FROM BASIC PRINCIPLES TO
COMPUTATIONALLY REFINED MODELS FOR A PRACTIC SYNTHESIS
OF THE NANO-COMPETENT POLYMERIC ANTIVIRALS

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Comments

● Good day, Dear Colleagues 
● With thankfulness for this opportunity. 
● I would like consider some aspects of not computer but the ‘own human brain-aided’ generation of fundamental strategies for design, synthesis, and testing the antiviral Drugs. 
● And after focusing on the key problems we can search their solutions using the computer-aided modeling capacity

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ANTIVIRAL DRUG DEVELOPMENT

Accumulated Basic Knowledge ↔ fundamentally predictable principles for

Targets identification ↔ Design ↔ Antivirals prognosis

Computer-Aided Modeling

Detailing - Clarifying the mechanisms & parameters for:
- **Viral Target** – Antiviral interaction in QSAR aspects
- **Synthesis optimization** toward the desired structures of the Antivirals

Bio Testing

Experimental verification of the
- **Design & Modeling Prognosis efficiency** or discovery of unpredicted data as new objects for analysis and reinvestigation
- **Prospects** of the **synthesized test-variants** for potential Antivirals for future biomedical advancement

Synthesis

- Realization of desired draggable (Macro) molecular constructs
- Stepwise preparation of drug-candidate samples for (bio) evaluations

Comments

- It can be summarized by the scheme. From basic knowledge supported Design toward pilot synthesis – bio-testing, and computational modeling if it is relevantly helpful
ANTIVIRAL DRUG DESIGN

1. FUNDAMENTALLY PREDICTABLE BASIC PRINCIPLES

Comments

- So, the Design and fundamentally predictable basic principles
First of all, small and macro molecules. The biologic life existence and evolution is naturally progressing from small toward great molecular forms. Under the temperature of our Planet only Polymeric chains and networks can covalently accumulate energy sufficient for stable resistance against chaotic dissociation. They capable of separate self-organizing up to bio life basis, starting from lipides to polysaccharides, proteins and nucleic acids.

Temperature & entropy stimulated dissociation & chaos

≈ 300 K (30°C) ± 50
1. FUNDAMENTALLY PREDICTABLE BASIC PRINCIPLES

1.1. Small and Macro Molecules

Molecular evolution of biological forms of Life

Covalently polymerized chains & networks
flexible, but stable self-organization

Extracellular Viral Particles =
max compressed inter-bio-polymeric complexes

HIV virion

Proteins with lipid envelope
RNA (2 copies) genome
Proteins replicated
Enzyme
Capsid
Spike

● Lipides
● Proteins
● Hydrocarbons
● Nucleic Acids

Comments

- The unique compact concentrates of these vital biopolymers, without small molecules ballast, represents extracellular viral particles, the virions. ......They are in fact the maximally compressed inter-bio-polymeric nano-complexes.
ANTIVIRAL DRUG DESIGN

1. FUNDAMENTALLY PREDICTABLE BASIC PRINCIPLES

1.1. Small and Macro Molecules

In view for counter intervention in Viral Life Cycle

Small Size ≤ 1 nm

Nano Scale ≥ 2 - 500 nm

Targets Stages

The Sub-nano sites of:

- Virions’ surface local points
- Virions’ surface full scale
- Virion Spikes*
- Cell Receptors*
- Passage of Ion Channels
- Cell membrane Raft –domains

The Nano objects:

- Virions’ surface full scale
- Virion Spikes
- Cell Receptors
- Cell membrane
- Virion Spikes
- Nucleic Acids & complexes
- Assembling pre-virion nano-units
- Cell membrane Raft –domains

Extracellular Viral Particles (Virions)

Virions Entry into Cells, and Uncoating

Intracellular Replication of Viral Species

Virions Assembly, Maturation, Release

Priority for the Poly-Antivirals nano-intervention

This fact is very important in view for any therapeutic counter intervention in a viral life cycle, which could be divided conditionally into 4 stages: I – extracellular Virions, II – Entry into Cell, III – Intracellular replication, and IV – assembly, maturation and release of new virions. Only the III stage involves small molecules as metabolites for biosynthesis, While other stages dominantly supported by biopolymeric macromolecules and their nano-complexes. Therefore, the stages I, II, and IV can and should be natural priority for adequate neutralization by exactly macromolecular scale drugs, that we named as “PolyAntivirals”. Just that is our general strategy for development of the PolyAntivirals.
Some Basic Principles/Criteria
toward
Poly-ANTIVIRALS
Design – Practical Synthesis

Comments
● The first task is formulation and implementation of same relevant principles and criteria for stepwise design and practical synthesis such products
ANTIVIRAL DRUG DESIGN

