Review Article

An Overview on Zika Virus and the Importance of Computational Drug Discovery

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Abstract

Since the beginning of the century, viral infection has become a widespread problem to public health globally. Recently, the latest newcomer of the *Flaviviridae* family, the Zika virus, has emerged as a potential global threat to human health. Due to lack of sufficient data at present, Zika infection has become a major cause of Zika fever, central nervous system malformations such as microcephaly, severe neuroimmunopathology, fetal abnormalities, and recently Guillain-Barre syndrome. The lack of genomic as well as proteomic information keeps the Zika virus under examination by a multitude of researchers. The rapid increase in Zika viral infections has been classified as a public health emergency by the World Health Organization. Neither preventive antiviral drugs nor effective vaccines are available on the market for combating Zika infection. Urgent innovative research is necessary to facilitate the development of protective and fruitful therapeutic agents, to improve lifespan of individuals throughout the world. Thus, we present this review to summarize the current research findings for Zika viral infection and to highlight the importance of computational drug discovery in the development of potent antiviral inhibitors against the Zika virus. We also anticipate that the information in this review will present a valuable opportunity for the prediction of efficient novel therapeutic remedies for controlling Zika.

Introduction

In 1947, Zika virus (ZIKV) was detected in the serum of a febrile sentinel rhesus monkey, found near the Zika forest in Uganda. Since then, the name of "Zika virus" has been kept on the basis of the primary location of its isolation region.[1] ZIKV is now classified as an emerging arbovirus (arthropod-borne virus) belonging to the *Flaviviruses* genus within the *Flaviviridae* family. and carrying a single-stranded positive-sense RNA. In addition to ZIKV, the *Flaviviruses* include dengue virus, yellow fever virus, St. Louis encephalitis virus, West Nile virus, Japanese encephalitis virus and tick-borne encephalitis virus.

Keywords: Zika virus**;** Zika fever**;** Drug targets**;** Computational drug discovery**;** Anti-ZIKV.

Abbreviations: ADME/T, absorption, distribution, metabolism, excretion and toxicity; C, capsid protein; CDD, computational drug discovery; CS, conserved sequence; E, envelope protein; MTase, methyltransferase; NS, non-structural protein; prM, precursor membrane protein; RDRP, RNA-dependent RNA polymerase; QSAR, quantitative structure–activity relationship; UTR, untranslated region; WHO, World Health Organization; ZIKV, Zika virus.

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ZIKV spreads mainly via infected mosquitoes, like the *Aedes aegypti* or the *Aedes albopictus* species, which are known to be associated with other disorders such as central nervous system malformations and Guillain-Barre syndrome, and generally cause fever, sweating, rashes, vomiting, and pains in muscles and the back of the eyes. The World Health Organization (WHO) declared ZIKV as a Public Health Emergency of International Concern in February 2016, due to the severity of ZIKV infections throughout the world. Based on WHO reports, more than 84 countries in the world provided evidence of ZIKA viral infections between April of 2007 and March of 2017. However, the future status of ZIKV distribution to the other regions of the world remains a mystery.[2,3]

Owing to the limited knowledge of ZIKV's genomic, proteomic and molecular mechanisms, neither potent drugs nor effective vaccines are presently available on the market. Thus, the present scenario has manifested a higher demand for safe and effective therapeutics to control the effects of ZIKV infection and to improve the quality of life and lifespan of individuals suffering with ZIKV infection. Developing a novel drug, from laboratory to market, takes time as well as money. Computational drug discovery (CDD) may help to identify a new antiviral therapeutic within less time and with less cost.

CDD is the most prominent approach currently used to search for novel drug candidates on a large scale, and it can be further implemented and optimized by inclusion of experimental and *in vivo* studies to validate the identified drug molecule against the targeted diseases.[4–6] Thus, this review presents the cutting edge knowledge related to ZIKV and its biological targets, and includes

Table 1. Historical review of the rise of ZIKV[12–14]

