DIALAT Ltd.

Data sheet
This data sheet provided for information only

2X Mas^{PLUS}Mix-2025

RESEARCH USE ONLY

Ready-to-use MasterMix for "anti-contamination" amplification

Cat.No.	Pack	Conc.
Dia-07120	**100 rnx**	**2 X**
Dia-07121	**500 rnx**	**2 X**

Stability:
2X Mas^{Plus}Mix stable for 24 months at -20°C, **or** for 12 months at +4°C storage without freezing.

CONTENT:
1X: SmarTaq Polymerase
UDG (Uracil DNA Glycosilase)
0.2mM dATP,dCTP,dGTP
0.1mM dTTP,dUTP
1,5 mM $MgCL_2$
Reaction Buffer components
Stabilizer/enhancer

DESCRIPTION
2X Mas^{Plus}Mix is a ready-to-use premix of all components for amplification of target DNA, contains **stabilizer/ enhancer,** which improves thermostabilization of enzyme during PCR amplification and storage**.**
2X Mas^{Plus} contains **dUTP, UDG** and **antibodies-blocked polymerase**, which are not active at ambient temperature (during PCR set-up) and activated automatically during the first PCR cycle at the temperature >70°C, preventing miss-priming and other artifacts formation. In addition,
2X Mas^{Plus}Mix prevents formation of false-positive amplicons, due to pre-amplification treatment of reaction mixture by UDG, what allows distracting possible unwanted contaminations.
It is no need for prolonged heating for activation of enzyme for PCR.
2X Mas^{Plus}Mix contains optimized buffer reagents, which greatly improve specificity of PCR with **complex**, **low-copy number DNA templates, multiplex PCR, "real-time" PCR, allowing** using very small initial quantities of DNA template.
One' can use an appropriate volume of **2X Mas^{Plus}Mix** for amplification reaction, depending on total final reaction volume.
Just place it into the tube/plate adds primers and template of choice mix all components and run PCR.
After PCR reaction running mix 5-10μl of reaction mixture with appropriative volume of "Loading Buffer" (for non-"real-time" mode PCR), apply to the gel and run electrophoresis.

Recommended PCR assay

50μl PCR assay		Final Conc.
25μl	**2X Mas^{Plus} -2025**	1X
0.2-1μM	each Primer	
Variable*	DNA Template	
To 50μl	PCR Grade Water	

*- depending on DNA template initial concentration

APPLICATIONS:
- "ani-contamination" PCR
- Primer extension
- Real-Time PCR (all types)
- Low-copy PCR (SmarTaq Polymerase)
- Multiplex PCR

SHIPPING CONDITIONS:
Should be shipped at ambient temperature. For long distance shipments preferably in **Blue Ice**

STORAGE CONDITIONS :
Store **2X Mas^{Plus}Mix** at -20°C (for long-term storage).

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General Protocol for amplification with 2XMas^{Plus}Mix-2025

Add and mix the following components:

Component	50μL reactions	25μL reactions	Final concentration
PCR grade Water	Up to 50 μL	Up to 25 μL	
2X Mas^{Plus} -2025	25 μL	12.5 μL	1X
Primers			0.3-0.5 μM each
Template DNA	optionally	optionally	1-50ng

In some cases, we recommends to optimize Mg concentration in the range 2.0-3.0mM
We recommend using 25μl reaction for the PCR with **2X Mas^{Plus}**

Cycling Protocol:

Cycle step	3-step amplification		Cycles
	T°C	Time	
Pre-amplification treatment	**47°C**	**2 min**	**1**
Initial Denaturation	**95°C**	**1-3 min**	**1**
Denaturation **Annealing** **Extension**	**95°C** **55-66*** **72°C**	**10 S** **5-10 S** **15-30 Sec/Kb****	**25-35**
Final extension	**72°C** **4°C**	**1-2 min** **hold**	**1**

Pre-amplification treatment needed for destruction of possible contamination (Uracil – containing amplicons) from the previous amplification by Uracil DNA-Glycosilase.
NOTE: If you used in the other amplification dNTP's not-containing dUTP (only regular dNTP's) UDG treatment is senselessly.

*Optimal Tm for the primer pair recommended as Tm of the lower primer, for the standard oligos <20nt.
To optimize amplification we recommend using gradient PCR amplification, to rich final amplification conditions in the short time.
**For non-complex DNA templates (plasmid DNA, phage DNA, BAC clone) extension time could be reduced up to 15 sec/Kb.
For complex DNA, templates (human DNA) strongly recommended to apply Extension time as 30 sec/Kb for the targets more than 1,5Kb

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