1. FUNDAMENTALLY PREDICTABLE BASIC PRINCIPLES

1.1. Small and Macro Molecules In view for counter intervention in Viral Life Cycle

1.2. Relevant Criteria for Macromolecular (Poly)Antivirals Synthesis

- Solubility in aqua-based physiological media → hydrophilic O / N groups
- Charge selectivity to Virions in competition with Cell’s Receptors → Poly Anions $n^-$

Comments
The 1st is solubility in aqua media, 2nd – Charge regulated selectivity toward the positively charged virions, used Coulomb forges to be attracted by negatively charged cell’s receptors. Exactly the polyanionic polymers may be most effective interceptors of virions in competition with cells, suppressing an adsorption of viruses on cells.
ANTIVIRAL DRUG DESIGN

1. FUNDAMENTALLY PREDICTABLE BASIC PRINCIPLES

1.1. Small and Macro Molecules

In view for counter intervention in Viral Life Cycle

1.2. Relevant Criteria for Macromolecular (Poly)Antivirals Synthesis

- Solubility in aqua-based physiological media → hydrophilic O/N groups
- Charge selectivity to Virions in competition with Cell’s Receptors → Poly Anions $n^-$
- Ability to bind Targets via combinations of ionic + H-bonds simultaneously
- Modifiability for said Ligands

Comments:
- The bio motivated criteria should be completed by chemical requirements for Ability to bind Targets double effective - using both ionic and hydrogen bonds simultaneously. The most suitable can be intrachain-inserted fragments of succinic acid, that easy obtained from copolymers of maleic anhydride via hydrolysis. Moreover, the anhydride centers are excellent points for covalent linkage of desired ligands in side positions of polymeric chain through aminolysis and/or esterification.
ANTIVIRAL DRUG DESIGN

1. FUNDAMENTALLY PREDICTABLE BASIC PRINCIPLES

1.1. Small and Macro Molecules
In view for counter intervention in Viral Life Cycle
Resulted Variant of some Basic Formulation for Synthetically Allowed Poly-Antivirals

1.2. Relevant Criteria for Macromolecular (Poly)Antivirals Synthesis

Required Combination of Properties

<table>
<thead>
<tr>
<th>Required Properties</th>
<th>Structural Determinants</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Polymerization degree $n$</td>
</tr>
<tr>
<td>Nano-competency</td>
<td>Determinant</td>
</tr>
<tr>
<td>Aqua-Solubility</td>
<td>Co-factor</td>
</tr>
<tr>
<td>Bio-Selectivity</td>
<td>Nano-Trigger</td>
</tr>
<tr>
<td>Non-Toxicity</td>
<td>Variator</td>
</tr>
<tr>
<td>Anti-Viral Efficiency</td>
<td>Directly Targeted</td>
</tr>
</tbody>
</table>

These leads us to the resulted variant of Basic Formulation of candidate for Polyantivirals. Here we have understandable set of macromolecular structure’s determinants allows us to regulate their desired properties: Nano-competency, Aqua-solubility, Charge-dependent Bio selectivity, as well: Non-toxicity and Anti-viral efficiency, which could be conducted through immune modulation and/or be directly targeted to virus objects in view for above mentioned priority within stages I, II and IV.
ANTIVIRAL DRUG DESIGN

1. FUNDAMENTALLY PREDICTABLE BASIC PRINCIPLES

1.1. Small and Macro Molecules

In view for counter intervention in Viral Life Cycle

1.2. Relevant Criteria for Macromolecular (Poly)Antivirals Synthesis

Resulted Variant of some Basic Formulation for SyntheticallyAllowed Poly-Antivirals

Non Toxicity = f (C--C)

Increasing the \( \text{LD}_{50} \) (mg/kg) = decreasing the toxicity

90 < 196 ≈ 170-300 < 386 ≈ >400 < 1230 < 1415 < 1600 < >2000 ≈ >2000

\( \text{In vivo, intraperitoneally (i.p.)} \)

Comments

- The next is history of pilot synthesis and selection of most prospective candidates in accordance with criteria of Non-toxicity & (next slide)
1.1. **Fundamentally Predictable Basic Principles**

**Macro Molecules** In view for counter intervention in **Viral Life Cycle**

1.2. **Relevant Criteria for Macromolecular (Poly)Antivirals Synthesis**

Resulted Variant of some Basic Formulation for Synthetically Allowed Poly-Antivirals

