

## Recombinant Leap-In transposon integration

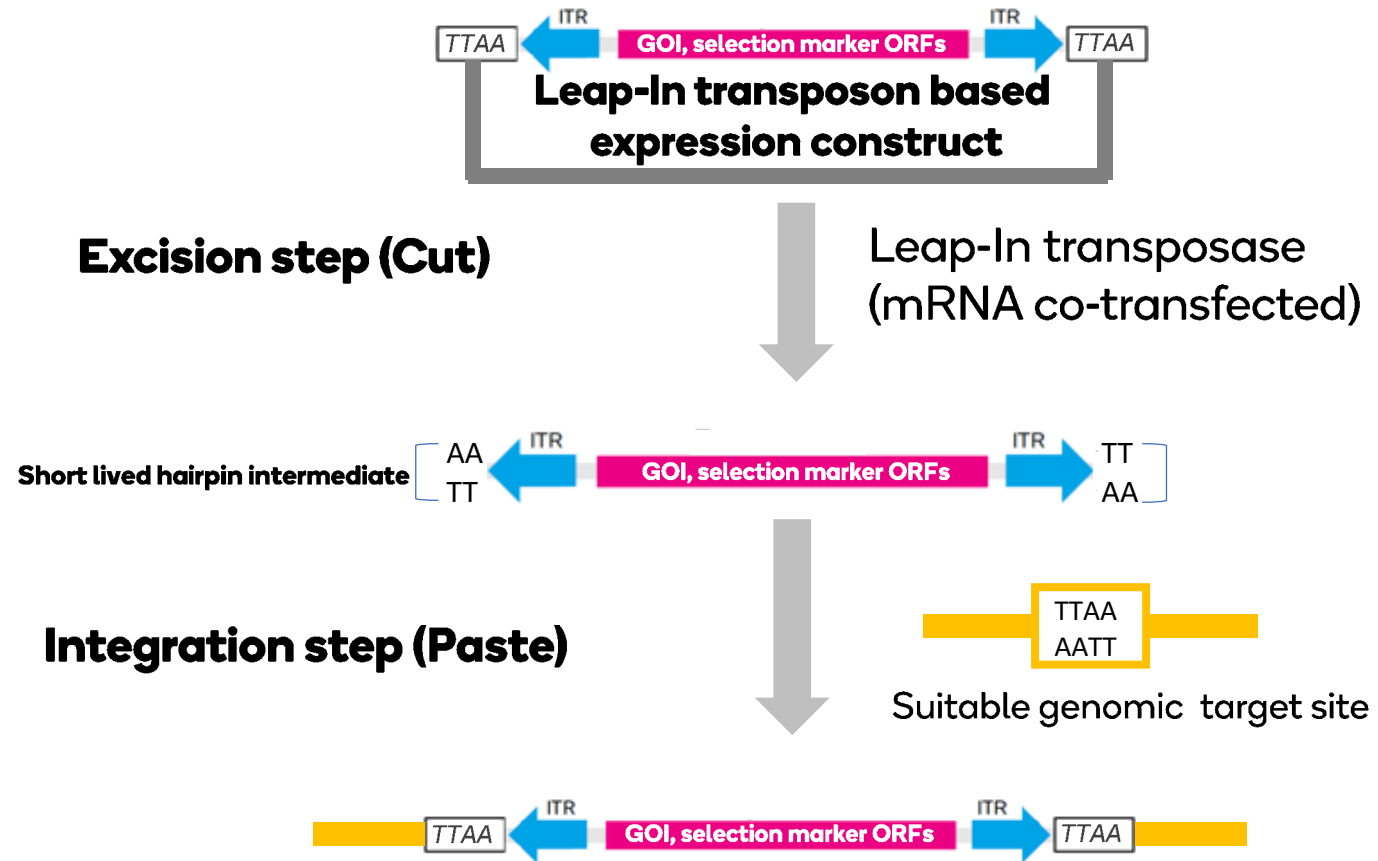


FIGURE 1

# Integration site 1

CHO genome

Transgene

CHO genome

...ACTGCCA TTAA CCTTTTT... *[expression construct]* ...AAAAGGG TTAA AAGACAA...

gi | 351516558 | ref | NW\_003614991.1 | :386716(-)

gi | 351516558 | ref | NW\_003614991.1 | : 386713(+)

# Integration site 2

CHO genome

Transgene

CHO genome

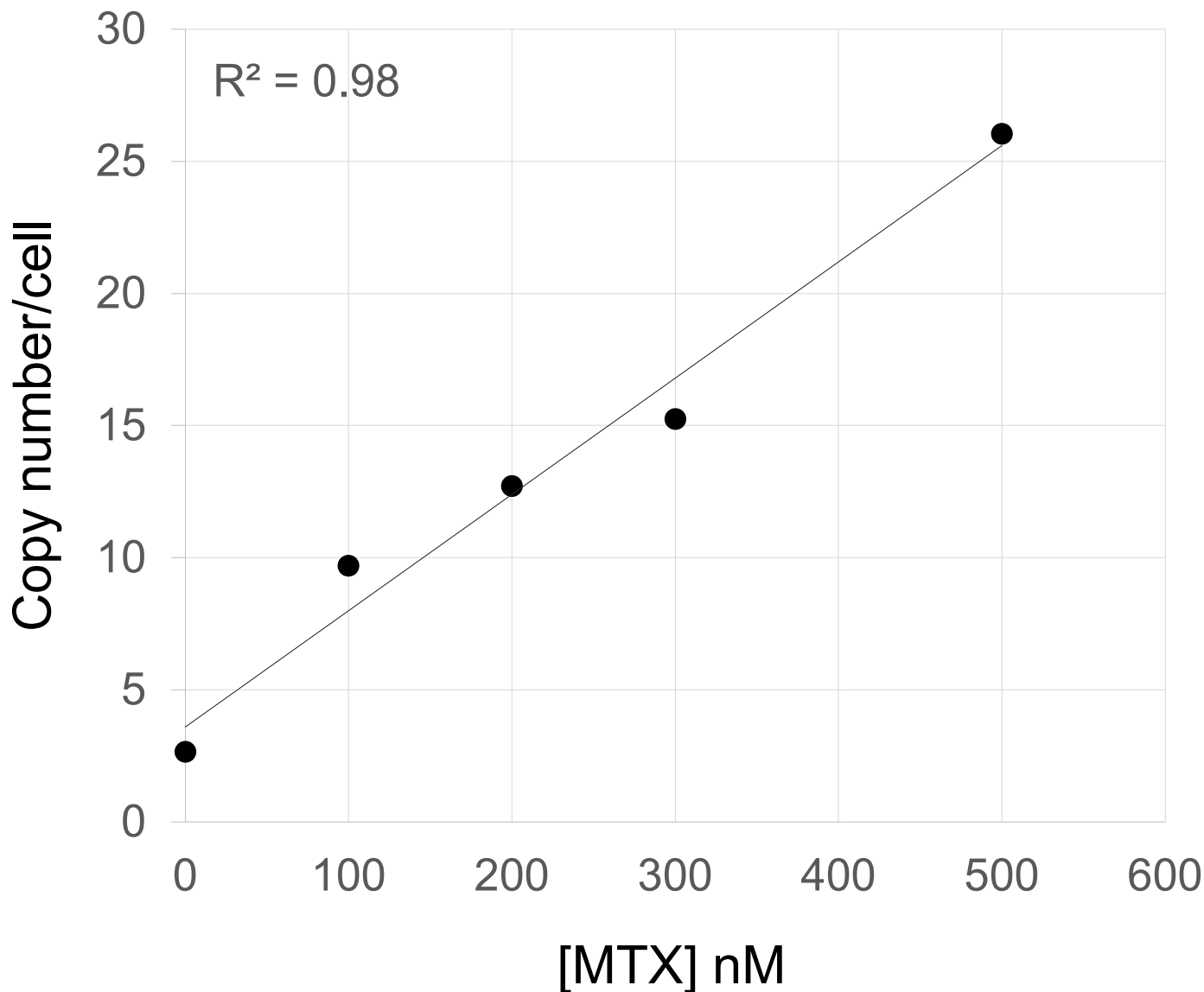
...AGGGTGT TTAA CCTTTTT... *[expression construct]* ...AAAAGGG TTAA TGTTCTG...

