## 129mmHuTg gradient 17.04.2015 (129Hg\_gradient\_17042015)

A gradient from 50°C-65°C annealing temperature was using a Primer Mix containing 20 of transgene primers and 10ul of actin in a final volume of 1ml. For each primer mix one master mix was prepared. 20ul of mm including DNA was pipetted in each well

	1x ul	14x	
Primers mix	8	112	ul
HotStar Taq	10	140	ul
DNA	2	28	ul

Thermocycler s 94°C 94°C 50-65°C 72°C	ettings 5min 30sec 1min 1min	x 25	Expected band size (bp)	description
72°C	10min		500	500 actin
10°C	for ever		750	)750 tg

## 129mmHuTg with actin

	50°C	50.5°C	51.4°C	53°C	54.8°C	56,6°C	58.4°C	60.2°C	62°C	63.6°C	64.5°C	65°C
2000 -												
600 -												
400 -												

60°C was chosen as optimal annealing temperature, but amount of actin was decreased to 5ul for further tests

## 129mmHuTg

Expected band size				add/remove expected band size		
		basepair	description			
		500.0	500 actin			
		750.0	750 tg			
Primer(s)				add/remove Primers		
		Primer	ID	Working Stock Concentration (µM)	µl for 1ml Stock	Sequence
	available	129m hutg-fwd	653	100µM	20.0µl	CAGTCATTATGGCGAACCTT
	available	129m hutg-rev	654	100µM	20.0µl	GACCTTCCTCATCCCACT
	available	Actin fwd	114	100µM	5.0µl	TGT TAC CAA CTG GGA CGA CA
	available	Actin rev	115	100µM	5.0µl	GAC ATG CAA GGA GTG CAA GA