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Viraffinity™

Virus and Viral Component Isolation

- Purifies whole infectious non-enveloped virus & non-infectious enveloped virus
- Complements ultracentrifugation, for viral proteomic enrichment 350X from plasma (Fagnoud et al, 2016)
- Isolates antigenic virions, enveloped and non-enveloped
- Enriches for viral nucleic acids
- Prepares viral samples for subsequent detection and analysis

Viraffinity™ is a unique water-insoluble elastomeric polyelectrolyte that has been engineered for the capture and recovery of viruses. Applications include: purification of whole infectious non-enveloped virus, virions, viral components, and sample preparation for subsequent detection and analysis. **Viraffinity™** is directly added to the sample that is then mixed and centrifuged. The centrifuged pellet contains polyelectrolyte-bound viruses that can then be recovered using a moderately alkaline pH solution.

Viraffinity™ is supplied as a suspension reagent ready for use. Viraffinity™ is also supplied as the separations component of the **ViraPrep™** application kits, for Lambda DNA & Mammalian Viruses.

Product	Size	Item No.
Viraffinity™ 15	15ml	V1062-15

Mammalian Virus & Lambda DNA Isolation Kits

ViraPrep™ Mammal contains **Viraffinity™** and all necessary buffers and protocols for mammalian virus and virion isolation. **ViraPrep™ Lambda** is a complete application kit containing **Viraffinity™** and all the necessary reagents for obtaining high purity DNA from plate or liquid lysates, suitable for amplification, automated fluorescent sequencing and other common molecular biology techniques. Please visit our website for more information.



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Virus	Titer	Ratio	% Bound ^b
HIV-1 ^a	7x10 ³ TCID ₅₀	1:2	96
HIV-1, human serum	7x10 ³ TCID ₅₀	1:2	80
Chimeric Human Rhinovirus ^a	10 ⁶ - 10 ⁸ pfu/ml	1:3	95
Dengue virus	After Sucrose density Ultra-centrifugation	1:2	
Adenovirus (Ad5d1309) ^a	10 ⁶ - 10 ⁸ pfu/ml	1:3	90
Reovirus Type 3 ^a	10 ⁶ - 10 ⁸ pfu/ml	1:3	50-80
Encephalomyocarditis (EMC) ^a	10 ⁷ TCID ₅₀	1:4	99
Porcine Parvovirus ^a	10 ⁷ TCID ₅₀	1:2	90
Unclassified Entero-Picornavirus ^a	10 ⁶ TCID ₅₀	1:4	90
Coxsackievirus A24 ^a	10 ⁶ - 10 ⁷ pfu/ml	1:2	70-95
Bacteriophage Lambda	10 ⁹ pfu/ml	1:5	>95

Ratio refers to the volumetric ratio of Viraffinity™ to original sample volume.

^a Tissue culture supernatants containing 1-10% Fetal Bovine Serum.

^b Based on infectivity.

Storage

Supplied as an aqueous suspension of a synthetic, anionic polyelectrolyte in buffer. The reagent should be kept sealed and stored at 4°C. Do not freeze. Viraffinity™ retains full activity when stored accordingly for approximately 1 year.

PROTOCOL

Purification of Infectious Virus, Virions and Viral Components

To date, viability of enveloped viruses after desorption of the virus has been anecdotal only. However, viability has been demonstrated when the viruses are non-enveloped although optimization for any given virus has been limited. In all cases, the recovery of virions, viral proteins or viral nucleic acids are good applications for Viraffinity™.

This protocol can be used for cell culture supernatants, clarified cell culture lysates, serum, plasma, cervical fluid, biological extracts or other types of samples.



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1. Add 1 volume of 60 mM MES, 150 mM NaCl, pH 6.5 to sample. Alternatively, for clarified cell culture samples, condition with 1:30 volume of 1 M MES, pH 6.5.
2. Incubate with 1:4 volume ratio, Viraffinity™:sample, based on initial sample volume. If necessary, the ratio can be adjusted according to the titer of sample, a minimum ratio of 1:5 Viraffinity™:sample, however, is recommended for quantitative recovery.
3. Mix well and let stand for 5 minutes at room temperature.
4. Pellet by centrifugation, 1,000 X G for 10 minutes. NOTE: Do not use maximum G-force to pellet as it makes subsequent steps difficult to re-suspend.
5. Decant and discard supernatant and wash the pellet with the equivalent starting volume of the sample. Use 60 mM MES, 150 mM NaCl, pH 6.5 for all washes. Repeat washing and pelleting steps 2 more times. Consider the 3 recovery options described in (6) before proceeding.
6. A. To recover and maintain viable non-enveloped virus, resuspend the pellet in 1 to 3 volumes (based on initial sample size) of 100 mM Tris-Base, 100 mM Borate pH 9.0. Optionally, a detergent such as 1% N-lauroyl sarcosine may be added to aid in recovery and purity.