gi | 351517626 | ref | NW\_003613923.1 | :540111(-)

gi | 351517626 | ref | NW\_003613923.1 | : 540108(+)

FIGURE 2

# MTX dose effect on copy#



**FIGURE 3**

# Copy number dependent productivity

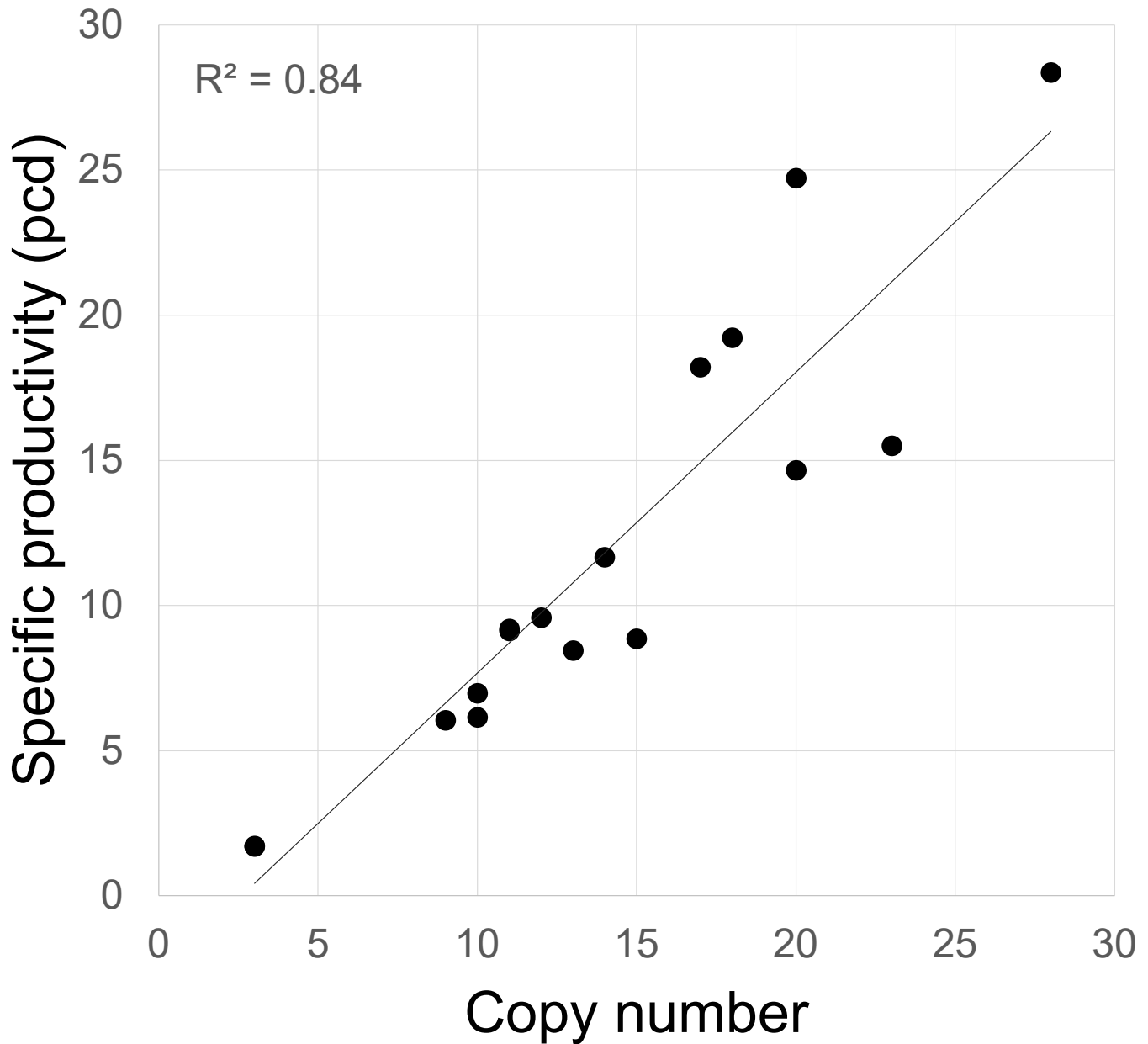
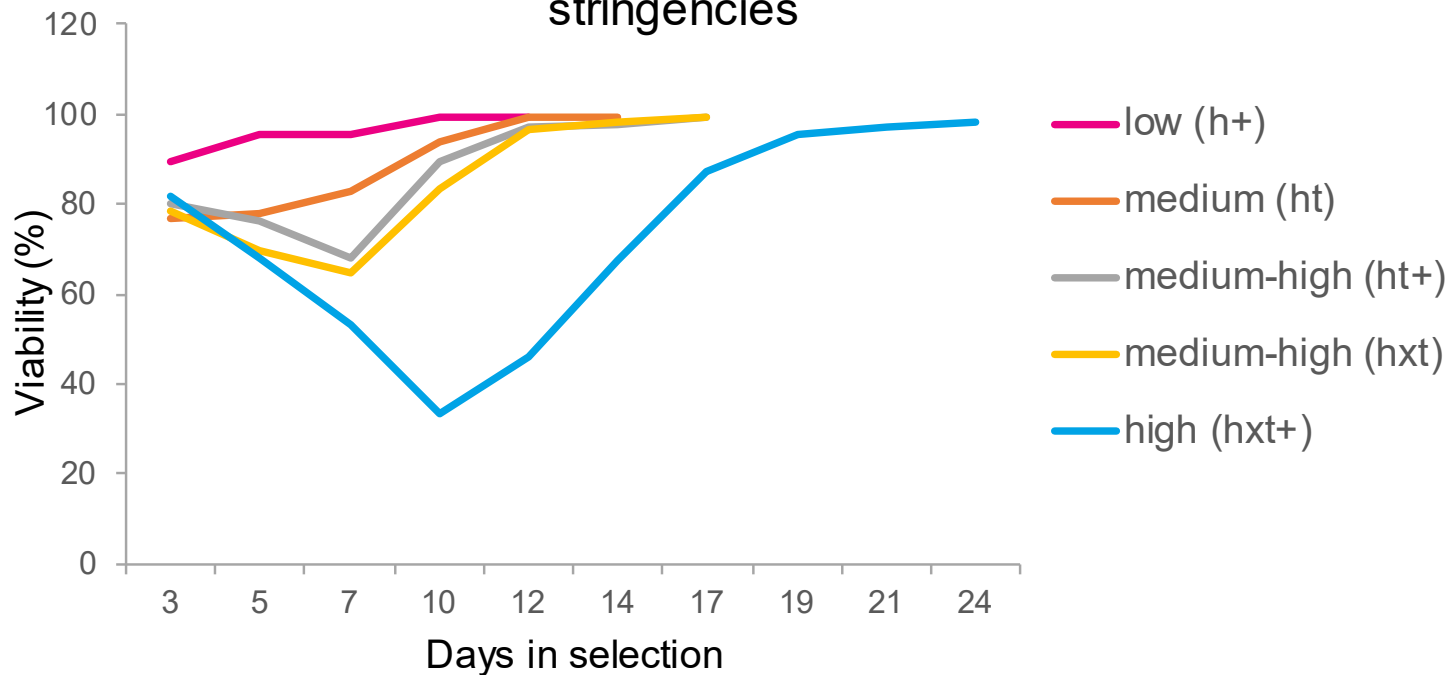


