

Supplementary

EGF-Coupled Gold Nanoparticles Increase the Expression of CNPase and the Myelin-Associated Proteins MAG, MOG, and MBP in the Septal Nucleus Demyelinated by Cuprizone

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Table S1. Descriptive statistics for rotarod, latency to fall.

8 rpm		15 rpm	
Group	Mean ± SE (seconds)	Group	Mean ± SE (seconds)
Ctrl	60 ± 0	Ctrl	60 ± 0
CPZ	60 ± 0	CPZ	54 ± 2.94
GNPs	60 ± 0	GNPs	56.43 ± 3.14
EGF	60 ± 0	EGF	51.75 ± 6.09
EGF-GNPs	58.06 ± 1.93	EGF-GNPs	57.87 ± 1.91
30 rpm		35 rpm	
Group	Mean ± SE (seconds)	Group	Mean ± SE (seconds)
Ctrl	54.9 ± 3.25	Ctrl	50.6 ± 4.23
CPZ	31.83 ± 5.12	CPZ	20.20 ± 5.68
GNPs	46 ± 5.36	GNPs	38.75 ± 7.70
EGF	33.87 ± 6.75	EGF	24.06 ± 6.30
EGF-GNPs	45.37 ± 4.96	EGF-GNPs	40.5 ± 6.51

Table S2. Rotarod test performance, statistical analysis for 3 WPI, Kruskal-Wallis H test.

Speed	Kruskal-Wallis test
8 rpm	$H = 0.314$
15 rpm	$H = 0.137$
30 rpm	$H = 0.026^*$
35 rpm	$H = 0.006^*$

Table S3. Rotarod test performance, statistical analysis for 3 WPI, Bonferroni correction for pair comparison.

Comparison	30 rpm	35 rpm
	p value	p value

Ctrl vs CPZ	<i>p</i> = 0.003*	<i>p</i> = 0.001*
Ctrl vs GNPs	<i>p</i> = 0.235	<i>p</i> = 0.244
Ctrl vs EGF	<i>p</i> = 0.010*	<i>p</i> = 0.005*
Ctrl vs EGF-GNPs	<i>p</i> = 0.267	<i>p</i> = 0.248
CPZ vs GNPs	<i>p</i> = 0.131	<i>p</i> = 0.055
CPZ vs EGF	<i>p</i> = 0.934	<i>p</i> = 0.840
CPZ vs EGF-GNPs	<i>p</i> = 0.111	<i>p</i> = 0.053
GNPs vs EGF	<i>p</i> = 0.192	<i>p</i> = 0.116
GNPs vs EGF-GNPs	<i>p</i> = 0.940	<i>p</i> = 0.993
EGF vs EGF-GNPs	<i>p</i> = 0.168	<i>p</i> = 0.114

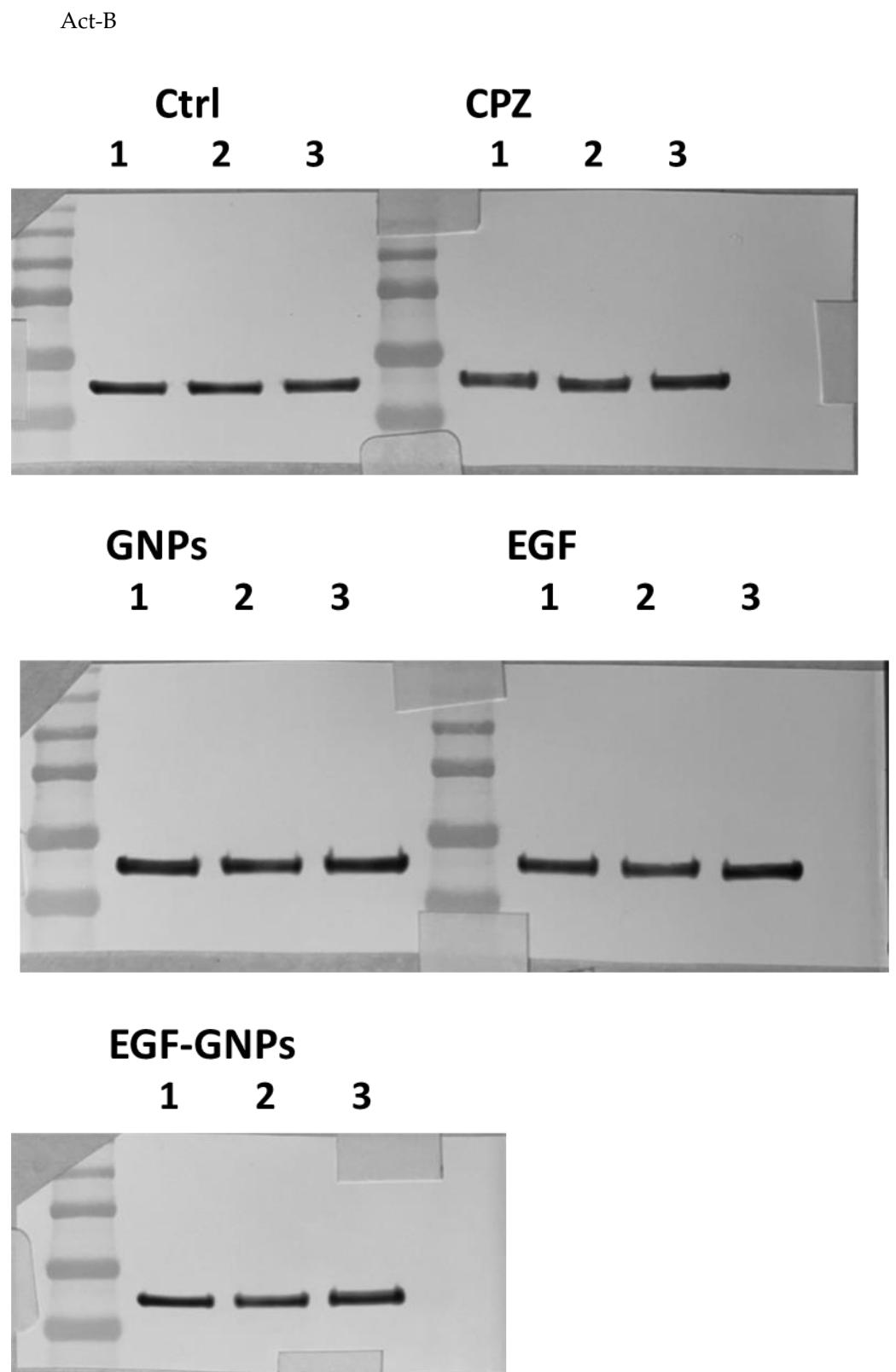
Table S4. Descriptive statistics for Western blot.

2 weeks post-injection		3 weeks post-injection	
CNPase		CNPase	
Group	Mean ± SE	Group	Mean ± SE
Ctrl	1.00 ± 0.03	Ctrl	1.00 ± 0.04
CPZ	0.81 ± 0.12	CPZ	0.94 ± 0.15
GNPs	1.29 ± 0.20	GNPs	0.97 ± 0.22
EGF	0.99 ± 0.12	EGF	0.69 ± 0.12
EGF-GNPs	1.26 ± 0.02	EGF-GNPs	1.09 ± 0.09
MAG		MAG	
Group	Mean ± SE	Group	Mean ± SE
Ctrl	1.00 ± 0.01	Ctrl	1.00 ± 0.06
CPZ	0.68 ± 0.19	CPZ	0.96 ± 0.06
GNPs	1.08 ± 0.19	GNPs	1.25 ± 0.15
EGF	0.82 ± 0.19	EGF	0.92 ± 0.13
EGF-GNPs	1.16 ± 0.05	EGF-GNPs	1.44 ± 0.06
MOG		MOG	
Group	Mean ± SE	Group	Mean ± SE
Ctrl	1.00 ± 0.17	Ctrl	1.00 ± 0.02
CPZ	0.32 ± 0.08	CPZ	1.21 ± 0.28
GNPs	0.90 ± 0.35	GNPs	1.30 ± 0.55
EGF	0.74 ± 0.33	EGF	0.72 ± 0.23
EGF-GNPs	0.95 ± 0.29	EGF-GNPs	1.36 ± 0.14
MBP		MBP	
Group	Mean ± SE	Group	Mean ± SE
Ctrl	1.00 ± 0.03	Ctrl	1.00 ± 0.09
CPZ	0.53 ± 0.32	CPZ	0.90 ± 0.27
GNPs	0.77 ± 0.31	GNPs	0.61 ± 0.08
EGF	0.53 ± 0.28	EGF	0.95 ± 0.23
EGF-GNPs	0.86 ± 0.06	EGF-GNPs	1.21 ± 0.26

