RESEARCH ARTICLE

REVISED Virtual screen for repurposing approved and experimental drugs for candidate inhibitors of EBOLA virus infection [version 2; peer review: 2 approved]

Veljko Veljkovic¹, Philippe M. Loiseau ², Bruno Figadere ², Sanja Glisic¹, Nevena Veljkovic¹, Vladimir R. Perovic¹, David P. Cavanaugh ³, Donald R. Branch⁴

¹Center for Multidisciplinary Research, University of Belgrade, Institute of Nuclear Sciences VINCA, P.O. Box 522, 11001 Belgrade, Serbia

²Antiparasitic Chemotherapy, UMR 8076 CNRS BioCIS, Faculty of Pharmacy Université Paris-Sud, Rue Jean-Baptiste Clément, F 92290- Chatenay-Malabry, France

³Bench Electronics, Bradford Dr., Huntsville, AL, 35801, USA

⁴Canadian Blood Services, Center for Innovation, 67 College Street, Toronto, Ontario, M5G 2M1, Canada

V2 First published: 02 Feb 2015, 4:34
<https://doi.org/10.12688/f1000research.6110.1>

Latest published: 16 Feb 2015, 4:34
<https://doi.org/10.12688/f1000research.6110.2>

Abstract

The ongoing Ebola virus epidemic has presented numerous challenges with respect to control and treatment because there are no approved drugs or vaccines for the Ebola virus disease (EVD). Herein is proposed simple theoretical criterion for fast virtual screening of molecular libraries for candidate inhibitors of Ebola virus infection. We performed a repurposing screen of 6438 drugs from DrugBank using this criterion and selected 267 approved and 382 experimental drugs as candidates for treatment of EVD including 15 anti-malarial drugs and 32 antibiotics. An open source Web server allowing screening of molecular libraries for candidate drugs for treatment of EVD was also established.

Keywords

Ebola virus , drug candidates , entry inhibitors , virtual screening

This article is included in the **Emerging Diseases and Outbreaks** gateway.

Open Peer Review

Approval Status

	1	2
version 2		
(revision)
16 Feb 2015
version 1
02 Feb 2015

1. **Patrick Butaye**, Ross University, Basseterre, Saint Kitts and Nevis

2. **Bruno Botta**, Sapienza University of Rome, Rome, Italy

Any reports and responses or comments on the article can be found at the end of the article.

This article is included in the **Ebola Virus** collection.

Corresponding author: Veljko Veljkovic (vv@vinca.rs)

Competing interests: No competing interests were disclosed.

Grant information: This work was supported by the Ministry of Education, Science and Technological Development of the Republic of Serbia (Grant no. 173001).

Copyright: © 2015 Veljkovic V *et al.* This is an open access article distributed under the terms of the [Creative Commons Attribution License](#), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. Data associated with the article are available under the terms of the [Creative Commons Zero "No rights reserved" data waiver](#) (CC0 1.0 Public domain dedication).

How to cite this article: Veljkovic V, Loiseau PM, Figadere B *et al.* **Virtual screen for repurposing approved and experimental drugs for candidate inhibitors of EBOLA virus infection [version 2; peer review: 2 approved]** F1000Research 2015, 4:34 <https://doi.org/10.12688/f1000research.6110.2>

First published: 02 Feb 2015, 4:34 <https://doi.org/10.12688/f1000research.6110.1>

REVISED Amendments from Version 1

According to Referees' suggestions we have made the following changes: (i) the explanation why some compounds from Ref. 3 are not included in Dataset1 was added in Material and methods, (ii) the explanation of the AQVN/ISM concept is moved from the Material and methods to Introduction. (iii) Results and discussion is divided in two separate sections.

See referee reports

Introduction

The current Ebola virus outbreak is one of the largest outbreaks of its kind in history and the first in West Africa. By January 14, 2015, a total of 21296 probable and confirmed cases, including 8429 deaths from Ebola virus disease (EVD), had been reported from five countries in West Africa - Guinea, Liberia, Nigeria, Senegal, and Sierra Leone (http://apps.who.int/iris/bitstream/10665/148237/2/roadmapsitrepre_14Jan2015_eng.pdf?ua=1). EVD with a high case-fatality rate of 40% and with currently no approved vaccine or therapy, represents a major public health threat.

In response to the current Ebola virus outbreak, the international community has urged for accelerated development of drugs against EVD but also has endorsed the clinical use of unregistered treatments for Ebola¹. Conventional time and the money consuming approach of drug development (> 10 years; > 2 billions \$) does not meet the current urgent need for anti-Ebola drugs. Repurposing or repositioning of existing drugs could overcome some of these obstacles and help in the rapid discovery and development of therapeutics for EVD, although this approach does not negate the need for some preclinical studies and clinical trials for validation of the proposed indications. Recently, results of two large repurposing screenings of Food and Drug Administration (FDA)-approved drugs have been reported. In the first study, Madrid and co-workers performed *in vitro* and *in vivo* (in mice) screening of 1012 FDA-approved drugs and selected 24 candidate entry inhibitors for Ebola virus². In the second study, 53 inhibitors of Ebola virus infection with $IC_{50} < 10 \mu M$ and selectivity index $SI > 10$ -fold have been identified by *in vitro* screening of 2816 FDA-approved drugs³. In the same study, an additional 95 drugs which are active against Ebola virus infection with $IC_{50} > 10 \mu M$ and $SI < 10$ -fold were also reported.

