NCI Practical session

Content

NOTE: This guide has been conceived in a progressive manner. You should go through it as it is written, since it is assumed that you already know how to do and interpret the examples given in previous sections.

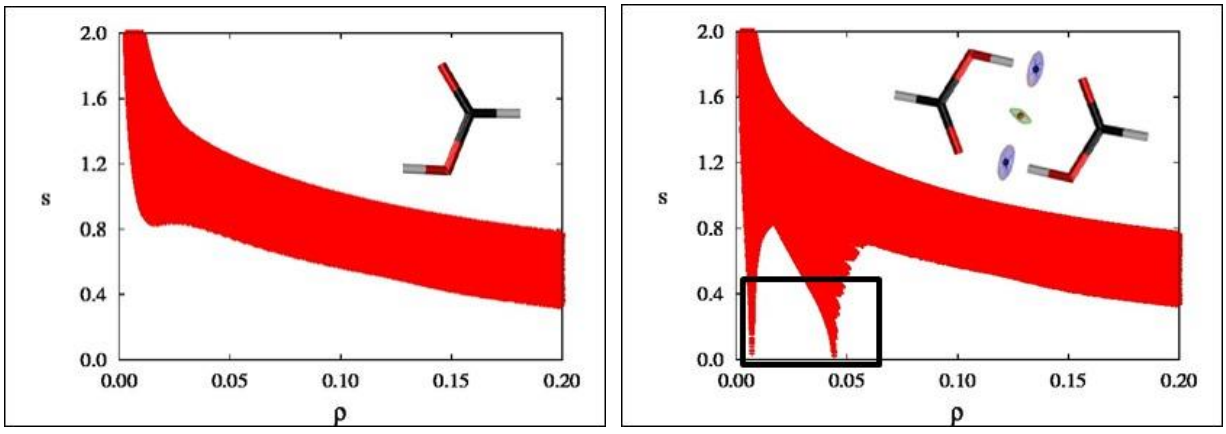
Theoretical background

The reduced density gradient

NCI (Non-Covalent Interactions) is a visualization index based on the density and its derivatives. It enables identification of non-covalent interactions. It is based on the peaks that appear in the reduced density gradient (RDG) at low densities. The reduced density gradient is given by:

$$s(r) = c\frac{|\nabla\rho(r)|}{\rho(r)^{4/3}}$$

where *c* is a constant. RDG enables to reveal the non-covalent interactions. When we plot the RDG as a function of the density across a molecule, we see that the main difference between the monomer (Figure 1a) and dimer (Figure 1b) cases is the appearance of steep peaks at low density.



a) Formic acid b) Formic acid dimer

Figure 1. s(ρ) for formic a) acid and b) formic acid dimer. The 3D representation of the peaks at low density is given in the insets: they appear in the dimer for the non-covalent interactions.

When we search for the points in 3D space giving rise to these peaks, non-covalent regions clearly appear in the (supra)molecular complex (insets in Figures 1a and 1b). They appear for the dimer case (Figure 1b) and reveal the non-covalent interactions: the hydrogen bonds (in blue) and the van der Waals interactions (in green). The coloring has been carried out in terms of the electron density. As we can see in Figure 1b, two peaks appear, a sharp one at very low densities which corresponds to the van der Waals interaction (the green isosurface) and a wider one at greater densities which corresponds to the symmetric hydrogen bonds (in blue).

The second eigenvalue

ρ approx. (a.u.)	λ_2	Typical shape	Interaction type	Example
>0.015	$\lambda_2 < 0$	Lentil	Strongly attractive	hydrogen bonds
>0.015	$\lambda_2 > 0$	Compact	Strongly repulsive	steric clash
<0.015	$\lambda_2 \simeq 0$	Sheets	Very weak	van der Waals

The interactions revealed by NCI correspond to both favourable and unfavourable interactions. In order to differentiate between them, the sign of the second density Hessian eigenvalue times the density is implemented. This value is able to characterize the strength of the interaction by means of the density, and its curvature thanks to the sign of the second eigenvalue. Some examples are collected in Figure 2. It can be seen that the method is applicable to small molecules as well as inorganic complexes.