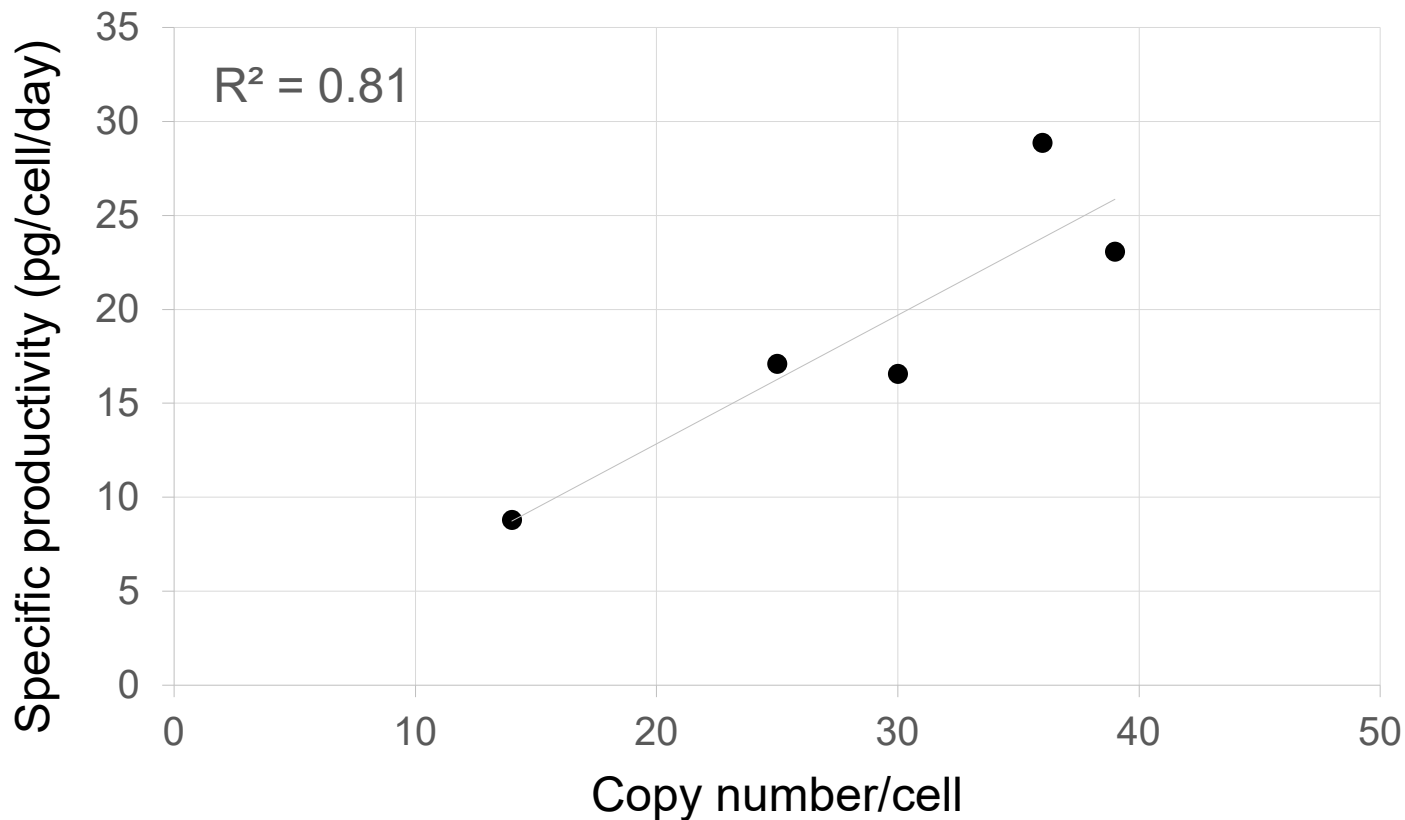
FIGURE 4

## Recovery of Leap-In pools at different selection stringencies



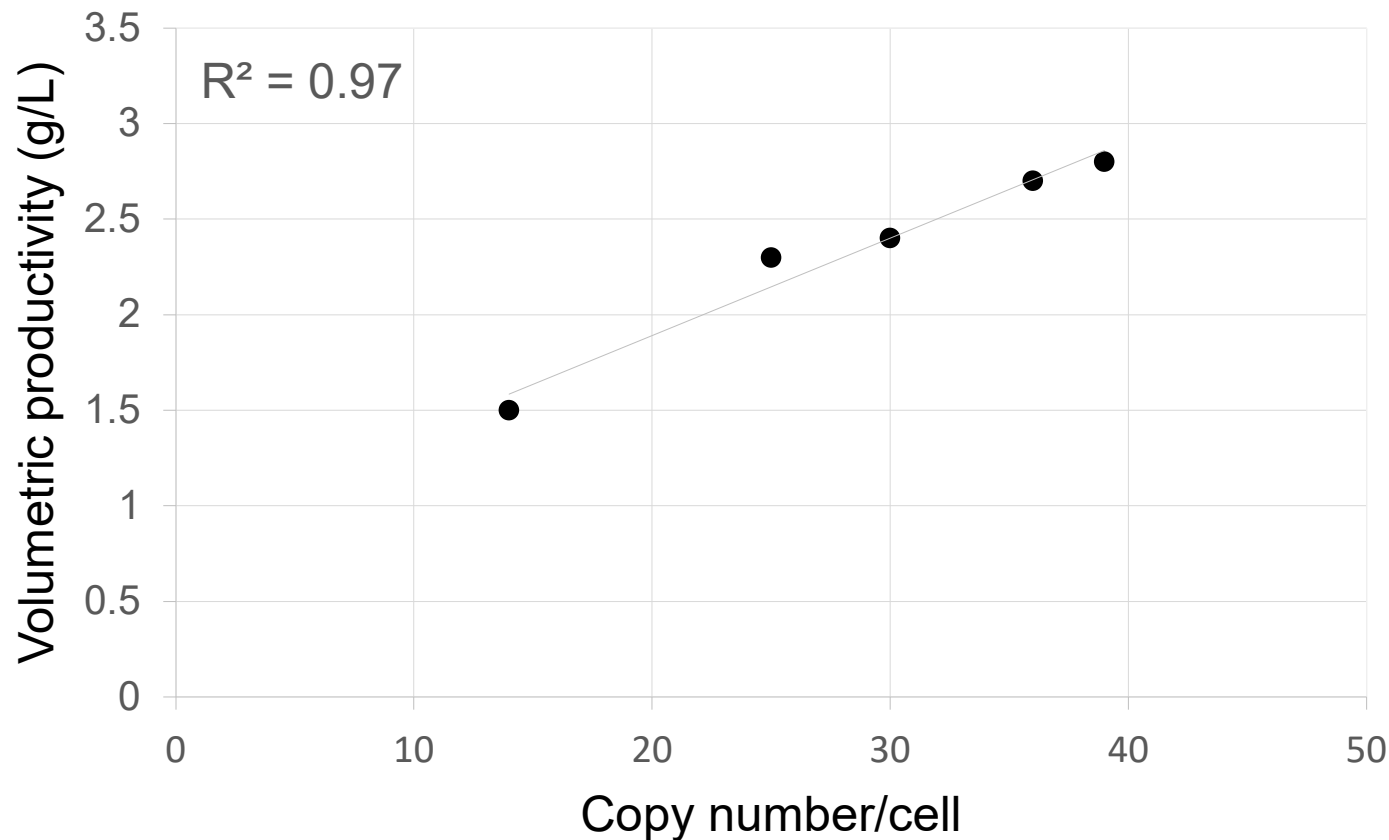
**FIGURE 5A**

## Copy number vs. Specific productivity



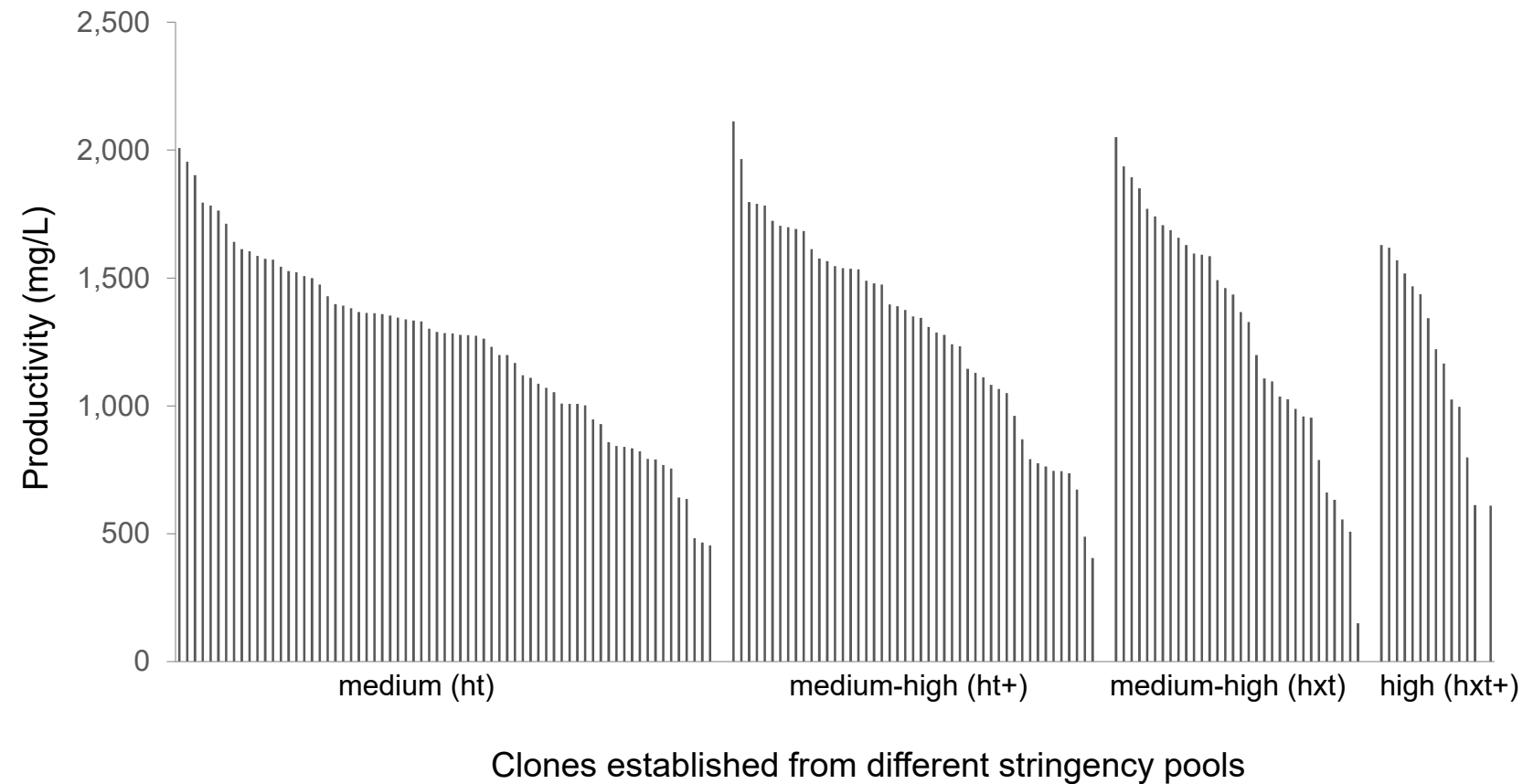
