



FMR-5S

Comparison Report

Vazyme FMR-5S: Vazyme FMR3



For Science For Health

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1. Reproducibility

(1) Experimental Design:

We conducted relative quantification experiments using three distinct qPCR reagents across different real-time PCR instruments: Vazyme FMR-5S, Vazyme FMR3 . The repeatability of each system was evaluated across instruments using the criterion of $CV \leq 3\%$ for 96 replicate wells. Specific reagents and procedures are detailed in the table below:

Reagents:	QN222-H23-ROX
procedures	37°C 2 min
	95°C 30 sec
	95°C 10 sec
	60°C 30 sec

] 40x

Note:

The entire 96-well plate is used to calculate the CV value, analyzing overall repeatability;
the red zone is for assessing central repeatability,
while the blue zone evaluates edge repeatability (edge effect).

	A	B	C	D	E	F	G	H	I	J	K	L
1												
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(2) Experimental Data

Vazyme FMR-5S			
	C_T Mean	C_T SD	$CV (\%) = \frac{SD}{Mean} \times 100\%$
Entire plate	28.23	0.10	0.36
Center	28.22	0.10	0.36
Edge	28.24	0.10	0.37

Vazyme FMR3			
	C_T Mean	C_T SD	$CV (\%) = \frac{SD}{Mean} \times 100\%$
Entire plate	29.25	0.26	0.88
Center	29.23	0.25	0.86
Edge	29.30	0.27	0.92

(3) Experimental Conclusion:

Performance was assessed by the CV value of entire-plate repeatability:
Vazyme FMR-5S (0.36%) > Vazyme FMR3 (0.88%)

Performance was evaluated based on edge effects using peripheral well CV values:

Vazyme FMR-5S (0.37%) > Vazyme FMR3 (0.92%)

Performance was judged according to CT values:

Vazyme FMR-5S (28.22) > Vazyme FMR3 (29.25)

To summary:

Vazyme FMR-5S > Vazyme FMR3

Vazyme FMR-5S demonstrated significantly better reproducibility with CV values nearly 2.5 times lower than Vazyme FMR3 (0.36% vs 0.88%), indicating superior well-to-well consistency. Additionally, FMR-5S exhibited earlier Ct values (28.22 vs 29.25), suggesting higher detection sensitivity.

2. Amplification Efficiency

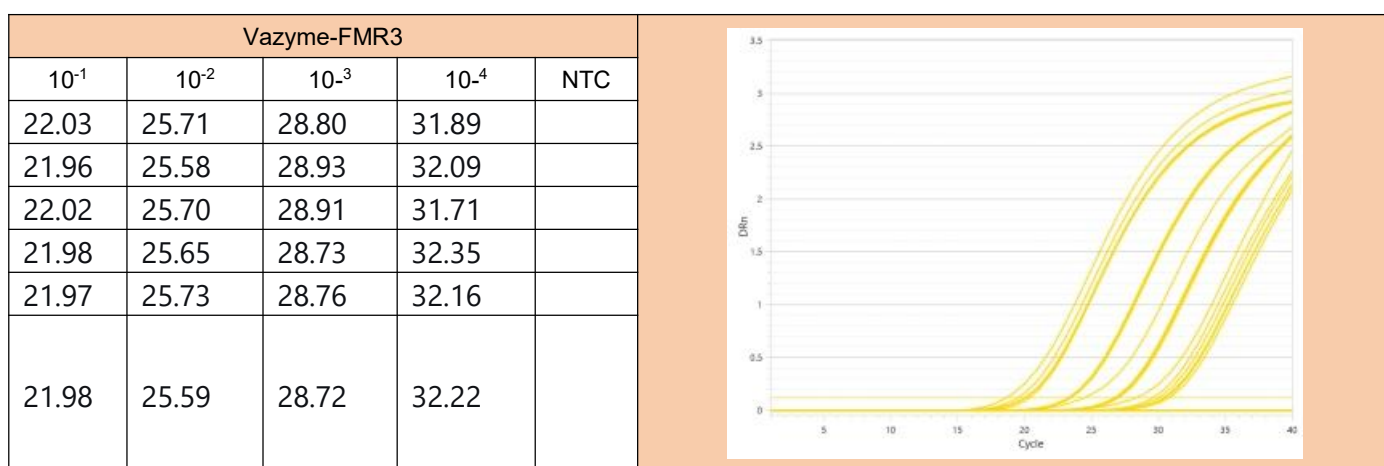
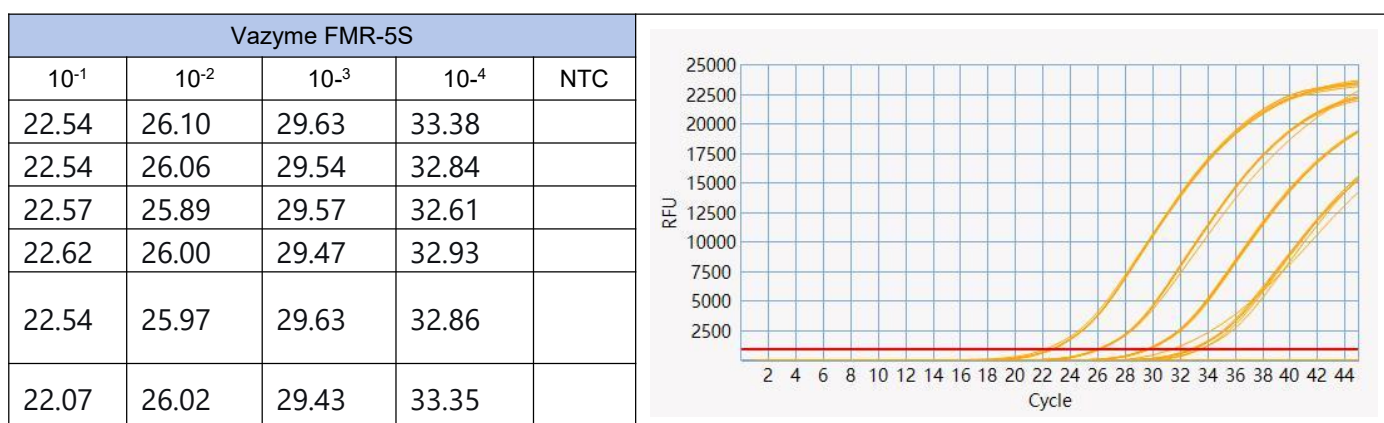
(1) Experimental Design:

Using 293 cDNA (100ng, 10ng, 1ng, 0.1ng) as the template, amplification was performed with Vazyme Probe-based reagent on different real-time PCR instruments including Vazyme FMR-5S and Vazyme FMR3. Amplification efficiency was compared across these systems.

Reagents:	QN222
procedures	37°C 2 min
	95°C 30 sec
	95°C 10 sec
	60°C 30 sec

] 40x

(2) Experimental Data



R ²	
Vazyme FMR-5S	Vazyme FMR3
0.9995	0.9997

(3) Experimental Conclusion:

The results of instrument performance assessment based on the amplification efficiency of Vazyme Probe-based reagent demonstrated:

Vazyme FMR-5S $\approx$ Vazyme FMR3

3. Limit of Detection (LOD)

(1) Experimental Design:

Using 293 cDNA at low concentrations (1ng, 0.5ng, 0.1ng, 0.05ng, 0.01ng) as template, amplification was performed with Vazyme QN222-H23-ROX probe system on two instruments: Vazyme FMR-5S and Vazyme FMR3. Each concentration had 12 technical replicates. Performance was evaluated based on positive detection rate and Ct value consistency at limiting dilution..

(2) Experimental Data:

Positive Detection Rate (%) — 12 replicates per concentration:

Concentration	Vazyme FMR-5S	Vazyme FMR3
1ng	12/12 (100%)	12/12 (100%)
0.5ng	12/12 (100%)	12/12 (100%)
0.1ng	12/12 (100%)	12/12 (100%)
0.05ng	12/12 (100%)	12/12 (100%)
0.01ng	12/12 (100%)	5/12 (41.7%)

Ct Values at 0.01ng (Mean $\pm$ SD of positive wells):

Instrument	n	Ct Mean	Ct SD
Vazyme FMR-5S	12	36.47	0.85
Vazyme FMR-3	5	37.10	0.74

(3) Experimental Conclusion:

Performance assessed by positive detection rate at 0.01ng (higher is better):

Vazyme FMR-5S (100%) > Vazyme FMR3 (41.7%).

Performance assessed by Ct consistency at 0.01ng (lower SD is better):

Vazyme FMR3 (0.74) > Vazyme FMR-5S (0.85)

To summary :

FMR-5S offers significantly more reliable detection at limiting dilution, with a clear advantage in low-template detection capability.

