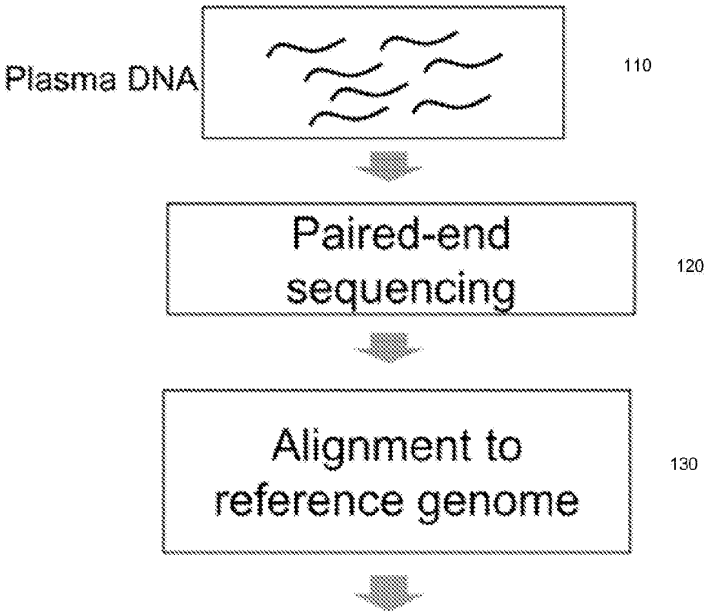
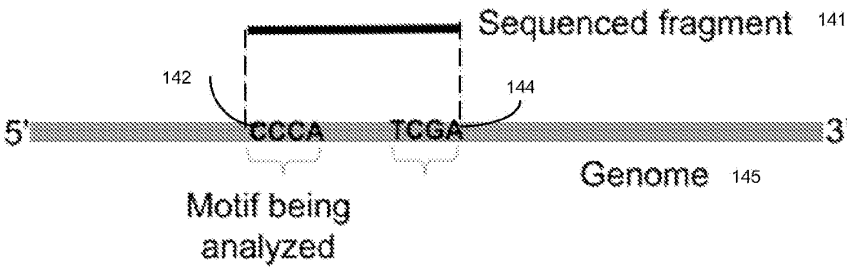


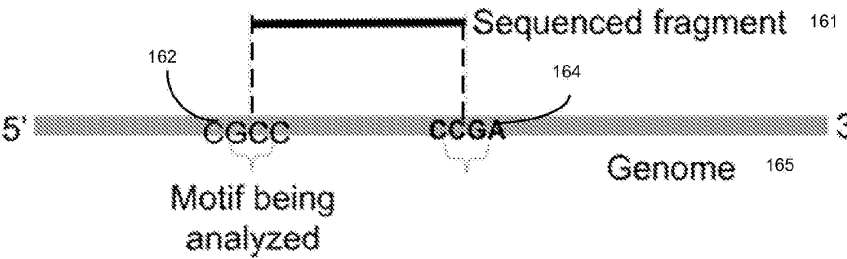
[Fig. 1]



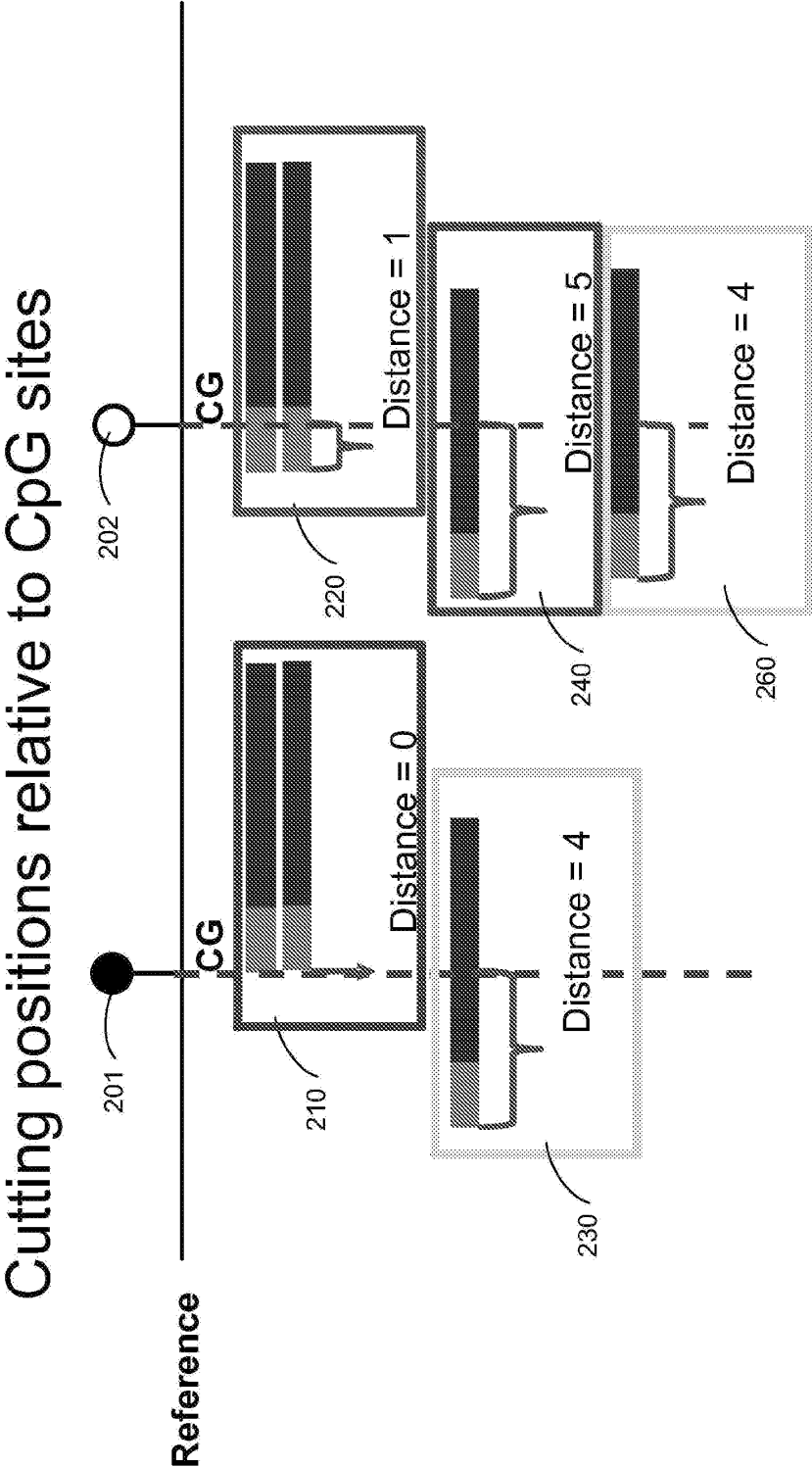
Technique 140 : 4-mer on the ends of a fragment



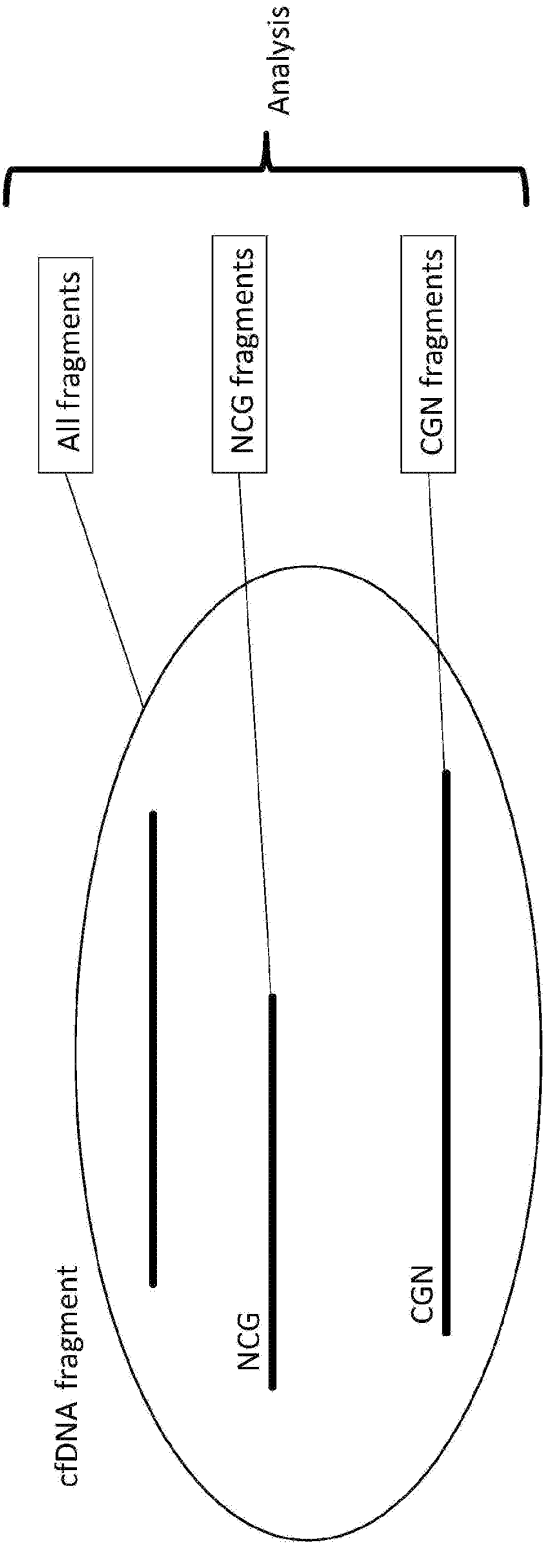
Technique 160 : 4-mer jointly constructed from 2-mer on the ends of a fragment plus 2-mer on the genomic regions adjacent to ends of a fragment