Although *in vitro* and *in vivo* screening for repurposing/repositioning of existing drugs could significantly accelerate discovery of new drugs these approaches are time-consuming and costly for screening of large drug libraries. Recently, we proposed a novel approach for *in silico* screening of molecular libraries for drug candidates⁴⁻⁸. This approach, which uses the average quasi valence number (AQVN) and the electron-ion interaction potential (EIIP), parameters determining long-range interaction between biological molecules, might hold a key to overcoming some of these obstacles in experimental screening by significantly reducing the number of compounds which should be *in vitro* and *in vivo* tested⁹. Among 3300 currently used molecular descriptors, AQVN and EIIP represent the unique physical properties which characterize the long-range interactions

between biological molecules⁴³. Small molecules with similar AQVN and EIIP values interact with the common therapeutic target, which allow establish criterions for virtual screening of molecular libraries for compounds with similar therapeutic properties⁴⁻⁹.

Here we develop the EIIP/AQVN-based criterion for virtual screening of molecular libraries for candidate drugs against Ebola virus infection. Using this criterion we screened DrugBank (<http://www.drugbank.ca>) and selected 267 approved and 382 experimental drugs as candidates for treatment of EVD. An open access portal allowing screening of molecular libraries for candidate drugs for treatment of EVD was established.

Material and methods

Molecular libraries

For screening of drugs for repurposing to select candidates for Ebola virus entry inhibitors, 1463 approved and 4975 experimental drugs from DrugBank (<http://www.drugbank.ca>) were screened. For development of the predictive criterion used in this analysis, the learning set (Dataset 1) encompassing 152 drugs which are selected as inhibitors of Ebola virus infection by *in vitro* and *in vivo* screening of 3828 FDA-approved drugs^{2,3}, was established. From this Dataset are excluded microtubule modulators from Ref. 3, which do not directly block the entry of Ebola virus, as well as compounds which previously were reported in Ref. 2. As control data sets 45,010,644 compounds from PubChem (<http://www.ncbi.nlm.nih.gov/pccompound>) and 49 Ebola virus entry inhibitors collected by data mining of literature and patents, were used. For screening of literature data the NCBI literature database PubMed (<http://www.ncbi.nlm.nih.gov/pubmed>) was used. For search of patents and patent applications we used the Free Patent Online browser (<http://www.freepatentsonline.com>).

Calculation of AQVN and EIIP parameters of organic molecules

Specific recognition and targeting between interacting biological molecules at distances > 5 Å are determined by the average AQVN and the EIIP¹⁰, which are derived from the general model pseudopotential^{11,12}. These parameters for organic molecules are determined by the following simple equations¹⁰:

$$EIIP = 0.25 \frac{Z^* \sin(1.04 \pi Z^*)}{2\pi} \quad (1)$$

Where Z^* is the average quasi-valence number (AQVN) determined by

$$Z^* = \frac{1}{N} \sum_{i=1}^m n_i Z_i \quad (2)$$

where Z_i is the valence number of the i -th atomic component, n_i is the number of atoms of the i -th component, m is the number of atomic components in the molecule, and N is the total number of atoms. EIIP values calculated according to equation 1 and equation 2 are expressed in Rydberg units (Ry).

Results

Previously, analyses of the EIIP/AQVN distribution of 45,010,644 compounds from the PubChem database (<http://www.ncbi.nlm.nih.gov/pccompound>) revealed that 92.5% of presented compounds are homogeneously distributed within EIIP and AQVN intervals (0.00 – 0.11 Ry) and (2.4 – 3.3), respectively). This domain of the EIIP/AQVN space, encompassing the majority of known chemical compounds, is referred to as the “basic EIIP/AQVN chemical space” (BCS)⁶. Analysis of the molecular training set (Dataset 1), encompassing 152 small molecule inhibitors of Ebola virus infection selected by *in vitro* screening of 3828 FDA approved drugs^{2,3}, show that 79% of these compounds are placed within AQVN and EIIP region (2.3 – 2.7) and (0.0829 – 0.0954 Ry), respectively (“Ebola Virus Infection Inhibitors Space”, EVIIS). The AQVN region (2.36 – 2.54) and the EIIP region (0.0912 – 0.0924 Ry) form the part of EVIIS which encompasses 55.5% of all drugs from the learning set (core EVIIS, cEVIIS). Literature data mining reveals 49 compounds with experimentally proved activity against Ebola virus infection (Table 1)^{13–29}. Most of these compounds 47 (95.9%) are placed within EVIIS (Table 1). Of note is that EVIIS and cEVIIS domains contain only 14.6% and 6.5% of compounds from PubChem, respectively. This confirms high specificity of clustering of Ebola virus infection inhibitors within the EIIP/AQVN space. Comparison of distributions of Ebola virus infection inhibitors and compounds from PubMed is given in Figure 1.

Dataset 1. FDA-approved drugs which are active against Ebola virus infection^{2,3}

<http://dx.doi.org/10.5256/f1000research.6110.d42876>

AQVN: average quasivalence number; EIIP: electron-ion interaction potential

It was shown that Ebola virus glycoprotein (GP)-mediated entry and infection is subordinated with a membrane-trafficking event that translocates a GP binding partner to the cell surface, which depends on microtubules^{30,31}. Consistently, microtubule inhibitors which block this trafficking process could decrease infection without interfering with the direct binding and translocation of the Ebola virus into cells. AQVN and EIIP values of microtubule modulators and transcription inhibitors with reported anti-Ebola virus activity are given in Table 2. As can be seen, all these compounds, which do not directly affect binding and internalization of Ebola virus, are located outside of EVIIS. This additionally confirms the specificity of the EVIIS domain.

In further analysis we used EVIIS as a filter for virtual screening for candidate Ebola virus infection inhibitors. In Dataset 2 622 approved and 1089 experimental drugs in Dataset 3 selected by EVIIS screening of 6532 drugs from DrugBank are reported. Using cEVIIS, we located 267 approved and 382 experimental drugs. This small molecular library represents a source of candidate drugs for treatment of Ebola virus disease (EVD), which can be further experimentally tested.