Key Events	Year
First confirmation of ZIKV in a rhesus monkey near the Zika forest of Uganda	1947
Recognition of a virus in the mosquito *Aedes africanus* in the Zika forest	1948
Serological evidence from the first human; detection of Zika antibodies in parts of Uganda and the United Republic of Tanzania	1951
ZIKV identification in various countries, such as Central African Republic, Sierra Leone, Egypt, Tanzania, Gabon, Angola, Liberia, Ivory Coast, Philippines, Malaysia, India, Pakistan, Thailand, Indonesia, Vietnam and Kenya	1952 to 1984
First major occurrence of Zika in humans in Yap Islands, Micronesia	2007
In Senegal, first case of sexual contact reported	2008
Identification of Zika infection in other locations, such as Cameroon, Maldives, Philippines and Thailand; the research community explains the lineages of African and Asian Zika viruses	2010 to 2012
The second outbreak of Zika is independently detected in French Polynesia, following in the Cook Islands, Chile and New Caledonia	2013 to 2014
15 May, the Ministry of Health confirms the presence and rapid circulation of ZIKV in Brazil and the Americas; at the end of December, local transmission of infection is reported in more than 45 countries, including Aruba, Bonaire, Cape Verde, Colombia, Curacao, Cost Rica, Dominican Republic, French Guyana, Guatemala, Haiti, Jamaica, Mexico, Paraguay, Samoa, Saint Martin, Suriname, Tonga and Venezuela; according to reports, outbreaks of ZIKV are also detected in some European countries, such as Denmark, Germany, Italy, France, The Netherlands, Spain, Switzerland, United Kingdom and Sweden	2015
February, the World Health Organization declares the ZIKV outbreak to be a Public Health Emergency of International Concern; March, Bangladesh's Health Ministry confirms the first case of Zika infection; Singapore's Ministry of Health and National Environment Agency announces a total of 90 cases of locally-transmitted ZIKV infection	2016
May, the Ministry of Health and Family Welfare, Government of India has evidence of ZIKV outbreak in Ahmedabad, Gujarat	2017

Abbreviation: ZIKV, Zika virus.

discussion on the significance of CDD for designing and developing anti-ZIKV therapeutic agents.

Search and selection criteria for ZIKV therapeutic agent candidates

The general approach taken for searching and selection of data for this review was based upon the keywords of "Zika virus", "Zika viral infection", "Zika forest", "ZIKV genome", "Zika outbreak", "ZIKV drug targets", "ZIKV inhibitors" and "computational drug discovery" using Google search engine, NCBI-PubMed, and standard scientific journals. We retrieved all selected articles, reviews and other literature in peer-reviewed journals. This strategy provided beneficial information about ZIKV, including its functional proteins and the exact role of CDD in the development of anti-ZIKV inhibitors.

Worldwide outbreak

The first conformation of Zika occurred in a simian near the Zika forest of Entebbe, at the side of Lake Victoria in Uganda in 1947. Subsequently, the first case of human infection was found in Nigeria, in the year of 1954. Later on, epidemic outbreaks of Zika infections were reported in various countries, such as Central African Republic, Sierra Leone, Egypt, Tanzania, Gabon, Philippines, Malaysia, India, Pakistan, Thailand, Indonesia, Vietnam and Kenya between the years of 1954 to 1983. The first major outbreak occurred in Yap Island, Micronesia in 2007. Soon after, in 2008, Senegal suffered a remarkable outbreak. From October 2013 to January 2014, the second major outbreak of Zika infection occurred in French Polynesia, followed by outbreaks in the Cook Islands, Chile and New Caledonia.

In the beginning of 2015, Zika infection was reported near the Solomon Islands, and that same year a massive outbreak occurred in Brazil and America, which were confirmed by the Ministry of Health on May 15, 2015. Finally, at the end of December 2015, local transmission of infection was reported in more than 45 countries, including Aruba, Bonaire, Cape Verde, Colombia, Curacao, Cost Rica, Dominican Republic, French Guyana, Guatemala, Haiti, Jamaica, Mexico, Paraguay, Samoa, Saint Martin, Suriname, Tonga and Venezuela, along with some European countries, like Denmark, Germany, Italy, France, The Netherlands, Spain, Switzerland, United Kingdom and Sweden. Recently, the Ministry of Health and Family Welfare of the Government of India has also reported evidence of ZIKV outbreak in Ahmedabad, Gujarat in May 2017.[1,2,7–11] Rapid increase in ZIKV infections are expected to come, with a greater number of cases anticipated in the near future. Before this happens, however, the scientific research community should come forward with an essential therapeutic agent to treat ZIKV infections.

A historical overview of ZIKV distribution across the globe is presented in Table 1.[12–14]

Transmission

It is believed that Zika infection has quickly spread all over the world in the relatively short period of time since its discovery.

