Supplementary Table 1. Optimization parameters for 5-azacytidine and 5-methyl-2'-deoxycytidine

	Declustering potential (V)	Entrance potential (V)	Collision energy (eV)	Collision cell exit potential (V)
5-Azacytidine	106	10	31	14
5-methyl-2'-deoxycytidine	86	10	31	8

Supplementary Table 2. Validation characteristics of 5-azacytidine

	LLOQ (5 ng/mL)	Low (15 ng/mL)	Medium (150 ng/mL)	High (400 ng/mL)	AULQ (4,000 ng/mL)	1mg/mL in 50% Methanol
n	15	15	15	15	15	
Average	5.45	14.67	157	414	4075	
Standard Deviation	0.52	1.08	11.01	28.42	321.16	
Accuracy (%)	109.1	97.8	104.4	103.4	101.9	
Within-day Precision (%)	9.8	5.1	5.5	5.4	6.4	
Between-day Precision (%)	**	6.4	5.2	5	5.5	
Stock Stability (-2-°C) (46 Days) (%)						92.4
Plasma Stability (-70°C) (166 Days) (%)		91.5		100.9	96.7	
Plasma Stability (-70°C) (323 Days) (%)		85.7		89.5	91.0	

** No additional information was observed as a result of performing the assay in different runs.

Supplementary Table 3. Matrix Effects and Recovery of 5-azacytidine (n=3)

Sample Name	Previous Published Assay[1]				Current Assay			
	Average Area Counts	Analyte Matrix Effect	Analyte Absolute Recovery	Analyte Combined Recovery	Average Area Counts	Analyte Matrix Effect	Analyte Absolute Recovery	Analyte Combined Recovery
QC Low Neat	2.06E+06				2.06E+06			
QC Low Post Spike	1.46E+06	-29%	5%	4%	1.01E+06	-51%	35%	17%
QC Low Pre Spike	7.32E+04				3.57E+05			
QC Mid Neat	2.21E+07				2.05E+07			
QC Mid Post Spike	1.48E+07	-33%	2%	1%	9.28E+06	-55%	36%	16%
QC Mid Pre Spike	2.37E+05				3.37E+06			
QC High Neat	5.75E+07				5.82E+07			
QC High Post Spike	3.72E+07	-35%	4%	3%	2.66E+07	-54%	34%	15%
QC High Pre Spike	1.58E+06				8.98E+06			