替换成离子，选择 SOL 溶剂分子即可。正常运行结束后，查看你的 topol.top 文件，可以看到最后几行的[molecules]中有 Protein_chain_A, JZ4, SOL 和 CL。

能量最小化

在每次执行 MD 任务之前，我们都需要先使用 grompp 程序生成 Run Input File。

```
grompp -f em_real.mdp -c solv_ions.gro -p topol.top -o em.tpr
```

其中此次最小化的参数文件如下。
最后执行最小化任务。

```
mdrun -v -deffnm em
```

em_real.mdp

```
; LINES STARTING WITH ';' ARE COMMENTS
title    = Minimization ; Title of run

; Parameters describing what to do, when to stop and what to save
integrator  = steep     ; Algorithm (steep = steepest descent
minimization)
emtol    = 1000.0    ; Stop minimization when the maximum force <
10.0 kJ/mol
emstep      = 0.01      ; Energy step size
nsteps      = 50000     ; Maximum number of (minimization) steps to
perform
energygrps  = Protein JZ4  ; Which energy group(s) to write to
disk

; Parameters describing how to find the neighbors of each atom and
how to calculate the interactions
nstlist     = 1          ; Frequency to update the neighbor list
and long range forces
ns_type     = grid      ; Method to determine neighbor list
(simple, grid)
rlist    = 1.0    ; Cut-off for making neighbor list (short range
forces)
coulombtype = PME    ; Treatment of long range electrostatic
interactions
rcoulomb = 1.0    ; long range electrostatic cut-off
rvdw     = 1.0    ; long range Van der Waals cut-off
pbc          = xyz      ; Periodic Boundary Conditions (yes/no)
```

其中 mdrun 命令的-v 代表 verbose，会输出较为详细的运行信息。-deffnm 代表此次任务输入和输出文件的前缀名称。如果正常结束，我们可以得到 em.gro,em.trr 等一系列输出文件。

如果此时你想查看一下最小化后配体结构是否正常（没有出现断键等异常情况），可以使用如下命令导出结果的坐标 PDB 文件并使用 PyMol 查看。

```
editconf -f em.gro -o test_em.pdb
```

同时，如果你想查看势能的变化过程，使用 g_energy 程序可以做到。

```
g_energy -f em.edr -o em_graph.xvg
```

并在选择 Potential – 11 后，输入 0，按回车结束。得到的 em_graph.xvg 文件是 xmgrace 画图软件专用的文本文件格式，但是其他软件也可以读取。这里我们用 Mathematica 读取并绘图。

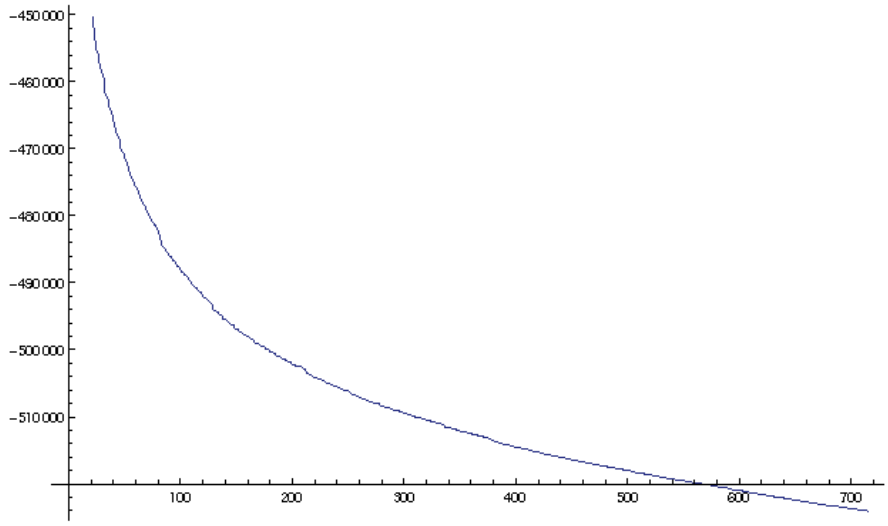


Figure 2: 能量极小化中势能随步数的变化情况

平衡

蛋白-配体复合物的平衡与单纯的蛋白的平衡类似，但是有不同：

- 配体上需要施加位置限制力(Position Restraints)
- 温度耦合的组合需要额外考虑处理

限制配体的位置

ACPYPE 脚本运行时，并没有生成配体的位置限制参数文件 itp，此时我们使用 GROMACS 的 genrestr 程序

```
genrestr -f ligand.acpype/ligand_GMX.gro -o pores_jz4.itp -fc 1000 1000 1000
```

Do not use the toolbar icons to apply numbering or bullets because you get different results than if you use the paragraph styles for lists.

