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Mahendra Kumar Trivedi, Alice Branton, Dahryn Trivedi, Gopal Nayak, Sambhu Charan Mondal, et al.. In vitro Evaluation of Biofield Treatment on Viral Load Against Human Immunodeficiency-1 and Cytomegalo Viruses. American Journal of Health Research, 2015, 3 (6), pp.338-343. hal-01467021

HAL Id: hal-01467021

<https://hal.science/hal-01467021>

Submitted on 14 Feb 2017

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***In vitro* Evaluation of Biofield Treatment on Viral Load Against Human Immunodeficiency-1 and Cytomegalo Viruses**

Mahendra Kumar Trivedi¹, Alice Branton¹, Dahryn Trivedi¹, Gopal Nayak¹, Sambhu Charan Mondal², Snehasis Jana^{2,*}

¹Trivedi Global Inc., Henderson, USA

²Trivedi Science Research Laboratory Pvt. Ltd., Bhopal, Madhya Pradesh, India

Email address:

publication@trivedisrl.com (S. Jana)

To cite this article:

Mahendra Kumar Trivedi, Alice Branton, Dahryn Trivedi, Gopal Nayak, Sambhu Charan Mondal, Snehasis Jana. *In vitro* Evaluation of Biofield Treatment on Viral Load Against Human Immunodeficiency-1 and Cytomegalo Viruses. *American Journal of Health Research*. Vol. 3, No. 6, 2015, pp. 338-343. doi: 10.11648/j.ajhr.20150306.14

Abstract: Viral load quantification is the amount of particular viral DNA or RNA in a blood samples. It is one of the surrogate biomarker of AIDS. High viral load indicates that the immune system is failed to fight against viruses. The aim of this study was to evaluate the impact of biofield treatment on HIV-1 and HCMV in terms of viral loads as surrogate marker. The viral load assay was performed on stored stock cultures of HIV infected human plasma samples before and after 7 days of biofield treatment using Roche COBAS[®] AMPLICOR analyzer. Viral load (HIV-1 RNA and HCMV DNAemia) was considered as surrogate marker for assessment of the impact of Mr. Trivedi's biofield treatment in HIV infected stored plasma samples. The viral load quantification of HIV-1 RNA in infected stored plasma samples was significantly reduced by 65% in biofield treated group as compared to control. Additionally, viral load of HCMV DNAemia in infected stored plasma samples was also reduced by 80% in the biofield treated group as compared to control. Because, children are more prone to HCMV infection and adults are generally liable to suffer from HIV-1 infection. As the biofield treatment has reduced HCMV DNAemia, it could be beneficial for HIV infected children populations. Altogether, data suggest that biofield treatment has significantly reduced the viral load quantification in HIV-1 and HCMV infected stored plasma samples and could be a suitable alternative treatment strategy for AIDS patients in near future.

Keywords: Human Immune Deficiency Virus, Biofield Treatment, Cytomegalo Virus, Viral Load, HIV RNA, HCMV DNAemia, AIDS, Surrogate Biomarker

1. Introduction

Human immunodeficiency virus type 1 (HIV-1) is the main causative agent of acquired immune deficiency syndrome (AIDS) [1]. HIV is a worldwide pandemic disease, and the number of infected peoples around the world increasing day by day. Recent estimate from World Health Organization (WHO) shows that 16.3 million people have died from AIDS since the beginning of the epidemic. Currently, around 34.3 million people alive with HIV infection, in which approximately 7% are young adults [2], infected with HIV type 1 (HIV-1) across the mucosal surfaces or by direct inoculation. The virus first attacks to dendritic cells (DCs) and subsequently spreads to cluster of

differentiation - 4 (CD⁴⁺) T lymphocytes [3]. Human cytomegalovirus (HCMV) is a vernacular name of the human herpes virus - 5, a highly host-specific virus of the herpesviridae family rarely causes symptoms. The target DNA sequence is specific, located within the HCMV DNA polymerase gene, and is not homologous to other members of the human herpes virus family. The pregnant women and immune weakened persons are highly prone to acquire infection by HCMV virus. It spreads through various body fluids, such as blood, urine, saliva, semen, and breast milk. It may also causes serious morbidity and mortality in organ transplant recipients, immunocompromised, HIV infected patients and congenitally infected newborns [4]. HCMV is mainly prevalent in kidney transplant patients. The viral 'threshold load' is very sensitive and specific for predicting