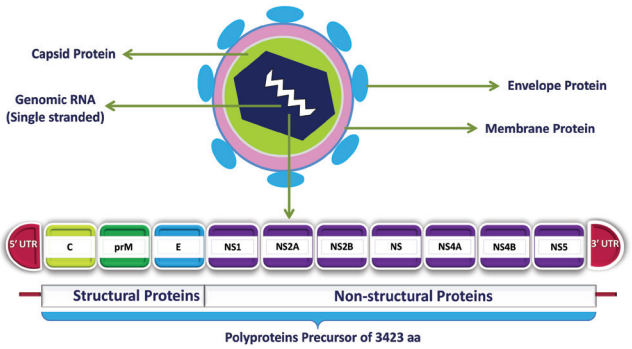

Fig. 1. Schematic representation of the ZIKV genome.

Generally, the primary host of ZIKV is humans. Like other diseases, such as Dengue and Chikungunya, the culprit for spread of ZIKV has been identified as an infected *Aedes* species mosquito's bite, which is now considered the main cause of transmission. Two subspecies of the *Aedes* mosquito, *A. aegypti* and *A. albopictus*, are reported to be major competent vectors for Zika infections.

Asia, Africa, America and Europe, including northern Austria, France, Germany and Spain, are the major regions for transmission, concerning both the *A. aegypti* and *A. albopictus* species. Other than *A. aegypti* and *A. albopictus*, various species of *Aedes* like *A. taylori A. africanus, A. furcifer, A. vittatus, A. polynesiensis, A. dalzieli, A. hirsutus* and *A. metallicus* have been identified as other sources of the viral transmission. Generally, humans serve as the primary host for the sylvatic transmission cycle of ZIKV, through direct contact (sex), mother to child, blood transfusion or in some cases through healthcare or laboratory practices. Additionally, the possible reservoirs can be wild animals, including non-primates and vertebrate species, who transmit through bites. From the reported research, it has been suggested that saliva is considered as one of the transmission vectors, having even higher frequency than blood. Hence, the evidence has thus far suggested these major reservoirs for the transmission of ZIKV infection globally.[1,3,7,15–19]

Structural genome of ZIKV

ZIKV belongs to the mosquito-borne *Flaviviruses* family. The first full-length genome of ZIKV was obtained from an African prototype strain named "MR 766", which had come from the sentinel monkey and was announced in the year of 2007.[20] The genome organization of ZIKV consists of a single-stranded positive-sense RNA with 10,794 bases making up a single long open reading frame flanked by two untranslated regions (UTRs; the 5′ UTR and the 3′ UTR). The open reading frame encodes for a polyprotein of 3,423 amino acids, with three structural proteins (the capsid (C), precursor membrane (prM) and envelope (E)) and non-structural proteins (NSs).

The ZIKV genome has the following order for these encoded proteins: 5′-C-prM-E-NS1-NS2A-NS2B-NS3-NS4A-NS4B-NS5-3′.[21–24] The 3′ terminus forms a loop structure, and the 5′ terminus has a methylated nucleotide cap which allows for cellular translation. The essential functions of genome cyclization and stabilization are carried out by the UTRs (5′ and 3′ terminal regions) in *Flaviviruses*, which contain evolutionarily conserved sequences (CSs). Older research has reported that the CSs (CS1, CS2 and CS3) may also play significant roles in the ZIKV genome of MR766, in particular. But, the organization of the CSs differs among the various *Flaviviruses*.[20,25]

Generally, the virion structure of Zika is 40 nm in diameter and in a spherical shape. It contains an enveloped proteins (E and M), nucleocapsids (within 25–30 nm in diameter, and surrounded by a host membrane-derived lipid bilayer), and non-segment regions. The surface proteins are organized in icosahedral-like symmetry.[1,24–26] The entire genomic structural view has been reproduced from the available information and is shown in Figure 1.[4,26] The available information on the ZIKV genome may be helpful in providing a detailed knowledge about the druggable targets in Zika, helping with the development of future therapeutic agents (drug or vaccine).

Role of structural proteins and NS proteins

The structural proteins and NS proteins have multi-transmembrane domains, which allows ZIKV to play an essential role in capsid formation, replication and assembly of the virus in the host. The importance of all these proteins, individually, is described below.[20,26–41]

C protein

The C protein associates with genomic RNA and is made up of 11 kDa. It functions in viral capsid formation to build a core of the mature virus particle.