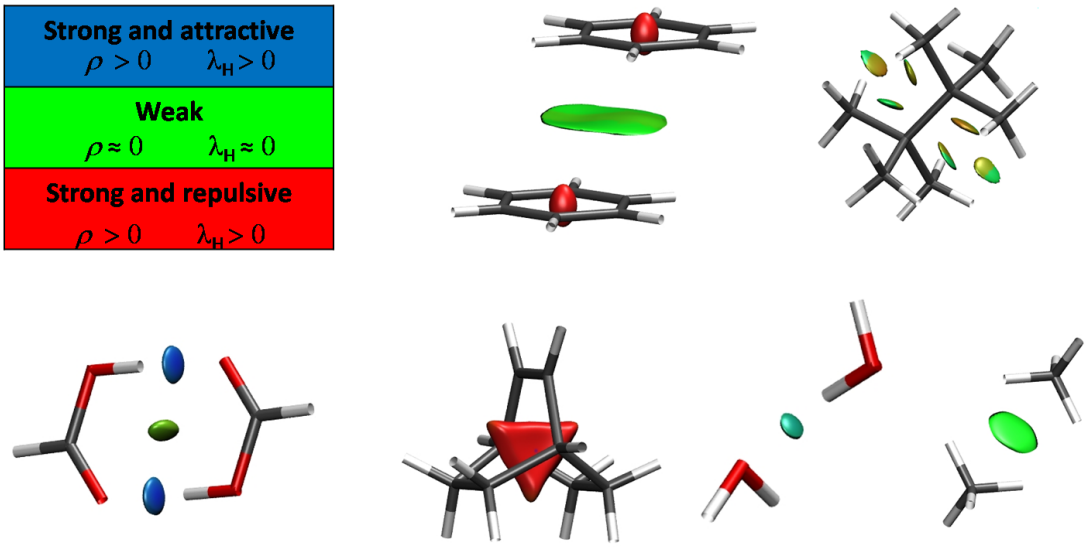


Figure 2. NCI isosurfaces colored according to sign(λ_2)ρ. From left to right: (top) benzene dimer, branched octane. (bottom) formic acid dimer, bicyclooctene, water dimer and methane dimer.

Promolecular densities

NCI (Non-Covalent Interactions) is applicable to promolecular densities, enabling the analysis of biomolecules (Figure 3). In this case, only the atomic coordinates are required as input. Both options (SCF and promolecular are implemented in the NCIPLOT).

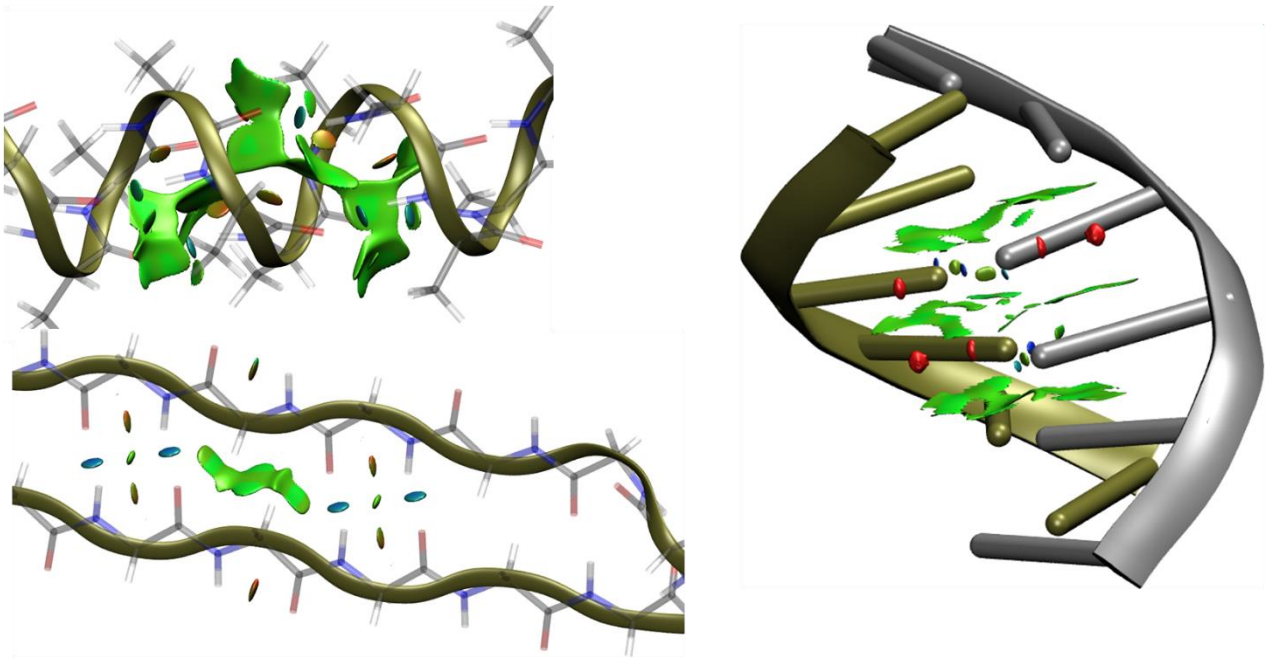


Figure 3. NCI isosurfaces from promolecular densities. Top left: α-helix, bottom left: β-sheet, right: DNA double helix.

Practical session guide

Preliminary step: the wavefunction file

Set out=wfn in the Route section and give the name of the wfn file at the end of the molecule specification.

The title first word (contiguous non blank characters) are used to form the file names in, it might be dangerous to use non alphanumerical characters.

Example: C_4H_4 tetrahedrane

```
# P HF/6-311++G(2df,2p) opt out=wfn
tetrahedrane
0,1
C -0.524831 0.524831 0.524831
C 0.524831 -0.524831 0.524831
C -0.524831 -0.524831 -0.524831
C 0.524831 0.524831 -0.524831
H -1.134794 1.134794 1.134794
H 1.134794 -1.134794 1.134794
H -1.134794 -1.134794 -1.134794
H 1.134794 1.134794 -1.134794

tetrahedrane.wfn
```

Obtaining NCI

Two options are given for Linux or Windows users. NCIPLOT allows to obtain all the features needed for the practical session. If you don't have access to a linux machine, you can install a public license for AIMALL which will allow you to carry out the first part of the TP.

