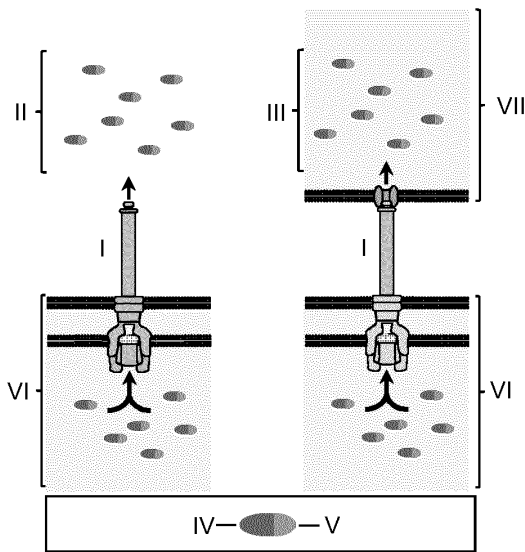
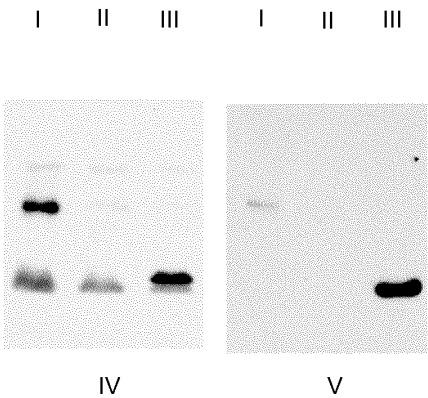


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Figure 1

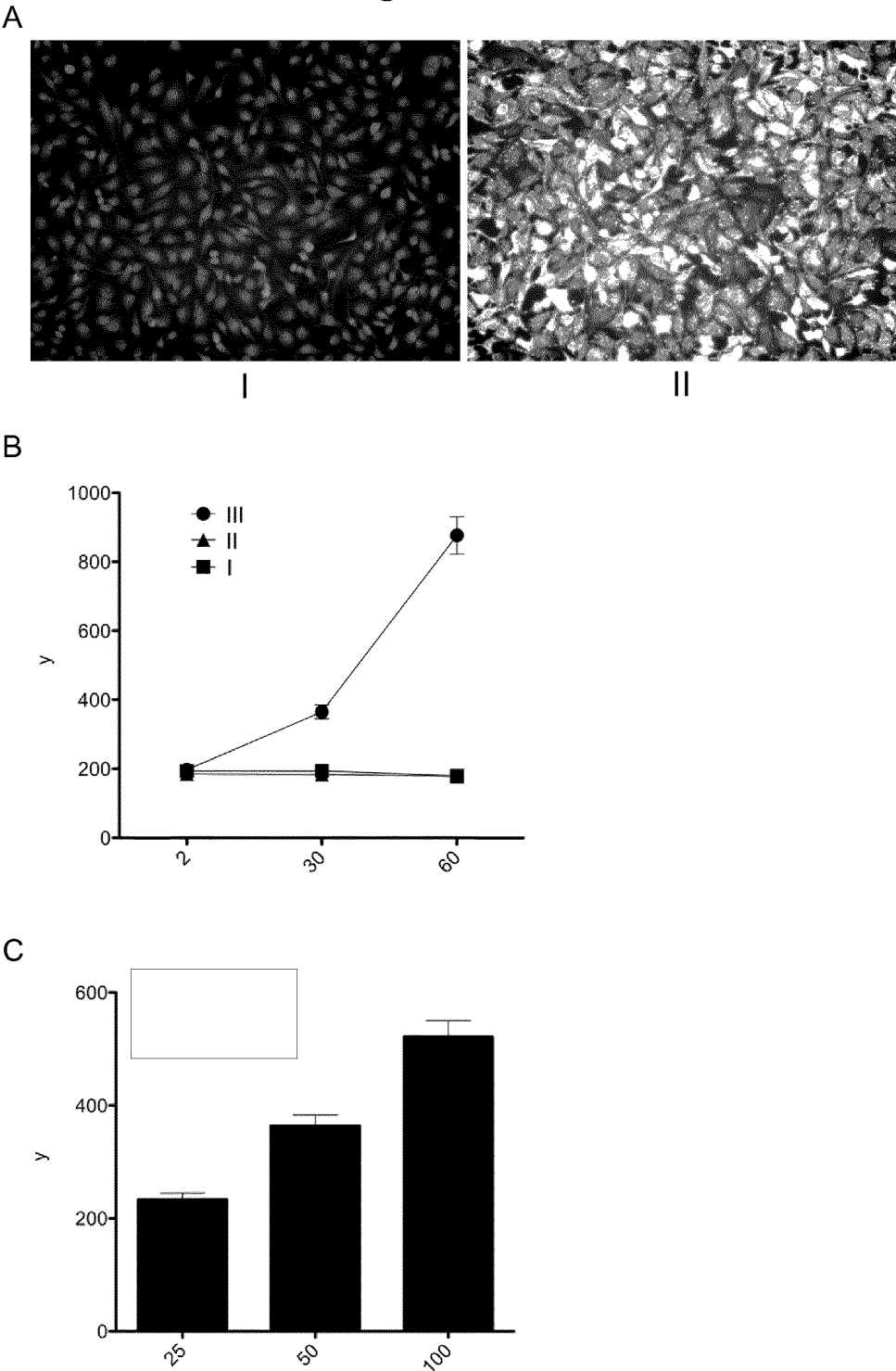
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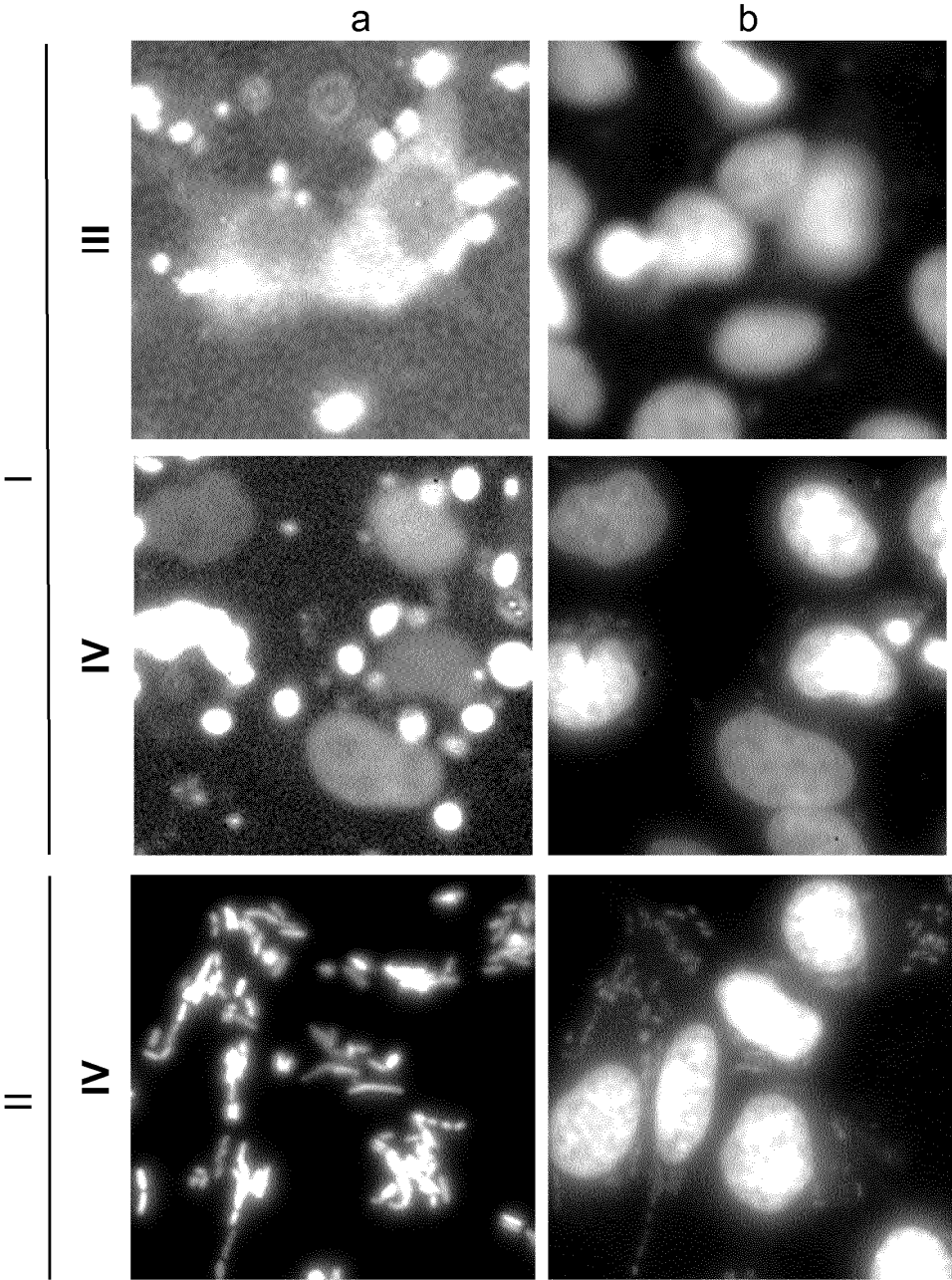
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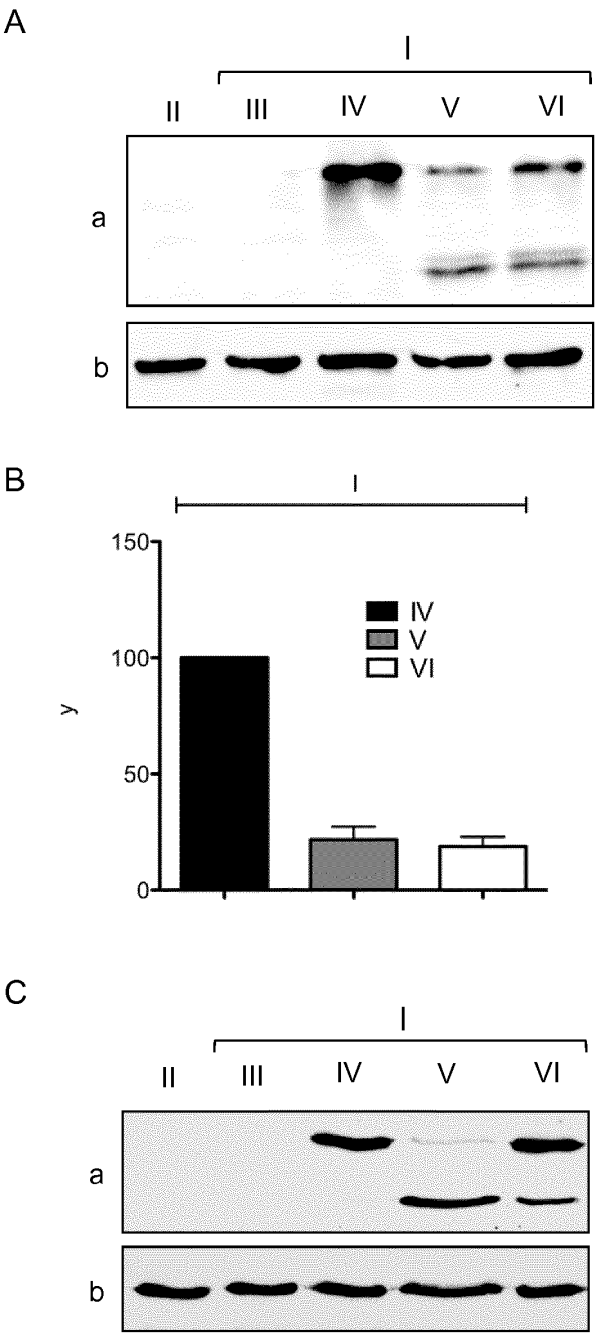
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Figure 2



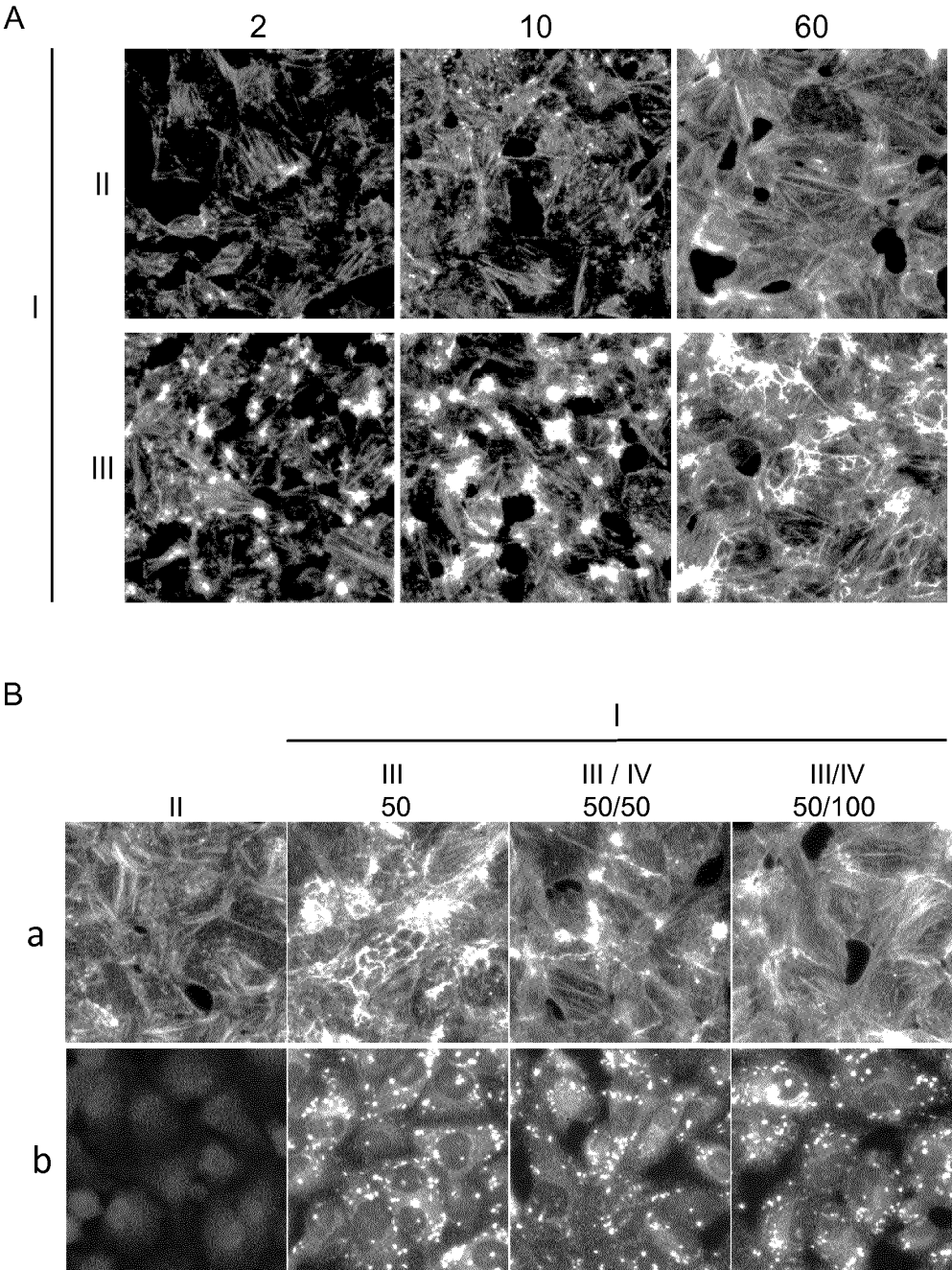
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Figure 3