Non Toxicity $= f(Ct^+)$

Increasing the $\text{LD}_{50}$ (mg/kg) = decreasing the toxicity

<table>
<thead>
<tr>
<th>$\text{Ct}^+$</th>
<th>Cu$^{**}$</th>
<th>Ni$^{**}$</th>
<th>Zn$^{**}$</th>
<th>K$^+$</th>
<th>Ba$^{**}$</th>
<th>H$^+$</th>
<th>Pt$^{**}$</th>
<th>Pd$^{**}$</th>
<th>Sr$^{**}$</th>
<th>Li$^+$</th>
<th>NH$_4^+$</th>
<th>Ca$^{**}$</th>
<th>Na$^{**}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>%</td>
<td>25</td>
<td>25</td>
<td>25</td>
<td>50</td>
<td>25</td>
<td>100</td>
<td>25</td>
<td>25</td>
<td>25</td>
<td>50</td>
<td>50</td>
<td>25</td>
<td>50</td>
</tr>
</tbody>
</table>

$\text{LD}_{50} =$

|-----------|----------|----------|-----------|--------|-----------|-------|-----------|-----------|-----------|-------|-----------|-----------|-----------|

In vivo, intraperitoneally (i.p.)
ANTIVIRAL DRUG DESIGN

1. FUNDAMENTALLY PREDICTABLE BASIC PRINCIPLES

1.1. Macro Molecules In view for counter intervention in Viral Life Cycle

Stages

I Virions

II V. Entry

III Replication

IV Assembly

Targets 2-500 nm

Virions’ surface (incl. Spikes)

Virion Spikes

Cell Receptors

Cell membrane Raft –domains

Fusion Mediators

Nucleic Acids & complexes

Assembling pre-virion nano-units

Cell membrane Raft –domains

1.2. Relevant Criteria for Macromolecular (Poly)Antivirals Synthesis

Resulted Variant of some Basic Formulation for Synthetically Allowed Poly-Antivirals

Non Toxicity = f (Ct+)

In vivo, intraperitoneally (i.p.)

LD50 = 90 190 470 > 1000 1100 1600 1700 2000 > 2000 > 2000 > 2000 > 2000 mg/kg

Anti-Viral Efficiency | Immune mediated

IFN = ≤ 10 ≤ 10 ≤ 80 ≤ 320 ≤ 80 ≤ 80 ≤ 10 ≤ 40 ≤ 40 ≤ 40 ≤ 10 ≤ 80 ≤ 320 mg/kg

Ig = --- --- 60 310 160 280 170 --- 60 --- --- 190 360 mg/kg

Comments

And Antiviral potency, at least, of an immune mediated mode
ANTIVIRAL DRUG DESIGN

1. FUNDAMENTALLY PREDICTABLE BASIC PRINCIPLES

1.1. Macro Molecules In view for counter intervention in Viral Life Cycle

1.2. Relevant Criteria for Macromolecular (Poly)Antivirals Synthesis

RESULTED VARIANT OF SOME BASIC FORMULATION FOR SYNTHETICALLY ALLOWED POLY-ANTIVIRALS

<table>
<thead>
<tr>
<th>Leaders for</th>
<th>Non Toxicity + immune stimulation:</th>
</tr>
</thead>
<tbody>
<tr>
<td>DVE(SA)SA</td>
<td>FSA</td>
</tr>
</tbody>
</table>

\[
\text{Ct}^+ = \begin{cases} \text{Cu}^{++}, & 25 \\ \text{Ni}^{++}, & 25 \\ \text{Zn}^{++}, & 25 \\ \text{K}^+, & 50 \\ \text{Ba}^{++}, & 25 \\ \text{H}^+, & 100 \\ \text{Pt}^{++(n+1b)}, & 25 \\ \text{Pd}^{++(n+1b)}, & 25 \\ \text{Sr}^{++}, & 25 \\ \text{Li}^+, & 50 \\ \text{NH}_4^+, & 25 \\ \text{Ca}^{++}, & 25 \\ \text{Na}^+, & 25 \end{cases} \%
\]

\[
\text{LD}_{50} = \begin{cases} 90 & \text{Cu}^{++} \\ 190 & \text{Ni}^{++} \\ 470 & \text{Zn}^{++} \\ > 1000 & \text{K}^+ \\ 1100 & \text{Ba}^{++} \\ 1600 & \text{H}^+ \\ 1700 & \text{Pt}^{++(n+1b)} \\ 2000 & \text{Pd}^{++(n+1b)} \\ > 2000 & \text{Sr}^{++} \\ 2000 & \text{Li}^+ \\ 2000 & \text{NH}_4^+ \\ > 2000 & \text{Ca}^{++} \\ > 2000 & \text{Na}^+ \end{cases} \text{mg/kg}
\]

\[
\text{Anti-Viral Efficiency} \quad \text{Immune mediated}
\]