both white blood cell and platelet quantification in kidney transplant patients. The threshold value is more than 10000 copies/mL considered as HCMV infection [5]. In infected adults, HIV viral load is predictive of progression to AIDS [6, 7] and HCMV DNAemia predicts progression to disease, particularly retinitis [8]. Several study reports have confirmed the relation between HIV-1 viral load and HCMV DNAemia levels in terms of prediction of disease (AIDS) progression in both adults [6, 7] and children [9,10]. Researchers found an excellent correlation between two viruses *i.e.* HIV infected adults are highly predictable for the development of HCMV end organ disease [11]. Hence, in this experiment the dual viruses were taken into consideration as a rational. Immunomodulatory therapies are currently used against HIV-1 infections which include mycophenolate mofetil, cyclosporine, interleukin-2 (IL-2), and various vaccines [12]. Although several treatment strategies are available against AIDS patients infected by HIV-1 such as antiretroviral agents and vaccination. But some difficulties are also present. Based on above lacunas, there is a need of an alternative way which may be useful to determine the viral loads by either enhancing the application of existing agents or by means of some alternative strategy or developing new drugs. Biofield treatment is an alternative approach which may be useful to improve these unfilled space associated with AIDS infected patients. Mr. Trivedi's unique biofield treatment (The Trivedi effect®) has been extensively contributes in scientific communities in several fields [13-16]. Therefore, authors interested, to evaluate the impact of biofield treatment on viral load in HIV and HCMV infected plasma samples.

2. Materials and Methods

The viral samples (HIV-1 and HCMV) as stored stock cultures were procured from department of microbiology laboratory, P.D. Hinduja National Hospital and Medical Research Centre, Mumbai. The viral load assay was performed on HIV patients stored plasma samples before and after treatment using Roche COBAS AMPLICOR analyzer according to manufacturer's instructions.

2.1. Biofield Treatment Strategy and Experimental Design

Two sets of each viral samples (HIV-1; 31 samples and HCMV; 5 samples) of HIV and HCMV infected stored

plasma were used in this experiment for determination of viral load quantification. The first sets of both viral samples were considered as control. No treatment was given to these sets. The second sets of both viral samples were subjected to Mr. Mahendra Trivedi's biofield treatment, considered as treated group. Both control and treated samples were analyzed after 7 days for viral load quantification in as per the standard protocols. An optimum precautionary measure was taken to maintain the cold chain throughout the experiment. The differences of viral load quantification before and after the treatment were noted.

2.2. COBAS® Amplicor HIV-1 Monitor Test

The COBAS® amplicor HIV-1 monitor test (v1.5) is an *in vitro* nucleic acid amplification test approved by food and drug administration (FDA) for the quantification of HIV-1 RNA in human plasma on the COBAS AMPLICOR™ analyzer. This technique is a gold standard automated solution for testing of HIV-1 viral loads in major pharmaceutical trials [17].

2.3. COBAS® Amplicor CMV Monitor Test

The COBAS® amplicor CMV monitor test is FDA approved *in vitro* amplification test for the quantification of HCMV DNA in human plasma on the COBAS AMPLICOR™ analyzer. This is an automated, sensitive, reliable, and specific method for quantification of cytomegalo viral loads in HCMV infected patients with high productivity (600 copies/mL to 100,000 copies/mL) [18].

3. Results and Discussion

Viral load (Fig. 1) means the amount of HIV RNA or HCMV DNA particles per milliliter of blood sample. Higher the viral titer indicates that the immune system is failed to fight against HIV or HCMV.

The viral loads of HIV-1 and HCMV expressed as copies/ml are shown in Table 1 and 2, respectively. HIV RNA and HCMV DNA (viral load) and CD4⁺ T lymphocyte cell count are the two surrogate markers of HIV patients [19].

All the values are expressed as (IU/mL); Serial number 1-31 denoted as viral stock human plasma samples.

