Dynamics of Nuclear Receptor Helix-12 Switch of Transcription Activation by Modeling Time-Resolved Fluorescence Anisotropy Decays

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Supplementary Information

1. Predicted anisotropy for H12 and H5

2. Parametrization of the Cysteine-Fluorescein fluorescent probe

3. Force-Field for Cysteine-Fluorescein

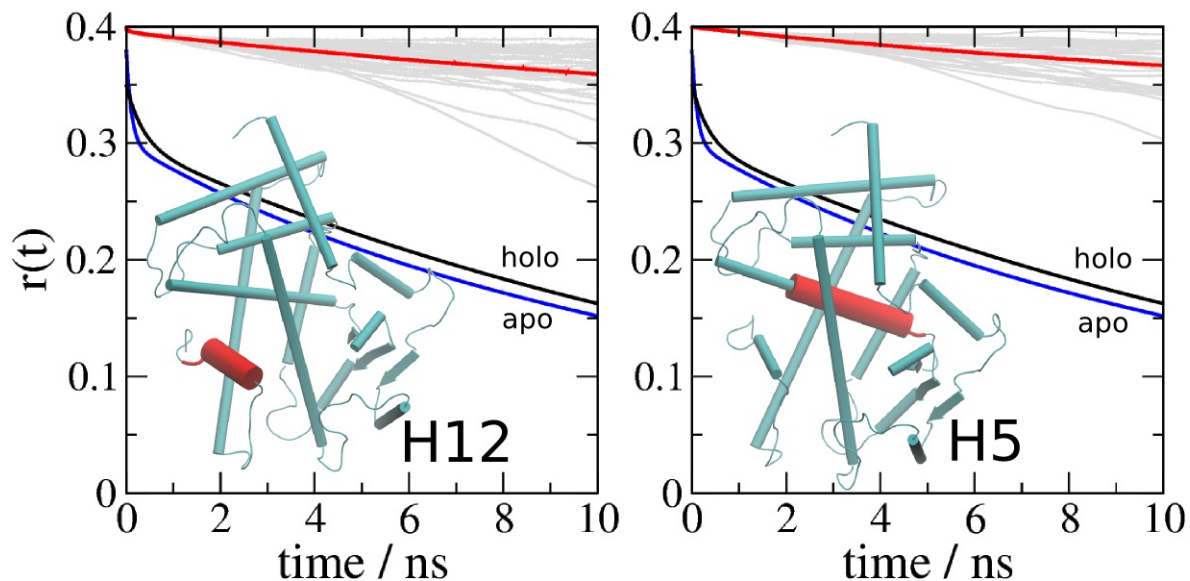## 1. Predicted anisotropy decays for H12 and H5

Figure S1. Reorientational dynamics of the Helix 12 (H12) and of Helix 5 (H5). H5 is a rigid helix buried in the protein core, which reorientates only as a function of protein overall rotation. The reorientational dynamics of these helices is mostly determined by protein tumbling, and are both much slower than the experimentally observed decay rates for holo and apo-receptors on short time-scales (black and blue curves). This is consistent with the interpretation that the experimental data is dependent mostly on the propensity of the probe to be attached or detached from the protein surface. Within this interpretation, this data indicates that the fluorescent probe must be detached from the protein surface, and thus display fast reorientational motions, in a significant fraction of the time, as indicated by the multiple fitting of Figure 8 (main text). The r(t) functions were computed from the reorientation of the vectors connecting the Cα atoms of Pro467 and Lys474 for H12, and Lys114 and Leu129 for H5, the range of residues being depicted in red in the inset figure.