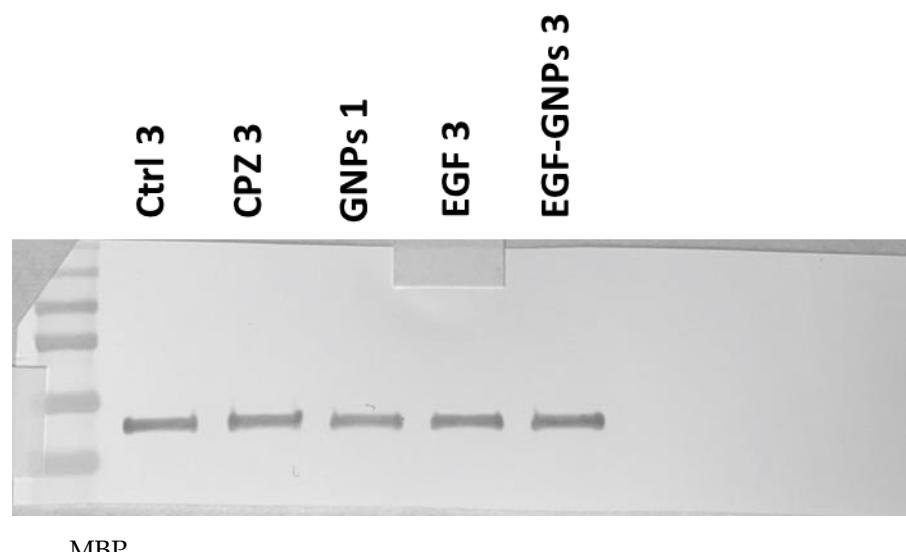
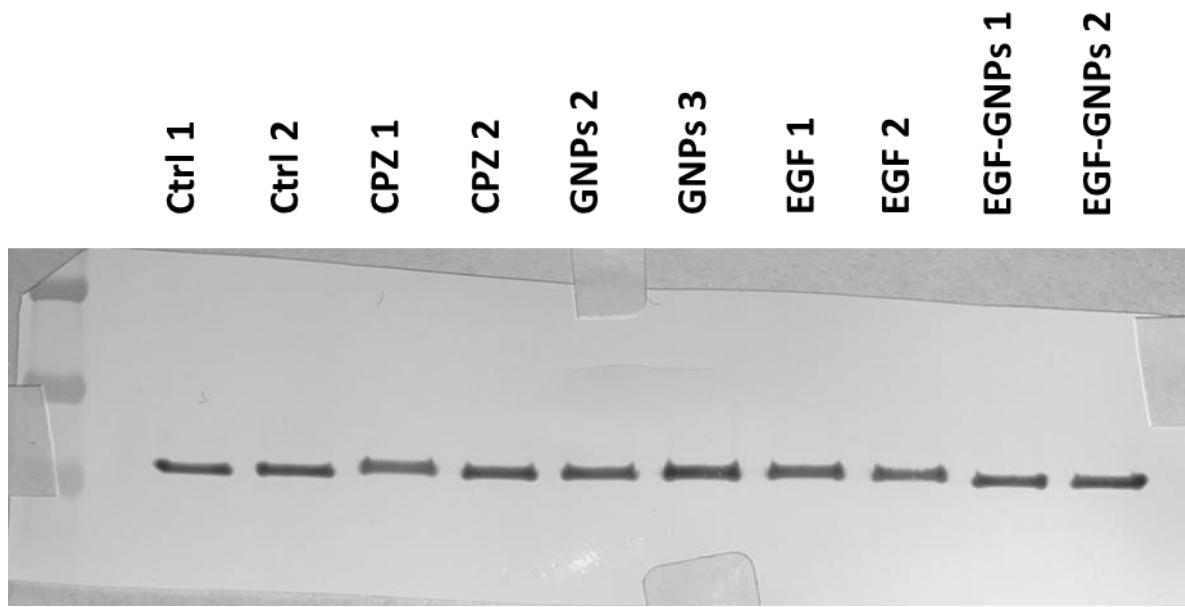
Table S5. Myelin proteins, statistical analysis for 2 WPI and 3 WPI, Mann-Whitney U test.

Comparison	2 WPI		CNPase		MAG		MOG		MBP	
	"U" value	<i>p</i> value	"U" value	<i>p</i> value	"U" value	<i>p</i> value	"U" value	<i>p</i> value	"U" value	<i>p</i> value
Ctrl vs CPZ	<i>U</i> = 3.000	<i>p</i> = 0.513	<i>U</i> = 2.000	<i>p</i> = 0.275	<i>U</i> = 0.000	<i>p</i> = 0.050*	<i>U</i> = 3.000	<i>p</i> = 0.513		

Ctrl vs GNPs	$U = 2.000$	$p = 0.275$	$U = 3.000$	$p = 0.513$	$U = 4.000$	$p = 0.827$	$U = 3.000$	$p = 0.513$
Ctrl vs EGF	$U = 3.000$	$p = 0.513$						
Ctrl vs EGF-GNPs	$U = 0.000$	$p = \textbf{0.050*}$	$U = 0.000$	$p = \textbf{0.050*}$	$U = 4.000$	$p = 0.827$	$U = 1.000$	$p = 0.127$
CPZ vs GNPs	$U = 1.000$	$p = 0.127$	$U = 2.000$	$p = 0.275$	$U = 2.000$	$p = 0.275$	$U = 2.000$	$p = 0.275$
CPZ vs EGF	$U = 2.000$	$p = 0.275$	$U = 3.000$	$p = 0.513$	$U = 2.000$	$p = 0.275$	$U = 4.000$	$p = 0.827$
CPZ vs EGF-GNPs	$U = 0.000$	$p = \textbf{0.050*}$	$U = 0.000$	$p = \textbf{0.050*}$	$U = 0.000$	$p = \textbf{0.050*}$	$U = 3.000$	$p = 0.513$
GNPs vs EGF	$U = 2.000$	$p = 0.275$	$U = 3.000$	$p = 0.513$	$U = 4.000$	$p = 0.827$	$U = 2.000$	$p = 0.275$
GNPs vs EGF-GNPs	$U = 3.000$	$p = 0.513$	$U = 3.000$	$p = 0.513$	$U = 4.000$	$p = 0.827$	$U = 3.000$	$p = 0.513$
EGF vs EGF-GNPs	$U = 0.000$	$p = \textbf{0.050*}$	$U = 1.000$	$p = 0.127$	$U = 2.000$	$p = 0.275$	$U = 3.000$	$p = 0.513$
3 WPI	CNPase	MAG	MOG	MBP				
Comparison	"U" value	p value						
Ctrl vs CPZ	$U = 3.000$	$p = 0.513$	$U = 2.000$	$p = 0.275$	$U = 3.000$	$p = 0.513$	$U = 4.000$	$p = 0.827$
Ctrl vs GNPs	$U = 3.000$	$p = 0.513$	$U = 2.000$	$p = 0.275$	$U = 4.000$	$p = 0.827$	$U = 0.000$	$p = \textbf{0.050*}$
Ctrl vs EGF	$U = 1.000$	$p = 0.127$	$U = 2.000$	$p = 0.275$	$U = 3.000$	$p = 0.513$	$U = 4.000$	$p = 0.827$
Ctrl vs EGF-GNPs	$U = 3.000$	$p = 0.513$	$U = 0.000$	$p = \textbf{0.050*}$	$U = 0.000$	$p = \textbf{0.050*}$	$U = 4.000$	$p = 0.827$
CPZ vs GNPs	$U = 4.000$	$p = 0.827$	$U = 1.000$	$p = 0.127$	$U = 4.000$	$p = 0.827$	$U = 2.000$	$p = 0.275$
CPZ vs EGF	$U = 2.000$	$p = 0.275$	$U = 4.000$	$p = 0.827$	$U = 1.000$	$p = 0.127$	$U = 4.000$	$p = 0.827$
CPZ vs EGF-GNPs	$U = 3.000$	$p = 0.513$	$U = 0.000$	$p = \textbf{0.050*}$	$U = 4.000$	$p = 0.827$	$U = 2.000$	$p = 0.275$
GNPs vs EGF	$U = 3.000$	$p = 0.513$	$U = 0.000$	$p = \textbf{0.050*}$	$U = 3.000$	$p = 0.513$	$U = 2.000$	$p = 0.275$
GNPs vs EGF-GNPs	$U = 3.000$	$p = 0.513$	$U = 2.000$	$p = 0.275$	$U = 3.000$	$p = 0.513$	$U = 0.000$	$p = \textbf{0.050*}$
EGF vs EGF-GNPs	$U = 1.000$	$p = 0.127$	$U = 0.000$	$p = \textbf{0.050*}$	$U = 1.000$	$p = 0.127$	$U = 4.000$	$p = 0.827$