**FIGURE 5B**

# Copy number vs. Volumetric productivity



**FIGURE 5C**

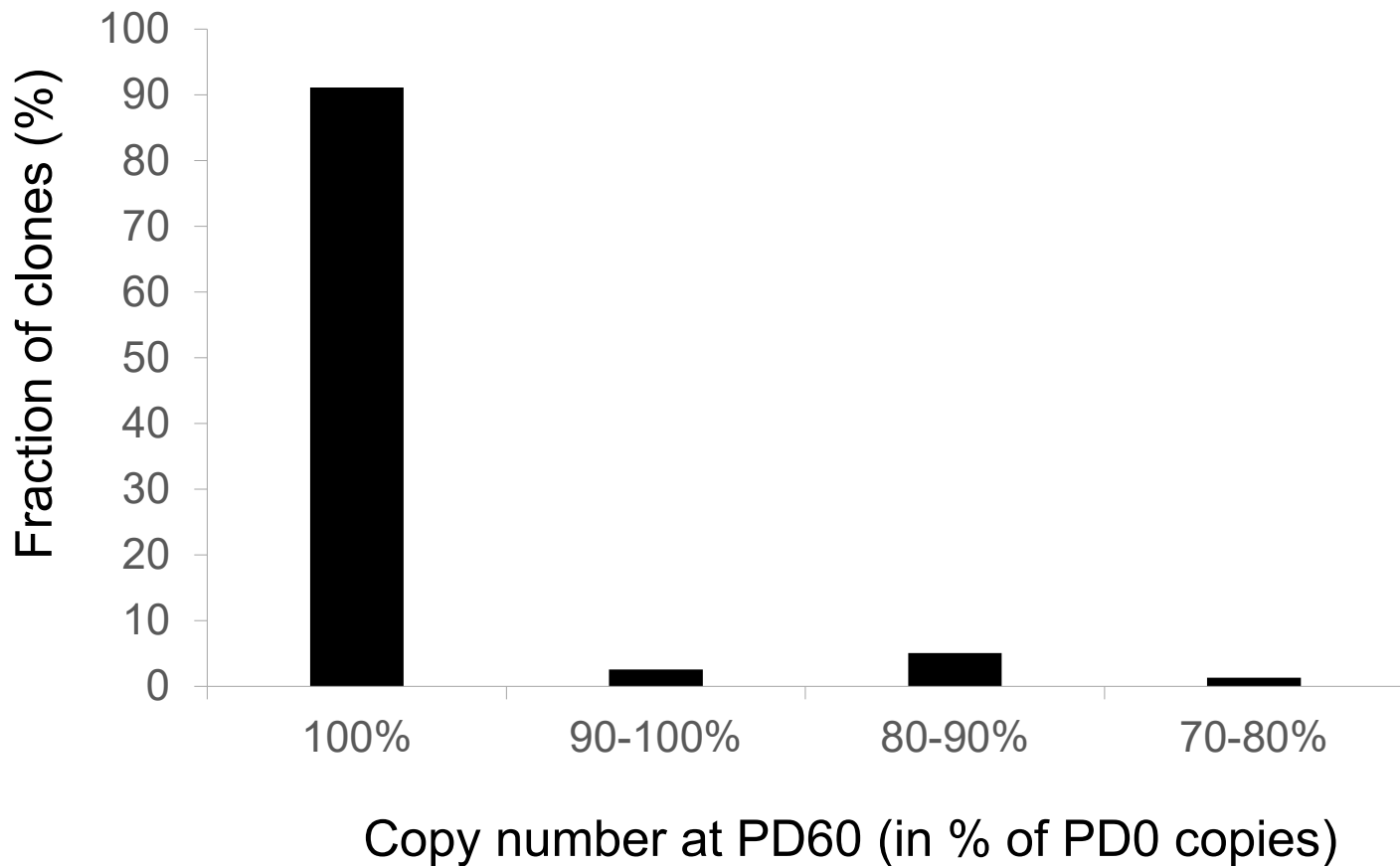
# Clonal volumetric productivity distributions in 7-day fed batch cultures