E protein

The largest envelope glycoprotein (E protein, □53 kDa) is the main virion surface protein and is involved in host cell binding, membrane fusion, penetration and hemagglutination during the viral replication cycle. A total of 180 E proteins comprise the icosahedral shell of the virion and represent one of the prime targets of neutralizing antibodies.

prM protein

Next is the structural protein prM (8 kDa). This protein first presents in an immature form, but a cellular furin-like protease enzyme positioned in the trans-Golgi cleaves it into pr peptide and M proteins, and releases mature virions from the cell. This protein forms heterodimers in complex with E protein and provides protections to E from degradation during assembly of virions.

The NS proteins play key roles during the replication cycle, as well as in the host immune response. The NS1, NS3 and NS5 proteins are highly conserved and large in form, whereas the NS2A, NS2B, NS4A, and NS4B proteins are hydrophobic and very small in form, and may be associated with membranes.

NS1 protein

The glycosylated protein NS1 is 46 kDa, with 12 conserved cysteine amino acid residues, and it has a major responsibility in viral replication. It connects with the host cell membrane using glycolipid and forms a homodimer inside the cell. The specific mutations by Asn130 and Asn207 in glycosylation disturbs replication and production of the virus. NS1 is necessary to begin the synthesis process of the viral RNA genome. The well-developed molecu-

lar mechanisms of the NS1 protein in the Dengue virus could be helpful to understand the structural features of NS1 in ZIKV.

NS2A and NS2B proteins

NS2A and NS2B are membrane associated proteins of 22 kDa and 14 kDa, respectively, and centered between hydrophobic regions. However, these proteins are not fully described and no specific function has been identified for either. They also do not have any enzymatic motifs identified. NS2A interacts with NS3 and NS5, while NS2B interacts with the C-terminal protease domain of NS3. These interactions could be helpful in the packaging and replication of RNA, and in the cleavage processes of the viral proteins.

NS3 protein

NS3 is a multifunctional domain protein of 70 kDa, and plays a central role in the capping and replication processes of ZIKV. The N terminal of NS3 harbors protease activity, whereas the C terminal harbors poly(A)-inhibited RNA triphosphatase (known as the RTPase) activity and RNA helicase activity, as well as Mg^{2+}-inhibited activity. Activities of the NS3 protease and NS3 RNA helicase are reported to be central to viral replication and survival of the ZIKV; thus, both could be drug targets for novel antiviral inhibitors against the viral infection.

NS4A and NS4B proteins

Like NS2A and NS2B, NS4A and NS4B are also small, hydrophobic and membrane-associated proteins, being 16 kDa and 27 kDa, respectively. The NS4A protein helps in localization of the replication complex to the membrane, regulation of membrane proliferation, and polyprotein processing; whereas, NS4B plays a significant role in viral replication, along with the NS3 protein.

NS5 protein

NS5 is the largest and most highly conserved protein (103 kDa) in the ZIKV genome. It is composed of two essential domains, known as the N-terminal methyltransferase (MTase) domain and the C-terminal RNA-dependent RNA polymerase (RDRP). Here, the N-terminal MTase helps in charging the RNA capping modification and in inhibition of the host innate immune response; meanwhile, the C-terminal RDRP supports the viral RNA synthesis. NS5 interactions with NS3 help in nuclear localization of the virus. Recently, the cocrystalized structure of MTase was reported in complex with S-adenosylmethionine (PDB: 5KQR). Thus, NS5 could be a favorable drug target for discovering new inhibitors that will combat Zika infection in the near future.

Zika as a drug target

Most of the *Flaviviruses* are pathogenic viruses, causing different viral infections in humans worldwide. Recently, ZIKV has emerged as a newcomer to public health concern, eliciting lethal worries. In comparison with dengue virus and West Nile virus, ZIKV presents the highest possibility to provide mechanistic insights for novel drug discovery. With the aid of various computational and experimental analyses, it is possible to approve an effective drug or therapeutic agents which could interfere with viral proteins to treat the infected patient.[42–45] Thus, ZIKV represents the latest druggable target in drug discovery. To date, no drug or vaccine have been established for treating Zika infections, but there is hope for the best, with either or both becoming available at the same time to slow down the Zika threat.