4. Six-Channel Consistency

(1) Experimental Design:

To evaluate six-channel detection consistency across different instruments, amplification was performed using Vazyme QN222 Probe qPCR Mix with a six-channel probe mix (FAM, VIC, ROX, CY3, CY5, CY5.5) in a single-well format. 24 evenly distributed test wells were used per instrument, with template concentration fixed at 1×10^4 copies/reaction..

Channel availability comparison:

(2) Experimental Data:

Instrument	FAM	VIC	ROX	CY3(TAMRA)	CY5	CY5.5	Total
Vazyme FMR-5S	√	√	√	√	√	√	6
Vazyme FMR3	√	√	√	×	√	√	5

a. Channel Availability

Vazyme FMR-5S supports all six required channels (FAM, VIC, ROX, CY3, CY5, CY5.5), providing full six-channel multiplexing capability without channel substitutions or limitations. Vazyme FMR3 supports five channels (FAM, VIC, ROX, CY5, CY5.5, it can be customized to 6 channels).

b. Six-Channel Ct Values

Instrument	FAM Mean (SD)	VIC Mean (SD)	ROX Mean (SD)	CY3 Mean (SD)	CY5 Mean (SD)	CY5.5 Mean (SD)	Max Channel ΔCt
Vazyme FMR-5S	25.60 (0.08)	24.61 (0.10)	25.98 (0.13)	28.10 (0.16)	25.17 (0.06)	24.72 (0.10)	3.49
Vazyme FMR3	24.33 (0.10)	23.21 (0.11)	21.62 (0.22)	—	24.56 (0.12)	22.42 (0.17)	2.94

c. Well-to-Well Reproducibility (24-well SD per channel)

Instrument	FAM SD	VIC SD	ROX SD	CY3 SD	CY5 SD	CY5.5 SD
Vazyme FMR-5S	0.08	0.10	0.13	0.16	0.06	0.10
Vazyme FMR3	0.10	0.11	0.22	—	0.12	0.17

(3) Experimental Conclusion:

Performance assessed by channel availability (more channels is better):

Vazyme FMR-5S (6 channels) > Vazyme FMR3 (5 channels).

Performance assessed by six-channel consistency (lower max ΔC_t is better, among available channels):

Vazyme FMR-3 (2.94) > Vazyme FMR-5S (3.49).

Performance assessed by well-to-well reproducibility (lower SD is better):

Vazyme FMR-5S (0.06–0.16) > Vazyme FMR-3 (0.10–0.22).

5. Fast Ramp Rate Performance

(1) Experimental Design:

To evaluate the practical utility of fast amplification protocols, we compared the total runtime and Ct values between standard and fast programs on two instruments: Vazyme FMR-5S and Vazyme FMR-3. Each condition had 4 technical replicates. Two qPCR reagents were tested: Vazyme QN222 Probe qPCR Mix and Vazyme Q713 SYBR qPCR Mix

Reagent	Program	Amplification Conditions
QN222	Standard	95°C 30s; [95°C 10s, 60°C 30s] × 40 cycles
	Fast	95°C 30s; [95°C 1s, 60°C 10s] × 40 cycles
Q713	Standard	95°C 30s; [95°C 10s, 60°C 30s] × 40 cycles + Melt curve
	Fast	95°C 30s; [95°C 1s, 60°C 7s] × 40 cycles + Melt curve

(2) Experimental Data:

a. QN222 Probe qPCR Mix — Standard vs Fast Program

Instrument	Program	Ct 1	Ct 2	Ct 3	Ct 4	Ct Mean	Ct SD	Runtime
Vazyme FMR-5S	Standard	25.80	25.92	25.92	25.75	25.85	0.08	64 min
	Fast	26.04	26.13	26.11	26.05	26.08	0.04	30 min
Vazyme FMR3	Standard	23.44	23.39	23.49	23.64	23.49	0.11	81 min
	Fast	23.99	24.02	24.12	24.02	24.04	0.06	38 min

b. Q713 SYBR qPCR Mix — Standard vs Fast Program

Instrument	Program	Ct 1	Ct 2	Ct 3	Ct 4	Ct Mean	Ct SD	Runtime
Vazyme FMR-5S	Standard	21.32	21.28	21.31	21.33	21.31	0.02	66 min
	Fast	22.13	22.11	22.16	22.15	22.14	0.02	32 min
Vazyme FMR3	Standard	20.80	20.48	20.43	20.46	20.54	0.18	88 min
	Fast	21.25	21.30	21.44	21.54	21.38	0.13	67 min

(3) Experimental Conclusion:

Performance assessed by fast program runtime (shorter is better):

Vazyme FMR-5S (30–32 min) > Vazyme FMR3 (38–67 min)

Performance assessed by fast program data reliability (Ct shift and SD):

Vazyme FMR-5S ≈ Vazyme FMR3

Both instruments demonstrated comparable fast-mode reliability, with FMR-5S showing slightly better reproducibility (SD = 0.02–0.04) compared to FMR-3 (SD = 0.06–0.13).