2. Parametrization of the Cysfluor fluorescent probe

The Cysteine-fluoresceine[1] probe was parametrized using a fragment based approach, to facilitate geometry optimization and convergence stability of ab-initio calculations. Quantum-chemical calculations were performed with Gaussian03[2] using the HF-631G(d,p) level of theory for charges, and MP2/6-31G(d,p) for dihedral calculations. Charges were computed for the optimized geometries using the Merz-Kollman charge model,[3] for the fragments of the Cysfluor probe which are represented in Figure S2(B-D).
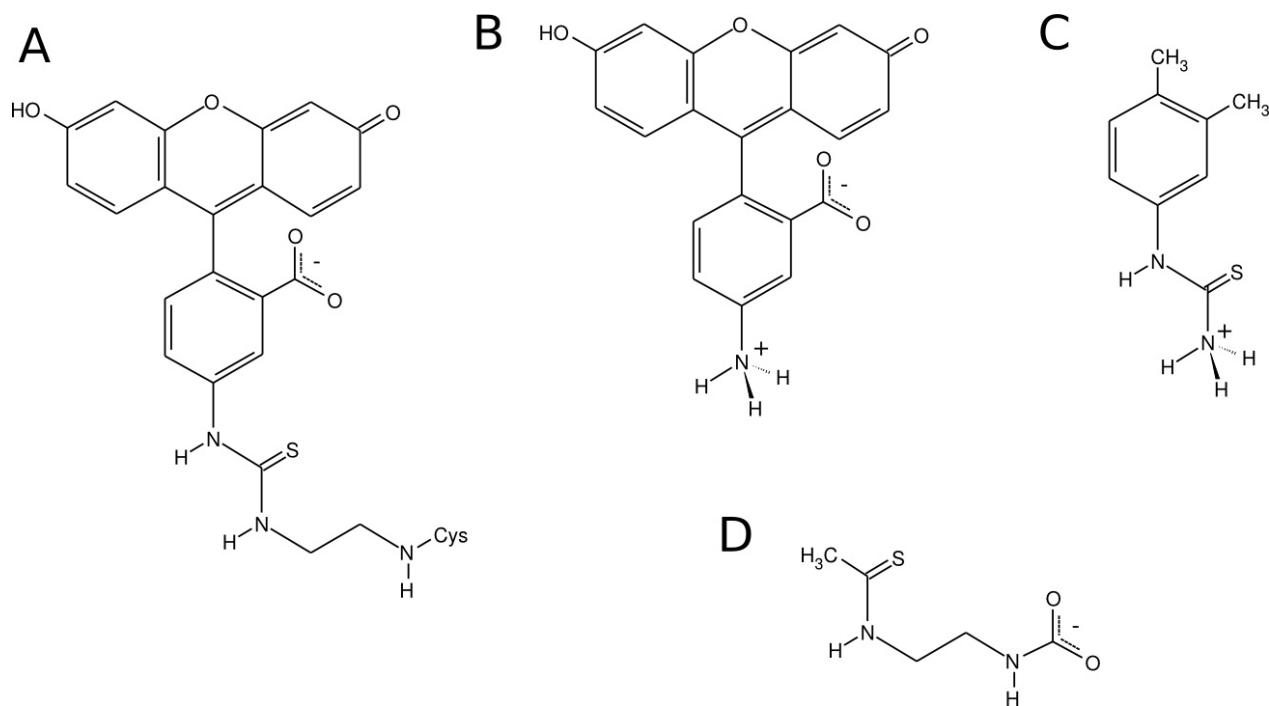

Figure S2. (A) Cysteine-fluoresceine residue and the (B-D) the fragments which were used for computing partial atom charges in quantum-mechanical calculations.

Charges for the whole Cysteine-fluorescein residue were obtained from the fragment calculations, except for the N-terminal nitrogen and hydrogen atoms, for which charges were copied from the corresponding N and HN atoms of amino-acid residues of the CHARMM27 force-field. Finally, all atom charges were minimally tuned to adjust the charge of the full Cysfluor residue. All charges are available in Supplementary Information 3, including the fragment-charges obtained from QM calculations.

We also parametrized three dihedral angles, for which we found no counterpart in the CHARMM27 set. The three angles are depicted in Figure S3A. For the parametrization, we computed optimized geometries at the MP2/6-31G(d,p) level of theory. Each angle under study was fixed to a value varying within 0 and 360° in 10° steps.
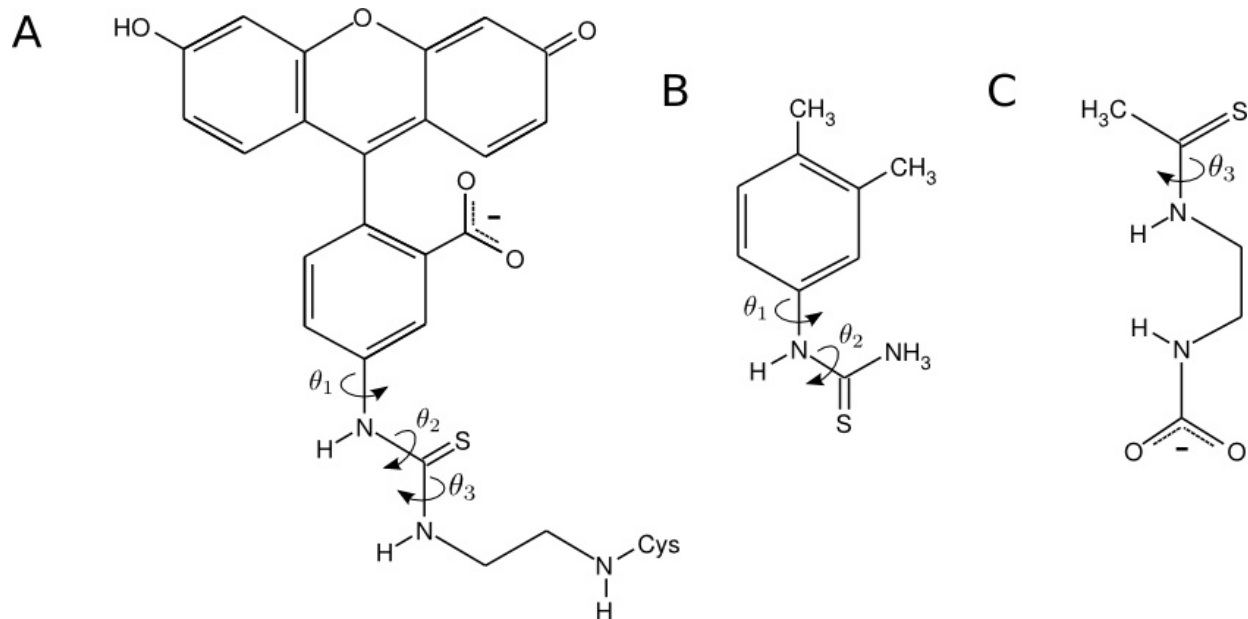

Figure S3. Molecular structure of (A) Cysfluor and (B) and (C) the molecular constructs used to compute the dihedral potentials which were not obtained from group analogy in the CHARMM set.

