

Supplementary information 2

Calcitriol promotes M2 polarization of tumor-associated macrophages in 4T1 mouse mammary gland cancer *via* the induction of proinflammatory cytokines

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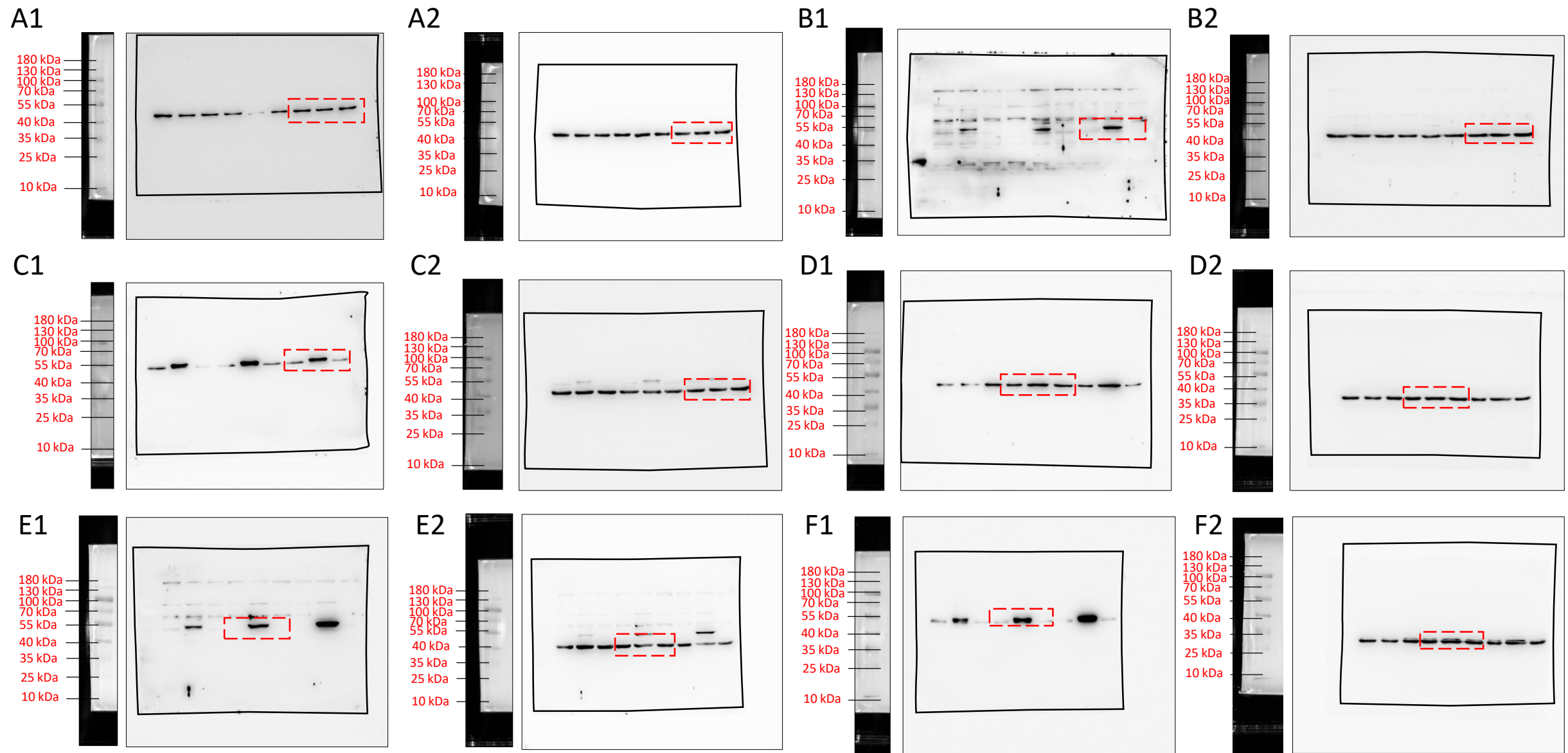


Figure S 1. **Uncropped western blot images corresponding to cropped blots presented in Figure 2 C in the manuscript.** A-C 4T1 cell line, D-F 67NR cell line. A1, D1 – CYP27B1; B1,E1 – CYP24A1; C1,F1 – VDR (vitamin D receptor); A2,B2,C2,D2,E2,F2 – loading control - β -actin. Chemiluminescence photographs with fragment of colorimetric picture showing protein ladder and the red box showing the cropping line. Visualization was performed on ChemiDoc Imaging System (Bio-Rad, Hercules, CA, USA) with exposure time estimated automatically.

