18.7.4. Current structure of the package

TNT presents different faces to different users. Some users simply want to run refinement; they see the shell interface. Others want to use the TNT programs in untraditional ways; they see the program interface. A few users want to change the basic calculations of TNT; they see the library interface.

The shell interface is the view of TNT that most people see. It is the most recent structural addition, having been added in release 5E in 1995. At this level, the restraints, weights and parameters of the model are described in the 'TNT control file' and the user performs particular calculations by giving commands at the shell prompt. For example, refinement is performed with the ‘tnt’ command and maps are calculated for examination with some graphics program with the ‘make_mats’ command. TNT is supplied with about two dozen shell commands. These commands allow the running of refinement, the conversion of the model to and from TNT’s internal format, and the examination of the model to locate potential problem spots. The TNT Users’ Guide describes the use of TNT at this level.

The program interface consists of the individual TNT programs along with their individual capabilities. TNT consists of the program Shift, which handles all the minimization calculations, a program for each module (restraints that fall into a certain format) and the vital crystal data. To add new functionality, one can either modify an existing program, write a new program using the TNT libraries as a start, or write a new program from scratch ignoring the TNT libraries. As long as a program can read and write files of the same format as the rest of TNT, it will work well with TNT, even if it does not share any code. A library exists, but is not copyrighted, that contains subroutines to read and write the crystallographic file formats used by the rest of TNT.

18.7.5. Innovations first introduced in TNT

TNT was not only designed to be an easy-to-use tool for the refinement of macromolecular models, but also as a tool for testing new ideas in refinement. Since its source code is designed to allow easy reordering of tasks and simple modifications, a number of innovations in refinement made their first appearance in TNT. These features include the following.

18.7.5.1. Identifying and restraining symmetry-related contacts (1982)

Without a search for symmetry-related bad contacts, it was quite common to build atoms into the same density from two different sides of the molecule. A number of models in the Protein Data Bank contain these types of errors because neither the refinement nor the graphics programs available at that time would indicate this type of error.

18.7.5.2. The ability of a single package to perform both individual atom and rigid-body refinement (1982)

Prior to TNT, one often started a refinement with rigid-body refinement using CORELS and then switched to another program. TNT was the first refinement package to allow both styles of refinement. One was not required to learn about two different packages when running TNT.

18.7.5.3. Space-group optimized FFTs for all space groups (1989)

This innovation allowed TNT to run efficiently in all space groups available to macromolecular crystals.

18.7.5.4. Modelling bulk solvent scattering via local scaling (~1989)

With a simple and quick model of the scattering of the bulk solvent in the crystal (Tronrud, 1997), the low-resolution data could be used in refinement for the first time. The inclusion of these data in the calculation of maps greatly improved their appearance.

18.7.5.5. Preconditioned conjugate-gradient minimization (1990)

This method of minimization (Axelsson & Barker, 1984; Tronrud, 1992) allows the direct inclusion of the diagonal elements of the second-derivative matrix and the indirect inclusion of its off-diagonal elements. An additional benefit is that it allows both positional parameters and B factors to be optimized in each cycle. Previously, one was required to hold one class of parameter fixed while the other was optimized. It is much more efficient and simpler for the user to optimize all parameters at once. This method, because it incorporates the diagonal elements directly, produces sets of B factors that agree with the diffraction data better than those from the simple conjugate-gradient method.

18.7.5.6. Restraining stereochemistry of chemical links to symmetry-related molecules (~1992)

It is not uncommon for crystallization enhancers to be found on a special position in the crystal. In addition, cross-linking the molecules in a crystal is often done for various reasons. In both cases, the model contains chemical bonds to a molecule or atoms in another asymmetric unit of the crystal. In order for the stereochemistry of these links to be properly restrained, it must be possible to describe such a link to the refinement program.

