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Geometric Manipulation of Flexible Ligands*

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Abstract

In recent years an effort has been made to supplement traditional methods for drug discovery by computer-assisted "structure-based design." The structure-based approach involves (among other issues) reasoning about the geometry of drug molecules (or ligands) and about the different spatial conformations that these molecules can attain. This is a preliminary report on a set of tools that we are devising to assist the chemist in the drug design process. We describe our work on the following three topics: (i) geometric data structures for representing and manipulating molecules; (ii) conformational analysis—searching for low-energy conformations; and (iii) pharmacophore identification—searching for common features among different ligands that exhibit similar activity.

1 Introduction

Most existing pharmaceutical drugs were found either by chance observation or by screening a large number of natural and synthetic substances [7]. In recent years there has been a growing tendency to supplement the traditional methods of drug discovery by *structure-based design*. The structure-based approach builds on the improved understanding of the molecular interaction underlying diseases, and attempts to predict the structure of a potentially active compound. The prediction can then be used either to synthetically construct such a compound, or to narrow down the screening process of existing substances. For a comprehensive survey and bibliography on the subject, see [3].

The fundamental assumption of structure-based drug design is that at the molecular level the key event leading to the desired effect of the drug is the recognition and binding of a small molecule (the *ligand*) to a specific site on a target macromolecule (the *receptor*) [3]. It is further assumed that at the binding site the ligand must present steric and electrostatic complementarity to that of the receiving pocket.

Depending on whether the structure of the receptor is known or not, two types of structure-based approach can be considered. If the structure of the binding site is known, the process then centers on finding (or devising) a ligand that will complement the binding site. Our work focuses on the case when the structure of the receptor is not known. In this case, a possible scheme is to start with a set of compounds, whose structure is known, and which have been observed to achieve the desired activity. The goal then is to extract the common features of the active compounds. These common features constitute a *pharmacophore*. With a pharmacophore at hand, the chemist can look for other compounds having similar features, but which may be more potent than any of the given compounds, or have other desirable properties such as non-toxicity.

A major source of difficulties in structure-based drug design is the flexibility of molecules to attain various *conformations*, namely different spatial configurations of their atoms. Properly handling flexible ligands has been identified as a major challenge in this field [21].

Together with our collaborators at the Computational Chemistry group at Pfizer Research Central we have chosen to concentrate on the following topics where algorithmic tools are needed to support the design process: (i) Data structures for representing molecule geometry and molecular surfaces. (ii) Conformational analysis — searching for low-energy conformation. (iii) Pharmacophore identification.

The first two components can be viewed as support tools for the pharmacophore identification part. However, although our current effort focuses on tools to support the drug design process when the receptor structure is not known, some of the tools that we have developed can also be used as building blocks to support the design when the receptor structure is known.

Software tools for drug design exist in abundance [3],

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[4]. Many algorithms with geometric flavor have been proposed and implemented in this domain. We believe that our work is innovative in the following aspects. In our study of data structures for representing molecule geometry we aim to develop techniques that are efficient under conformation change—namely techniques that will allow for efficient dynamic update of the structure as the molecule conformation changes. We have devised and analyzed several models for efficient dynamic maintenance of such structures; see Section 2. The goal of conformational search (Section 3) is to produce low-energy conformations of ligand molecules. These conformations constitute the input to the pharmacophore identification procedure. The conformational search is computationally expensive and may take hours on high speed workstations [21]. In our work, we aim at speed and efficiency of calculation, willing to find low-energy conformations that are not necessarily energy minima. Finally, in the pharmacophore identification component, described in Section 4, we consider a set of active molecules, each of which may be present in many low-energy conformations, and use inter-atom distances and molecular surface information to identify common structural elements of these molecules. Our search for pharmacophores is guided by user-specified size and accuracy requirements, and our software allows for an interactive refinement of the solutions thus obtained.

2 Geometric Data Structures

A prevailing approach to modeling the geometry of static molecules is to represent each atom as a ball of fixed radius in a fixed placement relative to the other atoms [24]. The radius assigned to each atom depends on the type of the atom. There are various sets of recommended values for atom radii, and a prevailing set is known as the *van der Waals* radii. In spite of its limitations, this model, which we will refer to as the *hard sphere model* of a molecule, is widely used.