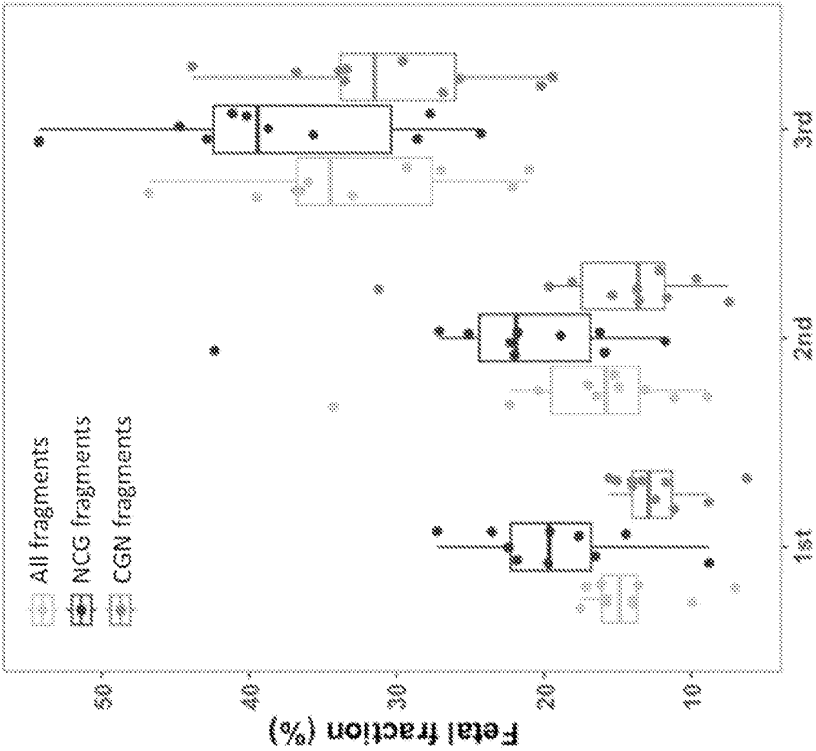
[Fig. 2]



[Fig. 3]



[Fig. 4A]



[Fig. 4B]

Trimester	Median fetal DNA fraction (%)		
	All fragments	NCG fragments	CGN fragments
1st	14.85	19.67	12.87
2nd	15.80	21.89	13.61
3rd	34.50	39.45	31.48

Trimester	Relative change to all fragments (%)	
	NCG fragments	CGN fragments
1st	32.46	-13.33
2nd	38.54	-13.86
3rd	14.35	-8.75

[Fig. 5A 5B]

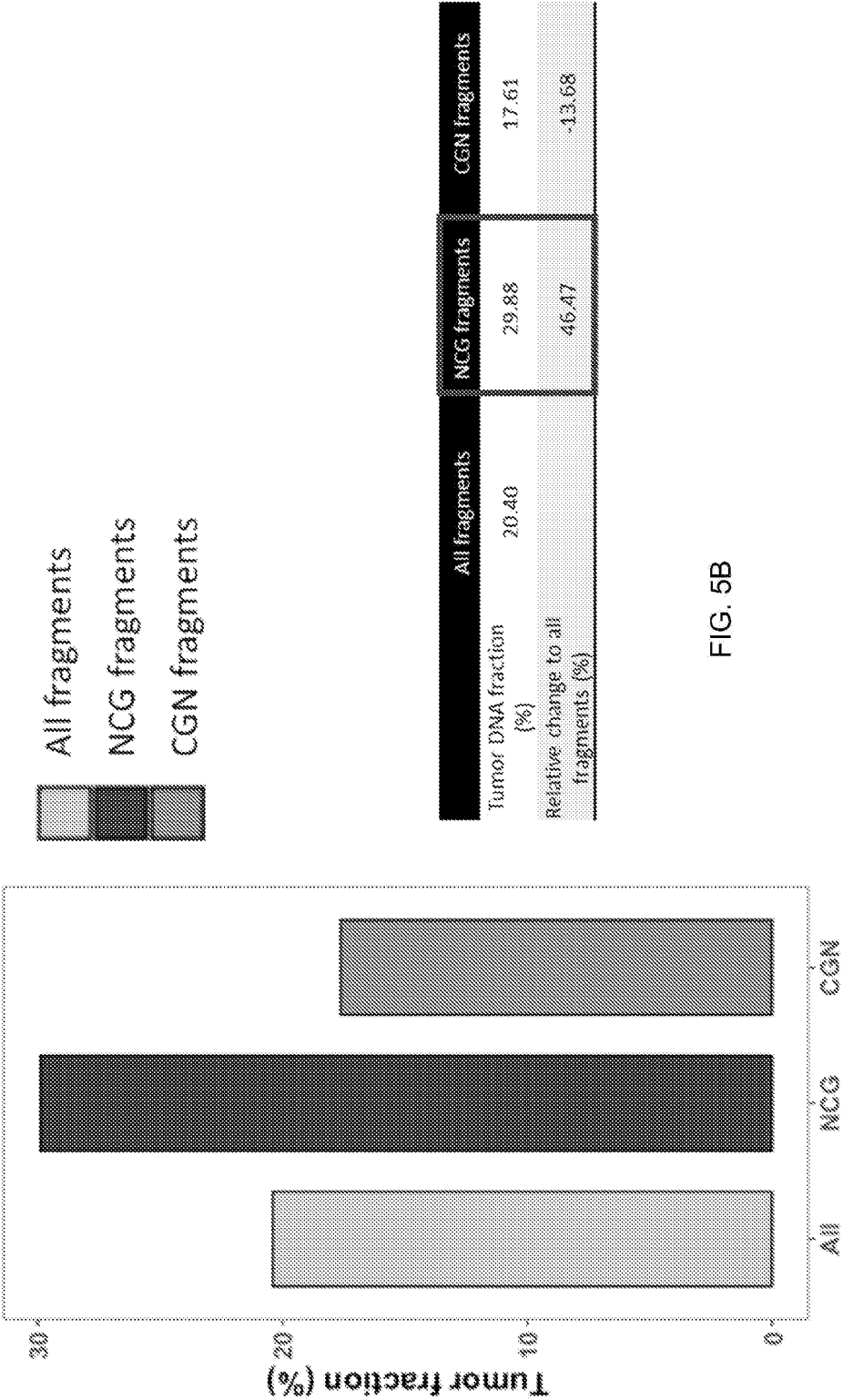
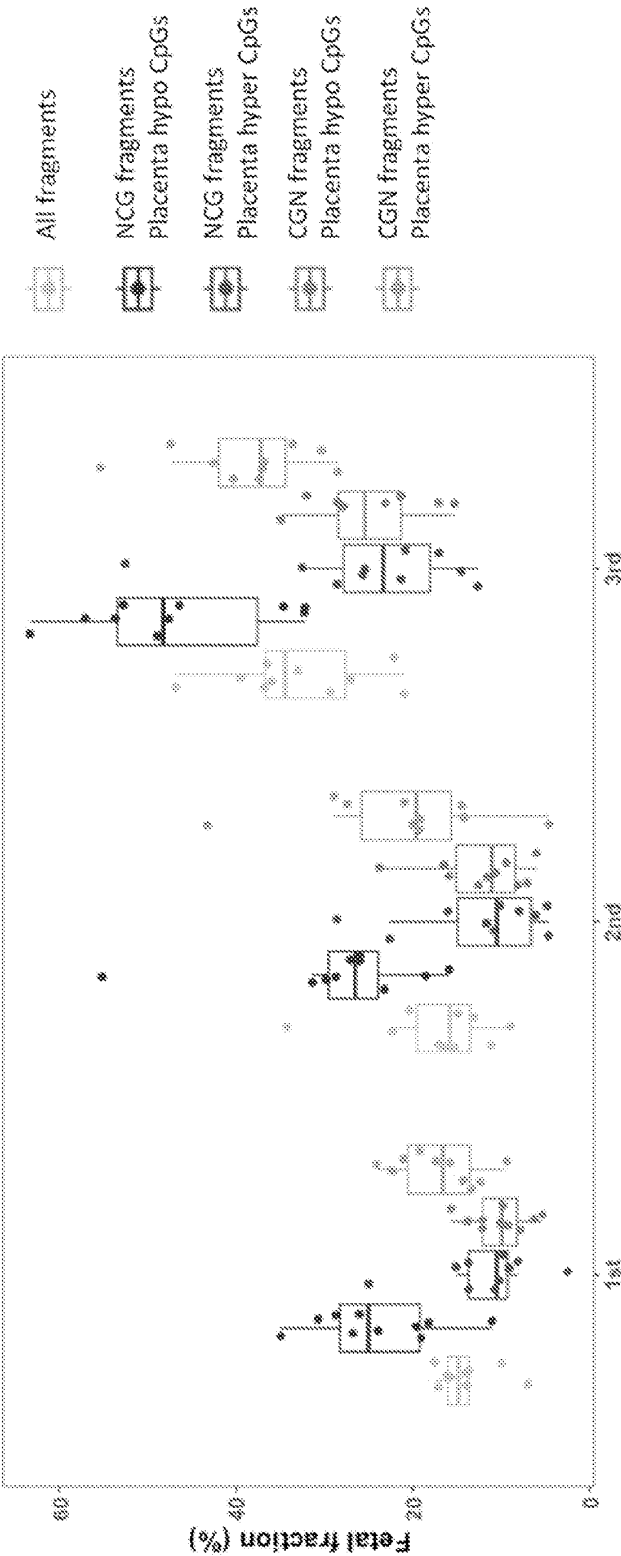


FIG. 5B

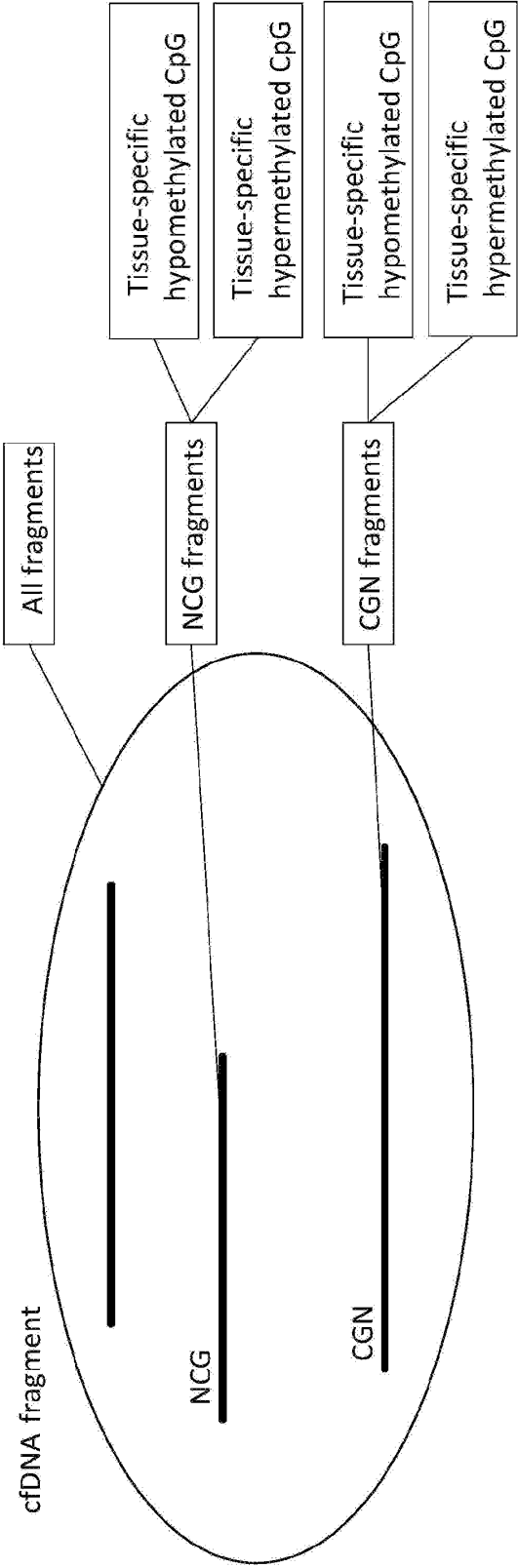
FIG. 5A

[Fig. 6]

