

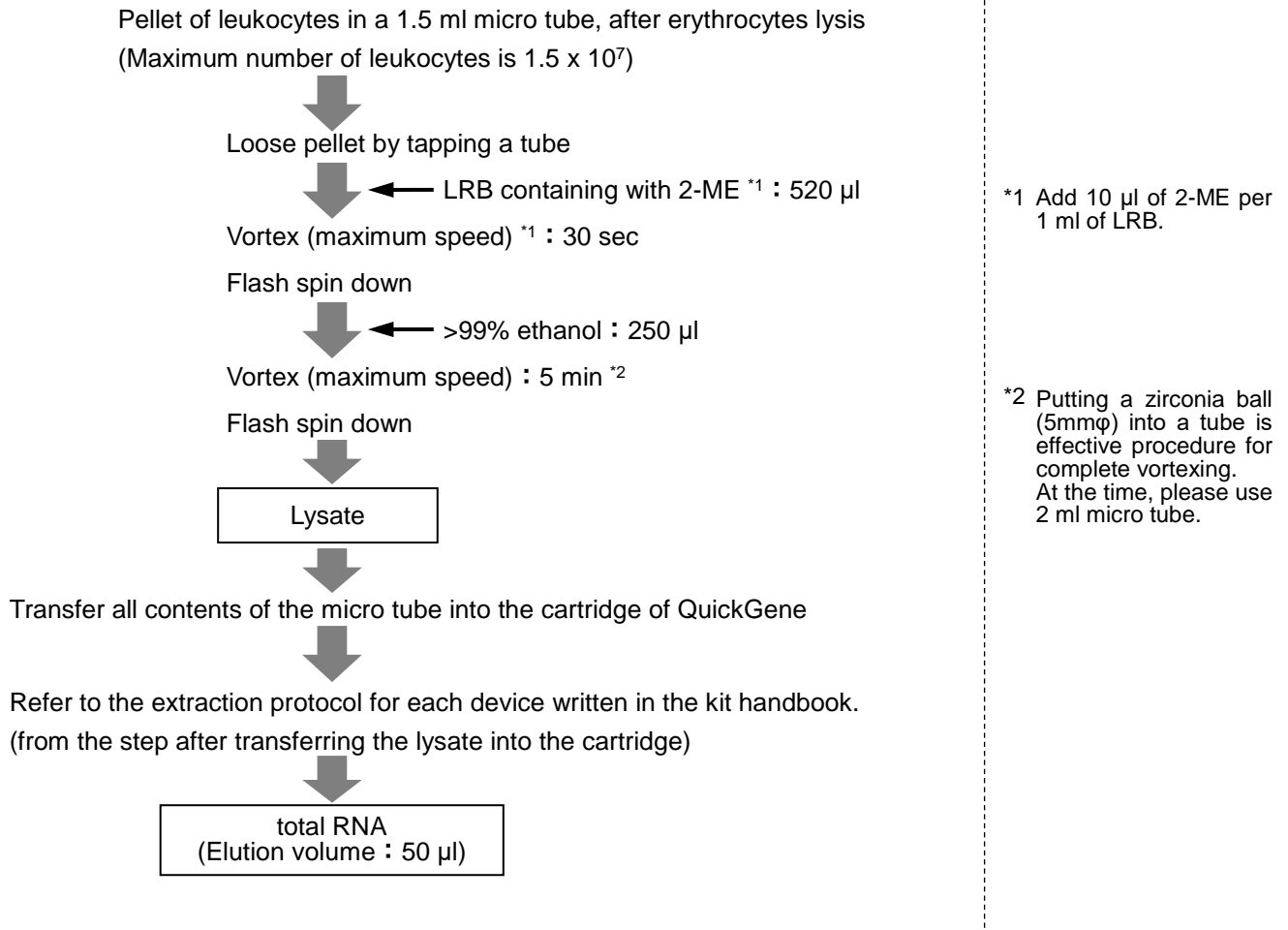


11. Total RNA Extraction from Blood of Animal

RA-a-1

Total RNA Extraction from Leukocyte

Protocol



Results

The yield of total RNA / Protein contamination : A260/280

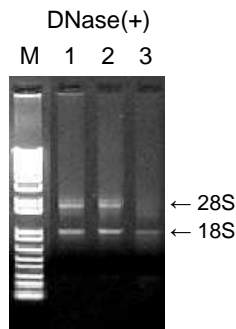
	Number of leukocytes (cells)	QuickGene		Spin column method (company A) *1		Automatic magnetic bead method *2	
		(μ g)	A260/280	(μ g)	A260/280	(μ g)	A260/280
With DNase treatment	2×10^6	0.6	2.20	0.4	2.04	0.7	2.46
	1×10^7	4.5	2.21	3.8	2.09	-	-
	1.5×10^7	6.5	2.10	-	-	-	-
Without DNase treatment	1.0×10^7	5.0	2.17	4.2	2.10	-	-

*1 : For spin column method, maximum number of leukocytes is 1×10^7 .

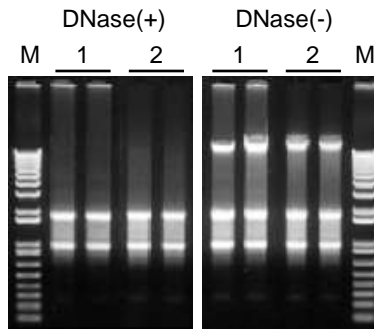
*2 : For automatic magnetic bead method, maximum number of leukocytes is 2×10^6 .

Electrophoresis of total RNA

Number of leukocytes : 2×10^6



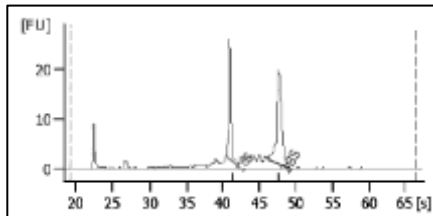
Number of leukocytes : 1×10^7



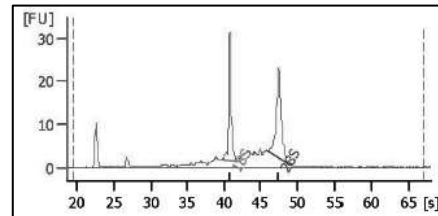
Electrophoresis condition : 1% Agarose / 1 x TAE
M : Marker (1Kb Plus DNA Ladder : Invitrogen)
1 : QuickGene
2 : Spin column method (A company)
3 : Automatic magnetic bead method

The quality of total RNA (with DNase treatment)