Figure 1. Distribution of compounds according to their average quasivalence number (AQVN) and electron-ion interaction potential (EIIP) values. (A) 45010644 compounds from the PubChem database (<http://www.ncbi.nlm.nih.gov/pccompound>); (B) FDA-approved drugs which are active against Ebola virus infection (Dataset 1)^{2,3}; (C) Entry inhibitors of Ebola virus (Table 1).

Table 1. Small-molecule entry inhibitors for Ebola virus.

Compound	Formula	AQVN	ELIP [Ry]	Reference
Chloroquine	C18H26ClN3	2.375	0.0941	16
Bafilomycin A1	C35H58O9	2.471	0.0960	17
Cytochalasin B	C29H37NO5	2.611	0.0810	17
Cytochalasin D	C30H37NO6	2.676	0.0672	17
Latruculion A	C22H31NO5S	2.667	0.0693	17
Jasplakinolide	C36H45BrN4O6	2.674	0.0676	18
Clomiphene	C26H28ClNO	2.526	0.0926	18
Toremifene	C26H28ClNO	2.526	0.0926	18
Chlorpromazine	C17H19ClN2S	2.600	0.0829	19
Amiodarone	C25H29I2NO3	2.567	0.0880	20
Dronedarone	C31H44N2O5S	2.578	0.0864	20
Verapamil	C27H38N2O4	2.535	0.0917	20
Clomiphene	C26H28ClNO	2.526	0.0926	21
AY-9944	C22H28Cl2N2	2.370	0.0938	21
Ro 48-8071	C23H27BrFNO2	2.505	0.0940	21
U18666A	C25H41NO2	2.290	0.0849	21
Terconazole	C26H31Cl2N5O3	2.687	0.0644	21
Triparanol	C27H32ClNO2	2.508	0.0941	21
Impramine	C19H24N2	2.444	0.0964	22
3.47	C34H43N3O5	2.635	0.0763	23
Cytochalasin B	C29H37NO5	2.611	0.0810	24
Cytochalasin D	C30H37NO6	2.676	0.0672	24
Latrunculin A	C22H31NO5S	2.667	0.0693	24
Jasplakinolide	C36H45BrN4O6	2.674	0.0676	24
NSC62914	C31H40O3	2.460	0.0962	25
Compound 1	C30H38N6O2	2.632	0.0770	26
Compound 2	C32H46N6	2.429	0.0963	26
Compound 3	C28H34N6O2	2.686	0.0635	26
Compound 5	C42H58N10O6	2.690	0.0635	27
Compound 8a	C17H23N3O3	2.695	0.0621	27
Compound 8b	C17H23N3O3	2.695	0.0621	27
Compound 8y	C16H20BrNO2	2.610	0.0812	27
Compound 15h	C15H20JN5O	2.667	0.0693	27
Compound 15k	C15H128Br3N5O	2.667	0.0693	27
Retinazone	C38H56Na3N5S2	2.385	0.0947	28
Compound 7	C17H12F4N2	2.467	0.0647	29
Brincidofovir*	C27H52N3O7P	2.467	0.0961	30
Hit compound 3	C25H35N3O2	2.494	0.0950	31
Hit compound 3.1	C21H24ClN3O2	2.667	0.0693	31
Hit compound 3.2	C20H29N3O2	2.518	0.0933	31
Hit compound 3.3	C30H35N3O2	2.556	0.0894	31
Hit compound 3.4	C25H32N4O3	2.656	0.0717	31
Hit compound 3.5	C20H23N3O2	2.708	0.0587	31
Hit compound 3.6	C25H33N3O2	2.540	0.09913	31
Hit compound 3.7	C22H27N3O3	2.691	0.0633	31
Hit compound 3.18	C26H37N3O2	2.471	0.0471	31
Hit compound 3.48	C34H43N3O5	2.635	0.0763	31
Hit compound 3.105	C34H40N6O2	2.658	0.0712	31
NSC 62914	C31H39O3	2.480	0.0957	32

*Experimental drug applied for treatment of Ebola patients in Liberia (<http://www.ox.ac.uk/news/2014-11-13-oxford-lead-trial-experimental-drug-ebola-patients>)

Table 2. Viral transcription inhibitors and microtubule modulators with anti-Ebola virus activity.

Compound	Formula	AQVN	EIIP [Ry]
Viral transcription inhibitors			
BCX4430	C11H15N5O3	3.000	0.0439
Favipiravir	C5H4FN3O2	3.467	0.1304
C-c3Ado	C12H16N4O3	2.914	0.0112
c3Nep	C12H14N4O3	3.030	0.0552
"D-like" 1'-6'-isoneplanocin	C11H12N5O3	3.194	0.1076
"L-like" 1'-6'-isoneplanocin	C11H12N5O3	3.194	0.1076
CMLDBU3402	C30H26BrN3O7	3.045	0.1343
Microtubule modulators			
Vinblastine	C13H8Cl2N2O4	3.310	0.0130
Vinorelbine	C45H54N4O8	2.721	0.0552
Vincristine	C46H56N4O10	2.759	0.0439
Colchicine	C22H25NO6	2.852	0.0121
Nocodazole	C14H11N3O3S	3.312	0.1298
Mebendazole	C16H13N3O3	3.143	0.0934
Albendazole	C12H15N3O2S	2.909	0.0092

Dataset 2. Approved and experimental drugs selected as candidate for treatment of EVD