Further enrichment in NCG ended DNA fragments from tissue-specific hypomethylated CpGs



[Fig. 7]



[Fig. 8A]

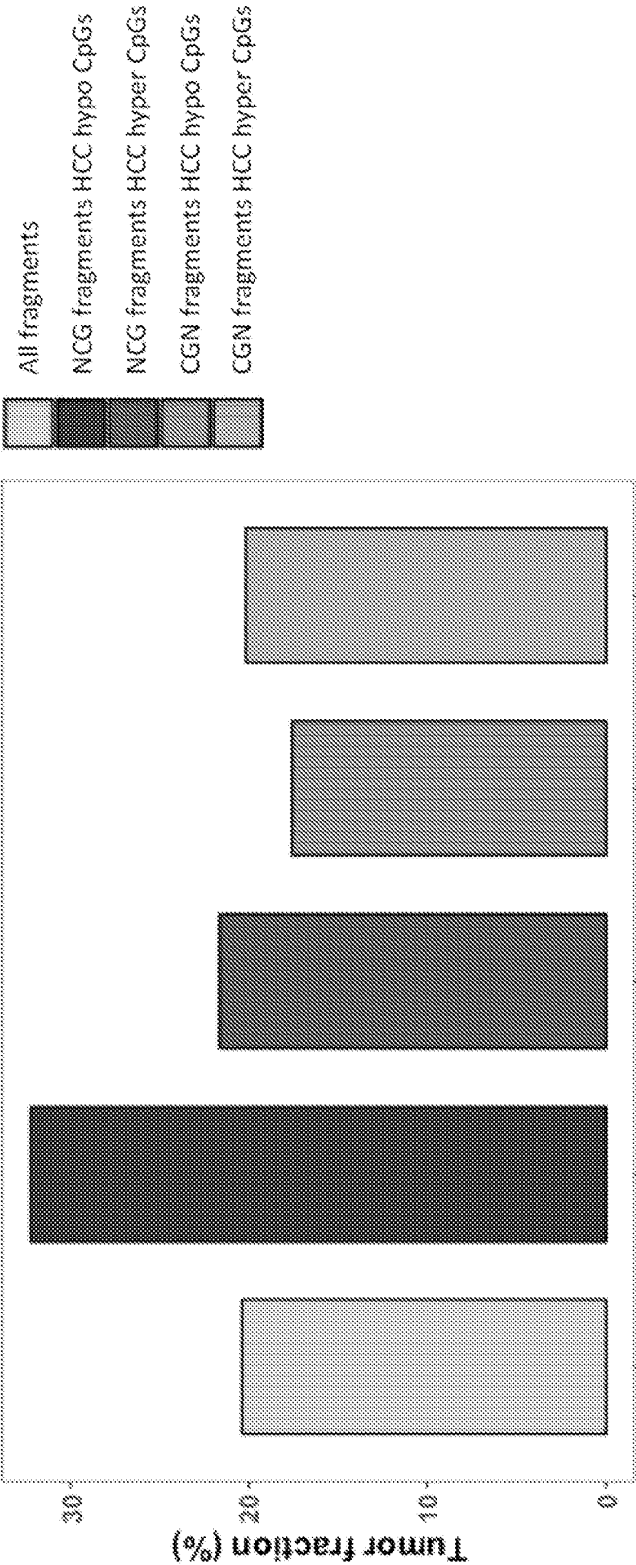
Trimester	Median fetal DNA fraction (%)			
	All fragments	NCG hypo fragments	NCG hyper fragments	CGN hypo fragments
1st	14.85	24.99	10.50	16.57
2nd	15.80	26.53	10.47	19.62
3rd	34.50	48.27	23.39	37.22

[Fig. 8B]

Trimester	Relative change to all fragments (%)			
	NCG hypo fragments	NCG hyper fragments	CGN hypo fragments	CGN hyper fragments
1st	68.28	-29.29	-33.13	11.58
2nd	67.91	-33.73	-30.00	24.18
3rd	39.91	-32.20	-26.23	7.88

[Fig. 9]

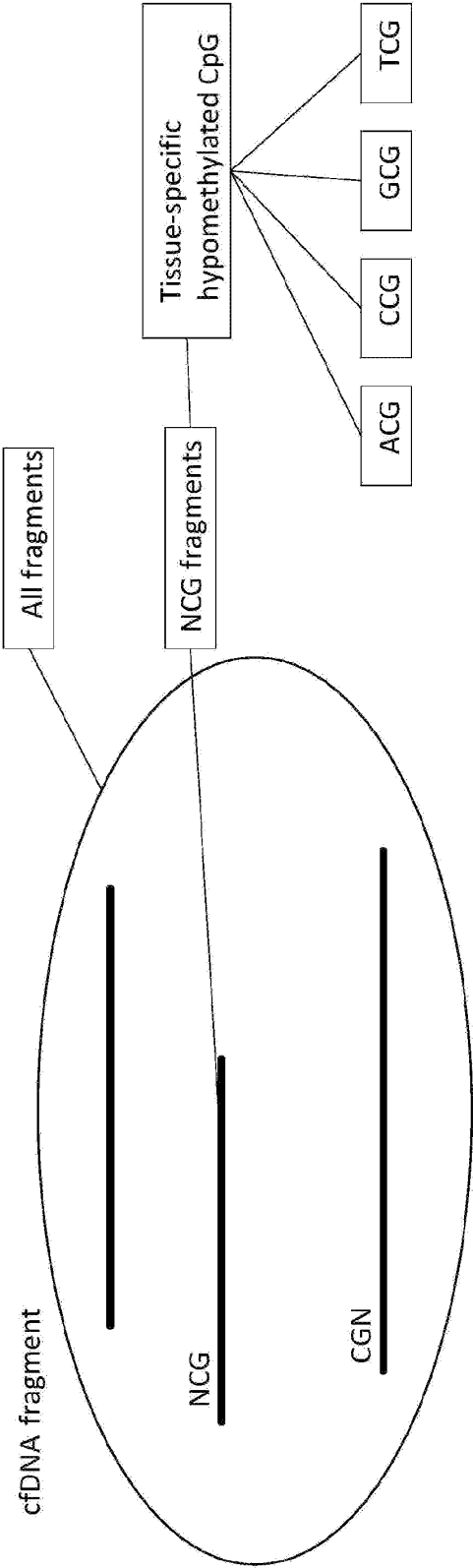
Tumor DNA is further enriched in NCG ended DNA fragments from HCC hypomethylated CpGs



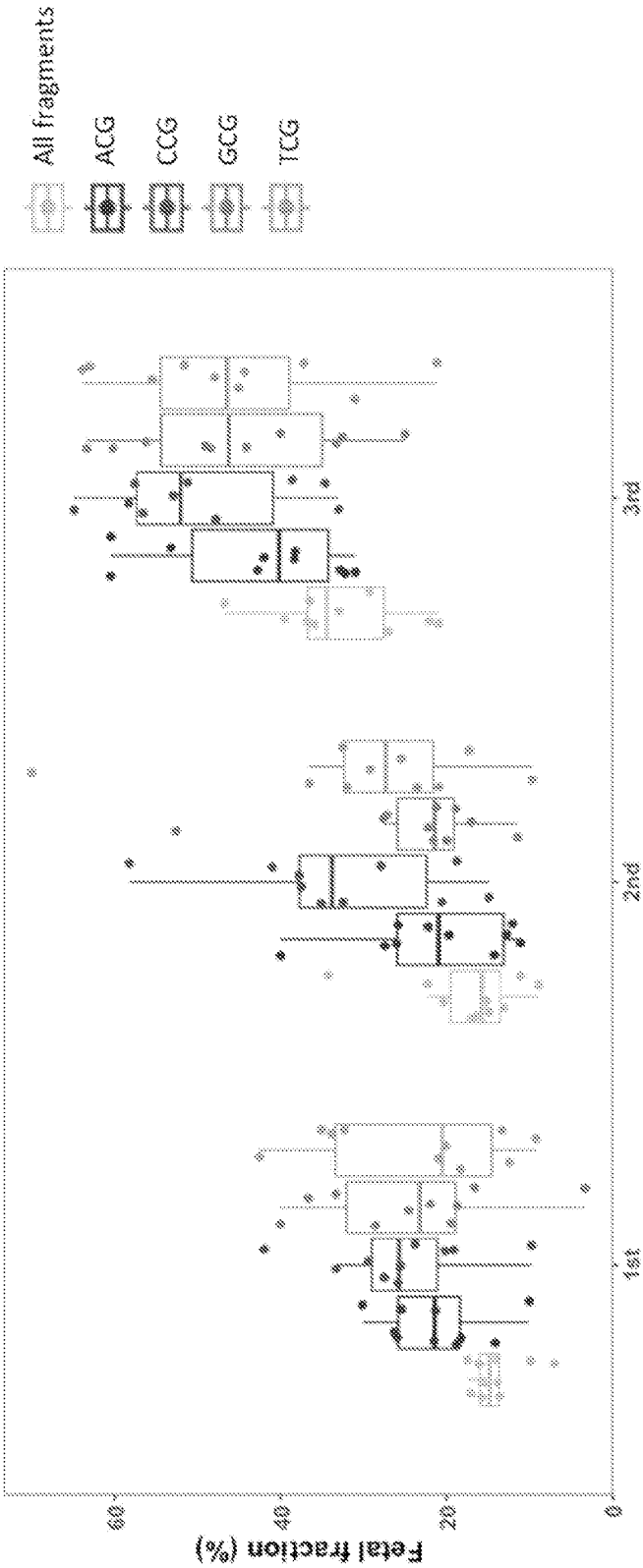
[Fig. 10]

All fragments		NCG hypo fragments		NCG hyper fragments		CGN hypo fragments		CGN hyper fragments	
Tumor DNA fraction (%)		32.25		21.67		17.50		20.17	
Relative change to all fragments (%)		58.10		6.21		-13.74		-1.14	

[Fig. 11]



[Fig. 12]