The QM energies were compared to the energies of the Molecular-Mechanics force field for the same geometries, and the force-field parameters of the dihedral angles were adjusted to obtain an optimal fit of the MM energies to the QM energies, according to standard parametrization procedures. The three dihedral angles requiring parametrization $(\theta_1, \theta_2$ and $\theta_3)$ were studied independently with *ab-initio* calculations of two different fragments of the CysFluor residue, represented in Figures S3B and C. QM and fitted MM energies as a function of each dihedral angle varied are represented in Figure S4. Rotations around all dihedral angles display quite high energy barriers (all greater than ~20 kcal mol^{-1}), mostly deriving from electronic delocalization, indicating that rotations will be restricted to the local minima defined by initial conditions at room temperature. All parameters are described in Supplementary Information Data 3.
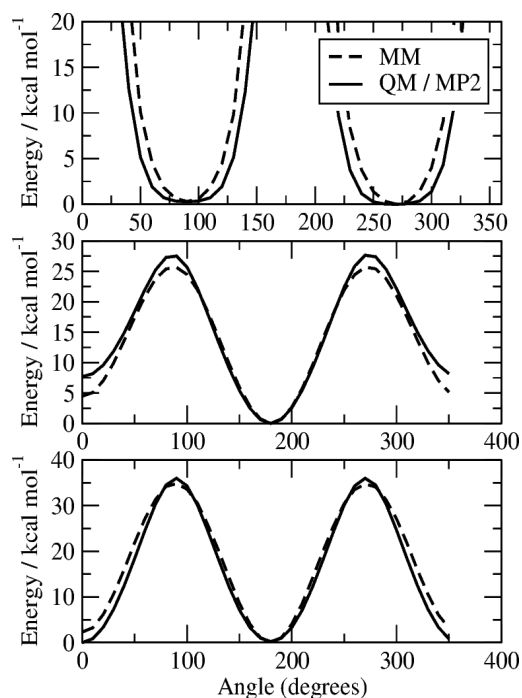
F**igure S4.** Overlap of the Quantum-Mechanical (QM) and Molecular-Mechanics (MM) energies as computed from ab-initio calculations and the adjusted CHARMM force field for the rotation of the parametrized dihedral angles.

These parameters were validated by reproducing the time-resolved anisotropy decay of the CysFluor probe free in solution, as shown in Figure S5. The anisotropy decays of the CysFluor probe were computed independently from 30 different 10 ns MD simulations following the same protocols as described in the main text, and the black line in Figure S5 represents the average decay obtained (the very short-time behavior of the experimental data is not reproduced because it is not exponential, which means that the experiment was not able to probe the anisotropy with that resolution).
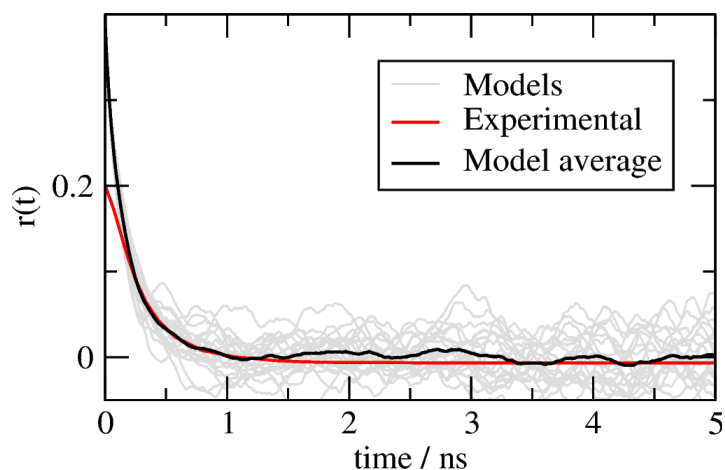
Figure S5. Reproduction of the time-resolved anisotropy decay of the CysFluor probe free in solution (experimental results from ref. 1). The experiment clearly did not capture the short-time scale decay, as it cannot be modeled as a sum of exponentials. The reproduction of the decay rates in intermediate (greater than 0.3 ns) and long time-scales is good, validating the parametrization of the probe and the simulation.