Option 1: NCIPLOT (Linux)

You can download NCIPLOT4 here: https://www.lct.jussieu.fr/pagesperso/contrera/nci-programs.html

The manual is at the download repository and also here:
http://www.lct.jussieu.fr/pagesperso/contrera/nci-programs.html

Basic run

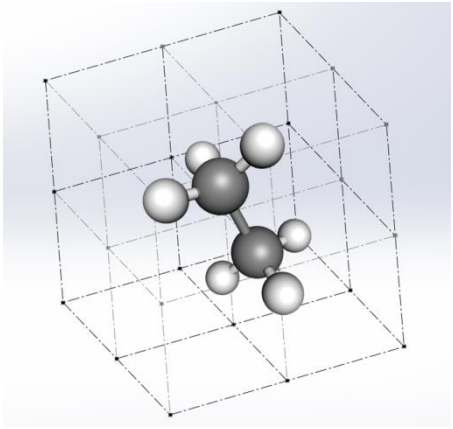
The minimum input for NCIPLOT is very simple:

1. Number of files to be read
2. Name of file(s) with extension `.wfn` for Gaussian calculations or `.xyz` for promolecular

Example

```
1
file.wfn/xyz
```

This will construct a paralepidid around the molecule:



And evaluate the reduced density gradient on a grid. For illustrative purposes, we have used a 2x2x2 grid in the ethane figure above.

Greater number of files allow to estimate inter- and intramolecular interactions separately.For example, 2 files are used for analyzing intermolecular interactions between a ligand and a protein. This is done with an extra keyword after the previous lines:

```
2
ligand.xyz
protein.xyz
```

```
INTERMOLECULAR
```

Once the input constructed, the code is invoked as follows:

nciplot.x < inputfile [> outputfile]

The main results are collected in four output files:

– name.dat file collects rho vs. RDG

– name-grad.cube file with RDG

– name-dens.cube file with sign(λ2) × ρ × 100

– name.vmd is a script for visualization of the results in VMD

Other options

a) Adaptative grids

NCIPLOT-4.0 features the implementation of the adaptive grid approach. Starting from a coarse grid to quickly explore the whole 3D space around the molecule, the user is prompted to define a succession of progressively finer grids to refine NCI results in regions where NCIs are actually detected. In this way, only NCI relevant points are computed accurately, resulting in a ∼ 10-fold speed-up.

In order to use this option, the number of adaptative grids and their relative size (finishing with "1," i. e., the basic INCREMENTS) are defined after the keyword CG2FG (Coarse Grid To Fine Grid). By default, the CG2FG option is set to 1 1 using only one level grid, i. e., without acceleration. In practice, we recommend to set CG2FG to 3 4 2 1 or 4 8 4 2 1 with 3- or 4-level grids for acceleration. This leads to an input looking as follows:

```
1
bigmolecule.xyz
CG2FG 4 8 4 2 1
```

b) Integrals

A second feature that is new in NCIPLOT4 is the optional definition of integration ranges, to assess the relative strength of NCIs in different regions (attractive, repulsive and van der Waals) of the system. This tool will be referred to collectively as Non-Covalent Interaction Integrals (NCIIs).

Starting from a standard 2D NCI plot, which allows the identification of NCIs by pairs of values of s and sign(λ2)ρ, integration regions can be defined as intervals on the sign(λ_2)ρ axis, which correspond to a window on NCIs of a given strength range. Collective integration of ρ in such ranges provides a single number, a NCII, which represents a measure of the given interaction strength window.

In order to define these ranges, the keyword RANGE is invoked, and the sign(λ_2)ρ intervals subsequently defined:

```
1
bigsystem.xyz
RANGE 3
-0.1 -0.015
-0.015 0.015
0.015 -0.1
```

This input defines 3 ranges of integration: [−0.1 −0.015], [−0.015 0.015] and [0.015 −0.1], to differentiate hydrogen bonds, van der Waals and steric crowding.

Visualization: vmd

Visualization is done through the cube files. We will be using vmd. Since visualization on the cluster might be slow, I recommend you copy all the visualization files (ELFCAR, .cube files) to your own computer and do the visualization locally.

VMD is free for academics. You can download it here:

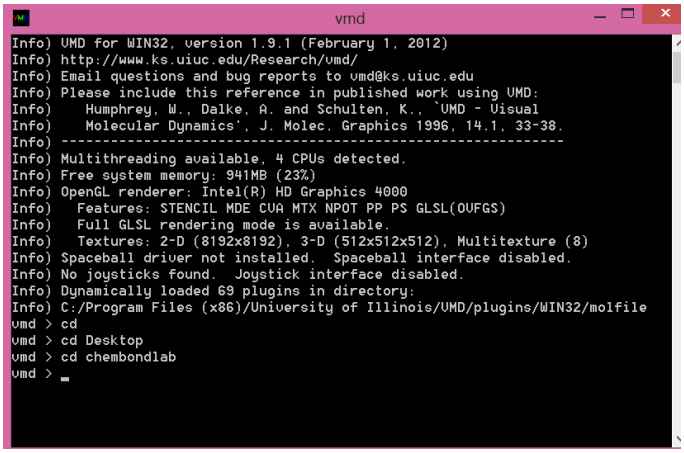
https://www.ks.uiuc.edu/Development/Download/download.cgi?PackageName=VMD

Loading the file

1-Go to the files location at the black vmd terminal:

>cd dir (dir=your working directory)

For example, I have transferred my files to a directory called “chembondlab” in my Desktop:

