Kazu Buffer Feedback Form

Name: Kim Maxson-Stein

Lab: (Brutnell lab)

Material: Setaria viridis

Primer set: Multiplex of SvF&R with HptsmallF&R

Amplification size (kb): ~225bp and 550bp

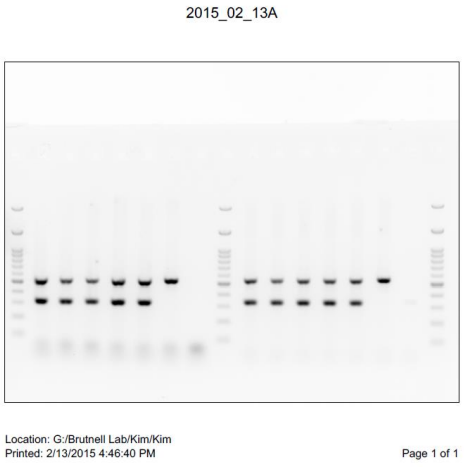
PCR taq: Go taq mix (2X) (from Kazu)

PCR amplification:

Reagent	Volume
Kazu DNA extract	2.5 µl
GoTaq Mix (2X)	7.5 µl
Forward primer (10uM)	2.5 µl
Reverse primer (10uM)	2.5 µl
	µl
	µl
H_2O	µl
Total volume	15 µl

Step	Temperature	Time	Cycles
Initial Denaturation	95℃	5 min	N/A
Denaturation	95℃	0.5min	
Annealing	58℃	0.5min	
Extension	72℃	1min	35
Final Extension	72℃	10min	N/A
Hold	12℃	Infin. min	N/A

PCR Gel Picture: (100bp ladders in lanes 1, 9, 17; 5 transgenic Setaria tillers, A10.1, and water check amplified with Kazu buffer in lanes 2-8, and amplified with Extract-N-Amp in lanes 10-16)



Would you prefer using Kazu Buffer over your current lab DNA extraction protocol? ☒ **Yes** ☐ **No**

Why or why not?

For applications like PCR genotyping and TaqMan copy number/zygosity screening, it is much quicker than a CTAB extraction. The downside is it cannot be used for Southern, but with the TaqMan assay I don't really need Southerns.

Other Comments:

I compared Kazu Buffer with Sigma Extract-N-Amp. This is the "quick" method of choice that the BTI transformation lab uses to confirm transgenic Setaria. Kazu buffer was just as effective at the genotyping PCR, and it was better at predicting copy number in the TaqMan assay, than Extract-N-Amp (see previous communication). There are a few extra steps in the Kazu buffer method, however if the price is competitive with Extract-N-Amp, this would not be prohibitive for me.