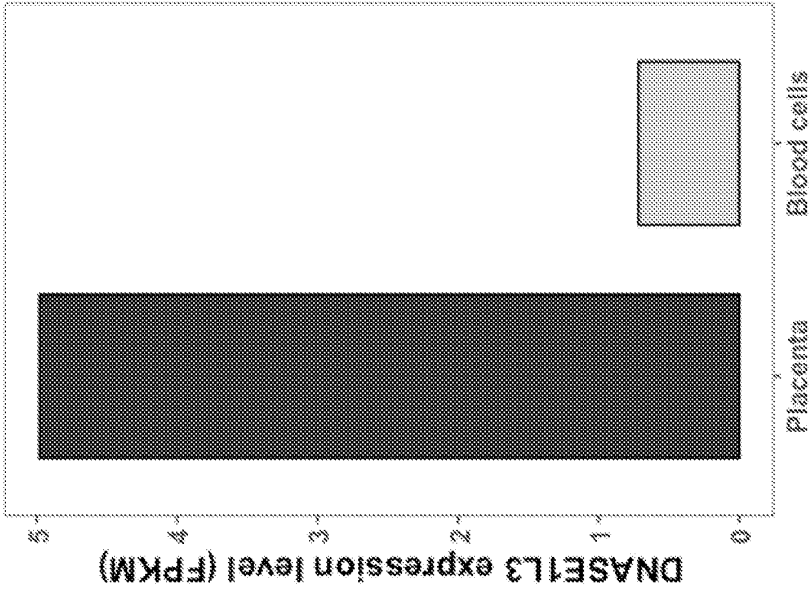
[Fig. 13A]

Trimester	Median fetal DNA fraction (%)				
	All fragments	ACG	CCG	GCG	TCG
1st	14.85	21.45	25.69	23.26	20.53
2nd	15.80	20.99	33.83	21.45	27.31
3rd	34.50	40.18	52.07	46.28	46.52

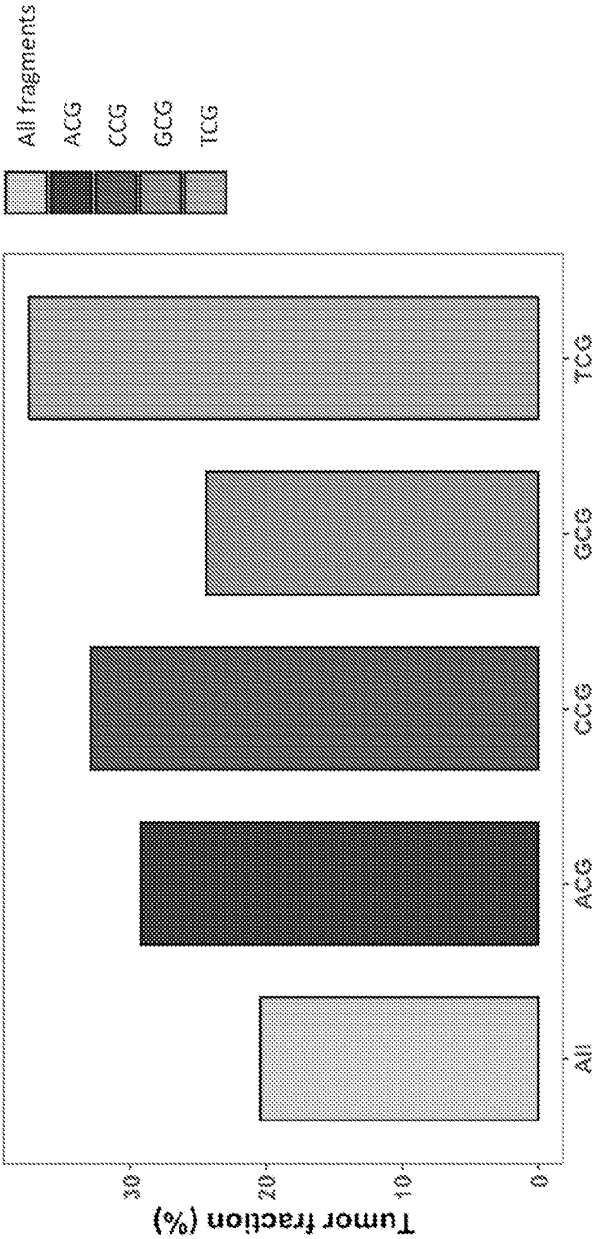
[Fig. 13B]

Trimester	Relative change to all fragments (%)			
	ACG	CCG	GCG	TCG
1st	44.44	73.00	56.63	38.25
2nd	32.85	114.11	35.76	72.85
3rd	16.46	50.93	34.14	34.84

[Fig. 14]
DNASE1L3 expression is higher in placenta tissues compared with blood cells



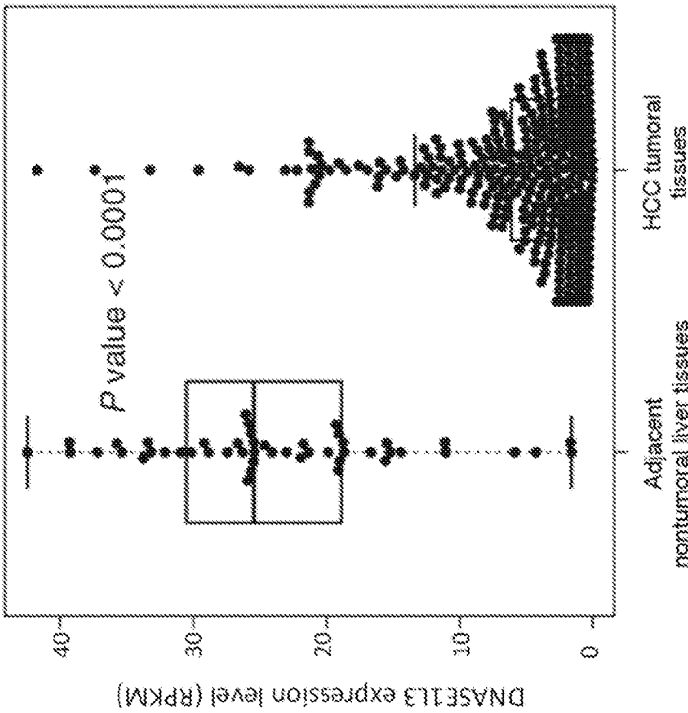
[Fig. 15A]



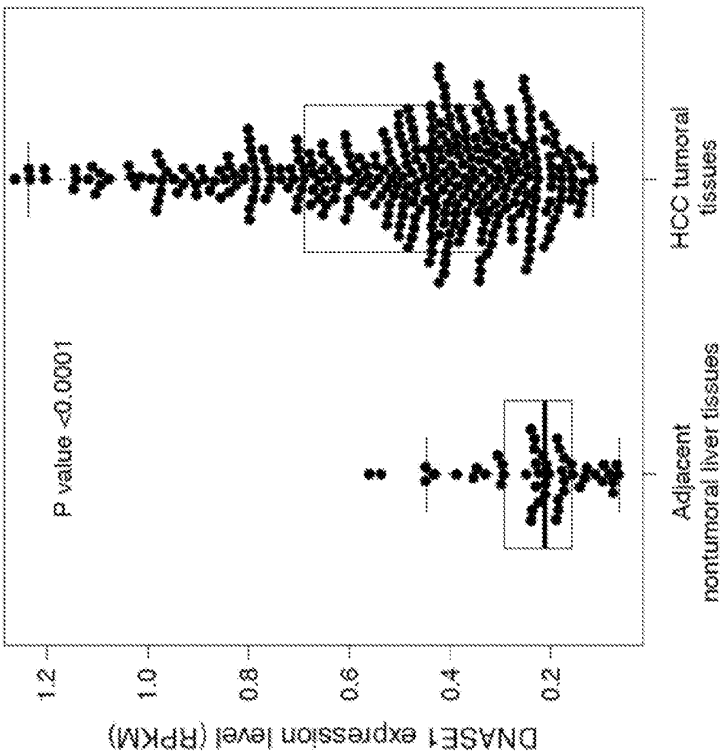
[Fig. 15B]

	All fragments	ACG	CCG	GCG	TCG
Tumor DNA fraction (%)	20.40	29.25	32.95	24.44	37.50
Relative change to all fragments (%)		43.38	61.51	19.83	83.82

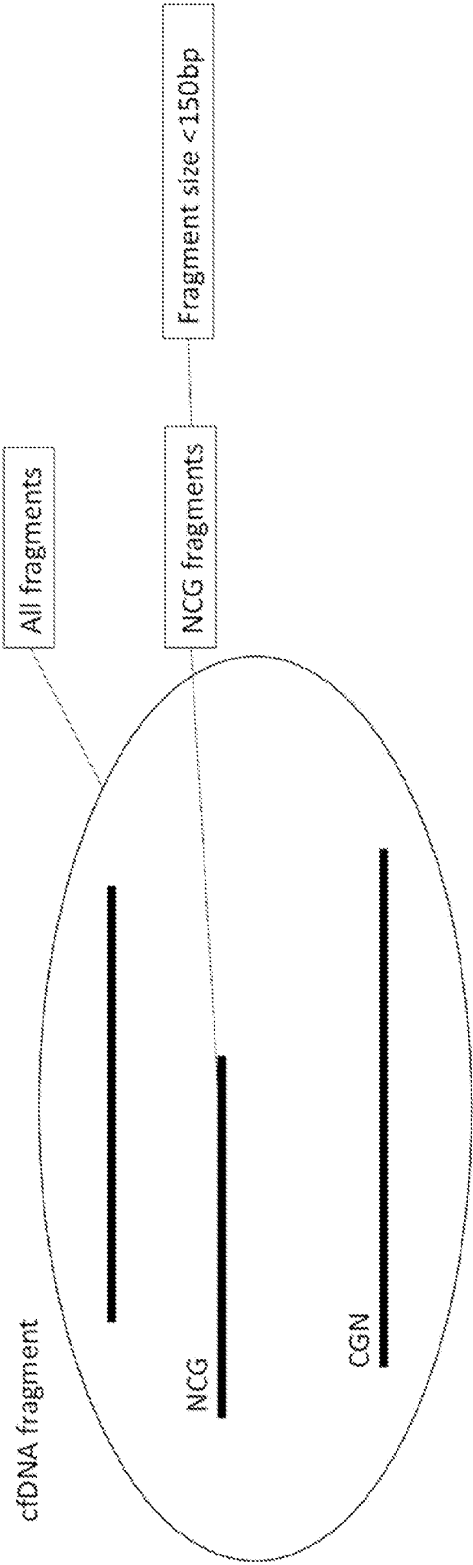
[Fig. 16A]