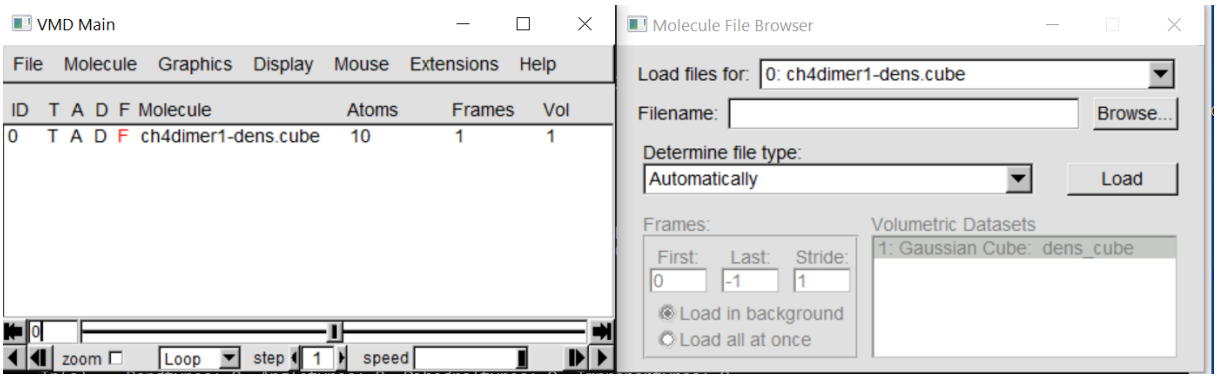


You can use DOS commands, e.g. to to see what is inside the repository:

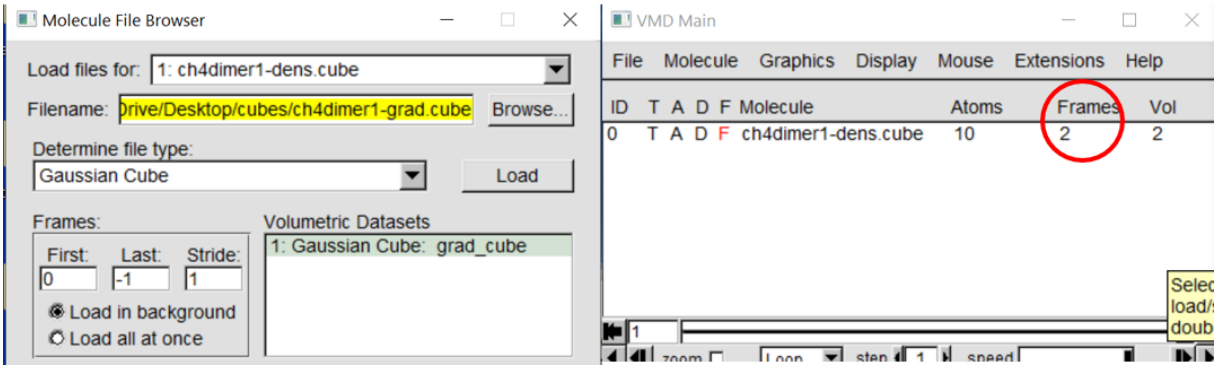
>dir

2-File > New Molecule>Browse

You choose the name-dens.cube and click Load. Without closing the window, you now choose the name-grad.cube file and click Load.

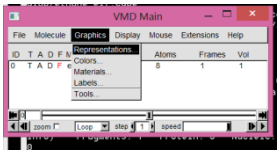


It is very important that you follow this order so that both files are uploaded one on top of the other. You can verify that you did it correctly by looking at the main vmd window. 2 frames should appear for your file:

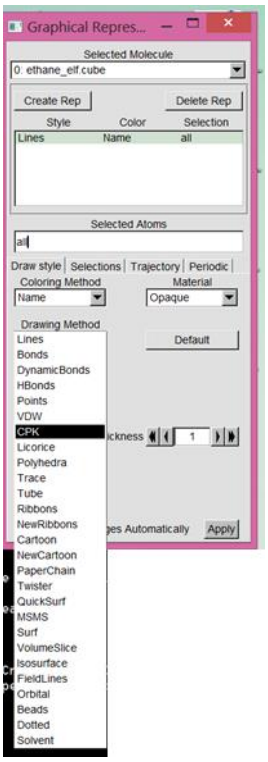


Drawing the surface

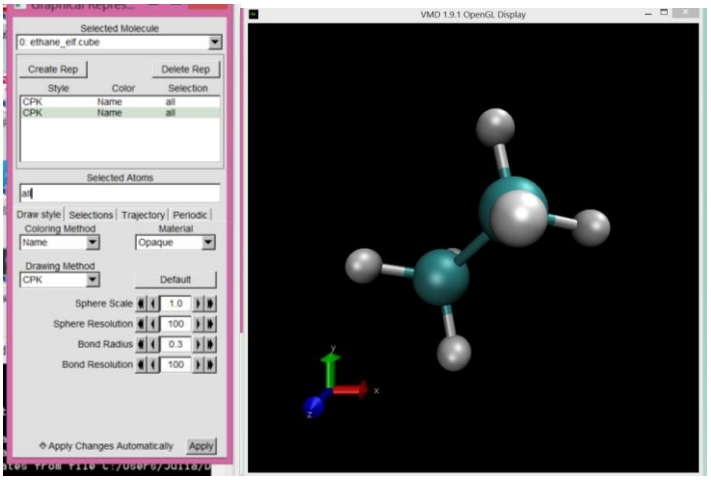
1-Graphics > Representations > Create Rep



You can change the rendering of the molecule into balls and sticks:

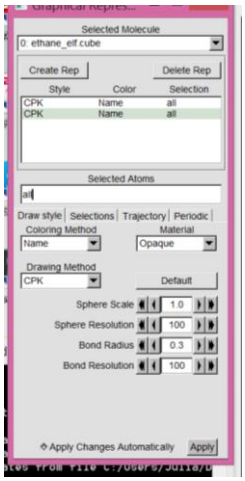


For a good quality picture, increase the sphere and bond resolution:



Now let's draw the NCI surface.