Various techniques in drug design use *molecular surfaces*. One type of molecular surfaces is simply the outer boundary of the union of the balls (or spheres) in the hard sphere model above. This type is often referred to as the van der Waals surface. There are two closely related types of surfaces: the solvent accessible surface [22] and Richard's smooth analytical surface [27]. See also [9, 10] and the survey by Mezey [24] for an extensive discussion on molecular surfaces.

A basic question in the geometric manipulation of molecules is the following: Given a hard sphere model of a molecule M and a query atom q, report the atoms of M intersected by q. It has been shown [15] that for a molecule with n atoms, an efficient data structure requiring $O(n)$ storage space can be constructed such that queries of the above type can be answered in $O(1)$ time each, after $O(n)$ expected preprocessing time. This data structure was used in [15] to obtain efficient algorithms for constructing molecular surfaces and for computing the visibility map of molecules.

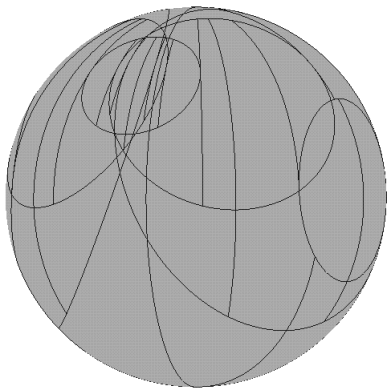

Figure 1: Partial trapezoidal decomposition of an arrangement of small circles on an atom sphere.

In the next subsection we describe an implementation of the above algorithm for computing the molecular surface. In Section 2.2 we extend the intersection data structure to the dynamic case.

2.1 Molecular Surfaces — Construction and Visualization

We construct an analytic representation of van der Waals molecular surfaces (the same procedure applies to solvent accessible surfaces as well). The representation consists of the patches that each atom sphere contributes to the outer surface, as well as the adjacency relations between patches on neighboring atoms that share a common arc. The basic procedure in our implementation of the computation of molecular surfaces is the construction of the subdivision (or *arrangement*) on each atom sphere s induced by the circles of intersection of other atom spheres with s. The arrangement is constructed incrementally, by adding one circle at a time, and maintaining the *trapezoidal decomposition*[1] of the current arrangement. See Figure 1 for an illustration.

We represent the arrangements of the circles using a quad-edge structure [12]. Once we obtain the arrangements for all atom spheres, we go on to construct the (van der Waals, or solvent accessible) molecular surface. This is achieved by adjusting between the individual atom arrangements so as to retain only the faces in those arrangements that contribute to the outer sur-

[1] The trapezoidal decomposition is a refinement of the arrangement of the circles by adding certain arcs of great circles through the poles. For more details on trapezoidal decomposition, see, e.g. [25]. In the implementation we extend such arcs only from points of tangency of original circles with great circles through the poles.

face, and by updating the edge information of edges that now border faces belonging to two different atom spheres (see [15] for details).

The arrangements on the spheres are used to compute the molecular surface area and the (possibly null) area contributed by each atom to the outer surface, and for graphic display of the intersection pattern of each atom with its neighbors (Figure 1). The information about area contribution of individual atoms is used in the pharmacophore identification module described below. Besides computing the outer surface, the program computes internal boundary components (bounding so-called *voids*) and outputs their surface area as well.

The implementation provides extensive facilities for interaction. It displays the molecule and the individual atom arrangements graphically and allows for recoloring of the atoms according to several parameters such as the level of contribution to the outer surface. The edges of the arrangement on each atom sphere are colored to distinguish between the contribution of the atom to the outer molecular surface and to each void (if any). The program also provides various statistics on the data structures it uses and allows for experimenting with and tuning up certain parameters that control the structures.

2.2 Dynamic Maintenance

We can model the flexibility of a molecule to attain different conformations as if the atoms were rigid links of a robot linkage, and the bonds between some of the atoms are rotational (or rotatable) joints (see Section 3 for more details on this simplified model). We describe these kinematic constraints by a graph where each node corresponds to an atom sphere, and an edge between nodes describes a constraint. An edge can be either rigid—there is a fixed relative displacement between the atoms that it connects, or rotatable—when there is a degree of freedom of rotation around a fixed line between the two atoms.

Suppose that we are given a sequence of update requests that are aimed at changing the conformation of the molecule. This is done by giving a sequence of joint angles to which we need to update the rotatable bonds. The sequence of updates is interleaved with intersection queries of the form: does a given query sphere intersect any of the atom spheres in the molecule at its current conformation. We are looking for a strategy to maintain a data structure, such that a sequence of updates and queries will be answered in optimal time, where the algorithm has the freedom to break or merge substructures.