<http://dx.doi.org/10.5256/f1000research.6110.d42877>

AQVN: average quasivalence number; EIIP: electron-ion interaction potential

Dataset 3. Experimental drugs selected as candidate for treatment of EVD

<http://dx.doi.org/10.5256/f1000research.6110.d42878>

AQVN: average quasivalence number; EIIP: electron-ion interaction potential

Discussion

Madrid and co-workers selected 24 drugs by *in vitro* screening of 1012 FDA-approved drugs, which are effective against Ebola virus infection². They also showed that among these compounds, four antimalarial drugs (chloroquine, hydroxychloroquine, amodiaquine and aminoquinoline-13) also are effective against Ebola virus infection *in vivo*². Among 53 compounds which effectively inhibit Ebola virus infection *in vitro*, which Kouznetsova and co-workers selected from 2816 approved drugs, are also three antimalarial drugs (mefloquine, chloroquine, amodiaquine)³. It was also suggested that application of chloroquine for prevention of virus transmission should be considered because this compound significantly inhibits Ebola virus infection¹³. Our analysis showed that 15 of 22 approved ant-malarial drugs (http://en.wikipedia.org/wiki/Antimalarial_medication) are located in EVIIS (Table 3).

Six 2-alkylquinolines have been also included in this study. This chemical series is promising as some derivatives exhibited anti-viral activity such as 2PQ, and 2QQ^{32,33} antimalarial activity such as 2PQ and 2PentQ2³⁴, antileishmanial activity such as 2PQ^{35,36} and neurotrophin-like activity on dopaminergic neurons such as 2QI15³⁷. These compounds exhibit some advantages in regard to their chemical synthesis with few steps and good yields as well as their chemical stability in tropical conditions of storage. Their combined effects against virus and Leishmania parasites suggested they could be an advantage for the treatment of Leishmania/HIV co-infections and they were considered as attractive enough to enter the pipeline of DNDi on 2010.

All these data strongly suggest that this class of drugs should be further investigated as a promising source of therapeutics for treatment of EVD. Anti-malarial drugs with dual activity should be of special interest because malaria represents the highest health-related disease in African countries with EVD.

Among 3828 FDA-approved drugs screened for anti-Ebola activity were six antibiotics which inhibit Ebola virus infection (azithromycin, erythromycin, spiramycin, dirithromycin, maduramicin, charitromycin)^{2,3}. All these antibiotics are within EVIIS and four of them are in cEVIIS. Analysis of 184 approved antibiotics (Dataset 4) showed that only 32 (17.4%) have AQVN and EIIP values in EVIIS, and that 11 of them are located within cEVIIS. Previously we reported domains of AQVN and EIIP which characterize different classes of antibiotics (Table 4)⁶. According to these data, among antibiotics some macrolides, pleuromutilins and aminoglycosides have the highest chance for inhibition of Ebola virus infection. Of note is that five of six antibiotics with experimentally proved

activity against Ebola virus infection (azithromycin, erythromycin, spiramycin, dirithromycin, clarithromycin) are macrolides. Antibiotics representing candidate Ebola virus infection inhibitors selected by EIIP/AQVN criterion are given in [Table 5](#).

Dataset 4. Approved antibiotics screened for candidate anti-Ebola drugs

<http://dx.doi.org/10.5256/f1000research.6110.d42879>

AQVN: average quasivalence number; EIIP: electron-ion interaction potential

Previous, we determined AQVN and EIIP domains characterizing different classes of anti-HIV drugs⁴⁻⁹. As can be seen in [Table 6](#), the EIIP/AQVN domain of CCR5 HIV entry inhibitors is within EVIIS, and domains of CXCR4 HIV entry inhibitors and HIV protease inhibitors partially overlaps EVIIS. The EIIP/AQVN domains of other classes of anti-HIV agents are located outside EVIIS. This indicates that some HIV entry inhibitors and HIV protease inhibitors could also be effective drugs against Ebola virus infection.

In conclusion, the presented results show that the EIIP/AQVN criterion can be used as an efficient filter in virtual screening of molecular libraries for candidate inhibitors of Ebola virus infection. Approved ([Dataset 2](#)) and experimental drugs ([Dataset 3](#)), anti-malarial

Table 3. Approved anti-malarial drugs selected as candidate drugs for EVD.

Compound	Formula	AQVN	EIIP [Ry]
Quinine	C ₂₀ H ₂₄ N ₂ O ₂	2.625	0.0784
Chloroquine	C ₁₈ H ₂₆ ClN ₃	2.375	0.0941
Amodiaquine	C ₂₀ H ₂₂ ClN ₃ O	2.638	0.0756
Proguanil	C ₁₁ H ₁₆ ClN ₅	2.606	0.0819
Mefloquine	C ₁₇ H ₁₆ F ₆ N ₂ O	2.524	0.0928
Primaquine	C ₁₅ H ₂₁ N ₃ O	2.600	0.0829
Halofantrine	C ₂₆ H ₃₀ Cl ₂ F ₃ NO	2.381	0.0945
Clindamycin	C ₁₈ H ₃₃ ClN ₂ O ₅ S	2.533	0.0919
Artemether	C ₁₆ H ₂₆ O ₅	2.553	0.0897
Piperaquine	C ₂₉ H ₃₂ Cl ₂ N ₆	2.609	0.0814
Artemotil	C ₁₇ H ₂₈ O ₅	2.520	0.0931
Dihydroartemisinin	C ₁₅ H ₂₄ O ₅	2.591	0.0844
Quinidine	C ₂₀ H ₂₄ N ₂ O ₂	2.625	0.0784
Cinchonidine	C ₁₉ H ₂₂ N ₂ O	2.591	0.0844
Artemisin	C ₁₅ H ₂₂ O ₅	2.667	0.0693

Table 4. AQVN and EIIP range of different antibiotics classes⁶.