\[
\text{IFN} = \begin{cases} \leq 10 & \text{Cu}^{++} \\ \leq 10 & \text{Ni}^{++} \\ \leq 80 & \text{Zn}^{++} \\ \leq 80 & \text{K}^+ \\ \leq 80 & \text{Ba}^{++} \\ \leq 10 & \text{H}^+ \\ \leq 40 & \text{Pt}^{++(n+1b)} \\ \leq 40 & \text{Pd}^{++(n+1b)} \\ \leq 40 & \text{Sr}^{++} \\ \leq 10 & \text{Li}^+ \\ \leq 40 & \text{NH}_4^+ \\ \leq 10 & \text{Ca}^{++} \\ \leq 320 & \text{Na}^+ \end{cases} \text{mg/kg}
\]

\[
\text{Ig} = \begin{cases} 60 & \text{Cu}^{++} \\ 310 & \text{Ni}^{++} \\ 160 & \text{Zn}^{++} \\ 280 & \text{K}^+ \\ 170 & \text{Ba}^{++} \\ 60 & \text{H}^+ \\ 60 & \text{Pt}^{++(n+1b)} \\ 60 & \text{Pd}^{++(n+1b)} \\ 190 & \text{Sr}^{++} \\ 360 & \text{Li}^+ \\ 50 & \text{NH}_4^+ \\ 25 & \text{Ca}^{++} \\ 25 & \text{Na}^+ \end{cases} \text{mg/kg}
\]

Comments:
- Finely we come to the displayed two copolymeric structures
1. FUNDAMENTALLY PREDICTABLE BASIC PRINCIPLES

1.1. Small and Macro Molecules

In view for counter intervention in Viral Life Cycle

Stages

1. Virions
   - Virions’ surface (incl. Spikes)
2. V. Entry
   - Virion Spikes
   - Cell Receptors
   - Cell membrane Raft –domains
   - Fusion Mediators
3. Replication
   - Nucleic Acids & complexes
   - Assembling pre-virion nano-units
   - Cell membrane Raft –domains
4. Assembly

Targets 2-500 nm

Leaders for Non Toxicity + immune stimulation:

- DVE(SA)SA
- FSA

Anti-Viral Efficiency Immune mediated

1.2. Relevant Criteria for Macromolecular (Poly)Antivirals Synthesis

Resulted Variant of some Basic Formulation for Synthetically Allowed Poly-Antivirals

in vivo, Protection, %

<table>
<thead>
<tr>
<th>TBV</th>
<th>RbV</th>
<th>EEEV</th>
</tr>
</thead>
<tbody>
<tr>
<td>65(-)</td>
<td>55(82)</td>
<td>65(95)</td>
</tr>
</tbody>
</table>

In vivo evaluations were performed under the lethally hard conditions up to 200 LD_{so} of Viruses

(in brackets – for combined: PolyAntivirals + Vaccine)

and following bio evaluations in vivo revealed highly significant capacity of these polyanionic compound protecting mice or rats against lethal doses of neuroviral infections.
The next step of the Polyantivirals development was oriented to combining the immune mediated potency with an additional capacity for direct anti-viral targeting impacts
**ANTIVIRAL DRUG DESIGN**

### 1. FUNDAMENTALLY PREDICTABLE BASIC PRINCIPLES

#### 1.1. Small and Macro Molecules *In view for counter intervention in Viral Life Cycle*

- Virions’ surface (incl. Spikes)
- Virion Spikes
- Cell Receptors
- Cell membrane Raft –domains
- Fusion Mediators
- Nucleic Acids & complexes
- Assembling pre-virion nano-units
- Cell membrane Raft –domains

**Targets** 2-500 nm

#### 1.2. Relevant Criteria for Macromolecular (Poly)Antivirals *Synthesis*

Resulted Variant of some Basic Formulation for Synthetically Allowed Poly-Antivirals

**NEXT GENERATIONS**

**The L-**co-SAR-programmed directly against Viral Targets

**Comments**

This task was taken into realization via chain’s side positions modification by desired combinations of ligand-expecting species, even if they are not active in small molecule forms.

