

Western Blot Overview

Enhanced Ca²⁺-dependent SK-channel gating and membrane trafficking in human atrial fibrillation

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Note: Given the limited availability of human atrial tissue samples, most gels were physically cut to enable incubation with different antibodies against proteins with distinct molecular weights. These fragments are shown in the subsequent slides. Unless otherwise indicated, samples are loaded in pairs: two Ctl samples followed by two cAF samples, etc. with molecular weight markers in the first and/or last lanes. Only samples that were used in the representative blots and/or quantitative analysis are marked.

SK1

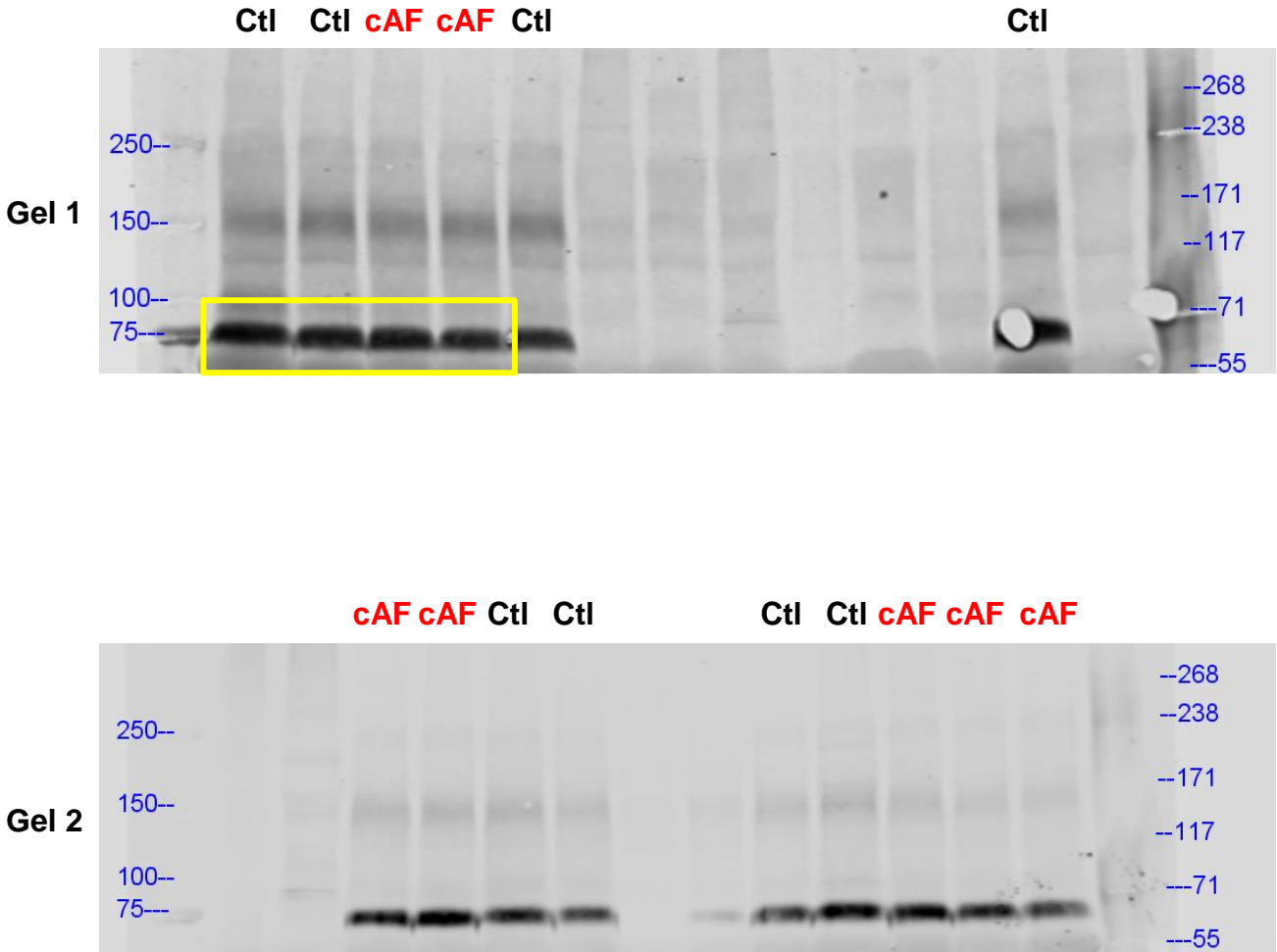


Figure 2 – Protein levels in RA cardiomyocytes

SK2

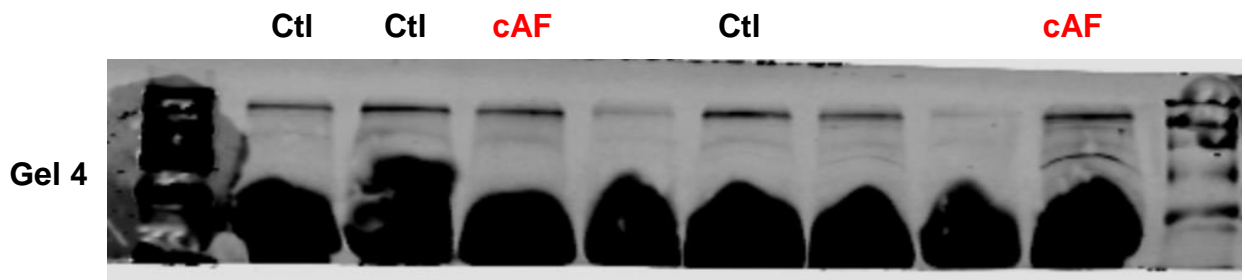
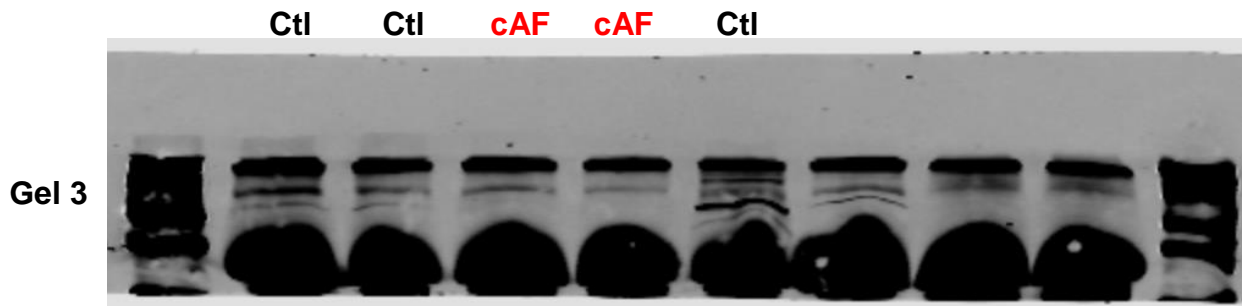
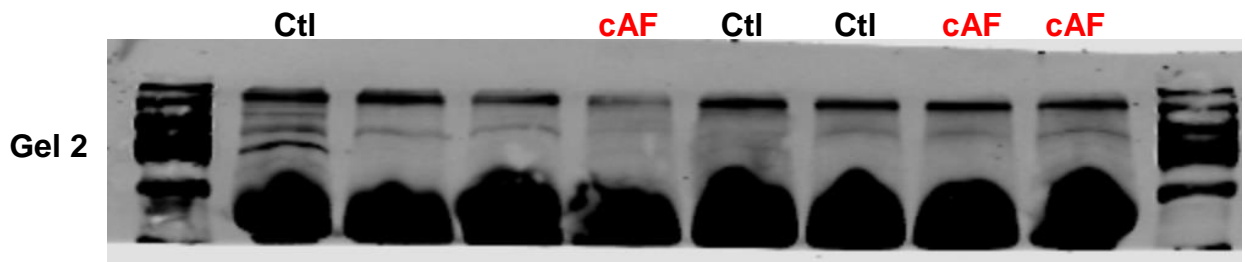
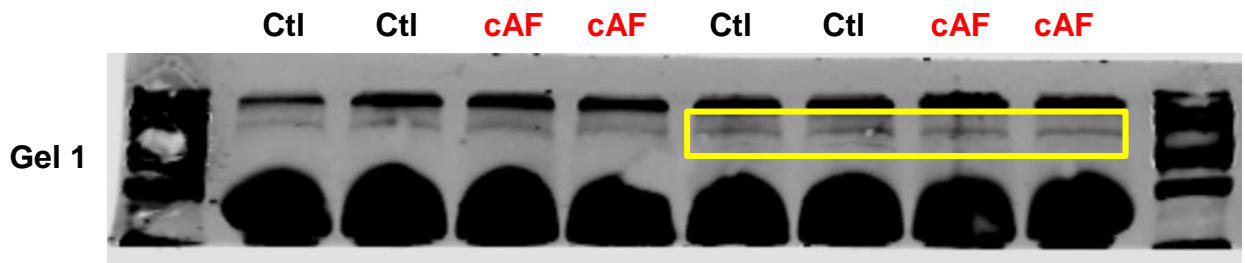


Figure 2 – Protein levels in RA cardiomyocytes

SK3

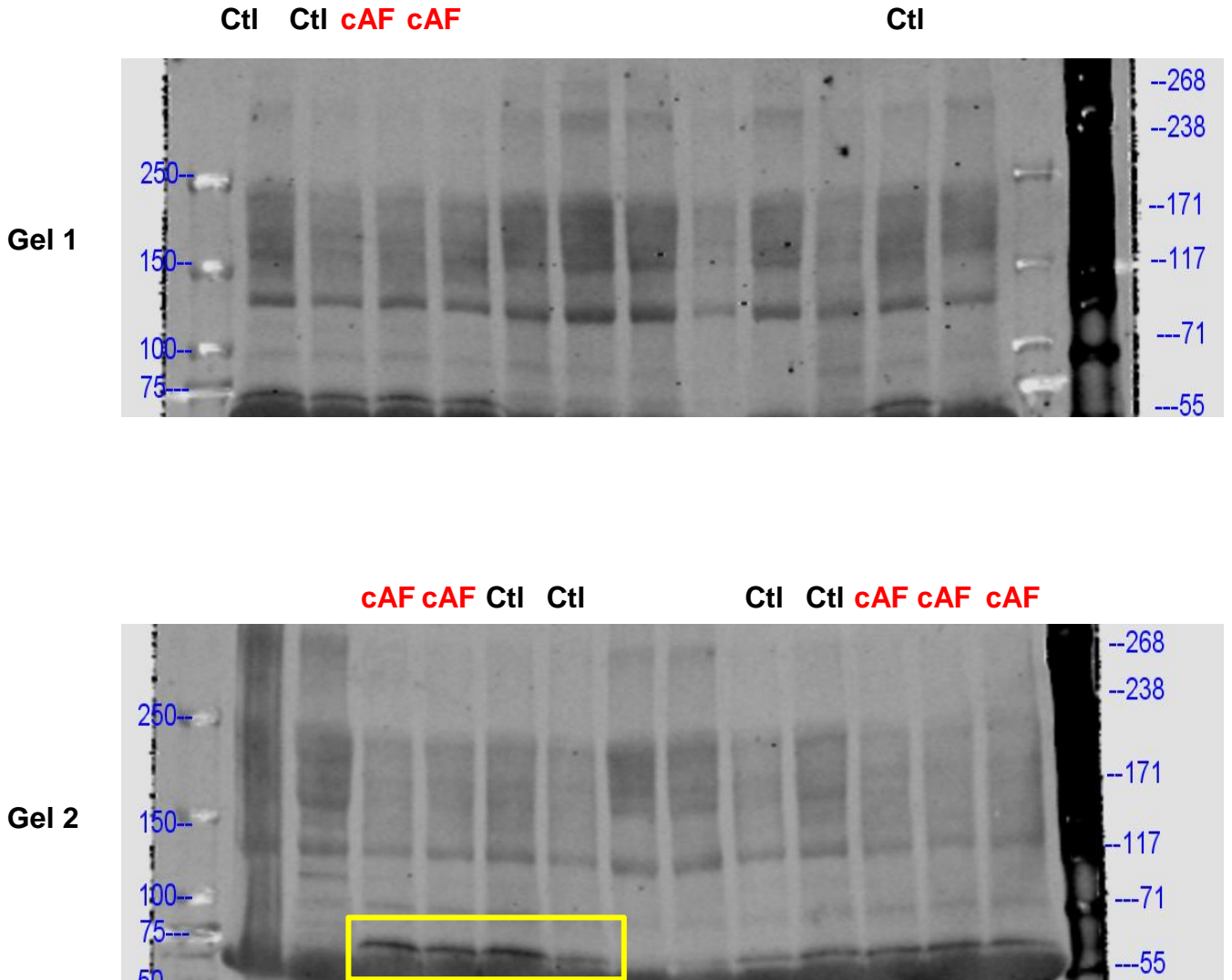


Figure 2 – Protein levels in RA cardiomyocytes

PP2Ac

Ctl

cAF

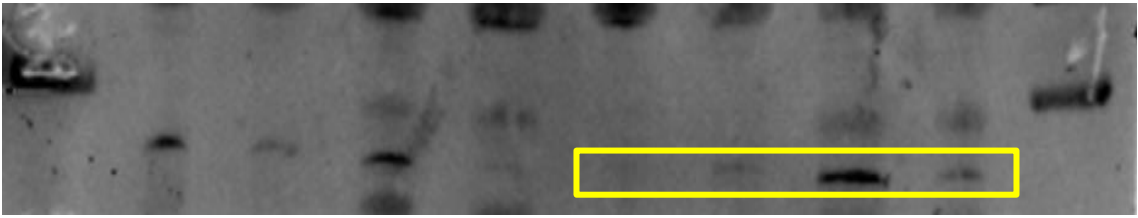
Ctl

Ctl

cAF

cAF

Gel 1



Ctl

cAF

cAF

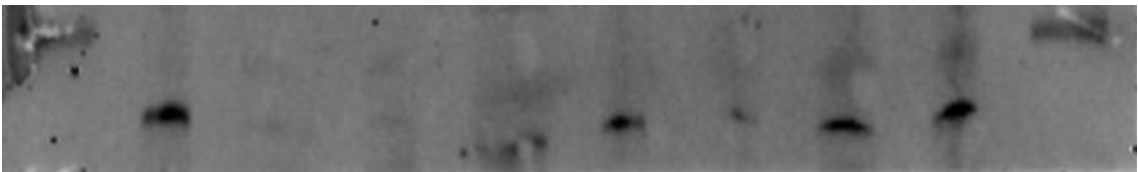
Ctl

Ctl

cAF

cAF

Gel 2



Ctl

Ctl

cAF

cAF

Ctl

cAF

cAF

Gel 3



Ctl

cAF

cAF

Ctl

Ctl

cAF

cAF

Gel 4



Figure 6 – Protein levels in RA cardiomyocytes

Total-CaM

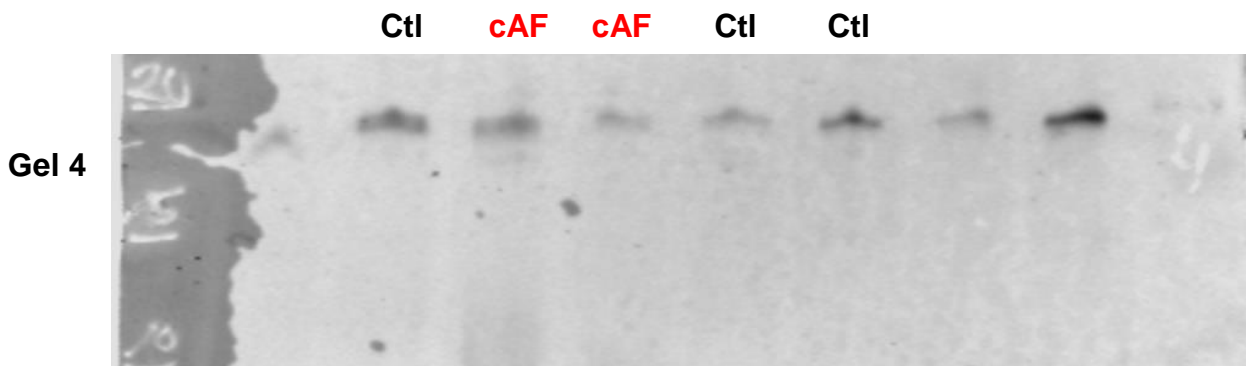
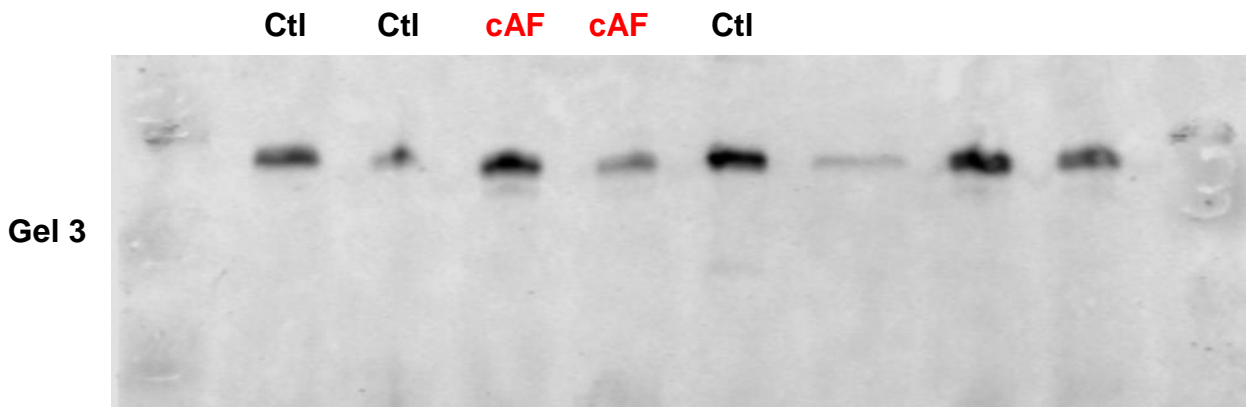
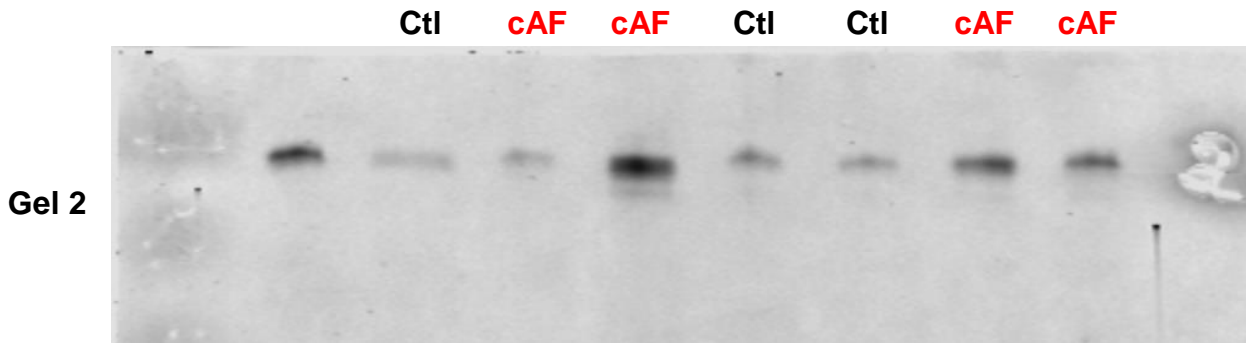
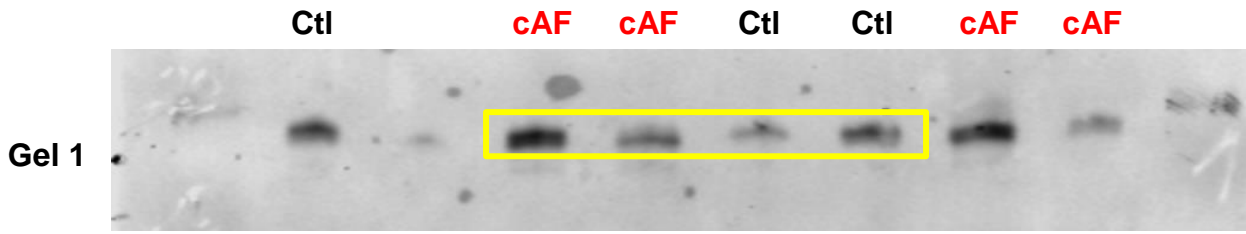