In [14] we study several models of dynamic maintenance of such structures, and devise maintenance algorithms for them. We give a worst-case optimal strategy for the case where the molecule graph is a tree. The key idea to achieving the efficiency in the worst-case is that of a *balanced decomposition* of the tree, namely, breaking up the tree into subtrees such that the number of subtrees (which corresponds to the cost of a query) is roughly the same as the size of each subtree (which corresponds to the worst case cost of an arbitrary update). We describe an efficient algorithm for constructing such a balanced decomposition.

We also show that obtaining the optimal solution for a given sequence of updates and queries, even when the molecule graph is a path, is an NP-hard problem and we present approximation algorithms for this case.

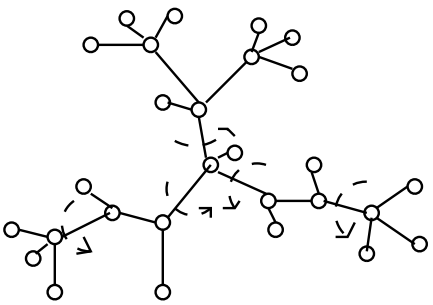

Figure 2: The molecule of valine with its torsional degrees of freedom. A ball indicates an atom, and a line—a bond between two atoms.

3 Conformational Search

Searching the conformational space of small compounds is an important operation in the process of pharmaceutical drug design [21]. Given a function that computes the energy of the molecule, the goal of the search is to produce low-energy conformations that are geometrically distinct. These conformations can provide the input to a pharmacophore generation procedure, or can be used to screen large databases of protein molecules for possible docking receptors.

When conformational search is conducted, the prevailing practice is to consider only the torsional degrees of freedom of the molecule (see Figure 2 for an example). Other degrees of freedom, such as bond lengths between atoms or bond angles between consecutive bonds, are often ignored since their variation does not drastically change the molecular conformation. The definition of empirical force fields that approximate the free energy of a molecule has been the focus of intensive research [21]. In reality, the conformation of a molecule is influenced both by intermolecular and intramolecular forces. However, the receptor site is frequently unknown and modeling a molecule in solution is difficult. As a result, conformational search is conducted with force fields which assume that the molecule is in vacuum. Typically these force fields have terms that involve pairs, triplets, and

quadruples of bonded atoms, as well as pairs of distant non-bonded atoms.

Conformational analysis is a very hard search problem when the considered molecule has more than 5 degrees of freedom. For a comprehensive survey of previous methods see [8, 21, 29]. During search, emphasis is placed on generating geometrically different conformations of a molecule within a user-defined energy interval. The underlying assumption is that one of these conformations will be adopted inside a receptor cavity. Selective generation of conformations with certain properties may also be desired. For example, if a pharmacophore is known, one may want to find low-energy conformations that retain the features of the pharmacophore at their relative positions. We describe below an implementation that generates low energy conformations of a molecule, and our preliminary efforts to efficiently organize the resulting conformations in clusters and provide feedback to conformational search.

3.1 General Framework

Inspired by our success with probabilistic techniques for robot motion planning [16], we have implemented a search procedure which randomly samples the conformational space of a small molecule. The search uses the Tripos Force Field [31] and is divided into three steps: generation of random conformations, minimization of these conformations, and grouping or clustering of the resulting conformations.

Generation of conformations. During this step, a large number of conformations, frequently tens of thousands, are generated at random over the conformational space of the molecule. In contrast with previous search methods that discretize each torsional angle, we obtain a random conformation by selecting each torsional degree of freedom uniformly from its allowed range. However, if information is available for the prefered values of a particular torsion angle (such information has been collected from crystallographic databases [19]), then it is easy to select the value of this torsion angle according to a distribution that reflects the above information. The resulting structure is stored only if it avoids self-collisions. These may result from intersections of the *hard spheres* of non-bonded atoms.