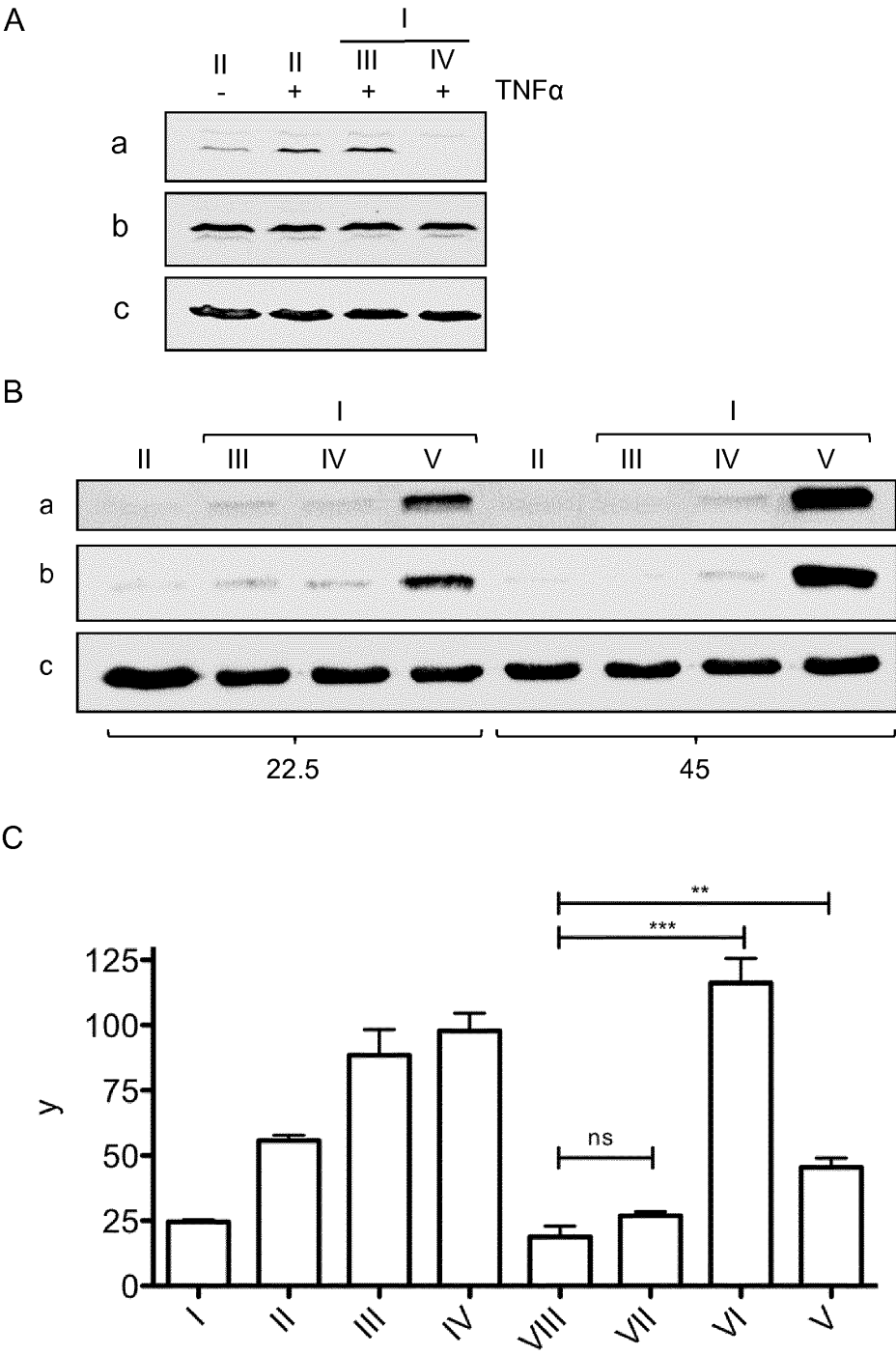
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Figure 4



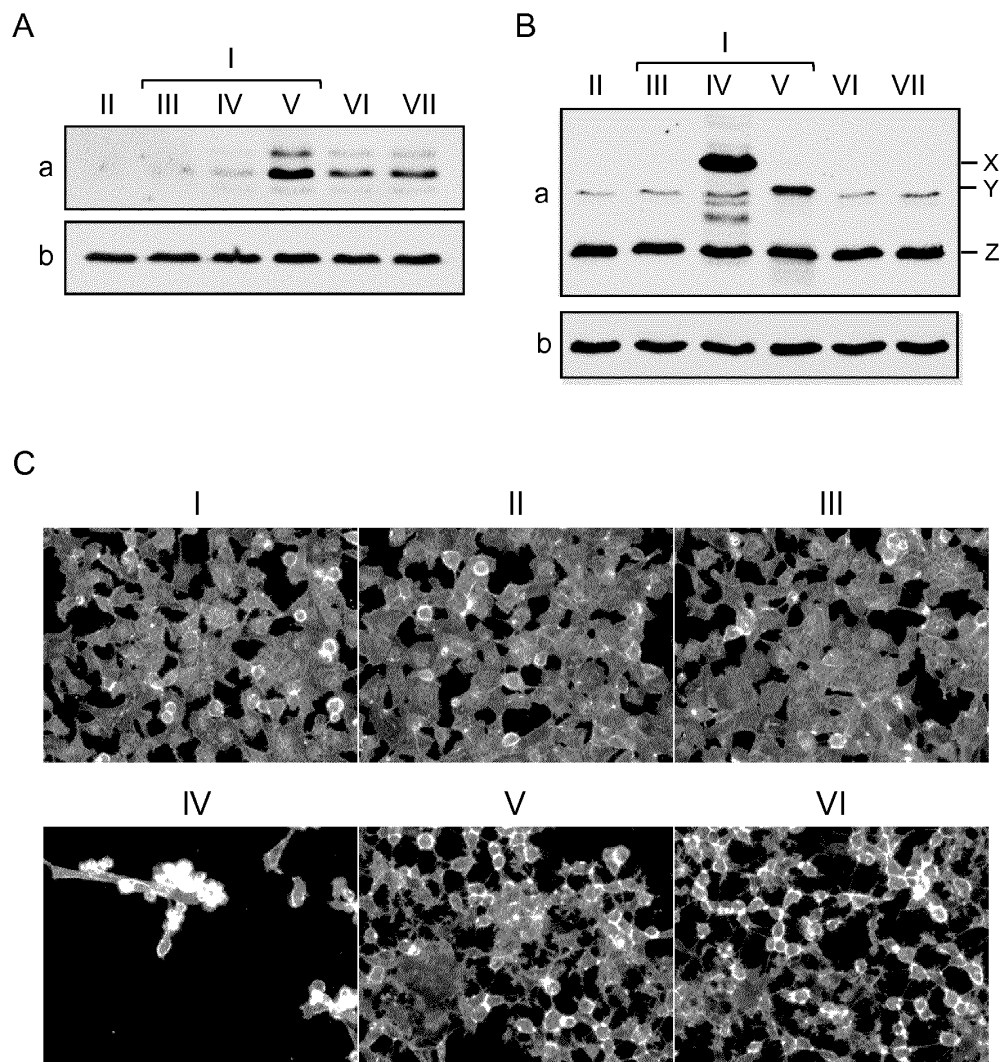
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Figure 5



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Figure 6

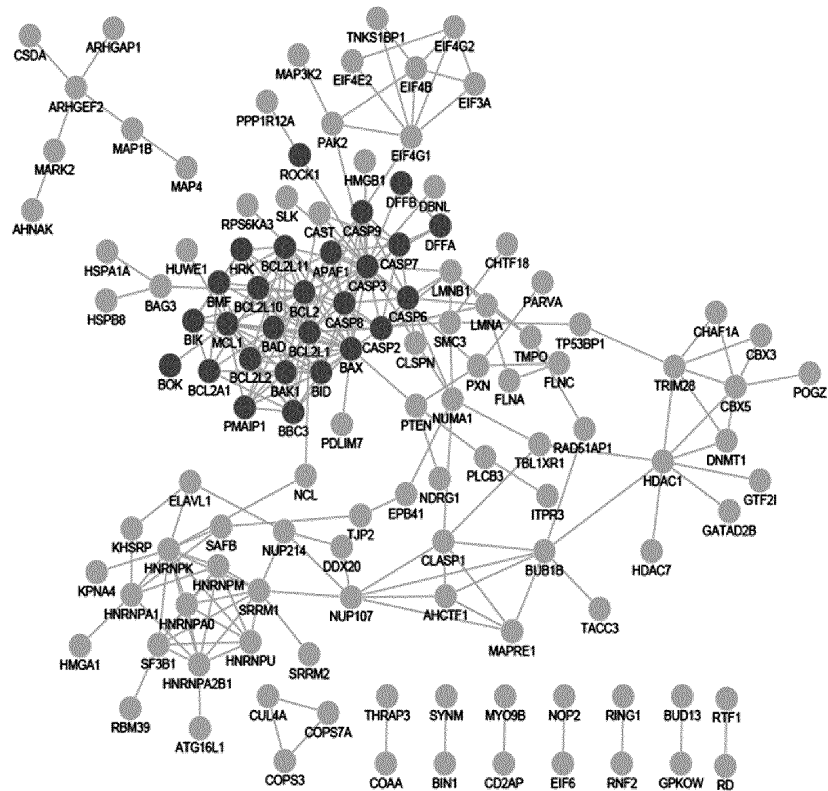


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Figure 7



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Figure 8

A



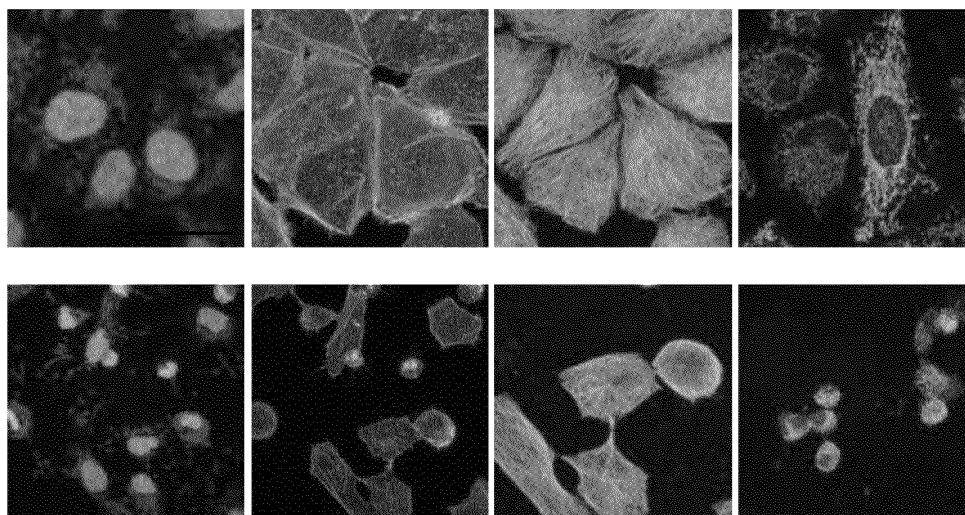
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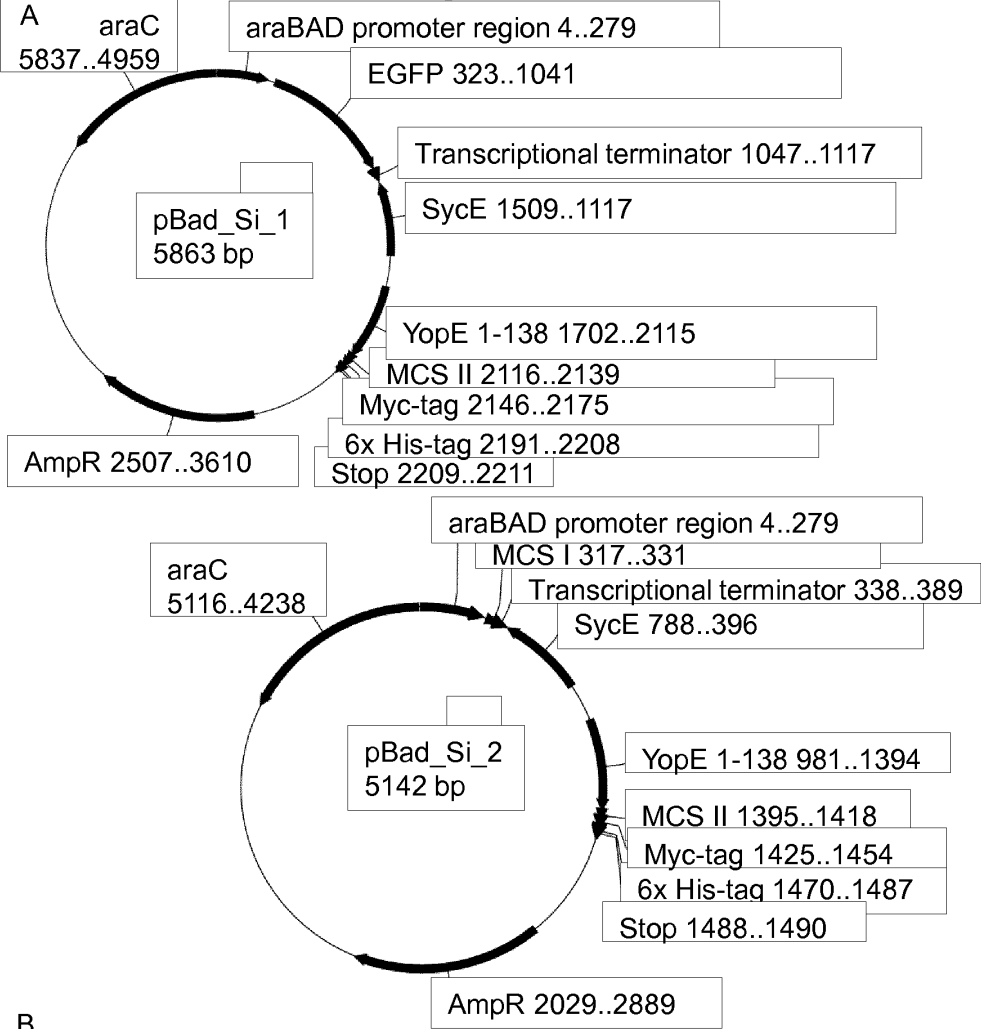
b

C

d

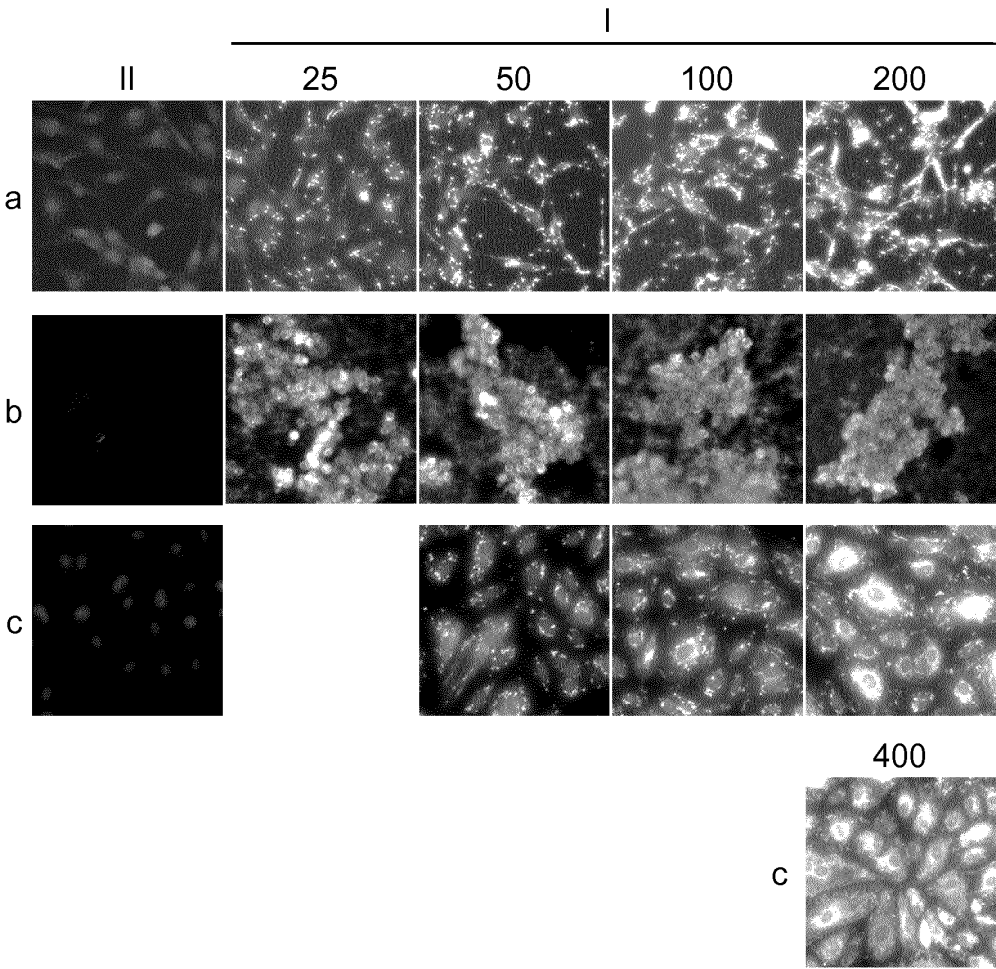


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Figure 9

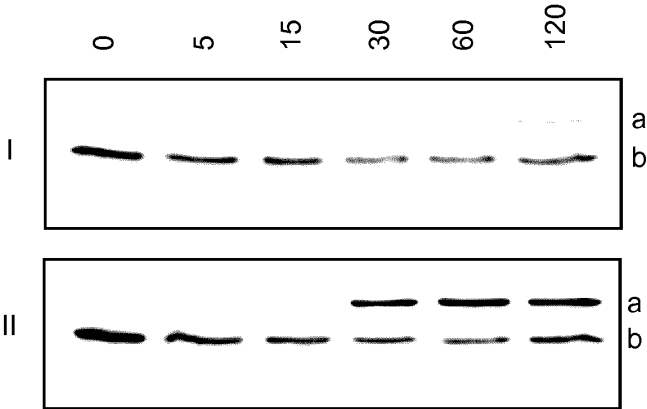


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6x His - tag ***
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Figure 10