Antibiotic class	AQVN	EIIP [Ry]
Penicillins	2.975 - 3.180	0.035 - 0.124
Cephalosporins	3.071 - 3.473	0.070 - 0.130
Carbapenems & Penems	2.973 - 3.059	0.022 - 0.066
Monobactams	3.166 - 3.581	0.100 - 0.134
Quinolines	2.760 - 3.060	0.003 - 0.065
Aminoglycosides	2.552 - 2.820	0.024 - 0.084
Tetracyclines	2.933 - 3.111	0.018 - 0.084
Macrolides	2.467 - 2.630	0.077 - 0.096
Pleuromutilins	2.395 - 2.473	0.095 - 0.096
Nitrofurans	3.652 - 3.826	0.010 - 0.086

Table 5. Antibiotics selected as candidate drugs for EVD.

Antibiotics	Formula	AQVN	EIIP [Ry]
Tiamulin	$C_{28}H_{47}NO_4S$	2.395	0.095
Retapamulin	$C_{30}H_{47}NO_4S$	2.434	0.096
Valnemulin	$C_{31}H_{52}N_2O_5S$	2.440	0.096
Azithromycin	$C_{38}H_{72}N_2O_{12}$	2.468	0.096
BC-3205	$C_{32}H_{51}N_2O_5S$	2.472	0.096
Dirithromycin	$C_{42}H_{78}N_2O_{14}$	2.500	0.095
Clarithromycin	$C_{38}H_{69}NO_{13}$	2.512	0.094
Surfactin	$C_{53}H_{93}N_7O_{13}$	2.518	0.093
Erythromycin	$C_{37}H_{67}NO_{13}$	2.525	0.093
Clindamycin	$C_{18}H_{33}ClN_2O_5S$	2.533	0.092
Roxithromycin	$C_{41}H_{76}N_2O_{15}$	2.537	0.092
Oleandomycin	$C_{35}H_{61}NO_{12}$	2.550	0.090
Gentamicin	$C_{21}H_{43}N_5O_7$	2.553	0.090
Spiramycin	$C_{43}H_{74}N_2O_{14}$	2.556	0.089
Mupirocin	$C_{26}H_{44}O_9$	2.557	0.089
Lincomycin	$C_{18}H_{34}N_2O_6S$	2.590	0.085
Netilmicin	$C_{21}H_{41}N_5O_7$	2.595	0.084
Astromicin	$C_{17}H_{35}N_5O_6$	2.603	0.082
Tylosin	$C_{46}H_{77}NO_{17}$	2.610	0.081
Kitasamycin	$C_{35}H_{59}NO_{13}$	2.611	0.081
Josamycin	$C_{42}H_{69}NO_{15}$	2.614	0.080
Telithromycin	$C_{43}H_{65}N_5O_{10}$	2.618	0.080
Telithromycin	$C_{43}H_{65}N_5O_{10}$	2.618	0.080
Verdamycin	$C_{20}H_{39}N_5O_7$	2.620	0.080
Midecamycin	$C_{41}H_{67}NO_{15}$	2.629	0.078
Troleandomycin	$C_{41}H_{67}NO_{15}$	2.629	0.078
Sisomicin	$C_{19}H_{37}N_5O_7$	2.647	0.074
Cethromycin	$C_{42}H_{59}N_3O_{10}$	2.649	0.073
Carbomycin A	$C_{42}H_{67}NO_{16}$	2.667	0.069
Dibekacin	$C_{18}H_{37}N_5O_8$	2.676	0.067
Echinocandin B	$C_{52}H_{81}N_7O_{16}$	2.692	0.063
Rifabutin	$C_{46}H_{62}N_4O_{11}$	2.699	0.061

Table 6. AQVN and EIIP range of anti-HIV drugs⁶.

Target	AQVN	EIIP [Ry]
CXCR4	2.16 - 2.53	0.062 - 0.096
CCR5	2.42 - 2.63	0.079 - 0.099
PI	2.61 - 2.78	0.040 - 0.080
NRTI/NtRTI	2.92 - 3.20	0.040 - 0.100
INI	3.00 - 3.20	0.044 - 0.116
Anti-HIV flavonoids	3.34 - 3.59	0.110 - 0.135

drugs (Table 3) and antibiotics (Table 5) selected by this criterion represents a valuable source of candidate therapeutics for treatment of EVD, some of which are already approved by FDA for treatment of other diseases which can be repurposed for use in EVD. We hope that these data, obtained by an *in silico* drug repurposing screen, will accelerate discovery of drugs for treatment of EVD, which are necessary in this ongoing emergency situation caused by the current unprecedented Ebola virus outbreak. To enable other researchers working on online EIIP/AQVN-based screening of different sources of small molecules for candidate Ebola drugs, we established an open web server (http://www.biomedconsulting.info/ebola_screen.php).

Data availability

The virtual screen for candidate inhibitors of EBOLA virus infection web tool is available at: <http://www.biomedconsulting.info/tools/ebolascreeen.php>. An archived version can be accessed at: <http://www.webcitation.org/6Vxtuoigx>³⁸

F1000Research: Dataset 1. FDA-approved drugs which are active against Ebola virus infection^{2,3}, [10.5256/f1000research.6110.d42876](https://doi.org/10.5256/f1000research.6110.d42876)³⁹

F1000Research: Dataset 2. Approved and experimental drugs selected as candidate for treatment of EVD, [10.5256/f1000-research.6110.d42877](https://doi.org/10.5256/f1000-research.6110.d42877)⁴⁰

F1000Research: Dataset 3. Experimental drugs selected as candidate for treatment of EVD, [10.5256/f1000research.6110.d42878](https://doi.org/10.5256/f1000research.6110.d42878)⁴¹

F1000Research: Dataset 4. Approved antibiotics screened for candidate anti-Ebola drugs, [10.5256/f1000research.6110.d42879](https://doi.org/10.5256/f1000research.6110.d42879)⁴²

Author contributions

Conceived and designed the study: VV SG NV. Developed the analysis tools: VP. Analyzed the data: VV SG NV DRB PPML BF DPC. Wrote the paper: VV DRB PPML.