3. CHARMM Topologies and Force-Field for Cysteine-Fluoresceine

Data 1. Topology, atom types and and charges

```
* CHARMM topology file for Cysteine-Fluoresceine
*
27 1


!
! The whole CysFluor molecule
!

RESI CYFL  -1.00
ATOM  C01   CA     0.34 !
ATOM  C02   CA    -0.45 !        H07              H06
ATOM  C03   CA     0.46 !         |                |
ATOM  C04   CA    -0.45 !  O04    C20     O03     C16     O05-H10
ATOM  C05   CA    -0.29 !    \\  /  \\   /   \   /  \\   /
ATOM  C06   CA    -0.27 !     C19     C11     C10     C15
ATOM  C07   CC     0.64 !      |       |       ||      |
ATOM  C08   CA     0.32 !     C18     C12     C09     C14
ATOM  C09   CA    -0.36 !    /  \\   /  \\   /   \  //   \
ATOM  C10   CA     0.68 !  H08    C17     C08     C13    H05
ATOM  C11   CA     0.59 !         |               |
ATOM  C12   CA    -0.26 !        H09              H04
ATOM  C13   CA     0.09 !         |                 O02        (TO PROTEIN C-TER)
ATOM  C14   CA    -0.56 !         |               //      -----|----------------
ATOM  C15   CA     0.73 !  H03    C04     C07  (-)  |  HN-N                      |
ATOM  C16   CA    -0.80 !    \   //  \   /    \     |    |     HB1               |
ATOM  C17   CA     0.01 !     C05     C03    O01    |    |     |                 |
ATOM  C18   CA    -0.52 !      |       ||           |  HA-CA--CB--SG   |-> Cysteine
ATOM  C19   CA     0.96 !     C06     C02           |    |     |    \  |  Not included
ATOM  C20   CA    -0.79 !    /  \\   /  \           |    |     HB2   HG1|
ATOM  O01   OC    -0.69 !  H02     C01    H01       |   O=C             |
ATOM  O02   OC    -0.69 !          |                 ----/----------------
ATOM  O03   OH1   -0.41 !         N01       H12 H13 H15  /
ATOM  O04   OS    -0.66 !        /   \       |   |   |  /
ATOM  O05   OH1   -0.67 !     H11      C21-N02- C -CA-N
```
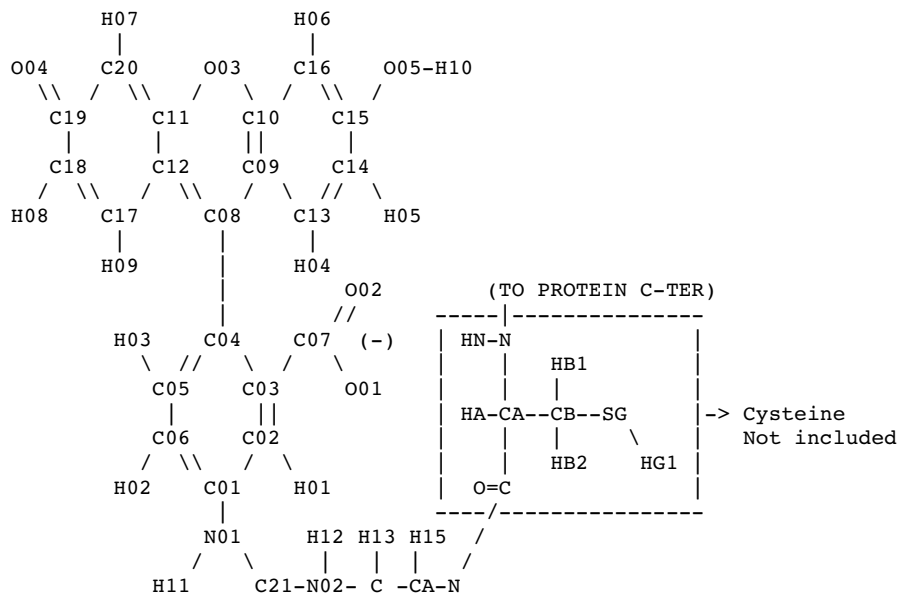
```
ATOM  N01   NH2  -0.37 !                                        ||       |  |   \
ATOM  H01   HP    0.23 !                                       S01     H14 H16  HN
ATOM  H02   HP    0.19 !
ATOM  H03   HP    0.11 !
ATOM  H04   HP    0.13 !
ATOM  H05   HP    0.23
ATOM  H06   HP    0.25
ATOM  H07   HP    0.24
ATOM  H08   HP    0.18
ATOM  H09   HP    0.14
ATOM  H10   H     0.45
ATOM  C21   C     0.24
ATOM  N02   NH2  -0.16
ATOM  S01   S    -0.50
ATOM  H11   H     0.34
ATOM  H12   H     0.21
ATOM  C     CT2  -0.31
ATOM  CA    CT2   0.48
ATOM  H13   HA    0.11
ATOM  H14   HA    0.11
ATOM  HA    HA   -0.04
ATOM  H16   HA   -0.04
ATOM  N     NH1  -0.47
ATOM  HN    H     0.30


BOND N    HN  N    CA  CA   HA  CA  H16 CA  C    C   H13 C    H14
BOND C    N02 N02 H12 N02 C21 C21 S01 C21 N01 N01 H11 N01 C01
BOND C01 C02 C02 H01 C02 C03 C03 C07 C07 O02 C07 O01 C03 C04