Lists use paragraph styles that have been defined to include a numbering (list) style.

Four principal categories of lists are used in our books. Use the type that best suits the purpose:

- Procedures or instructions where steps should be done in a specific order should use numbered lists with Arabic numbers.
- For items set out in a list for easy reference, but where the sequence is not important (as in this list), use a bullet list.
- Simple lists are similar to bullet lists, but without the bullets. Use simple lists only when all of the list items are no longer than one line.
- Definition lists are used mainly for short explanations of fields or options on dialogs. They are in two parts: the “definition term” and the “definition” itself.

Examples and descriptions of each type of list follow.

Bullet lists

The template makes available two types of paragraph style families for bullet lists:

OOoList1 using circular bullet symbols (Unicode U+2022)

OOoList2 using dash bullet symbols (U+2013)

The *OOoList2* paragraph styles should only be used for second-level bullet lists where the first level is also a bullet list. To reach the second level, click the **Demote** icon on the Bullets and Numbering toolbar.

This paragraph is in *OOoTextBody_ListIntro* style. Same as *OOoTextBody* but includes a “keep with next paragraph” parameter. It also has less space before the first entry in a list.

- Sample bullet list (*OOoList 1 Start* style)
- Another item in the bullet list (*OOoList 1 Cont.*)

  And a continuation item (no bullet character), using *OOoList_TextBody_L1*
- Another item in the bullet list (*OOoList 1 Cont.*)
  - A sublist item (use *OOoList 2 Start*, then the **Demote** icon to style this as level 2)
  - Another sublist item
- Final item in main list (*OOoList 1 End*). Use this style for the very last ‘‘paragraph” entry in a first-level list so it can later be redefined (if we wish) to provide more vertical space preceding the following paragraph.

If you need to add a paragraph within the list and you want the indentation to be aligned with that of the list element, you need to use *OOoList_TextBody_L1* (for first-level list items) or *OOoList_TextBody_L2* for second-level list items.

Numbered lists

This template includes three Numbered paragraph style families that you can use to create simple or nested lists. Normally we should not nest procedural lists beyond three levels, and only use the third levels when really necessary.

The three numbering styles are defined as follows:

OOoNum 123: Arabic number followed by)

OOoNum abc: small case letter followed by)

OOoNum iii: Roman numeral followed by .

In general numbered lists should use Arabic numbers for the first level, small case letters for the second and Roman numerals for the third level.

Sample paragraph in *OOoTextBody_ListIntro* style introducing a numbered list.

1) Sample numbered list item (*OOoNum 123 Start* style)
2) Another item in the numbered list (*OOoNum 123 Cont.*)

   And a continuation item (no number), using *OOoList_TextBody_L1*
3) Another item in the numbered list (*OOoNum 123 Cont.*)
   a) A second-level list item ((using alphabetic “numbers”) —use **Demote** icon and *OOoNum abc Start* to style.
   b) Another second-level list item (*OOoNum abc Cont.*).
      i. Now we have a third-level nested list. It uses lowercase roman numerals—use **Demote** icon and *OOoNum iii Start* to style.
      ii. Another third-level list item
   c) A final second-level list item

4) Final item in first level (*OOoNum 1 End*).

Simple lists

Simple lists are similar to bullet lists, but without the bullets. The list items use the *OOoSimpleList* style.

Apples
Oranges
Pears
Plums
Bananas
Peaches

Definition lists(词条，术语，名词解释)

Definition lists are typically used for describing options on a dialog box. This list type can be used instead of third-level or fourth-level subheads, especially if you do not want any such subheads to appear in an automatically generated ToC.

Example of definition term
Example of a definition. All lines are indented from the left.

Another definition term
Here is the definition of this term: xxxxxxxxx xxxxxxxxx xxxxxxxx xxxxxxxxxx

Specific uses of lists

The following list is an example that uses hierarchical items in a list definition. Note that the separator (see highlights) uses an en-dash with a normal space on either side.

