PSS2 PCR Protocol

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Abstract

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Guidelines

Primers:

attP_F502: CTC CTC AAC TGC CAG GAC TC (PSS2 target 502bp upstream of att: 90315-90334) attP_R141: CCC AACAAG CTC TCA GGA AG (PSS2 target 141bp downstream of att rev:

91010-91029)

Reaction Mix

Reagent Amt. Final Conc.

Water 18.25µl 10x Buffer (no Mg) 2.5μ l =1x25mM MgCl₂ 1.5µl =1.5 mMdNTP mix, 10mM each 0.5µl $=200 \mu M$ ea. Primer F502, 10µm 0.5μ l $=0.2 \mu M$ Primer R141, 10mM 0.5μ l $=0.2 \mu M$ $0.25\mu l = 0.05U/\mu l$ Sigma Tag 5U/µl

DNA (viral lysates) 1.0μl Total 25μl

Cycling

95°C 3 min

95°C 15sec

60°C 30sec 35 cycles

72°C 30sec

72°C 10min

15°C Hold

Protocol