- We propose an ability of they activation to detectable levels of purposed bioactivity due to a rational conintegration together on the prepared platforms of polymeric chains.
- Similarly macromolecular programming the protein and nucleic acid biopolymeric chains by certain combinations of side-groups we hope to find novel artificial combinations of so called “synthetic” polymers which could programmed these polymers toward virus-specific targets.
ANTIVIRAL DRUG DESIGN

1. FUNDAMENTALLY PREDICTABLE BASIC PRINCIPLES

1.1. Small and Macro Molecules

In view for counter intervention in Viral Life Cycle

1.2. Relevant Criteria for Macromolecular (Poly)Antivirals Synthesis

Resulted Variant of some Basic Formulation for Synthetically Allowed Poly-Antivirals

Basic Design

Model. Synth. Testing

Macro Molecules

In view for counter intervention in Viral Life Cycle

1.1. Relevant Criteria for Macromolecular (Poly)Antivirals Synthesis

Resulted Variant of some Basic Formulation for Synthetically Allowed Poly-Antivirals

II. Virions

V. Entry

III. Replication

IV. Assembly

Comments

And today we have, at least, seven original generations that possess many-folds more higher and widen antiviral activity in comparison with known small molecule prototypes. The correspondent indexes of selectivity for inhibition of HIV, Influenza, Cytomegalo viruses, as examples, are shown in the Table. The markers to the left indicate the most expected virus-specific targets for the each generation in correspondence with stages of a viral life cycle.

In vivo evaluations were performed under the lethally hard conditions up to 200 LD50 of Viruses (in brackets – for combined: PolyAntivirals + Vaccine)
Modeling
the interactions between
Poly-ANTIVIRALS (of 1-4 generations)
and Viral Fusion mediated Proteins
gp41 (HIV), HA2 (Influenza), Gp2 (Ebola)
...

Comments
Of course, some of that polymeric compounds should be investigated in details applying modern possibilities of computer-aided modeling
2. MODELING the POLY-ANTIVIRALS ↔ VIRAL TARGETS

2.1. Macro Molecular Products synthesized within the generations 1, 2, 3, 4

Stages

I. Virions
- Virions’ surface (incl. Spikes)
- Virion Spikes
- Cell Receptors
- Cell membrane Raft – domains
- Fusion Mediators

II. V. Entry
- No Side Ligands
- -Z-SO3⁻
- -Z-Nb
- -Z-Ad

III. Replication
- Nucleic Acids & complexes
- Assembling pre-virion nano-unit
- Cell membrane Raft – domains

IV. Assembly
- Strongly effective • Antiviral protection + • Drug resistance prevention
- Polymer capacity for multipoint covering the viral Nano-Targets

Next Generations
The \( \text{L} \)-co-SAR-programmed directly against Viral Targets

<table>
<thead>
<tr>
<th>No</th>
<th>(-Z-\text{L})</th>
<th>(m/n)</th>
<th>(\text{in vitro, SI} = \text{CC}<em>{50}/\text{IC}</em>{50})</th>
<th>(\text{in vivo, Protection, }%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>No Side Ligands</td>
<td>0</td>
<td>10-100 ≤ 30 ≤ 350</td>
<td>65(-) 55(82) 65(95)</td>
</tr>
<tr>
<td>2</td>
<td>-Z-SO3⁻</td>
<td>≤ 0.80</td>
<td>&gt;680 →10000</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>-Z-Nb</td>
<td>0.1-0.3</td>
<td>&gt;3300 &gt;2140</td>
<td>240</td>
</tr>
<tr>
<td>4</td>
<td>-Z-Ad</td>
<td>0.1-0.3</td>
<td>&gt;1100 →10000</td>
<td>25</td>
</tr>
</tbody>
</table>

Comments

- First of all, there are generations 3 and 4 where the norbornane (terpenoid related) and adamantane-derived side ligands has been used, in comparison with 1 and 2 generations.
ANTIVIRAL DRUG DESIGN

2. MODELING the POLY-ANTIVIRALS ↔ VIRAL TARGETS

2.1. Small and Macro Molecular Products synthesized within the generations 1, 2, 3, 4

Viruses of Influenza type A block the very small gate of ion (proton) channel between 4 nano-scale molecules M2

Amino derivatives of Adamantane (Ad), Norbornane (Nb) block the very small gate of ion (proton) channel between 4 nano-scale molecules M2

Amantadine, Rimantadine, Deitiforin, …

are well known to be a fairly good inhibitors of some Influenza type A viruses,

but without any significant protection against other viruses

Objectively:

the small size → the highly limited effectiveness & applicability

- within the only size adequate - very specific targets, which can be not typical for other viruses
- because of easily allowed Drug Resistance
  – by simplest (one point) mutations of virus
- in virtue of an enhanced Permeability through bio-protective barriers → resulted in hardly controllable risks of toxicity

Comments

● The small molecule prototypes such as rimantadine and similar are well known to be a fairly good inhibitors of some Influenza type A viruses, but without any significant protection against other viruses.
● However, considered here approach to the ‘Polyantivirals’ implies novel possibilities for powerful amplification of the small molecules’ potency due to their rational cointegration into the polyanionic macromolecules.
2. MODELING the POLY-ANTIVIRALS ↔ VIRAL TARGETS

2.1. Small and Macro Molecular Products synthesized within the generations 1, 2, 3, 4

Stage II

Virus Entry

Rimantadine + many variations

Amino derivatives of Adamantane (Ad), Norbornane (Nb)

HIV-1

Stage II Entry, Sub-Step Fusion of Vir. Envelope with Cell Membrane mediated by glycoprotein (gp41)₃ = nano-Target?