>Graphics>Create rep



And choose to draw the isosurface and its properties:

a.Drawing Method : Isosurface

b.Isovalue (0.3-0.5 are good values)

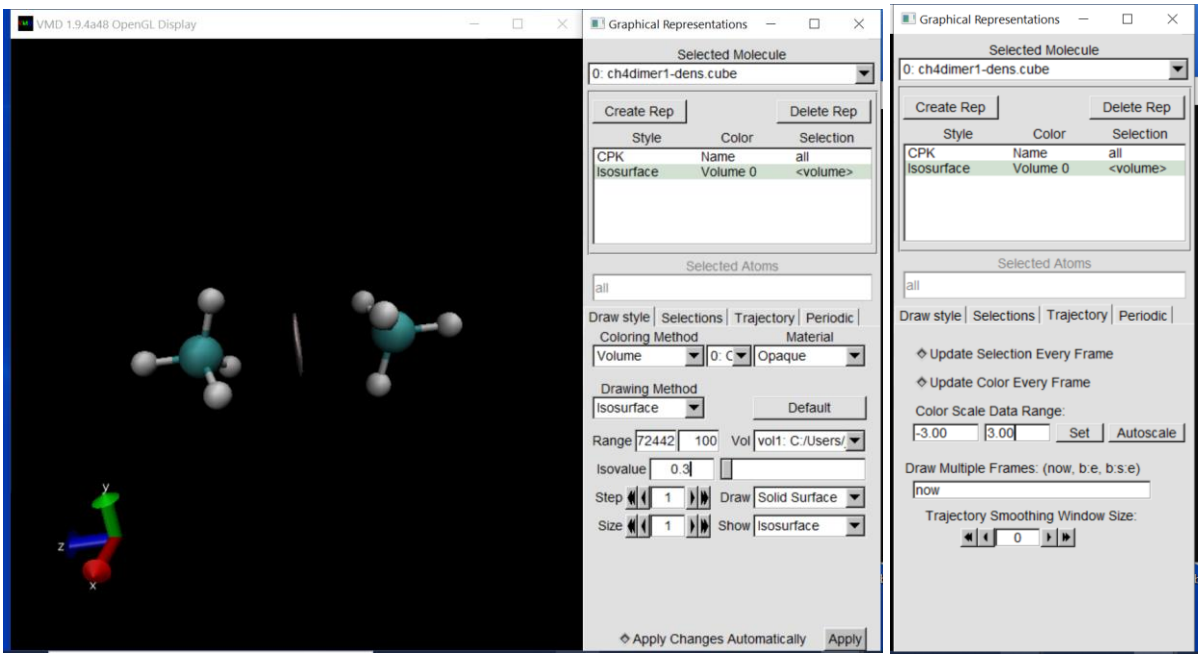
c. Draw: Solid surface

d. Coloring Method: Volume

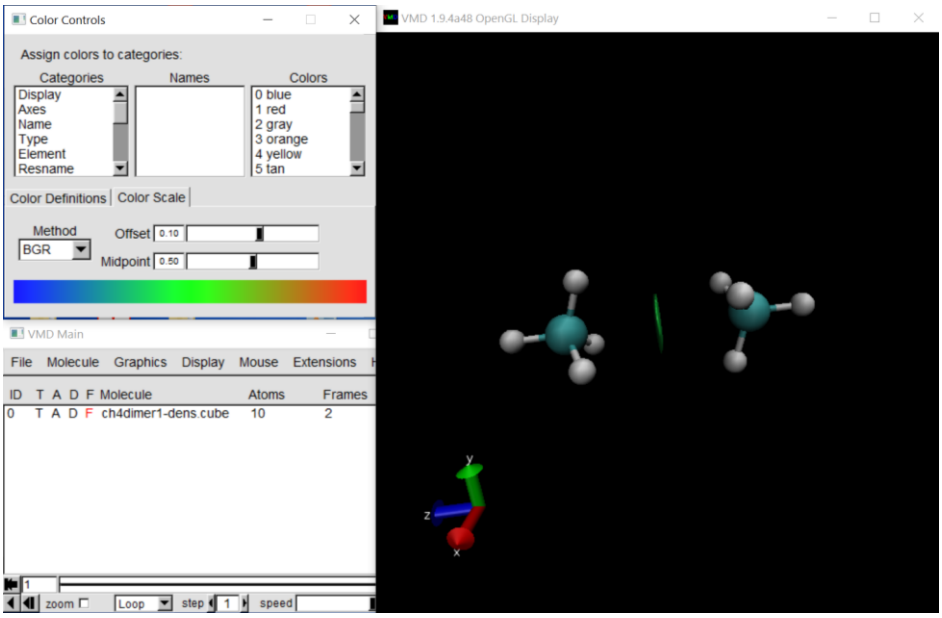
e.In Trajectory (3rd tab): Color Range Data Range :

-3	3

f. Show surface (or tap Enter key)



In order to obtain the blue-green-red NCI colors you have to go to: Graphics > Colors > Color Scale and choose: BGR

