

ABL's HIV p24 ELISA

ACCURATE LENTIVIRUS QUANTIFICATION FOR INDUSTRIAL PROCESSES

Advanced BioScience Laboratories, Inc. (ABL) boasts a distinguished track record of more than 40 years in human immunodeficiency virus (HIV) research. Our best-in-class p24 ELISA kits are born from that legacy and are available for use in various retrovirus applications.

p24 ELISA Kits: p24 is a reliable marker for HIV, and p24 ELISA kits are commonly used to determine HIV titers. ABL is proud to have pioneered one of the first p24 ELISA kits on the market. Unlike other commercial p24 kits, all essential components in ABL's p24 ELISA kit are derived from native viruses instead of recombinant sources.

- **p24 protein standard:** Purified from a cell line which constitutively produces HIV-1.
- **Capture antibodies:** Two carefully selected monoclonal antibodies that recognize distinct epitopes, and are derived from mice injected with native p24 protein.
- **Detection antibody:** HRP-conjugated polyclonal antibody purified from HIV-positive human plasma.

Challenge: One of the key challenges with manufacturing Lentiviruses (LVV) is quantifying the virus produced at different stages of the production process. There is a particular lack of correlation between genomic copy (GC titer) and p24 results of in-process lentivirus samples, notably in early-stage and low-concentration samples.

Study Design: Quantitation of p24 from LVV in-process samples were evaluated using the ABL p24 ELISA kit and ELISA kits from other vendors. The genomic titer was measured by qRT-PCR. Samples were divided into the following sub-categories for analysis:

- **Total Samples**
- **Lower Titer Samples:**
GC Titer < 4.0E+09 vg/mL
- **Higher Titer Samples:**
GC Titer ≥ 4.0E+09 vg/mL
- **Early Step Samples:**
Harvest, IEX Load
- **Later Step Samples:**
IEX, TFF Ret. & Sterile filtrate

Figure 1: Key components of ABL's p24 sandwich ELISA kit. Catalog #5421 & #5447.

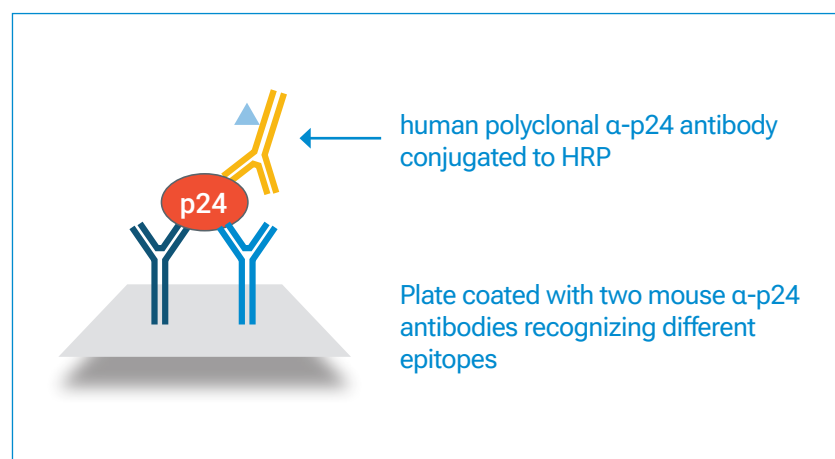


Table 1: Cost & Operation Comparisons

Mfr	Price	Operation time (hrs)	# of steps
ABL	\$	2-3.5	3
Kit-B	\$	2-3.5	4
Kit-C	\$\$\$	< 2	2
Kit-D	\$\$\$	> 3.5	4
Kit-E	\$\$	> 3.5	3

