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Traditional medicine protocol

Disease transmission
Investigation
Cause identification
Prevention
Diagnosis
Therapeutic plan
Public health policy
Vaccine
Resistance
Drugs
Knowledge-based
Evidence-based
Lab results
Computer assisted

Avian Influenza A (H5N1) in 10 Patients in Vietnam

The NEW ENGLAND JOURNAL OF MEDICINE

Avian Influenza A (H5N1) in 10 Patients in Vietnam

Thi Tram, M.D., Nguyen Pham Linh, M.D., Nguyen Thi Dai, M.D., Lam Thi Hue, M.D., Nguyen Thi Thu, M.D., Le Thi Phuong Nga, M.D., Tran Thi Thanh, B.E., and Vo Thi Dinh, M.D.

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Now let’s get back to the basic...
Maximum parsimony algorithm for finding minimum number of mutations on a tree

Ancestor:

---

Rat Mouse Human Chicken
ATGACA ATGACA ACCAAA ACCCAT

Tree A

Eight mutations

---

Tree B

Five mutations

---

Sites 819-824 in cytochrome oxidase 1

Chicken ACCCAT
Mouse ATGACA
Rat ATGACA
Human ACCCAA

---

Rat Mouse Human Chicken
ATGACA ATGACA ACCAAA ACCCAT

Tree B

Eight mutations

---

Rat Mouse Human Chicken
ATGACA ATGACA ACCAAA ACCCAT

Tree A

Eight mutations

---

Reliable / Accuracy?
Probable transmission of HIV from orthopedic surgeon to patients in France

- In July 1995, an orthopedic surgeon practicing in the hospital of Saint-Germain-en-Laye had been accidentally infected with HIV while performing surgery 12 years earlier.
- In September 1995, he asked the director to screen all of his former patients and informed the press of his request.
- In October 1995, the French Ministry of Health offered HIV testing to patients who had been operated on by the surgeon.
- Letters were mailed to 3004 patients (May 1983 to October 1993)
  - 1218 (41%) patients responded, 218 were reported by their families to have died and had no info on sero status, 983 (33%) reported HIV test results, 982 were negative for HIV and 1 was positive for HIV.

Probable transmission of HIV from orthopedic surgeon to patients in France

- A 53-year-old surgeon received a diagnosis of AIDS in March 1994
- He stopped performing surgery in October 1993
- He was married, had only heterosexual contacts with his wife, denied using injection drugs, never received a blood tx, no history of STD.
- In May 1983, he had a needle stick injury while operating on a patient who had a multiple transfusions. This patient later died
  - 30 days after an accident, he developed a symptoms consistent with the HIV seroconversion syndrome
  - The surgeon reported frequent opportunities for blood exposure as well as frequent actual blood exposures
  - He often tightened suture wires with his fingers and sometimes tied sutured with the needle still attached percutaneous injuries with sharp objects previous in contact with blood occurred as frequently as once a week.
  - These injuries sometimes (but rarely) involved blood from an HIV infected patient.

Probable transmission of HIV from orthopedic surgeon to patients in France

- A 67-year-old French woman who had undergone an invasive procedure in June 1992 (placement of a total hip prosthesis with bone graft)
  - She was widowed in 1986 and had no sexual intercourse a few years before the death of her husband.
  - The donors of the 2 units of packed RBC also tested negative for HIV (one in April 1994 and one in May 1994)

Transmission of HIV to a patient during an invasive dental procedure

CBS' news program "60 Minutes" revisited the case of Jensen Beach, Florida, dentist David Acer a case that came to the nation's attention in 1990 when one of Dr. Acer's patients, 23-year-old college student Kimberly Bergalis, went public with her story. Both died of AIDS: Acer in 1990 and Bergalis in 1991.

At least 5 patients involved

Patient A: young college student woman
Patient B: elderly woman, married > 25 yr, her spouse tested -ve for HIV Ab
Patient C: young man, multiple heterosexual partners (all HIV seronegative)
Patient D: a man with AIDS with established risk factors for HIV infection
Patient E: a woman whose epidemiologic and lab tests has not been completed
Patient F: Patient G

All denied sexual contact with the dentist.
The viruses of the dentist and patients A, B and C are closely related in their V3 sequences with an average difference of 3.4%.
The average viral sequence difference for patient D and seven control patients was approximately 13% (range 8% - 15%).