ACT-B: duplicates used as representative images in the main text



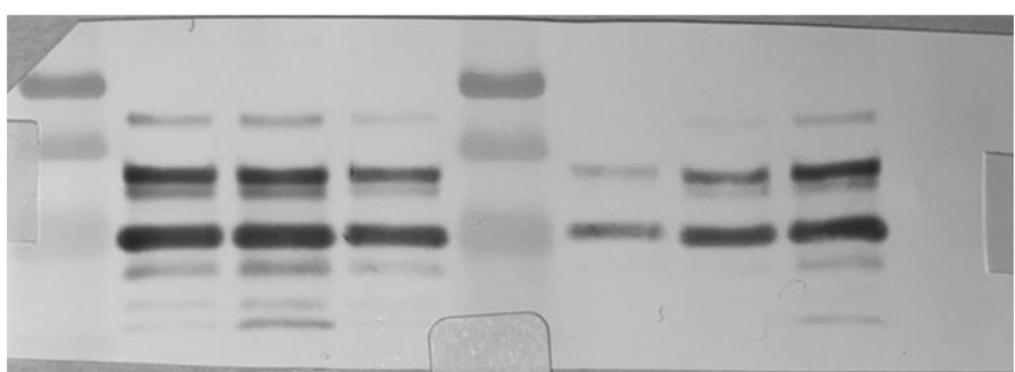
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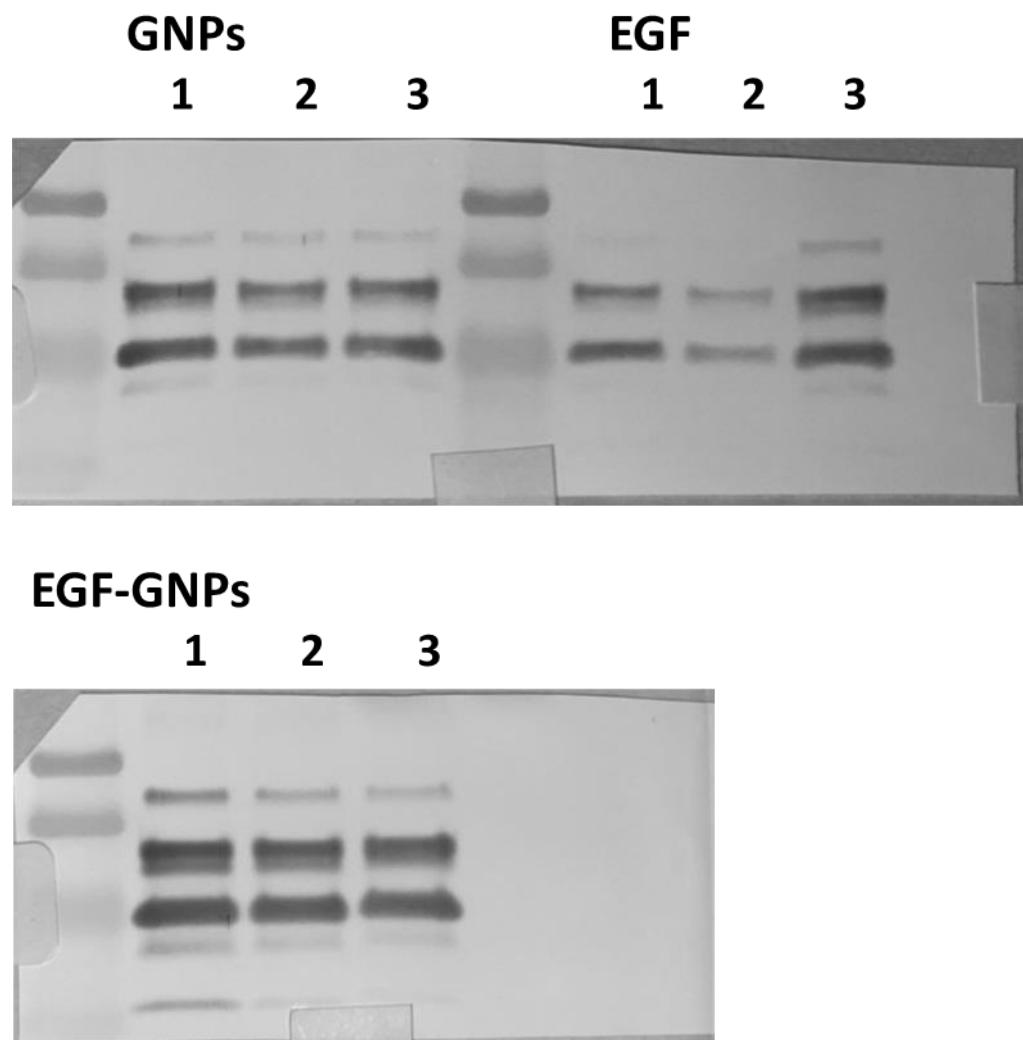
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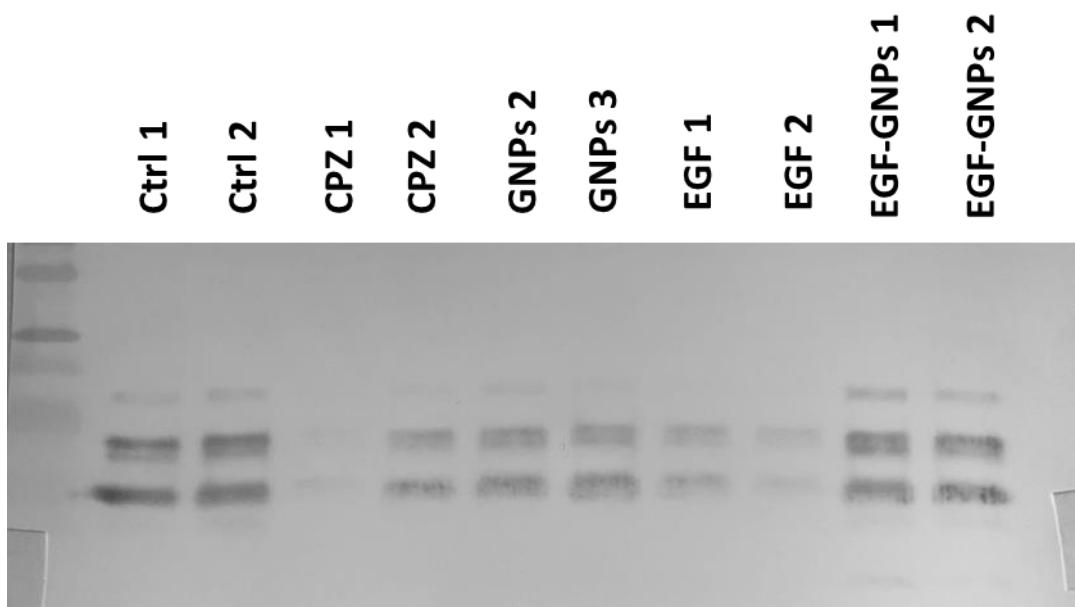
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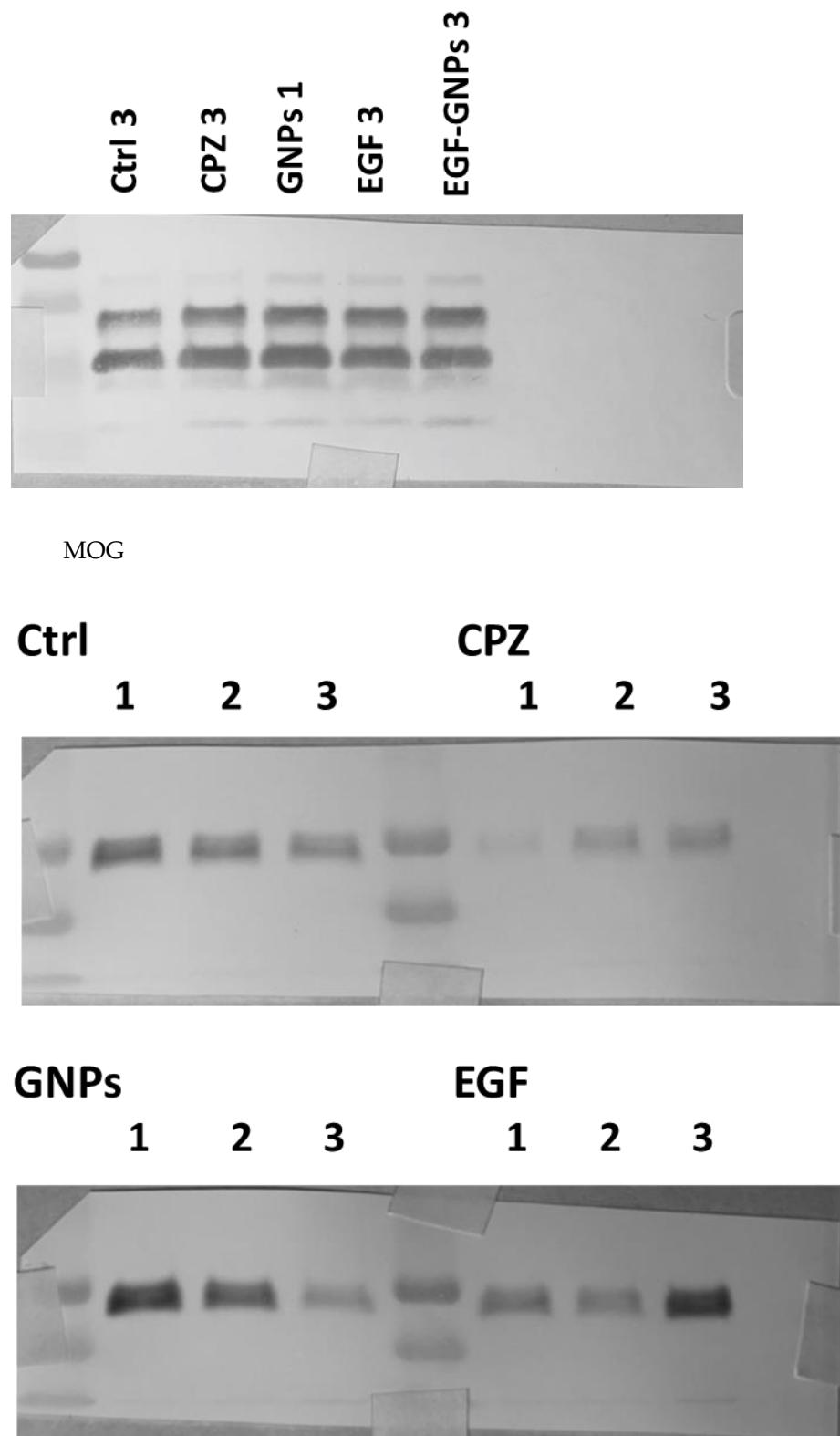
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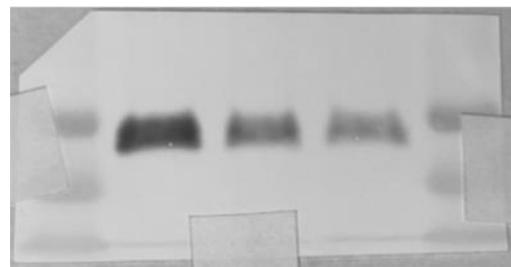
MBP: duplicates used as representative images in the main text