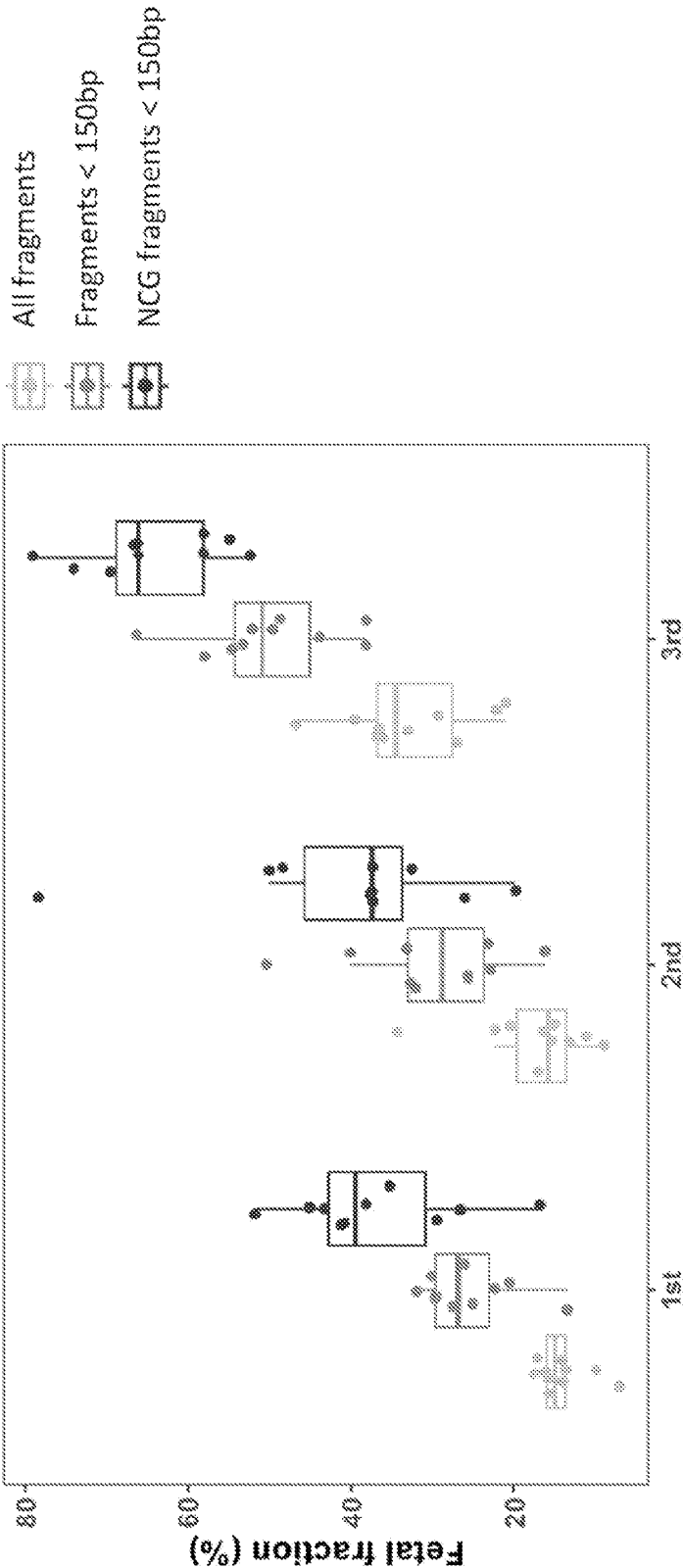
[Fig. 16B]



[Fig. 17]



[Fig. 18]



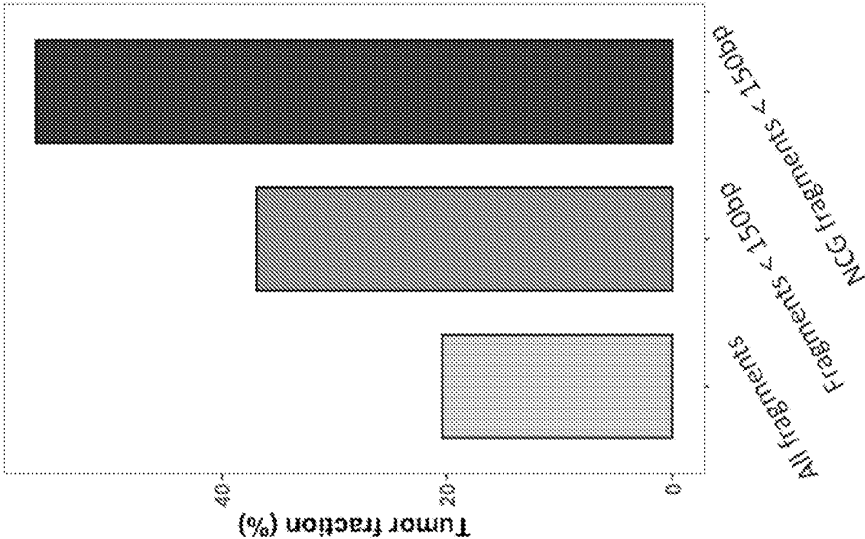
[Fig. 19A]

Trimester	Median fetal DNA fraction (%)		
	All fragments	<150bp	NCG + <150bp
1st	14.85	26.78	39.44
2nd	15.80	28.84	37.34
3rd	34.50	50.88	66.17

[Fig. 19B]

Trimester	Relative change to all fragments (%)	
	<150bp	NCG + <150bp
1st	80.35	165.60
2nd	82.55	136.30
3rd	47.48	91.81

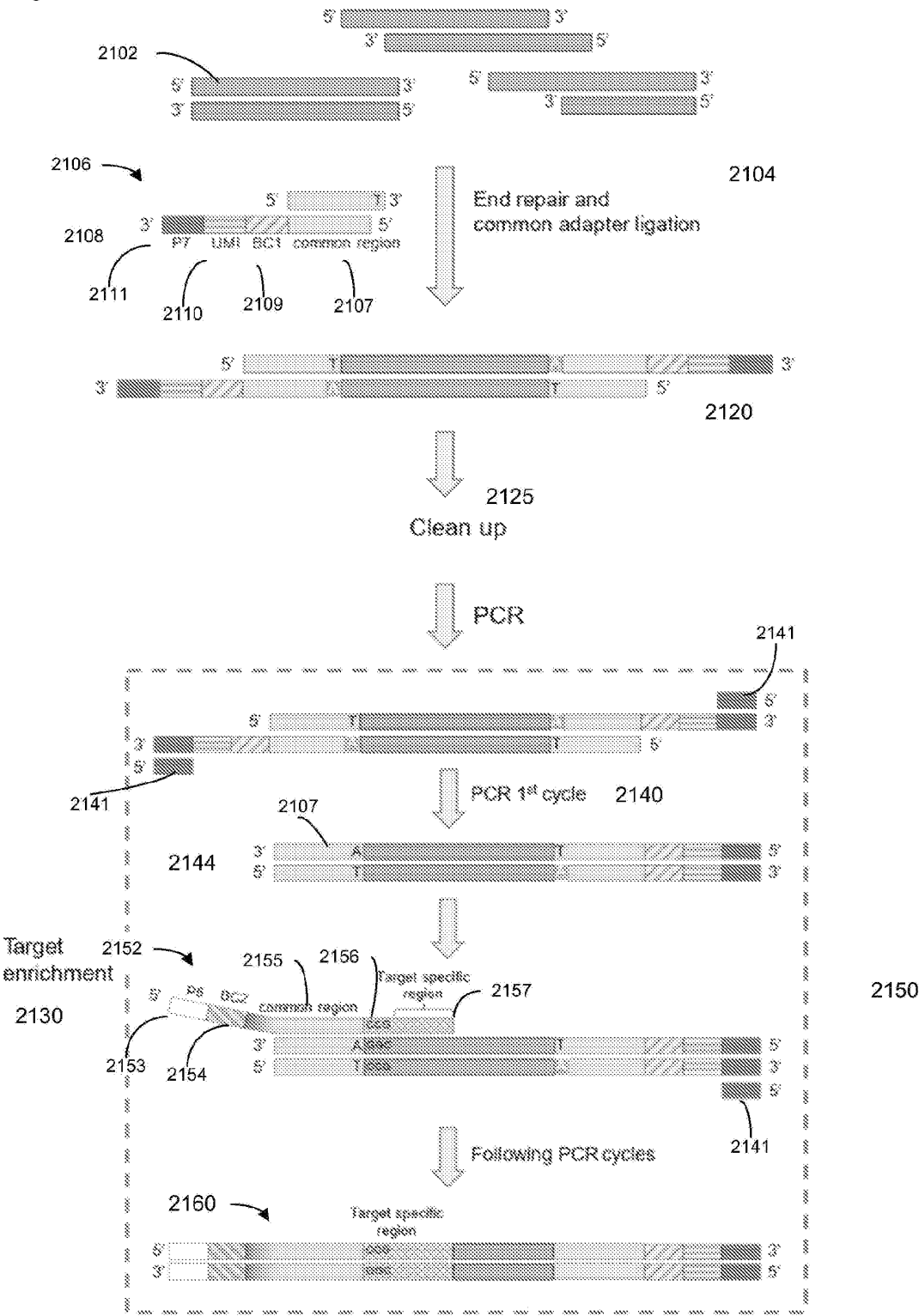
[Fig. 20A]



[Fig. 20B]

All fragments		<150bp	NCG + <150bp
Tumor DNA fraction (%)	20.40	36.88	56.52
Relative change to all fragments (%)		80.78	177.07

[Fig. 21]



[Fig. 22A 22B]