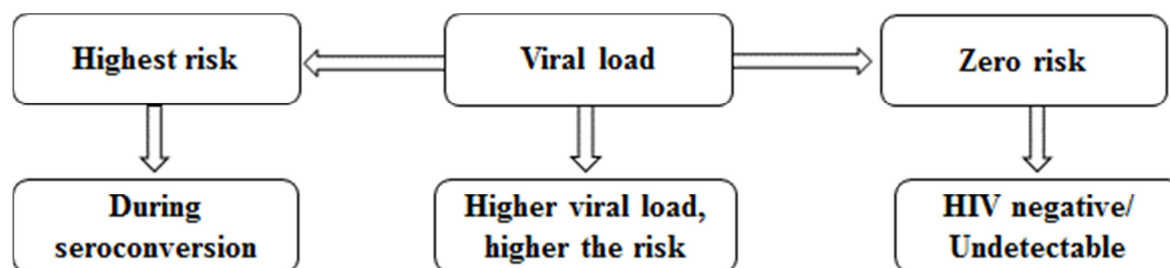


Figure 1. Schematic diagram related to viral load and its risk manifestation.

Table 1. Viral load of human immunodeficiency virus type-1 (HIV-1) in infected stored plasma samples.

S. No.	Viral load Control	Log10 (Control)	Viral load Treatment	Log10 (Treatment)	Change in Viral Load Log10 (Treatment) - Log10 (Control)
1.	91900	4.96	18500	4.27	-0.70
2.	179000	5.25	69300	4.84	-0.41
3.	159000	5.20	121000	5.08	-0.12
4.	10300	4.01	4110	3.61	-0.40
5.	107000	5.03	103000	5.01	-0.02
6.	1650000	6.22	3320000	6.52	0.30
7.	172000	5.24	122000	5.09	-0.15
8.	7820	3.89	2320	3.37	-0.53
9.	1540	3.19	3060	3.49	0.30
10.	34400	4.52	1320	3.12	-1.40
11.	69300	4.84	174000	5.24	0.40
12.	2190	3.34	1760	3.25	-0.09
13.	1860	3.27	881	2.94	-0.32
14.	17400	4.24	8690	3.94	-0.30
15.	216000	5.33	297000	5.47	0.14
16.	321000	5.51	440000	5.64	0.14
17.	17400	4.24	17400	4.24	0.00
18.	9840	3.99	8540	3.93	-0.06
19.	10300	4.01	3140	3.50	-0.52
20.	99200	5.00	65700	4.82	-0.18
21.	1650000	6.22	2900000	6.46	0.24
22.	290000	5.46	291000	5.46	0.00
23.	156000	5.19	401000	5.60	0.41
24.	3070000	6.49	1450000	6.16	-0.33
25.	587000	5.77	807000	5.91	0.14
26.	36500	4.56	22100	4.34	-0.22
27.	138000	5.14	68400	4.84	-0.30
28.	6560	3.82	4230	3.63	-0.19
29.	1900000	6.28	2580000	6.41	0.13
30.	32800	4.52	20800	4.32	-0.20
31.	42700	4.63	18600	4.27	-0.36

All the values are expressed as (IU/mL); Serial number 1-31 denoted as viral stock human plasma samples.

Table 2. Viral loads of human cytomegalo virus (HCMV) in infected stored plasma samples.

S. No.	Viral load Control	Log10 (Control)	Viral load Treatment	Log10 (Treatment)	Change in Viral Load Log10 (Treatment) - Log10 (Control)
1.	86900	4.94	78200	4.89	-0.05
2.	18100	4.26	8760	3.94	-0.32
3.	40500	4.61	35000	4.54	-0.06
4.	1570	3.20	1360	3.13	-0.06
5.	697	2.84	<600	2.78	-0.07

All the values are expressed as (IU/mL); Serial number 1-5 denoted as viral stock human plasma samples.