EGF-GNPs

1 2 3



MOG: duplicates used as representative images in the main text

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CPZ 2

GNPs 2

GNPs 3

EGF 1

EGF 2

EGF-GNPs 1

EGF-GNPs 2



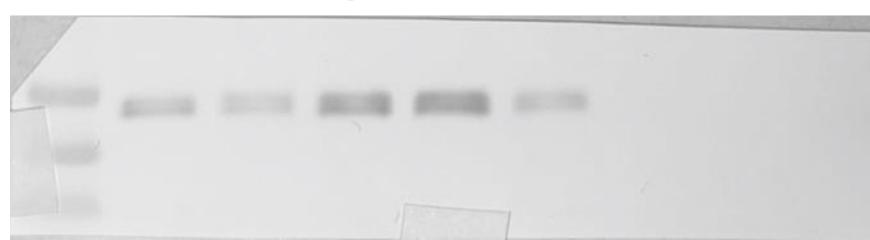
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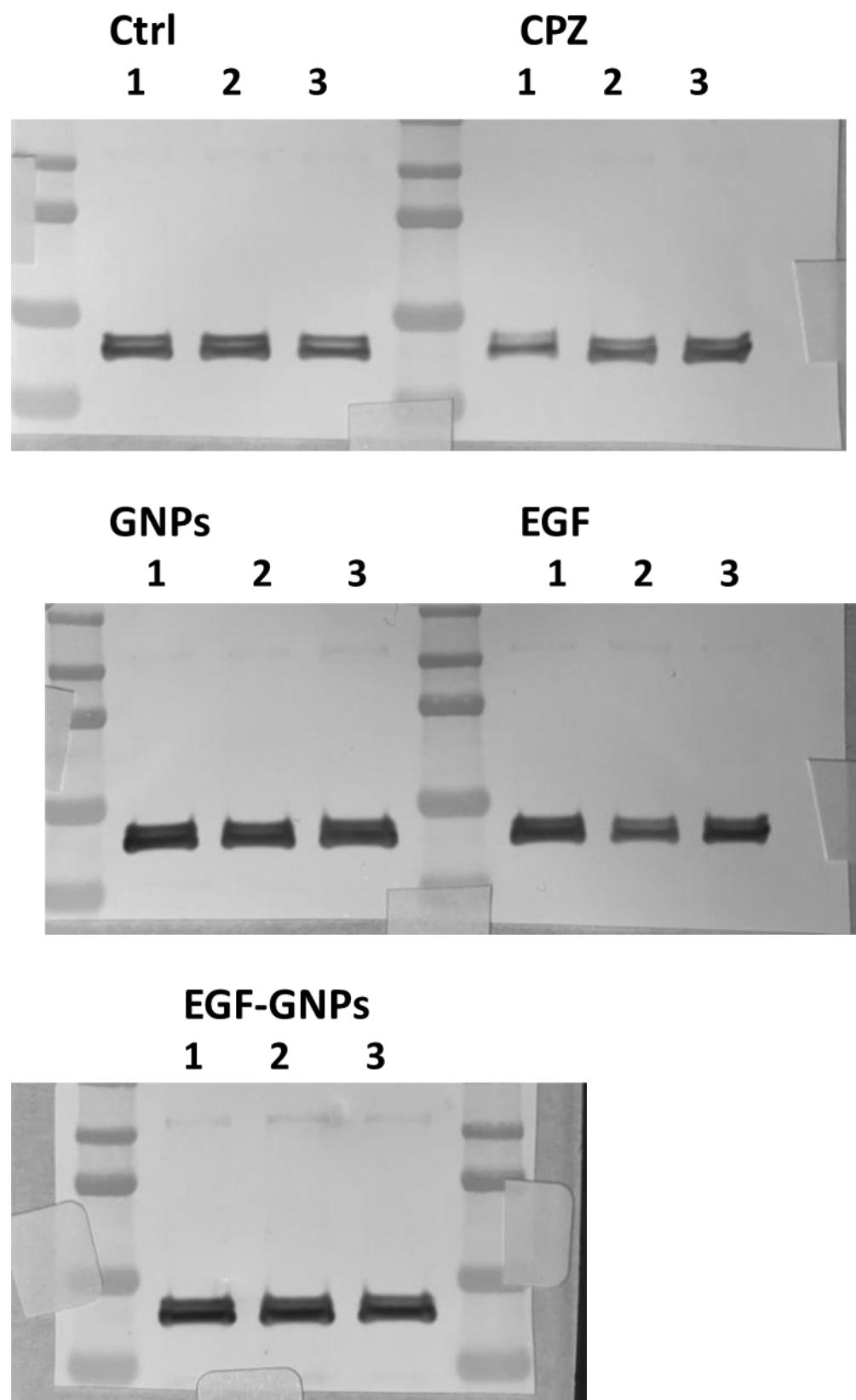
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EGF 3