Figure 6 – Protein levels in RA cardiomyocytes

Thr80-CaM

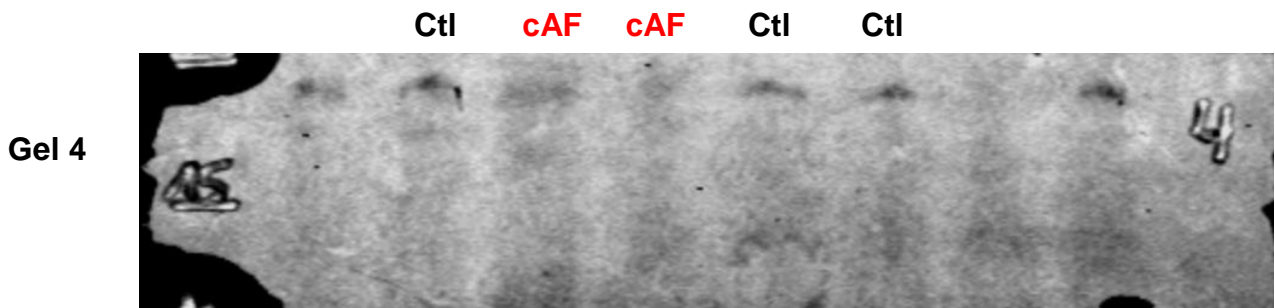
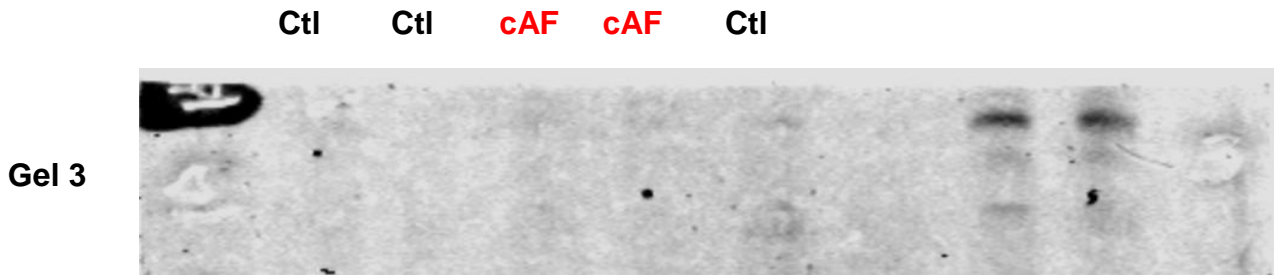
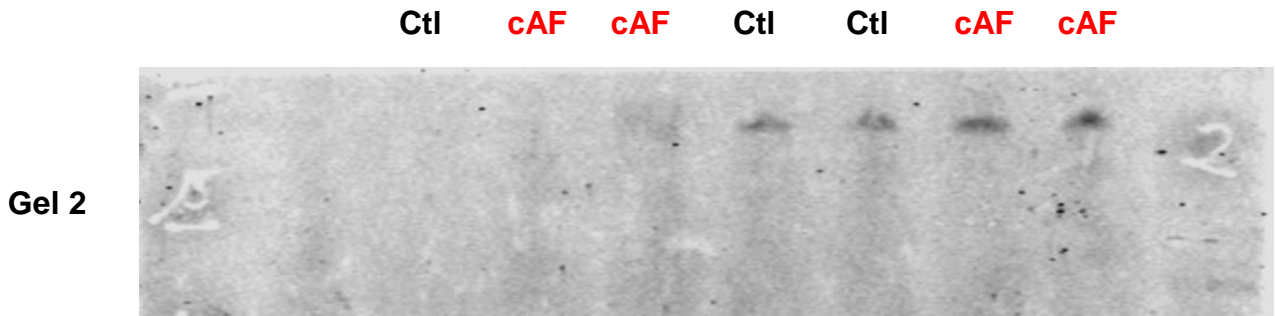
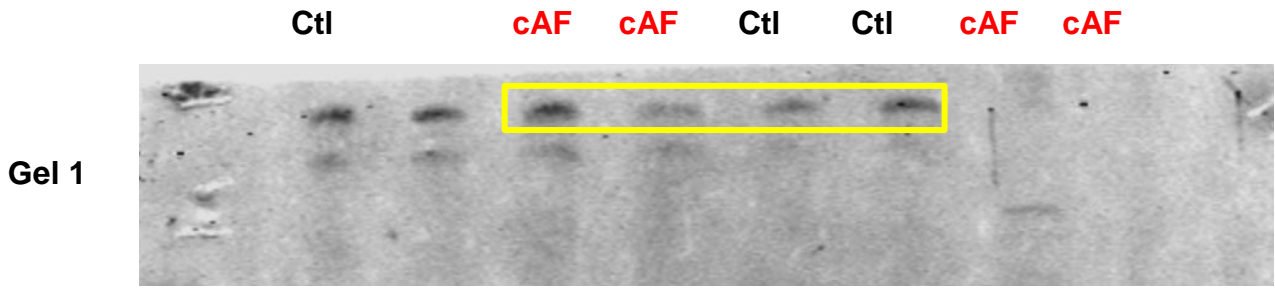


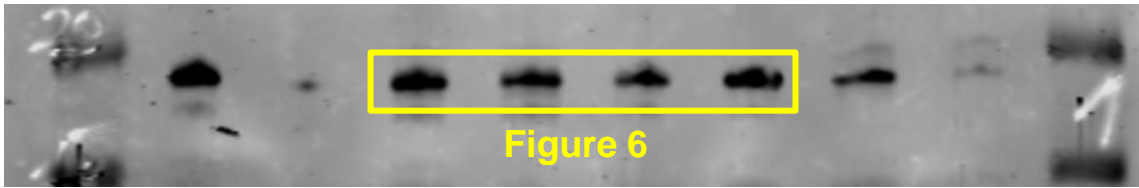
Figure 6 – Protein levels in RA cardiomyocytes

Troponin C

Used for

CaM	CaM	CaM	CaM	CaM	CaM	CaM	CaM
PP2Ac	PP2Ac	PP2Ac	PP2Ac	PP2Ac	PP2Ac	PP2Ac	PP2Ac
SK2	SK2	SK2	SK2	SK2	SK2	SK2	SK2
Ctl	Ctl	cAF	cAF	Ctl	Ctl	cAF	cAF

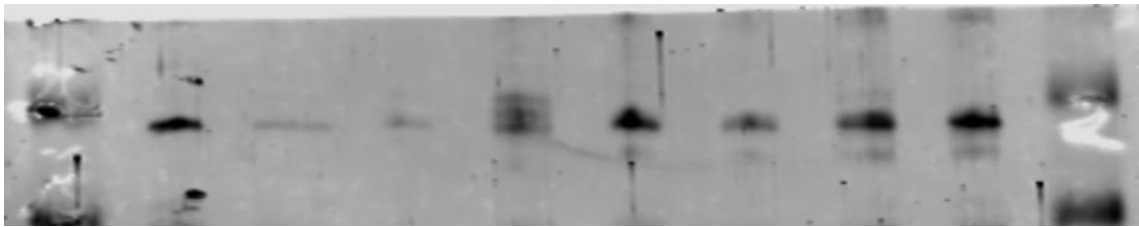
Gel 1



Used for

CaM	CaM	CaM	CaM	CaM	CaM	CaM	CaM
PP2Ac	PP2Ac	PP2Ac	PP2Ac	PP2Ac	PP2Ac	PP2Ac	PP2Ac
SK2		SK2	SK2	SK2	SK2	SK2	SK2
Ctl	Ctl	cAF	cAF	Ctl	Ctl	cAF	cAF

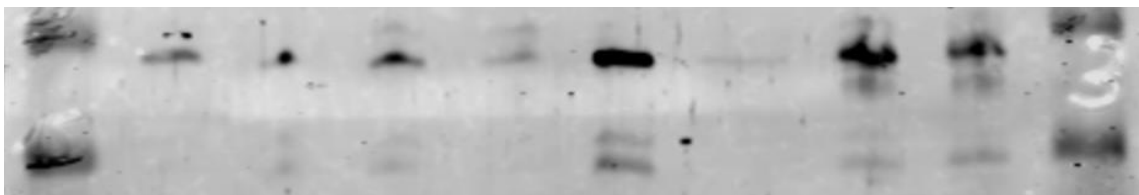
Gel 2



Used for

CaM	CaM	CaM	CaM	CaM	CaM	CaM	CaM
PP2Ac	PP2Ac	PP2Ac	PP2Ac	PP2Ac	PP2Ac	PP2Ac	PP2Ac
SK2	SK2	SK2	SK2	SK2			
Ctl	Ctl	cAF	cAF	Ctl		cAF	cAF

Gel 3



Used for

CaM	CaM	CaM	CaM	CaM	CaM	CaM	CaM
PP2Ac	PP2Ac	PP2Ac	PP2Ac	PP2Ac	PP2Ac	PP2Ac	PP2Ac
SK2	SK2	SK2		SK2			SK2
Ctl	Ctl	cAF	cAF	Ctl	Ctl	cAF	cAF

Gel 4



Figure 2 + 6 – Protein levels in RA cardiomyocytes

Troponin C

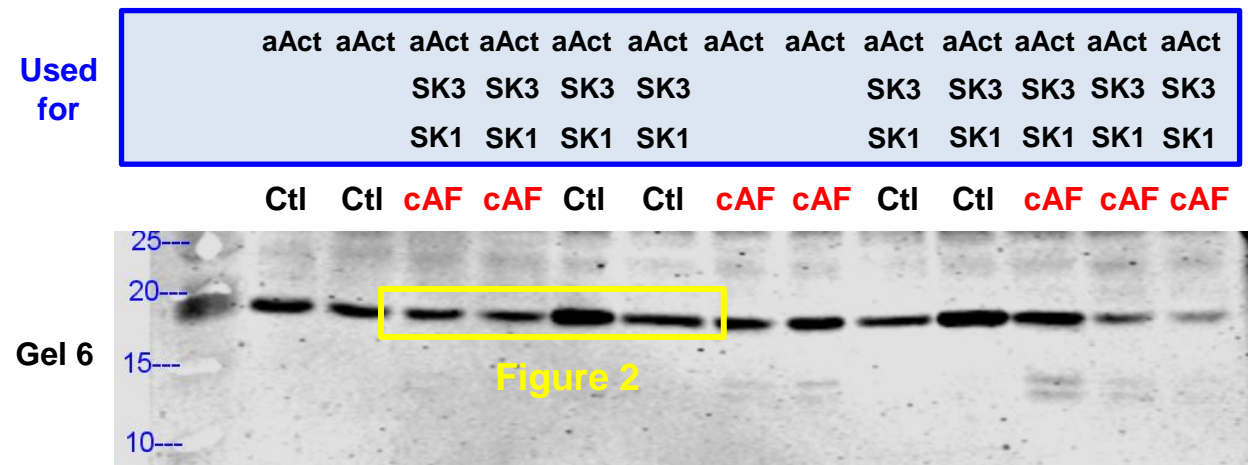
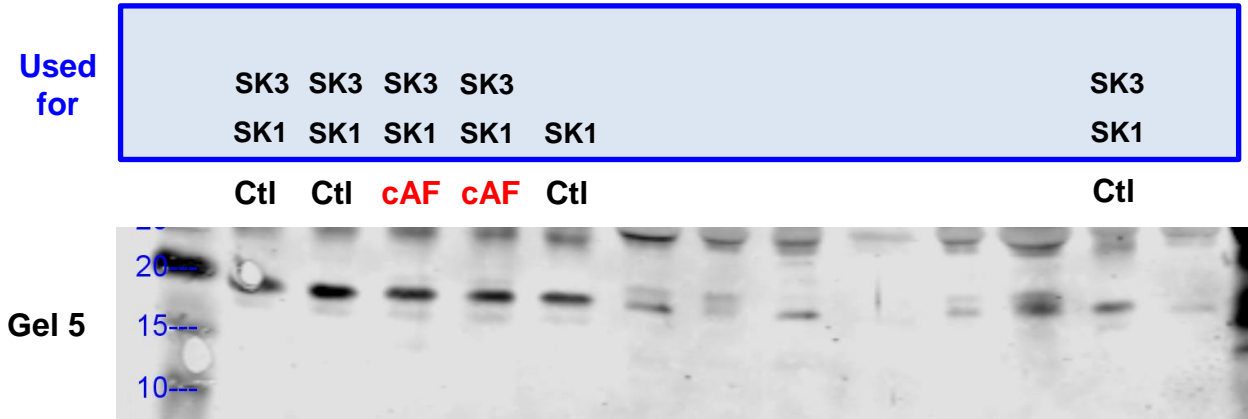