BOND C04 C05 C05 H03 C05 C06 C06 H02 C06 C01
BOND C04 C08 C08 C09 C09 C10 C09 C13 C13 H04 C13 C14 C14 H05
BOND C14 C15 C15 O05 O05 H10 C15 C16 C16 H06 C16 C10 C10 O03
BOND O03 C11 C11 C12 C11 C20 C20 H07 C20 C19 C19 O04 C19 C18
BOND C18 H08 C18 C17 C17 H09 C17 C12 C12 C08
IMPR C07 C03 O01 O02
IMPR N01 H11 C01 C21
IMPR C21 S01 N01 N02

PATCH FIRST NTER LAST NONE


!
! The CysFluor ring part
!

RESI RING  0.00
ATOM  C01   CA     0.556  !
ATOM  C02   CA    -0.626  !          H07              H06
ATOM  C03   CA     0.471  !           |                |
ATOM  C04   CA    -0.454  !  O04  C20     O03  C16    O05-H10
ATOM  C05   CA     0.094  !   \\  /  \\   /  \  /  \\  /
ATOM  C06   CA    -0.517  !    C19    C11    C10    C15
ATOM  C07   CC     0.651  !     |      |      ||      |
ATOM  C08   CA     0.326  !    C18    C12    C09    C14
ATOM  C09   CA    -0.359  !    / \\   /  \\  /  \  // \
ATOM  C10   CA     0.687  !  H08    C17    C08    C13    H05
ATOM  C11   CA     0.595  !          |      |      |
ATOM  C12   CA    -0.257  !         H09     |     H04
ATOM  C13   CA     0.096  !                 |            O02
ATOM  C14   CA    -0.558  !                 |           //
ATOM  C15   CA     0.736  !        H03    C04    C07  (-)
ATOM  C16   CA    -0.801  !          \  //   \  /   \
ATOM  C17   CA     0.015  !          C05    C03    O01
ATOM  C18   CA    -0.521  !           |      ||
ATOM  C19   CA     0.968  !          C06    C02
ATOM  C20   CA    -0.788  !          /  \\  /  \
ATOM  O01   OC    -0.737  !        H02    C01    H01
ATOM  O02   OC    -0.635  !                |
ATOM  O03   OH1   -0.412  !              N01(+)
ATOM  O04   OS    -0.663  !              /  |  \
ATOM  O05   OH1   -0.669  !           H11   |   HX1
ATOM  N01   NH3   -0.758  !                HX2
ATOM  H01   HP     0.260  !
ATOM  H02   HP     0.229  !
ATOM  H03   HP     0.123  ! Obs: *X* atoms do not belong to the
```

```
ATOM  H04   HP        0.136   !        complete cys-fluor
ATOM  H05   HP        0.239   !
ATOM  H06   HP        0.263
ATOM  H07   HP        0.251
ATOM  H08   HP        0.189
ATOM  H09   HP        0.145
ATOM  H10   H         0.459
ATOM  H11   HC        0.418
ATOM  HX1   HC        0.415
ATOM  HX2   HC        0.433


BOND C01 C02 C02 C03 C03 C04 C05 C06 C06 C01 C04 C05
BOND C01 N01 C02 H01 C03 C07 C04 C08 C05 H03 C06 H02
BOND N01 H11 N01 HX1 N01 HX2
BOND C07 O02 C07 O01
BOND C08 C09 C09 C10 C10 O03 O03 C11 C11 C12 C12 C08
BOND C09 C13 C10 C16 C11 C20 C12 C17
BOND C13 H04 C13 C14 C14 H05 C14 C15 C15 O05 O05 H10
BOND C15 C16 C16 H06
BOND C20 H07 C20 C19 C19 O04 C19 C18 C18 H08 C18 C17 C17 H09
IMPR C07 C03 O01 O02

PATCH FIRST NONE LAST NONE


!
! The CysFluor sulfamide part:
!

RESI SLMD 1.00
ATOM  C01   CN2       0.347   !