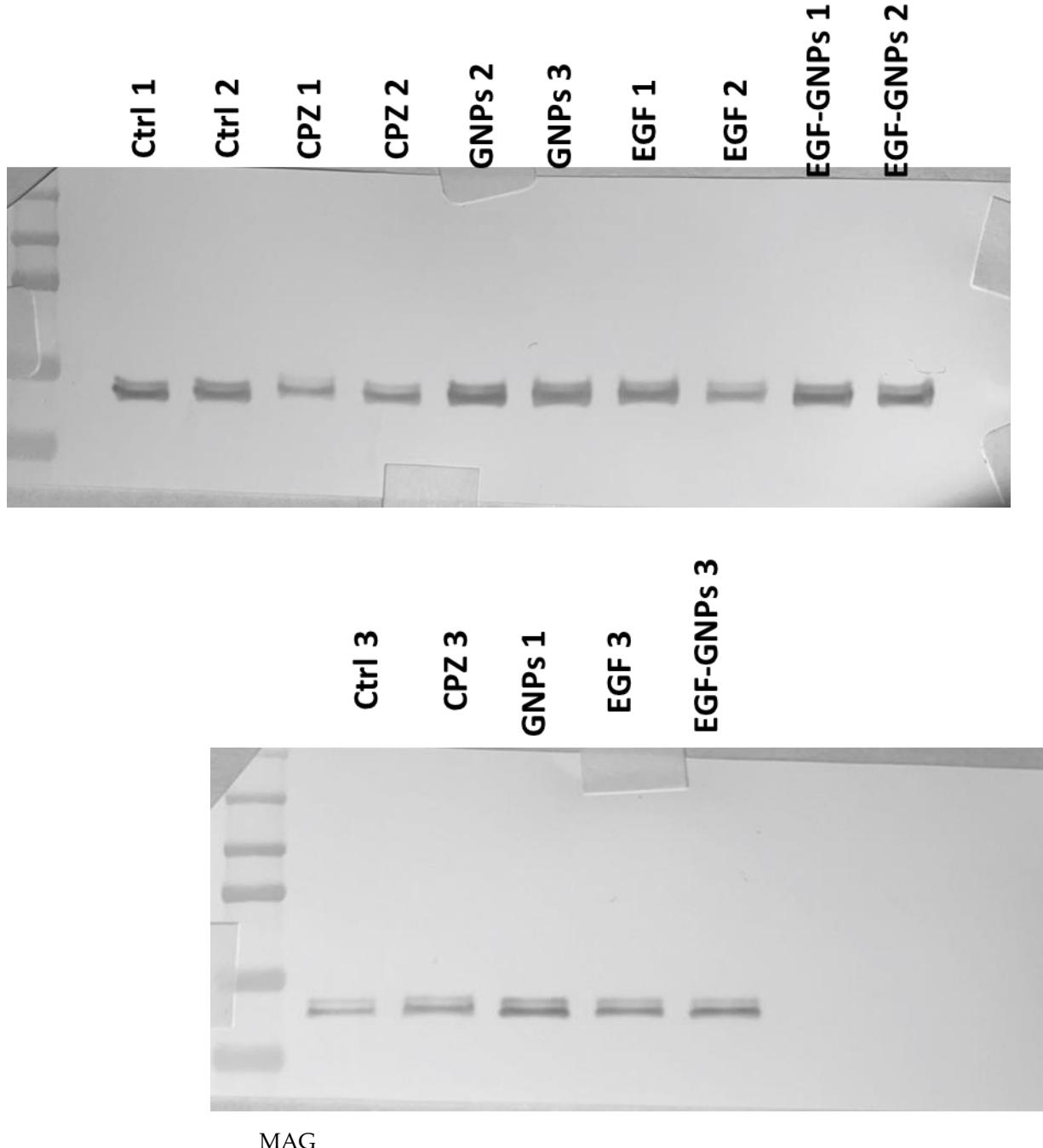
EGF-GNPs 3

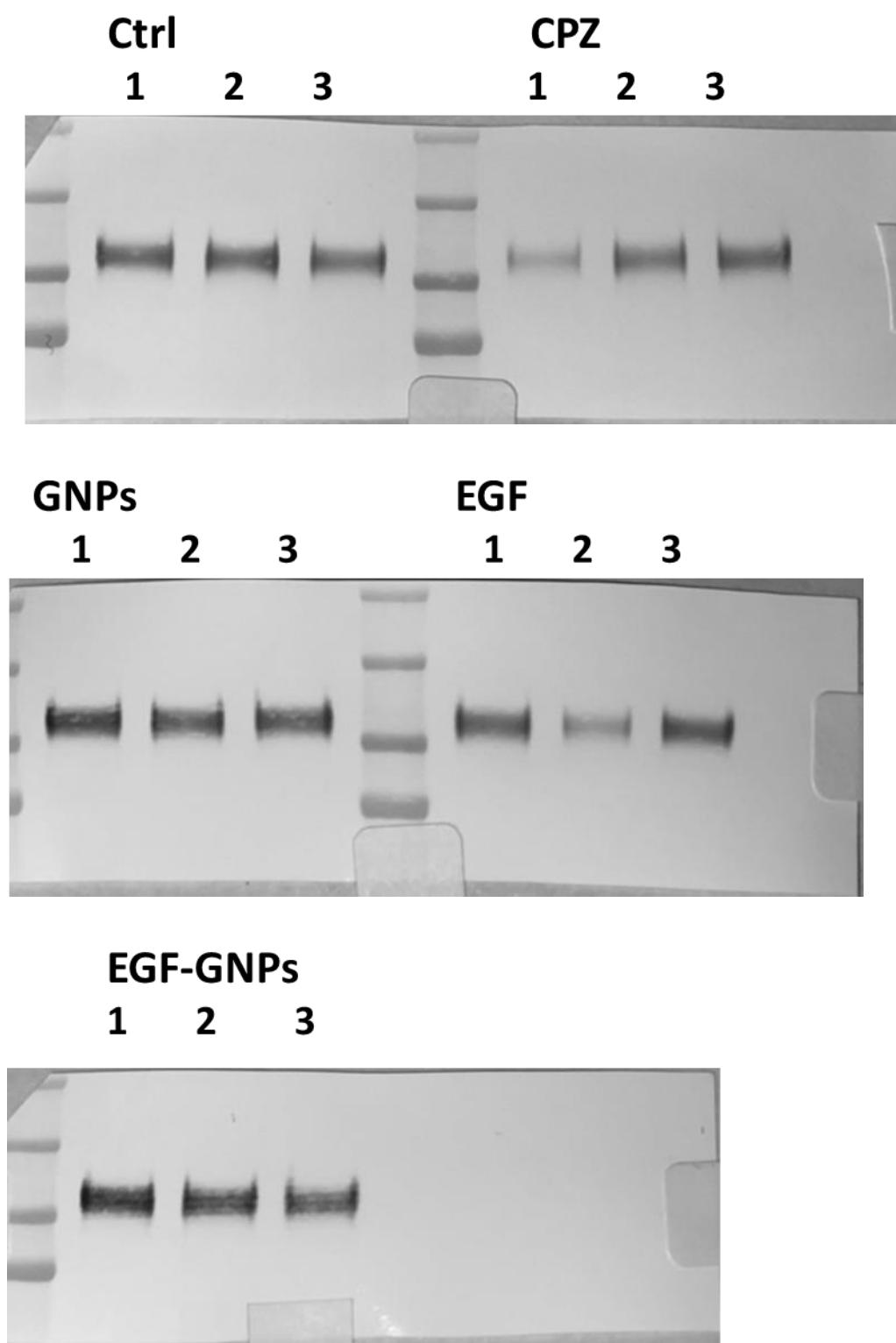


CNPase



CNPase: duplicates used as representative images in the main text





MAG: duplicates used as representative images in the main text