6B. To recover virions or viral proteins, resuspend the pellet in 1 to 3 volumes (based on initial sample size) of elution buffer to pH 10.5. Optionally, detergents such as Triton or SDS can be used in conjunction with the buffer to isolate viral proteins. As an option to recover viral proteins for proteomic analysis, 5 min at 70 °C in SDS-buffer can be used.

6C. To recover viral nucleic acids, resuspend the pellet in 1 to 3 volumes (based on initial sample size) of a lysis buffer of the users choice. These may contain chaotropes or detergents such as Triton or SDS which are compatible with Viraffinity™. Temperatures to 65°C may also be used to assist with lysis.
7. Pellet by centrifugation using maximum G-force for 10 minutes. Recover supernatant.



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References

Dengue Virus

Romain Fragnoud, Marie Flamand, Frederic Reynier, Philippe Buchy, Vasna Duong, Alexandre Pachot, Glaucia Paranhos-Baccala and Frederic Bedin. **Differential proteomic analysis of virus enriched fractions obtained from plasma pools of patients with dengue fever or severe dengue.** BMC Infectious Diseases (2015) 15:518. <http://www.ncbi.nlm.nih.gov/pubmed/26572220>

Norovirus

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Hanta Virus

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Mihalic, K. A., et al, *Development of a Chemiluminescent Western Blot for Detecting Hantaan-Specific Antibodies*, poster American Society of Tropical Medicine and Hygiene Meeting, October 1997.

Human Mouth Virus

Metagenomic Analysis of the Human Mouth Virus Population and Characterisation of Two Lytic Viruses, Al-Jarbou, Ahmed, Theses, Dept. of Infection. Immunity and Inflammation. 2009

Lepidoptera: Gelechiidae

An isometric virus of the potato tuber moth Tecia solanivora (Povolny) (Lepidoptera: Gelechiidae) has a tri-segmented RNA genome Jean-Louis Zeddou et. al Journal of Invertebrate Pathology, Volume 99, Issue 2, October 2008, Pages 204-211

Human Immunodeficiency Virus Type 1

Activity of the Small Modified Amino Acid -Hydroxy Glycineamide on In Vitro and In Vivo Human Immunodeficiency Virus Type 1 Capsid Assembly and Infectivity Antimicrobial Agents and Chemotherapy, October 2008, p. 3737-3744, Vol. 52, No. 10 Samir Abdurahman

Mutation in the loop C-terminal to the cyclophilin A binding site of HIV-1 capsid protein disrupts proper virus assembly and infectivity Samir Abdurahman, Stefan Höglund, Anders Höglund and Anders Vahne Retrovirology 2007, 4:19doi:10.1186/1742-4690-4-19



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Poliovirus type 1 (PV1), Hepatitis A virus (HAV), Norwalk virus

[Detection Methods for Human Enteric Viruses in Representative Foods](#). Leggitt, Paris R.1; Jaykus, Lee-Ann, *Journal of Food Protection®*, Volume 63, Number 12, December 2000 , pp. 1738-1744(7).

Polio Virus Type 1 and Calcivirus

[Rapid Detection of Foodborne Viruses from Minimally Processed Foods](#). *Microbial Detection Methods* 2000

Polio Virus

Ting, W.T. E., E. M. Nielson, and C.C. Tseng. 1997. [The use of Viraffinity matrix to concentrate waterborne polioviruses for RT-PCR detection](#). Abstr. Q-169. p.484. In Abstracts of the 97th General Meeting of the American Society for Microbiology 1997, American Society for Microbiology, Washington, D.C.

Bacteriophage Lambda DNA

Hitti, J., et al, [Fast and Convenient Purification of Bacteriophage Lambda DNA with Viraffinity Matrix](#), poster Cold Spring Harbor Conference on Genome Mapping & Sequencing, May 1997.

Related ViraPrep™ References

Human Mouth Virus

Al-Jarbou, Ahmed N. "[Metagenomic Analysis of the Human Mouth Virus Population and Characterisation of Two Lytic Viruses](#)." PhD diss., University of Leicester, 2008.

CONTACT US

We welcome your questions, and comments regarding our products.

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