ATOM  C02   CA       -0.448   !
ATOM  C03   CA        0.149   !              HX6 HX5 HX4
ATOM  C04   CA        0.175   !                \  |  /
ATOM  C05   CA       -0.286   !                 CX1      HX1
ATOM  C06   CA       -0.274   !                  |        |
ATOM  C07   CT3      -0.301   !        H03      C04      C07-HX2
ATOM  C21   C         0.049   !          \    //   \   /   \
ATOM  N01   NH2      -0.366   !           C05      C03     HX3
ATOM  N02   NH3      -0.046   !           |        ||
ATOM  S01   S        -0.151   !          C06      C02
ATOM  H01   HP        0.241   !         /   \\   /   \
ATOM  H02   HP        0.205   !       H02    C01      H01
ATOM  H03   HP        0.220   !               |
ATOM  H11   H         0.346   !              N01         H12
ATOM  H12   HC        0.276   !             /   \   (+) /
ATOM  CX1   CT3      -0.397   !           H11    C21-N02-HX7
ATOM  HX1   HA        0.126   !                  ||    \
ATOM  HX2   HA        0.104   !                  S01    HX8
ATOM  HX3   HA        0.104   !
ATOM  HX4   HA        0.132   !
ATOM  HX5   HA        0.150   !   Obs: *X* atoms do not belong to the
ATOM  HX6   HA        0.126   !        complete cys-fluor
ATOM  HX7   HC        0.268
ATOM  HX8   HC        0.251


BOND N02 H12 N02 HX7 N02 HX8
BOND N02 C21 C21 S01 C21 N01 N01 H11
BOND N01 C01 C01 C02 C02 C03 C03 C04 C04 C05 C05 C06 C06 C01
BOND C02 H01 C03 C07 C07 HX1 C07 HX2 C07 HX3
BOND C04 CX1 CX1 HX6 CX1 HX5 CX1 HX4
BOND C05 H03 C06 H02
IMPR N01 H11 C01 C21
IMPR C21 S01 N01 N02

PATCH FIRST NONE LAST NONE


!
! Cys-fluor: the tail part
!

RESI TAIL -1.00
ATOM  C21   C         0.255   !
```
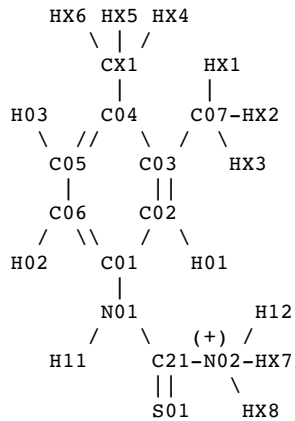
```
ATOM  C22   CT2    -0.315  !    HX1 HX2                              O06
ATOM  C23   CT2     0.492  !      \ |                               /
ATOM  C24   CC      1.014  !       CX1      H12 H13 H15      C24 (-)
ATOM  S01   S      -0.498  !      /  \       |   |   |      /  \
ATOM  O06   OC     -0.840  !    HX3    C21-N02-C22-C23-N03     OX1(-to CYS)
ATOM  N02   NH2    -0.155  !           ||        |   |   |
ATOM  N03   NH2    -0.831  !           S01      H14 H16 H17
ATOM  H12   H       0.224  !
ATOM  H13   HA      0.118  !
ATOM  H14   HA      0.118  !   Obs: *X* atoms do not belong to the
ATOM  H15   HA     -0.041  !        complete cys-fluor
ATOM  H16   HA     -0.041
ATOM  H17   H       0.335
ATOM  CX1   CT3    -0.303
ATOM  OX1   OC     -0.847
ATOM  HX1   HA      0.112
ATOM  HX2   HA      0.112
ATOM  HX3   HA      0.091


BOND CX1 C21 C21 N02 N02 C22 C22 C23 C23 N03 N03 C24
BOND CX1 HX1 CX1 HX2 CX1 HX3
BOND C21 S01 N02 H12 C22 H13 C22 H14 C23 H15 C23 H16
BOND N03 H17 C24 O06 C24 OX1
IMPR C24 N03 O06 OX1

PATCH FIRST NONE LAST NONE
```