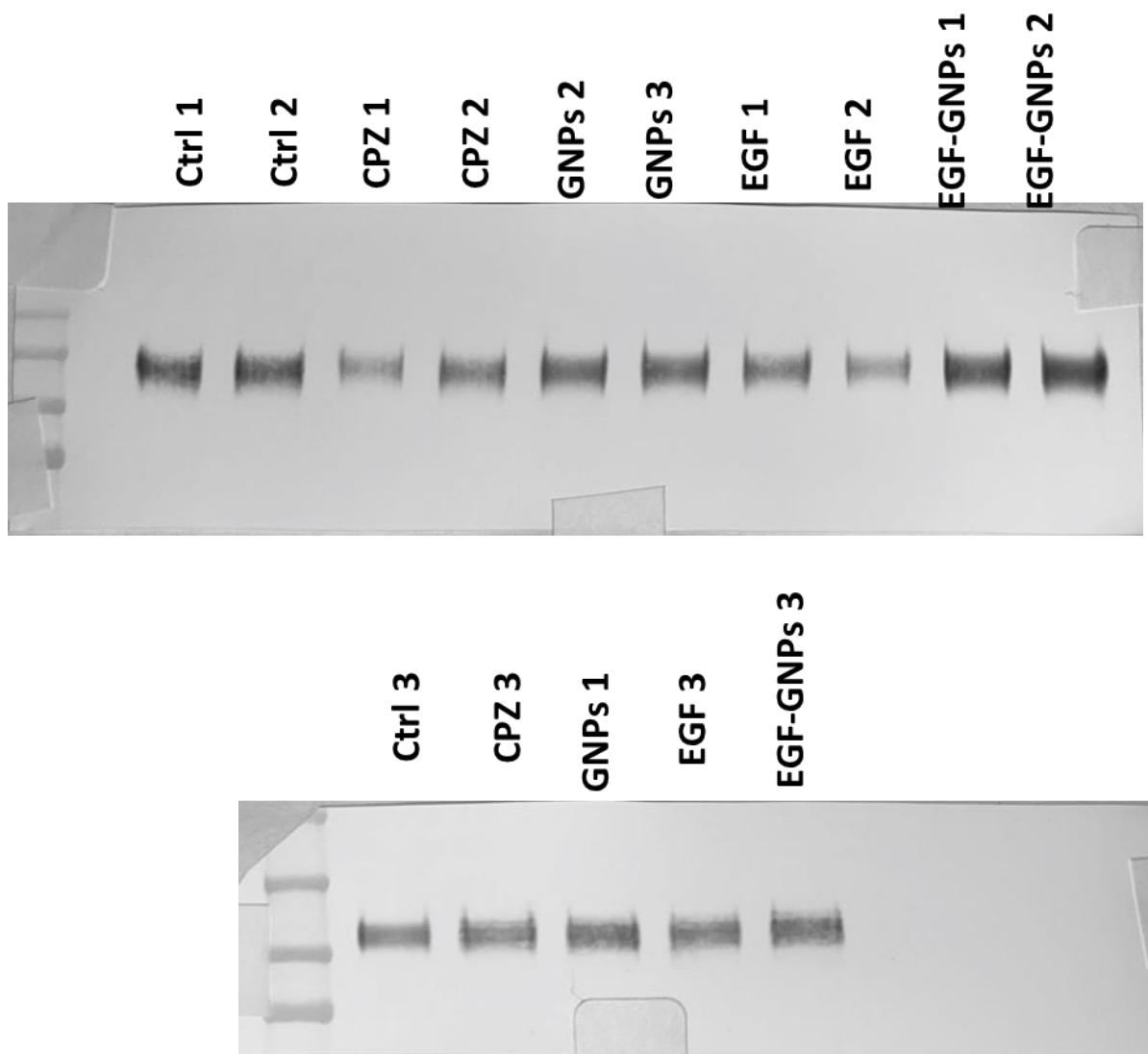
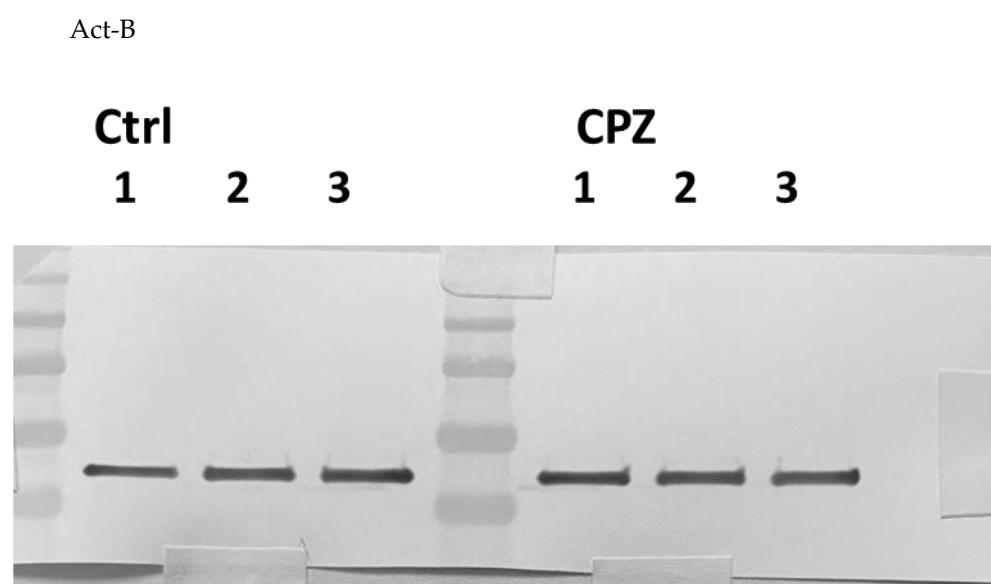
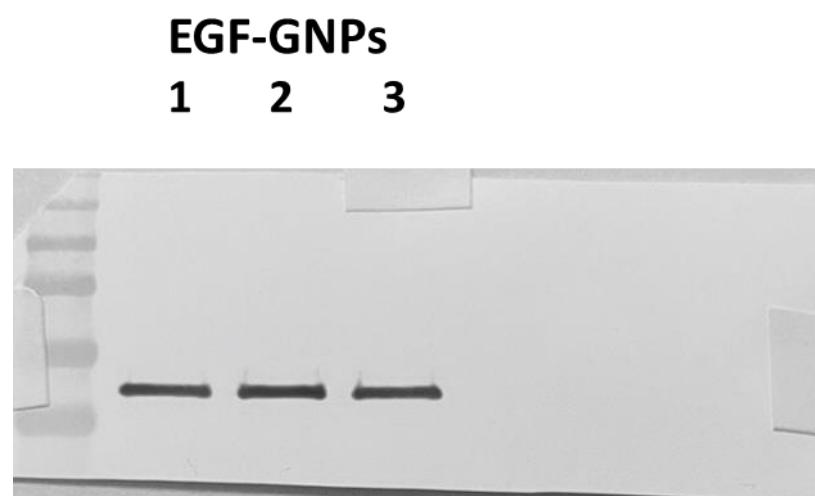
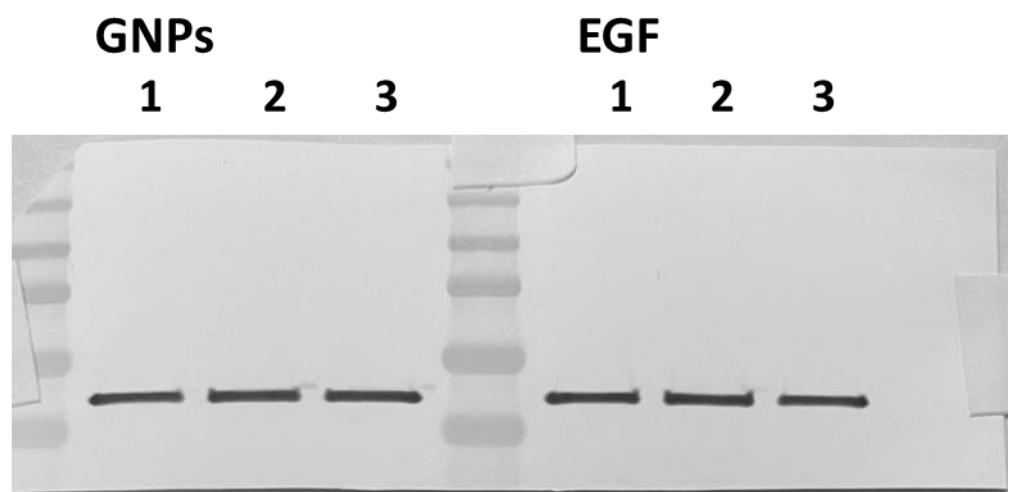
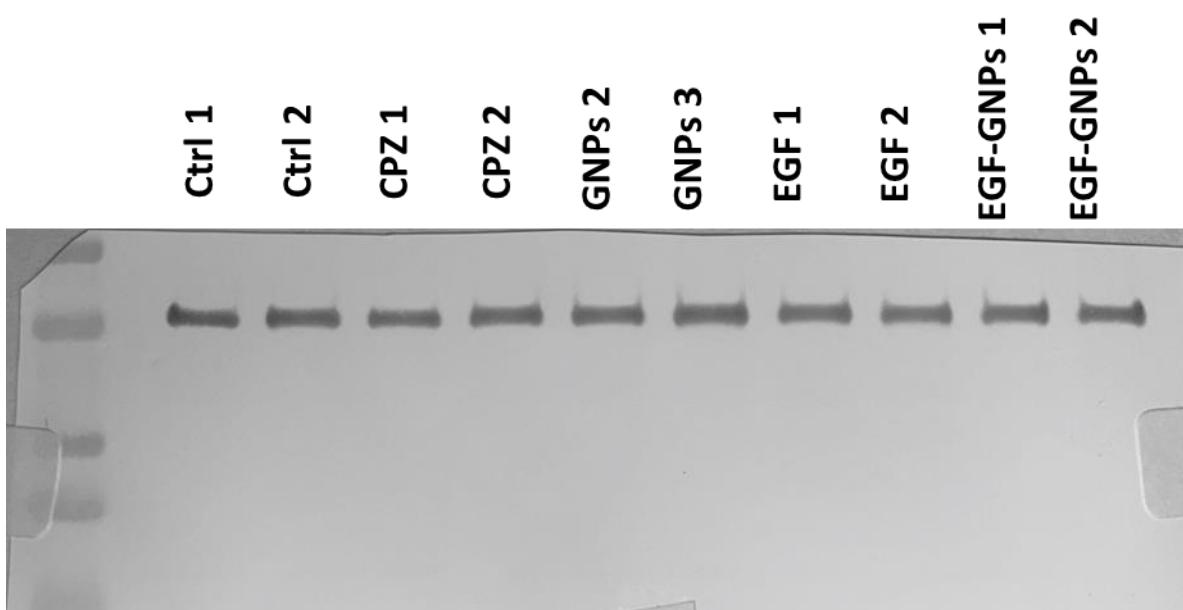