Import – Ignore font settings
Select this option to have LibreOffice ignore all font settings when importing. The fonts that were defined in the HTML Page Style will be used.

Export
To optimize the HTML export, select a browser or HTML standard from the **Export** box. If **LibreOffice Writer** is selected, specific LibreOffice Writer instructions are exported.

Export – LibreOffice Basic
Enable this option to include LibreOffice Basic macros (scripts) when exporting to HTML format. You must activate this option *before* you create the LibreOffice Basic blah blah blah

These template instructions show many lists with run-in-line terms. For example:

OOoTextBody—used for most paragraphs of text other than headings and lists.

Note that this is a normal paragraph, in whatever paragraph style is proper in that location (for example, it may be within a larger list). The term is separated from the text using an em-dash with no spaces on either side. Use *OOoEmphasis* for the term.

Figures

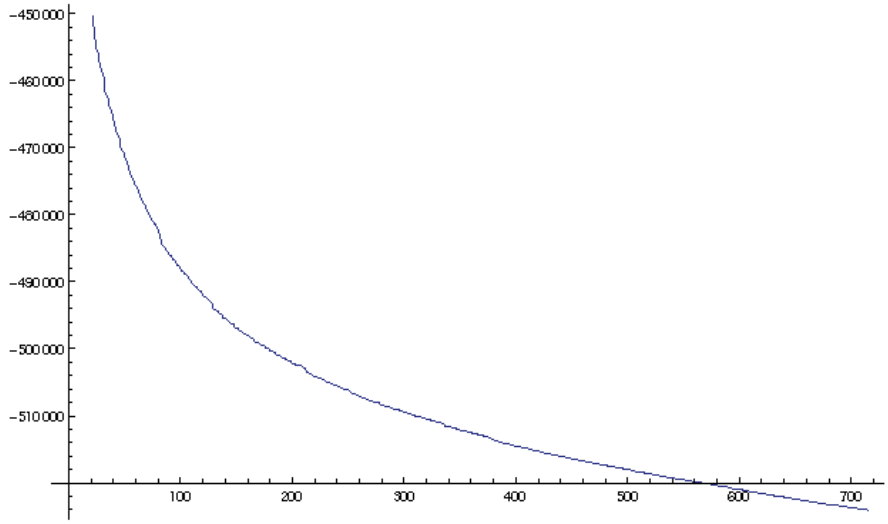
Illustrations (called Figures) and their caption use these styles:

OOoFigure—used for paragraphs containing figures. This type of paragraph does not contain any text, only the graphic. Its next style is *OOoTextBody*.

OOoFigureCaption—used for the captions to figures.

In case the illustration contains several elements you want to highlight (for example, descriptions of the icons on a toolbar such as that of Figure 3), you should insert a borderless table for the legend. Style with *OOoTableText(Caption)* for the description and *OOoTableText(CaptionNumber)* for the sequential number.

In this template the font size of these two styles is fixed to 10pt, therefore it is recommended to use a 0.75cm column width for the numbers column while the description columns should have roughly the same width.



1 Open Styles and Formatting Window	**5** Align Right	**10** Numbering On/Off
2 Apply Style	**6** Justified	**11** Bullets On/Off
3 Align Left	**7** Line Spacing: 1	**12** Decrease Indent
4 Centered	**8** Line Spacing: 1.5	**13** Increase Indent
	9 Line Spacing: 2	**14** Paragraph format dialog

Figure 3: Formatting toolbar icons description in tabular format

Inserting figures (images)

We use two types of illustrations (called Figures) in our books: with captions or without captions. Usually, only small, closely cropped illustrations (and pictures of icons or other controls) have no caption.

Most images should go in a separate paragraph containing no text. Small images are often best placed in a table beside the text. Icon images can either go in a table next to text, or be anchored as a character within the text. Avoid inserting “floating” images, as these often become displaced from their intended location.

To insert an image, such as a screen capture, in its own paragraph:

1) Place the cursor at the end of the paragraph that will be above the image. Press *Enter* once to create a blank paragraph.