Simulation of number of reads needed for T21 diagnosis

Fetal fraction = 5%
Specificity = 100%

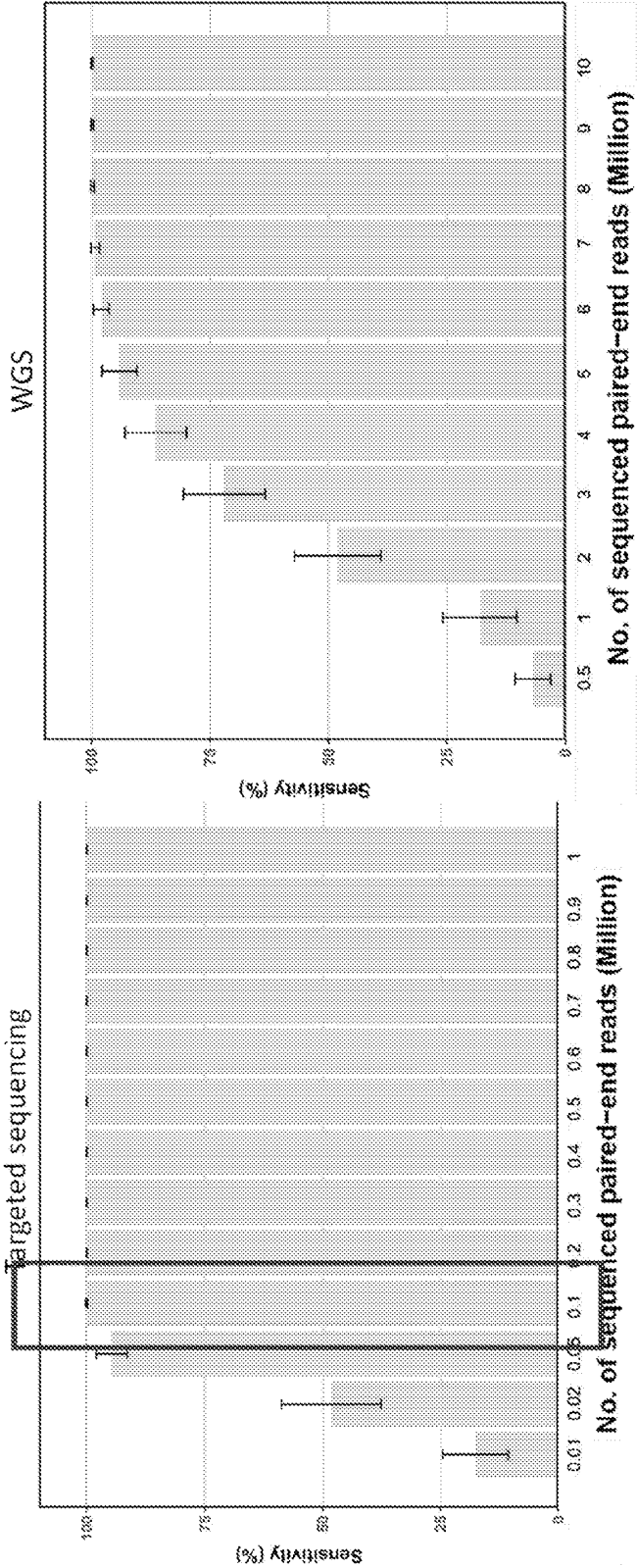


FIG. 22B

FIG. 22A

[Fig. 23]
Example ddPCR workflow

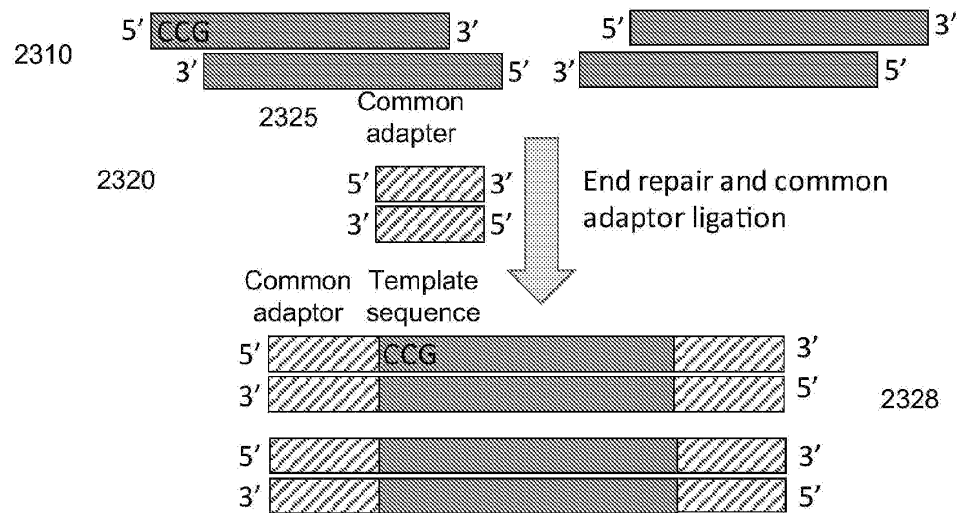
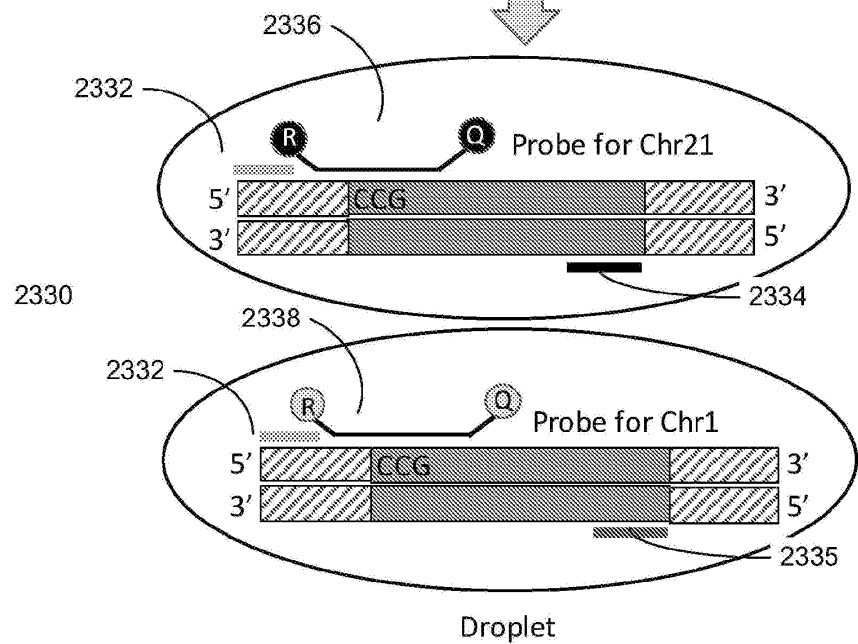


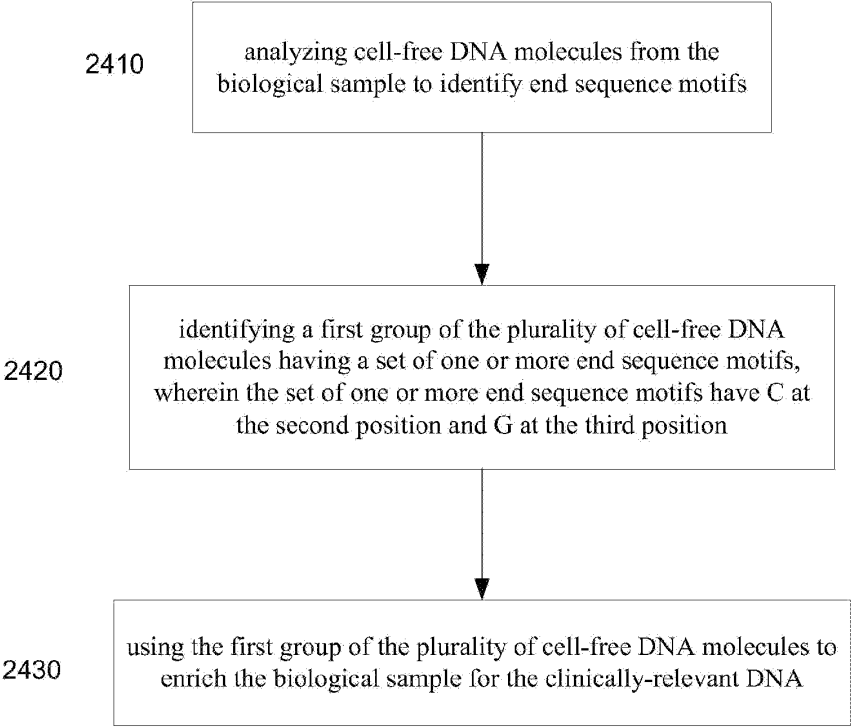
FIG. 23



- (R) Reporter
- (Q) Quencher
- Common forward primer
- Regional specific reverse primer for chr1
- Regional specific reverse primer for chr21

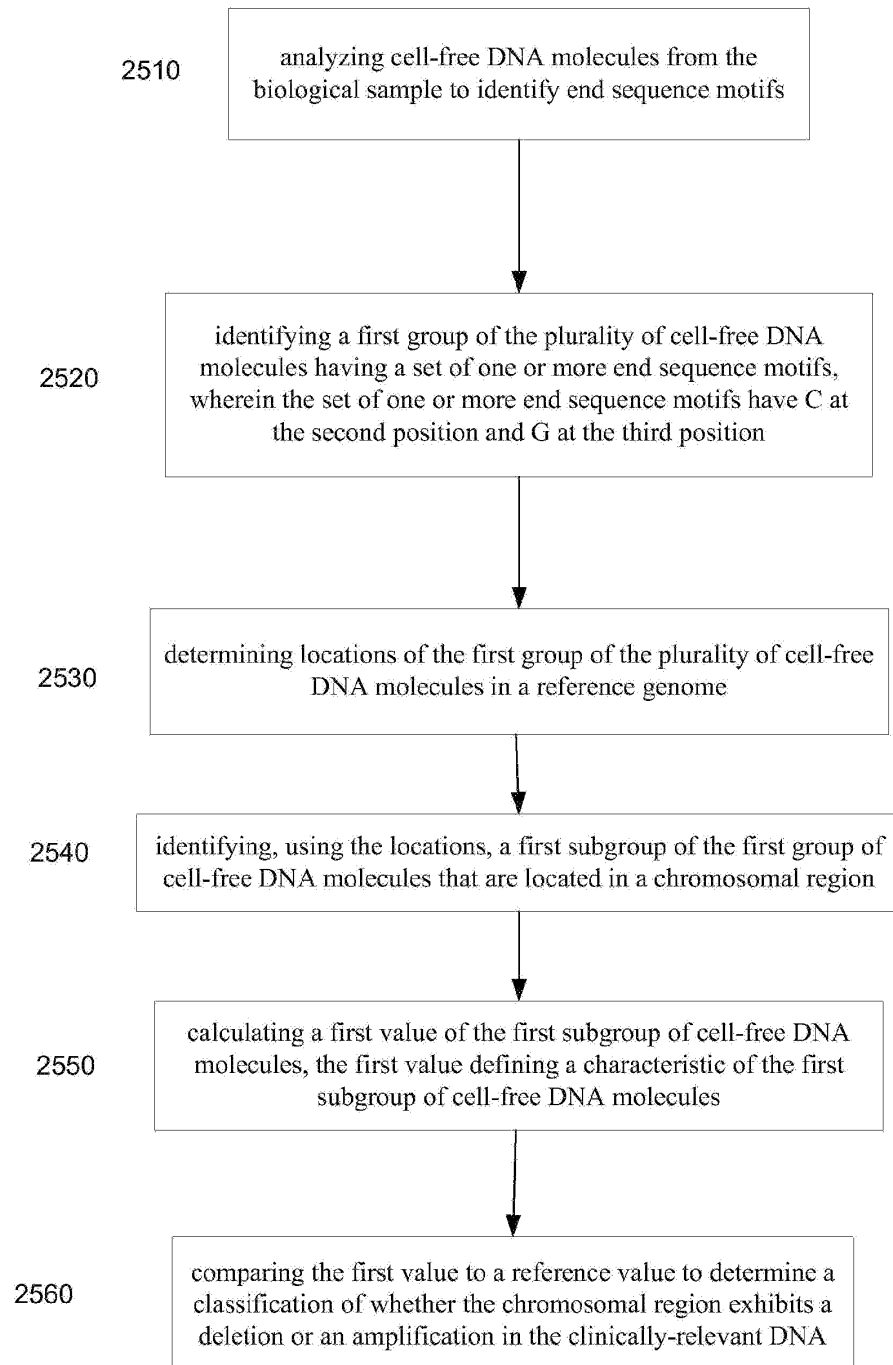
[Fig. 24]