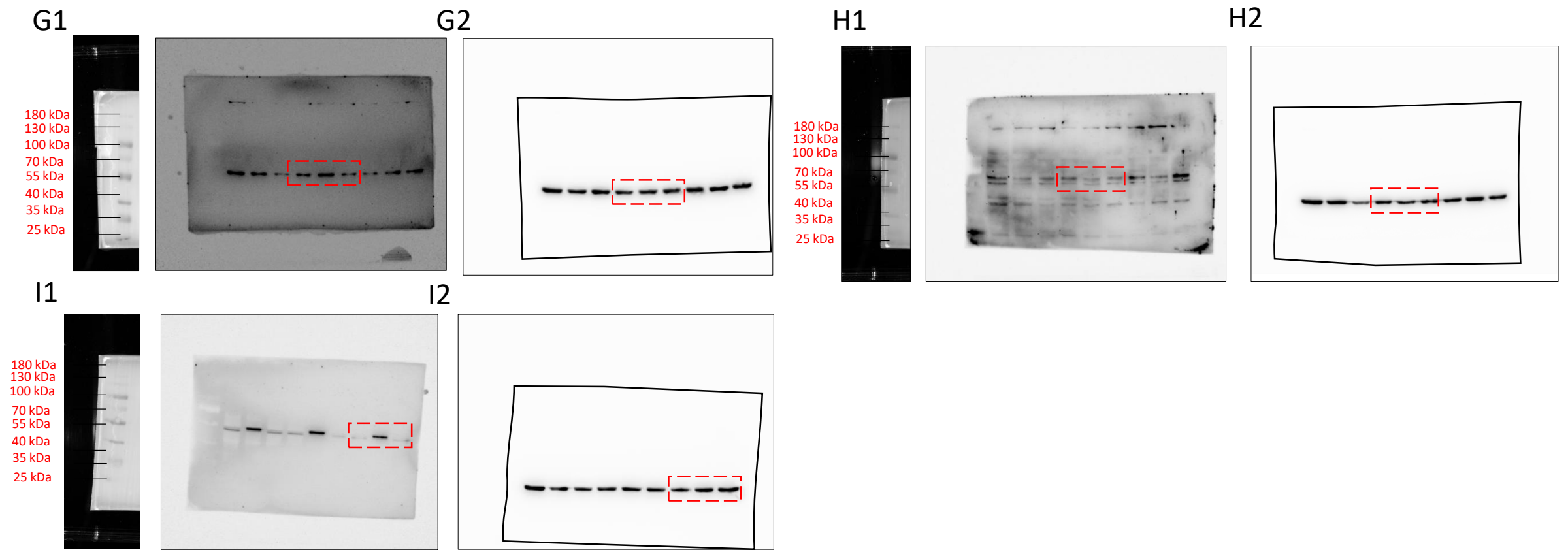


Figure S 1 c.d. **Uncropped western blot images corresponding to cropped blots presented in Figure 2 C in the manuscript.** G-I E0771cell line. G1 – CYP27B1; H1 – CYP24A1; I1 – VDR (vitamin D receptor); G2,H2,I2 – loading control - β -actin. Chemiluminescence photographs with fragment of colorimetric picture showing protein ladder and the red box showing the cropping line. Visualization was performed on ChemiDoc Imaging System (Bio-Rad, Hercules, CA, USA) with exposure time estimated automatically.