Data 2. Bonds, angles and dihedrals

```
* CHARMM parameter file Cysteine-Fluoresceine
*

NONBONDED nbxmod  5 atom cdiel shift vatom vdistance vswitch -
cutnb 14.0 ctofnb 12.0 ctonnb 10.0 eps 1.0 e14fac 1.0 wmin 1.5
                !adm jr., 5/08/91, suggested cutoff scheme
!
!V(Lennard-Jones) = Eps,i,j[(Rmin,i,j/ri,j)**12 - 2(Rmin,i,j/ri,j)**6]
!
!epsilon: kcal/mole, Eps,i,j = sqrt(eps,i * eps,j)
!Rmin/2: A, Rmin,i,j = Rmin/2,i + Rmin/2,j
!
!atom  ignored    epsilon     Rmin/2    ignored    eps,1-4      Rmin/2,1-4
!

BONDS
!
!V(bond) = Kb(b - b0)**2
!
!Kb: kcal/mole/A**2
!b0: A
!
!atom type     Kb          b0
!
CA    NH3   230.0      1.488  ! b0 from optimal 631g(d,p), Kb from CT3 CA
CA    NH2   230.0      1.488  ! b0 from optimal 631g(d,p), Kb from CT3 CA
CA    CC    230.0      1.565  ! b0 from optimal 631g(d,p), Kb from CT3 CA
CA    OS    334.3      1.206  ! b0 from optimal 631g(d,p), Kb from OH1 CA
CA    CN2   305.0      1.440  ! b0 from optimal 631g(d,p), Kb from CA CA
C     S     300.0      1.620  ! b0 from optimal 631g(d,p), Kb from well intuition
C     NH2   430.0      1.320  ! b0 from optimal 631g(d,p), Kb CC NH2
CN2   NH2   430.0      1.440  ! b0 from optimal 631g(d,p), Kb CC NH2
C     NH3   430.0      1.500  ! b0 from optimal 631g(d,p), Kb CC NH2

ANGLES
!
!V(angle) = Ktheta(Theta - Theta0)**2
!
!V(Urey-Bradley) = Kub(S - S0)**2 !
!Ktheta: kcal/mole/rad**2
!Theta0: degrees
```

```
!Kub: kcal/mole/A**2 (Urey-Bradley)
!S0: A
!
!atom types          Ktheta    Theta0   Kub      S0
!
!
CA   NH3  HC      49.3     109.5     0.0    0.000  ! tetha0 from 631g(d,p), Kb from HA CT3 CA
CA   NH2  H       49.3     109.5     0.0    0.000  ! tetha0 from 631g(d,p), Kb from HA CT3 CA
CA   CA   CC      45.8     116.3     0.0    0.000  ! tetha0 from 631g(d,p), Kb from CT3 CA CA
CA   CA   NH3     45.8     117.8     0.0    0.000  ! tetha0 from 631g(d,p), Kb from CT3 CA CA
CA   CA   NH2     45.8     117.8     0.0    0.000  ! tetha0 from 631g(d,p), Kb from CT3 CA CA
CA   CC   OC      40.0     114.1     0.0    0.000  ! tetha0 from 631g(d,p), Kb from OC CC CT3
CA   OH1  CA      40.0     122.2     0.0    0.000  ! tetha0 from 631g(d,p), Kb from CA CA CA
CA   CA   OS      40.0     122.9     0.0    0.000  ! tetha0 from 631g(d,p), Kb from CA CA CA
CN2  CA   HP      30.000   120.00    22.00   2.15250 ! from CA CA HP
CA   CA   CN2     45.800   122.3000 ! from CA CA CT3
CA   CN2  CA      40.0     117.6     0.0    0.000  ! tetha0 from 631g(d,p), Kb from CA CA CA
C    NH2  CN2     50.0     126.5     0.0    0.000  ! tetha0 from 631g(d,p), Kb from CC NH2 H
C    NH2  CA      50.0     126.5     0.0    0.000  ! tetha0 from 631g(d,p), Kb from CC NH2 H
CA   CN2  NH2     40.0     119.4     0.0    0.000  ! tetha0 from 631g(d,p), Kb from CA CA CA
NH2  C    NH3     50.000   111.3     0.0    0.000  ! tetha0 from 631g(d,p), Kb from NH1 CT1 C
NH2  C    NH2     50.000   111.3     0.0    0.000  ! tetha0 from 631g(d,p), Kb from NH1 CT1 C
CN2  NH2  H       50.0     118.8     0.0    0.000  ! tetha0 from 631g(d,p), Kb from NH1 CT1 C
C    NH3  HC      50.0     108.0     0.0    0.000  ! tetha0 from 631g(d,p), Kb from NH1 CT1 C
C    NH2  H       50.0     114.7     0.0    0.000  ! tetha0 from 631g(d,p), Kb from CC NH2 H
NH2  C    S       50.0     131.0     0.0    0.000  ! tetha0 from 631g(d,p), Kb from NH1 CT1 C
NH3  C    S       50.0     109.7     0.0    0.000  ! tetha0 from 631g(d,p), Kb from NH1 CT1 C
C    NH2  CT2     50.0     127.2     0.0    0.000  ! tetha0 from 631g(d,p), Kb from CC NH2 H
CT1  NH2  HA      50.0     117.3     0.0    0.000  ! tetha0 from 631g(d,p), Kb from CC NH2 H
CC   NH2  CT2     50.0     121.9     0.0    0.000  ! tetha0 from 631g(d,p), Kb from CC NH2 H
NH2  CC   OC      40.0     114.3     0.0    0.000  ! tetha0 from 631g(d,p), Kb from CC NH2 H
CT3  C    S       40.0     121.3     0.0    0.000  ! tetha0 from 631g(d,p), Kb from CA CA CA
CT3  C    NH2     40.0     113.9     0.0    0.000  ! tetha0 from 631g(d,p), Kb from CA CA CA
HA   CT1  NH3     50.0     117.3     0.0    0.000  ! tetha0 from 631g(d,p), Kb from CC NH2 H
CT1  CT2  NH2     40.0     113.9     0.0    0.000  ! tetha0 from 631g(d,p), Kb from CA CA CA
HA   CT1  HB      36.000   115.0000 ! ALLOW    PEP
CT2  NH2  H       50.0     117.3     0.0    0.000  ! tetha0 from 631g(d,p), Kb from CC NH2 H