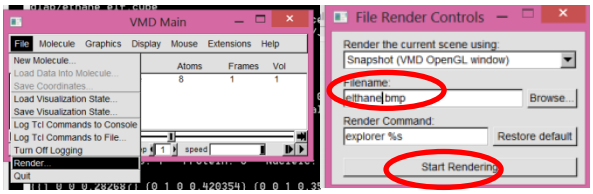


Other settings

- Changing the background color to white
  Graphics>Colors>Display (in Categories)> 8 white (in Colors)
- Getting rid of the axes
  Display>Axes>Off
- Not showing perspective
  Display>Orthographic

Saving your results

- Saving your picture
  File > Render > Filename



- Saving your state for ulterior use
  File > Nom du fichier: yourname.vmd

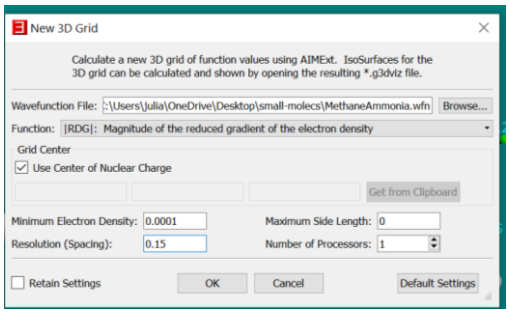
Option 2: AIMALL (Windows)

You can download a public license for AIMALL which can be used for up to 12 atoms following the instructions here: http://aim.tkgristmill.com/register.html

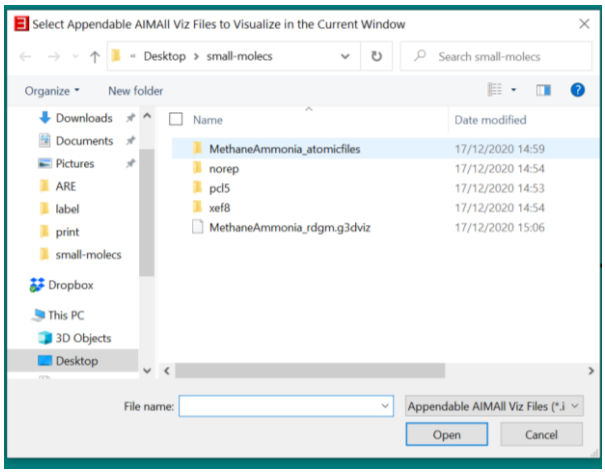
Once you have installed it:

1. Click on AIMStudio icon 
2. File> Open in New window

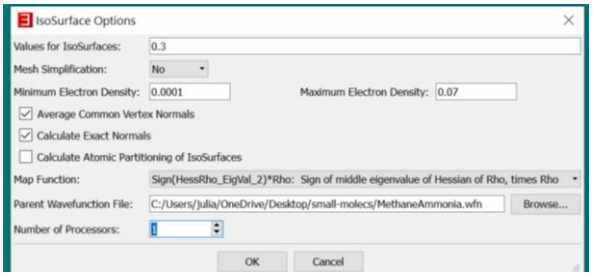
3. Run > AIMQB
4. Isosurfaces > New 3D grid
   Choose:
   Function: |RDG|: Magnitude of the reduced gradient of the electron density



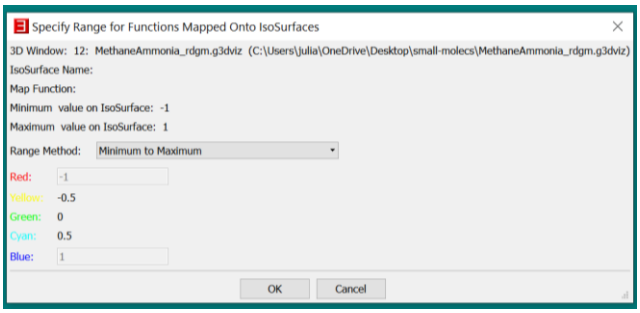
5. File > Open in current Window
   Choose: “name_rdgm.g3dviz”



6. The Isosurface option window will pop up.
   Choose:
   Values for isosurfaces=0.3-0.5
   Maximum electron density=0.7
   Map function: Sign(HessianRho_EigVal_2)*Rho
   Parent wavefunction file: name.wfn



Note that the coloring is not the default one for NCIPLOT, but given as follows:

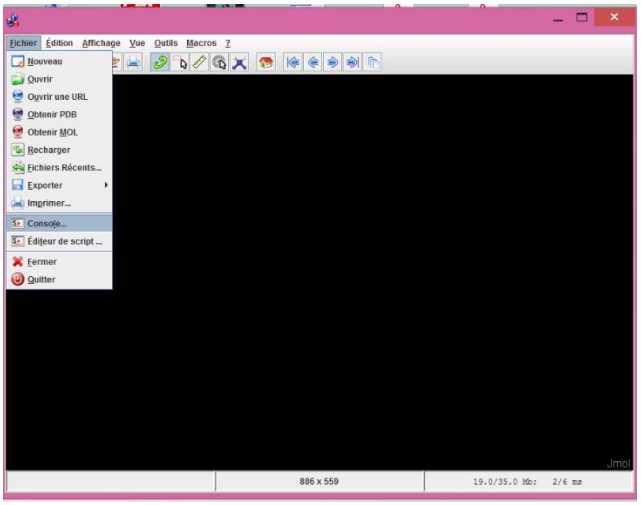


Option 3: Jmol (Windows)