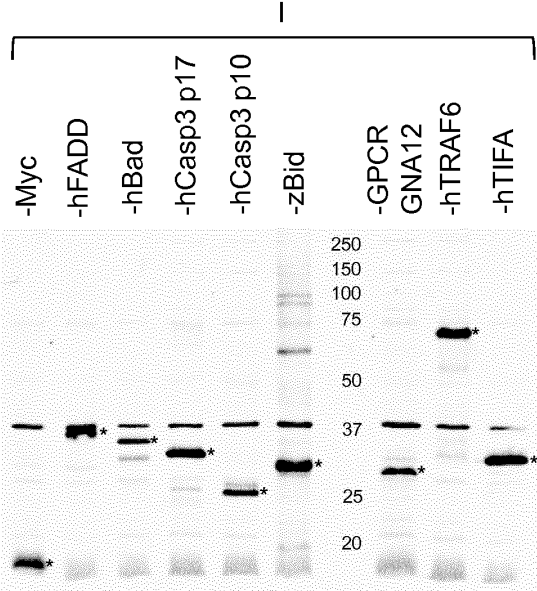


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Figure 11

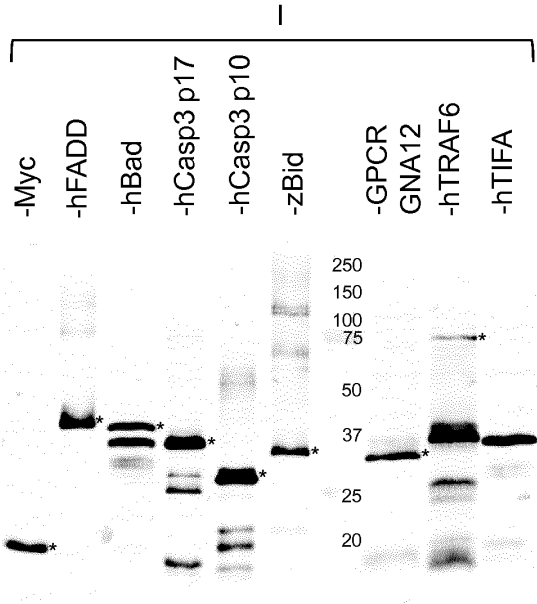


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Figure 12

A

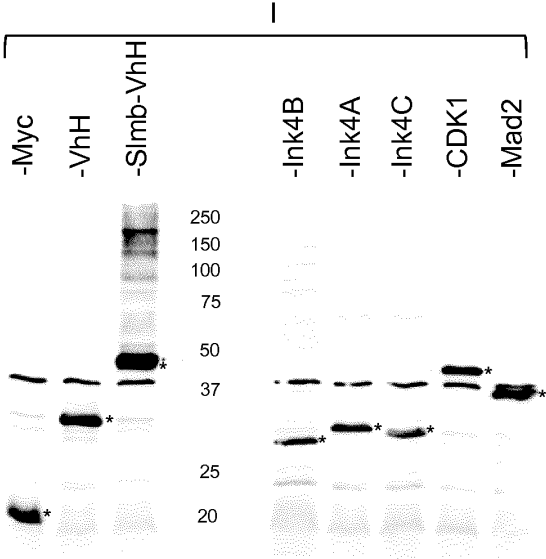


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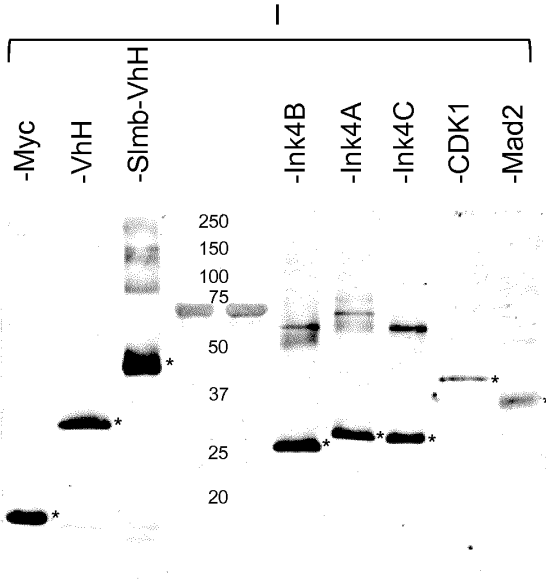


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Figure 13

A



B



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Figure 14A

Strain Name	Background strain	Insert/relevant characteristics	Protein to be delivered by T3SS	Backbone plasmid	Resulting plasmid name	Primers. Si_Nr.:	resistances	Reference
Δ HOPEMT	Y. enterocolitica Δ yopH,O,P,E,M,T	MRS40 pIML421 [yopH Δ 1-352, yopO Δ 65-558, yopP23, yopE21, yopM23, yopT135]					Nal	Iriarte and Cornelis, 1998
Δ HOPEMT asd yopB	Y. enterocolitica Δ yopH,O,P,E,M,T Δ yopB	MRS40 pIML421 [yopB Δ 89-217, yopH Δ 1-352, yopO Δ 65-558, yopP23, yopE21, yopM23, yopT135]					Nal Kan	
Δ HOPEMT asd	Y. enterocolitica Δ yopH,O,P,E,M,T Δ asd	MRS40 asd Δ 292-610 pIML421 [yopH Δ 1-352, yopO Δ 65-558, yopP23, yopE21, yopM23, yopT135]					Nal	Kudryashev et al., 2013
Δ HOPEMT asd inv	Y. enterocolitica Δ yopH,O,P,E,M,T Δ asd Δ invA	MRS40 asd Δ 292-610 invA Δ 352-2225::aphA-3 pIML421 [yopH Δ 1-352, yopO Δ 65-558, yopP23, yopE21, yopM23, yopT135]				445/446, 447/448, 449/450	Nal Kan	
Δ HOPEMT asd inv yadA	Y. enterocolitica Δ yopH,O,P,E,M,T Δ asd Δ invA Δ yadA	MRS40 asd Δ 292-610 invA Δ 587-836 (vector cointegration) yadA Δ 89-354::aphA3 pIML421 [yopH Δ 1-352, yopO Δ 65-558, yopP23, yopE21, yopM23, yopT135]				352/353, 354/355, 356/357	Nal Kan Tet	

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Figure 14B

Strain Name	Background strain	Insert/relevant characteristics	Protein to be delivered by T3SS	Backbone plasmid	Resulting plasmid name	Primers. Si_Nr.:	resistances	Reference
Δ HOPEMT asd pBad_Si1	<i>Y. enterocolitica</i> Δ yopH,O,P,E,M,T Δ asd	EGFP (Arabinose inducible), SycE-YopE1-138-MycHis fragment		pBad- MycHisA (Invitrogen)	pBad_Si_1	285/286 (EGFP), 287/288 (sycE- YopE1- 138)	Nal Amp	
Δ HOPEMT asd pBad_Si2	<i>Y. enterocolitica</i> Δ yopH,O,P,E,M,T Δ asd	SycE-YopE1-138-MycHis fragment	YopE1- 138- MycHis	pBad- MycHisA (Invitrogen)	pBad_Si_2	287/288 (sycE- YopE1- 138)	Nal Amp	
Δ HOPEMT asd pSi_16	<i>Y. enterocolitica</i> Δ yopH,O,P,E,M,T Δ asd		YopE1- 138- IpgB1	pBad_Si_2	pSi_16	292/293	Nal Amp	
Δ HOPEMT asd pSi_20	<i>Y. enterocolitica</i> Δ yopH,O,P,E,M,T Δ asd		YopE1- 138- SopE	pBad_Si_2	pSi_20	296/297	Nal Amp	
Δ HOPEMT asd pSi_22	<i>Y. enterocolitica</i> Δ yopH,O,P,E,M,T Δ asd		YopE1- 138- Rac1 Q61L	pBad_Si_2	pSi_22	299/300	Nal Amp	
Δ HOPEMT asd pSi_24	<i>Y. enterocolitica</i> Δ yopH,O,P,E,M,T Δ asd		YopE1- 138- RhoA Q61E	pBad_Si_2	pSi_24	301/302	Nal Amp	

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Figure 14C

Strain Name	Background strain	Insert/relevant characteristics	Protein to be delivered by T3SS	Backbone plasmid	Resulting plasmid name	Primers. Si_Nr.:	resistances	Reference
Δ HOPEM T asd pSi_28	Y. enterocolitica Δ yopH,O,P,E,M,T Δ asd		YopE1-138-SopE-MycHis	pBad_Si_2	pSi_28	296/306	Nal Amp	
Δ HOPEM T yopB asd pSi_28	Y. enterocolitica Δ yopH,O,P,E,M,T Δ yopB Δ asd		YopE1-138-SopE-MycHis	pBad_Si_2	pSi_28	296/306	Nal Amp	
Δ HOPEM T asd pSi_30	Y. enterocolitica Δ yopH,O,P,E,M,T Δ asd		YopE1-138-SopB	pBad_Si_2	pSi_30	307/308	Nal Amp	
Δ HOPEM T asd pSi_37	Y. enterocolitica Δ yopH,O,P,E,M,T Δ asd		YopE1-138-FADD	pBad_Si_2	pSi_37	367/386	Nal Amp	
Δ HOPEM T asd pSi_38	Y. enterocolitica Δ yopH,O,P,E,M,T Δ asd		YopE1-138-OspF	pBad_Si_2	pSi_38	317/318	Nal Amp	
Δ HOPEM T asd pSi_43	Y. enterocolitica Δ yopH,O,P,E,M,T Δ asd		YopE1-138-BepG 715-end	pBad_Si_2	pSi_43	324/351	Nal Amp	

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Figure 14D

Strain Name	Background strain	Insert/relevant characteristics	Protein to be delivered by T3SS	Backbone plasmid	Resulting plasmid name	Primers. Si_Nr.:	resistances	Reference
Δ HOPEMT asd pSi_51	<i>Y. enterocolitica</i> Δ yopH,O,P,E,M,T Δ asd		YopE1-138-Rac1 Q61L-MyhHis	pBad_Si_2	pSi_51	299/339	Nal Amp	
Δ HOPEMT yopB asd pSi_51	<i>Y. enterocolitica</i> Δ yopH,O,P,E,M,T Δ yopB Δ asd		YopE1-138-Rac1 Q61L-MyhHis	pBad_Si_2	pSi_51	299/339	Nal Amp	
Δ HOPEMT asd pSi_53	<i>Y. enterocolitica</i> Δ yopH,O,P,E,M,T Δ asd		YopE1-138-Slmb1-VhH4	pBad_Si_2	pSi_53	341/342	Nal Amp	
Δ HOPEMT asd pSi_57	<i>Y. enterocolitica</i> Δ yopH,O,P,E,M,T Δ asd		YopE1-138-Bad	pBad_Si_2	pSi_57	346/347	Nal Amp	
Δ HOPEMT asd pSi_64	<i>Y. enterocolitica</i> Δ yopH,O,P,E,M,T Δ asd		YopE1-138-SptP	pBad_Si_2	pSi_64	364/365	Nal Amp	
Δ HOPEMT asd pSi_70	<i>Y. enterocolitica</i> Δ yopH,O,P,E,M,T Δ asd		YopE1-138-NLS-Slmb1-VhH4	pBad_Si_2	pSi_70	369/342	Nal Amp	

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Figure 14E

Strain Name	Background strain	Insert/relevant characteristics	Protein to be delivered by T3SS	Backbone plasmid	Resulting plasmid name	Primers. Si_Nr.:	resistances	Reference
ΔHOPEMT asd pSi_85	Y. enterocolitica ΔyopH,O,P,E,M,T Δasd		YopE1-138-Bid	pBad_Si_2	pSi_85	387/391	Nal Amp	
ΔHOPEMT asd pSi_87	Y. enterocolitica ΔyopH,O,P,E,M,T Δasd		YopE1-138-t-Bid	pBad_Si_2	pSi_87	389/391	Nal Amp	
ΔHOPEMT asd pSi_97	Y. enterocolitica ΔyopH,O,P,E,M,T Δasd		YopE1-138-Caspase3 p17	pBad_Si_2	pSi_97	403/406	Nal Amp	
ΔHOPEMT asd pSi_103	Y. enterocolitica ΔyopH,O,P,E,M,T Δasd		YopE1-138-GPCR GNA12	pBad_Si_2	pSi_103	410/413	Nal Amp	
ΔHOPEMT asd pSi_106	Y. enterocolitica ΔyopH,O,P,E,M,T Δasd		YopE1-138-Caspase3 p10/12	pBad_Si_2	pSi_106	417/420	Nal Amp	
ΔHOPEMT asd pSi_111	Y. enterocolitica ΔyopH,O,P,E,M,T Δasd		YopE1-138-IpgD	pBad_Si_2	pSi_111	423/424	Nal Amp	
ΔHOPEMT asd pSi_112	Y. enterocolitica ΔyopH,O,P,E,M,T Δasd		YopE1-138-Slmb1- VhH4-NLS	pBad_Si_2	pSi_112	341/425	Nal Amp	

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Figure 14F

Strain Name	Background strain	Insert/relevant characteristics	Protein to be delivered by T3SS	Backbone plasmid	Resulting plasmid name	Primers. Si_Nr.:	resistances	Reference
Δ HOPEMT asd pSi_116	Y. enterocolitica Δ yopH,O,P,E,M, T Δ asd		YopE1-138-z-Bid	pBad_Si_2	pSi_116	428/430	Nal Amp	
Δ HOPEMT asd pSi_117	Y. enterocolitica Δ yopH,O,P,E,M, T Δ asd		YopE1-138-z-t-Bid	pBad_Si_2	pSi_117	429/430	Nal Amp	
Δ HOPEMT asd pSi_118	Y. enterocolitica Δ yopH,O,P,E,M, T Δ asd		YopE1-138-BepA E305- end	pBad_Si_2	pSi_118	433/435	Nal Amp	
Δ HOPEMT asd pSi_119	Y. enterocolitica Δ yopH,O,P,E,M, T Δ asd		YopE1-138-BepA	pBad_Si_2	pSi_119	434/435	Nal Amp	
Δ HOPEMT asd pSi_120	Y. enterocolitica Δ yopH,O,P,E,M, T Δ asd		YopE1-138-ET1	pBad_Si_2	pSi_120	436/437	Nal Amp	
Δ HOPEMT asd pSi_121	Y. enterocolitica Δ yopH,O,P,E,M, T Δ asd		YopE1-138-z-BIM	pbad_Si_1	pSi_121	438/439	Nal Amp	
Δ HOPEMT asd pSi_124	Y. enterocolitica Δ yopH,O,P,E,M, T Δ asd		YopE1-138-VhH4 nanobody recognizing EGFP	pBad_Si_2	pSi_124	451/373	Nal Amp	