Now we are going to find a magic bullet.

Cytomegalovirus retinitis is most common ocular opportunistic infection in immunocompromised or immunosuppressed patients, i.e. patients with AIDS or organ transplant recipients.
In AIDS patients, it usually seen in patients with CD4+ < 50 per mm$^3$
Clinical features: May be asymptomatic, blurry or cloudy vision, floaters, light flashes, loss of central or peripheral vision, vitreous cells with mild anterior chamber inflammatory reaction.
Management: Intravenous anti-viral agents such as gancyclovir, cidofovir and foscarnet which may be used individually or in combination. Alternative intravitreal gancyclovir implant (the effect lasts in 6-10 months)
Virtual screening of HIV-1 protease inhibitors against human cytomegalovirus protease using docking and molecular dynamics

Research question:


Virtual screening of HIV-1 protease inhibitors against human cytomegalovirus protease using docking and molecular dynamics

Substrate binding site of CMV protease

Virtual screening of HIV-1 protease inhibitors against human cytomegalovirus protease using docking and molecular dynamics

List of six HIV-1 protease inhibitors

Virtual screening of HIV-1 protease inhibitors against human cytomegalovirus protease using docking and molecular dynamics

Binding modes/energies of HIV-1 protease inhibitors to the CMV protease

APV IDV LPV
NFV RTV SQV
-15.70 7.66x10^-9 -14.26 9.37x10^-9 -13.43 1.76x10^-6
-12.87 5.93x10^-7 -15.99 3.69x10^-7 -14.22 4.35x10^-8

Virtual screening of HIV-1 protease inhibitors against human cytomegalovirus protease using docking and molecular dynamics

Summary

- This study provides evidence for the inhibitory activity of two approved inhibitors, amprenavir and indinavir, against the CMV protease.
- Including either of these two inhibitors in a HAART regimen should help to control the CMV viral load in HIV-1-infected patients.
- The activity of the CMV protease would be inhibited soon after starting HAART, in contrast to inhibition by promoting immune system restoration, which may take several weeks.
- Structural studies of human herpes proteases indicates homology among several subtypes.

Virtual screening of HIV-1 protease inhibitors against human cytomegalovirus protease using docking and molecular dynamics

Identifying inhibitors of the SARS Coronavirus protease

Coronavirus Main Protease (S1Pr) Structure: Basis for Design of Anti-SARS Drugs

AG7088 may be a good starting point for drug development.
In vitro evidence showed that AG7088 failed to inhibit SARS CoV.
Protease (protein hydrolase) catalyze amide (peptide) bond hydrolysis in protein or peptide substrates:

\[
\text{R} - \text{R} - \text{O} - \text{N} = \text{C} - \text{C} - \text{N} - \text{H} + \text{H}_2\text{O} \rightarrow \text{R} - \text{R} - \text{O} - \text{N} = \text{C} - \text{C} - \text{N} - \text{H} + \text{H} - \text{N} - \text{C} - \text{O}
\]

acid
amine

Coronavirus cysteine protease has three important parts in their active site:

1) Specificity pocket a pocket which interacts with the side chains of specific amino acids
2) Catalytic dyad two residues, histidine and cysteine, are present in the active site
3) Oxyanion hole two free amide \(N-H\) groups available to donate hydrogen bonds
Identifying inhibitors of the SARS Coronavirus protease

Heptad-repeat-2 mutations enhance stability of enfuvirtide-resistant HIV-1 gp41 hairpin structure

T-20 resistance HR1 mutations
- G36D/S
- I37V
- V38A/E/M
- Q40H
- N42D/E/S/T
- N43D/K/S
- L45M
Heptad-repeat-2 mutations enhance stability of enfuvirtide-resistant HIV-1 gp41 hairpin structure