You can download Jmol at: http://jmol.sourceforge.net/

You can proceed as follows:

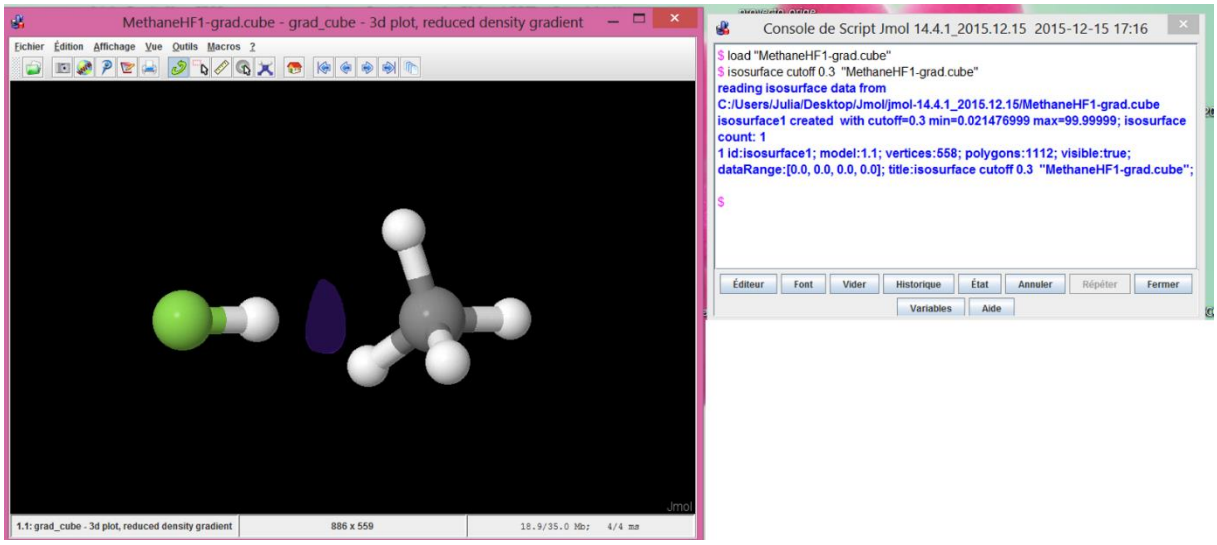
1. Click on Jmol.jar
2. Open a console with File > Console



3. Write in the console (+Intro)

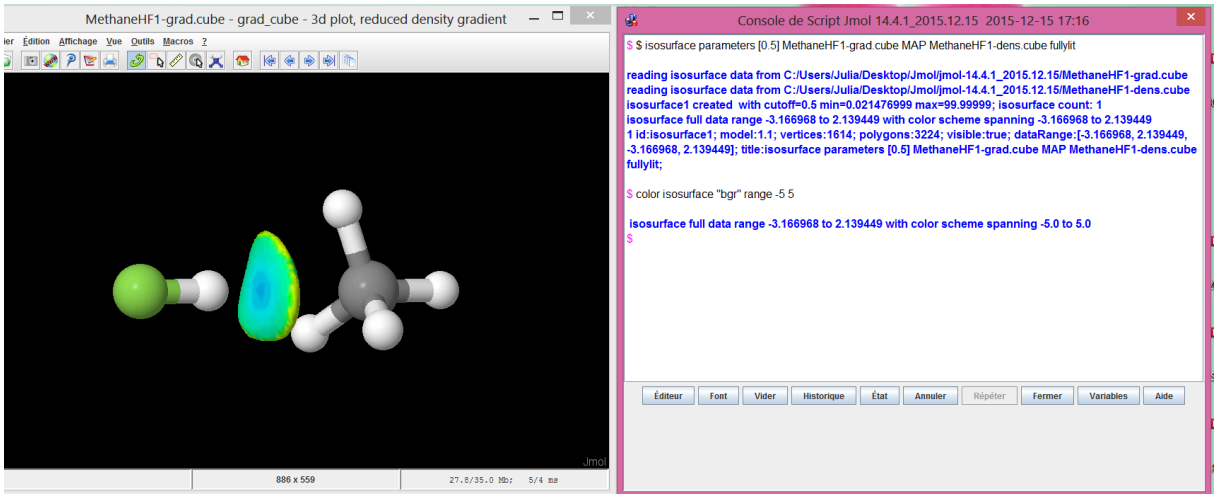
```
load "MethaneHF1-grad.cube" ↲
isosurface cutoff 0.5 "MethaneHF1-grad.cube" ↲
```

The `cutoff` is the isovalue that you can change at your wish.



4. You can map the density to color the isosurface as follows:

```
isosurface parameters [0.5] "MethaneHF1-grad.cube" MAP
"MethaneHF1-dens.cube" fullylit ↲
```



5. Other options for visualizing NCI are listed here https://chemapps.stolaf.edu/jmol/docs/examples-12/new2.htm
   Note that you can directly construct the promolecular NCI from the xyz!

