Extra Buffer (15 mM $MgCl_2$)

Conc.: 10x Buffer

Cat. No.: 733-2303

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-	10x Extra Buffer 15 mM $MgCl_2$
ID No. Cap colour	5100510 Blue
733-2303	3 x 1.5 ml

Store at –20 °C

General Description

VWR offers 3 different buffers to allow the customer to choose the optimal buffer system for a specific amplification process. VWR Buffers are usually supplied in 10x formulations with 15 mM $MgCl_2$ included but are also available as Mg^{2+} free buffers, detergent free buffers as well as Mg^{2+} and detergent free buffers.

Key Buffer

Composition: Tris-HCl pH 8.5, $(NH_4)_2S0_4$, 15 mM $MgCl_2$*, 1% Tween® 20*. Buffers including ammonium (NH_4^+) usually give a superior amplification signal (high yield) in many primer-template systems (figure 1). Ammonium in the buffer minimizes the need for optimization of the $MgCl_2$ concentration or the annealing temperature for most primer-template systems.

Extra Buffer

Composition: Tris-HCl pH 8.5, KCl, 15 mM $MgCl_2$*, 1% Triton X-100*. Extra Buffer is the traditional potassium (K^+) buffer. Extra Buffer promotes high specificity (figure 1) and careful optimization of primer annealing temperatures may be required.

Combination Buffer

Composition: Tris-HCl pH 8.7, KCl, $(NH_4)_2SO_4$, 15 mM $MgCl_2$*, 1% Tween® 20*. Combination Buffer is a proprietary mixture of K^+ and NH_4^+. This buffer combines high specificity with good product yield and high tolerance to optimization of primer annealing temperatures and Mg^{2+} concentrations due to its balanced ammonium-potassium formulation.

Magnesium

Mg^{2+} is required for polymerase activity. Low Mg^{2+} concentrations increase the fidelity but with too low Mg^{2+} concentrations the polymerase will not work. The Mg^{2+} concentration available in the reaction is dependent on several parameters e.g. the presence of chelators or the dNTP concentration. Therefore the Mg^{2+} concentration should be optimized.

Tween, Triton

Non-ionic detergents are used to prevent the polymerase to stick to the walls of the tube, to stabilize the polymerase and increase yield. However, these agents might increase non-specific amplification or interfere with downstream reactions. Tween can be used to neutralize SDS contaminations in the DNA template.

Recommended Storage and Stability

Long term storage at -20 °C. Product expiry at -20 °C is stated on the label.
Option: Store at +4 °C for up to 6 months.

Quality Control

Each lot of buffer is functionally tested in PCR.

Kit Components

10x Extra Buffer

Tris-HCl pH 8.5, KCl, 15 mM $MgCl_2$, 1% Triton X-100.

Determining the optimal buffer system for your application

By running three reactions using the three VWR buffers, the optimal reaction conditions should be found for the specific application. The final concentration of the buffer in the reaction should be 1x.

1. 1x Key Buffer
2. 1x Extra Buffer
3. 1x Combination Buffer (not shown in the figure)

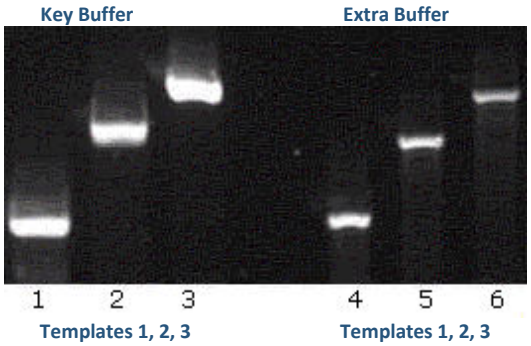

Figure 1: Amplification of three different cDNA templates using Key Buffer versus Extra Buffer.

Optimising the Mg^{2+} concentration

In some applications, optimization of $MgCl_2$ is required for best results. For this reason, 25 mM $MgCl_2$ is included in all buffer kits. Table 1 provides the volume of 25 mM $MgCl_2$ to be added to the master mix using the VWR buffer kit, if a higher $MgCl_2$ concentration is required.