Continent comments

That has been first demonstrated experimentally by our research group in relation to rimantadine resistant viruses of Influenza and HIV. Particularly, the gp41, being key mediator of HIV fusion, was found could be the most probable nano-target of anti-HIV protection by these Polyantivirals. Extraction from literature data includes modelling works, and analysis these data lead us to NHR region of gp41, as a most probable target.
2. MODELING the POLY-ANTIVIRALS ↔ VIRAL TARGETS

2.1. Small and Macro Molecular Products synthesized within the generations 1, 2, 3, 4

Virus Entry

Rimantadine + many variations

Amino derivatives of Adamantane (Ad), Norbornane (Nb)

Basic Design
Model
Testing

Stage II

Poly-Antivirals, Generations 3 and 4

HIV-1, Stage II Entry, Fusion, (gp41)3, (NHR)3+ - cationic domain = nano-Target?

Local attacks scattered in space & time

Unstable Binding

Binding strongly Stabilized by Polymeric Chain

Polymer chain – combines small precursors together, supporting:

- multi-point cooperative attacks
- mutual reinforcing in space & time
- Polymer Charge-selective orientation toward the cationic Target

Chain flexibility allows ligand-cooperating adaptation to the Target

DOCKING

Comments

- The small molecule carbocycles inefficiency against HIV fusion is well explained by the Docking modeling. The binding energies are too slow to provide a stable blocking. The observed multiple binding-permitted sites allows to conclude that these small molecules capable of only local attacks, scattered in space and time. But they incapable working together in mutual coordination against the target. However the required possibility can be achieved hypothetically on basis of integrated polymeric chains.

≤ 60 kcal/mol

≤ 20 kcal/mol
2. MODELING the POLY-ANTIVIRALS ↔ VIRAL TARGETS

2.1. Small and Macro Molecular **Products synthesized within the generations 1, 2, 3, 4**

**Stage II**

**Virus Entry**

---

**DOCKING**

**Poly-Antivirals, Generations 3 and 4**

**HIV-1, Stage II Entry, Fusion, (gp41)$_3$, (NHR)$_3^+$ - cationic domain = nano-Target?**

**Electrostatic Potential (EP)**

**H-bonding Sites (HB)**

**Lipophilic Potential (LP)**

**Cavity Depth (CD)**

---

- The Docking and MD exploration undertaken in collaboration with Vladimir Tsvetkov has been launched from pre-investigation of the Target parameters. Particularly it is typical nano-object with scale up to 5 nm in length, enriched by positive charged amino acids potentially sensitive to polyanionic chains.
2. MODELING the POLY-ANTIVIRALS ↔ VIRAL TARGETS

2.1. Small and Macro Molecular Products synthesized within the generations 1, 2, 3, 4

**Basic Design**

**Synth.**

**Testing**

---

**Stage II**

**Virus Entry**

---

**Step 1** Separate backbone unit or pendant anchor (small molecule) models, e.g.:

\[
X \quad - \quad Y - \text{Anc} = \quad -16.6 \quad -15.1 \quad -16.8 \quad -18.7 \text{ kcal/mol}
\]

- 60-70% - L1
- 50-60% - L2
- 100% - L3

**Step 2** Polymeric chain motifs

- L1
- L2
- L3

**Step 3** Extrapolation toward

1) \( n \rightarrow 30-50 \)
2) effective Anc \( \geq 9 \)

(\( \geq 6-8\% \) from all X)

Predicted \( \Delta G_{\text{min}} \leq 200 \text{ kcal/mol} \)

---

Comments

- Then docking of small size structural fragment from backbone of polymeric chain, as well side-ligands and linkers were performed with following stepwise elongation of models toward bigger parts of polymeric antiviral molecule till the scale that permitted by docking program applied. Further the data obtained were used for extrapolations to scale comparable with original polymer compound.
ANTIVIRAL DRUG DESIGN