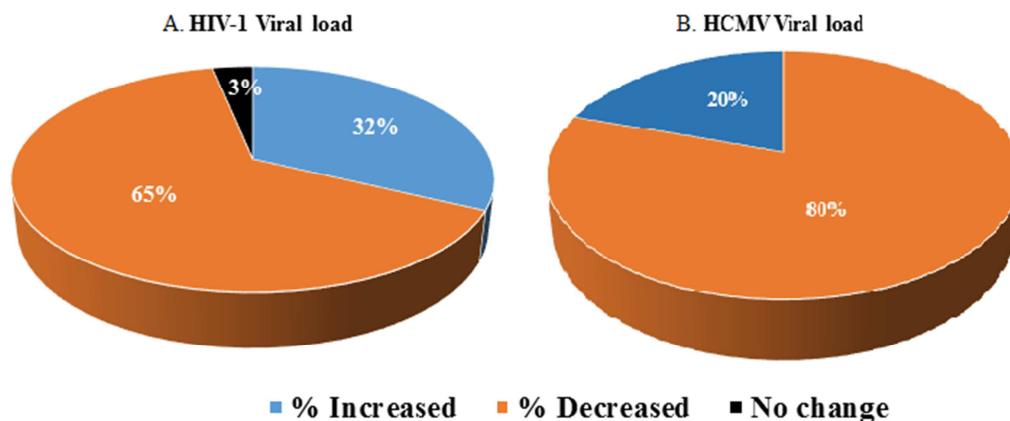


Figure 2. Percentage change of viral load of A. (HIV RNA) in human immunodeficiency virus type-1 (HIV-1) and B. (HCMV DNA) in human cytomegalo virus (HCMV) after biofield treatment.

All the values are expressed as (IU/mL); Serial number 1-5 denoted as viral stock human plasma samples

In this experiment, viral load was considered as surrogate biomarker for assessment of the impact of Mr. Trivedi's biofield treatment. Study was carried out in total thirty one infected human stored plasma samples. The result showed

that viral load of HIV RNA in infected plasma samples were reduced by 65% out of thirty one samples after biofield treatment as compared to control. In addition, viral load were increased by 32% in biofield treated group and 3% unchanged as compared to control (Figure 2A and 3).

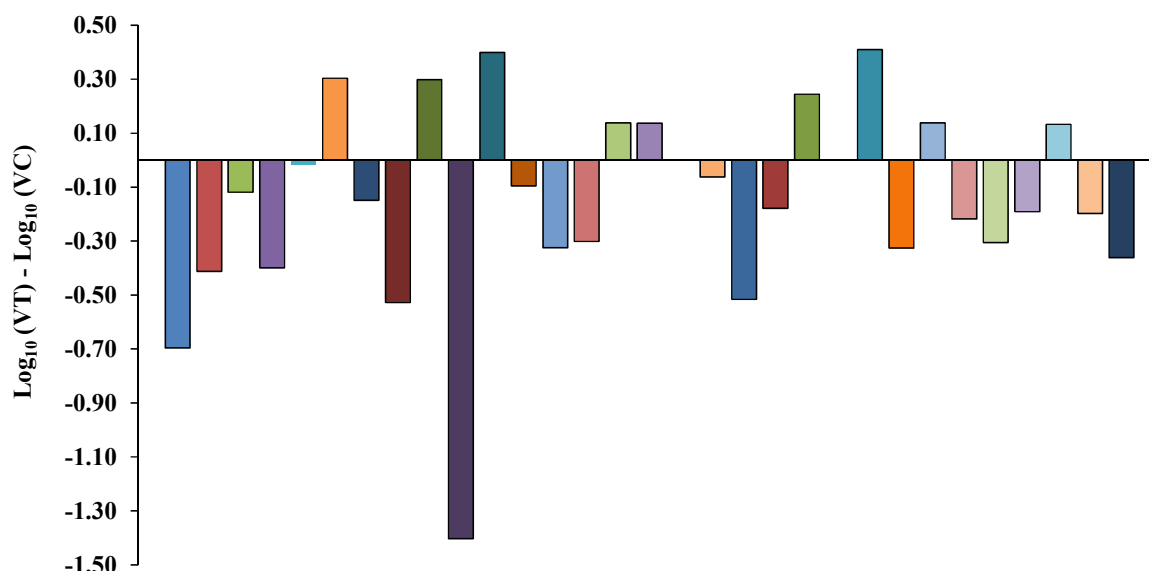


Figure 3. Difference in viral load of HIV RNA of 31 viral stock human plasma samples after biofield treatment. VT- Viral load in treatment (IU/mL); VC- Viral load in control (IU/mL).