Minimization. An efficient minimizer [5, 28] is used to obtain conformations that are at local energy minima. It is important to note that minimization is the most time consuming step during conformational search, and that its efficiency is crucial for the performance of the search. A large part of the minimization time is spent towards the end of the procedure, when the process tries to meet certain user-defined stop criteria. Relaxing these criteria reduces significantly the running time at the expense of accepting conformations that are not

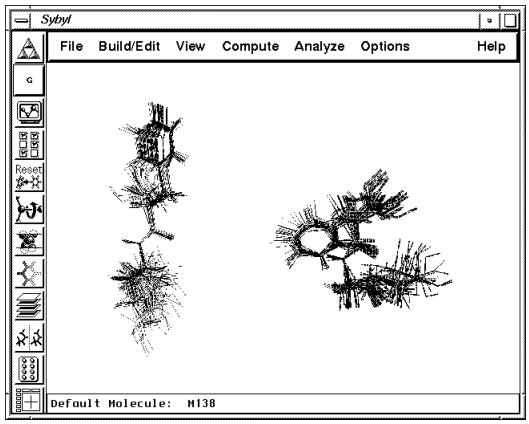

Figure 3: Two clusters of an actual drug molecule.

at local energy minima. However, experiments show that conformations very close to a local minimum usually have similar geometries. These conformations are later grouped by our implementation in clusters as explained in the next paragraph. For the purpose of most applications (i.e., pharmacophore generation) only one conformation per cluster is retained. Hence, no relevant information is likely to be lost by avoiding the full minimization of conformations that belong to the same cluster. In the framework of conformation generation and minimization, the research described in 2.2 may prove beneficial in reducing the time spent on collision checking and on the calculation of the energy function terms.

Distinct geometric conformations. Interpreting a set of a few thousands of conformations is challenging and can benefit from extensive research in the area of clustering [17]. Currently, our system partitions conformations into clusters by using an easy-to-compute measure of distance between conformations. In particular, three distinct atoms a_1, a_2, a_3 are first selected in the molecule. The distance of two conformations is defined as the sum of the Euclidean distances of their corresponding atoms, after identifying atoms a_1, directions a_1a_2, and planes $a_1a_2a_3$ of the two conformations. Clustering is performed by putting a given conformation in an existing cluster if its distance from the 'center' of that cluster is less than a predefined value. If no such cluster is found, a new cluster is formed. Each torsional degree of freedom of the 'center' of the cluster is the average of the corresponding torsional angles of the conformations in the cluster. Despite its simplicity, this procedure seems to perform well in practice when a few thousands of conformations are involved. Examples of clusters for a molecule with 12 degrees of freedom are shown in Figure 3. It is interesting to investigate further the relationship between cluster generation and the

extent to which minimization is performed. Additionally, randomization and hashing techniques may reduce the complexity of clustering and permit its efficient use with very large numbers of conformations.

4 Pharmacophore Identification

In the literature, pharmacophore identification techniques model a molecule either as a graph (where atoms are vertices and bonds are edges) [30], or as a set of points in 3-dimensional space [6]. The second approach is often justified by arguing that pharmacophores tend to be non-local — bond information seems to have less of an influence on ligand-receptor binding than other properties such as hydrophobicity, hydrogen bonding, and electrostatic fields (see, e.g., [11]). Therefore, we model a molecule as a set of *labeled* points in three dimensions, where each point represents the center of an atom. In fact we expand each point into a small sphere of radius ϵ centered at that point, where ϵ represents uncertainty in locating atoms. Labeling allows us to generalize the notion of compatibility between atoms, so that we can take into consideration other structural/chemical properties of the atoms without having to change our algorithms. One type of labeling that we use is the surface area contributed by an atom to the outer molecular surface (see Section 2.1).

Since each molecule may exist in one of many low-energy conformations, the representation of a molecule consists of a collection of one or more point sets, each corresponding to one such conformation. The problem is now to determine one or more point sets that are congruent to a subset of some conformation in each molecule, where the only transformations permitted are translations and rotations. Note that allowing molecules to have multiple conformations makes the problem much harder, because now for any pair of molecules, we may potentially have to compare each possible pair of conformations in these molecules to determine even one pharmacophore.

It is known that if we restrict each molecule to one conformation, finding the largest such subset is NP-hard; from the point of approximate solutions, it is known to be hard to approximate [1] and only weak positive results are known [18]. There exist polynomial-time solutions for the problem if we consider only two point sets [2], and these could be used to build solutions over a larger number of point sets. However, these solutions are inefficient. In the next subsection, we describe an implementation that provides a fast heuristic to determine pharmacophores between two point sets. Subsequently, we outline a scheme whereby such pharmacophores could be used to reconstruct a solution for many point sets efficiently.