Mapping of HR1/HR2 residue-residue interactions

<table>
<thead>
<tr>
<th>HR1</th>
<th>Corresponding HR2</th>
</tr>
</thead>
<tbody>
<tr>
<td>G36</td>
<td>Q141 / N145</td>
</tr>
<tr>
<td>V38</td>
<td>Q139 / Q142</td>
</tr>
<tr>
<td>Q40</td>
<td>S138 / Q141</td>
</tr>
<tr>
<td>N42</td>
<td>Q139 / Q142</td>
</tr>
<tr>
<td>N43</td>
<td>L134 / S138</td>
</tr>
<tr>
<td>L45</td>
<td>S138 / Q139</td>
</tr>
</tbody>
</table>

RAPDF reference
- Residue-specific all-atom probability discriminatory function (RAPDF) score
- http://compbio.washington.edu/

Comparison of wild-type and mutant surface structures
Heptad-repeat-2 mutations enhance stability of enfuvirtide-resistant HIV-1 gp41 hairpin structure

Designing enfuvirtide derivatives against resistant strains

Prediction of HIV-1 protease inhibitor resistance using a protein-inhibitor flexible docking approach

HIV drug susceptibility testing background
- Monitoring of disease progression: CD4 count, viral load
- Emergence of antiretroviral drug resistant strains
- Monitoring of drug resistance: genotyping, phenotyping
- Phenotyping: expensive, takes time, direct, easy to interpret
- Genotyping: cheap, fast turn around time, indirect, interpretation problems
- Computational methods for genotypic susceptibility interpretation
- Knowledge based: needs large genotype/phenotype data for training
- Physic based: studies interaction between viral enzyme and drugs
- Accuracy of individual and combined methods

Prediction of HIV-1 protease inhibitor resistance using a protein-inhibitor flexible docking approach

Knowledge based machine learning: rule based, decision tree, SVM, ANN


Downloaded 25 HIV-1 protease-inhibitor complexes that their experimental binding energies are available. Missing atoms or residues were checked and fixed.

Water molecules and sodium-chloride ions were added until the shell was 10 Å thick.

Molecular dynamics simulations protocol

Protein-inhibitor docking protocol
- Protein-inhibitor binding energy calculation: AutoDock 3.0.5
  \[ \Delta G_{\text{binding}} = \Delta G_{\text{vdW}} + \Delta G_{\text{elec}} + \Delta G_{\text{bond}} \]
  - \( \Delta G_{\text{vdW}} \): 12-6 Lennard-Jones potential, dispersion/repulsion
  - \( \Delta G_{\text{elec}} \): Coulombic electrostatic potential
  - \( \Delta G_{\text{bond}} \): Directional 12-10 potential
- Set up the ligand & macromolecule:
  - Ligand: Add all hydrogens, compute charges
  - Distinguish aliphatic and aromatic carbons
  - Choose non & rotatable bonds
  - Macromolecule: Add polar H, assign charges
- Pre-compute AutoGrid Maps for all atom types
- Perform dockings of ligand to target
Prediction of HIV-1 protease inhibitor resistance using a protein-inhibitor flexible docking approach

Correlation between experimentally-determined and calculated binding energy

Docking only 0.01 ps MD + Docking

0.1 ps MD + Docking 1 ps MD + Docking

0.38 0.53

0.87 0.79

Accuracy of each method

Accuracy of the consensus

Coverage SCWRL and SCWRL v2.95

Docking with dynamics Protocol

Mutation of the side chain

Wild type crystal structure

Mutant structure

Side chain replacement

Query Sequence

Template

Docking with dynamics protocol

Accuracy of the consensus

Docking with dynamics

Rule-based

SVM

Consensus

Prediction of HIV-1 protease inhibitor resistance using a protein-inhibitor flexible docking approach

Prediction of HIV-1 protease inhibitor resistance using a protein-inhibitor flexible docking approach
Prediction of HIV-1 protease inhibitor resistance using a protein-inhibitor flexible docking approach

http://protinfo.compbio.washington.edu/pirspred/

Application of Computational Biology in Veterinary Medicine

Traditional medicine (and prevention) protocol

- Disease transmission
- Investigation
- Prevention
- Diagnosis
- Therapeutic plan
- Drugs
- Vaccine
- Resistance
- Public health
- Policy

Knowledge-based
Evidence-based
Lab results
Computer assisted

References

- http://compbio.washington.edu/cv.html