18.7.5.7. Knowledge-based B-factor restraints (~1994)

When the resolution of the diffraction data set is less than about 2Å, the individual B factors of a refined model are observed to vary wildly from atom to atom, even when the atoms are bonded to one another. This pattern is not reasonable if one interprets the B factor as a measure of the vibrational motion of the atom. Traditionally, one applies an additional restraint on the B factors of the model, where the ideal value for the difference in B factor for two bonded atoms is zero.
Since it is clear from examinations of higher-resolution models that the $B$ factors generally increase from one side of a bond to the other (e.g. moving from the main chain to the end of a side chain), the traditional restraint is flawed. A restraint library was generated (Tronrud, 1996) where each bond in a residue is assigned a preferred increment in $B$ factor and a confidence (standard deviation) in that increment.

18.7.5.8. Block-diagonal preconditioned conjugate-gradient minimization with pseudoinverses (1998)

With this enhancement, TNT’s minimizer treats the second-derivative matrix as a collection of $5 \times 5$ element blocks along its diagonal, one block for each atom. While this method improves the rate of convergence for noncrystallographic symmetry restraints, its most significant feature is that it allows the refinement of atoms located on special positions without special handling by the user.

18.7.5.9. Generalization of noncrystallographic symmetry operators to include shifts in the average $B$ factor (1998)

It is rather common in crystals containing multiple copies of a molecule in the asymmetric unit for one or more molecules to have a higher $B$ factor than the others. If the transformation that generates each copy of the molecule consists only of a rotation and translation of the positions of the atoms, the difference in $B$ factors cannot be modelled. The transformations used in TNT now consist of a rotation, translation, a $B$-factor shift and an occupancy shift.

18.7.6. TNT as a research tool

TNT was intended not only as a tool for performing refinement, but as a tool for developing new ideas in refinement. While most of the latter has been done by TNT’s authors, several others have made good use of TNT in this fashion. If one has an idea to test, the overhead of writing an entire refinement package to perform that test is overwhelming. TNT allows modification at a number of levels, so one can choose to work at the level that allows the easiest implementation of the idea. Several examples follow.

18.7.6.1. Michael Chapman’s real-space refinement package

At Florida State University, Chapman has implemented a real-space refinement package, principally intended for the refinement of virus models, using TNT. He was able to use TNT’s minimizer and stereochemical restraints unchanged along with programs he developed to implement his method. More information about this package can be found at http://www.sb.fsu.edu/~rsref.

18.7.6.2. Randy Read’s maximum-likelihood function

When Navraj Pannu wanted to implement Read’s maximum-likelihood refinement functions (Pannu & Read, 1996) in TNT, he chose not to implement it as a separate program, but modified TNT’s source code to create a new version of the program Rfactor, named Maxfactor.

18.7.6.3. J. P. Abrahams’ likelihood-weighted noncrystallographic symmetry restraints

Abrahams (1996) conceived the idea that because some amino-acid side chains can be expected to violate the noncrystallographic symmetry (NCS) of the crystal more than others, one could develop a library of the relative strength with which each atom of each residue type would be held by the NCS restraint. He chose to determine these strengths from the average of the current agreement to the NCS of all residues of the same type. For example, if the lysine side chains do not agree well with their NCS mates, the NCS will be loosely enforced for those side chains. On the other hand, if almost all the valine side chains agree well with their mates, then the NCS will be strongly enforced for the few that do not agree well.

He chose to implement this idea by modifying the source code for the TNT program NCS. Since the calculations involved in implementing this idea are simple, the extent of the modification was not large.

18.7.6.4. The Buster refinement package

Bricogne & Irwin (1996) have developed a maximum-likelihood refinement package using TNT. Not only are TNT’s minimizer and stereochemical restraints used, but many of the calculations of the maximum-likelihood residual’s derivatives are performed using TNT programs. While Bricogne and co-workers have not needed to modify TNT programs to implement their ideas, there is ongoing collaboration between them and TNT’s authors on the development of commands that allow access to some previously internal calculations. More information about Buster can be found at http://www.globalphasing.com/buster/.

18.7.7. Current status of TNT

TNT is no longer being actively developed at the University of Oregon. The principal use of TNT today is from within the Buster package of Global Phasing, Ltd. Over the last ten years, they have made considerable modifications to their branch of the source code which have greatly increased the radius and rate of convergence of the minimizer and expanded the scope of the distributed library of standard groups in the ideal stereochemistry library. These enhancements are only available via their distribution of Buster.

References