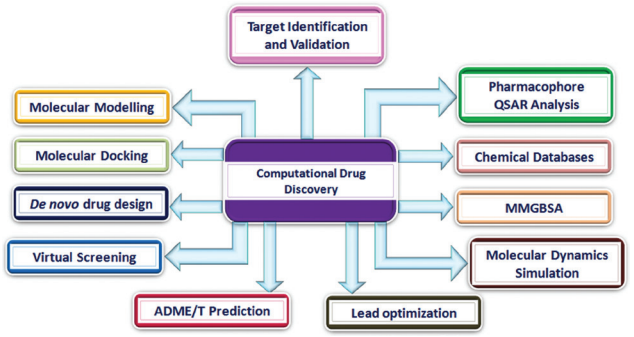

Fig. 2. Approaches of computational drug discovery.

Importance of CDD

Discovering an innovative drug or vaccine to a specific target is one of the greatest tasks in the field of medicine. The main objectives of drug discovery are to first recognize a novel drug molecule and then process the lead discovery of definite candidates, followed by various experimental studies, like pre-clinical and *in vivo*.[46–48] Therefore, introducing a drug into the market is one of the most time consuming (taking 10–15 years) and costliest (US $500–1,000 million) endeavors of drug discovery. Many factors can be responsible for failure in the early or later stages of this endeavor, for example poor pharmacological properties, side effects or lack of effectiveness. Consequently, it is advantageous to use computational approaches to facilitate the discovery of novel drug candidates within a short period of time and without spending much money before proceeding onto experimental clinical trials.

The reported compounds in drug discovery that have been identified using computational approaches offer an excellent platform toward the prediction of therapeutic agents.[49–52] Thus, the current scenario of CDD is aimed at a special contribution for the development of novel inhibitors in a very short period. CDD has already had tremendous impact on the understanding of medicinal science. CDD approaches have yielded excellent and speedy responses in different stages of antiviral drug discovery, and have helped in analyses of the biological as well as the medical data. As such, these methods have become an integral part of medicinal drug discovery and are believed to be complimentary to experimental studies.[53–55]

Herein, we have presented the idea of CDD approaches playing a role in the discovery of antiviral inhibitors for ZIKV. To date, there are no approved specific drugs or vaccines to fight against Zika infection. The major studies on biological targets of ZIKV (prM, E and NS) and their biological mechanisms using computational approaches, such as homology modelling, molecular docking, high-throughput virtual screening, *in silico* absorption, distribution, metabolism and excretion predictions, and molecular dynamics simulation may provide an opportunity to lead the drug discovery process.[56–58] The general concept of CDD methods is shown in Figure 2, and a list of all computational software

Table 2. Computational software and databases used in drug discovery[59–73]

Computational Approach	Software or Server Name	Remarks
Homology or molecular modeling	Modeller, SWISS-MODEL	Helps in prediction of protein structure
Molecular docking & virtual screening	AutoDock Vina, Schrodinger Suite	Docking is utilized for the receptor-ligand interaction and screening helps in searching for a new compound candidate aimed towards the specific biological target
Chemical libraries	Chembridge, Drug Bank, Lifechemicals, Maybridge, Zinc	Collection of chemical compounds for use in screening
ADME/T	Swiss ADME, admetSAR	For the prediction of pharmacokinetics and drug likeness properties of chemical compounds
Molecular dynamics simulation	Amber, Desmond, Gromacs	For obtaining the dynamic behavior and stability of a biomolecular system
QSAR	Schrodinger Suite	Shows the structural descriptors of a compound
MMGBSA	Schrodinger Suite	To compute binding free energy
De novo drug design	e-LEAD3D	Designing new-fangled from the beginning

Abbreviations: ADME/T, absorption, distribution, metabolism, excretion and toxicity; QSAR, quantitative structure–activity relationship.

available for drug discovery, including databases, is given in Table 2.[59–73]

Homology modeling is a well-defined process for generating the structural properties of a biological target.[74] Ekins and colleagues modeled all 15 proteins of ZIKV (NS5, FtsJ, NS4B, NS4A, HELICc, DEXDc, peptidase S7, NS2B, NS2A, NS1, E stem, glycoprotein M, propeptide, capsid and glycoprotein E) using homology modeling methodology,[75] providing a detailed structural knowledge of ZIKV for further understanding of its genomic, proteomic and molecular mechanisms to aid in the development of drug discovery. Molecular docking is a versatile approach in drug discovery, and could provide an informational, accruable geometry and stable interaction of the ligand within the targeted protein. A challenging task of finding a novel drug candidate can be overcome by high-throughput virtual screening, which may be helpful in screening of large chemical libraries based on the structural and ligand information. At present, many of molecules have been identified using molecular docking and a virtual screening process for the NS proteins of ZIKV.