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Figure 14G

Strain Name	Background strain	Insert/relevant characteristics	Protein to be delivered by T3SS	Backbone plasmid	Resulting plasmid name	Primers. Si_Nr.:	resistances	Reference
Δ HOPEMT asd pSi_132	Y. enterocolitica Δ yopH,O,P,E,M, T Δ asd		YopE1-138-TEV protease S219V	pBad_Si_2	pSi_132	463/464	Nal Amp	
Δ HOPEMT asd pSi_140	Y. enterocolitica Δ yopH,O,P,E,M, T Δ asd		YopE1-138-EGFP	pBad_Si_2	pSi_140	477/476	Nal Amp	
Δ HOPEMT asd pSi_143	Y. enterocolitica Δ yopH,O,P,E,M, T Δ asd		YopE1-138-Cdk1	pBad_Si_2	pSi_143	478/479	Nal Amp	
Δ HOPEMT asd pSi_145	Y. enterocolitica Δ yopH,O,P,E,M, T Δ asd		YopE1-138-Mad2	pBad_Si_2	pSi_145	482/483	Nal Amp	
Δ HOPEMT asd pSi_147	Y. enterocolitica Δ yopH,O,P,E,M, T Δ asd		YopE1-138-Ink4A	pBad_Si_2	pSi_147	486/487	Nal Amp	
Δ HOPEMT asd pSi_150	Y. enterocolitica Δ yopH,O,P,E,M, T Δ asd		YopE1-138-Ink4B	pBad_Si_2	pSi_150	492/493	Nal Amp	
Δ HOPEMT asd pSi_151	Y. enterocolitica Δ yopH,O,P,E,M, T Δ asd		YopE1-138-Ink4C	pBad_Si_2	pSi_151	494/495	Nal Amp	

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Figure 14H

Strain Name	Background strain	Insert/relevant characteristics	Protein to be delivered by T3SS	Backbone plasmid	Resulting plasmid name	Primers. Si_Nr.:	resistances	Reference
Δ HOPEMT asd pSi_153	Y. enterocolitica Δ yopH,O,P,E,M,T Δ asd		YopE1-138-TIFA	pBad_Si_2	pSi_153	558/559	Nal Amp	
Δ HOPEMT asd pSi_156	Y. enterocolitica Δ yopH,O,P,E,M,T Δ asd		YopE1-138-2x TEVsite - ET1	pBad_Si_2	pSi_156	504/505	Nal Amp	
Δ HOPEMT asd pSi_159	Y. enterocolitica Δ yopH,O,P,E,M,T Δ asd		YopE1-138- 2xTEVsite - EGFP - NLS	pBad_Si_2	pSi_159	511/513	Nal Amp	
Δ HOPEMT asd pSi_160	Y. enterocolitica Δ yopH,O,P,E,M,T Δ asd		YopE1-138- 2xTEVsite - NLS - EGFP	pBad_Si_2	pSi_160	512/476	Nal Amp	
Δ HOPEMT asd pSi_161	Y. enterocolitica Δ yopH,O,P,E,M,T Δ asd		YopE1-138-2x TEVsite - INK4C	pBad_Si_2	pSi_161	508/509	Nal Amp	
Δ HOPEMT asd pSi_164	Y. enterocolitica Δ yopH,O,P,E,M,T Δ asd		YopE1-138-2x TEVsite - Flag - INK4C	pBad_Si_2	pSi_164	515/509	Nal Amp	
Δ HOPEMT asd pSi_166	Y. enterocolitica Δ yopH,O,P,E,M,T Δ asd		YopE1-138-murine Traf6	pBad_Si_2	pSi_166	561/562	Nal Amp	

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Figure 14I

Strain Name	Background strain	Insert/rel evant character istics	Protein to be delivered by T3SS	Backbone plasmid	Resulting plasmid name	Primers. Si_Nr.:	resist ances	Referenc e
Δ HOPEMT asd pSi_318	Y. enterocolitica Δ yopH,O,P,E,M,T Δ asd		YopE1-138-Y. enterocolitica codon optimized murine tBid BH3 part	pBad_Si_2	pSi_318	677/678	Nal Amp	
Δ HOPEMT asd pSi_322	Y. enterocolitica Δ yopH,O,P,E,M,T Δ asd		YopE1-138-Y. enterocolitica codon optimized murine Bax BH3 part	pBad_Si_2	pSi_322	682/683	Nal Amp	
S. enterica SL1344 Δ aroA	S. enterica subsp. enterica serovar Typhimurium SL1344 Δ aroA							Hoiseth and Stocker, 1981
S. enterica Δ aroA pSi_266	S. enterica SL1344 Δ aroA		SteA1-20	pBad- MycHisA (Invitrogen)	pSi_266	580/612	Amp	
S. enterica Δ aroA pSi_267	S. enterica SL1344 Δ aroA		SteA	pBad- MycHisA (Invitrogen)	pSi_267	580/613	Amp	

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Figure 14J

Strain Name	Background strain	Insert/relevant characteristics	Protein to be delivered by T3SS	Backbone plasmid	Resulting plasmid name	Primers. Si_Nr.:	resistance	Reference
S. enterica Δ aroA pSi_268	S. enterica SL1344 Δ aroA		SopE1-80	pBad-MycHisA (Invitrogen)	pSi_268	614/615	Amp	
S. enterica Δ aroA pSi_269	S. enterica SL1344 Δ aroA		SopE1-104	pBad-MycHisA (Invitrogen)	pSi_269	614/616	Amp	
S. enterica Δ aroA pSi_270	S. enterica SL1344 Δ aroA		SteA1-20-S. enterica codon optimized murine tBid	pSi_266	pSi_270	synthetic construct	Amp	
S. enterica Δ aroA pSi_271	S. enterica SL1344 Δ aroA		SteA-S. enterica codon optimized murine tBid	pSi_267	pSi_271	synthetic construct	Amp	
S. enterica Δ aroA pSi_272	S. enterica SL1344 Δ aroA		SopE1-80-S. enterica codon optimized murine tBid	pSi_268	pSi_272	synthetic construct	Amp	
S. enterica Δ aroA pSi_273	S. enterica SL1344 Δ aroA		SopE1-104-S. enterica codon optimized murine tBid	pSi_269	pSi_273	synthetic construct	Amp	

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Figure 14K

Strain Name	Background strain	Insert/relevant characteristics	Protein to be delivered by T3SS	Backbone plasmid	Resulting plasmid name	Primers. Si_Nr.:	resistances	Reference
Δ HOPEMT asd pSi_362	Y. enterocolitica Δ yopH,O,P,E,M,T Δ asd		YopE1-138-Y. enterocolitica codon optimized Ink4A 84-103	pBad_Si_2	pSi_362	745/746	Nal Amp	
Δ HOPEMT asd pSi_363	Y. enterocolitica Δ yopH,O,P,E,M,T Δ asd		YopE1-138-Y. enterocolitica codon optimized Cdk2/cyclin inhibitory peptide I	pBad_Si_2	pSi_363	747/748	Nal Amp	
Δ HOPEMT asd pSi_364	Y. enterocolitica Δ yopH,O,P,E,M,T Δ asd		YopE1-138-Y. enterocolitica codon optimized p21 peptide 10 (DtoA)	pBad_Si_2	pSi_364	749/750	Nal Amp	
Δ HOPEMT asd pSi_366	Y. enterocolitica Δ yopH,O,P,E,M,T Δ asd		YopE1-138-Y. enterocolitica codon optimized p21 core of peptide 10 and 11	pBad_Si_2	pSi_366	753/754	Nal Amp	
Δ HOPEMT asd pSi_367	Y. enterocolitica Δ yopH,O,P,E,M,T Δ asd		YopE1-138-Y. enterocolitica codon optimized p21 peptide 17-33	pBad_Si_2	pSi_367	755/756	Nal Amp	

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Figure 14L

Strain Name	Background strain	Insert/relevant characteristics	Protein to be delivered by T3SS	Backbone plasmid	Resulting plasmid name	Primers. Si_Nr.:	resistances	Reference
Δ HOPEMT asd pSi_368	<i>Y. enterocolitica</i> Δ yopH,O,P,E,M,T Δ asd		YopE1-138-Y. enterocolitica codon optimized pep5 of G1/S cyclin D2	pBad_Si_2	pSi_368	757/758	Nal Amp	
<i>S. enterica</i> Δ aroA pSi_333	<i>S. enterica</i> SL1344 Δ aroA		SteA-Ink4a-MycHis	pSi_267	pSi_333	703/704	Amp	
<i>S. enterica</i> Δ aroA pSi_334	<i>S. enterica</i> SL1344 Δ aroA		SopE1-104-Ink4a- MycHis	pSi_269	pSi_334	703/704	Amp	
<i>S. enterica</i> Δ aroA pSi_335	<i>S. enterica</i> SL1344 Δ aroA		SteA-Ink4c-MycHis	pSi_267	pSi_335	705/706; 707/708; 705/708	Amp	
<i>S. enterica</i> Δ aroA pSi_336	<i>S. enterica</i> SL1344 Δ aroA		SopE1-104-Ink4c- MycHis	pSi_269	pSi_336	705/706; 707/708; 705/708	Amp	
<i>S. enterica</i> Δ aroA pSi_337	<i>S. enterica</i> SL1344 Δ aroA		SteA-Mad2-MycHis	pSi_267	pSi_337	709/710	Amp	

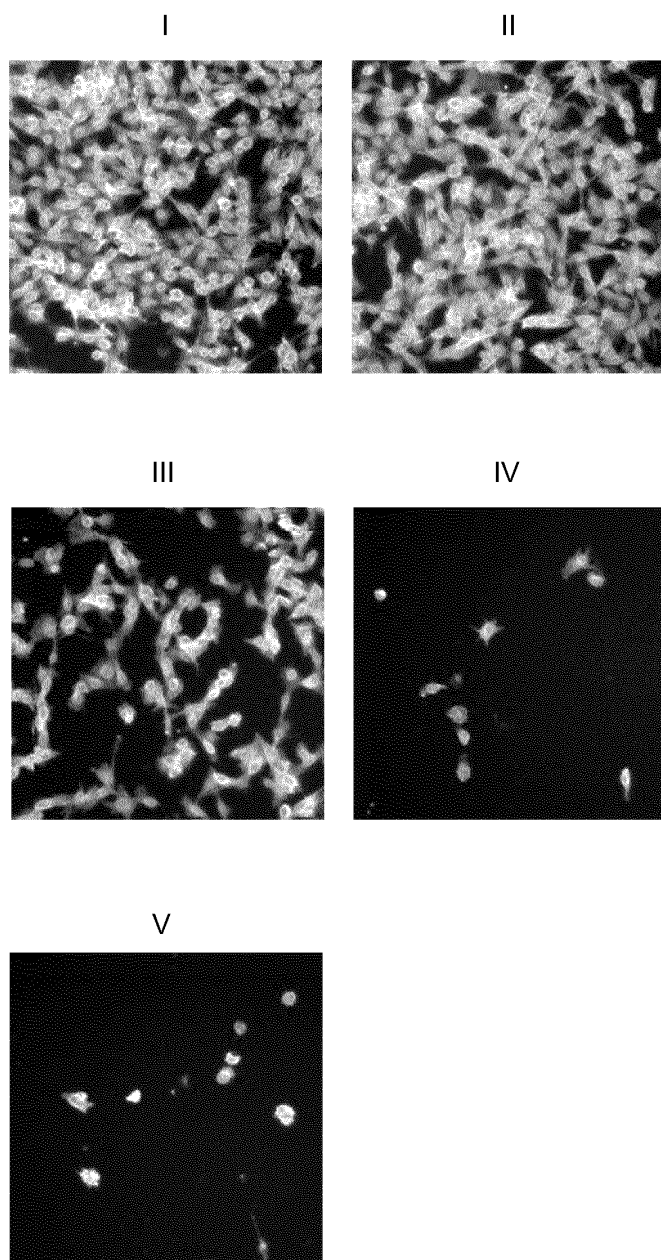
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Figure 14M

Strain Name	Background strain	Insert/relevant characteristics	Protein to be delivered by T3SS	Backbone plasmid	Resulting plasmid name	Primers. Si_Nr.:	resistance	Reference
S. enterica Δ aroA pSi_338	S. enterica SL1344 Δ aroA		SopE1-104-Mad2-MycHis	pSi_269	pSi_338	709/710	Amp	
S. enterica Δ aroA pSi_339	S. enterica SL1344 Δ aroA		SteA-Cdk1-MycHis	pSi_267	pSi_339	711/712	Amp	
S. enterica Δ aroA pSi_340	S. enterica SL1344 Δ aroA		SopE1-104-Cdk1-MycHis	pSi_269	pSi_340	711/712	Amp	
Δ HOPEMT asd pSi_315	Y. enterocolitica Δ yopH,O,P,E,M,T Δ asd		YopE1-138-Y. enterocolitica codon optimized murine tBid	pBad_Si_2	pSi_315	synthetic construct	Nal Amp	
Δ HOPEMT asd pSi_236	Y. enterocolitica Δ yopH,O,P,E,M,T Δ asd		YopE1-138-Ubiquitin	pBad_Si_2	pSi_236	585/586	Nal Amp	
Δ HOPEMT asd pSi_237_II	Y. enterocolitica Δ yopH,O,P,E,M,T Δ asd		YopE1-138-Ubiquitin-Flag-INK4C-MycHis	pSi_236	pSi_237_II	588/509	Nal Amp	

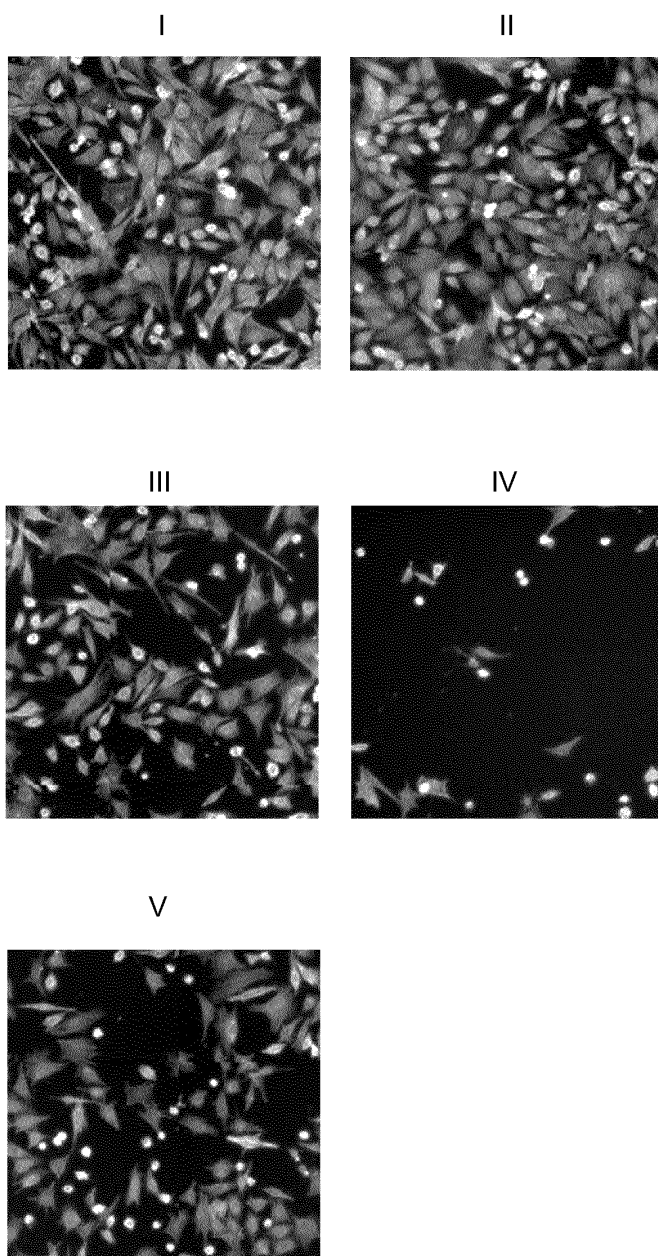
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Figure 14N

Strain Name	Background strain	Insert/relevant characteristics	Protein to be delivered by T3SS	Backbone plasmid	Resulting plasmid name	Primers. Si_Nr.:	resistance	Reference
Δ HOPEMT _{asd} pSi_357	Y. enterocolitica Δ yopH,O,P,E,M,T Δ asd		YopE1-138-Y. enterocolitica codon optimized murine tBid BH3	pBadSi_2	pSi_357	733/735	Nal Amp	
Δ HOPEMT _{asd} pSi_358	Y. enterocolitica Δ yopH,O,P,E,M,T Δ asd		YopE1-138-Y. enterocolitica codon optimized murine Bax BH3	pBad_Si_2	pSi_358	736/738	Nal Amp	
Δ HOPEMT _{asd} pSi_371	Y. enterocolitica Δ yopH,O,P,E,M,T Δ asd		YopE1-138-(Y. enterocolitica codon optimized murine tBid BH3) ₂	pSi_357	pSi_371	733/734	Nal Amp	
Δ HOPEMT _{asd} pSi_373	Y. enterocolitica Δ yopH,O,P,E,M,T Δ asd		YopE1-138-(Y. enterocolitica codon optimized murine tBid BH3) ₂	pSi_358	pSi_373	733/734	Nal Amp	