2400



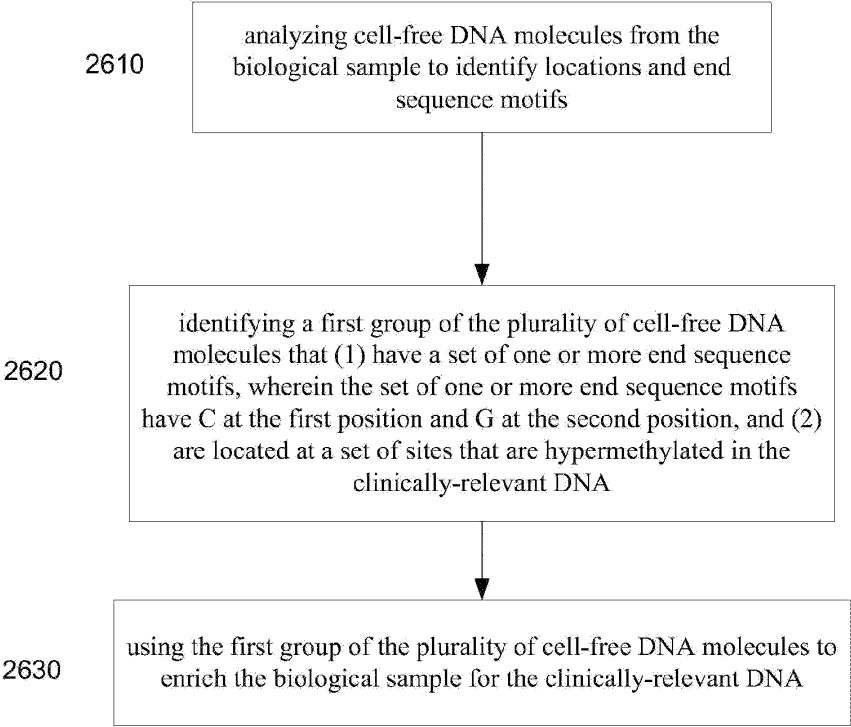
[Fig. 25]

2500

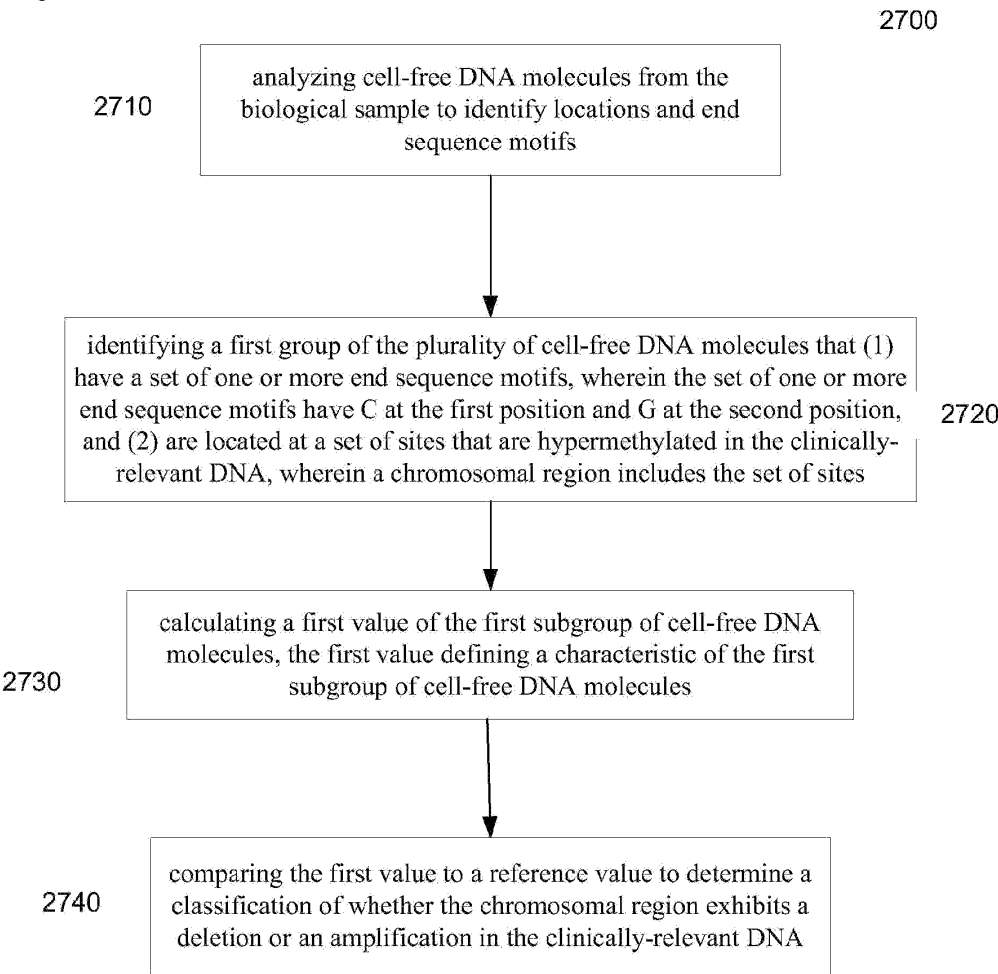


[Fig. 26]

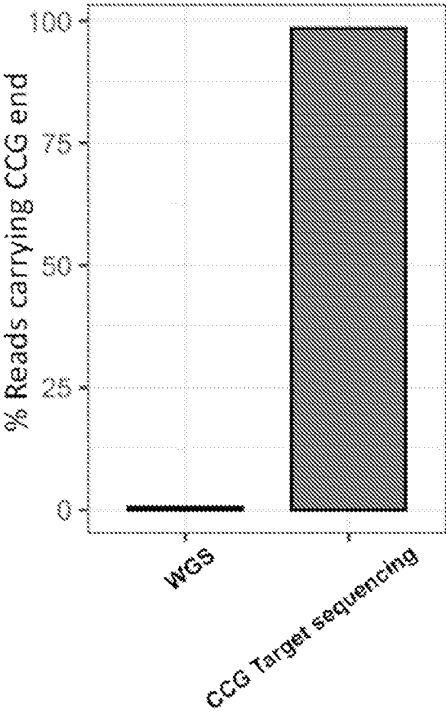
2600



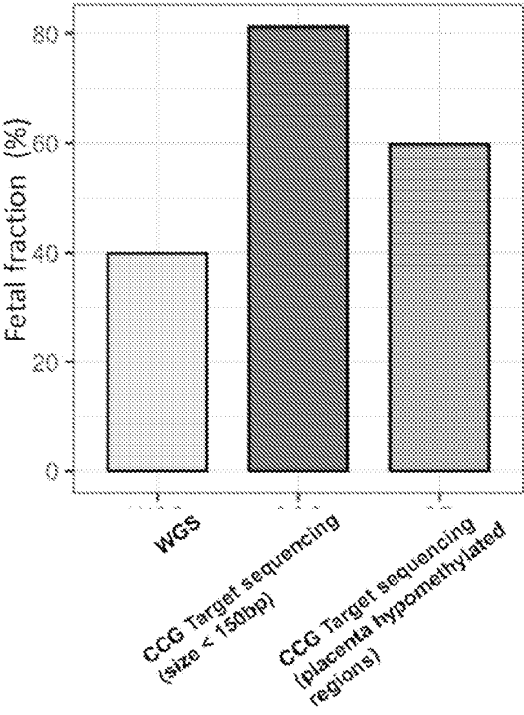
[Fig. 27]



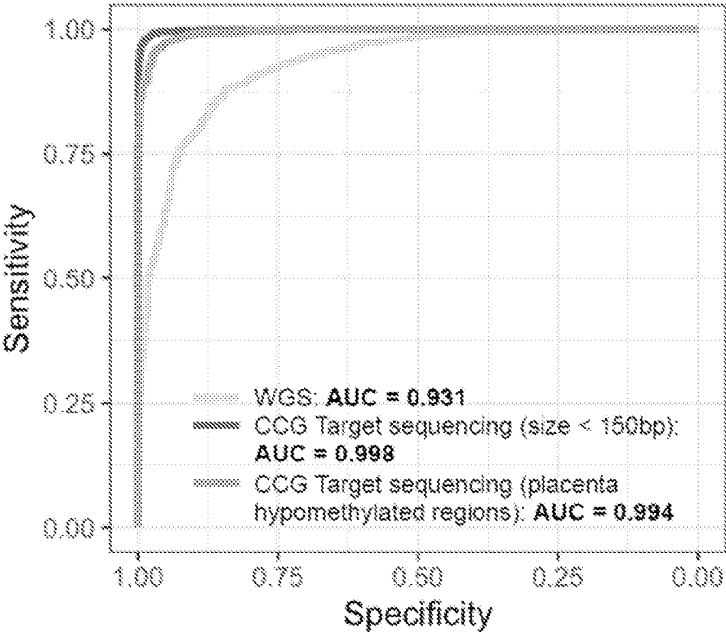
[Fig. 28A]



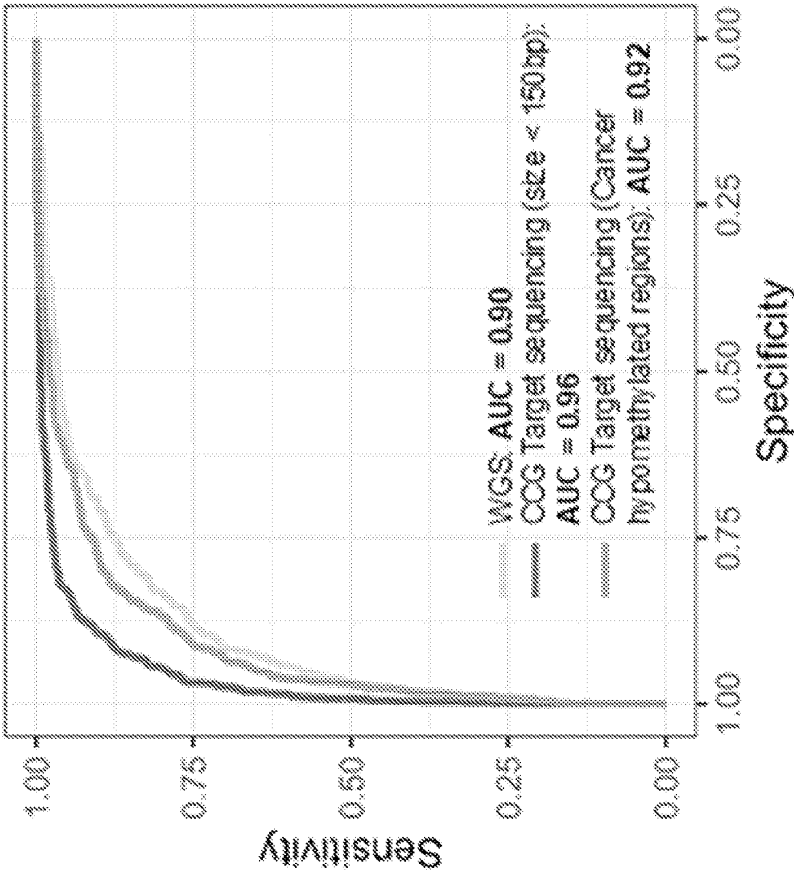
[Fig. 28B]



