



Report on the efficacy of Biotek Disinfectant against human immunodeficiency virus type 1 measured using destruction of viral genome as a marker

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Quadralene Ltd.

Human immunodeficiency virus RNA degradation

Introduction

Human immunodeficiency virus type 1 (HIV) infection, which is the cause of AIDS, is a pandemic affecting almost all countries. Homosexual and bisexual men represent the largest number of AIDS cases in Western Europe and North America but, in 1993, over half of all new cases occurred in injecting drug users. In addition to these modes of transmission, rare occurrences of casual contact with infected individuals by, for example, nursing staff, and exposure of non-intact skin to blood after a road traffic accident and a sports injury have been reported to result in HIV infection. HIV has been isolated from a variety of body fluids, however only contacts with blood, semen, other genital secretions and breast milk have been implicated as sources of infection.

Difficulties exist in measuring the effectiveness of disinfectants against the virus. Virus can be grown in the laboratory in permissive cell lines but a high level of containment is required making the process expensive and rarely available. Viral nucleic acid is usually readily detectable in the serum of chronically infected patients, often at high titres. This protocol uses an assay for measuring the concentration of viral nucleic acid (which in the case of HIV is RNA) in serum following exposure of the sample to the disinfectant, and to distilled water as control. The loss of detectable viral RNA is used as a marker of virus 'killing'. The assay is likely to underestimate the effectiveness of disinfectants against HIV because the RNA molecule detected is relatively resistant to chemical degradation; it is, however, essential for infectivity and so its disappearance following treatment is a reliable indication of virus inactivation.

Protocol

The source of the HIV RNA for this test was a patient with well-documented HIV infection. The serum had a high titre ($>10^6$ HIV-1 RNA copies/ml) of HIV RNA, and was HIV antibody positive. 2.5 μ l aliquots of the serum sample were treated in a suspension test without the addition of a high protein load by adding,

- a. 997.5 μ l of 5% Biotek Disinfectant, or
- b. 997.5 μ l of distilled water

These treatments were performed at room temperature ($\sim 21^\circ\text{C}$) for a contact time of 5 minutes. Following this an extraction procedure (High Pure Viral Nucleic Acid extraction kit; Roche; according to the manufacturers instructions) was used to isolate viral RNA from the solution. Residual disinfectant was removed at this stage.

The concentration of remaining detectable viral RNA was measured using a very sensitive quantitative assay, the results of which are expressed as copies of RNA detected per millilitre.

Results

5% Biotek Disinfectant treatment	Water treatment
NOT detected (<100 HIV RNA copies/ml)	3,615 HIV RNA copies/ml

Comment

To be considered successful in this protocol a disinfectant must be able to reduce the concentration of HIV RNA to an undetectable level, which, with the sensitivity of the assay used, is less than 100 copies/ml. After 5 minutes contact time to Biotek Disinfectant at a concentration of 5%, HIV RNA was not detectable in the serum sample and Biotek Disinfectant was therefore successful in this indirect estimation of its activity against HIV.

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