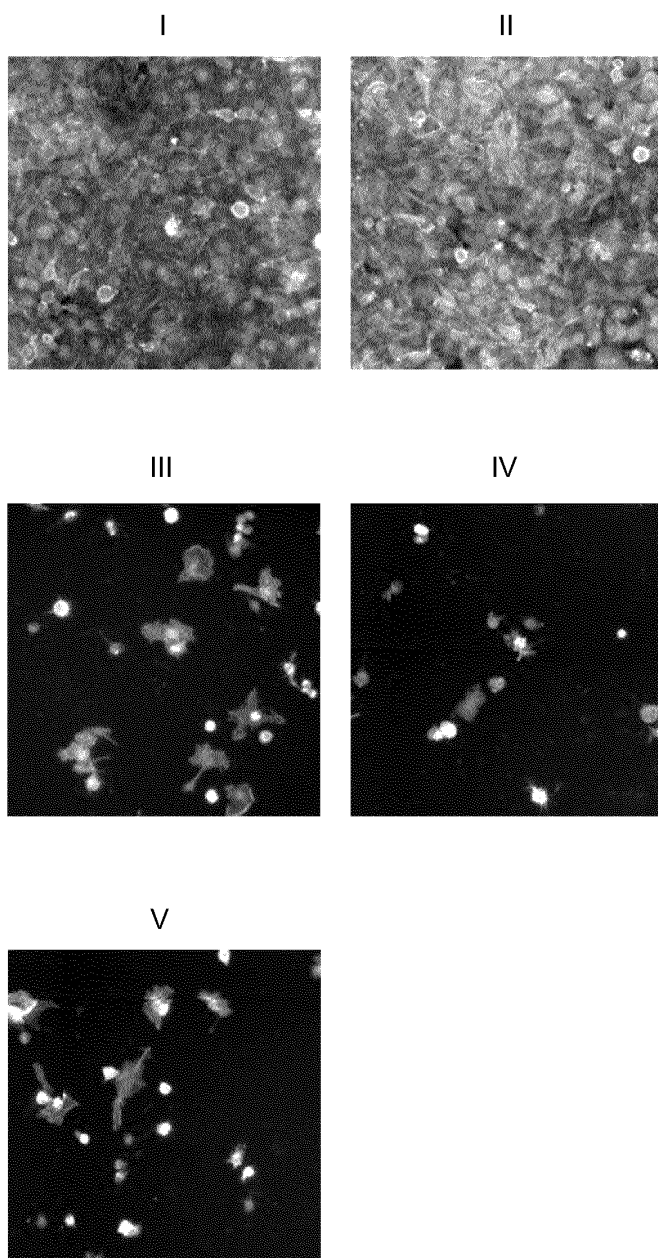
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Figure 15



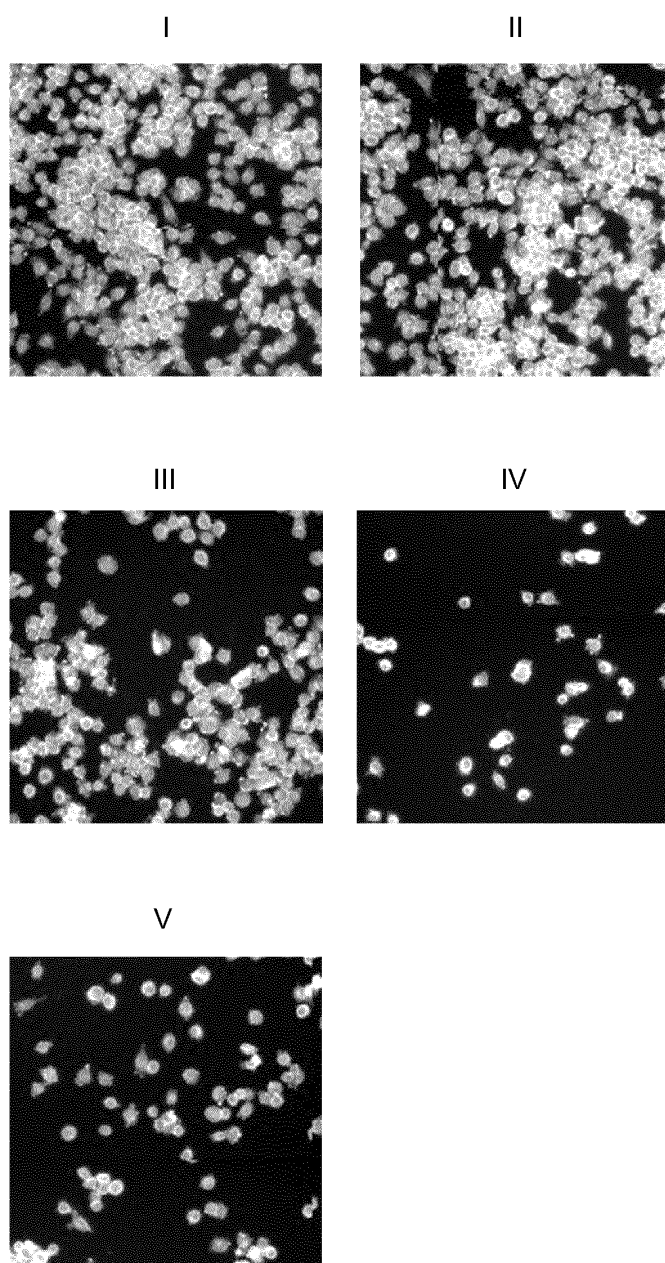
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Figure 16



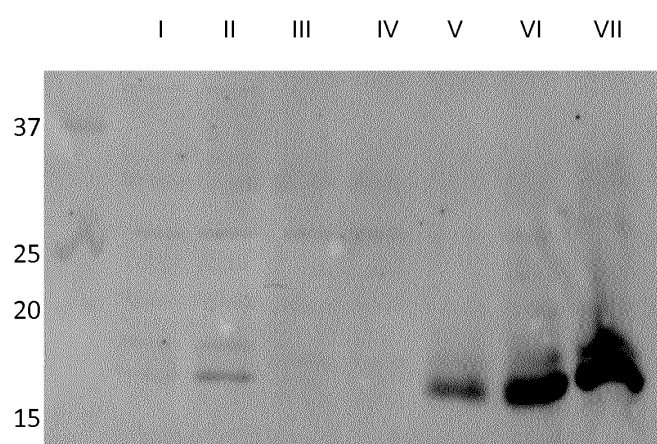
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Figure 17



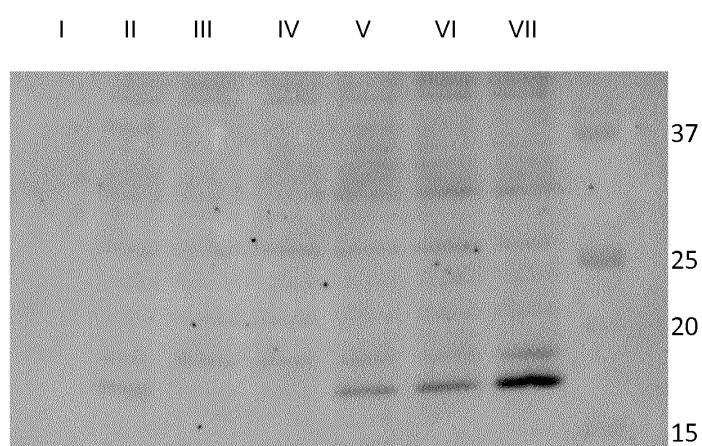
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Figure 18



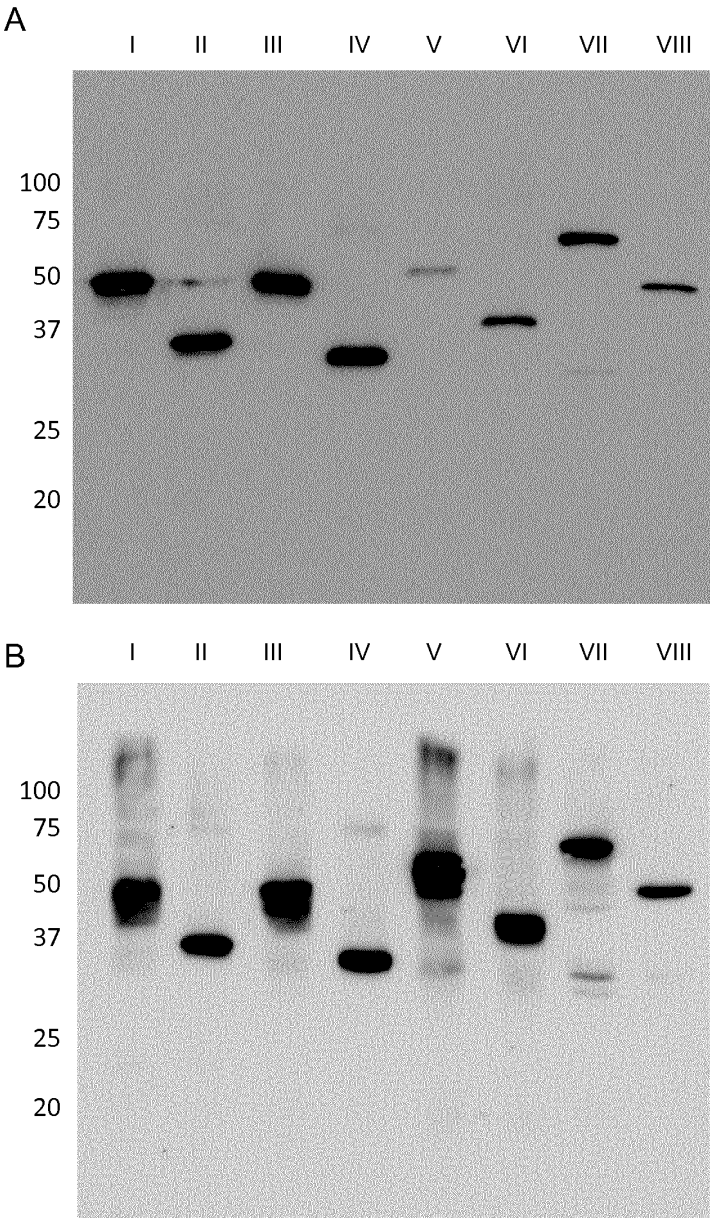
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Figure 19



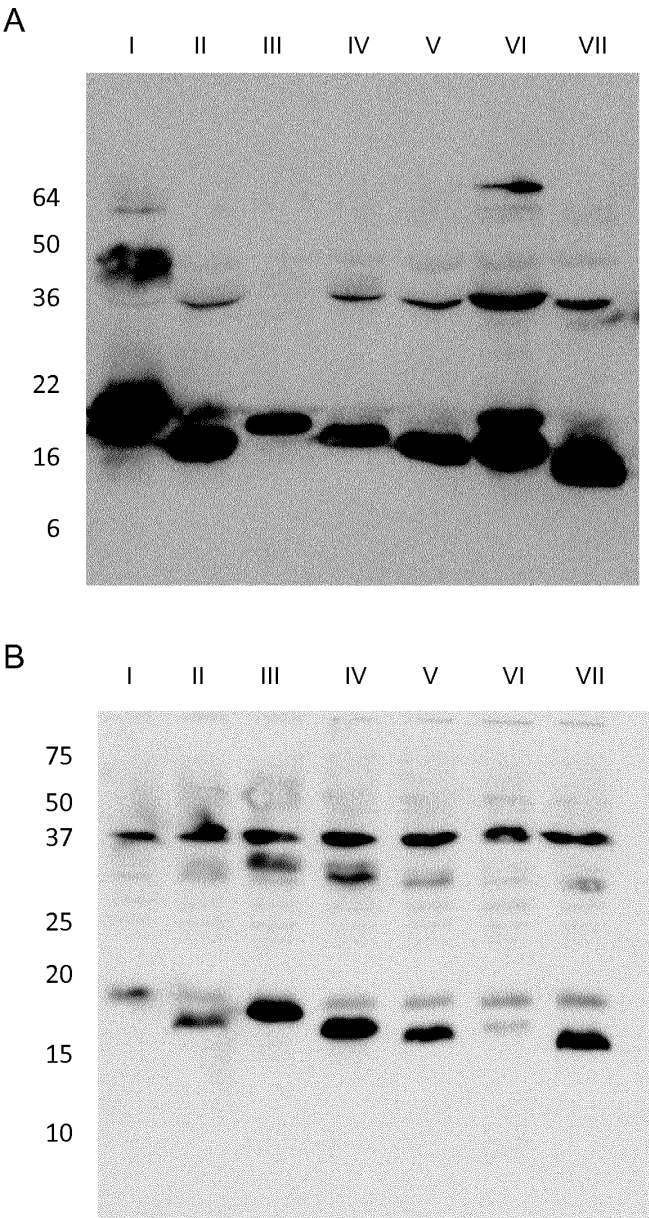
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Figure 20



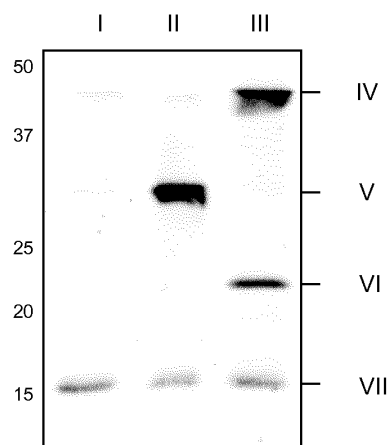
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Figure 21



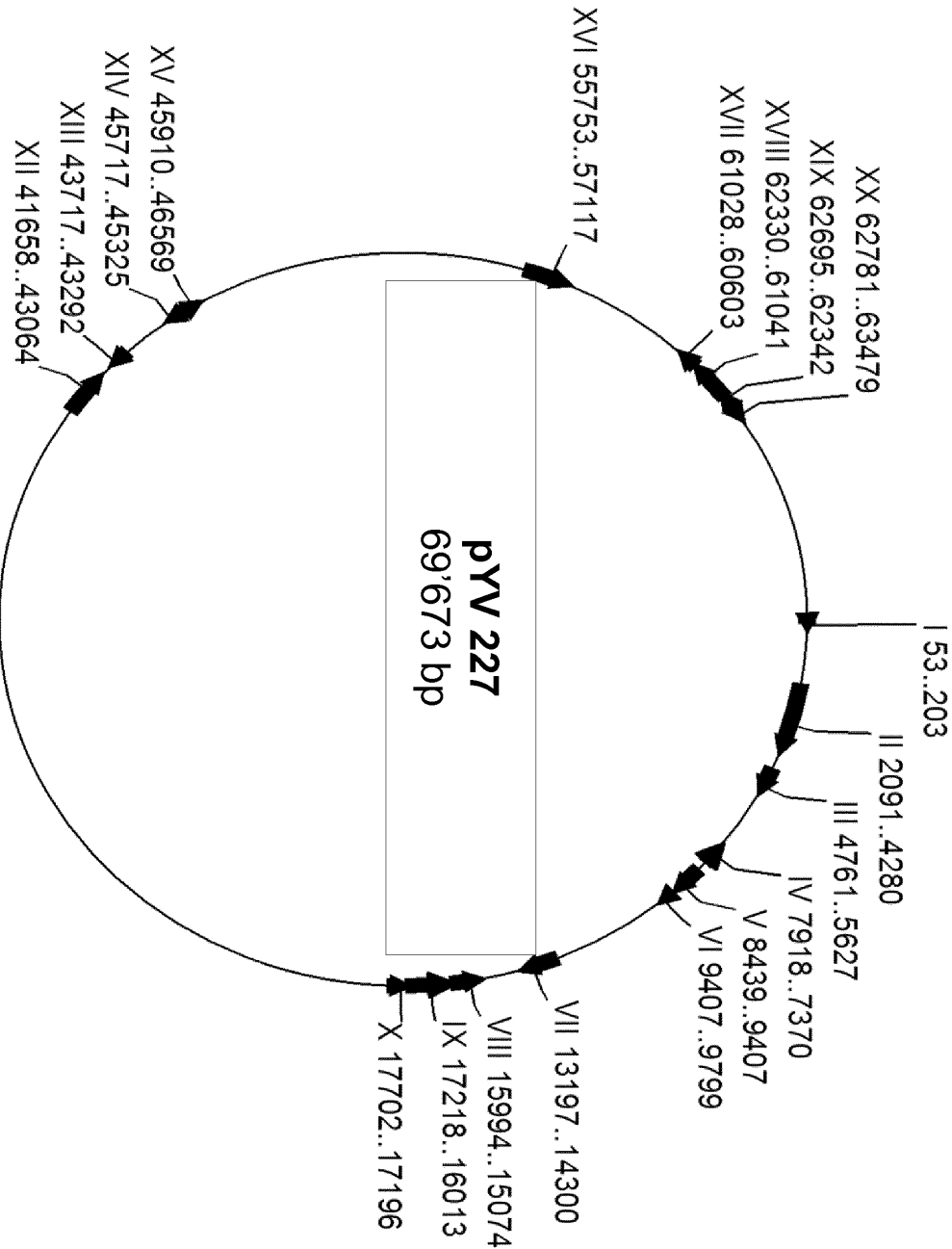
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Figure 22