6. SNP Genotyping (Allelic Discrimination)

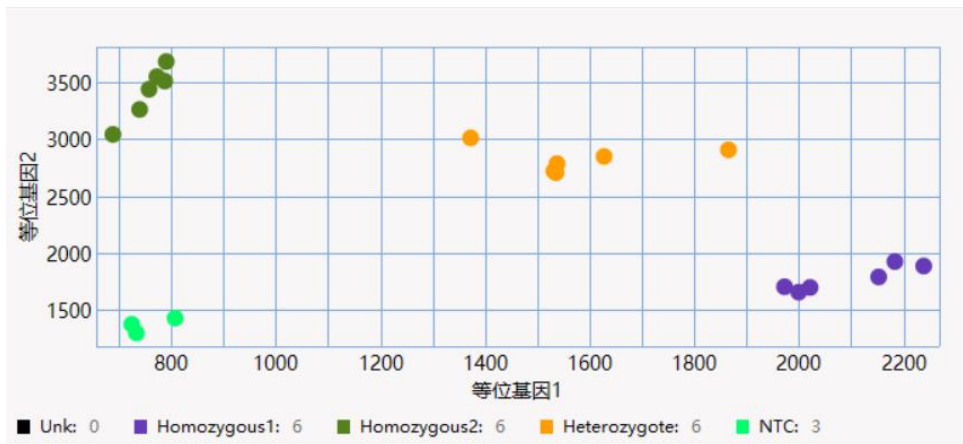
(1) Experimental Design:

Using Vazyme QK121 with FAM-labeled wild-type probe and HEX-labeled mutant probe, three known genotype standards (Wild-type, Mutant, Heterozygous) were tested across two instruments: Vazyme FMR-5S and Vazyme FMR-3. Each genotype had 6 technical replicates. End-point fluorescence signals (FAM/HEX) were read after 40 cycles to evaluate allelic discrimination performance. NTC controls were included for each instrument.

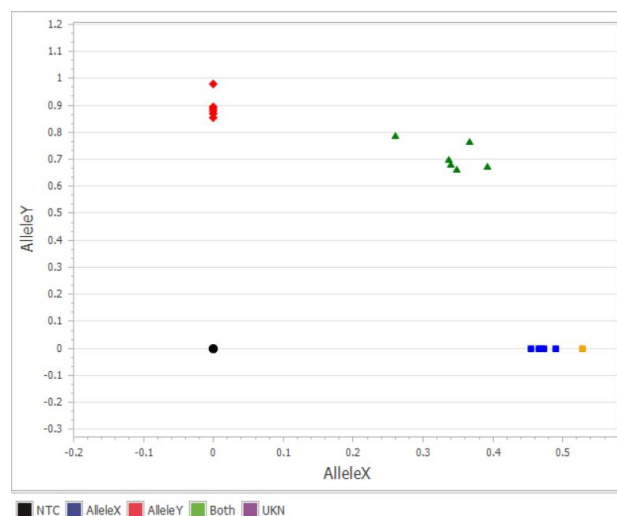
(2) Experimental Data:

a. Allelic Discrimination Scatter Plot Summary

Vazyme FMR-5S:



Vazyme FMR3



b. Genotyping Performance Summary

Instrument	WT (6 reps)	MUT (6 reps)	HET (6 reps)	Overall Accuracy
Vazyme FMR-5S	6/6 (100%)	6/6 (100%)	6/6 (100%)	18/18 (100%)
Vazyme FMR3	6/6 (100%)	6/6 (100%)	6/6 (100%)	18/18 (100%)

(3) Experimental Conclusion:

Performance assessed by genotyping accuracy (higher is better):

Vazyme FMR-5S (100%) = Vazyme FMR3 (100%).

Performance assessed by cluster separation and compactness:

Both Vazyme FMR-5S and Vazyme FMR-3 achieved 100% genotyping accuracy, confirming that both instruments are highly reliable for SNP genotyping applications. WT and MUT clusters were clearly separated on both instruments, with heterozygous clusters readily distinguishable. These results demonstrate that both instruments provide robust and accurate SNP genotyping performance.