**FIGURE 6**

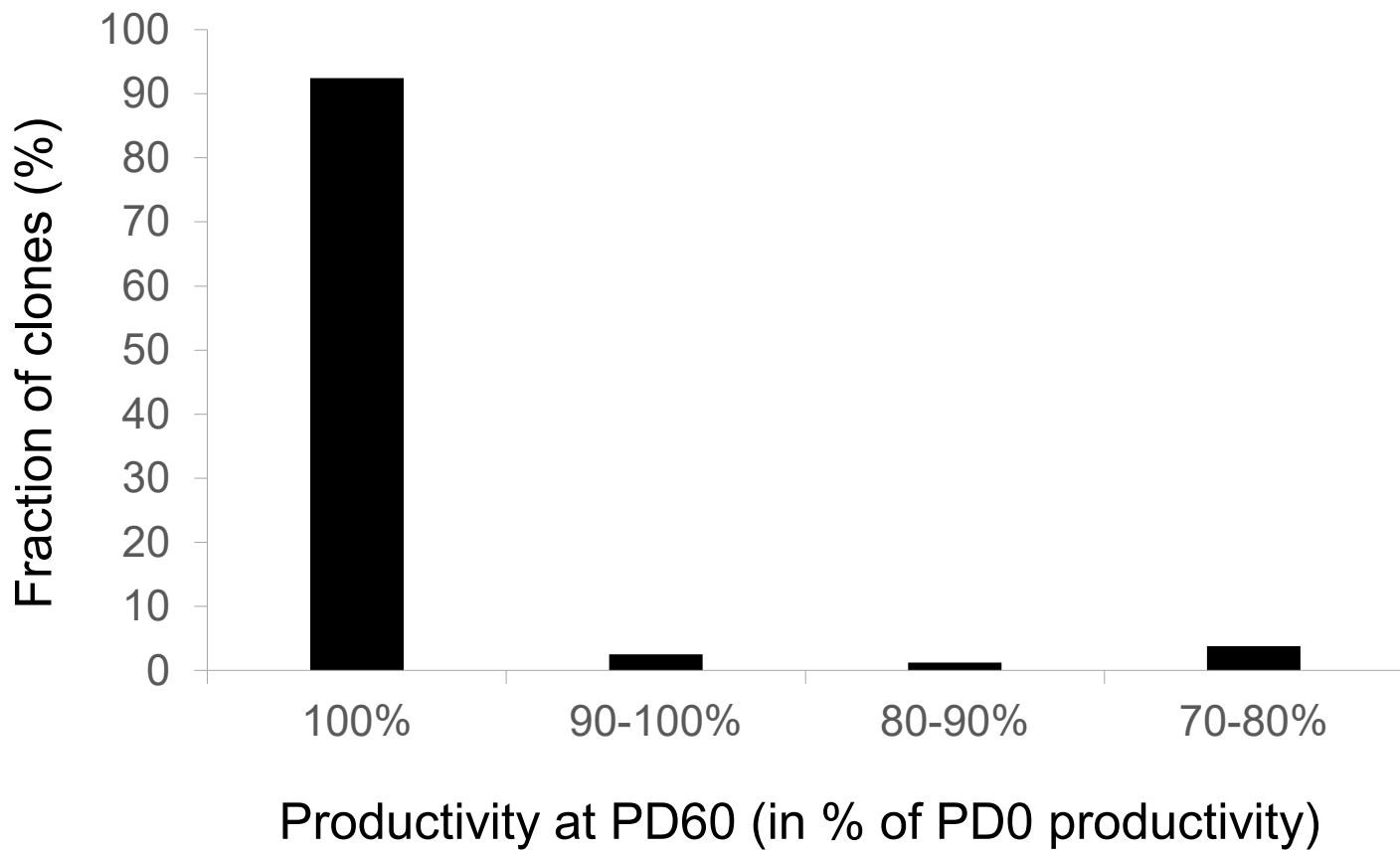


# Copy number stability

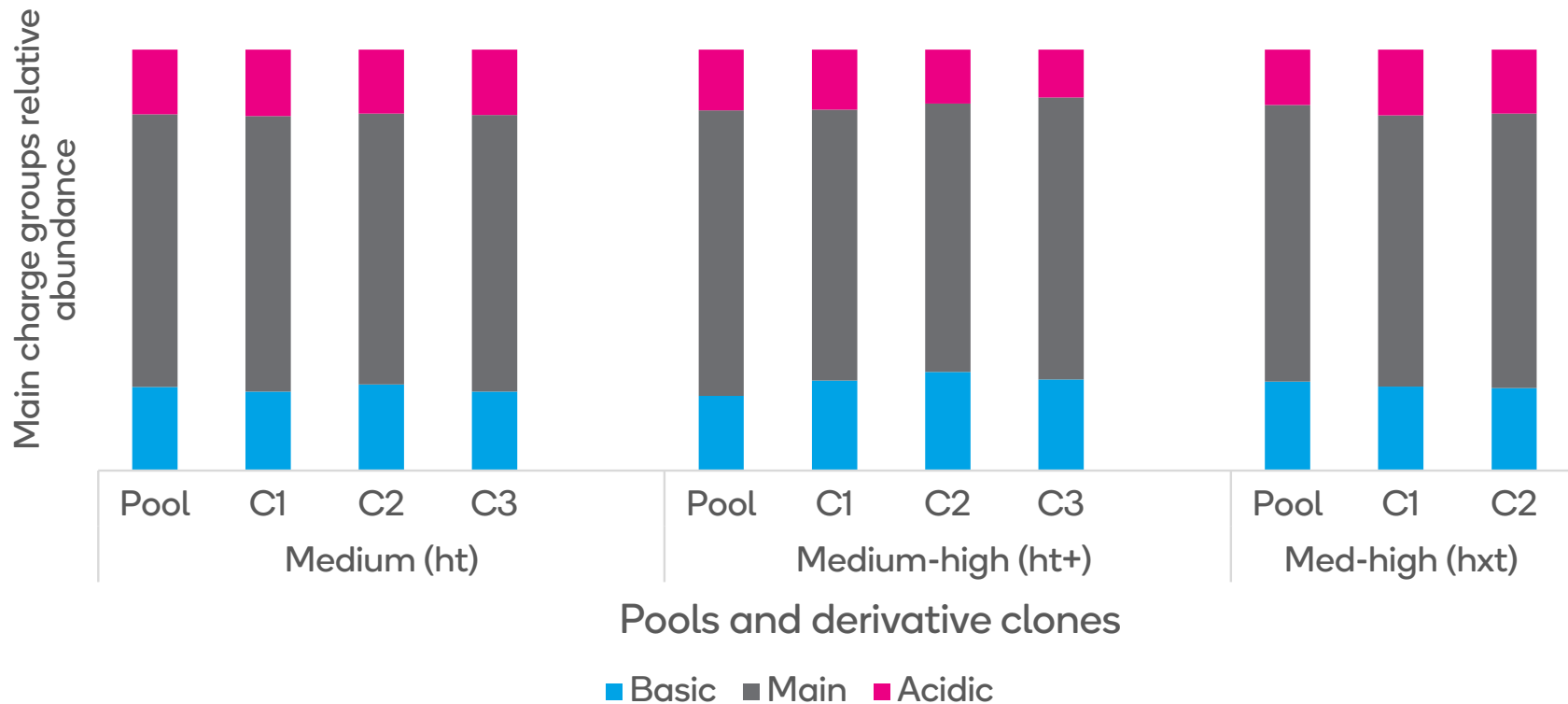


**FIGURE 7A**

# Expression stability

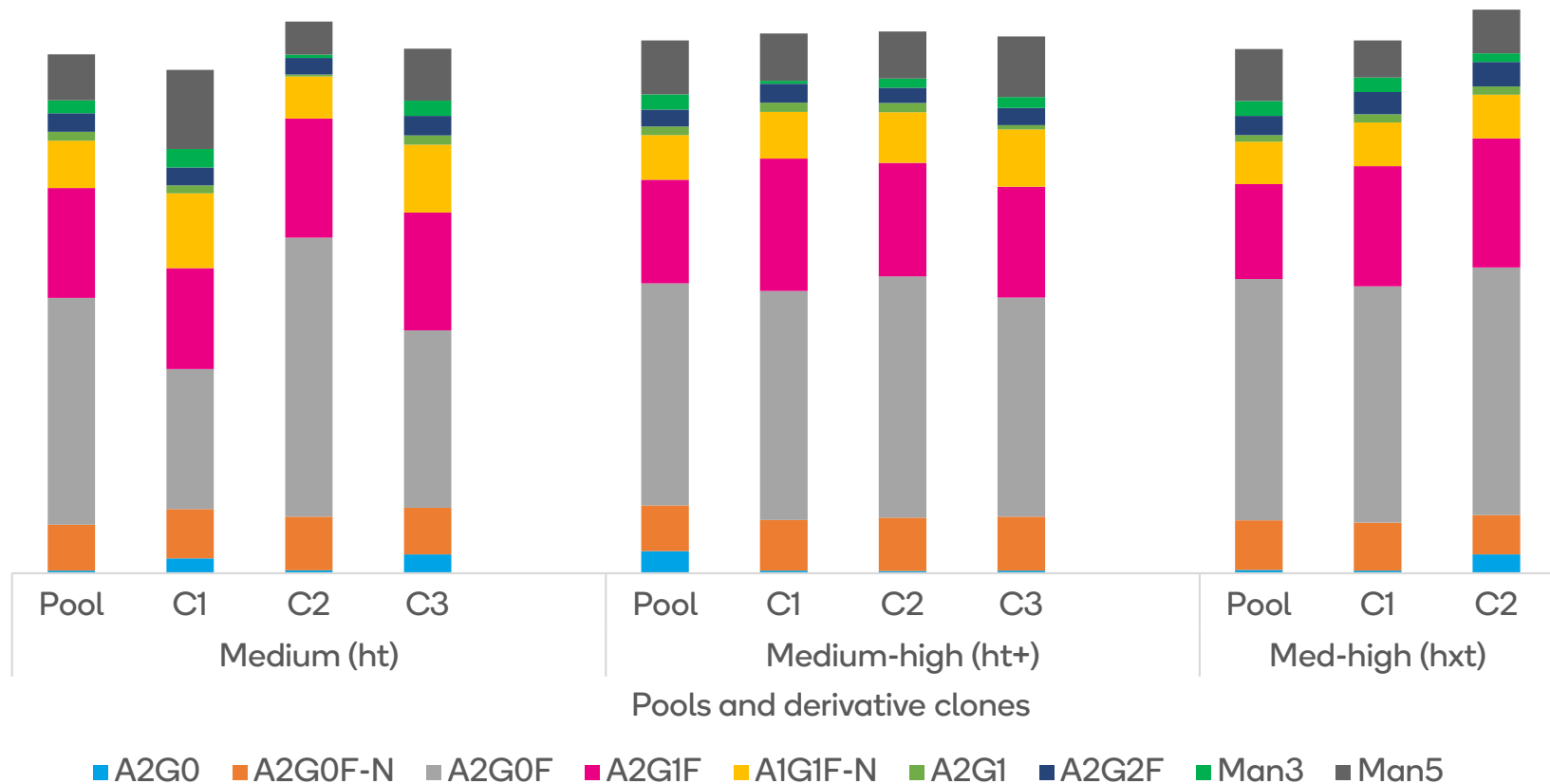


**FIGURE 7B**

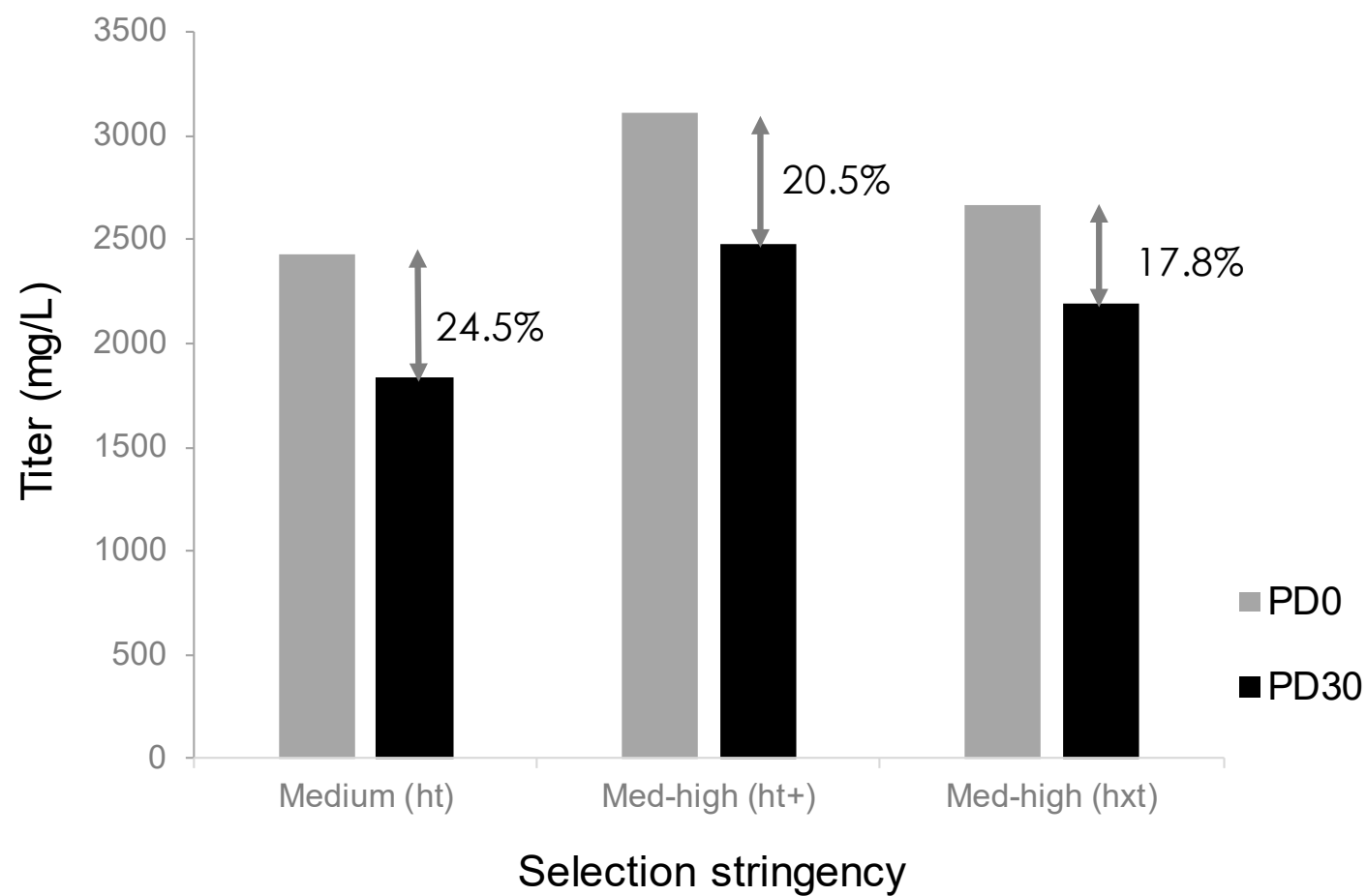


**FIGURE 8A**

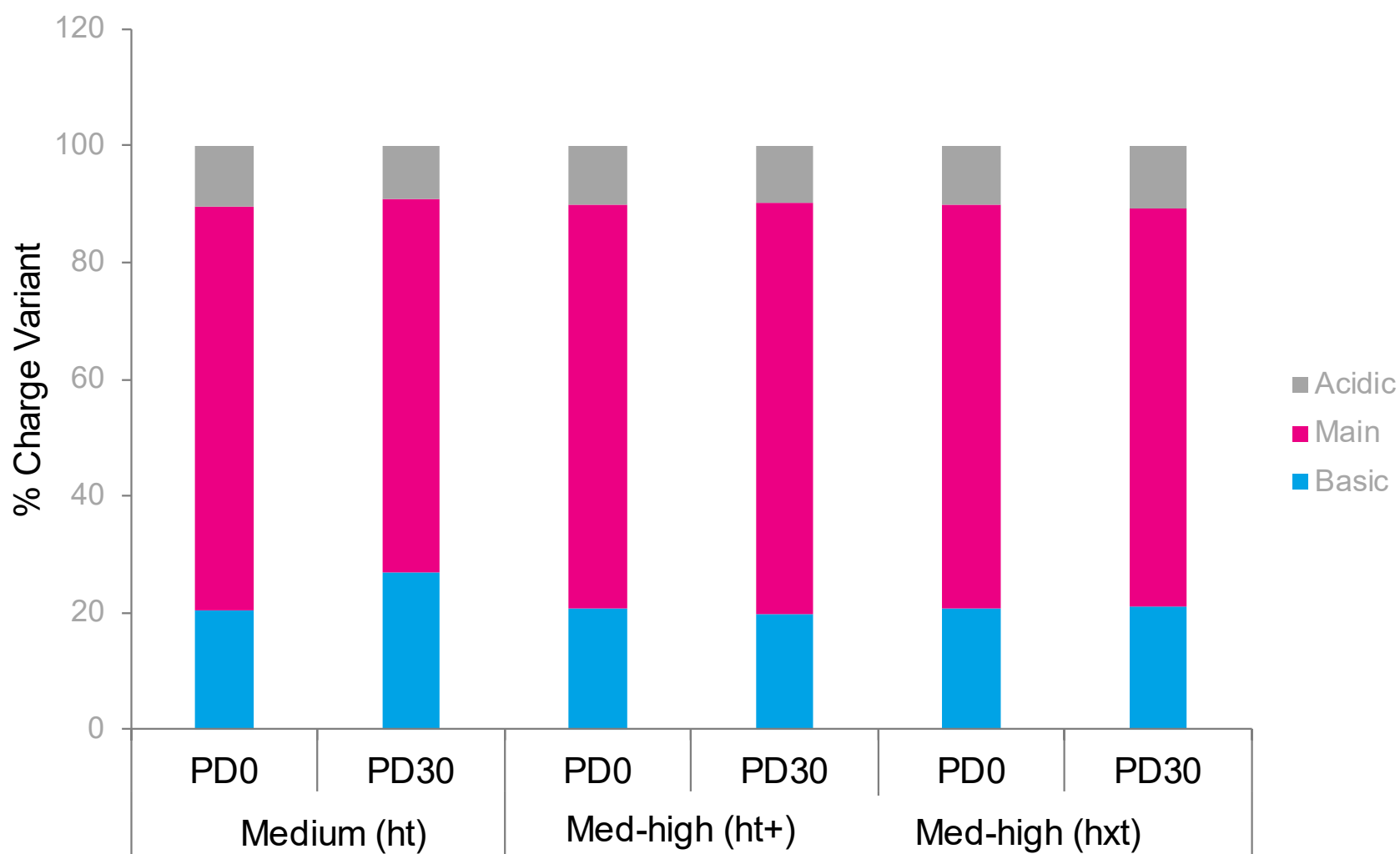
Common glycoforms relative abundance