Figure 2 + Online Figure XXIII – Protein levels in RA CMs

SK2

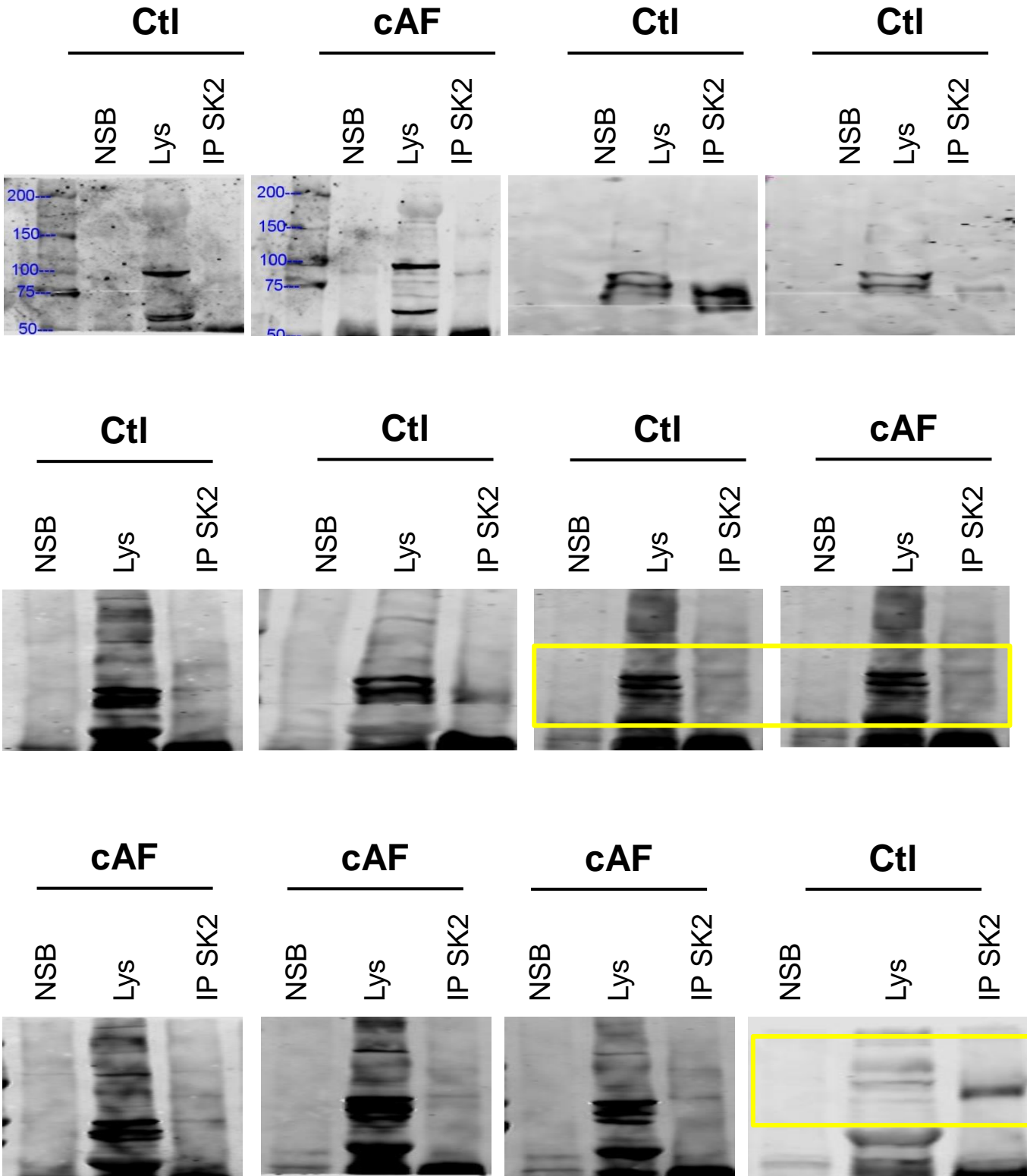


Figure 6 - colPs

SK2

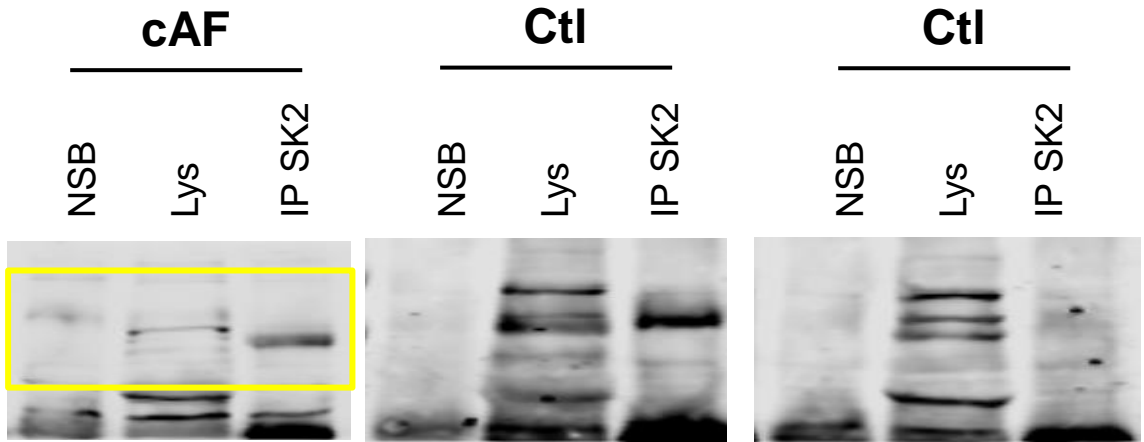
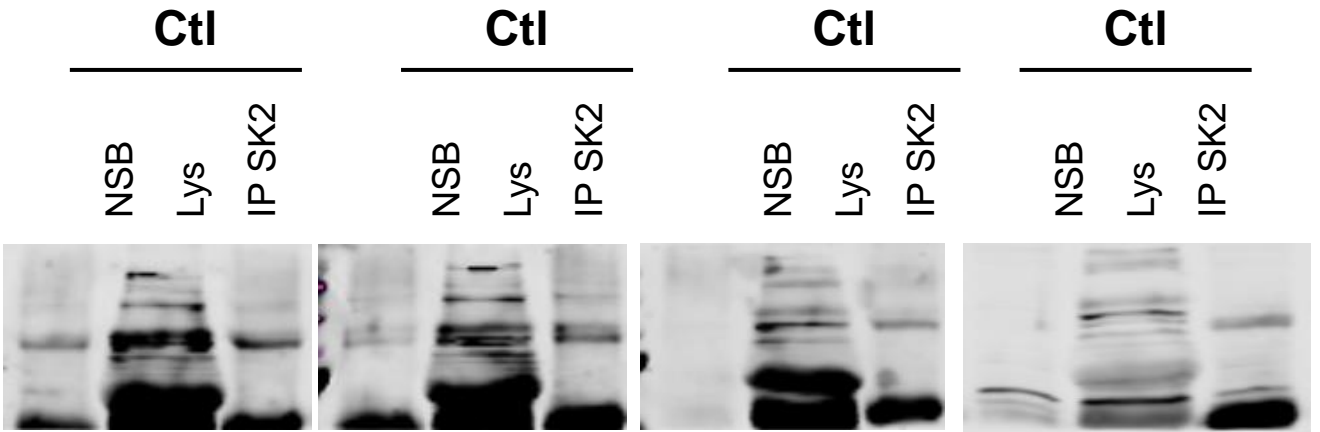


Figure 6 - colPs

α -Act2

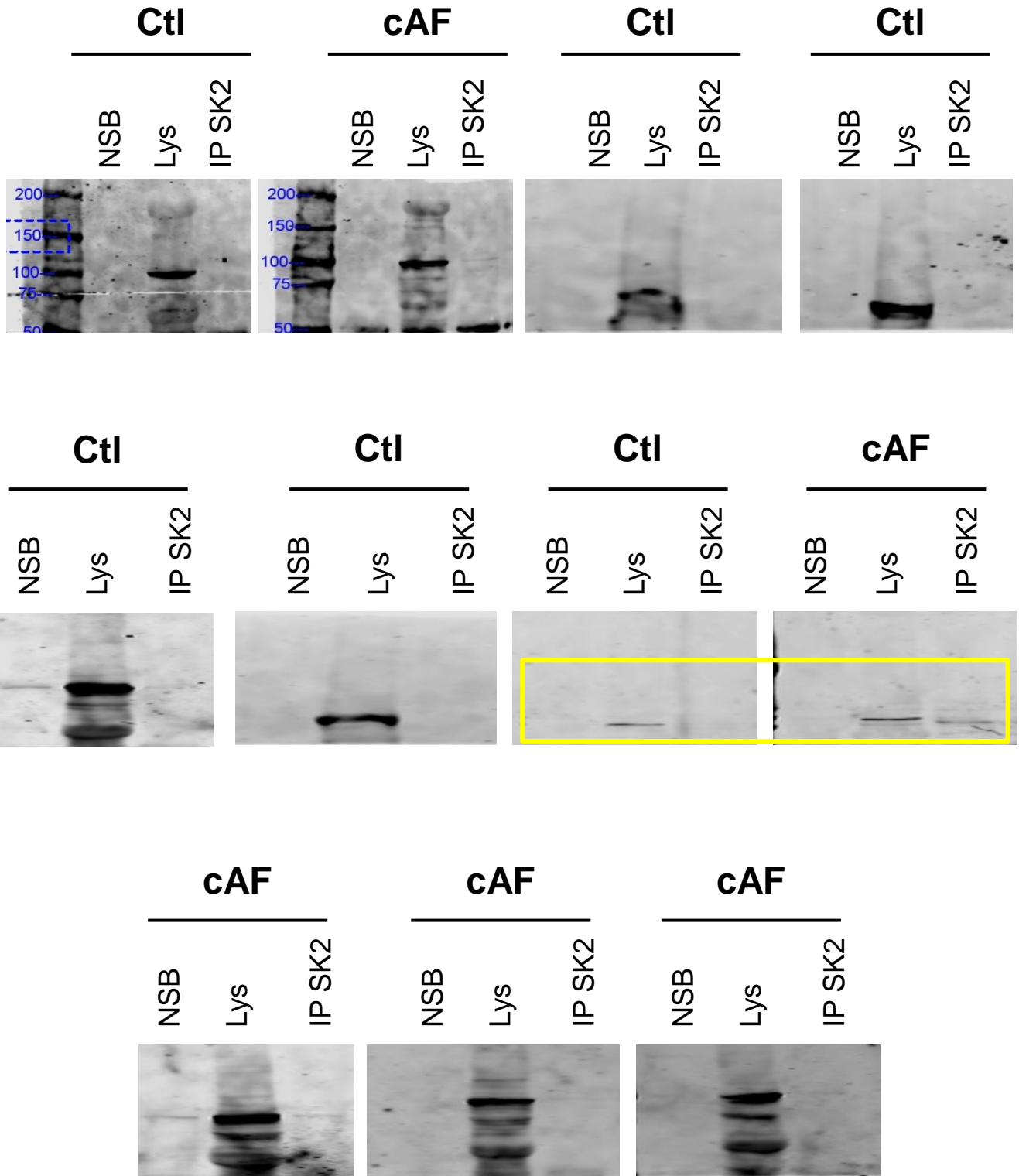


Figure 6 - colPs

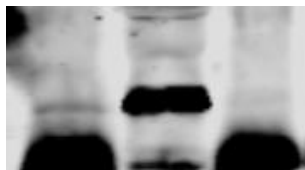
PP2Ac

Ctl

NSB

Lys

IP SK2

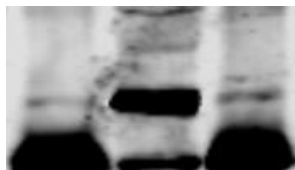


Ctl

NSB

Lys

IP SK2

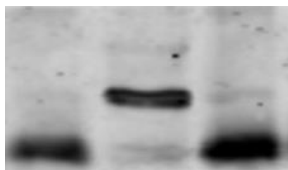


Ctl

NSB

Lys

IP SK2



Ctl

NSB

Lys

IP SK2



Ctl

NSB

Lys

IP SK2



cAF

NSB

Lys

IP SK2



Ctl

NSB

Lys

IP SK2



cAF

NSB

Lys

IP SK2



Ctl

NSB

Lys

IP SK2



cAF

NSB

Lys

IP SK2



cAF

NSB

Lys

IP SK2



cAF

NSB

Lys

IP SK2



Figure 6 - colPs

PP2Ac

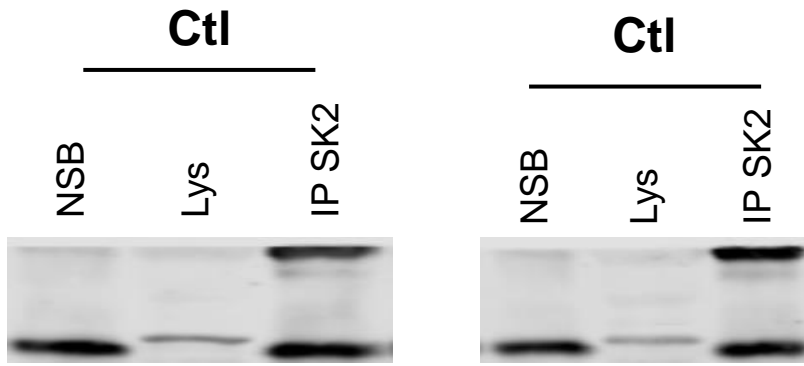
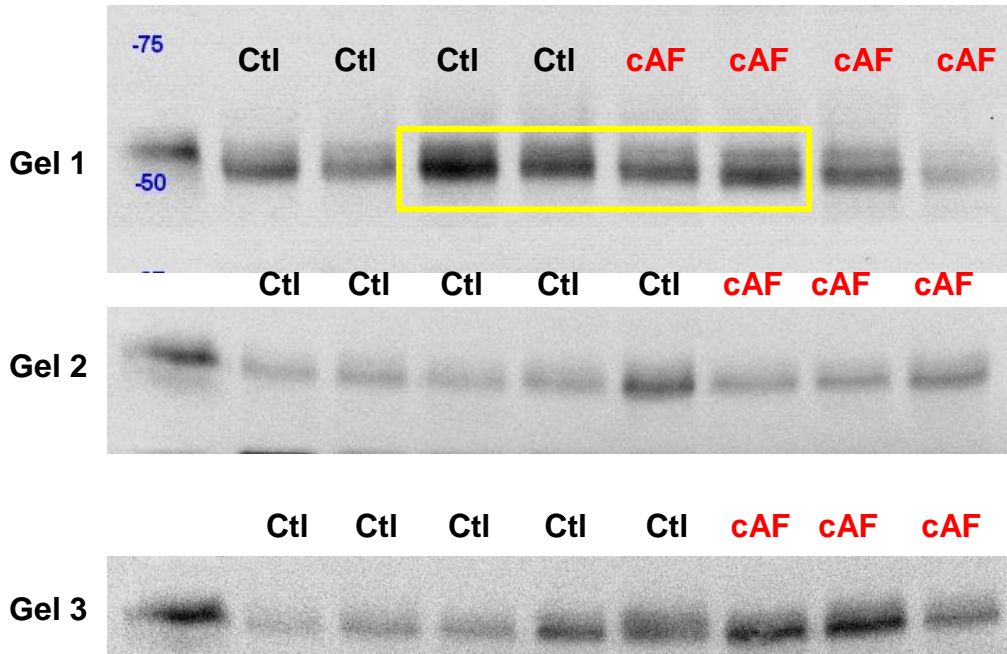
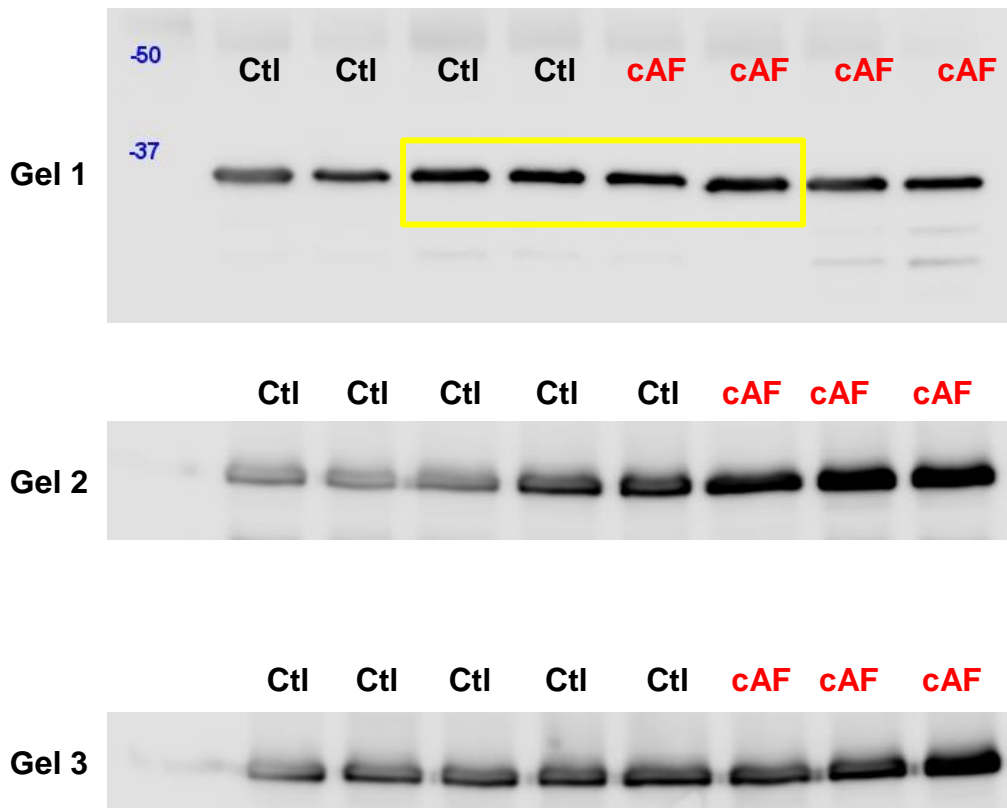