QuickGene (Number of leukocytes : 1×10^7)



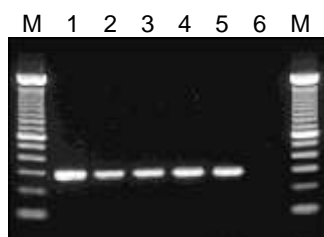
Spin column method (company A) (Number of leukocytes : 1×10^7)



	Number of leukocytes	QuickGene	Spin column method (company A) ^{*1}	Automatic magnetic bead method ^{*2}
RIN	2×10^6	7.7	6.5	5.0
	1×10^7	9.2	8.8	-
28S / 18S	2×10^6	1.5	0.8	0.0
	1.0×10^7	1.6	1.2	-

Other

RT-PCR



M : Marker (100bp DNA Ladder : Invitrogen)
1 : Positive control
2,3 : QuickGene
4,5 : Spin column method (A company)
6 : Negative control

Real Time PCR

Number of copied *GAPDH* per $1\mu\text{g}$ of total RNA (For isolation from 1×10^7 leukocytes)

QuickGene	3.15×10^7
Spin column method (company A)	1.11×10^7

Used model : Real Time PCR system Roche LightCycler
Used reagents : LightCycler FastStart DNA Master SYBR Green I
LightCycler Human GAPDH Primer Set

Common protocol is usable for the following

No Data

Depending on sample and storage conditions, nucleic acid may not be extractable.
Therefore, we cannot guarantee accurate data.
The extracted nucleic acid contains unintended acid (ex: when extracting DNA, RNA is also extracted).

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