Version: VWR 10/2019

Made in Denmark

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Table 1. Additional volume (µl) of $MgCl_2$ per 50 µl reaction using a 10x Buffer including 15 mM $MgCl_2$

Final $MgCl_2$ conc. in reaction (mM)	1.5	2.0	2.5	3.0	3.5	4.0	4.5
Volume of 25 mM $MgCl_2$	0	1	2	3	4	5	6

When using Mg^{2+} free buffers, the addition of $MgCl_2$ is imperative. Table 2 provides the volume of 25 mM $MgCl_2$ to be added to the master mix using Mg^{2+} free buffers.

Table 2. Additional volume (µl) of $MgCl_2$ per 50 µl reaction using a Mg^{2+} free 10x Buffer

Final $MgCl_2$ conc. in reaction (mM)	0.5	1.0	1.5	2.0	2.5	3.0	3.5	4.0	4.5
Volume of 25 mM $MgCl_2$	1	2	3	4	5	6	7	8	9

Related Products

Description	Cat. No.
VWR Taq DNA Polymerase	
VWR Glycerol Free Taq DNA Polymerase (1000 units) with 10x Key Buffer with 10x Extra Buffer	**733-1817**
VWR Taq DNA Polymerase (500 units) with 10x Key Buffer with 10x Extra Buffer	**733-1301**
VWR Taq DNA Polymerase (500 units) with 10x Extra Buffer (Mg^{2+}-free and Triton-free)	**733-1304**
VWR Taq DNA Polymerase (1000 units) with 10x Key Buffer (Tween-free) with 10x Extra Buffer (Triton-free)	**733-1307**
VWR Taq DNA Polymerase (500 units) with 10x Key Buffer (Mg^{2+}-free)	**733-1311**
VWR Red Taq DNA Polymerase (2500 units) with 10x Key Buffer with 10x Extra Buffer	**733-1323**
VWR Taq DNA Polymerase Master Mix*	
VWR Taq DNA Pol. 1.1x Master Mix, 1.5 mM $MgCl_2$ final conc. (2500 reactions)	**733-1314**
VWR Taq DNA Pol. 2x Master Mix, 1.5 mM $MgCl_2$ final conc. (2500 reactions)	**733-1316**
VWR Red Taq DNA Pol. 1.1x Master Mix, 1.5 mM $MgCl_2$ final conc. (2500 reactions)	**733-1318**
VWR Red Taq DNA Pol. 2x Master Mix, 1.5 mM $MgCl_2$ final conc. (2500 reactions)	**733-1320**
VWR TEMPase Hot Start DNA Polymerase	
VWR TEMPase Hot Start DNA Polymerase (500 units) with 10x Key Buffer with 10x Combination Buffer	**733-1331**
VWR Glycerol Free TEMPase DNA Polymerase (500 units) with 10x Key Buffer	**733-2555**
VWR TEMPase Hot Start Master Mix**	
VWR TEMPase Hot Start 2x Master Mix K, 1.5 mM $MgCl_2$ final conc. (2500 reactions)	**733-2582**
VWR Blue TEMPase Hot Start 2x Master Mix K, 1.5 mM $MgCl_2$ final conc. (2500 reactions)	**733-2585**
VWR Multiplex Master Mix	
VWR Multiplex 2x Master Mix, (2500 reactions)	**733-2569**
VWR High Fidelity DNA Polymerase	
VWR AccuPOL DNA Polymerase (250 units) with 10x Key Buffer	**733-1324**
VWR AccuPOL DNA Polymerase (250 units) with 10x Key Buffer (Mg^{2+} free and Tween free)	**733-1328**
VWR PCR accessories	
Betaine Enhancer Solution 5 M, 5 ml (500 reactions)	**733-1361**
dNTP Mix, 40 mM (10 mM of each dATP, dCTP, dGTP & dTTP) (40 µmol, 2x 500 µl)	**733-1363**
dNTP Set, 100 mM of each dATP, dCTP, dGTP & dTTP (4x 25 µmol, 4x 250 µl)	**733-1364**
VWR PCR grade water (6 x 5 ml)	**733-2573**
VWR Loading Buffer Red (5 x 1 ml) – available in 4 different colors	**733-2574**

*Also available with 2 mM $MgCl_2$
**Also available as 2x Master Mix C based on Combination Buffer

Tween® 20 is a registered trademark of ICI Americas, Inc.

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