Figure 6 – colIPs

SK4

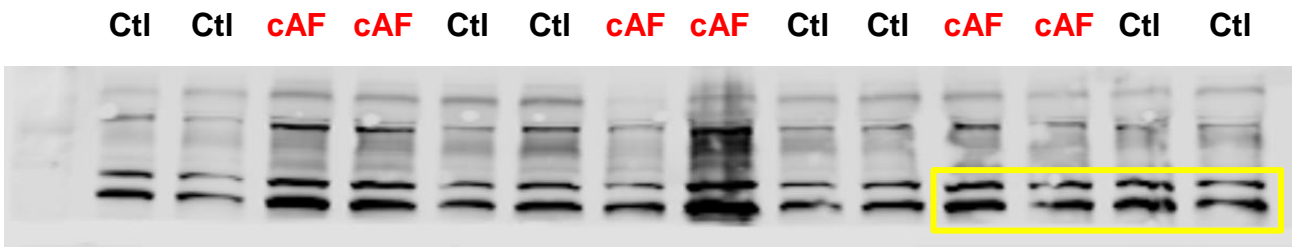


GAPDH

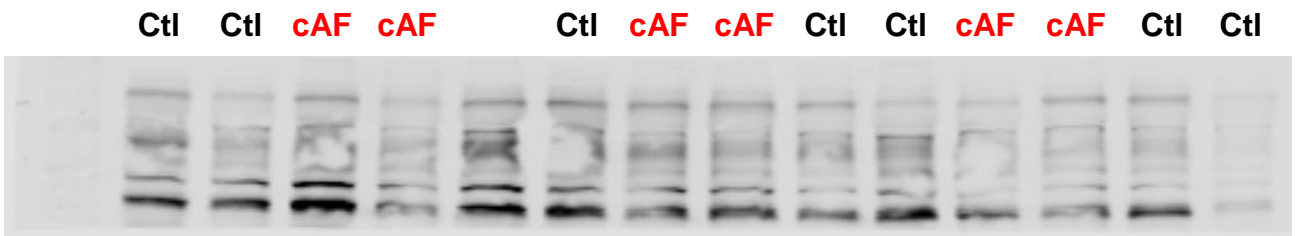


BKCa

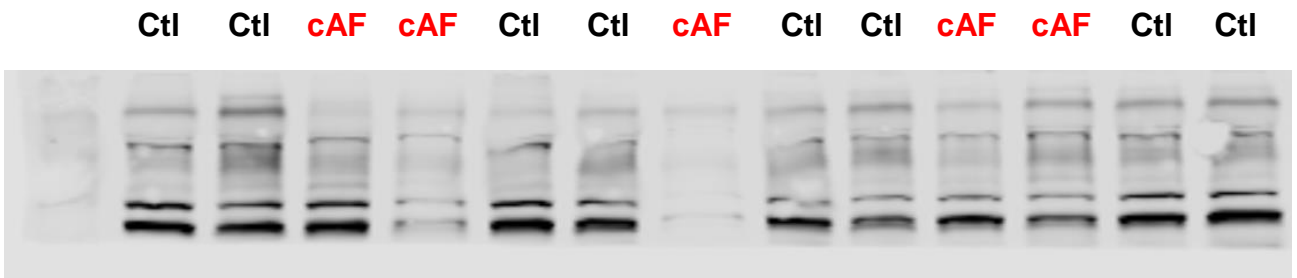
Gel 1



Gel 2

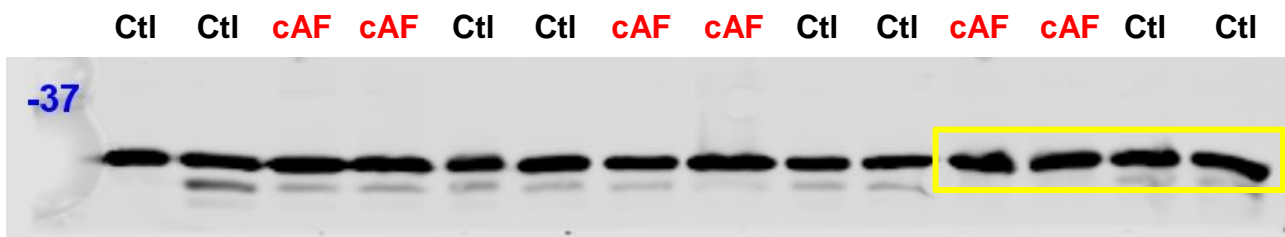


Gel 3

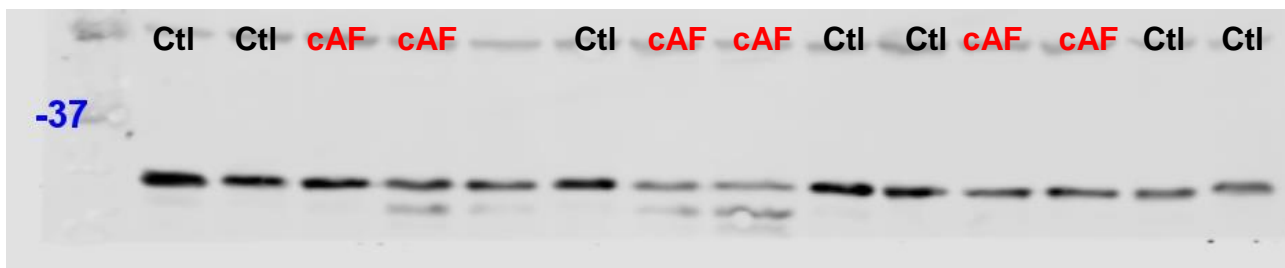


GAPDH

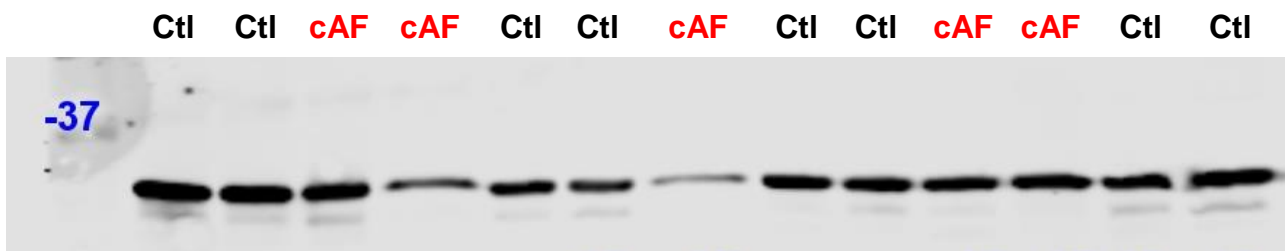
Gel 1



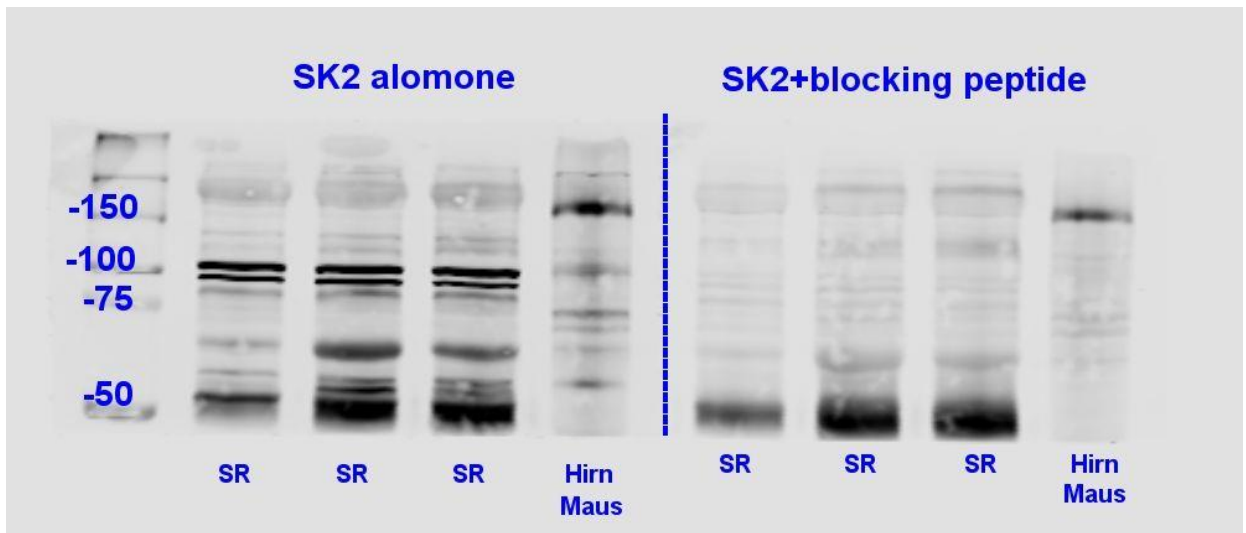
Gel 2



Gel 3

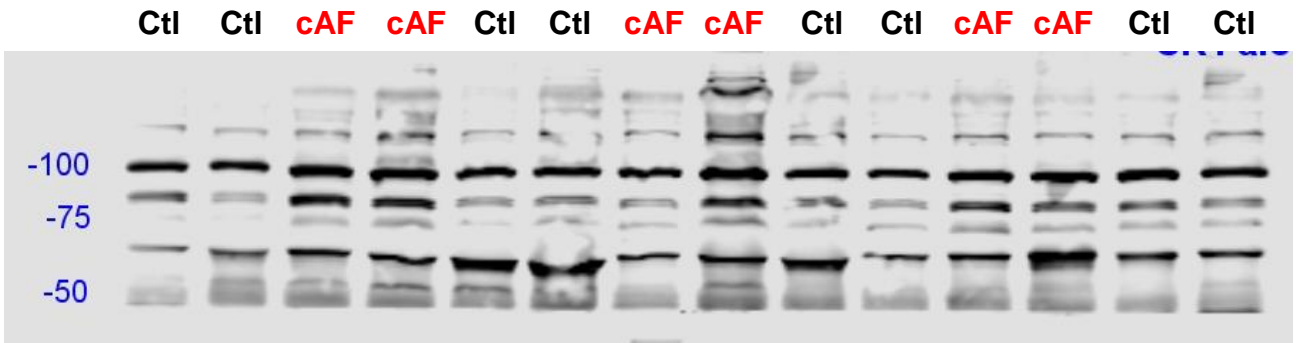


SK2 antibody + blocking peptide

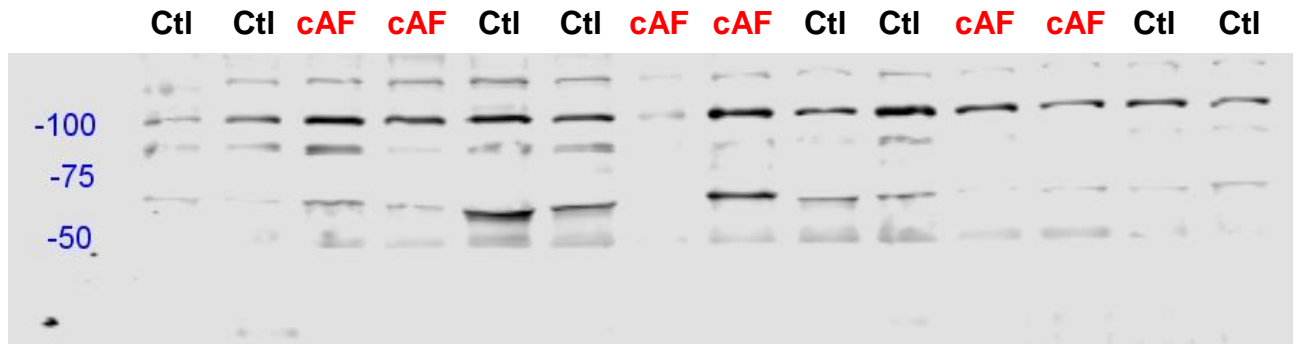


SK1

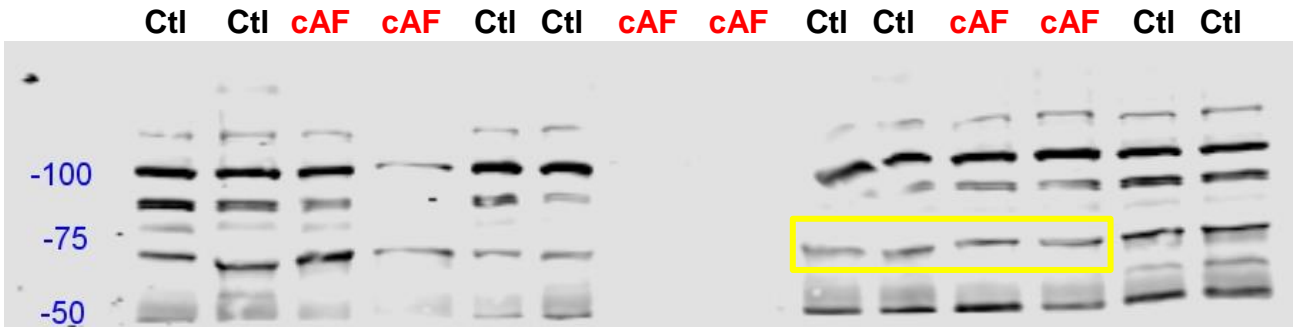
Gel 1



Gel 2



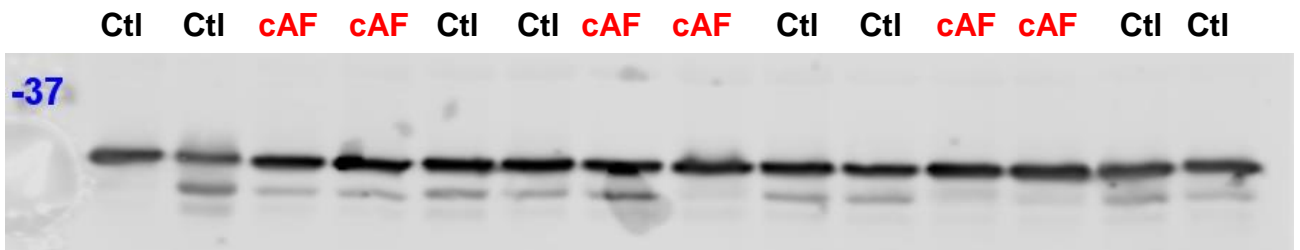
Gel 3



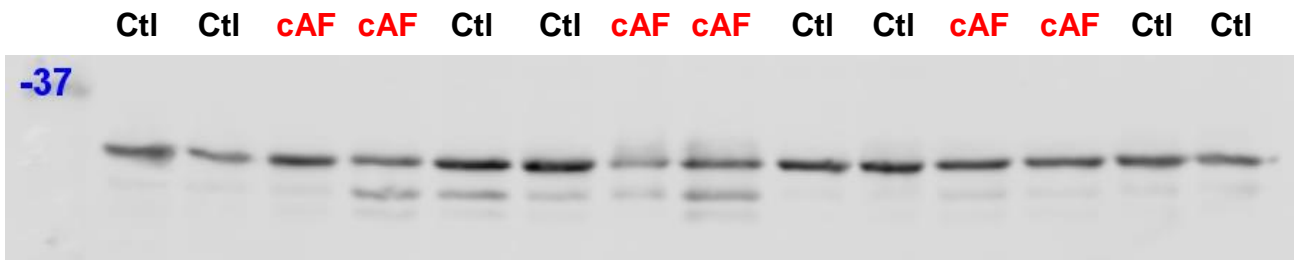
Online Figure XIII – Protein levels in RA whole-tissue homogenates

GAPDH

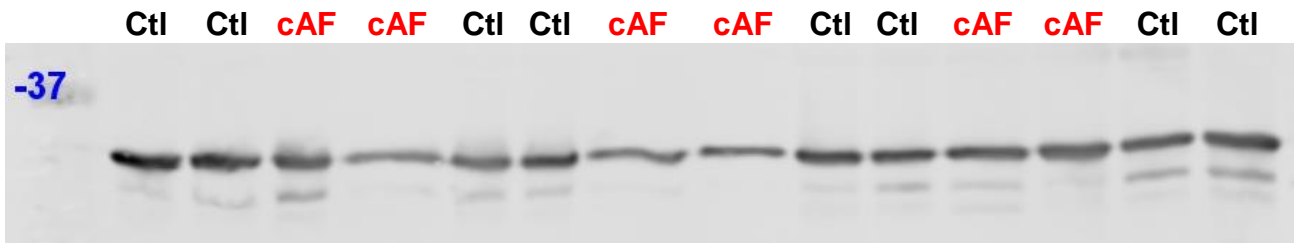
Gel 1



Gel 2

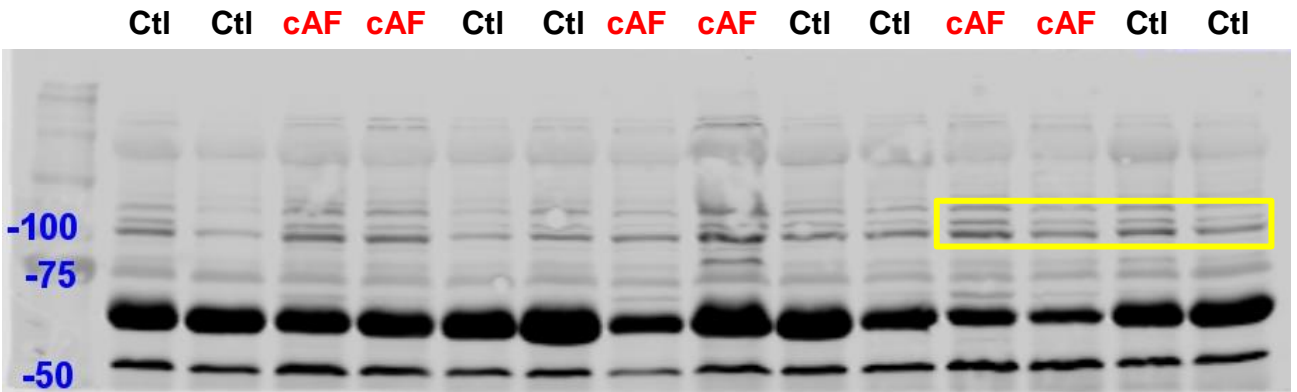


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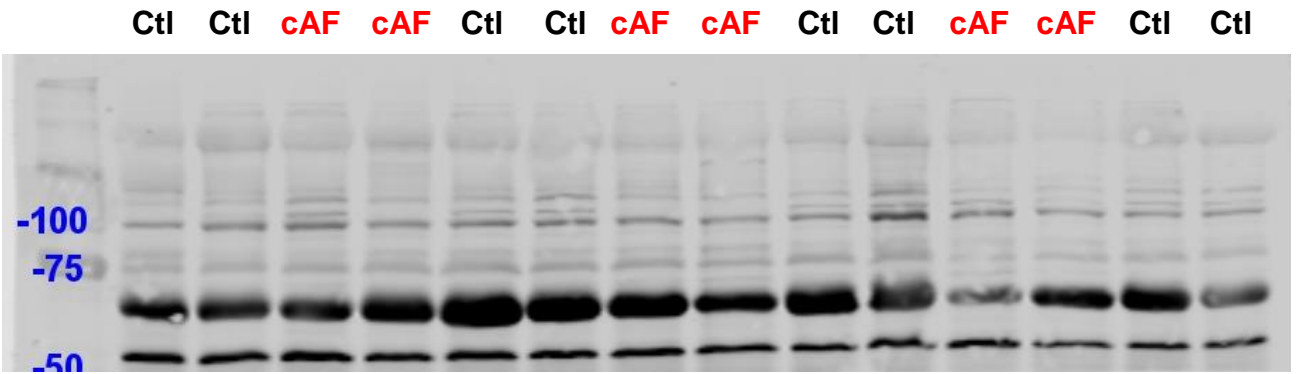