Competing interests

No competing interests were disclosed.

Grant information

This work was supported by the Ministry of Education, Science and Technological Development of the Republic of Serbia (Grant no. 173001).

Acknowledgments

The authors would like to gratefully acknowledge networking support by the COST Action CM1307.

References

- Enserink M: **Infectious diseases. Debate erupts on 'repurposed' drugs for Ebola.** *Science*. 2014; **345**(6198): 718–9.
[PubMed Abstract](#) | [Publisher Full Text](#)
- Madrid PB, Chopra S, Manger ID, *et al.*: **A systematic screen of FDA-approved drugs for inhibitors of biological threat agents.** *PLoS One*. 2013; **8**(4): e60579.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Kouznetsova J, Sun W, Martínez-Romero C, *et al.*: **Identification of 53 compounds that block Ebola virus-like particle entry via a repurposing screen of approved drugs.** *Emerg Microb Infect*. 2014; **3**: e84.
[PubMed Abstract](#) | [Publisher Full Text](#)
- Veljkovic V, Veljkovic N, Este J, *et al.*: **Application of the EIIP/ISM bioinformatics concept in development of new drugs.** *Curr Med Chem*. 2007; **14**(4): 441–53.
[PubMed Abstract](#) | [Publisher Full Text](#)
- Veljkovic V, Mouscadet JF, Veljkovic N, *et al.*: **Simple criterion for selection of flavonoid compounds with anti-HIV activity.** *Bioorg Medic Chem Lett*. 2007; **17**(5): 1226–32.
[PubMed Abstract](#) | [Publisher Full Text](#)
- Veljkovic N, Glisic S, Perovic V, *et al.*: **The role of long-range intermolecular interactions in discovery of new drugs.** *Exp Opin Drug Disc*. 2011; **6**(12): 1263–70.
[PubMed Abstract](#) | [Publisher Full Text](#)
- Maga G, Veljkovic N, Crespan E, *et al.*: **New *in silico* and conventional *in vitro* approaches to advance HIV drug discovery and design.** *Exp Opin Drug Discov*. 2013; **8**(1): 83–92.
[PubMed Abstract](#) | [Publisher Full Text](#)
- Veljkovic N, Glisic S, Prljic J, *et al.*: **Simple and general criterion for "*in silico*" screening of candidate HIV drugs.** *Curr Pharm Biotechnol*. 2013; **14**(5): 561–9.
[PubMed Abstract](#) | [Publisher Full Text](#)
- Tintori C, Veljkovic N, Veljkovic V, *et al.*: **Computational studies of the interaction between the HIV-1 integrase tetramer and the cofactor LEDGF/p75: insights from molecular dynamics simulations and the informational spectrum method.** *Proteins*. 2010; **78**(16): 3396–408.
[PubMed Abstract](#) | [Publisher Full Text](#)
- Veljkovic V: **A theoretical approach to preselection of carcinogens and chemical carcinogenesis.** *New York: Gordon & Breach*. 1980.
[Reference Source](#)
- Veljkovic V, Slavic I: **Simple general-model pseudopotential.** *Phys Rev Lett*. 1972; **29**: 105–7.
[PubMed Abstract](#) | [Publisher Full Text](#)
- Veljkovic V: **The dependence of the Fermi energy on the atomic number.** *Phys Lett*. 1973; **45A**(1): 41–2.
[PubMed Abstract](#) | [Publisher Full Text](#)
- Wool-Lewis RJ, Bates P: **Characterization of Ebola virus entry by using pseudotyped viruses: identification of receptor-deficient cell lines.** *J Virol*. 1998; **72**(4): 3155–60.
[PubMed Abstract](#) | [Free Full Text](#)
- Yonezawa A, Cavois M, Greene WC: **Studies of Ebola virus glycoprotein-mediated entry and fusion by using pseudotyped human immunodeficiency virus type 1 virions: involvement of cytoskeletal proteins and enhancement by tumor necrosis factor alpha.** *J Virol*. 2005; **79**(2): 918–26.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Johansen LM, Brannan JM, Delos SE, *et al.*: **FDA-approved selective estrogen receptor modulators inhibit Ebola virus infection.** *Sci Transl Med*. 2013; **5**(190): 190ra79.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Bhattacharyya S, Warfield KL, Ruthel G, *et al.*: **Ebola virus uses clathrin-mediated endocytosis as an entry pathway.** *Virology*. 2010; **401**(1): 18–28.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Gehring G, Rohrmann K, Atenchong N, *et al.*: **The clinically approved drugs amiodarone, dronedarone and verapamil inhibit filovirus cell entry.** *J Antimicrob Chemother*. 2014; **69**(8): 2123–31.
[PubMed Abstract](#) | [Publisher Full Text](#)
- Shoemaker CJ, Schornberg KL, Delos SE, *et al.*: **Multiple cationic amphiphiles induce a Niemann-Pick C phenotype and inhibit Ebola virus entry and infection.** *PLoS One*. 2013; **8**(2): e56265.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Carette JE, Raaben M, Wong AC, *et al.*: **Ebola virus entry requires the cholesterol transporter Niemann-Pick C1.** *Nature*. 2011; **477**(7364): 340–3.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Côté M, Misasi J, Ren T, *et al.*: **Small molecule inhibitors reveal Niemann-Pick C1 is essential for Ebola virus infection.** *Nature*. 2011; **477**(7364): 344–8.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Yonezawa A, Cavois M, Greene WC: **Studies of Ebola virus glycoprotein-mediated entry and fusion by using pseudotyped human immunodeficiency virus type 1 virions: involvement of cytoskeletal proteins and enhancement by tumor necrosis factor alpha.** *J Virol*. 2005; **79**(2): 918–26.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Panchal RG, Reid SP, Tran JP, *et al.*: **Identification of an antioxidant small-molecule with broad-spectrum antiviral activity.** *Antiviral Res*. 2012; **93**(1): 23–9.
[PubMed Abstract](#) | [Publisher Full Text](#)
- Selaković Z, Osenica D, Eaton B, *et al.*: **A limited structural modification results in a significantly more efficacious diazachrysen-based filovirus inhibitor.** *Viruses*. 2012; **4**(8): 1279–88.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Yermolina MV, Wang J, Caffrey M, *et al.*: **Discovery, synthesis, and biological evaluation of a novel group of selective inhibitors of filoviral entry.** *J Med Chem*. 2011; **54**(3): 765–81.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Kesel AJ, Huang Z, Murray MG, *et al.*: **Retinazone inhibits certain blood-borne human viruses including Ebola virus Zaire.** *Antivir Chem Chemother*. 2014; **23**(5): 197–215.
[PubMed Abstract](#) | [Publisher Full Text](#)