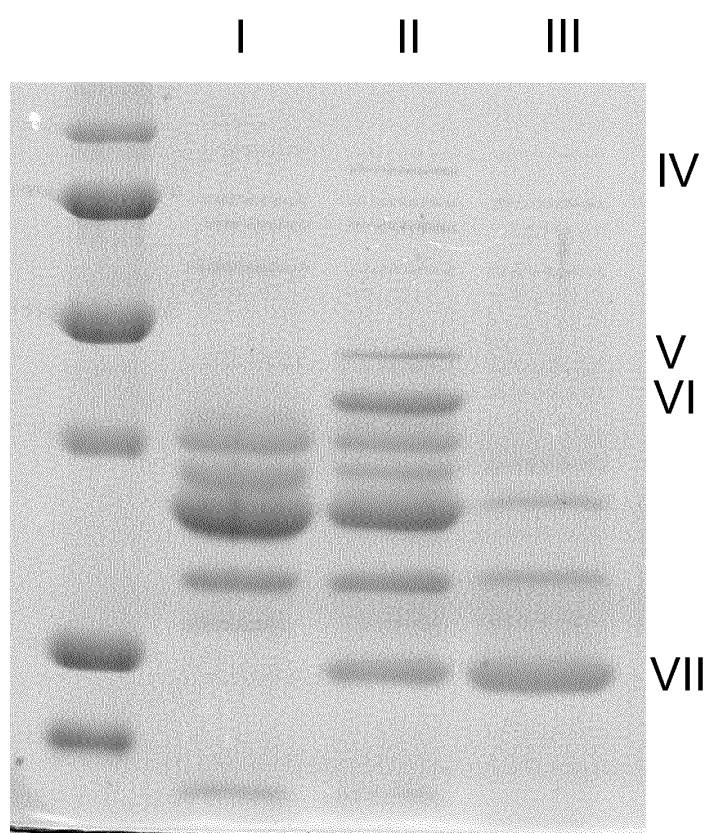
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Figure 23



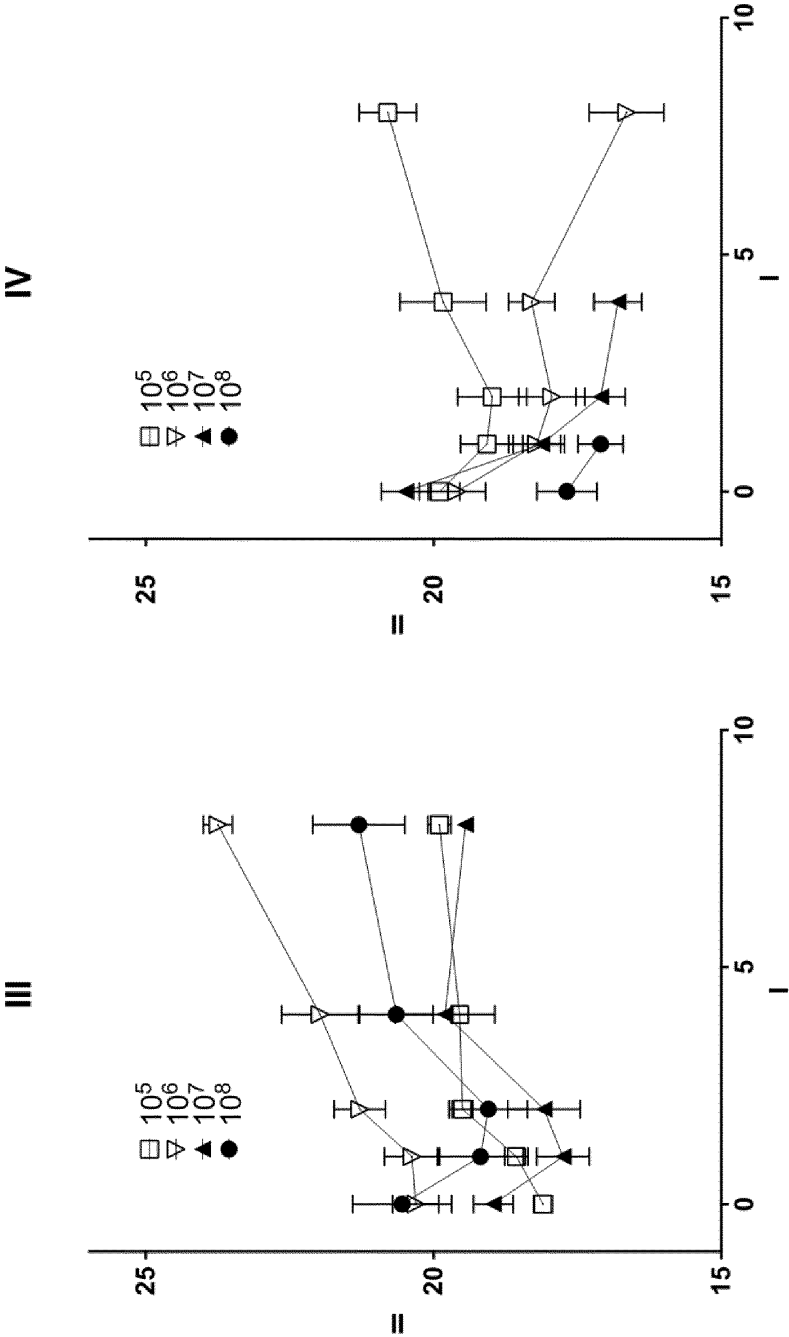
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Figure 24



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Figure 25

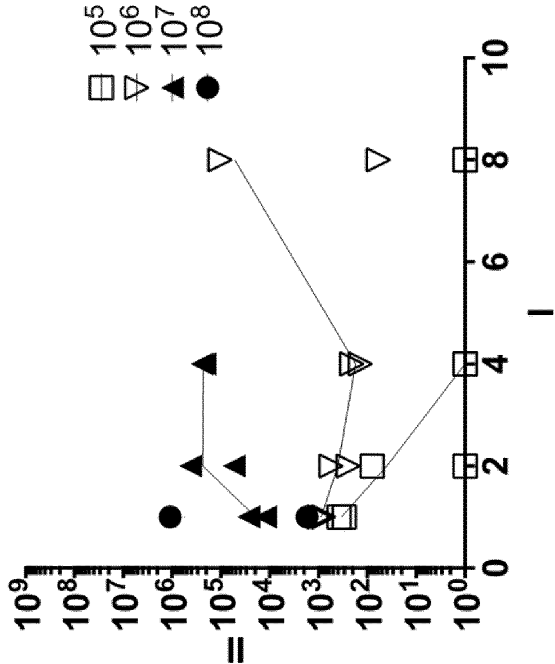


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Figure 26

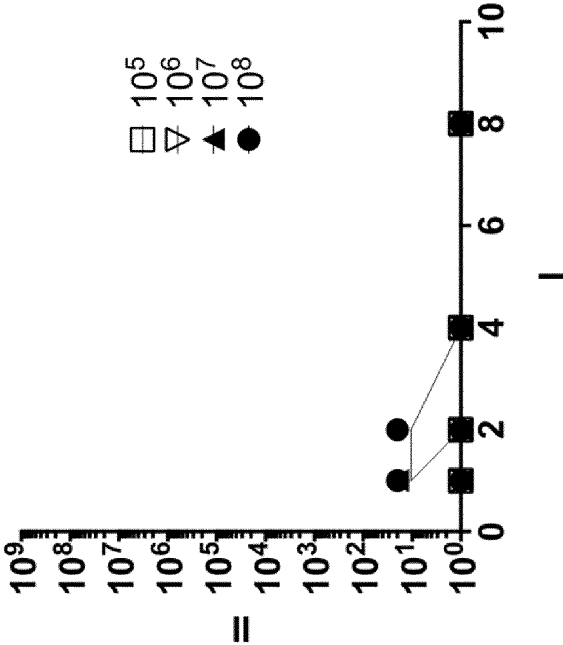


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Figure 27

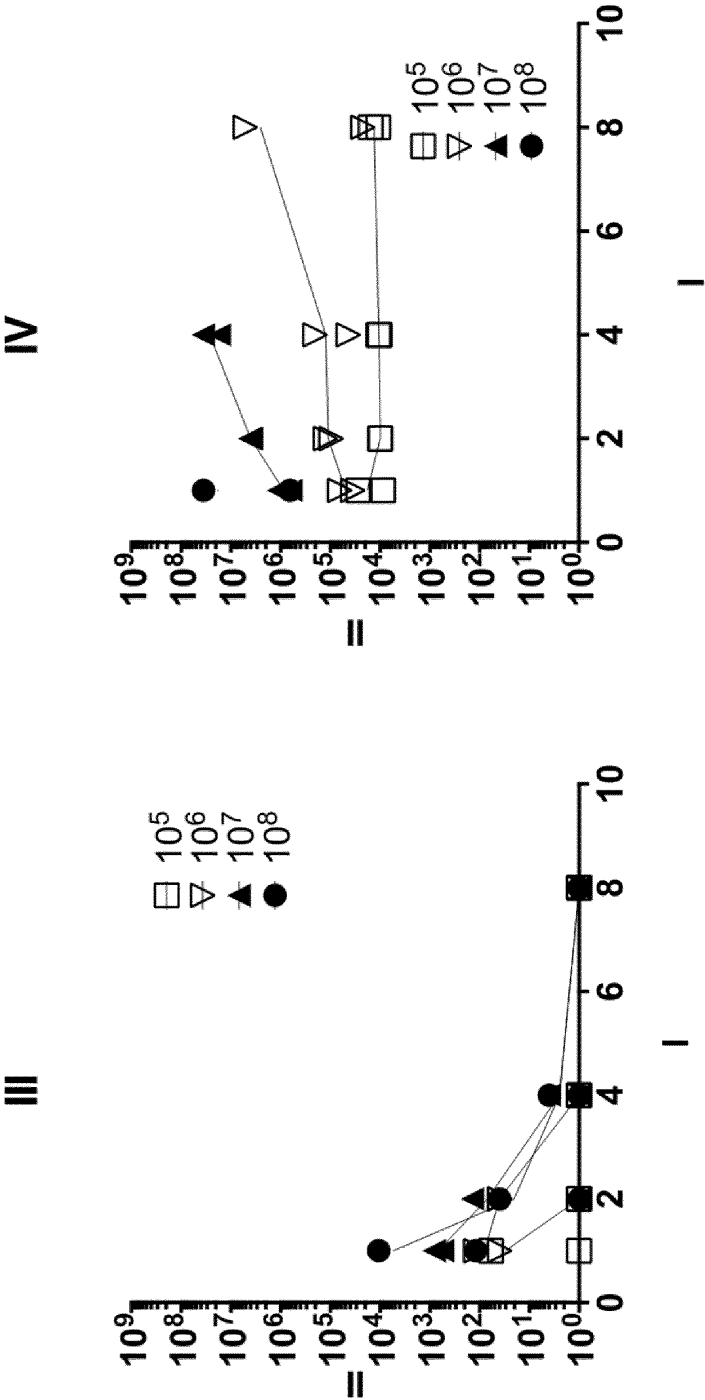
IV



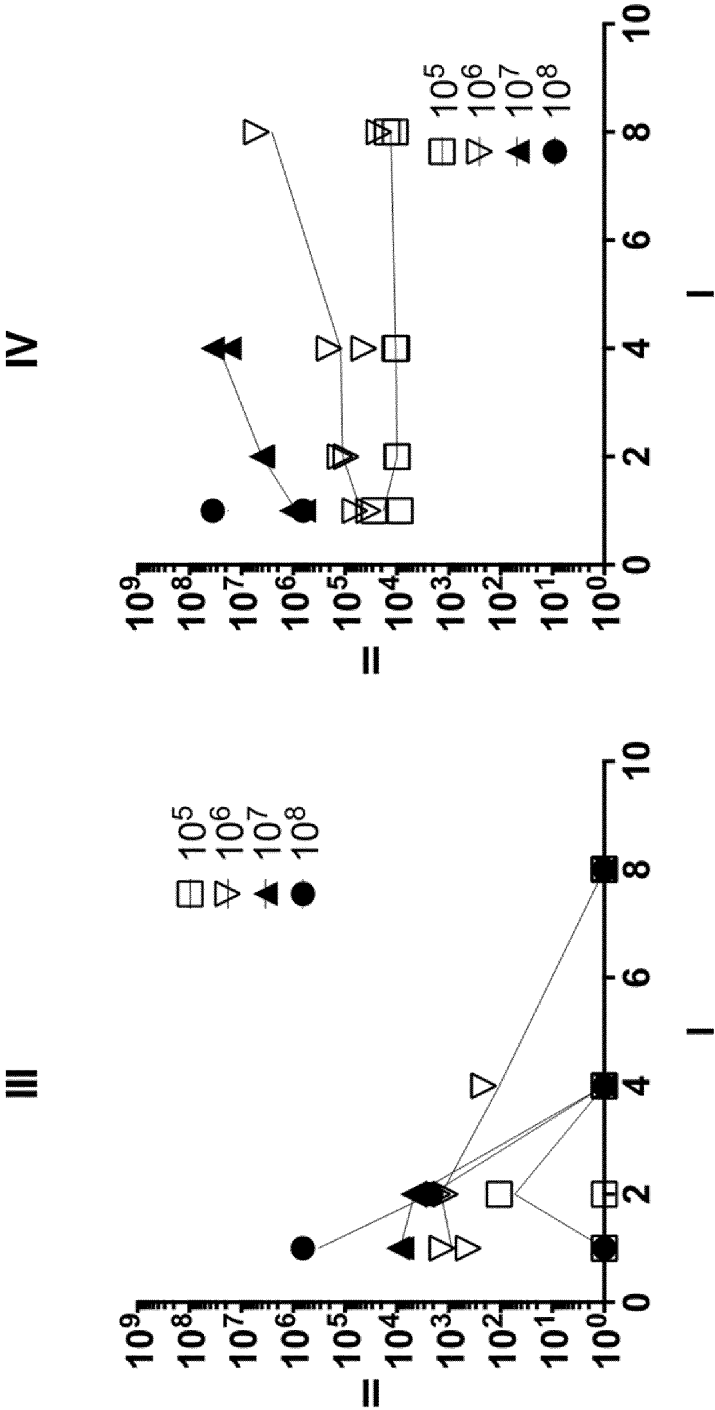
III



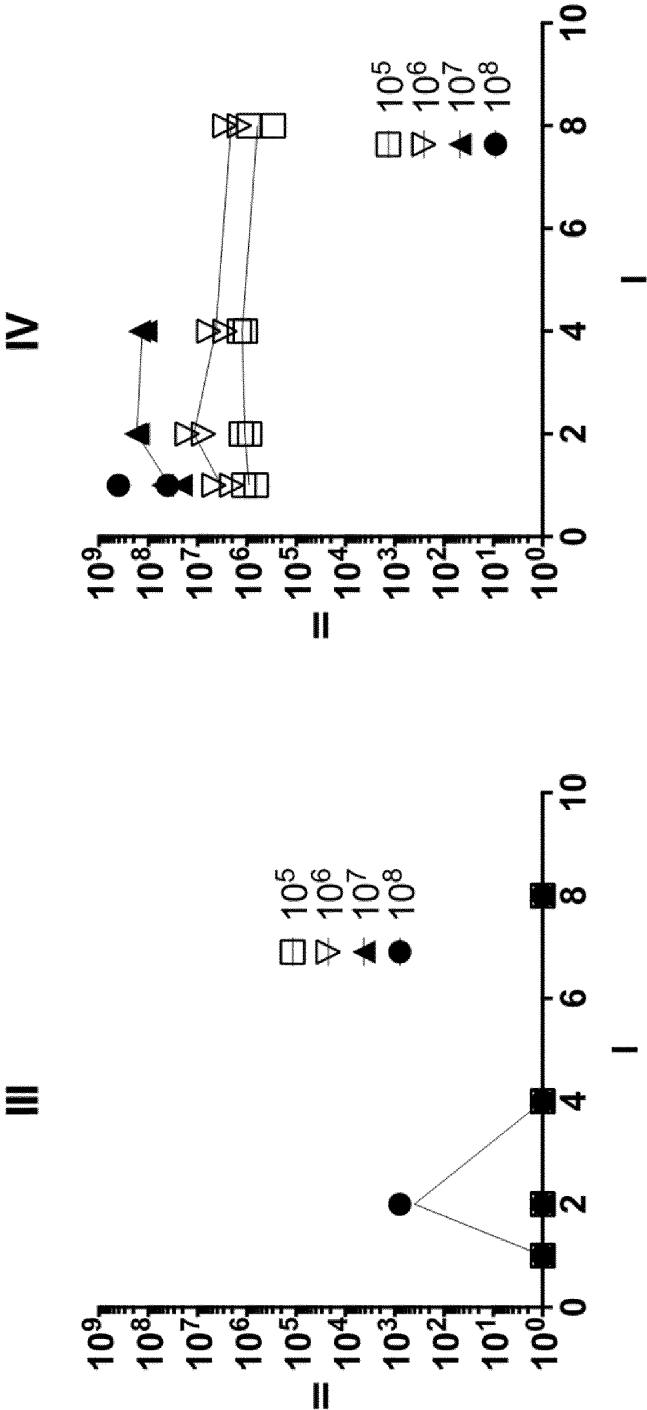
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Figure 28