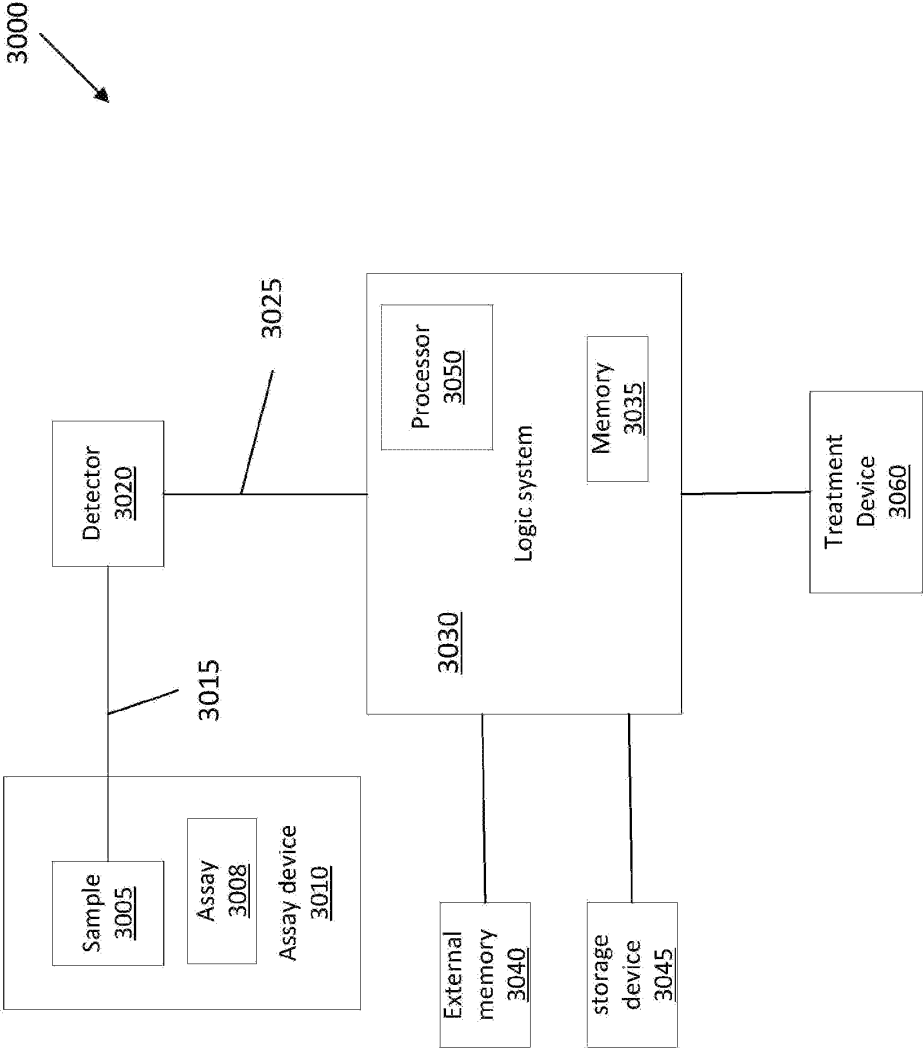
[Fig. 28C]



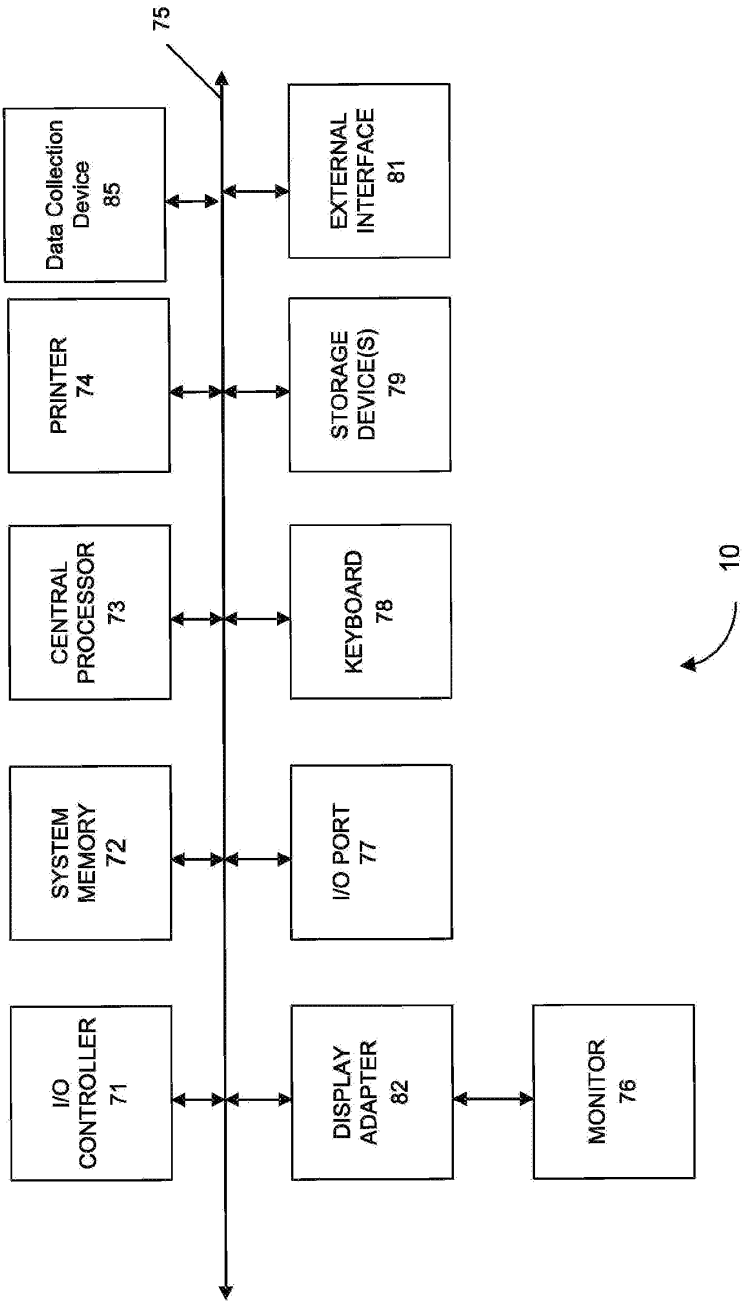
[Fig. 29]



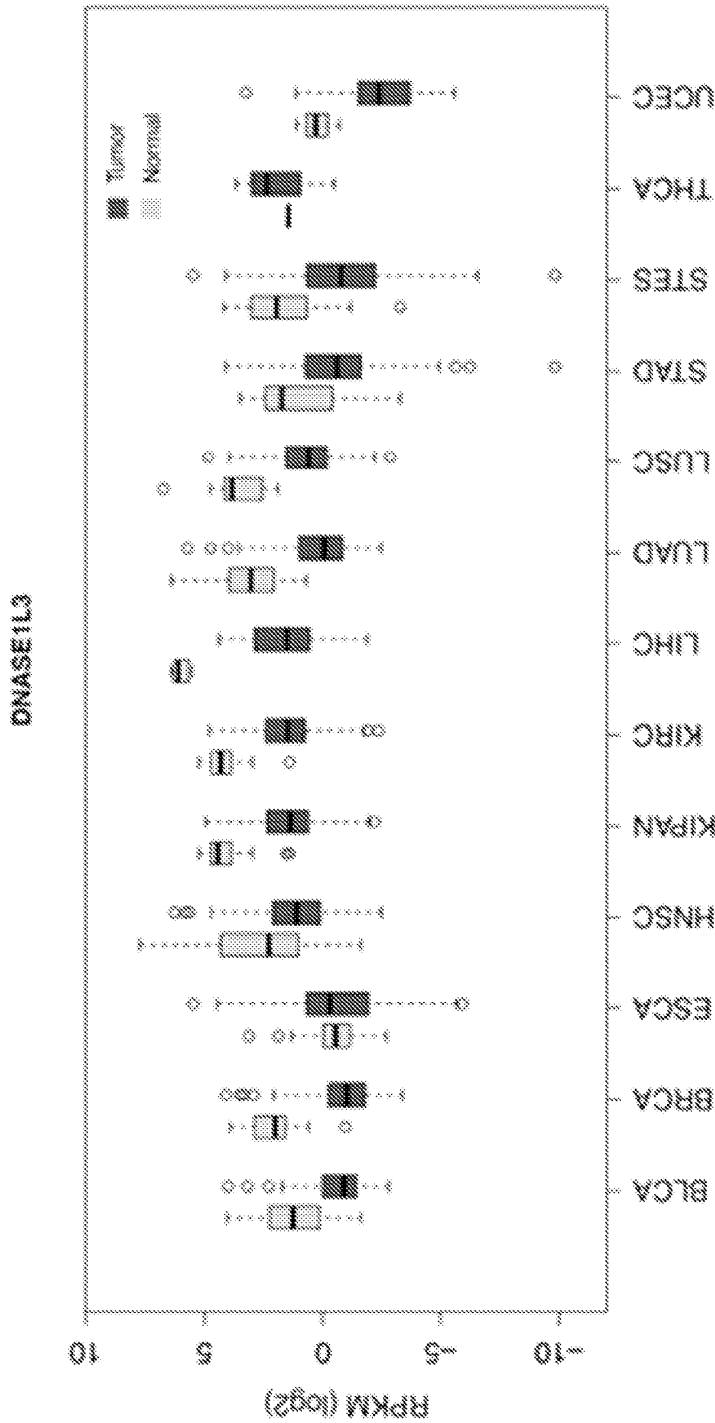
[Fig. 30]



[Fig. 31]



[Fig. 32]



[Fig. 33]