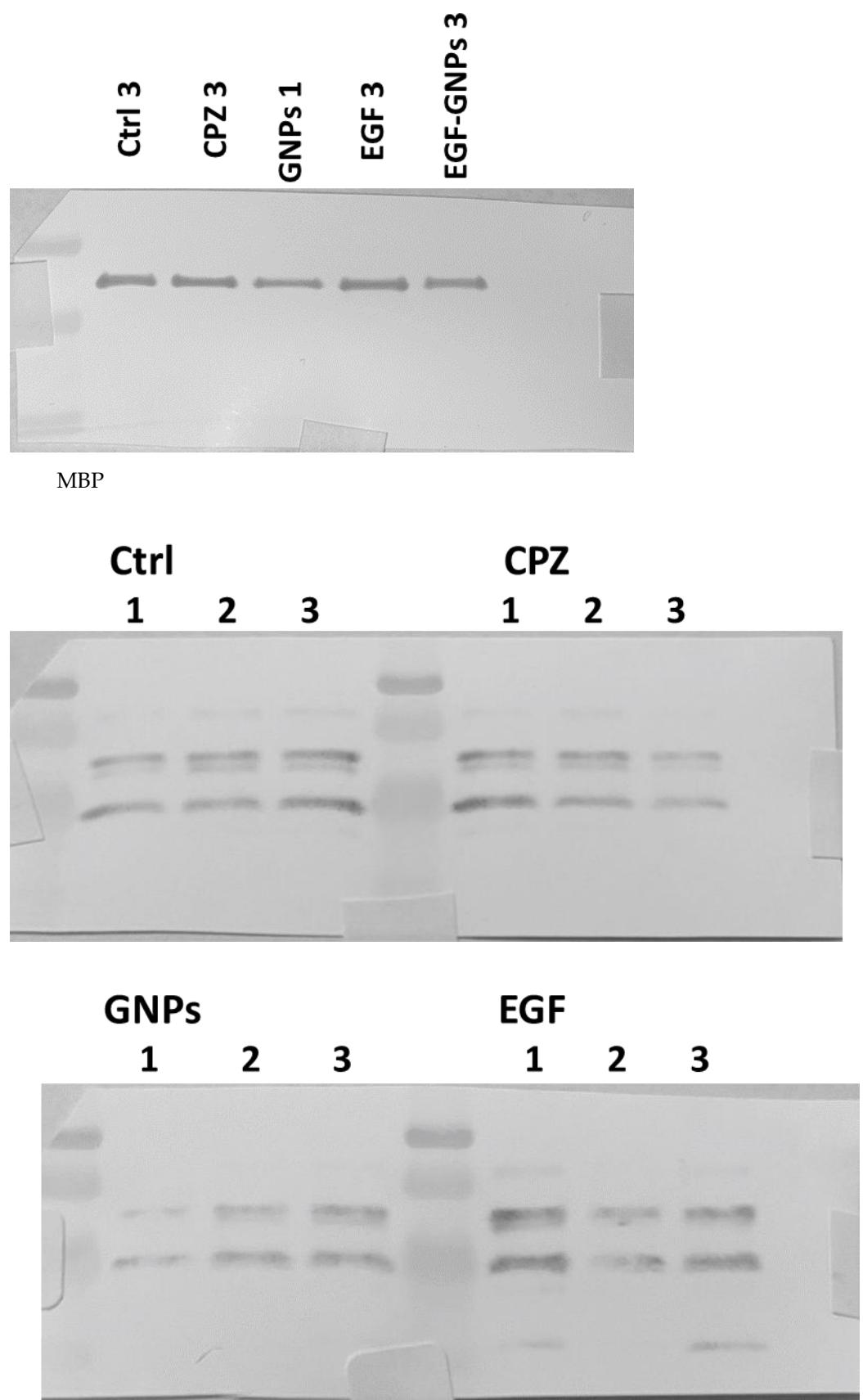
Figure S1. Blots obtained at 2 weeks of recovery. Every subject for each group was assigned with a number from 1 to 3. The duplicates were organized in a different arrangement, we used the numbers to identify each animal in every blot.





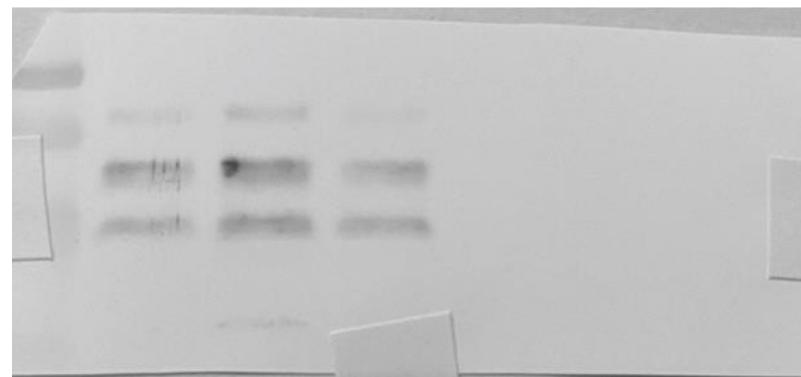
Act-B: duplicates used as representative images in the main text.





EGF-GNPs

1 2 3



MBP: duplicates used as representative images in the main text

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Ctrl 2

CPZ 1

CPZ 2

GNPs 2

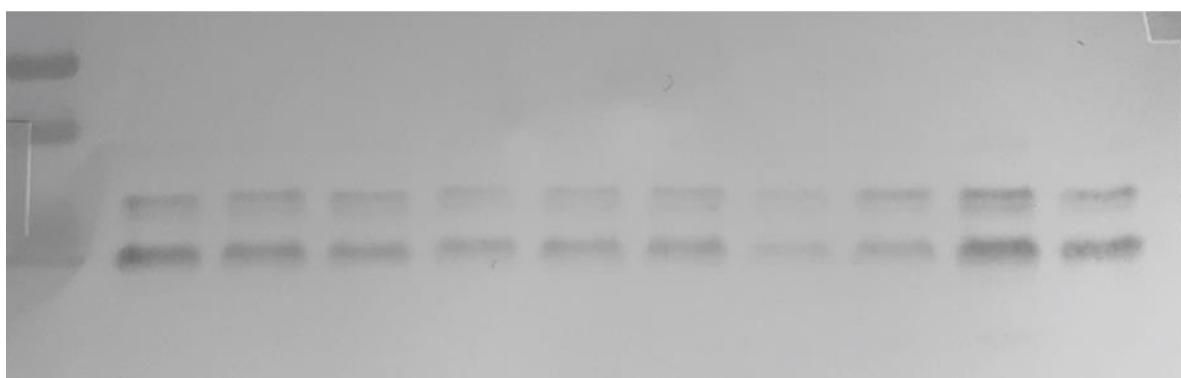
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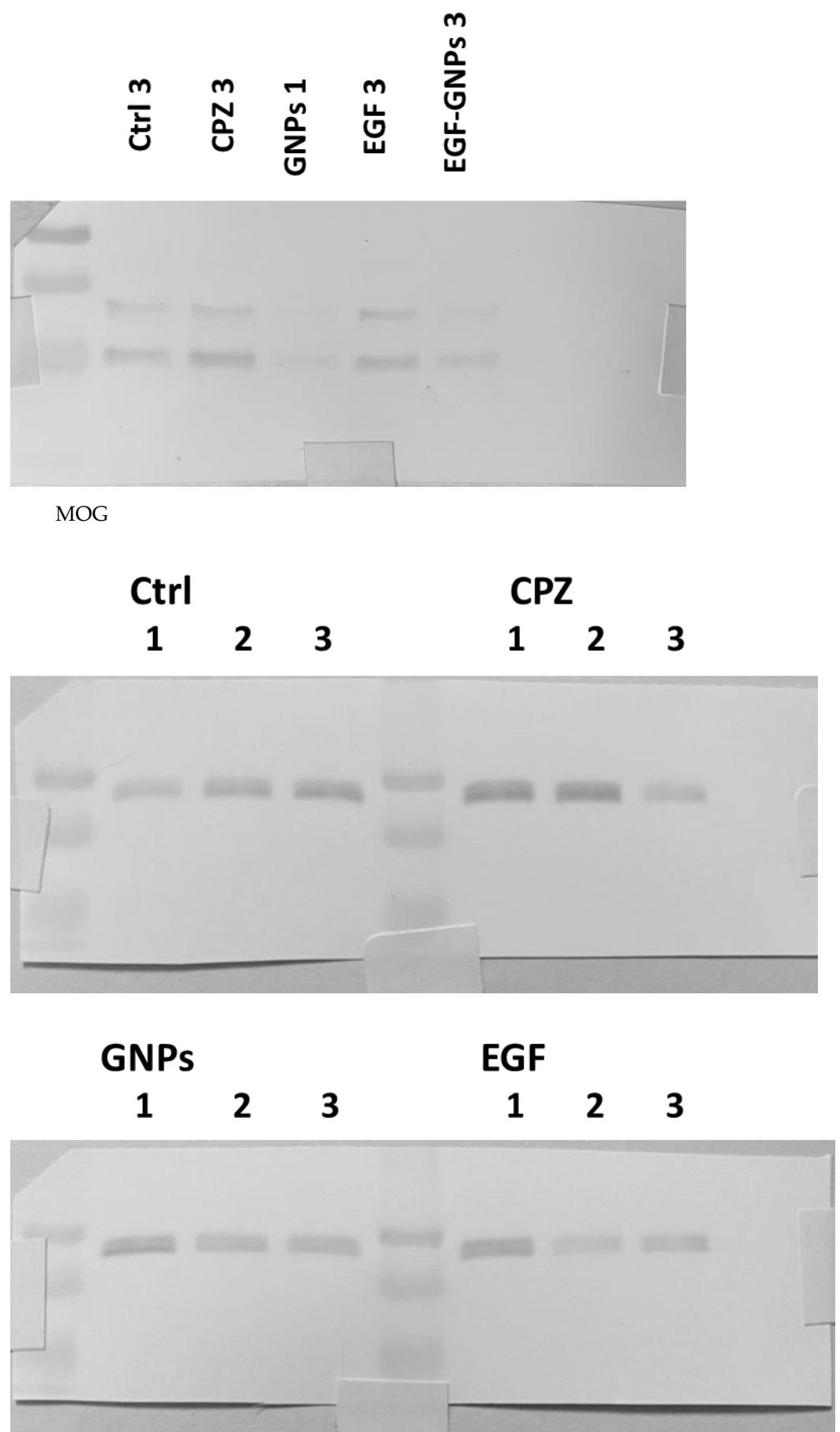
EGF 1