Many drugs may fail in the early or later stages of drug development, due to poor pharmacokinetics and toxicity, with high cost to the industries. Thus, the *in silico* absorption, distribution, metabolism, excretion and toxicity properties' prediction method can be utilized to find a successful drug without facing any issues during experimental studies. In drug discovery, molecular dynamics simulation can be used to investigate the internal interactions of protein-ligand complexes and to access the dynamic behavior with conformational changes under a physiological environment. A few more computational approaches, like quantitative structure–activity relationship (QSAR), MMGBSA and *de novo* drug design can be very useful in current drug discovery of ZIKV inhibitors. QSAR may represent the relations of structural descriptor properties (electronic, hydrophobic, steric and topologic) of a compound with biological activity. MMGBSA is valuable for computing the relative binding free energy for improving the docking score in a biomolecular system. *De novo* drug design, which means "from new or fresh fangled" in the Latin language, allows for processing the new-fangled with desired pharmacological properties from the beginning.[76–83] Based on informative knowledge, we suggest that the competing drug discovery task using computational approaches will involve higher configuration of a computing facility, like a graphical processing unit, cluster-based workstation or supercomputers. Herein, the key principle of CDD approaches can be illustrated for future drug designing to combat ZIKV infection.

Recent discoveries of antiviral inhibitors for ZIKV inhibition

In the 21st century, discovering a new drug is one of the most demanding fields of medicinal science, but is necessary to fight such viral diseases as Dengue, Ebola, human immunodeficiency virus and influenza. Population growth and environmental vectors present an opportunity to these emerging viruses and should be recognized as a major warning to civilization. In the race of these emerging viruses, the latest epidemic newcomer *"*Zika*"* has entered society and established itself as a severe cause of microcephaly, neurological problems and autoimmune disorders.[1–3] But, there is a reminder from the last 20 years that the antiviral therapeutic agents have become a safer means by which to respond to a viral emergency.

Some of the recent remarkable discoveries of antiviral therapeutics have provided great hope to slowing down the global spread of ZIKV infection. The first defense against ZIKV is the inhibition of virus entry into the host cell, and this was successfully inhibited by an active compound, epigallocatechin gallate, derived from a green tea molecule in Vero E6 cells and showing high antiviral activity. The molecular mechanism of inhibition was clearly explained by computational studies, using molecular docking and dynamics simulation processes. Many researchers have studied the epigallocatechin gallate as a nutrient-rich agent without adverse results.[84,85] Another more potent peptide inhibitor (GQASNGVFVIHWGKFDSFGIAV) from the Japanese encephalitis virus has a reported IC50 of 3.93 nM for inhibiting ZIKV.[86] Recently, two zinc molecules, ZINC33683341 and ZINC49605556, were identified by CDD as effective candidates against the ZIKV envelope protein for blocking receptor binding, possibly representing future drug candidates.[87] Finally, nanchangmycin, a polyether from *Streptomyces nanchangensis*, was also investigated for its ability to inhibit ZIKV using human osteosarcoma cells (U2OS).[88]

Table 3. List of effective inhibitors against ZIKV

Sl. No.	Compound/Drug Molecule	Mode of Action	References
1.	Epigallocatechin gallate	Inhibition of virus entry into the host cell	84,85
2.	GQASNGVFVIHWGKFDSFGIAV	Peptide inhibitor for viral infection	86
3.	ZINC33683341/ ZINC49605556	Blocking receptor binding	87
4.	Nanchangmycin	Inhibitor of ZIKV entry	88
5.	6-methylmercaptopurine riboside	Inhibit viral replication	89
6.	Sofosbuvir	Inhibit the polymerase activity of NS5 RDRP	90,91
7.	NITD008	Premature termination of RNA synthesis	92
8.	Sinefungin	Inhibit the methyltransferase activity of viral replication	93
9.	Tetrapeptide-boronic acid, berberin, and myricetin	Inhibition of NS2B-NS3 protease activity of ZIKV	94–96
10.	Gemcitabine and 5-fluorouracil	Blocking pyrimidine biosynthesis	97,98
11.	Bortezomib, mycophenolic acid, and daptomycin (Federal Drug Administration-approved drugs)	Inhibits ZIKV	99
12.	Obatoclax	Reducing acidity of endolysosomal vesicles	100
13.	Niclosamide	Inhibit ZIKV replication	101
14.	T-705 and T-1105	Inhibit ZIKV replication	102
15.	Nitazoxanide	AntiZIKV potential, probably through targeting of the viral postattachment step	103

