Universal PCR Primers for Bacteria (for other domains or specific groups, check specific literature):

Primer	5´-3	E coli location	Tm ° C	Ref.	Length
8F	AGAGTTTGATCCTGGCTCAG	8	55	Zhou et al 1997	1533 nuc
1541R	AAGGAGGTGATCCAGCCGCA	1541	55		
27F	AGAGTTTGATCCTGGCTCAG	27	50	Lane et al 1991	1464 nuc
1492R	GGTTACCTTGTTACGACTT	1492	50		

PCR reaction for standard screenings with the Promega GoTaq kit

(comment, if you use other kits other conditions may apply):

Components	Amount (µl) – for a 25 µl PCR reaction volume
5x Buffer with 20 mM Magnesium For standard applications: Promega Green GoTaq reaction buffer + Mg_2^+ M791A	5
2,5 mM Nucleotide mix For standard applications: Promega U151B	0,5
Taq For standard applications: Polymerase Promega M830B	0,125
Primer F (Forward) 100 pmol	0,113
Primer R (Reverse)100 pmol	0,113
DNA … ng/µl	1 (or more)
MQ total volume **25 µl**	18,15

Comment: The PCR reaction can be scaled up to 50 µl, and components can be exchanged (e.g. PCR buffer and Taq polymerase can be exchanged to high fidelity polymerases based on e.g. Pfu or Phusion-Finnzymes.
Advice: When preparing a master mix for several reactions, add one extra sample just in case.

Standard PCR Programme:

Step	Temperature (° C)	Time (sec)	
Lid	95	∞	
Pre-Denaturation	95	120	
Denaturation	95	40	
Annealing	55	40	Cycles: 35
Elongation	72	90	
Final elongation	72	600	
Cooling	4	∞	

Where appropriate, change time, temperature, cycles.