4.1 The Two-Set Problem

Our implementation is based on the following observation: if we consider the multi-set of numbers formed by measuring the distance between any two atoms in a molecule, and compare these multi-sets for two molecules, then if the multi-sets are identical, it is likely that the molecules are congruent as well. Our algorithm computes these sets for two molecules, and sorts them, merging the two sorted lists. Then it clusters similar distances together, where each pair of similar edges (one from each molecule) *witnesses* a match between the corresponding endpoints. Note that labels on the points and edges can be taken into account at this stage. Clearly, if the set of witnesses for a match is large, that match has a good chance of being a part of a pharmacophore.

We use this to generate, for each point in a set, a ranking of the points in the other set, a high rank implying that the match is likely to occur in a pharmacophore. This is used as the input to a stable marriage algorithm [13], which generates a matching between points of the two sets. Now, we take triplets of these pairs and generate a transformation mapping each point to its match. This transformation can then be applied to the entire point set to determine the common subset it induces. Note that we can repeat the process of generating stable marriages and producing transformations to generate more solutions of varying sizes.

Our implementation has a graphic interface to display the results of the search. Various tolerances, such as errors in point locations and distance measurement, and minimum subset sizes, can be specified. Multiple solutions can be displayed simultaneously, and the user can change parameters and view the results in real time.

4.2 Extension to Multiple Molecules

Previous attacks on the problem often use a clique-finding approach similar to the above to determine common subsets. This is then extended to multiple molecules in a natural way, where one molecule is compared with each of the rest simultaneously, and then the procedure is iterated [23]. This approach is simple, but computationally expensive, and cannot handle situations where a pharmacophore may exist in most, but not all molecules. It should also be noted that these methods do not address the case where molecules can be in one of many conformations.

In general, it is easier to determine whether a given substructure exists in a conformation than to search for some substructure that exists in both conformations. Therefore, our approach to solving the multiple molecule problem involves techniques similar in spirit to geometric hashing [32], a technique used for pattern recognition in computer vision, and recently also

in computational biology [26]. Basically, we use a *probe* to obtain pharmacophores from two sets of conformations, using techniques like the one above, or using a simpler random search procedure. Then, we hash all the conformations of all the molecules into a table, using a hash function of a geometric nature defined on pairs or triplets of atoms, and taking all such subsets. With every such table entry, we associate the conformation which contributed to a hit on it. Now, if we hash the pharmacophores in a similar fashion, we can identify which conformations are hit frequently, and so can deduce which molecules are likely to possess at least one conformation containing the pharmacophore.

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References

[1] T. Akutsu, M. Halldorsson, On the approximation of largest common point sets,*Proc. of the International Symposium on Algorithms and Computations, Springer-Verlag Lecture Notes on Computer Science* **834** (1987), pp. 405–413.

[2] H. Alt, K. Melhorn K., H. Wagener and E. Welzl, Congruence, Similarity, and Symmetries of Geometric objects, *Discrete Comput. Geom.* **3** (1988), pp. 237–256.

[3] L.M. Balbes, S.W. Mascarella, D.B. Boyd, A perspective of modern methods for computer aided drug design, *Reviews in Computational Chemistry* **5** (1994), VCH Publishers Inc., pp. 337–379.

[4] D.B. Boyd, Compendium of molecular modeling software, *Reviews in Computational Chemistry* **4** (1993), VCH Publishers Inc., pp. 229–257.

[5] R.P. Brent, *Algorithms for finding zeros and extrema of functions without calculating derivatives*, Ph.D. Thesis, Stanford University, 1971.

[6] Brint, A. T. and Willet P, Algorithms for the identification of three-dimensional maximal common substructures. *J. Chem. Inf. Comput. Sci.* **27** (1987), pp. 152–158.

[7] C.E. Bugg, W.M. Carson, and J.A. Montgomery, Drugs by design, *Scientific American*, December 1993, pp. 92–98.

[8] D.E. Clark, G. Jones, P. Willet, P.W. Kenny, and R.C. Glen, Pharmacophoric pattern matching in files of three-dimensional chemical structures: Comparison of conformational searching algorithms for flexible searching, *J. Chem. Inf. Comput. Sci.* **34** (1994), pp. 197–206.