EGF 2

EGF-GNPs 1

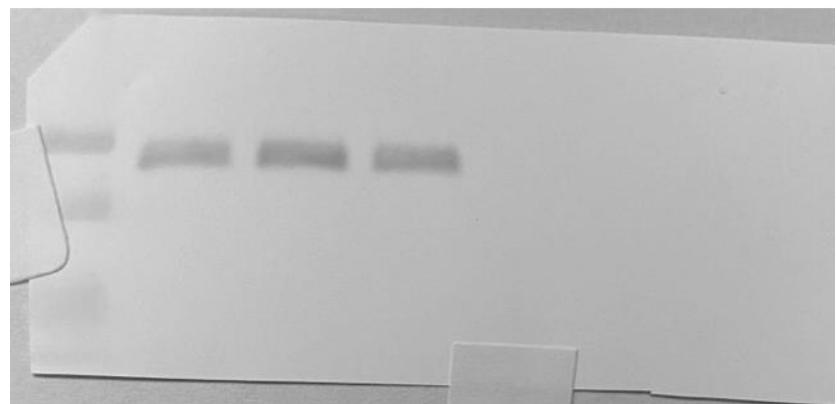
EGF-GNPs 2





EGF-GNPs

1 2 3



MOG: duplicates used as representative images in the main text

Ctrl 1

Ctrl 2

CPZ 1

CPZ 2

GNPs 2

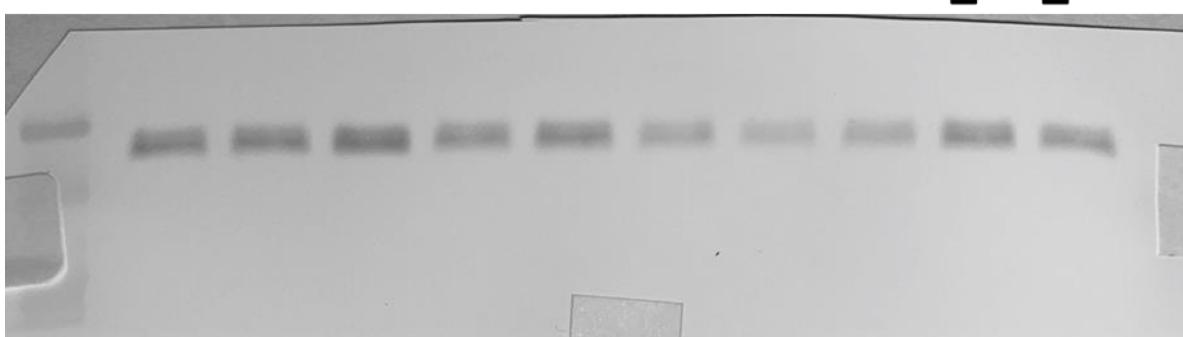
GNPs 3

EGF 1

EGF 2

EGF-GNPs 1

EGF-GNPs 2



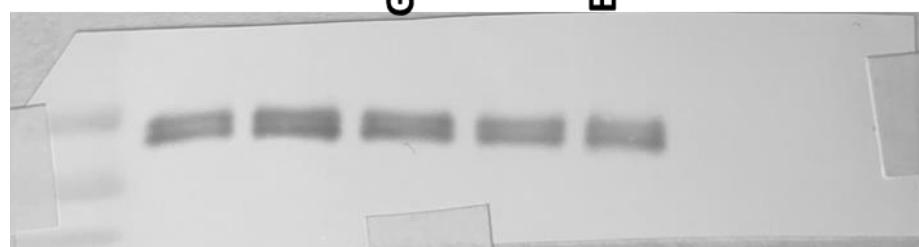
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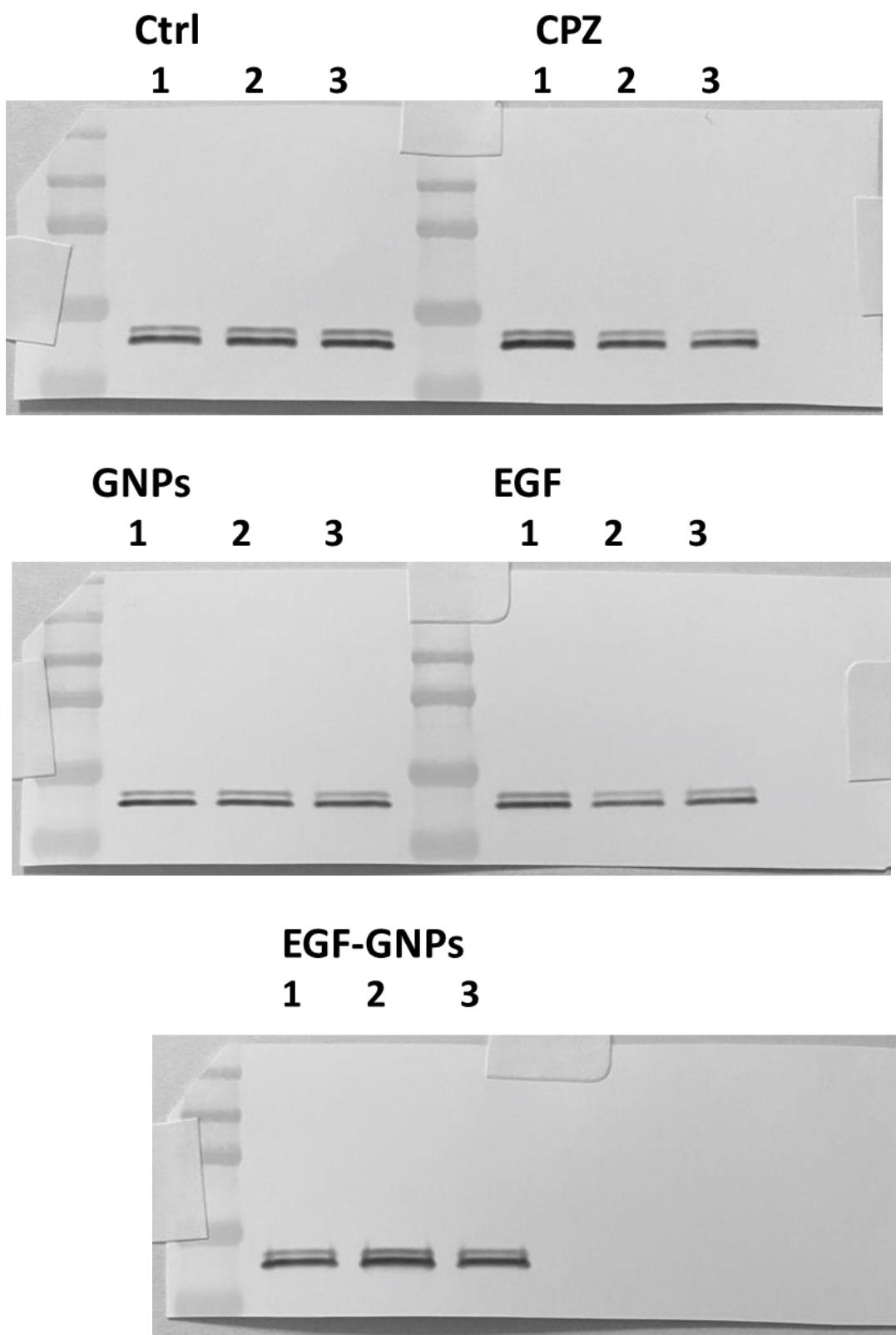
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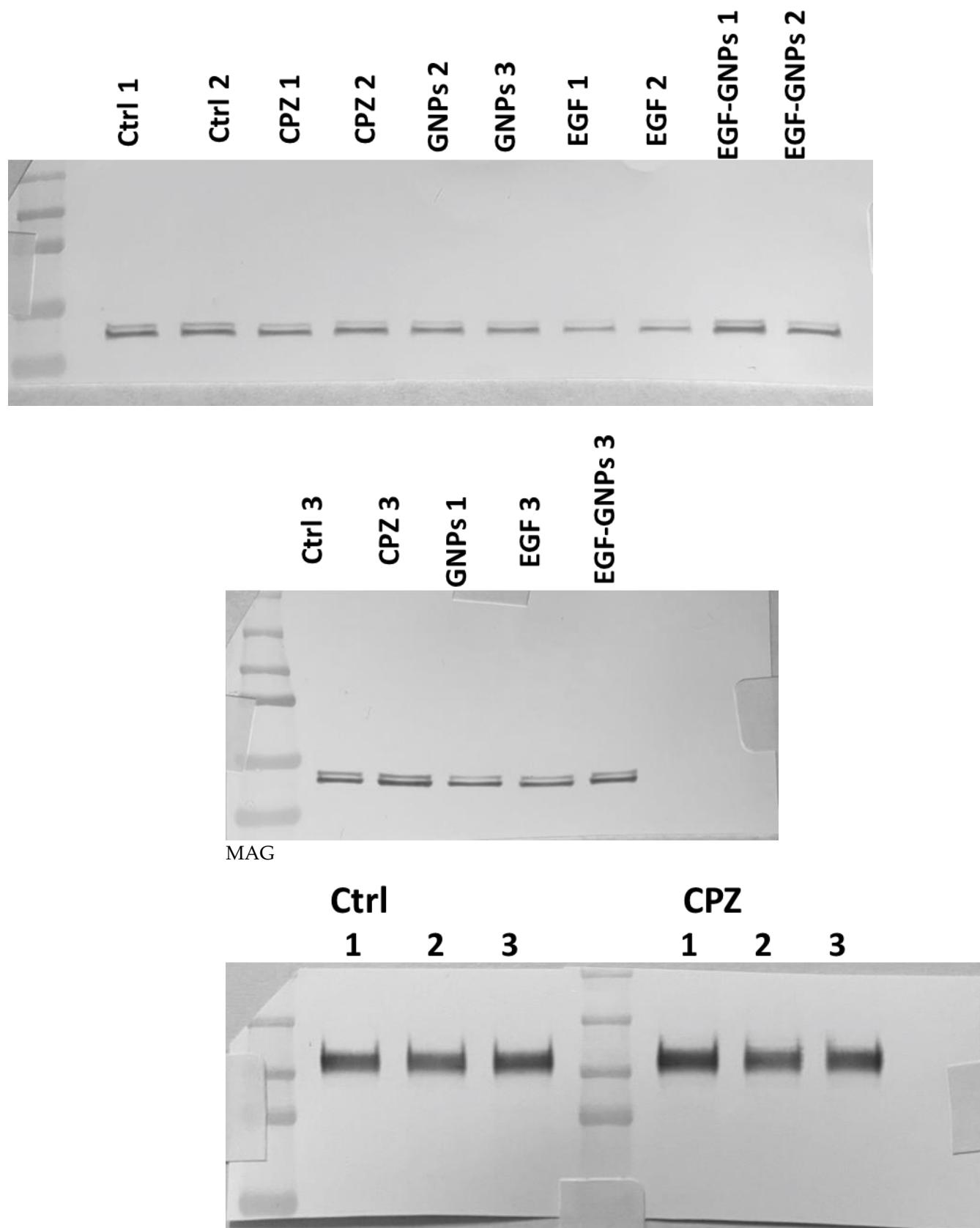
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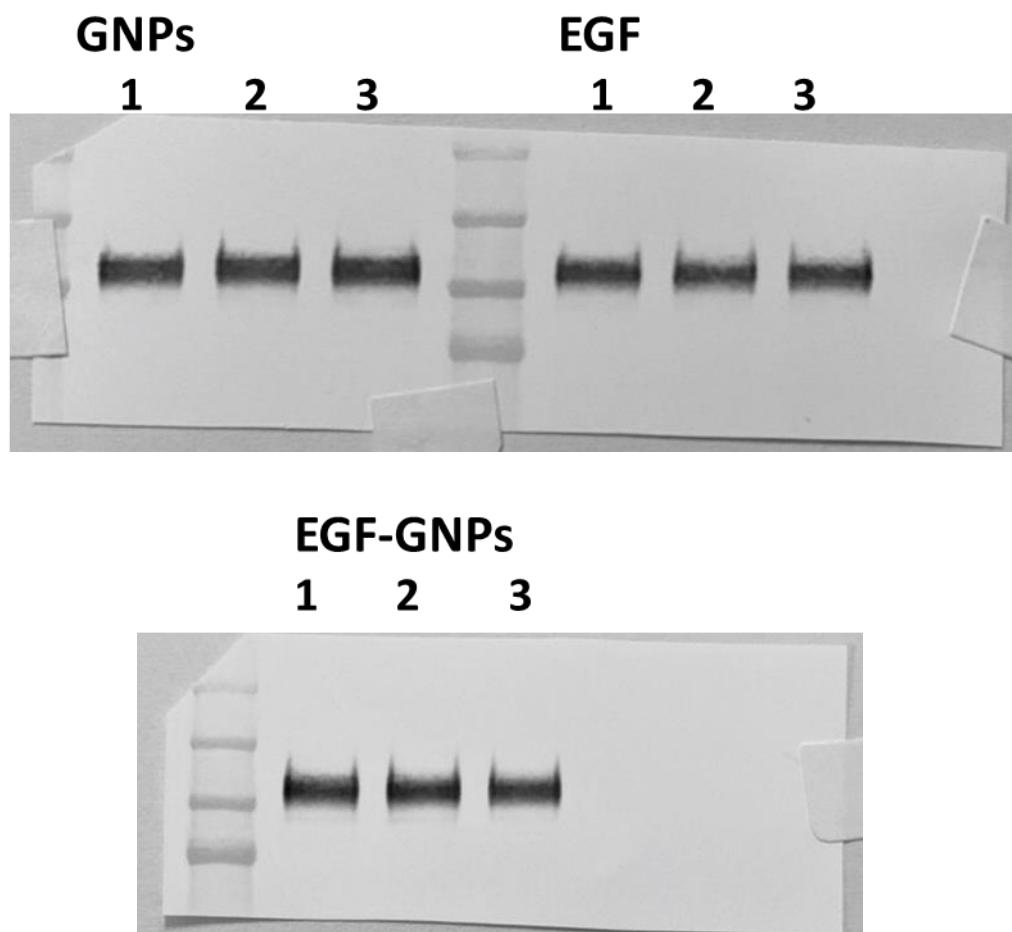


CNPase

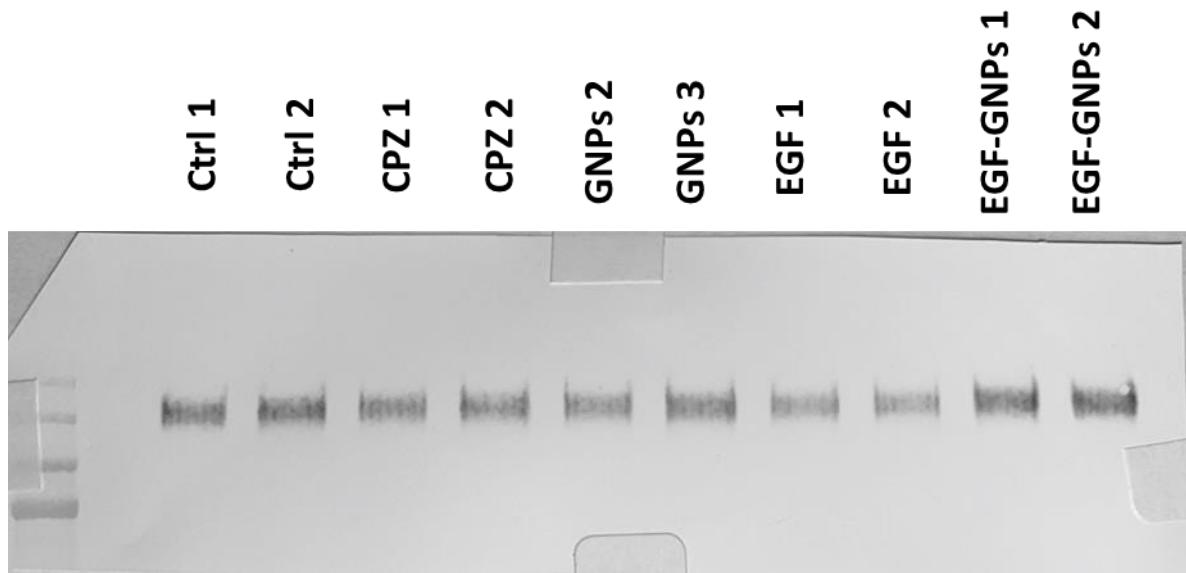


CNPase: duplicates used as representative images in the main text





MAG: duplicates used as representative images in the main text



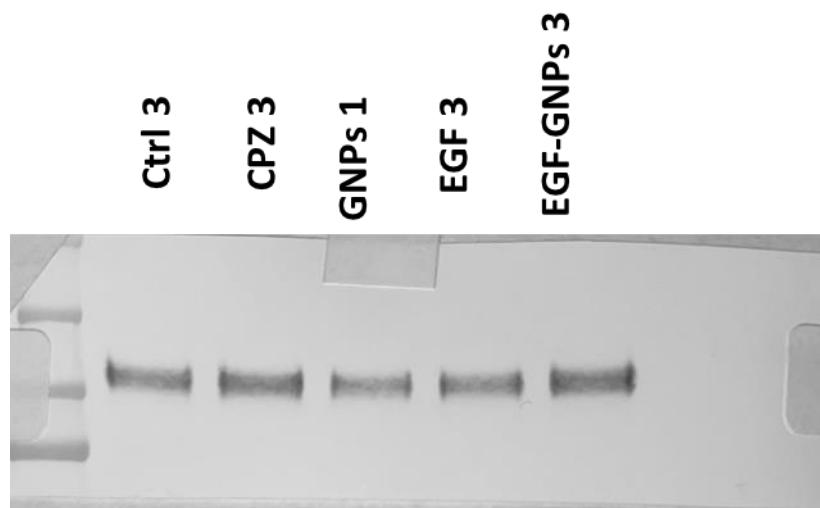


Figure S2. Blots obtained at 3 weeks of recovery.