The First Structures of T Cell Receptors Bound to Peptide–MHC

Kai W. Wucherpfennig

J Immunol 2010; 185:6391-6393; doi: 10.4049/jimmunol.1090110
http://www.jimmunol.org/content/185/11/6391

Why The JI?

• Rapid Reviews! 30 days* from submission to initial decision
• No Triage! Every submission reviewed by practicing scientists
• Speedy Publication! 4 weeks from acceptance to publication

*average

References
This article cites 26 articles, 11 of which you can access for free at:
http://www.jimmunol.org/content/185/11/6391.full#ref-list-1

Subscription
Information about subscribing to The Journal of Immunology is online at:
http://jimmunol.org/subscription

Permissions
Submit copyright permission requests at:
http://www.aai.org/About/Publications/JI/copyright.html

Email Alerts
Receive free email-alerts when new articles cite this article. Sign up at:
http://jimmunol.org/alerts
The First Structures of T Cell Receptors Bound to Peptide–MHC

Kai W. Wucherpfennig

The structure of the MHC class I molecule HLA-A2 reported in 1987 by Bjorkman et al. (1) had revealed how peptide Ags are presented to T cells: peptides are buried in the long and deep groove of the MHC molecule, flanked on each side by a long α helix. The structure immediately suggested that TCRs recognize the compound surface formed by the MHC helices and the embedded peptide (2), and a number of groups started to work on the extremely challenging problem of crystallizing a complex of peptide–MHC bound to a TCR. This quest was driven by several conceptually important problems. Among these, the most important question was how T cells recognize MHC despite the vast diversity within the T cell repertoire. Would there be a general binding mode of TCR on the MHC helices that explained MHC restriction, or would TCRs be able to bind to MHC proteins in many different ways? If a general binding mode indeed existed, how could it accommodate the vast repertoire of TCR sequences and the polymorphic nature of MHC molecules? In addition, would MHC class I and class II proteins be recognized in a similar manner? Thus, the goal of this effort was to provide a definitive molecular understanding for the concept of “MHC restriction”, revealed by the elegant functional experiments of Zinkernagel and Doherty (3), for which both received the Nobel Prize in Physiology or Medicine in the same year these two structures were published.

Prior to publication of the HLA-A2 structure, only a few immunologists had anticipated the decisive contributions that structural biology was going to make to their field, but with this single structure it became crystal clear that protein structures could provide deep insights into intractable immunological problems. For noncrystallographers, it is important to appreciate that, with this type of work, you really have nothing in your hands, despite years of effort, until there are crystals that diffract to a sufficient resolution. The initial structure of HLA-A2 had been determined using protein purified from a human B cell line, and the groove therefore contained a complex mixture of peptides. In 1987, it was therefore not even known how a recombinant MHC protein with a single bound peptide could be generated. Furthermore, expression of soluble TCRs in quantities sufficient for serious structural work proved to be very difficult, even though clever approaches that yielded small quantities were reported. On the MHC side, it became clear that the MHC helices actually folded around the offered peptide and that it was impossible to generate stable, empty MHC class I molecules that could later be loaded with peptide. Years later, cell biological studies showed that nascent MHC class I chains become part of a peptide loading complex in the endoplasmic reticulum, in which the MHC class I molecule is held by chaperones in a conformation suitable for rapid peptide acquisition (4). Therefore, one solution to the MHC class I expression problem was to generate the molecules by refolding the MHC class I heavy chain and β2-microglobulin from subunits expressed in Escherichia coli in the presence of peptide. This approach, reported 5 y after the initial HLA-A2 structure, provided a reliable source of stable, single peptide–MHC complexes that crystallized on their own (5). A second solution was to express the MHC class I molecule in insect cells lacking components of the peptide loading complex and to culture these cells with the peptide of interest (6). The two groups also found different solutions to the TCR expression problem, either by refolding from individual chains expressed in E. coli or by expression in insect cells (7, 8). On top of these technical challenges in generating the two components, there was also the remaining question of whether the complex would indeed crystallize, given the low affinity and fast off rate for TCR binding reported by Mark Davis and colleagues (9). Clever photo-crosslinking approaches had been developed (10), but in crystallization trials, the protein concentration was far above the $K_d$ of the interaction so that most TCR and peptide–MHC molecules would be expected to be in the bound state.

The two groups that succeeded in 1996 came well prepared and had laid the groundwork for this important advance. Don Wiley’s group had determined the crystal structure of HLA-A2 in 1987 in collaboration with Jack Strominger’s laboratory and later developed the E. coli approach to MHC class I refolding. Ian Wilson’s group had determined the structures of mouse MHC class I proteins with bound single peptides from viral antigens in 1992, after having teamed up with Per Peterson’s group, which had developed the approach for MHC class I expression in insect cells (6). The two structures of TCR–peptide–MHC complexes were published in short succession in the fall of 1996, the Garcia et al. paper (7) (Wilson laboratory) in October, and the Gaborczi et al. paper (11) (Wiley laboratory) in November. The Wilson laboratory built on its early success with mouse H2-Kb and used the well-characterized 2C TCR with the low-affinity dEV8 self-peptide (12).
moved in the last figure to the most exciting result: the structure of the complex. The Wiley laboratory built on its work with the human HLA-A2 molecule and used a high-affinity antiviral TCR (A6 TCR) specific for a Tax peptide from HTLV-1, a human retrovirus associated with adult T cell leukemia and a chronic inflammatory CNS disease. The paper described many salient features of this complex, including the atomic contacts between TCR and peptide as well as MHC.