26. Basu A, Li B, Mills DM, *et al.*: **Identification of a small-molecule entry inhibitor for filoviruses.** *J Virol.* 2011; **85**(7): 3106–19.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
27. Bishop BM: **Potential and emerging treatment options for Ebola virus disease.** *Ann Pharmacother.* 2015; **49**(2): 196–206.
[PubMed Abstract](#) | [Publisher Full Text](#)
28. Cuninham J, Lee K, Ren T, *et al.*: **Small molecules inhibitors of Ebola and Lassa fever viruses.** 2014.
[Reference Source](#)
29. Panchal RG, Reid SP, Tran JP, *et al.*: **Identification of an antioxidant small-molecule with broad-spectrum antiviral activity.** *Antiviral Res.* 2012; **93**(1): 23–9.
[PubMed Abstract](#) | [Publisher Full Text](#)
30. Dube D, Schornberg KL, Shoemaker CJ, *et al.*: **Cell adhesion-dependent membrane trafficking of a binding partner for the ebolavirus glycoprotein is a determinant of viral entry.** *Proc Natl Acad Sci USA.* 2010; **107**(38): 16637–42.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
31. Yonezawa A, Cavois M, Greene WC: **Studies of Ebola virus glycoprotein-mediated entry and fusion by using pseudotyped human immunodeficiency virus type 1 virions: involvement of cytoskeletal proteins and enhancement by tumor necrosis factor alpha.** *J Virol.* 2005; **79**(2): 918–26.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
32. Fakhfakh MA, Fournet A, Prina E, *et al.*: **Synthesis and biological evaluation of substituted quinolines: potential treatment of protozoal and retroviral co-infections.** *Bioorg Med Chem.* 2003; **11**(23): 5013–23.
[PubMed Abstract](#) | [Publisher Full Text](#)
33. Fournet A, Mahieux R, Fakhfakh MA, *et al.*: **Substituted quinolines induce inhibition of proliferation of HTLV-1 infected cells.** *Bioorg Med Chem Lett.* 2003; **13**(5): 891–4.
[PubMed Abstract](#) | [Publisher Full Text](#)
34. Gantier JC, Fournet A, Munos MH, *et al.*: **The effect of some 2-substituted quinolines isolated from *Galipea longiflora* on *Plasmodium vinckei petteri* infected mice.** *Planta Med.* 1996; **62**(3): 285–6.
[PubMed Abstract](#) | [Publisher Full Text](#)
35. Campos Vieira N, Vacus J, Fournet A, *et al.*: **Antileishmanial activity of a formulation of 2-n-propylquinoline by oral route in mice model.** *Parasite.* 2011; **18**(4): 333–6.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
36. Nakayama H, Loiseau PM, Bories C, *et al.*: **Efficacy of orally administered 2-substituted quinolines in experimental murine cutaneous and visceral leishmaniases.** *Antimicrob Agents Chemother.* 2005; **49**(12): 4950–6.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
37. Schmidt F, Champy P, Séon-Ménier B, *et al.*: **Chemicals possessing a neurotrophin-like activity on dopaminergic neurons in primary culture.** *PLoS One.* 2009; **4**(7): e6215.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
38. Perovic VR: **Virtual screen for candidate inhibitors of EBOLA virus infection.** *F1000Res.* 2015.
[Reference Source](#)
39. Veljkovic V, Loiseau PM, Figadère B, *et al.*: **Dataset 1 in: Virtual screen for repurposing approved and experimental drugs for candidate inhibitors of EBOLA virus infection.** *F1000Research.* 2015.
[Data Source](#)
40. Veljkovic V, Loiseau PM, Figadère B, *et al.*: **Dataset 2 in: Virtual screen for repurposing approved and experimental drugs for candidate inhibitors of EBOLA virus infection.** *F1000Research.* 2015.
[Data Source](#)
41. Veljkovic V, Loiseau PM, Figadère B, *et al.*: **Dataset 3 in: Virtual screen for repurposing approved and experimental drugs for candidate inhibitors of EBOLA virus infection.** *F1000Research.* 2015.
[Data Source](#)
42. Veljkovic V, Loiseau PM, Figadère B, *et al.*: **Dataset 4 in: Virtual screen for repurposing approved and experimental drugs for candidate inhibitors of EBOLA virus infection.** *F1000Research.* 2015.
[Data Source](#)
43. Todeschini R, Consoni V: **Molecular descriptors for chemoinformatics.** *Weinheim: Wiley VCH Verlag.* 2009.
[Publisher Full Text](#)

Open Peer Review

Current Peer Review Status:

Version 2

Reviewer Report 19 February 2015

<https://doi.org/10.5256/f1000research.6601.r7701>

© 2015 Butaye P. This is an open access peer review report distributed under the terms of the [Creative Commons Attribution License](#), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Patrick Butaye

School of Veterinary Medicine, Ross University, Basseterre, Saint Kitts and Nevis

The revision is approved

Competing Interests: No competing interests were disclosed.