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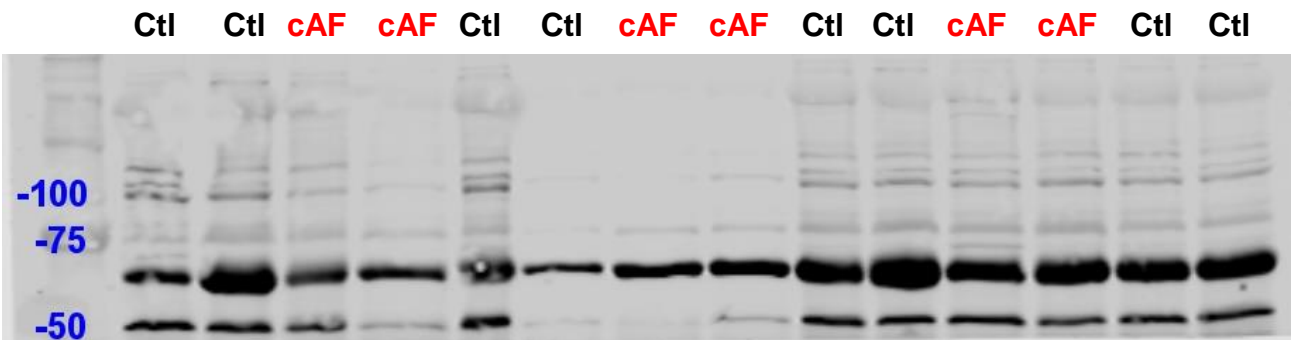
Gel 1



Gel 2



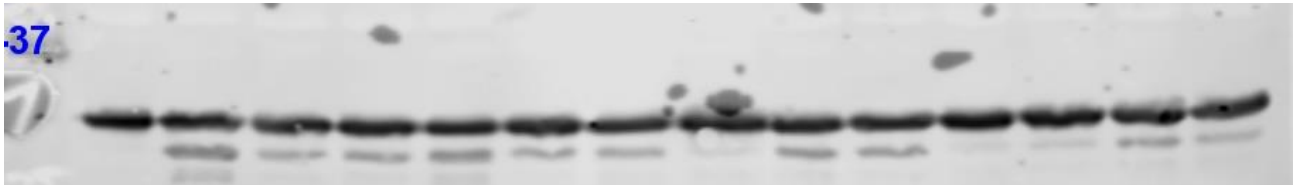
Gel 3



GAPDH

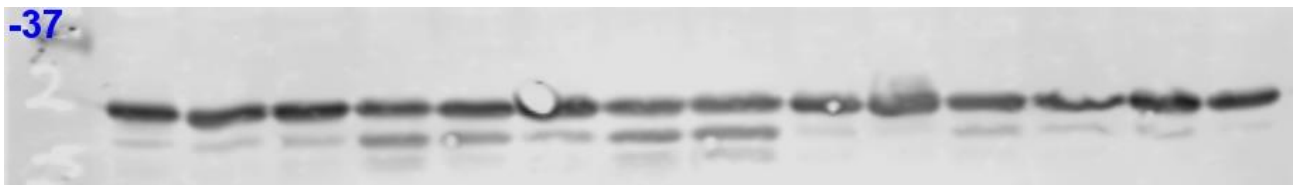
Gel 1

Ctl Ctl cAF cAF Ctl Ctl cAF cAF Ctl Ctl cAF cAF Ctl Ctl



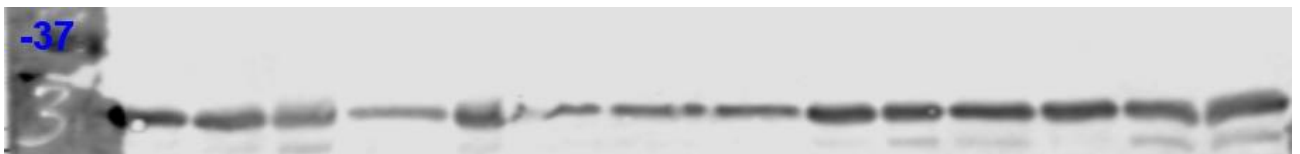
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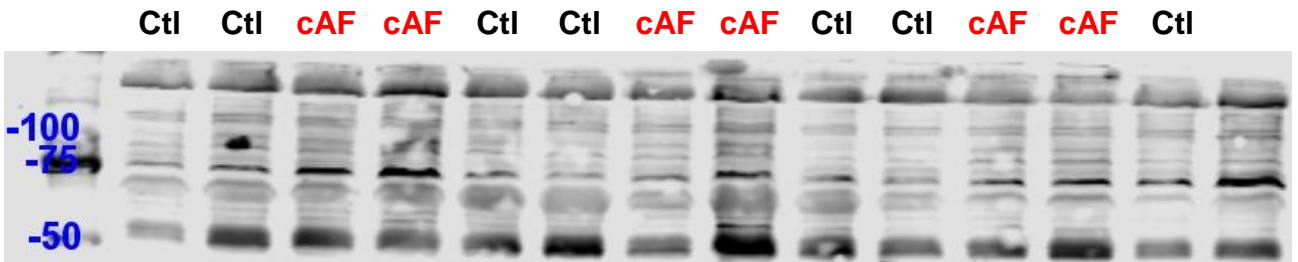
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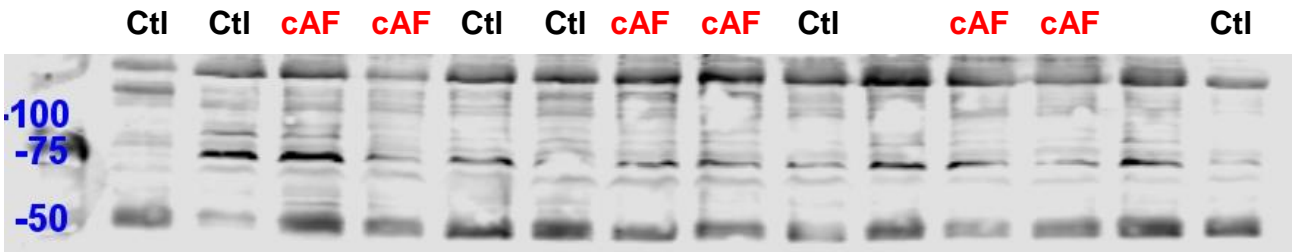


SK3

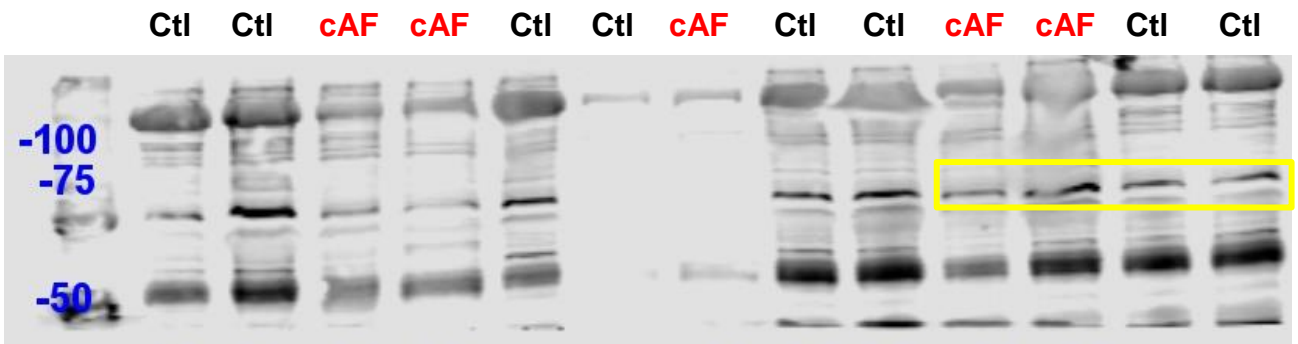
Gel 1



Gel 2

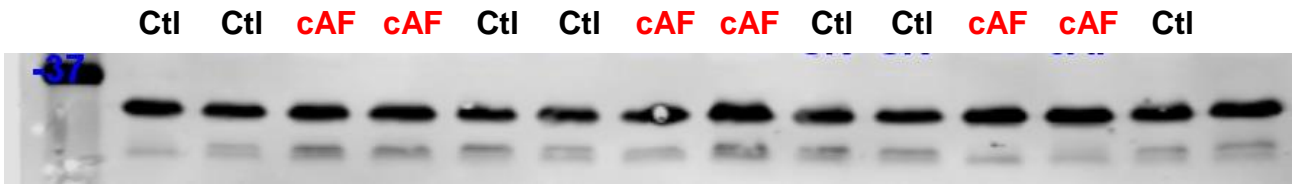


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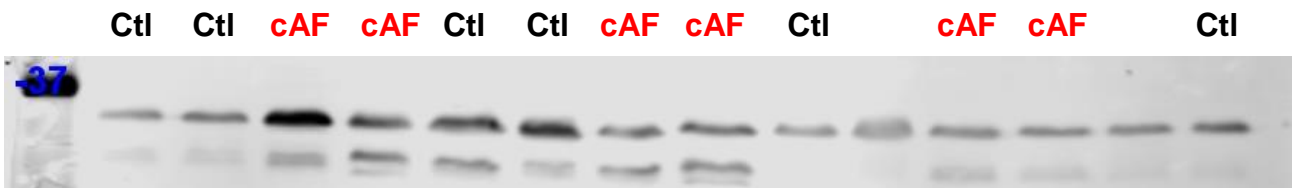


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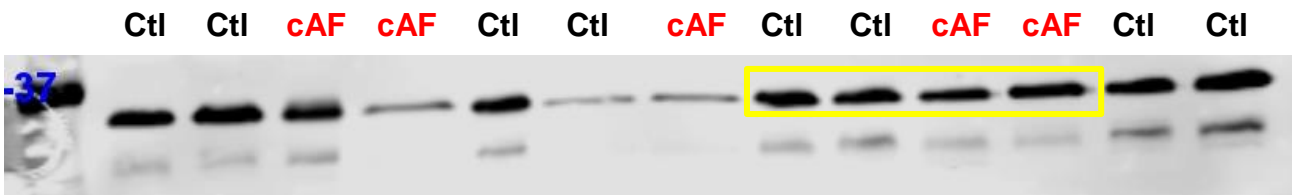
Gel 1



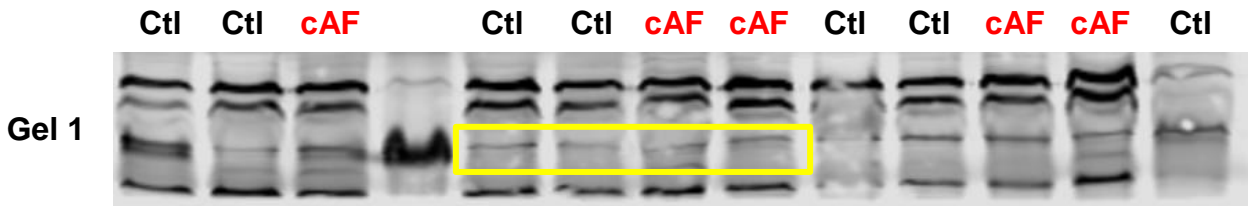
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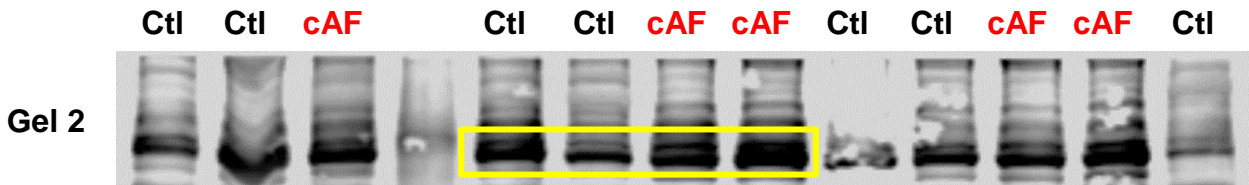
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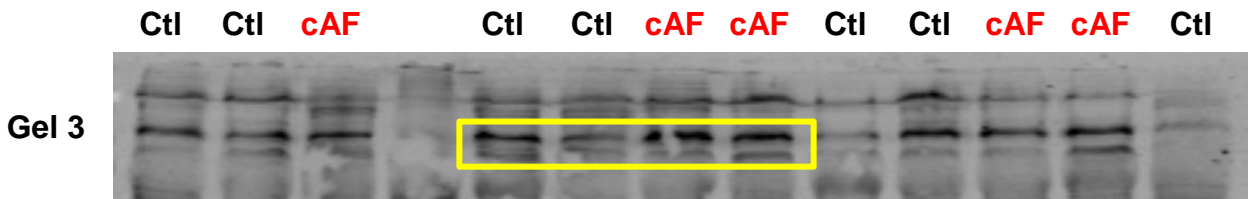
SK1



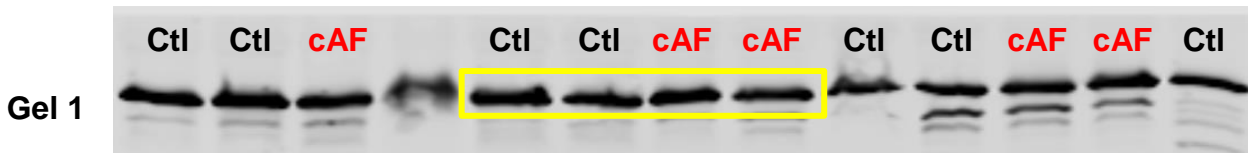
SK2



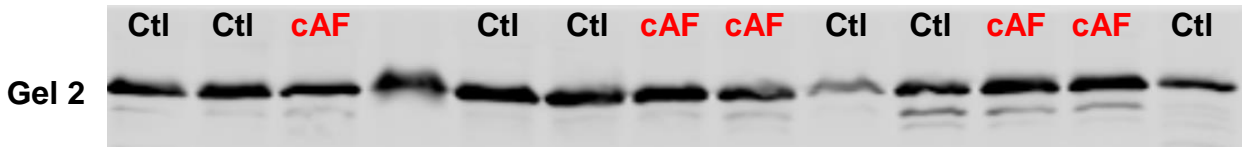
SK3



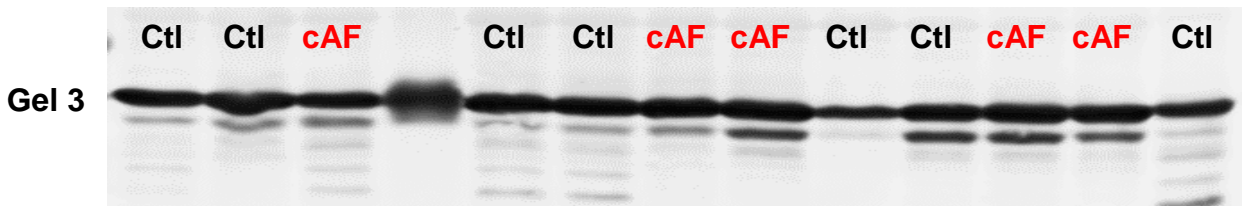
GAPDH



GAPDH



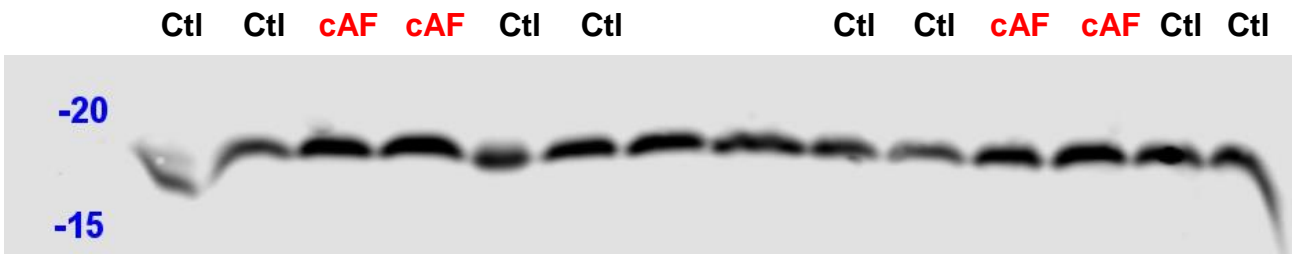
GAPDH



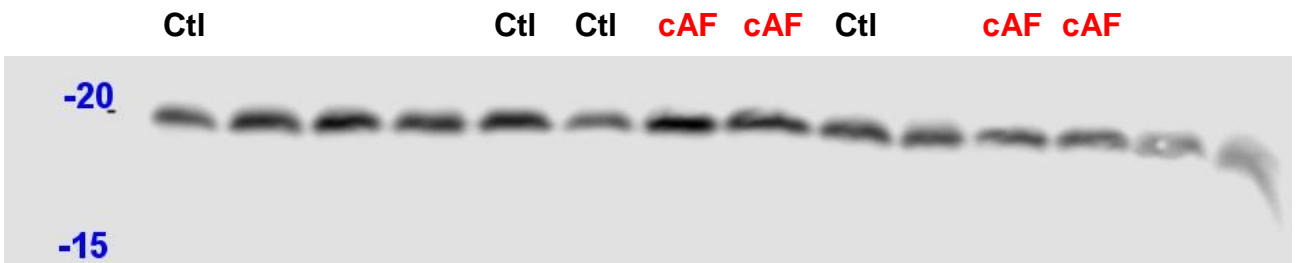
Online Figure XIII – Protein levels in LA whole-tissue homogenates