What do we see in these structures, or better, what do we learn? Both papers described a similar binding solution, a diagonal orientation of the TCR on peptide–MHC that buried most of the exposed surface of the peptide underneath the TCR footprint. This result was very exciting because these two TCRs were biologically very different from each other: They bound mouse versus human MHC, had a low versus high affinity for their peptide–MHC ligand, and recognized self versus viral peptides. The similarity in the overall binding mode, despite these biological differences, immediately suggested that this binding solution was probably general. Both papers reported that their respective TCR had a rather flat binding surface for peptide–MHC, and the Wiley group proposed a structural rationale for this general binding mode. They showed that the surface of MHC class I and class II molecules was actually not flat, but that there were two “peaks” near the N termini of the α helical regions. The diagonal orientation allowed the TCR to fit down between the two highest points of the MHC molecule so that it could make contacts along the length of the peptide. Prior work by Stan Nathenson’s laboratory had suggested a diagonal TCR footprint based on mutational analysis of 59 antiviral T cell clones restricted by H2-Kd (13). The similar diagonal orientation in both structures, combined with these functional data, provided solid experimental support for the idea that this binding mode was general.

How do TCRs recognize the bound peptide? The diagonal docking solution positioned the most diverse loops of the TCR (CDR3α and CDR3β) over the center of the peptide. Such a result had been predicted because this organization would provide the greatest discrimination power for peptide recognition (14). In both structures, the CDR3 loops formed a cavity in the center of the binding surface, and in the A6 TCR structure this pocket was occupied by the central P5 residue of the Tax peptide (a tyrosine). The two CDR3 loops of A6 TCR made extensive contacts to the Tax peptide, contacting residues 4, 5 (CDR3α) and 5, 6, and 8 (CDR3β). The germline-encoded CDR1 loops of both chains also contributed to T peptide recognition, with CDR1α positioned over the peptide N terminus and CDR1β over the C terminus. Mark Davis’ laboratory had earlier proposed that the TCRβ-chain was placed over the C-terminal part of the I-Eα–bound cytochrome C peptide, based on cleverly designed experiments with single-chain TCR transgenic mice (15).

What are the consequences of the diagonal binding mode for MHC binding? The diagonal positioning placed both CDR2 loops over the MHC helices, CDR2α over the MHC α2 helix, and CDR2β over the MHC α1 helix. It also allowed the CDR1 loops to contact the MHC helices as well as the bound peptide. Placement of the germline-encoded CDR1 and CDR2 loops over the MHC helices provided a structural solution for the biological problem of MHC restriction. The CDR1 and CDR2 loops were shorter and had less conformational diversity than did the CDR3 loops. The less diverse features of TCR were thus positioned over the MHC helices and the most diverse loops over the bound peptide.

After publication of these two papers, attempts were made to identify conserved MHC residues required for binding by all TCRs (16, 17). Even though some MHC residues were identified, TCR contact residues on MHC proteins could not be identified. Subsequent work by the Garcia and Kappler laboratories showed that all mouse TCRs that used Vβ8 and recognized I-A proteins showed similar binding interactions of the germline-encoded CDR1 and CDR2 loops with the I-A α1 helix (18, 19). Mutation of key contact residues of the Vβ8 CDR2 loop greatly reduced thymocyte numbers in mice expressing such mutated TCRβ-chains, suggesting that these MHC contacts were important for T cell development (20). These results led to the hypothesis that individual Vα and Vβ domains have preferred binding interactions with MHC proteins because of the coevolution of individual V-genes with MHC (21).

Later structural characterization of MHC class II-restricted TCRs confirmed the general principles outlined in these two papers, even though MHC class II-bound peptides can exit the groove on both ends (22). The Wiley group used the accessibility of the peptide C terminus to develop a clever approach for production of stable MHC class II–peptide–TCR complexes (23). They took advantage of the long α helix of MHC class II-bound peptides and attached the peptide sequence to the N-terminus of the TCRβ-chain; stable complexes resulted from binding of the TCR-tethered peptide to the MHC molecule. This technique enabled structural characterization of autocrine TCRs with low affinity for their peptide–MHC ligand (24, 25).

A large number of structures have now been determined that have dealt with important topics, such as immunodominance, autoimmunity, tumor immunity, and allo-MHC recognition (26). The García et al. (7) and Garboczi et al. (11) papers deserve to be considered classics in immunology. They provided the structural explanation for MHC restriction, a phenomenon that had intrigued and baffled immunologists for more than two decades. They are testament to what can be accomplished by the right mix of excellence, good judgment, and persistence, with the little dose of good luck that is required for all crystallographic endeavors.

Disclosures
The author has no financial conflicts of interest.

References