2. MODELING the POLY-ANTIVIRALS ↔ VIRAL TARGETS

2.1. Small and Macro Molecular Products synthesized within the generations 1, 2, 3, 4

Stage II

Virus Entry

DOCKING + MOLECULAR DYNAMICS

- 63.0 ± 9.7 kcal/mol

- 59.1 ± 12.5 kcal/mol

- 61.8 ± 14.5 kcal/mol

Comments

The studies were completed by molecular dynamics, particularly, of n=11 oligomer equipped by 3 anchors, manually positioned to be well capable both for axial and belting types binding. This model demonstrated good binding energy nearly 60 kcal/mol, sufficient for stable while dynamically adaptable fixation on the target.
2. MODELING the POLY-ANTIVIRALS ↔ VIRAL TARGETS

2.1. Small and Macro Molecular Products synthesized within the generations 1, 2, 3, 4

- Basic Design
  - Model
  - Synth.
  - Testing

**Stage II**

**Virus Entry**

**Antiviral Drug Design**

**Poly-Antivirals**

**Virus Entry**

- **Small Molecules**
  - ΔE kcal/mol
  - Antiviral SI
  - ≤ [-12.7]
  - ≤ [-13.8] → [-15.6]
  - ≤ [-16.7]
  - ≤ [-18.4]

- **Poly-Antivirals**
  - ΔE kcal/mol
  - Antiviral SI
  - No detectable and statistically significant anti-HIV activity
  - Binding Energy
  - Binding Mode Regulation:
    - A – Axial
      - n ≥ 4-6
      - m → 0
    - B – Belting
      - n ≈ 4-12
      - m ≈ 2-3
    - C – Combined
      - n ≥ 30-50
      - m ≥ 9

- DOCKING + MD

**Vladimir B. Tsvetkov, Alexander V. Serbin, et al.**

*Biotechnologia – 2012. – № 1. – С. 72-89;

*The results obtained are in well correlation with experimentally detected anti-HIV-1 activity in vitro*

*Finally, the tree modes of the target binding were found (axial, belting, combined) in dependence on degree of polymerization and in comparison with grade of side modification by hydrophobic carbocyclic ligands (anchors) and their configuration*
In view of important role of the polymerization degree, related to molecular mass, the next task of special modeling was oriented toward the problem of the mass control under practical synthesis of precursors for polymeric chains.
3.1. Providing the **MM** and **MMD** of **Polymeric Basis** required for the purposed bioactivity

The widely used methods for control of **MM** and **MMD** required in *practical synthesis*:

\[(\text{MM}) \text{ – Molecular Mass and their distribution (MMD)}\]

\[n \text{ – degree of polymerization } \leftrightarrow \text{ the Polymeric Chain’s length / size}\]

**Comments**


3.1. Providing the MM and MMD of Polymeric Basis required for the purposed bioactivity

- In studied case the best MM-controlling results were observed for reversible three thiocarbonate RAFT-agent
- However this method have one by-effect: inserting the threethiocarbonate residue in center of chain and the benzene cycles – in both tails of the chain
3. POLY-ANTIVIRALS SYNTHESIS CONTROL FOR CHAIN SIZE

3.1. Providing the MM and MMD of Polymeric Basis required for the purposed bioactivity

The control required for Practical Synthesis:

- **Modeling** the possible bio consequences:
  - DOCKING + MD
  - - 50-70 kcal/mol
  - - 20-30 kcal/mol

- **Reverse corrections** for the Synthesis methodology

**Basic Design**
- **Model**
- **Synth.**
- **Testing**

**Comments**
- So prior usage these methods we need estimate the possible bio consequences
- The MD shown significant decreasing the binding energy crucially required for purposed antiviral activity. Therefore it indicates the necessity to find reverse corrections of the practical synthesis methodology to prevent or remove the undesired micro insertions within polymeric chain
3.1. Providing the **MM** and **MMD** of Polymeric Basis required for the purposed bioactivity

**Basic Design Model.**

**Testing Synth.**

**Providing the MM and MMD of Polymeric Basis required for the purposed bioactivity**

With RAFT residues

*Sum* = $|\text{-50-70}|$ kcal/mol

Coulomb + hydration

*Sum* = $|\text{-20-30}|$ kcal/mol
The second unique problem of the practical synthesis appeared as polymeric chain isomerism.
3.2. Clearing mechanisms and conditions of Synthesis for chain isomers (↔ bio activity?)

\[ 4 \text{ routes} \cdot 2^6 \text{ stereo isomers} = 256 \text{ isomeric variants of chin backbone units?} \]

Synthesis of the Parent Polymeric backbone, [-DVE(MA)-MA-]_n, which is reactive for the following conversions toward Poly-ANTIVIRALS

Controlled portion aminolysis (L'= COOH, CONHR), esterification (L'= COOH, COOR), final hydrolyzis (L'= COOH↔COO⁻+H⁺);

BIO ACTIVE Poly-ANTIVIRAL GENERATIONS [1,2,3,4,5,6,7, presented above]

Comments

Alternating radical cyclo copolymerization of DVE with MA hypothetically could assume up to 256 variations of isomerism. Of course, some of them are predictable preferable. But no reliable information in this regard was found.
3.2. Clearing mechanisms and conditions of Synthesis for chain isomers ($\leftrightarrow$ bio activity?)