Total-CaM

Gel 1



Gel 2

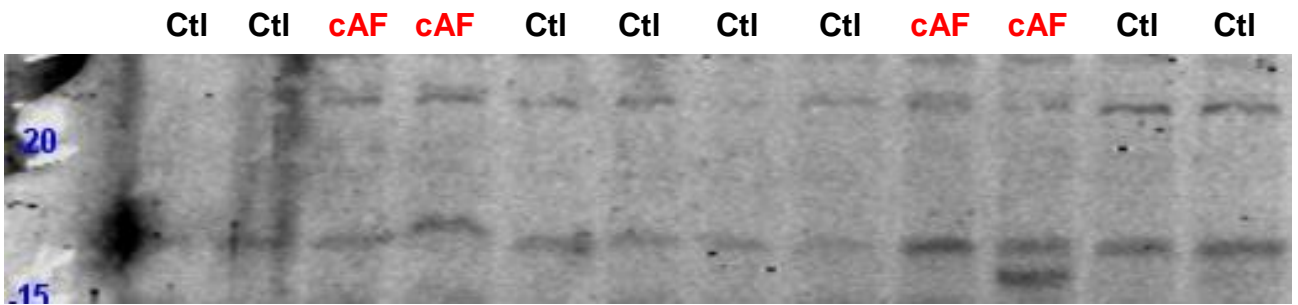


Gel 3

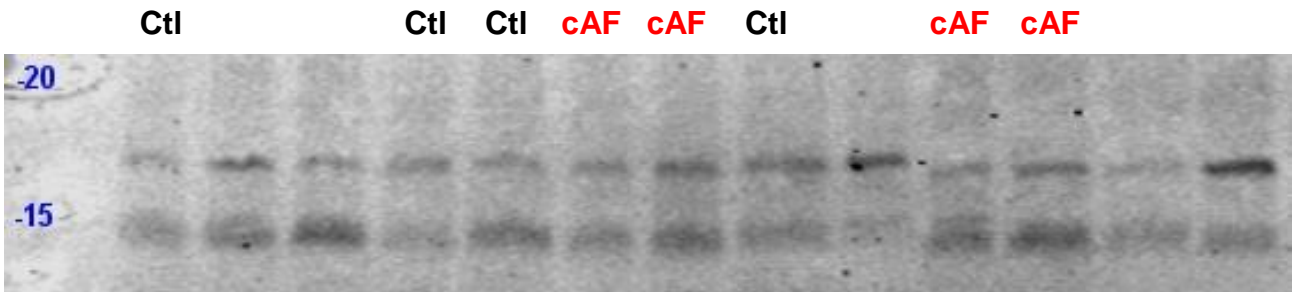


Thr80-CaM

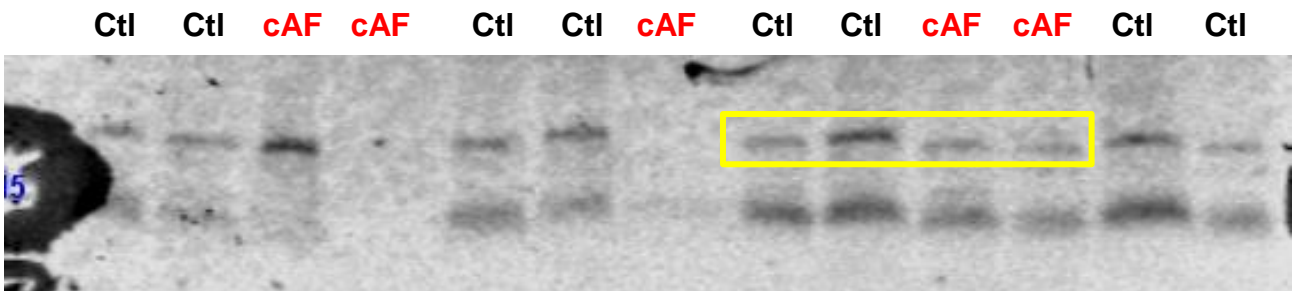
Gel 1



Gel 2

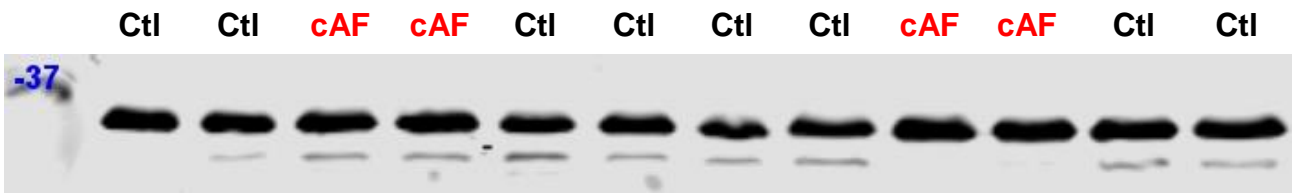


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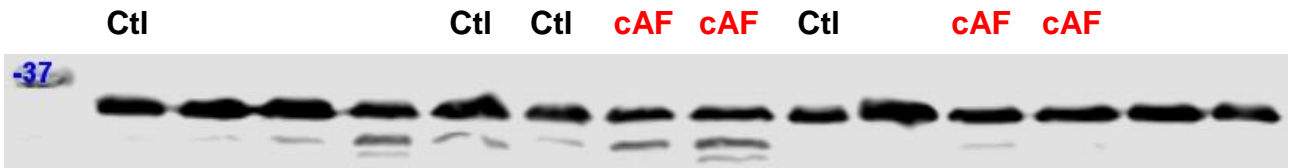


GAPDH

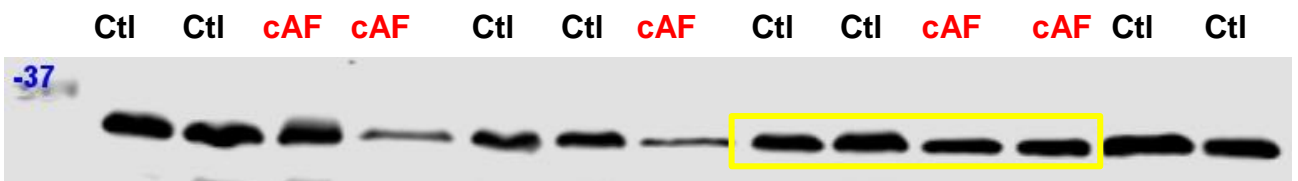
Gel 1



Gel 2

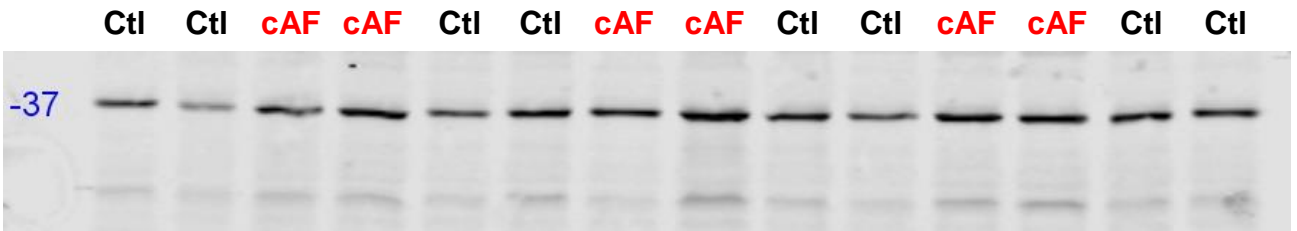


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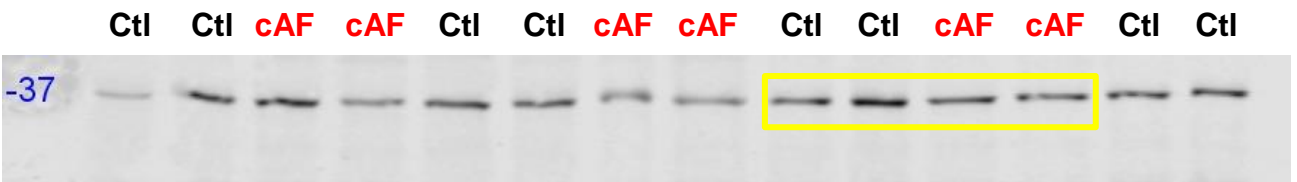


CKII α

Gel 1



Gel 2

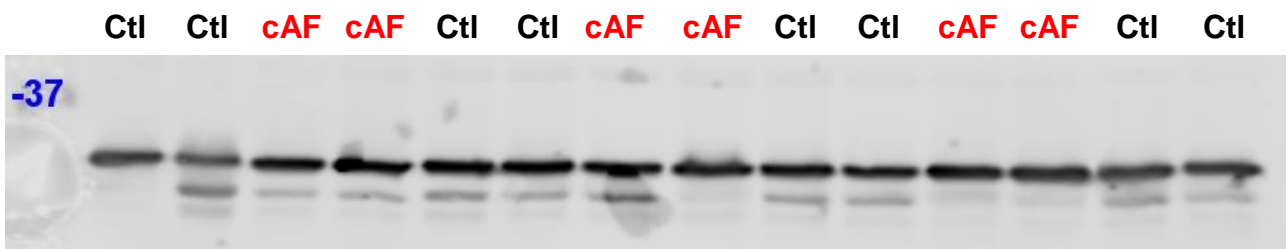


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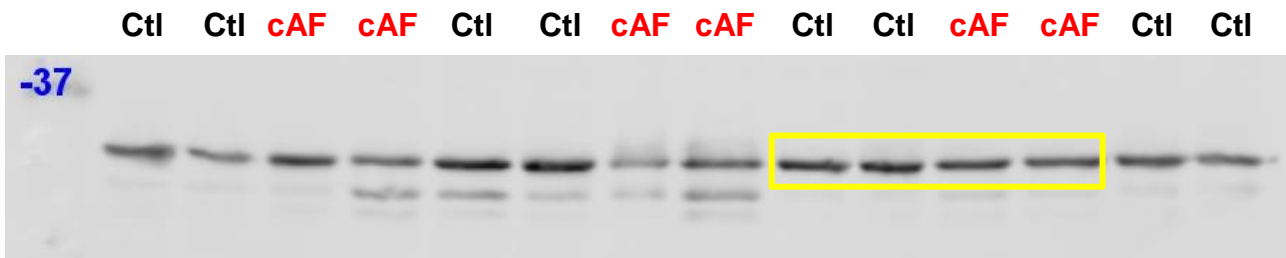