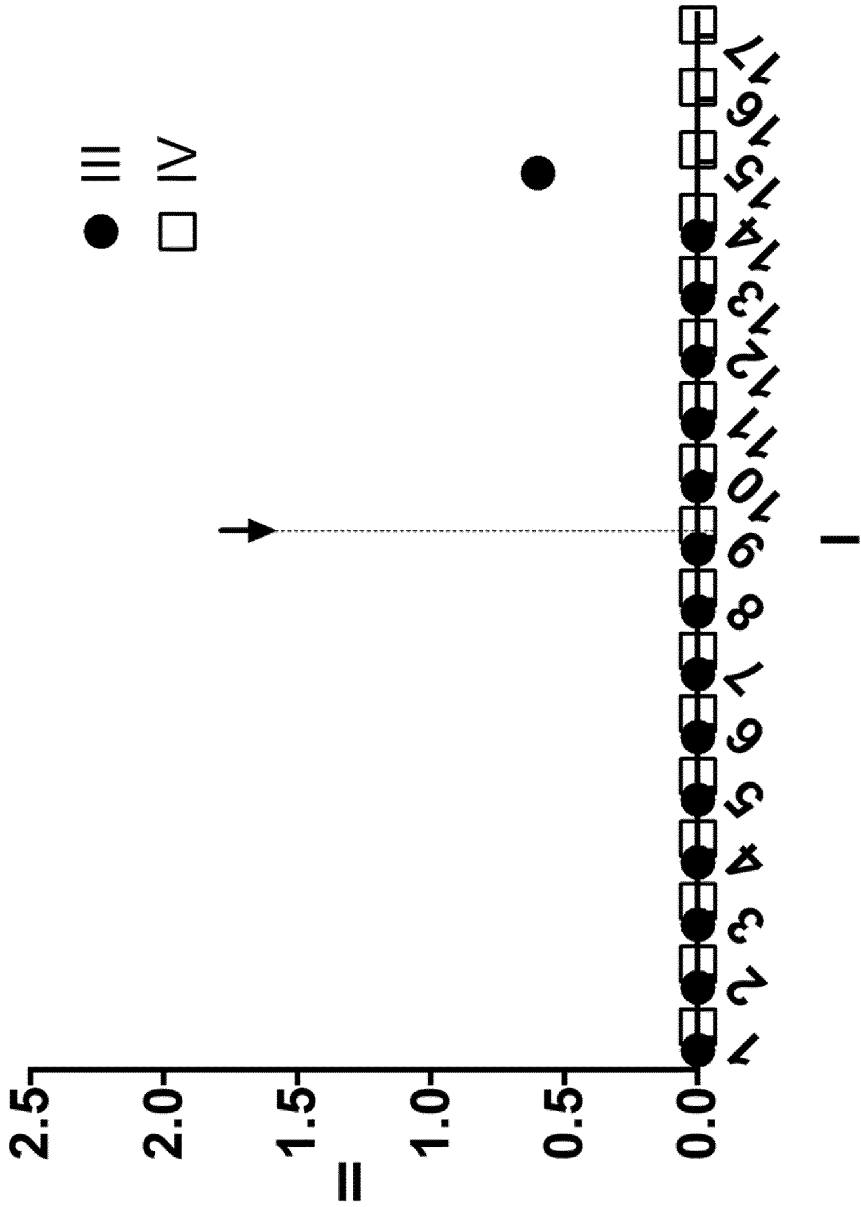
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Figure 29



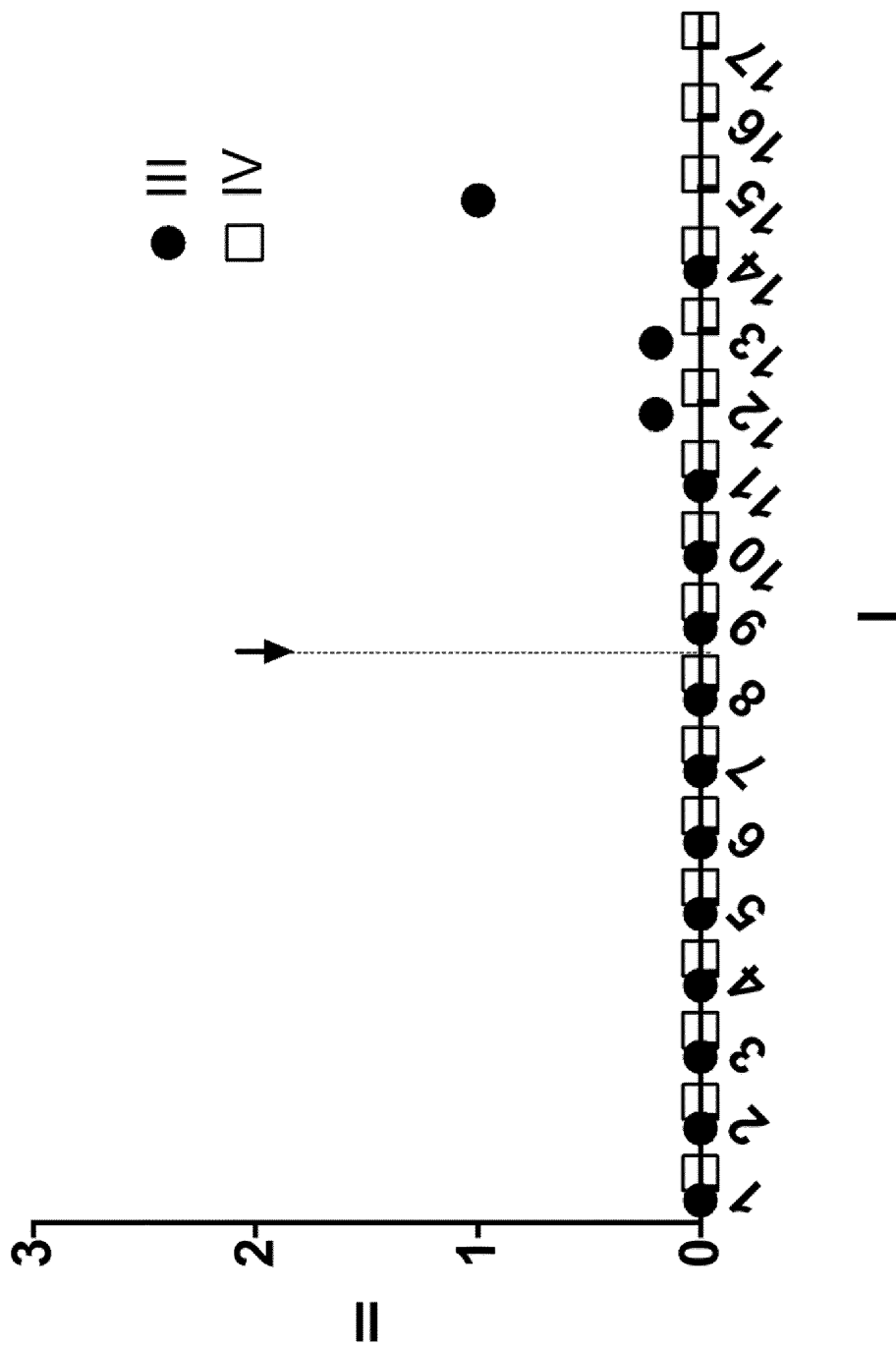
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Figure 30



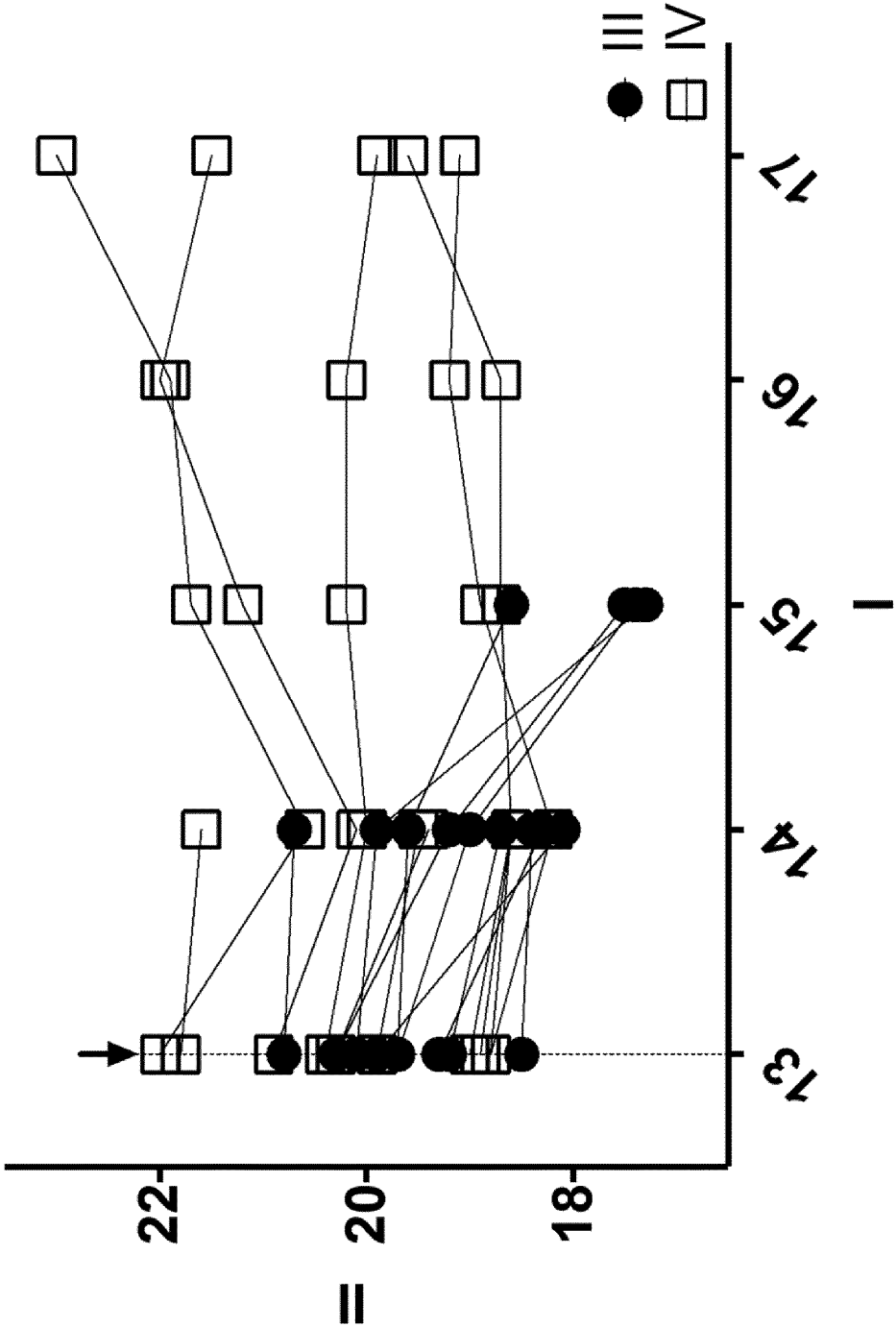
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Figure 31



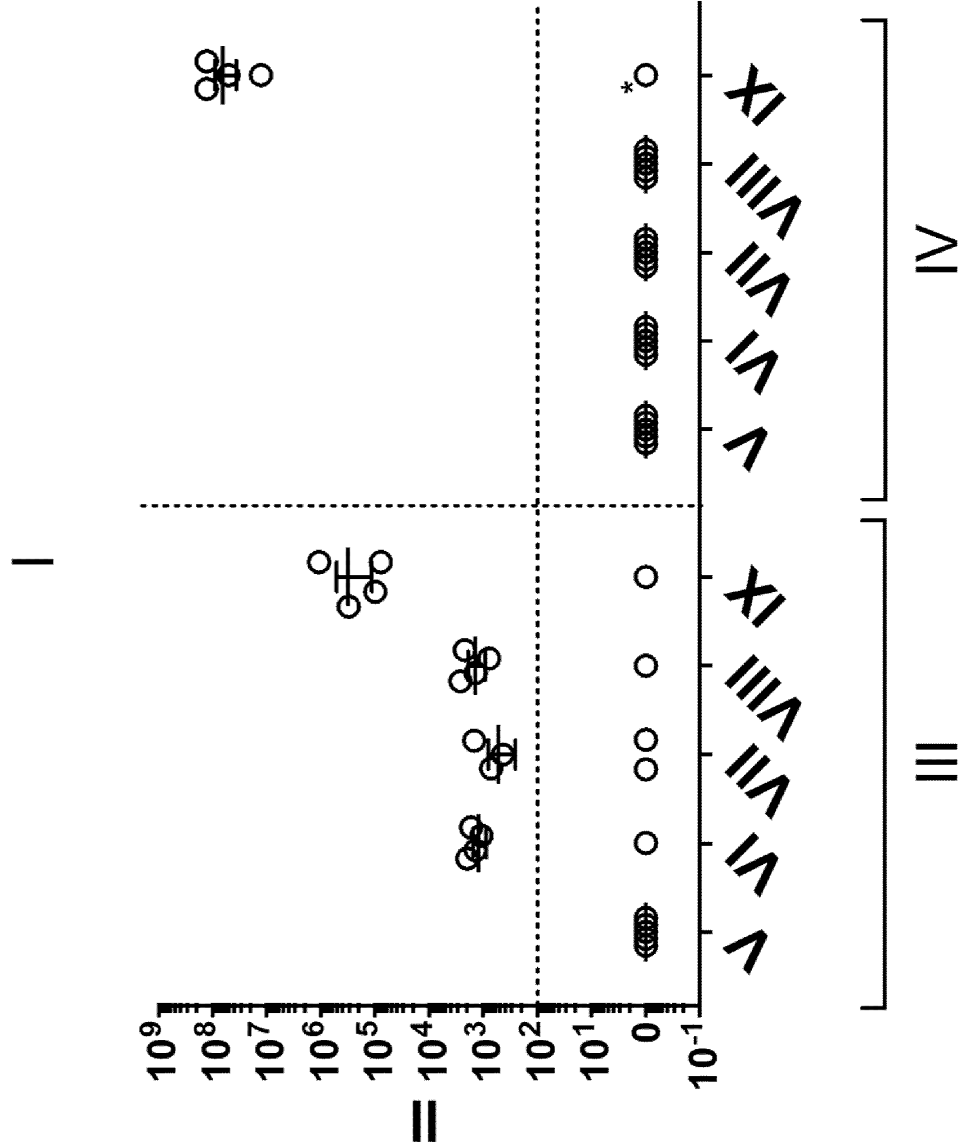
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Figure 32