I confirm that I have read this submission and believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard.

Version 1

Reviewer Report 11 February 2015

<https://doi.org/10.5256/f1000research.6544.r7633>

© 2015 Botta B. This is an open access peer review report distributed under the terms of the [Creative Commons Attribution License](#), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Bruno Botta

Department of Chemistry and Pharmaceutical Technology, Sapienza University of Rome, Rome, Italy

To identify drug candidates against Ebola virus infections is surely an urgent need, especially in light of recent virus outbreaks registered mostly in Africa. In this respect, Veljkovic's article is presented in a timely manner and offers a fast and reliable opportunity to screen among large databases to reposition old drugs against Ebola.

The experimental design relies on a consolidated methodology, developed by some of the authors and successfully applied in multiple projects. Overall, the manuscript is clear and few minor editing would be necessary, in my personal opinion, to improve its consistency.

In Materials and Methods, a dataset of 152 drugs that counteract Ebola virus *in vitro* and *in vivo* has been selected as training set. However, it seems that an inconsistency does exist within this number. As authors have reported, these anti-Ebola drugs have been identified by Madrid (24 molecules) and Kouznetsova (53 molecules and 95 weaker drugs). Accordingly, the total number of FDA-approved drugs identified in these studies is 172. Why authors used a smaller set of 152? Is there any structural redundancy? A clarification of this discrepancy would improve the reproducibility of the work.

Finally, if I understood properly authors have selected more than 500 molecules (including FDA-approved and experimental drugs) as anti-Ebola candidates by means of *in silico* screening and suggest that further *in vitro/in vivo* tests should be performed on these molecules. In my opinion, this number is still too large for enabling efficient and fast *in vitro* and/or *in vivo* assays. Experimental testing of this set would require significant efforts. Just for comparison, the number of candidates selected in silico by authors is about half of those selected by Madrid by means of HTS (ref 2). Is there a way to prioritize small molecules by using the EIIP/AQVN-based approach, and to provide a lower number of compounds to be submitted to experimental evaluation? Authors should comment on this point, because the advantage of using the EIIP/AQVN-based screening in silico appears to be limited in the current version of the manuscript.

Competing Interests: No competing interests were disclosed.

I confirm that I have read this submission and believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard.

Author Response 13 Feb 2015

Veljko Veljkovic, University of Belgrade, Institute of Nuclear Sciences VINCA, P.O. Box 522, 11001 Belgrade, Serbia

The aim of the work was not only to reduce the number of candidate drugs for EVD but to select all approved drugs which will efficiently target GP or its receptor. Further filtering of these candidate anti-Ebola drugs by other structural tools (pharmacophoric modeling, docking studies, etc.) will reduce the number of compounds for experimental testing.

Competing Interests: No competing interests were disclosed.

Reviewer Report 11 February 2015

<https://doi.org/10.5256/f1000research.6544.r7552>

© 2015 Butaye P. This is an open access peer review report distributed under the terms of the [Creative Commons Attribution License](#), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Patrick Butaye

School of Veterinary Medicine, Ross University, Basseterre, Saint Kitts and Nevis

This manuscript deals with the *in silico* analysis of molecules for their activity against Ebola Virus (EBV). They started from a reference library of compounds who have previously demonstrated *in vitro* and/or *in vivo* activity against EBV and analyzed these compounds by the determination of their EIIP and AQVN. With these data, they scanned a larger collection of compounds with unknown activity against EBV and selected possible candidates to test for their activity *in vitro* and/or *in vivo*. This is a straight forward manuscript however it may be better structured. The part of the M&M dealing with the EIIP and AQVN is more appropriate to go into the introduction since this is background information of the calculation. For clarity of the manuscript is also better to separate the results section as it is difficult to follow now. Start with the analysis of the compounds with known activity, the two datasets, and then proceed with results from the unknown dataset. Then in the discussion, the different products of interest may be evaluated. This will largely increase the readability. Upon the antibiotics, it would be good to elaborate a bit on how they work on EBV, since common sense tells that antibiotics do not work on viruses. A better explanation on how these products may interact and inhibit/kill EBV would also be good.

Competing Interests: No competing interests were disclosed.

I confirm that I have read this submission and believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard.

Author Response 13 Feb 2015

Veljko Veljkovic, University of Belgrade, Institute of Nuclear Sciences VINCA, P.O. Box 522, 11001 Belgrade, Serbia

The antibiotics presented in this article are not selected because of their antibiotic activity but they are proposed as candidate entry inhibitors of Ebola virus (drug repurposing).

Competing Interests: No competing interests were disclosed.

The benefits of publishing with F1000Research:

- Your article is published within days, with no editorial bias
- You can publish traditional articles, null/negative results, case reports, data notes and more
- The peer review process is transparent and collaborative
- Your article is indexed in PubMed after passing peer review
- Dedicated customer support at every stage

For pre-submission enquiries, contact research@f1000.com

F1000Research