GAPDH

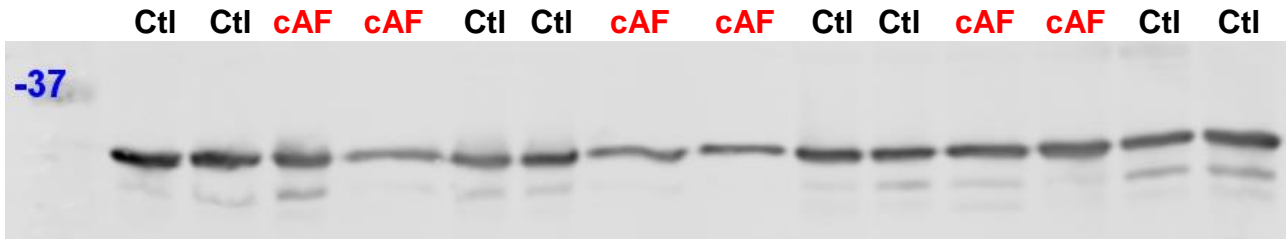
Gel 1



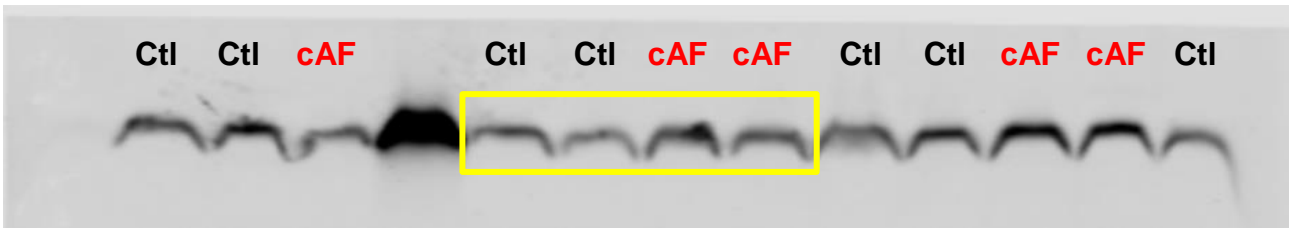
Gel 2



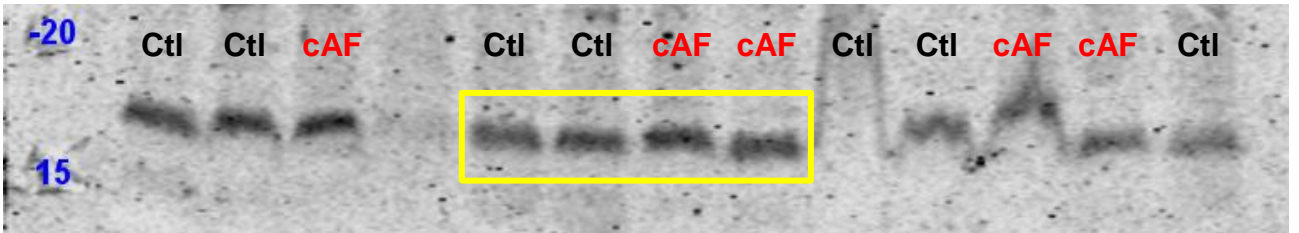
Gel 3



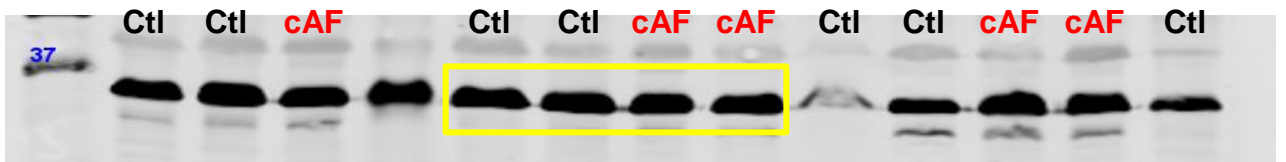
Total-CaM



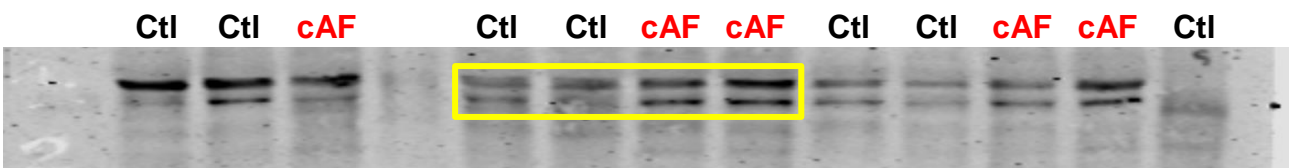
Thr80-CaM



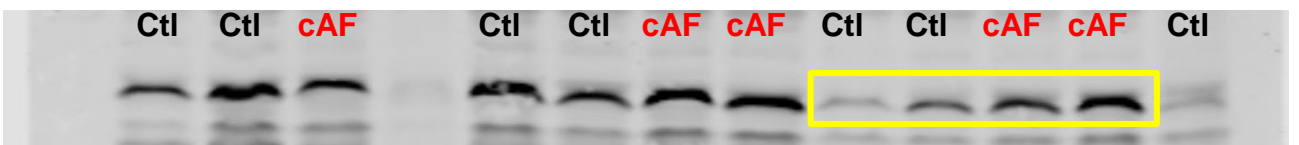
GAPDH



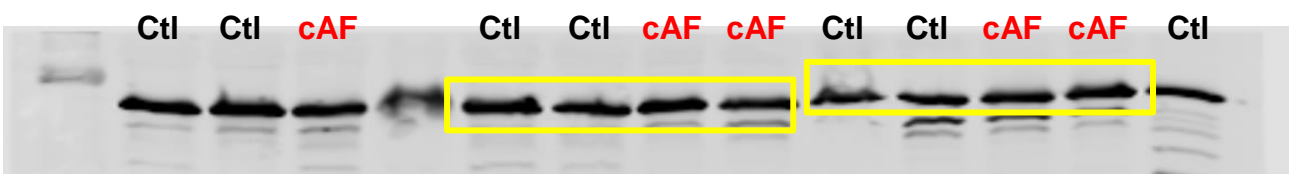
CKII α



PP2Ac



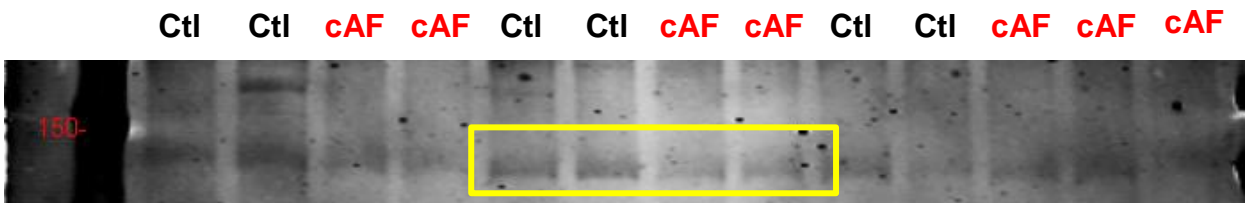
GAPDH



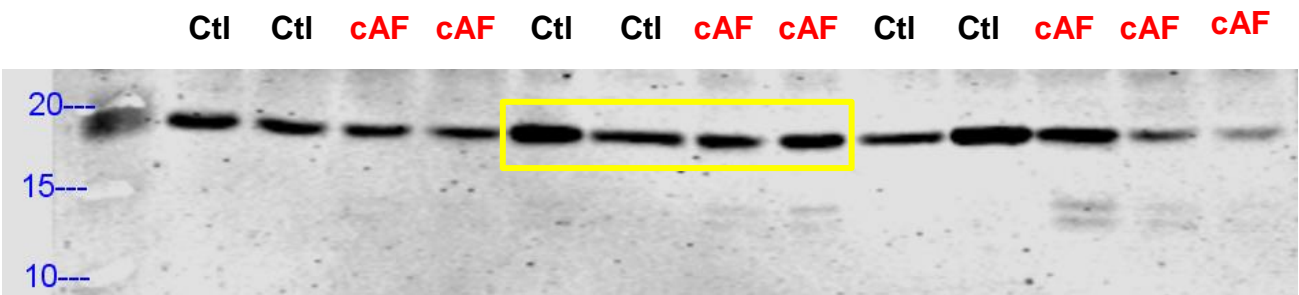
α -Act2
LA whole-tissue homogenates



α -Act2
RA cardiomyocytes



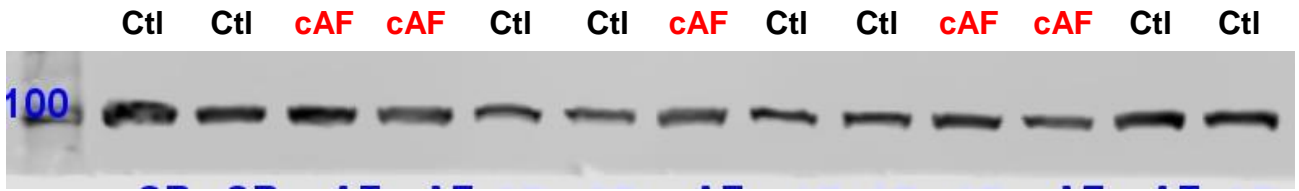
TnC
RA cardiomyocytes



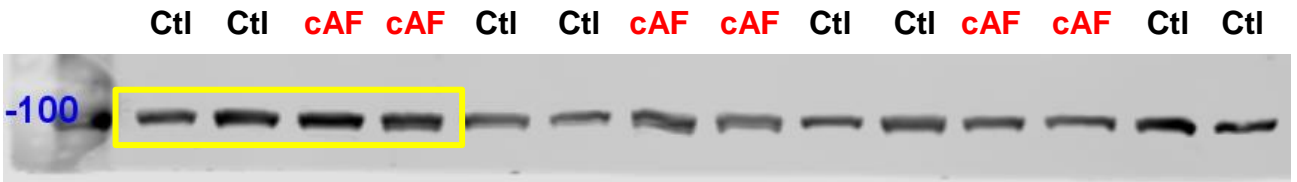
Same gel as shown on Slide 9

α -Act2

Gel 1



Gel 2

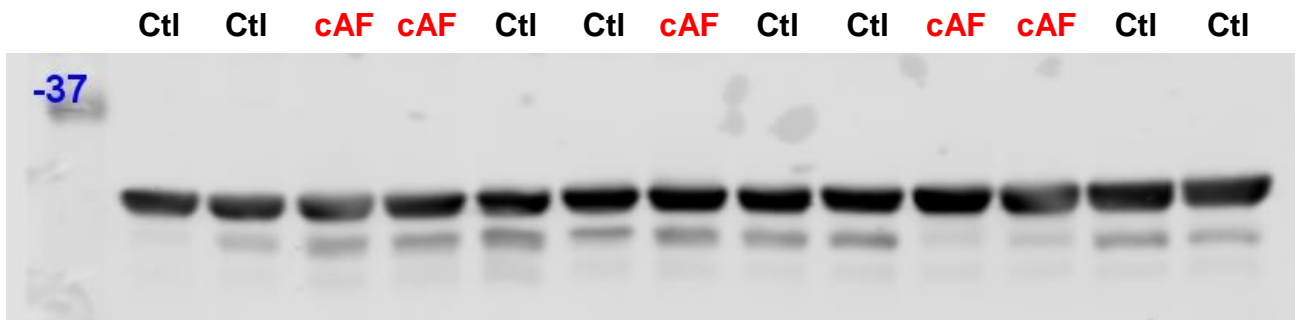


Gel 3

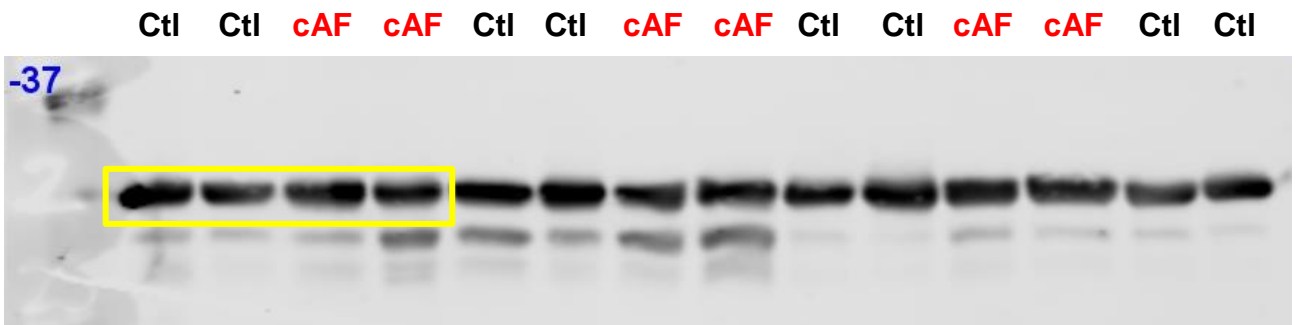


GAPDH

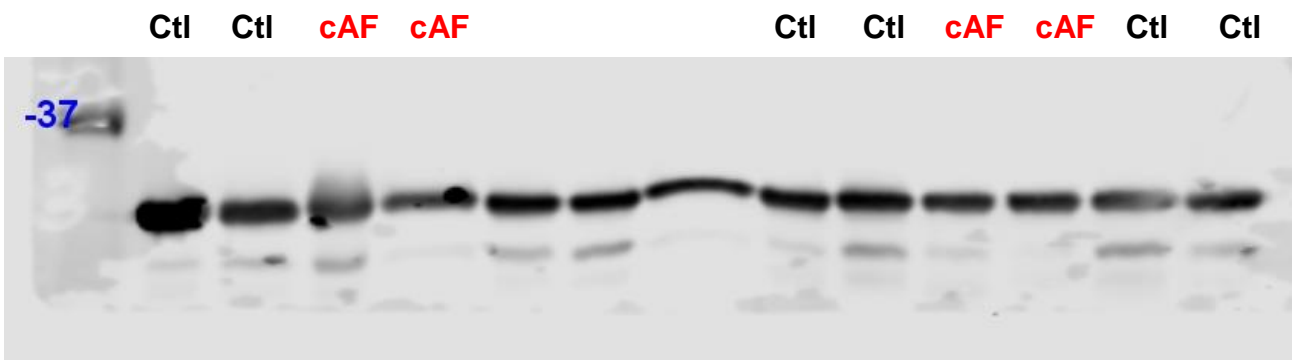
Gel 1



Gel 2

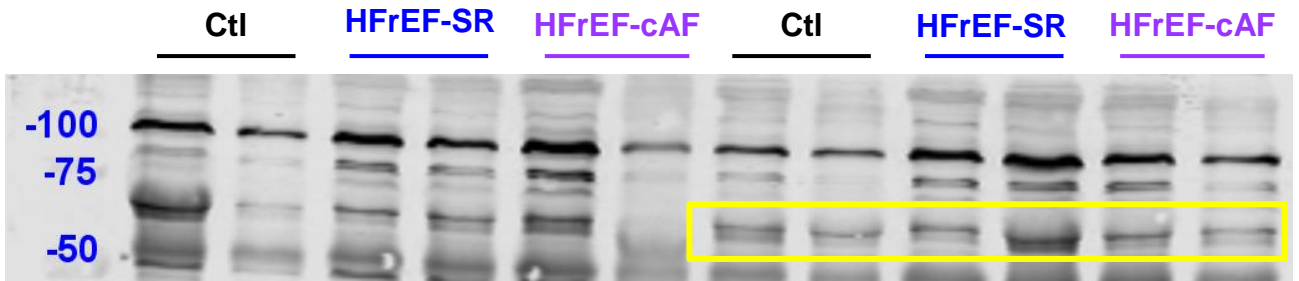


Gel 3

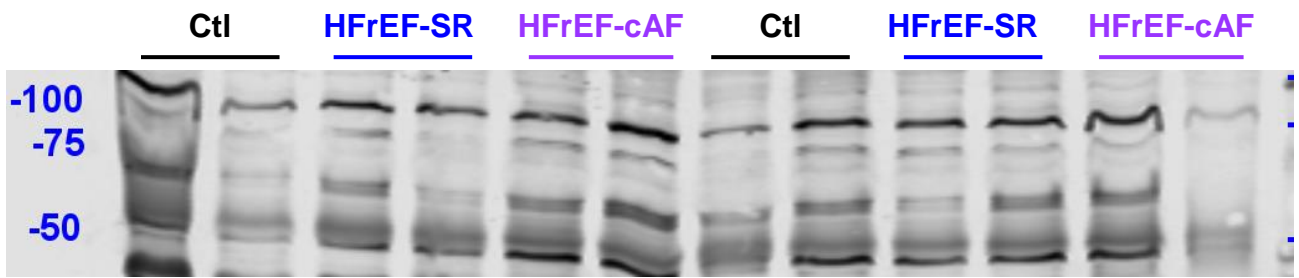


SK1

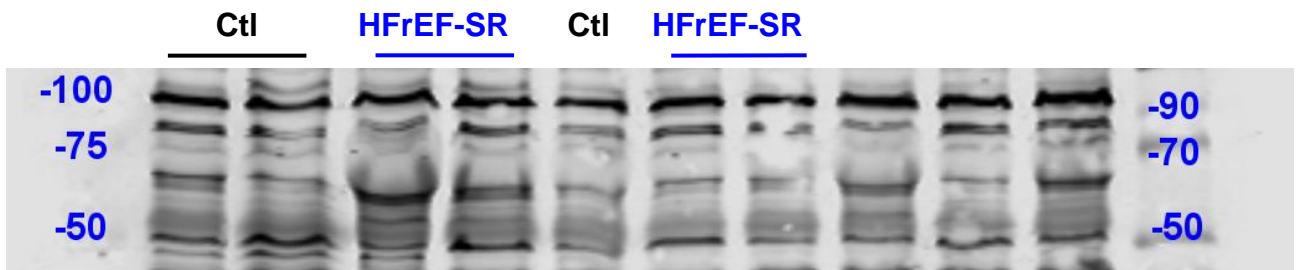
Gel 1



Gel 2

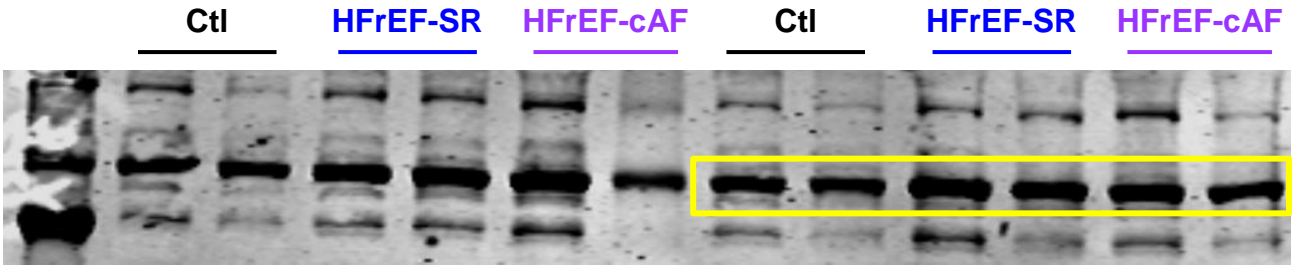