[9] M.L. Connolly, Solvent-accessible surfaces of proteins and nucleic acids, *Science* **221** (1983), pp. 709–713.

[10] M.L. Connolly, Analytical molecular surface calculation, *J. of Applied Crystallography* **16** (1983), pp. 548–558.

[11] R.C. Glen, G.R. Martin, A.P. Hill, R.M. Hyde, P.M. Woollard, J. Salmon, J. Buckingham and A. Robertson, Computer-aided design and synthesis of 5-substituted Tryptamins and their pharmacology at the 5-HT Receptor: discovery of compounds with potential anti-migraine properties, *J. Med. Chem.*, **38** (1995), pp. 3566-3580.

[12] L.J. Guibas and J. Stolfi, Primitives for the manipulation of general subdivisions and the computation of Voronoi diagrams, *ACM Transactions on Graphics*, **4** (1985), pp. 74–123.

[13] D. Gusfield and R. W. Irving, *The stable marriage problem: structure and algorithms*. MIT Press, Cambridge, 1989.

[14] D. Halperin, J.-C. Latombe and R. Motwani, Dynamic maintenance of kinematic structures, Manuscript, 1995.

[15] D. Halperin and M.H. Overmars, Spheres, molecules, and hidden surface removal, *Proc. 10th ACM Symposium on Computational Geometry*, Stony Brook, 1994, pp. 113–122.

[16] L.E. Kavraki, *Random networks in configuration space for fast path planning*, Ph.D. Thesis, Stanford, 1995.

[17] L. Kaufman and P.J. Rousseeuw, *Finding groups in data an introduction to cluster analysis*, Wiley, NY, 1990.

[18] S. Khanna, R. Motwani, and Frances F. Yao, Approximation algorithms for the largest common subtree problem, Report No. STAN-CS-95-1545, Department of Computer Science, Stanford University (1995).

[19] G. Klebe and T. Mietzner, A fast and efficient method to generate biologically relevant conformations, *J. of Computer Aided Molecular Design* **8** (1994), pp. 583-606.

[20] A.R. Leach, A survey of methods for searching the conformational space of small and medium size molecules, *Reviews in Computational Chemistry* **2** (1991), VCH Publishers Inc., pp. 1–55.

[21] T. Lengauer, Algorithmic research problems in molecular bioinformatics, *IEEE Proc. of the 2nd Israeli Symposium on the Theory of Computing and Systems*, 1993, pp. 177–192.

[22] B. Lee and F.M. Richards, The interpretation of protein structure: Estimation of static accessibility, *J. of Molecular Biology* **55** (1971), pp. 379–400.

[23] Y.C. Martin, M.G. Bures, E.A. Danaher, J. DeLazzer, I. Lico, and P.A. Pavlik, A fast new approach to pharmacophore mapping and its application to dopaminergic and benzodiazepine agonists, *J. of Computer-Aided Molecular Design* **7** (1993), pp. 83–102.

[24] P.G. Mezey, Molecular surfaces, in *Reviews in Computational Chemistry*, Vol. I, K.B. Lipkowitz and D.B. Boyd, Eds., VCH Publishers, 1990, pp. 265–294.

[25] K. Mulmuley, *Computational Geometry: An Introduction Through Randomized Algorithms*, Prentice Hall, New York, 1993.

[26] R. Norel, D. Fischer, H.J. Wolfson, and R. Nussinov, Molecular surface recognition by a computer vision-based technique, *Protein Engineering* **7** (1994), pp. 39–46.

[27] F.M. Richards, Areas, volumes, packing, and protein structure, in *Annual Reviews of Biophysics and Bioengineering* **6** (1977), pp. 151–176.

[28] D.A. Pierre, *Optimization theory with applications*, Dover, NY, 1986.

[29] A. Smellie, S.D. Kahn, and S.L. Tieg, Analysis of conformational coverage: 1. Validation and estimation of coverage, *J. Chem. Inf. Comput. Sci*, 35(1995), pp. 285-294.

[30] Y. Takahashi, Y. Satoh and S. Sasaki, Recognition of largest common structural fragment among a variety of chemical structures, *Analytical Sciences* **3** (1987), pp. 23–28.

[31] Tripos Associates Inc., Sybyl Manual, St. Louis, MO.

[32] H. J. Wolfson, Model-based object recognition by geometric hashing, *Proc. of the 1st European Conference on Computer Vision* (1990), pp. 526-536.