- load 4watertest-1.xyz; isosurface NCI;color isosurface "bgr" range -0.03 0.03 # promolecular
- load gb-nci-dens.cube; reset;center {-2.6583672E-5 0.025006354 5.3048134E-6}; rotate z -36.33; rotate y 60.68; rotate z 81.14;
- isosurface parameters [0.35 1] NCI # promolecular (intramolecular; requires smaller cutoff)
- isosurface parameters [0.5 0 0 0 0 0.01] NCI "" # NCIPLOT density-cube only (requires scaling factor of 0.01)
- isosurface parameters [0.5] gb-nci-grad.cube MAP gb-nci-dens.cube fullylit # direct NCIPLOT results
- isosurface parameters [0.5] NCI "gb-nci-cubegen.cube" # generic CUBEGEN density file (no scaling required; the different grid gives a slight offset)

Exercises

In this exercise we will analyze non-covalent interactions in real molecules. If you have access to a Linux machine, you can download NCIPLOT here. If you are working under a Windows environment, you can register for a free version of AIMALL here. Note that the free AIMALL version does not allow more than 12 atoms, so you will not be able to do the second part of the exercise. AIMALL will enable you to have the densities of the NCI peaks. Visualization (but no quantification) is available with Jmol here.

Exercises 1 and 2 can be carried out with any of the three codes. Exercise 3 (quantification) can only be carried out with NCIPLOT.

1. Looking at the hydrogen molecule

This exercise is a connection with the model system in the previous exercise. We will now study H_2 molecule at several distances.

1. Calculate NCI at d=2.0 and 2.5Å.[1] Visualize the isosurface s=0.3 and a range in between -3 to 3. What kind of interaction do you see for each distance?
2. As we have seen in Exercise 1, we can also use NCI to visualize covalent interactions. It suffices to look at higher densities. You can do that by adding the CUTPLOT keyword in NCIPLOT:[2]

```
1
File.wfn
CUTPLOT 2.0 0.3
```

[1] You should calculate the wfn files first (e.g. CCSD/aug-cc-pvtz). If you cannot make wavefunction calculations use a promolecular density with an xyz format (explained in footnote 3).

[2] The value 2.0 corresponds to the density up to which interactions will be analyzed, i.e. ρ=0-2.0 au will be plotted. The value 0.3 corresponds to the isosurface that will be analyzed if the vmd file is used.

You can also visualize it with AIMALL introducing the value 2.0 at the Maximum Electron Density entry.

The difference from non-covalent to covalent is easy to visualize in the shape. What do changes do you observe?

3. The color of the surface is given by the density. What is the density at the point s=0? You can use `gnuplot` to plot s(ρ) (information is in file.dat) or use AIMALL to calculate the density at the critical point (BCP) between the hydrogens (which is equivalent to the point s=0).

	Color	**Shape**	**Density at s=0**	**Interaction type (non-covalent/covalent)**
d=0.7 Å				
d=2.0 Å				
d=2.5 Å				

4. Instead of using wavefunction files, you can use `xyz` files with promolecular densities. This is easily done for example for 0.7 Å as:[3]

```
2

H 0.0 0.0 0.0
H 0.0 0.0 0.7
```

Do the same for d=2.0 and d=2.5 Å. Evaluate the agreement between the promolecular approximation and the real wavefunction. Do they agree better at long or short distances? Why?

What is the difference between the functions used to describe the atoms with the promolecular approximation and the approach used with Python?

2. Methane: analysis of different non-covalent interaction types

Download the methane files. This exercise will enable you to see the difference between localized and delocalized interactions. Whereas localized interactions are mainly between two

[3] The "2" corresponds to the number of atoms, then there is a blank line and finally, the list of xyz atom positions

atoms (like the ones we saw for H_2), delocalized one are between several atoms (so the surface spreads out).

1. Visualize Methane dimer, CH_4-H_2O, CH_4-HF and CH_4-NH_3. Which interaction is local (mainly between two atoms) or delocalized?

Dimer	**NCI shape**	**Interaction type (localized/delocalized)**
CH_4- CH_4		
CH_4-H_2O		
CH_4-HF		
CH_4-NH_3		

3. Towards bigger systems and size dependency of non-covalent interactions

Download the files for benzene, naphthalene and anthracene. These systems have one, two and three fused rings.

1. Calculate NCI. Since in this case we are using rather big systems, we will choose promolecular densities, faster to calculate. We will further accelerate the calculation with an adaptative grid:

```
2
BenzeneA.xyz
BenzeneB.xyz
INTERMOLECULAR
CG2FG 3 4 2 1
```

2. Visualize the non-covalent interactions in the parallel conformation (P) and the T-shape conformation (T) for benzene and classify them as delocalized or localized.
3. Visualize now naphthalene and anthracene P and T conformations. What happens as the size increases for each interaction type?
4. You can quantify this effect with NCIPLOT. Use the following input type to obtain the volume of the non-covalent interactions the parallel and T-shape series with respect to the number of units (from 1 unit in benzene to 3 in anthracene).

INPUT EXAMPLE:

```
2
BenzeneA.xyz
BenzeneB.xyz
INTERMOLECULAR
RANGE 3
-0.07 -0.01
-0.01 0.01
0.01 0.07
```

OUTPUT EXAMPLE:

```
---------------------------------------------------------------------

                INTEGRATION DATA

---------------------------------------------------------------------

 Integration  over the volumes of rho^n

---------------------------------------------------------------------

 n=1.0        :       0.70727487
```

According to this output, the volume of the NCI region in benzene dimer is 0.70727487 au.

Benzene units	**Parallel conformation volume integral**	**T-shape conformation volume integral**
1		
2		
3		

Plot these data. What interaction becomes favored with size? Check the fit to a straight line of the local interaction, what does this mean?