\$: 450-600 \$\$: 600-750 \$\$\$: 750-900



GOOD CORRELATION WITH GC, PARTICULARLY IN LOW-TITER AND EARLY-STEP SAMPLES

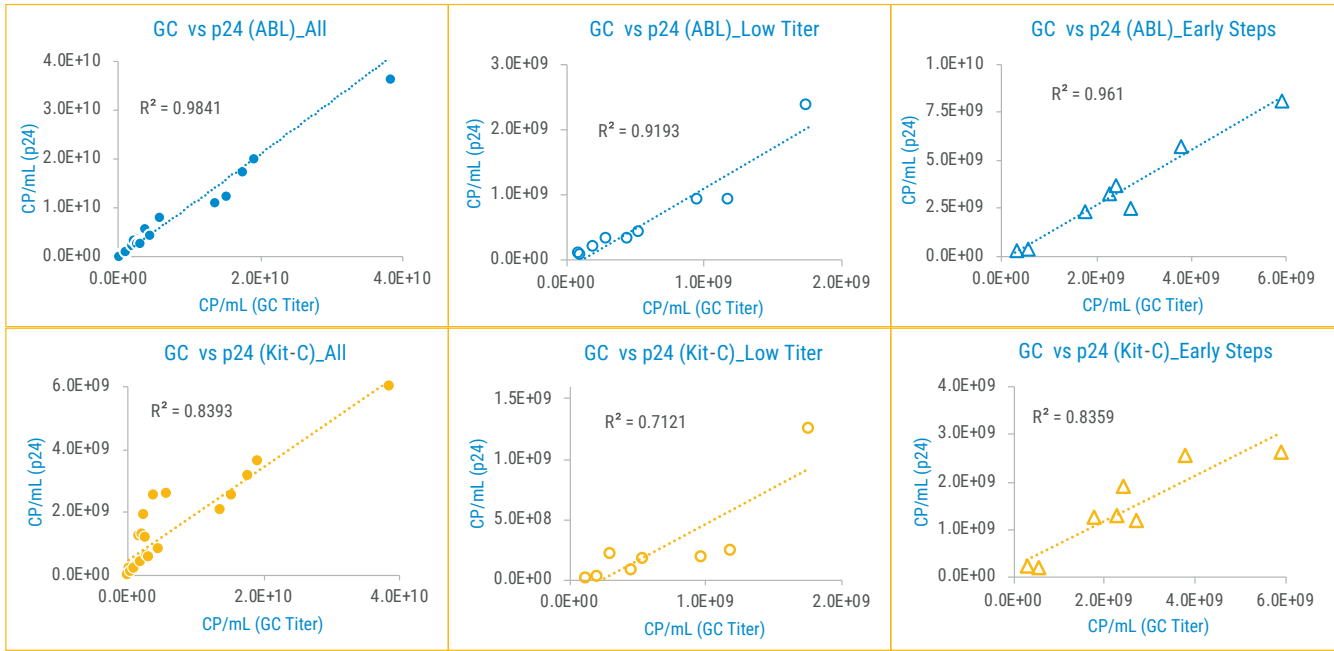


Figure 2: (above) Representative graphs comparing p24-derived with genomic copies (GC)-derived viral particle concentrations (VP/mL) using linear regression.

Table 2: (right) displays R² values for each kit and sample sub-categories, with darker gold indicating stronger correlation.

Mfr	All	Low Titer	High Titer	Early Step	Late Step
ABL	0.984	0.919	0.981	0.961	0.992
Kit-B	0.976	0.890	0.971	0.816	0.981
Kit-C	0.839	0.712	0.825	0.836	0.990
Kit-D	0.981	0.877	0.980	0.845	0.988
Kit-E	0.989	0.877	0.990	0.852	0.998

vg: virus genomic copy number, measured by qRT-PCR. VP: Virus Particles. VP conc. (from p24) = p24 conc. (ng/mL) x 1.0 E+07. VP conc. (from GC) = GC conc. (vg/mL) ÷ 2.

A FAVORABLE AVERAGE P24/GC RATIO WITH LESS VARIATION (CV %) AMONG LVV IN-PROCESS SAMPLES

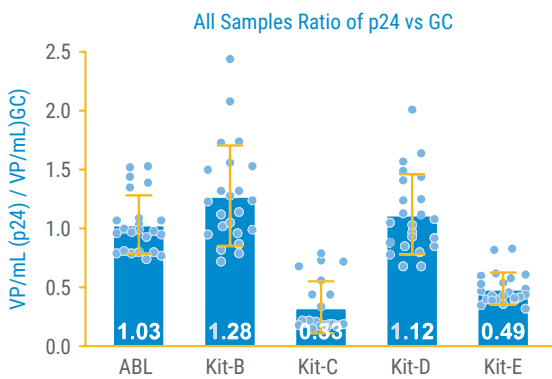


Figure 3: (above) Bar graph displaying average ratios and sample distributions for all kits in "All samples" subcategory.

Mfr	VP/ng p24	Total		Lower Titer		Higher Titer		Early Step		Later Step	
		Ave	CV%	Ave	CV%	Ave	CV%	Ave	CV%	Ave	CV%
ABL	1.00E+07	1.03	24%	0.94	21%	1.09	24%	1.25	23%	0.92	12%
Kit-B	1.25E+07	1.28	33%	0.99	26%	1.46	28%	1.63	30%	1.09	23%
Kit-C	1.00E+07	0.33	67%	0.34	70%	0.32	67%	1.88	28%	0.59	7%
Kit-D	1.00E+07	1.12	30%	0.89	26%	1.27	24%	1.37	27%	0.97	20%
Kit-E	1.00E+07	0.49	27%	0.43	25%	0.53	26%	0.61	25%	0.43	6%

Table 3: (above) lists average ratios and variation (CV %) for all kits across all subcategories with dark gold indicating strong correlation.

Conclusions for ABL's p24 ELISA kit: TOP CHOICE for quantifying Lentiviral Vector (LVV) in-process samples

- Cost-effective, moderate incubation time and reasonable operation steps.
- Good correlation with GC, particularly in low-titer and early step samples.
- A favorable average p24/GC ratio with less variation (CV%) among LVV in-process samples.