Quantum chemical investigation of chain isomerism regulation

**Methods**

Approbation and selection of known Methods in search for

- CPU-time saving, but
- precisely adequate to estimation of the $E_a$ and $\Delta H_r$

in reference to experimental data about related reactions

Boris D. Bolshchikov
Alexander V. Serbin,
et al.

*Polymer.*

*Biomed Chem (Russia).*
2019, 65 (2) 133-151J

*Macromolecular Chemistry and Physics.*

*Biomed Chem (Russia).*
2019, 65 (2) 133-151J

*Macromolecular Chemistry and Physics.*

Comments

- Then a special quantum chemical studies in combination with kinetics modeling were performed in collaboration with Boris Bolshchikov
3.2. Clearing mechanisms and conditions of Synthesis for chain isomers (↔ bio activity?)

Quantum chemical investigation of chain isomerism regulation

Stepwise sub-Reactions Map

Stepwise sub-Reactions’ energy (the $E_a$ and $\Delta H_r$) Map

Comments

*All considerable routes of sub reactions and activation energies and enthalpies were estimated, mapped and analyzed.*
3.2. Clearing mechanisms and conditions of Synthesis for chain isomers (↔ bio activity?)

Quantum chemical investigation of chain isomerism regulation

\[ f(\text{concentrations}) + \text{Kinetics Modeling} \]

Comments
- Crucial points and factors of kinetic and thermodynamic control of isomerism, as well as quantitative estimations of isomeric variations in polymeric chain were determined.
- The computational prognosis for possible variations of special experimental conditions allowing switching isomerism from furan-related cyclization toward pyran-related alternative were found and formulated in practical recommendations.
3.2. Clearing mechanisms and conditions of Synthesis for chain isomers (↔ bio activity?)

Quantum chemical investigation of chain isomerism regulation

Pyranose-like cyclo- isomerism = polysaccharide – related mimicry

HIV-1, gp41 - NHR₃

- 173 kcal/mol

Furanose-like cyclo- isomerism = Nucleic Acids – related mimicry

- 195 kcal/mol
Current Results & Prospects

Comments
- And finally Current results & Prospects
ANTIVIRAL DRUG DESIGN

Poly-Antivirals

<table>
<thead>
<tr>
<th>№</th>
<th>Ligand</th>
<th>m/n</th>
<th>in vitro, SI = CC50/IC50</th>
<th>in vivo, Protection, %</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>No Side Ligands</td>
<td>0</td>
<td>10-100</td>
<td>≤ 30</td>
</tr>
<tr>
<td>2</td>
<td>Z-SO₃⁻</td>
<td>≤ 0.80</td>
<td>&gt;680</td>
<td>→10000</td>
</tr>
<tr>
<td>3</td>
<td>Z-Nb</td>
<td>0.1-0.3</td>
<td>&gt;3300</td>
<td>&gt;2140</td>
</tr>
<tr>
<td>4</td>
<td>Z-Ad</td>
<td>0.1-0.3</td>
<td>&gt;1100</td>
<td>→10000</td>
</tr>
<tr>
<td>5</td>
<td>Z-Chol</td>
<td>≤ 0.03</td>
<td>&gt;220</td>
<td>5400</td>
</tr>
<tr>
<td>6</td>
<td>Z-Pept Cell Receptors</td>
<td>≤ 0.02</td>
<td>→10000</td>
<td>-</td>
</tr>
<tr>
<td>7</td>
<td>Z-Pept Viral (HIV, MA)</td>
<td>≤ 0.01</td>
<td>+</td>
<td>← Test-Samples are completed for bio evaluations</td>
</tr>
</tbody>
</table>

Comments:
The set of novel high effective Polyantivirals was designed, synthesized, successfully tested and partially modeled with great use for theory and practice of antiviral drug development. Some other part of polyantiviral generations needs future studies basing on modeling techniques.
Current groundwork opens wide horizon for future advancing, including inhibition of other danger viruses. Especially concerning the viruses with similar biomolecular mechanisms of entry into cells, where viruses can be stopped at the very initial stage.
ANTIVIRAL DRUG DESIGN

Basic Design
Model → Synth. → Testing

### Comments
- Current groundwork opens wide horizon for future advancing, including inhibition of other danger viruses.
- Especially concerning the viruses with similar biomolecular mechanisms of entry into cells, where viruses can be stopped at the very initial stage.
Thank you for your attention