2) With the cursor in the blank paragraph, apply the *OOoFigure* style to the paragraph.
3) With the cursor still in the blank paragraph, choose **Insert > Picture > From File**.
4) Select the desired image.
5) Click **Open**. The image is placed in the document.
6) Right-click the image and choose **Anchor > As Character**.
7) If the image needs a caption, right-click and choose **Caption** from the pop-up (context) menu.
8) Choose **Figure** as the *Category*. Type the caption's text. Click **OK** to save.
9) Apply the *OOoFigureCaption* style to the caption.
10) Select the image (not the frame around the image+caption), right-click and choose **Anchor > As Character**. (When you created the caption, the anchor in step 6 became the anchor for the frame around the image+caption.)

Figure 4 shows an example of a figure with a caption.

January	February	March
April	May	June
July	August	September
October	November	December

Figure 4: This is a figure caption.

Caution

Do not anchor graphics **To Page**. This works fine in standalone chapters, but when the chapter becomes part of a master document (for example, when compiling a full book), anything anchored To Page disappears.

If you do use a floating image, go to the *Type* page of the Picture dialog box and make sure that the **Follow text flow** option is *not* selected in the *Position* section; this provides better compatibility with MS Word.

Footnotes

Here[3] is an example of a footnote, whose text is displayed at the bottom of the page where the footnote was inserted. The numbering restarts at 1 for each chapter. Its paragraph style is *OOoFootnote*.

Tables

OOoTableCaption—used for captions to tables.

OOoTableHeader—used for table headers.

OOoTableText—used for table text.

Lists in tables

As the font size of the *OOoTableText* paragraph style is smaller than the *OOoTextBody* style, in order to create a list in a table you need to use one of the following two families of paragraph styles: for bullet list use *OOoListTNC*, while for numbered lists use *OOoNumberedTNC*.

3 Any footnotes will appear at the bottom of the page. Notice the indenting of the second and subsequent lines of the footnote (*OOoFootnote*).

As for lists in the main text body, the paragraph introducing the lists has to be formatted with the special paragraph style which in this case is *OOoTableText_listIntro*.

Use the above styles whenever you enter a list in a table, such as in a tip, note or caution.

Note that, given the numbered lists in a table can only use Arabic numbers and bullet lists only the round symbol.

Table 1: Example of numbered and bullet lists in a table

Example of list styles usage in a table	
Numbered list	**Bullet list**
The introductory paragraph style is *OOoTableText_listIntro*. 1) The first element of the list uses the *OOoNumberedTNC Start* style. 5) The second and subsequent elements of the numbered list use the *OOoNumberedTNC Cont* style. 2) The last element of the numbered list uses the *OOoNumberedTNC End* style.	The introductory paragraph style is *OOoTableText_listIntro*. • The first element of the bulleted list uses the *OOoListTNC Start* style. • The second and subsequent elements of the Bulleted list use the *OOoNumberedTNC Cont* style. • The last element of the Bulleted list uses the *OOoListTNC End* style.

A sample table

Below is an example table, with all the spacing set to our standard. You can copy and paste it into your documents, or copy it into an AutoText, or create a Table AutoFormat. Change the width of the columns as needed.

Table 2: This is a table caption.

Column 1	***Column 2***

Cross-referencing

It is possible to insert a cross-reference (also known as an xref or x-ref) to a heading, a figure, a table, a bookmark, a footnote, or a numbered item in a list.

For example, to insert a cross reference to the name and page of this section proceed as follows:

1) Press *Ctrl+F2* to open the Fields dialog box.
2) Go to the **Cross-References** page and select:
   a) **Headings** from the *Type* list
   b) The heading called **Cross Referencing** in the *Selection* list
   c) **Reference** in the *Insert Reference to* list
3) Click **Insert**.
4) Type the text **on page** and then repeat steps 2 and 3, taking care to select **Page** in substep 2c.

Inserting frames

For floating frames, make sure that the **Follow text flow** option is *not* selected in the *Position* section of the *Type* page of the Frame dialog box; this provides better compatibility with MS Word.

On the *Wrap* page of the Frame dialog box, use a vertical spacing of 9, 18, or 36 points, depending upon what follows the frame. This spacing is set in the *Bottom* field in the *Spacing* subsection. Usually, let the vertical spacing of the preceding paragraph style determine the *Top* vertical spacing (use the 0pt default). Select the **None** option in the *Settings* subsection for typical situations.