DIHEDRALS
!
!V(dihedral) = Kchi(1 + cos(n(chi) - delta))
!
!Kchi: kcal/mole
!n: multiplicity
!delta: degrees
!
!atom types            Kchi    n   delta
!

CA   CA   CA   CC     3.10    2   180.00  ! From CA CA CA CA
CA   CA   CC   OC     0.23    2   180.00  ! From CT3 CT2 CA CA
CA   CA   NH3  HC     0.99    2   180.00  ! From H OH1 CA CA
CA   CA   CA   NH3    3.10    2   180.00  ! From CA CA CA CA and analogs
CA   CA   CA   NH2    3.10    2   180.00  ! From CA CA CA CA and analogs
CC   CA   CA   HP     4.20    2   180.00  ! From HP CA CA CT3
CA   CA   OH1  CA     3.10    2   180.00  ! From CA CA CA CA
CA   CA   CA   OS     3.10    2   180.00  ! From CA CA CA CA
OS   CA   CA   HP     4.20    2   180.00  ! From HP CA CA CT3
NH3  CA   CA   HP     4.20    2   180.00  ! From HP CA CA CT3
NH2  CA   CA   HP     4.20    2   180.00  ! From HP CA CA CT3
CN2  CA   CA   CA     3.10    2   180.00  ! From CA CA CA CA
CA   CN2  CA   CA     3.10    2   180.00  ! From CA CA CA CA
CN2  CA   CA   HP     4.20    2   180.00 ! From CA CA CA HP
CN2  CA   CA   CT3    4.20    2   180.00 ! From HP CA CA CT3
CA   CA   NH2  C      1.60    1   180.00  ! From CT3 C     NH1   CT1
CA   CN2  NH2  C      1.60    1   180.00  ! From CT3 C     NH1   CT1
CA   CA   NH2  C      1.60    1   180.00  ! From CT3 C     NH1   CT1
CA   CN2  NH2  H      1.60    1   180.00  !
CA   CA   NH2  H      1.60    1   180.00  !
CA   CN2  CA   HP     4.20    2   180.00 ! From CA CA CA HP
CA   CA   CN2  NH2    4.20    2   180.00 ! From CA CA CA HP
CT3  CA   CA   CT3    4.20    2   180.00 ! From HP CA CA CT3
NH2  C    NH3  HC     1.60    1   180.00  !
```

```
NH2  CN2  CA   HP        0.99   2   180.00  ! From H OH1 CA CA
S    C    NH3  HC        1.60   2   180.00  !
C    NH2  CT2  CT2       0.20   1   180.00 ! ALLOW PEP
C    NH2  CT2  HA        0.00   3     0.00 ! ALLOW PEP
CT2  CT2  NH2  CC        2.50   2   180.00 !  ALLOW PEP
CT1  CT2  NH2  C         2.50   2   180.00 !  ALLOW PEP
CT2  CT2  NH2  H         0.99   2   180.00  ! From H OH1 CA CA
CT1  CT2  NH2  H         0.99   2   180.00  ! From H OH1 CA CA
CT2  NH2  CC   OC        3.20   2   180.00 ! ALLOW PEP PRO
CC   NH2  CT2  HA        0.1600 3     0.00 ! ALLOW PEP PRO POL
S    C    CT3  HA        3.23   2   180.00 !
OC   CC   NH2  H         3.20   2   180.00 ! ALLOW PEP PRO
NH2  C    CT3  HA        1.60   1   180.00
H    NH2  CT2  HA        1.60   1   180.00
! The following dihedrals had to be parametrized by fitting the
! corresponding HF/631G(d,p) QM energy curve

NH3  C    NH2  H         3.23   2   180.00  !
S    C    NH2  H         3.23   2   180.00  !
CN2  NH2  C    NH3       3.23   2   180.00  ! From CT3  C     NH1  CT1
CA   NH2  C    NH2       3.23   2   180.00  ! From CT3  C     NH1  CT1
CN2  NH2  C    S         3.23   2   180.00  ! From CT3  C     NH1  CT1
CA   NH2  C    S         3.23   2   180.00  ! From CT3  C     NH1  CT1
H    NH2  C    CT3       4.65   2   180.00
CT2  NH2  C    CT3       4.65   2   180.00 !  ALLOW PEP
CT2  NH2  C    S         4.65   2   180.00  ! From CT3  C     NH1  CT1
CT2  NH2  C    NH2       3.23   2   180.00  ! From CT3  C     NH1  CT1
NH2  C    NH2  H         3.23   2   180.00  !




!
!
IMPROPER
!
!V(improper) = Kpsi(psi - psi0)**2
!
!Kpsi: kcal/mole/rad**2
!psi0: degrees
!note that the second column of numbers (0) is ignored
!
!atom types            Kpsi                   psi0
!
NH2 H  CN2 C         20.0000       0        0.0000 ! ALLOW   PEP POL ARO
NH2 H  CA  C         20.0000       0        0.0000 ! ALLOW   PEP POL ARO
C   S  NH2 NH3       96.0000       0        0.0000 ! ALLOW   PEP POL ARO ION
C   S  NH2 NH2       96.0000       0        0.0000 ! ALLOW   PEP POL ARO ION

NONBONDED nbxmod  5 atom cdiel fshift vatom vdistance vfswitch -
cutnb 14.0 ctofnb 12.0 ctonnb 10.0 eps 1.0 e14fac 1.0 wmin 1.5
!
!V(Lennard-Jones) = Eps,i,j[(Rmin,i,j/ri,j)**12 - 2(Rmin,i,j/ri,j)**6]
!
!epsilon: kcal/mole, Eps,i,j = sqrt(eps,i * eps,j)
!Rmin/2: A, Rmin,i,j = Rmin/2,i + Rmin/2,j
!
!atom  ignored    epsilon     Rmin/2   ignored    eps,1-4       Rmin/2,1-4
!


END
```

Supporting References

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3. Singh, U. C. & Kollman, P. A. (1984). An approach to computing eletrostatic charges for molecules. *J. Comp. Chem.* **5**, 129-145.