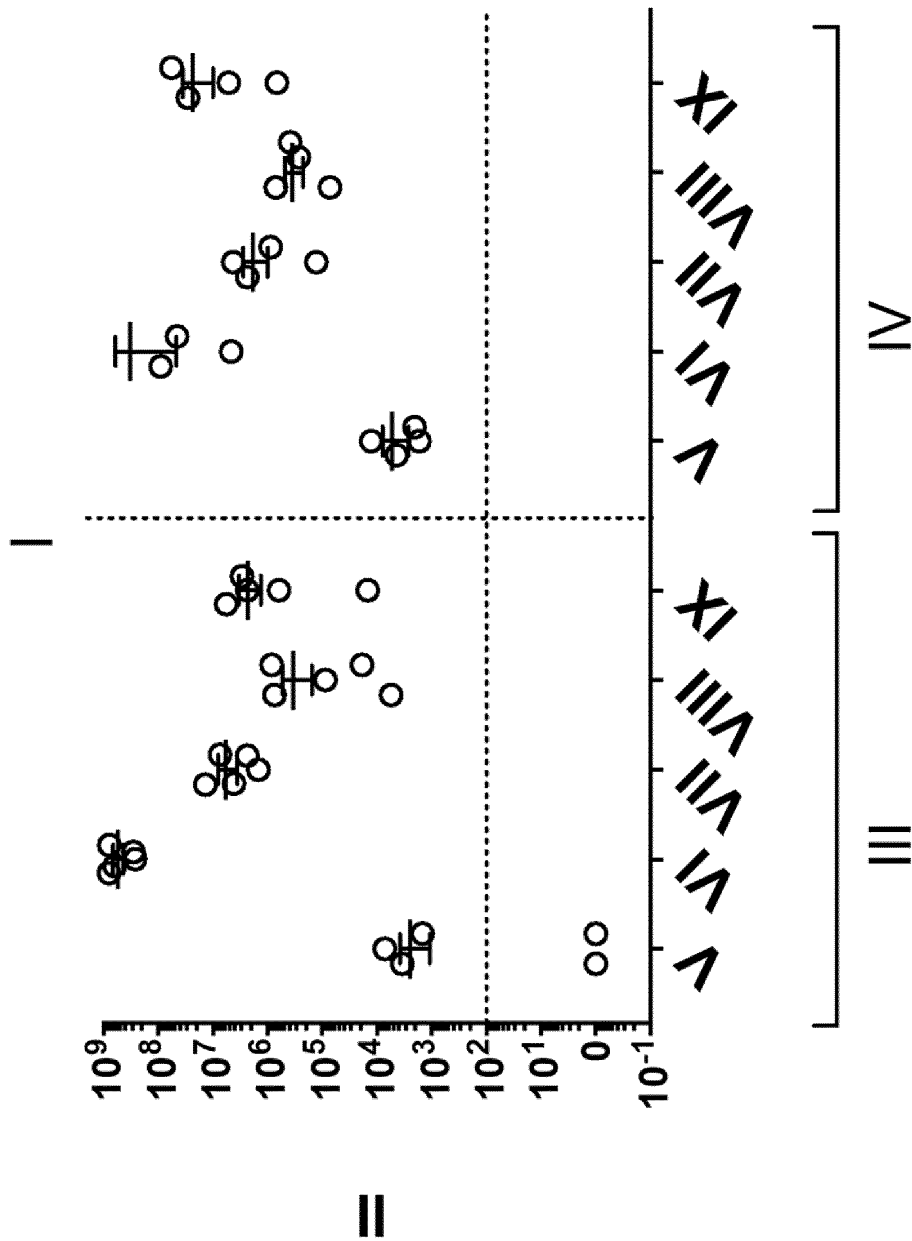
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Figure 33



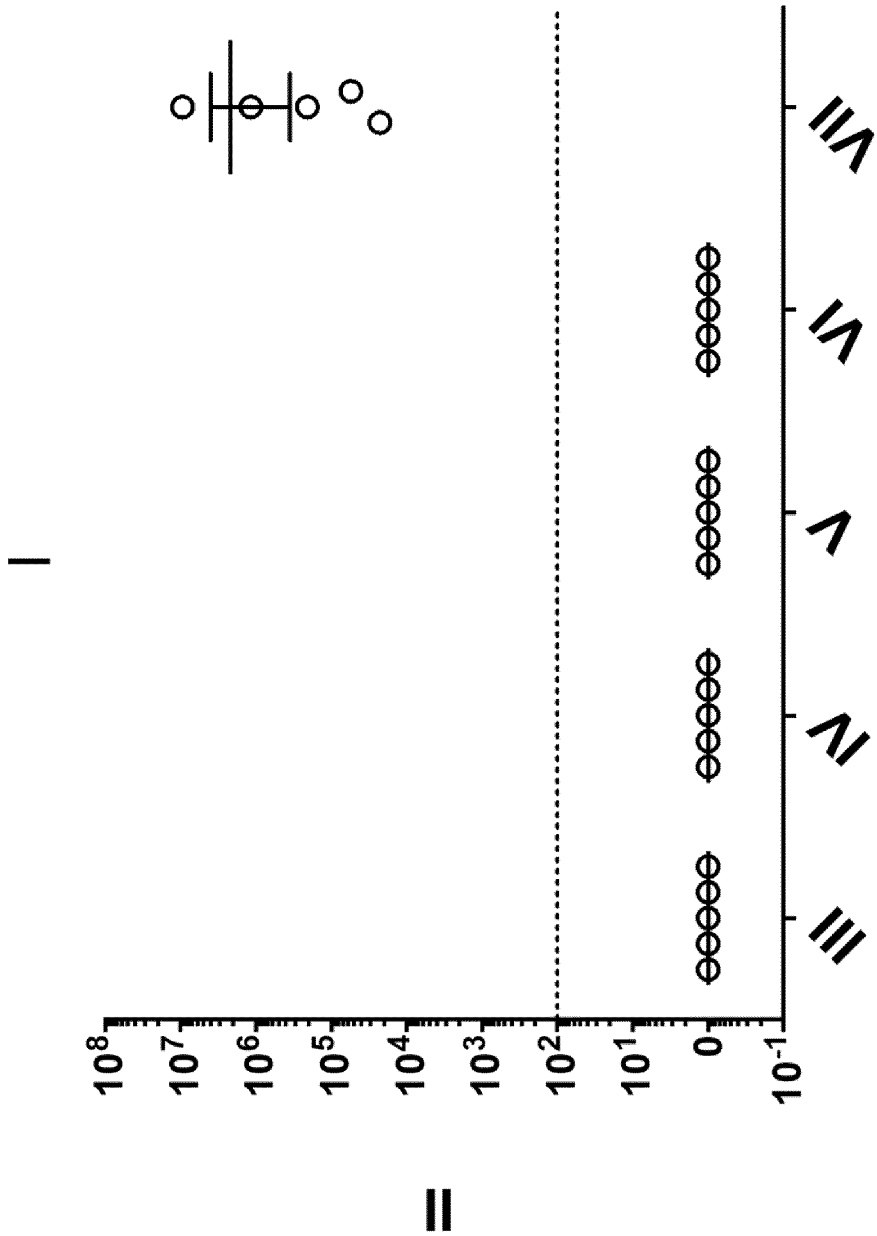
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Figure 34



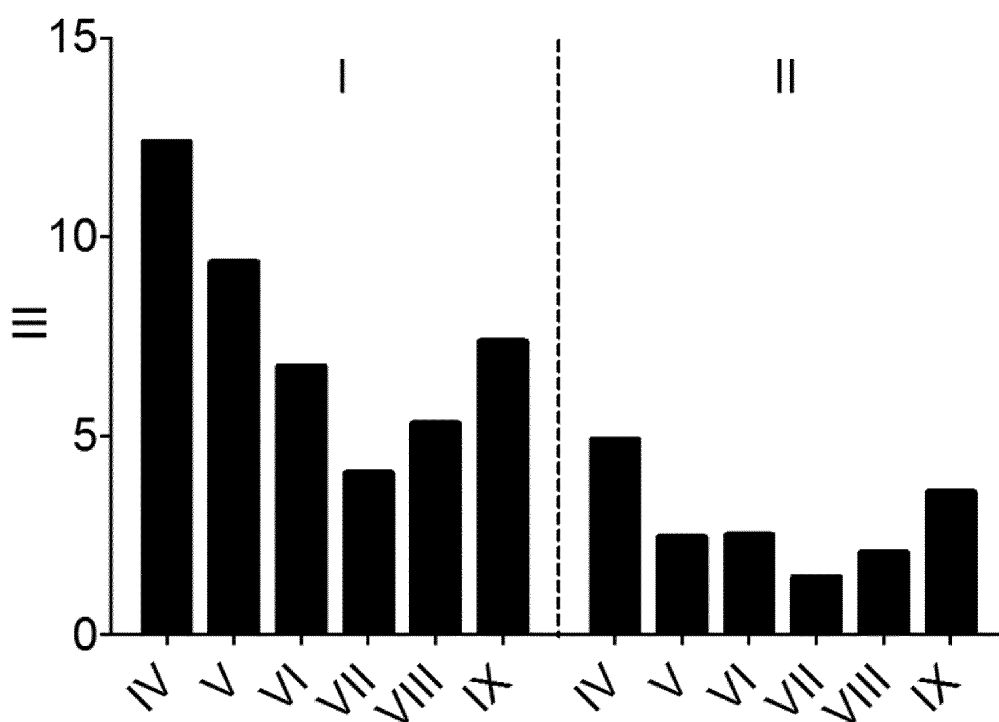
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Figure 35



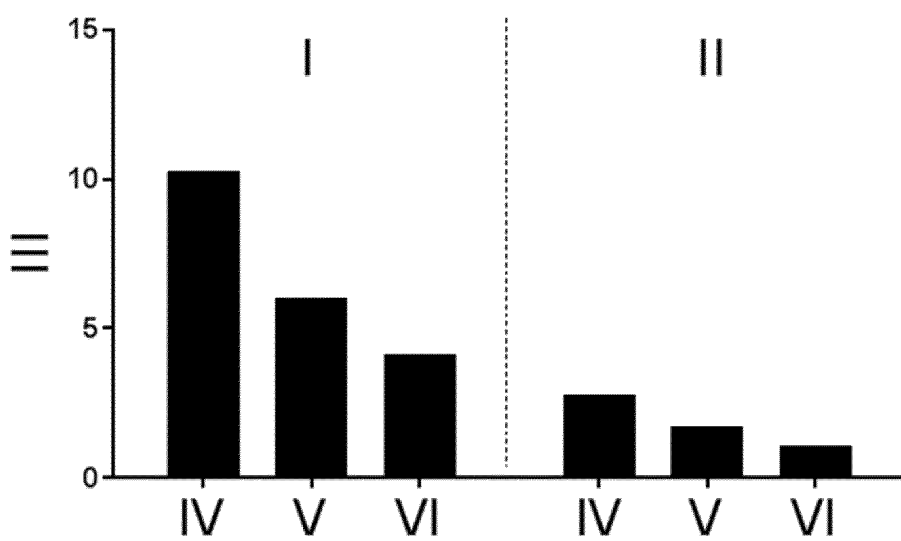
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Figure 36



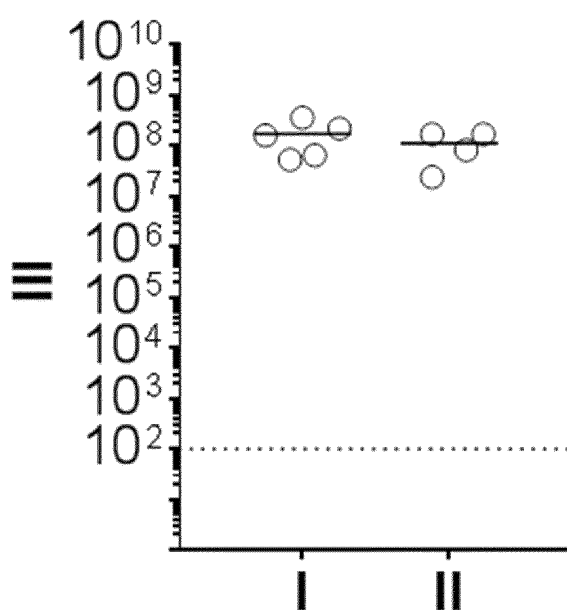
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Figure 37



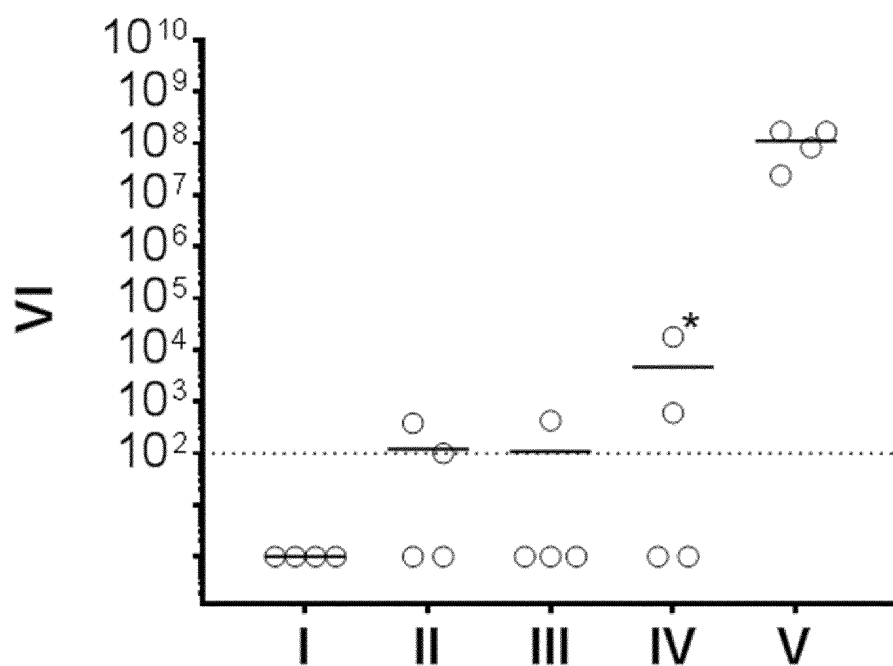
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Figure 38



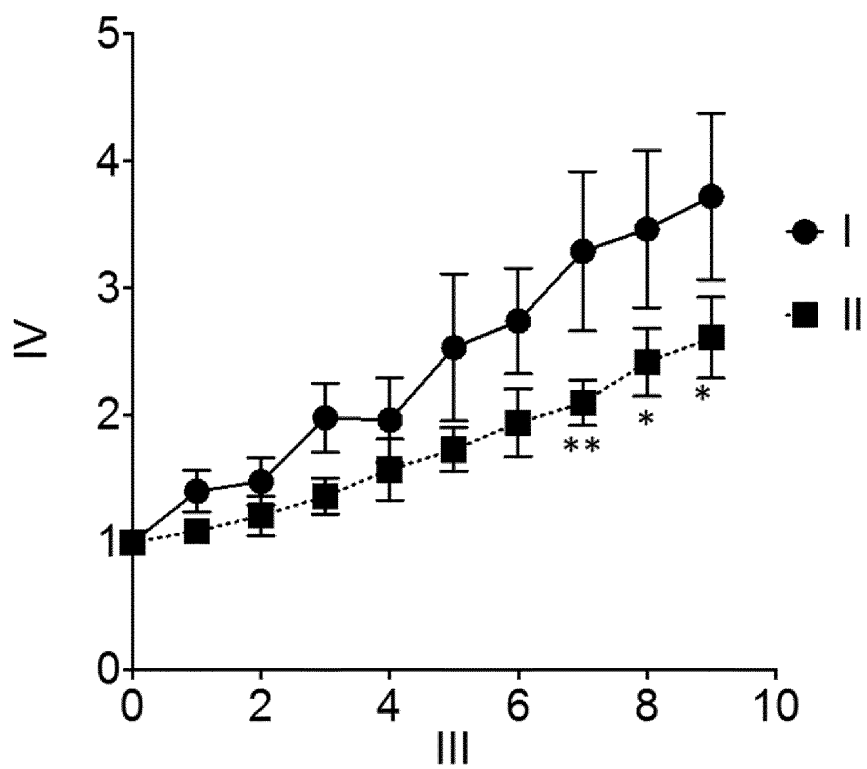
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Figure 39



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Figure 40



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Figure 41



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Figure 42