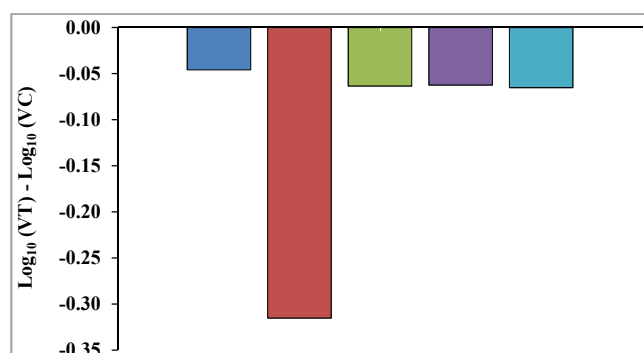


Figure 4. Difference in viral load of HCMV DNA of 5 viral stock human plasma samples after biofield treatment. VT- Viral load in treatment (IU/mL); VC- Viral load in control (IU/mL).

According to Greene *et al.* National Center for Complementary and Alternative Medicine (NCCAM), about 2000 reported that bioelectromagnetic based therapy *i.e.* biofield is used as an effective [20], and alternative therapy in HIV/AIDS infected patients [21]. This study findings also explored in pros of biofield treatment against HIV infections. This study further emphasized to HCMV that are responsible for progression of AIDS. Viral load quantification of HCMV DNA were determine in total five infected stored plasma samples. The results showed that viral load of HCMV DNA in infected stored plasma samples were significantly reduced by 80% after biofield treatment as compared to control. The study outcomes

showed a significant improvement of biofield treatment. Besides, the viral load of HCMV DNA were increased by 0% and unable to detect about 20% in biofield treated group as compared to control (Figure 2B and 4).

Because, 20% of HCMV DNA are inactive not in multiplying state but infectious. Based on existing literatures, HCMV load in the plasma samples of HIV infected or AIDS patients is an important factor in the pathogenesis of retinitis [22]. Another study data indicates that the risk of HCMV disease and death occurs in AIDS patients is directly proportional to the quantity of HCMV DNA (viral load) in plasma, and the viral load quantification is a better predictor than CD⁴⁺ T lymphocyte count [23]. The viral particles *i.e.* HIV RNA and HCMV DNA those are in multiplying state, possibly affected by Mr. Trivedi's biofield treatment. The specific frequencies of electromagnetic radiation which matches with the resonance frequencies of RNA or DNA, probably disintegrate the respective viral RNA/DNA and disrupted thus reduced the viable viral titer and *vice versa* [24,25]. According to a recent report regarding biofield treatment which was later approved by the German equivalent of the FDA emphasized that cancer patients have experienced healing with biofield treatment. Nowadays, many scientists and cutting edge practitioners believed that the secrets of regeneration and healing lie not only on costly medical drugs or expensive medical treatments, but also in the body's own Quantum Energy Biofield (QEB) [26]. Mr. Trivedi's biofield treatment has shown to improve overall productivity in the field of agriculture, biotechnology,

material science and others field also, without utilization of any drugs. So, it is assumed that biofield treatment would be cost effective as compared to the existing treatment strategy.

Overall, the study data indicates that biofield treatment significantly reduced the viral load of HIV-1 and simultaneously eliminate the viral load of HCMV from infected human plasma samples. Based on the obtained results, it is presumed that biofield treatment could be novel, cost effective and an alternative advanced strategy towards AIDS patients.

4. Conclusion

To summarize, the study results showed significant (65%) reduction of HIV RNA viral load from infected plasma samples in the biofield treated group. Experimental data also showed 80% elimination of HCMV DNAemia from infected plasma samples after biofield treatment. It is assumed that Mr. Trivedi's biofield treatment could be beneficial to improve the viral loads in HIV-1 infected AIDS patients specially children with high level of HCMV DNAemia.

Acknowledgement

The Authors gratefully acknowledged to Trivedi Science, Trivedi Testimonials and Trivedi Master Wellness and P.D. hinduja national hospital and medical research centre, Mumbai, India for their support.

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