

Symptom	Possible cause	Solution
Extraction cannot be performed properly. Or yield is extremely low.	Inadequate preparation of extraction reagent	Please confirm the protocol of the extraction kit in use. Please confirm that the reagent in use is not precipitated. If the reagent requires addition of ethanol, etc. before use, confirm that the required amount of ethanol has been added.
	Ingredients derived from the sample are affecting extraction performance.	Ingredients of the culture solution may be affecting extraction efficiency. Consider centrifuging the cell suspension and replacing the medium with PBS.
When nucleic acid was extracted from a sample prepared by adding mycoplasma strains to a solution that does not contain any cells such as medium or saline, no nucleic acid was obtained.	The amount of nucleic acid in the sample is too small.	The amount of nucleic acid contained in the sample may be smaller than the minimum amount the extraction kit can extract. The DNeasy Blood&Tissue Kit that we recommend encourages use of carrier DNA when the amount of nucleic acid obtained is estimated to be 10ng or less.
Inner control group yields negative result.	Extraction reagent is remaining.	(1) If extraction reagent is remaining in the column and mixed with eluted nucleic acid solution, it may hinder PCR reaction. Confirm that no reagent is remaining in the column before performing elution, and if there is any remaining reagent, remove them by centrifugation or pipetting.
	Excessive amount of sample genome	(2) When an excessive amount of genomes is brought into PCR, PCR reaction may be hindered. Reduce the amount of genome and perform PCR again.
	Fluorescence detection setting is not optimized.	(3) Please reconfirm the PCR program and fluorescence detection settings. When the Y-axis scale of the amplification curve shown in the analysis display is too large, amplification curve near the lower end of the detection limit can be invisible. If this is the case, adjust the scale properly.