Gel 3

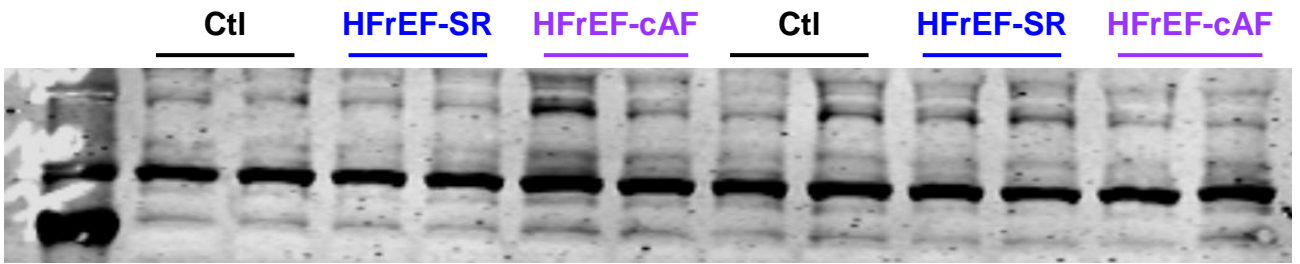


SK2

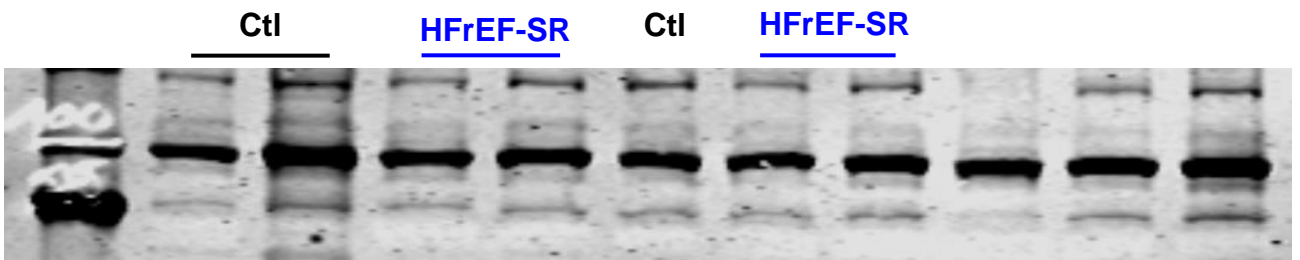
Gel 1



Gel 2

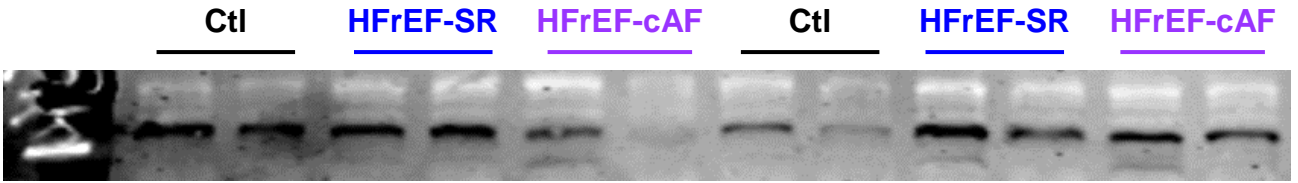


Gel 3

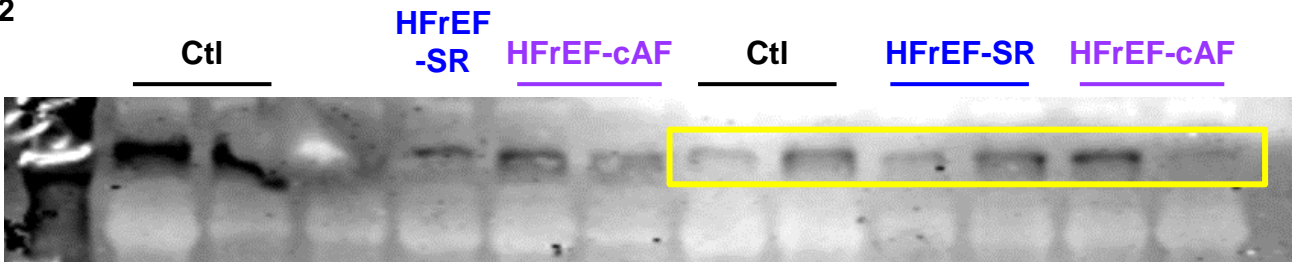


SK3

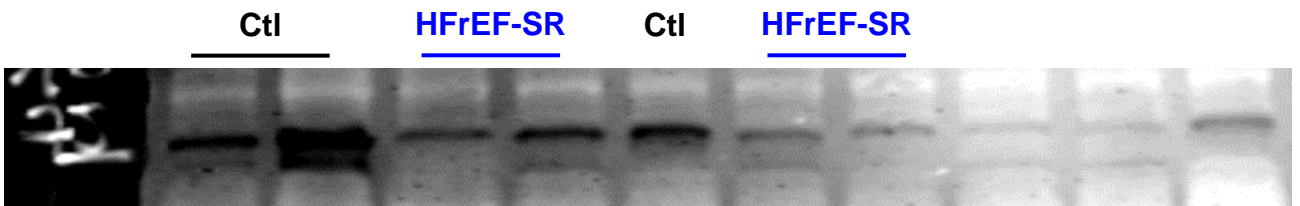
Gel 1



Gel 2

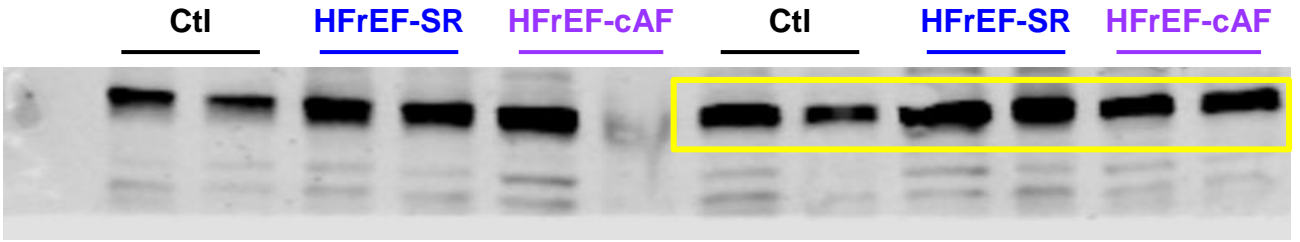


Gel 3

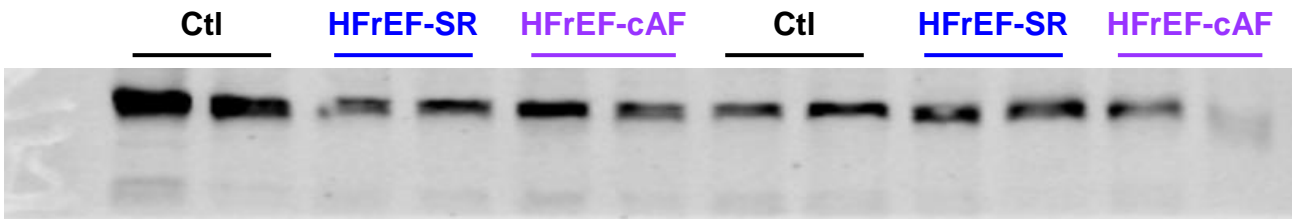


PP2Ac

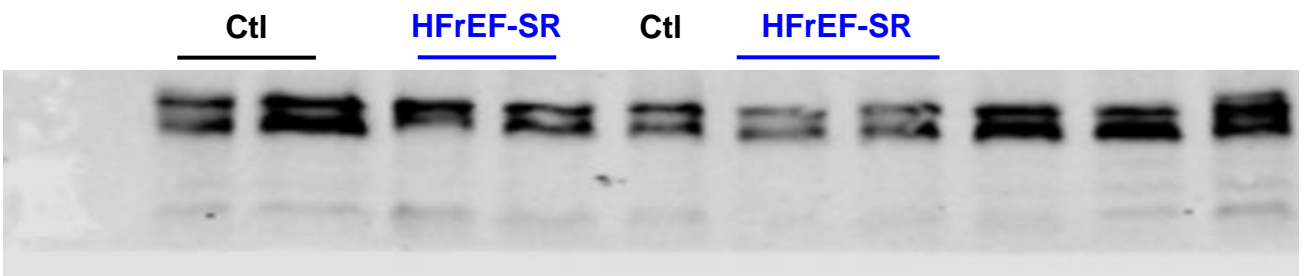
Gel 1



Gel 2

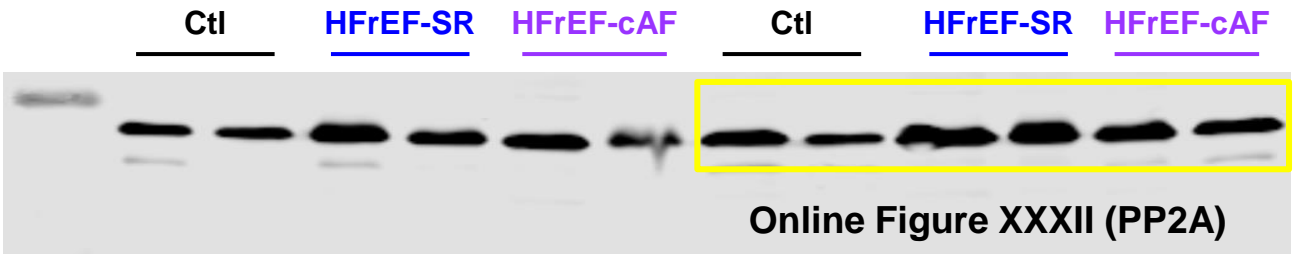


Gel 3

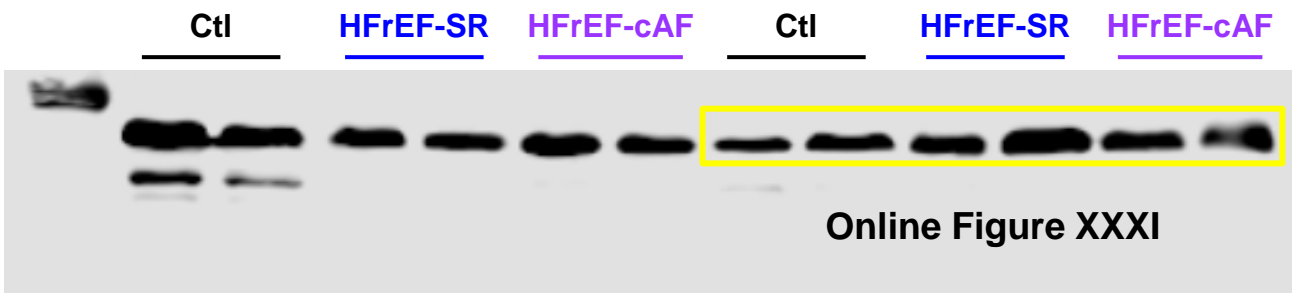


GAPDH

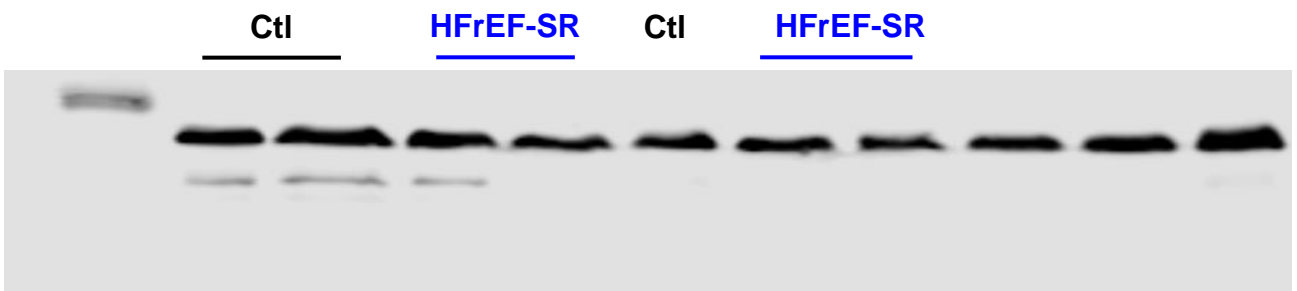
Gel 1



Gel 2

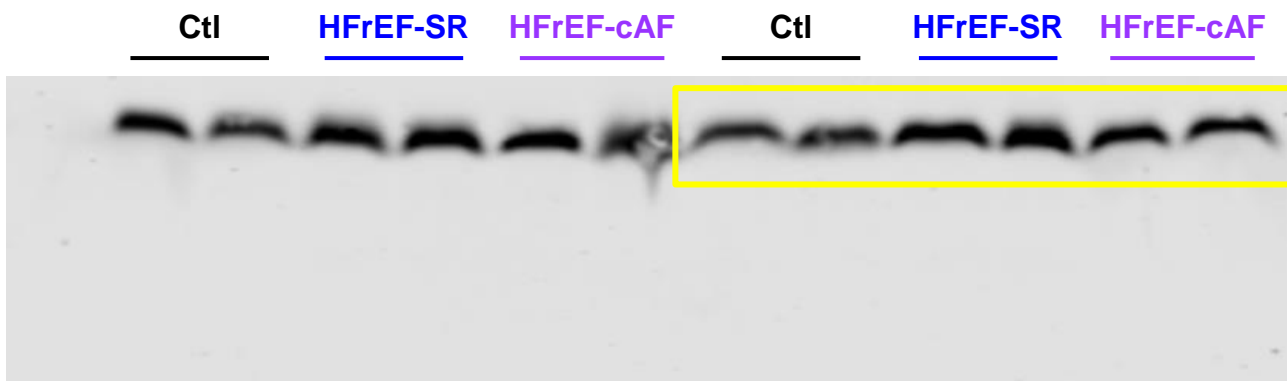


Gel 3

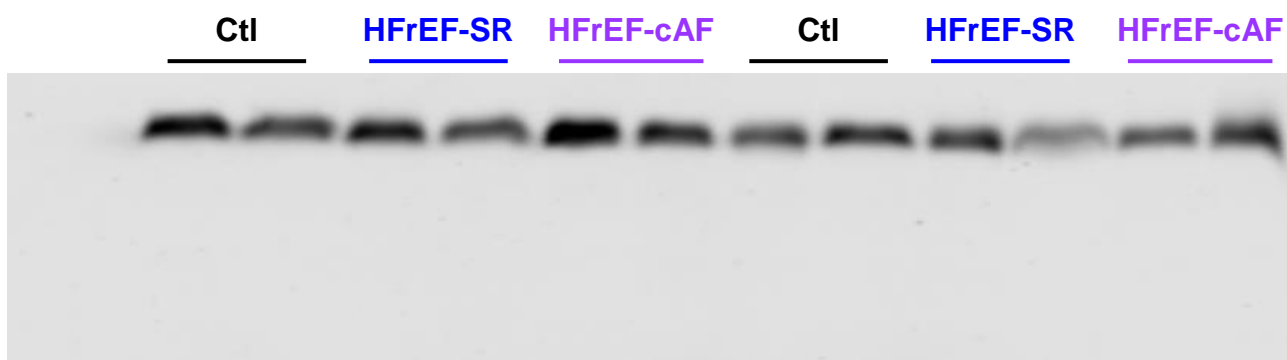


Total-CaM

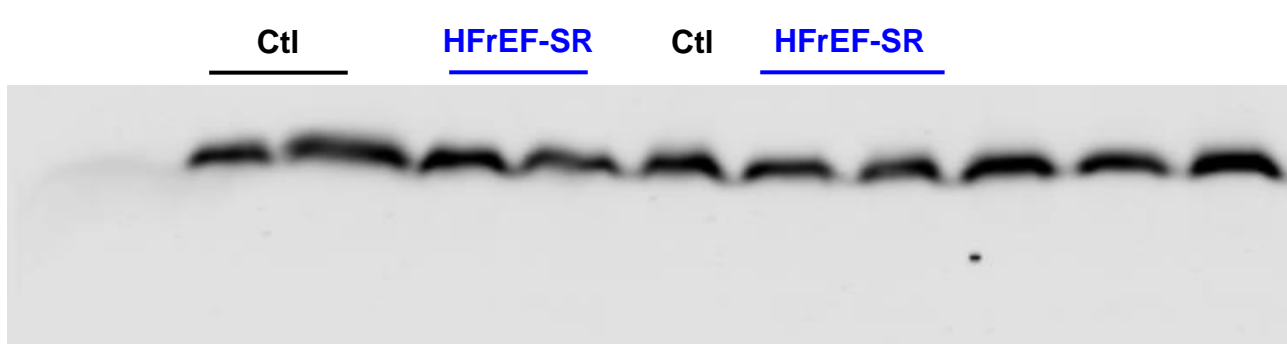
Gel 1



Gel 2

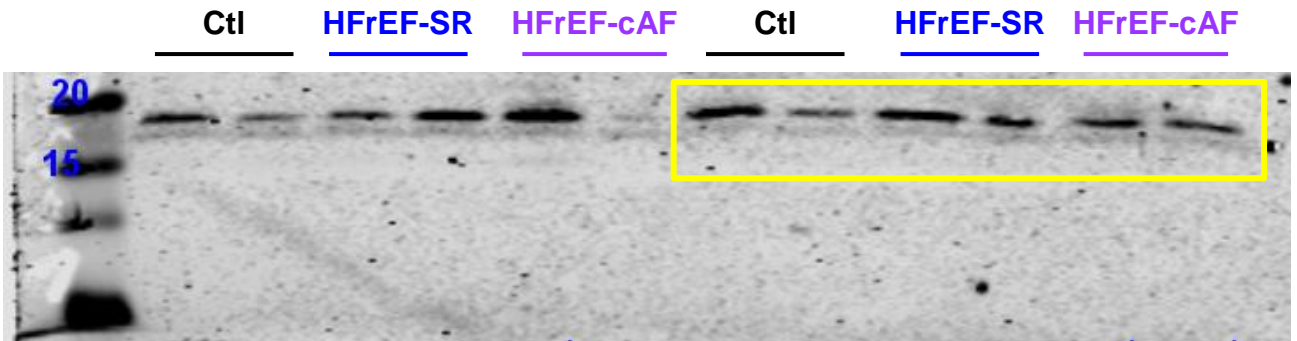


Gel 3

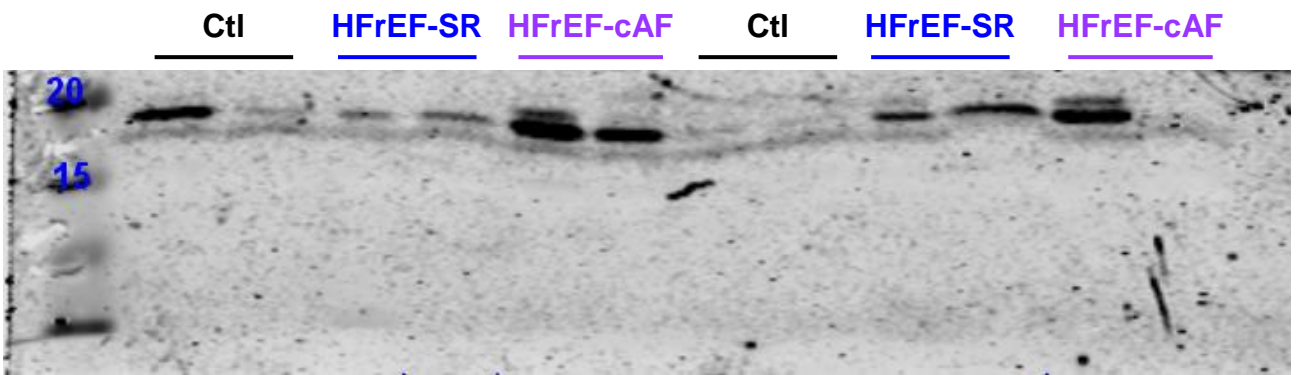


Thr80-CaM

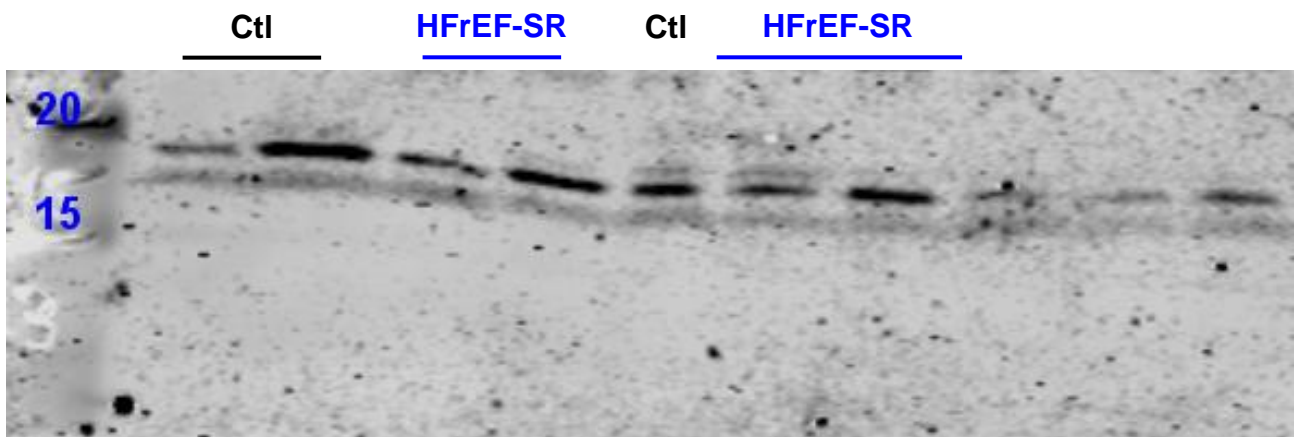
Gel 1



Gel 2

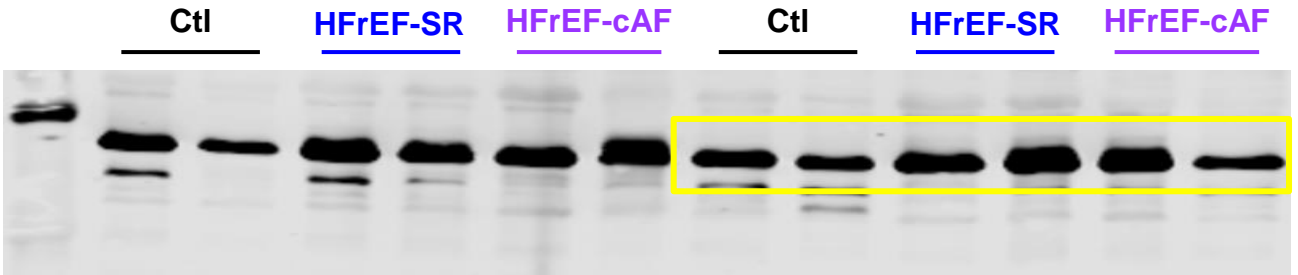


Gel 3

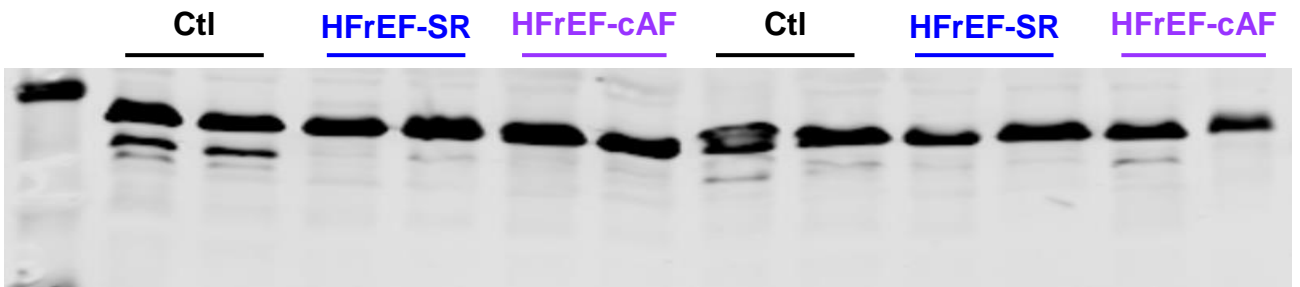


GAPDH

Gel 1



Gel 2



Gel 3