Starting a new chapter from the template

The LibreOffice User Guide chapter template is a template file (`*.ott`). To use it:

1) Download the template from the English Content > Documentation > Resources space on the LibreOffice Alfresco site.
2) Store this file anywhere you like on your computer.
3) Import the template into “My Templates” using the Template Management window (reached through **File > Templates > Organize**).
4) Start a new chapter from the template using **File > New > Templates and Documents**.
5) On the title page:
   - Replace the **n** after the word **Chapter** with the chapter number. Be sure to keep the two blank spaces following the n; these are required for correct spacing of the chapter number and title when the chapter becomes part of a book.
   - Replace the words **Type Chapter Name Here** with the name of the chapter. Use headline-style capitalization.
   - Replace the words **Type optional subtitle** here with the subtitle of the chapter, or delete this line if the chapter has no subtitle. Use sentence-style capitalization for the subtitle.
   - Be sure to keep the blank paragraph below the subtitle. This paragraph has a style of *OOoPageBreak* and is required for correct pagination when the chapter becomes part of a book.
6) Go to **File > Properties**. On the *Description* page, change the title from the chapter template’s filename to the title—not the filename—of the chapter.
7) On the first page of text, be sure that the first heading is the first paragraph on the page, as in this template. The style of the first heading paragraph in this template has been modified to have a page-break-before attribute (on the Text Flow tab of the Paragraph dialog) that changes the page style from *OOoFrontMatter* to *OOoPageStyle*. To ensure that this attribute is retained, it’s best to replace only the words “背景和软件准备” and not delete the paragraph marker.

Note	See the document titled *Producing LibreOffice User Guides* for instructions on updating the chapter template from the website, and updating a chapter from a changed template.

Updating the table of contents

The table of contents is already formatted and generated.

Do not forget to update the existing table of contents when your document is complete.

1) Place the cursor in the table (the gray field under the Contents heading on the third page).
2) Right-click and choose **Update Index/Table** from the pop-up menu.

Another way to update the ToC is to use the Tool menu (**Tools > Update > Update All**). This method also updates all indexes, tables, and fields. When updating the ToC, it is advisable to turn off edit-change tracking because if any heading entries in the ToC were edited, the edit-tracking could be displayed in the ToC for some edited entries. If that occurs, just turn off the display of edit-change tracking and update again.

Using tips, notes, and cautions

Below are samples of the tips, notes, and cautions for the guides. You can copy and paste these or use the AutoText feature to have LibreOffice remember them for you. These are 1-row, 2-column tables that do not break across pages. Both columns are preset to vertically center their contents.

Tip	A tip describes practical but nonessential information that does not fit into the text flow.

Note	A note contains information related to the text. It might be an explanation, a comment, or a statement meant to catch attention.

Caution 	Use cautions for operations that could result in data loss.

AutoText

Predefined samples of tables and figures

Samples of the Tip, Note, and Caution tables in the previous section can be easily inserted into a document by employing AutoText. After they are inserted, you can replace their content with your own content. To set this up, first install the dedicated `*.bau` AutoText category file onto your computer. For a US English setup for Windows, the default folder path for a BAU file is: `C:\Program Files\LibreOffice 3\Basis\share\autotext\en-US`.
You can change this path for other folder locations, if you so choose. See the AutoText instructions in the online LibreOffice Help (*F1*) for advanced AutoText topics, or you can conduct a web search for discovering other fine points.

AutoText entries for a figure (along with its caption) and for other tables (with captions) are also included in the BAU file for this template: LO_3_4_chapter_template.bau.

Using AutoText

As mentioned above, AutoText can be employed by authors or editors to insert preformatted sample tables and figures in your document as a timesaving tool—as placeholders to replace with your own content.

Be sure to select your desired AutoText entries from the appropriate category in the AutoText dialog box: **Edit > AutoText** (*Ctrl+F3*). Select the category—`LO_3_4_chapter_template`, in this case —and then select the desired AutoText entry and press the **Insert** button. Check the **Show**

preview check box in order to see a very rough display of the AutoText entries. Afterward, you will substitute your own text or graphics, replacing the contents of the preformatted placeholders.

Note	Creating an AutoText entry of a graphic for the `*.bau` category file by the template designer requires that the picture or anchored frame be anchored as a character, and it must be followed by one or more text characters before being selected as an AutoText entry. For the sample Figure in this template guide, a blank space character was used for the text following the sample graphic. So, after inserting an AutoText figure into your document, you can simply delete that extra space, if you choose. You can also change the anchoring for an AutoText-imported figure afterward to something other than **As character**, in case you desire a different anchoring system.

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