**FIGURE 8B**



**FIGURE 9A**



**FIGURE 9B**

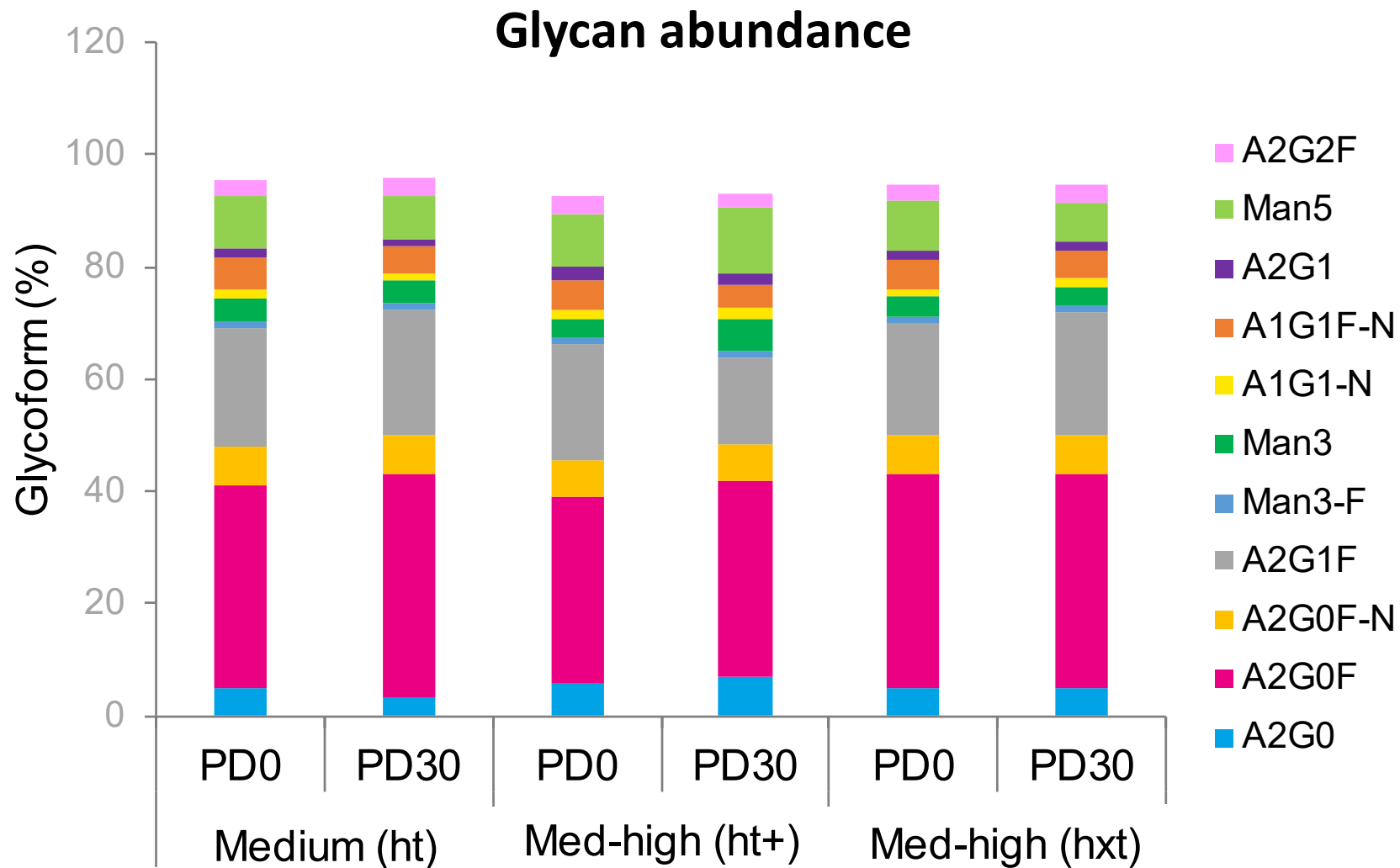


FIGURE 9C

Clone	Transposition	Non-homologous recombination
963-0	24/25	1/25
964-5	23/24	1/24
AT-G8	58/59	1/59

**TABLE 1**



Selection stringency		Copy/cell	Productivity on day 10	
Designation	Strength		Volumetric (g/L)	Specific (pcd)
h+	low	14	1.5	8.8
ht	medium	25	2.3	17.1
hxt	medium-high	30	2.4	16.55
ht+	medium-high	39	2.8	23.05
hxt+	high	36	2.7	28.85

**TABLE 2**

	Selection stringency				
	level	low	medium-high		high
		ht	ht+	hxt	hxt+
% of cells in	Q1	27.5	42.55	46.88	57
	Q2	49.3	36.17	34.38	29
	Q3	18.8	17.02	15.63	14
	Q4	4.3	4.26	3.13	0
Number of clones tested		69	47	32	14
Outgrowth rate		72%	49%	33%	15%

**TABLE 3**

	Productivity (mg/L)		Rel. stdev%
	Pool	Clonal Average	
CLD program 1	3367	3824	6.24
CLD program 2	3900	4480	7.21
CLD program 3	3588	3621	17.73
CLD program 4	4432	4483	4.75
CLD program 5	3125	3039	20.17
CLD program 6	3600	3891	5.38
CLD program 7	4500	5333	4.72
CLD program 8	3856	4024	3.98
CLD program 9	4299	4970	9.23

**TABLE 4**