Abbreviations: NS, non-structural protein; RDRP, RNA-dependent RNA polymerase; ZIKV, Zika virus.

De Carvalho and co-workers identified a thiopurine nucleoside analogue from the prodrug azathioprine[89]: 6-methylmercaptopurine riboside as a potent drug to inhibit the viral replication of ZIKV, showing its remarkable antiviral activity in both Vero and human neuronal cell lines. Some other analogues, such as sofosbuvir and NITD008, have also been shown as potent inhibitors capable of inhibiting the polymerase activity of NS5 RDRP and affecting the premature termination of RNA synthesis, respectively. A potent antifungal and antiparasitic molecule, sinefungin (a derivative of adenosine), has been reported as a novel inhibitor toward the inhibition of MTase activity in ZIKV replication.[90–93]

A few potent inhibitors, like tetrapeptide-boronic acid, berberin and myricetin, have been analyzed computationally as well as experimentally for their ability to inhibit NS2B-NS3 protease activity in ZIKV.[94–96] Two more inhibitors, gemcitabine and 5-fluorouracil, have been reported to impede pyrimidine biosynthesis to exert ZIKV inhibition.[97,98] In searching for antiviral inhibitors against ZIKV, three Federal Drug Administration-approved drugs (bortezomib, mycophenolic acid and daptomycin) have been shown as effective for the inhibition of ZIKV, which suggests their utility as future resources for drug designing.[99] A list of effective inhibitors against the druggable targets of ZIKV is presented in Table 3.[84–103]

Conclusions and future outlook

Globally, people are facing old, newly-developing or redeveloping viral diseases, and have been since the beginning of the century. These include Dengue, influenza, chikungunya and human immunodeficiency virus, and now Zika. Previously, Zika was considered as innocuous, but the speedy outbreak of ZIKV to pandemic status has become a tantalizing mystery for the world (to find a way to slow down it). According to WHO, the current scenario represents Zika as a major challenging concern to public health, and soon it may spread over the entire world through its vectors (mainly, the *Aedes* mosquito species); this spread may be helped along by travelling of humans worldwide.

Despite enormous research efforts in the last few years, the following questions remain unanswered: "*What kind of genetic characteristics does ZIKV have?";* "*What are the other vectors that support Zika virulence?*"; and "*How do we combat Zika?*". The pathological and biological mechanisms are still not fully identified, leaving us with an incomplete understanding of the growth of ZIKV infection rates. Moreover, many Zika viral structures are unsolved, mainly for the NS proteins. Many of the molecules identified as having antiZIKV activity have failed in experimental testing, and no drug or vaccine has yet been able to come onto the market. Continued strong research is necessary for developing a superior drug against ZIKV, with less side effects. Thus, improved diagnostic techniques and effective therapeutics may be beneficial to avert Zika infection.

The rapid advancement of computational studies for drug discovery can also help in uncovering the detailed mechanisms of ZIKV pathogenesis. CDD provides rapid procedures to screen the relevant information for a new drug, including its pharmacokinetic properties and biological effects. The implementation of the described tools and techniques in this review can be valuable for identifying and evaluating biological drug targets of ZIKV on the genomic, proteomic and molecular levels to help push forward the design of a potent drug candidate. In addition, appropriate prevention of mosquito bites and local vector control could be helpful to avert infection.

This review has summarized the worldwide outbreak of Zika, describing the structure of the genome, ZIKV transmission, important biological proteins, and the developed inhibitors of ZIKV. Now is the time for researchers, industry, funding agencies and policy makers to make history in medicinal science by improving our ability to combat Zika worldwide.

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Conflict of interest

The authors have no conflicts of interest related to this publication.

Author contributions

Conception and performance of the present study and writing of the manuscript (UP, SKS).

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