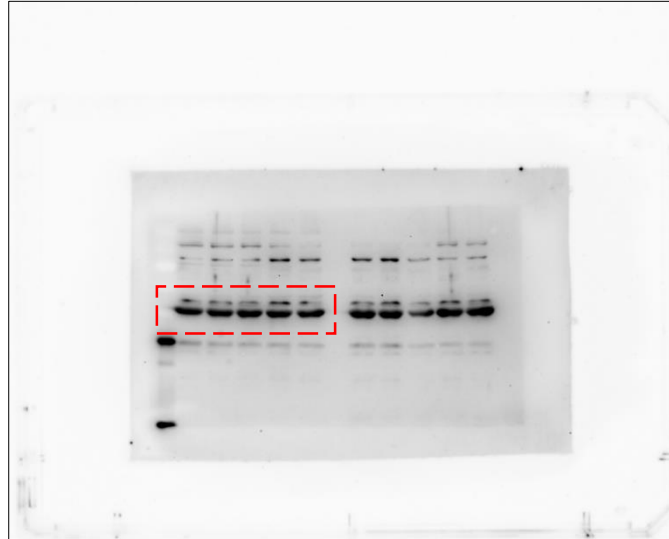
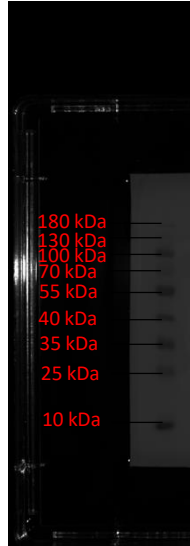
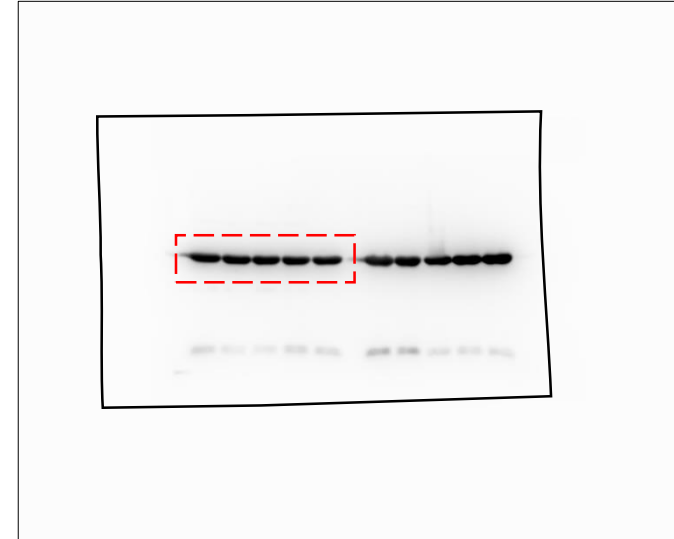
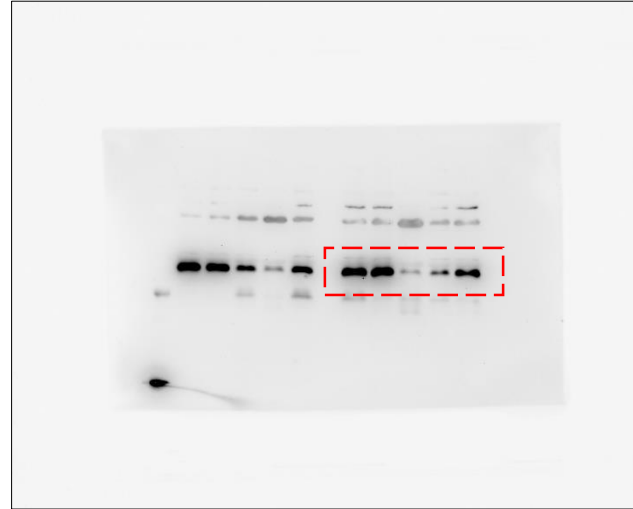
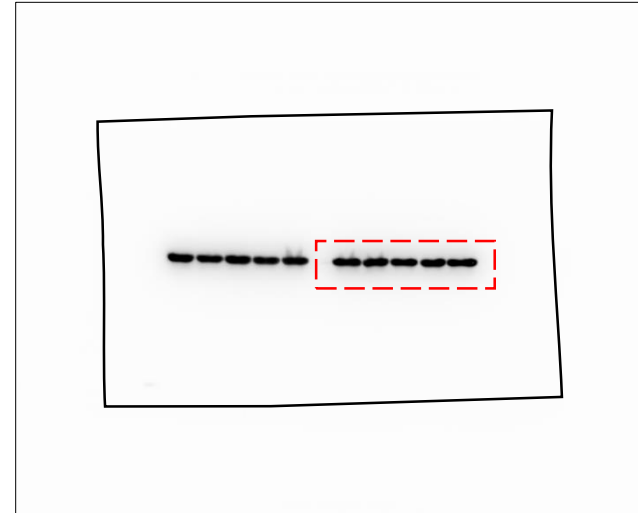
A1**A2**

Figure S 2. **Uncropped blot images corresponding to cropped blots presented in Figure 4 E in the manuscript.** A1 – EpCAM, A2 – loading control – β -actin. Chemiluminescence photographs with fragment of colorimetric picture showing protein ladder and the red box showing the cropping line. Visualization was performed on ChemiDoc Imaging System (Bio-Rad, Hercules, CA, USA) with exposure time estimated automatically.

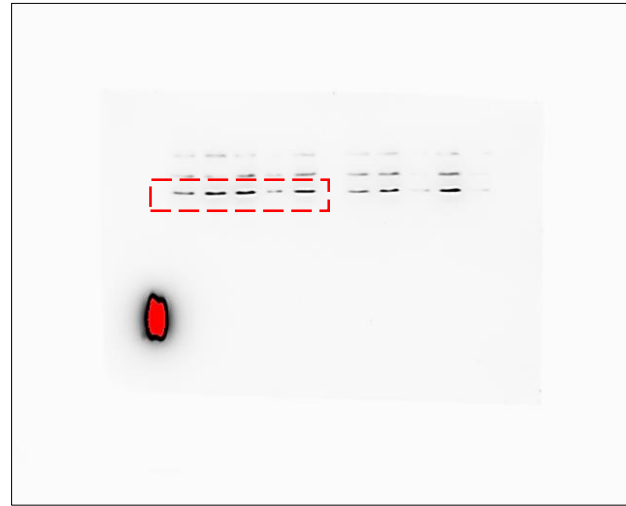
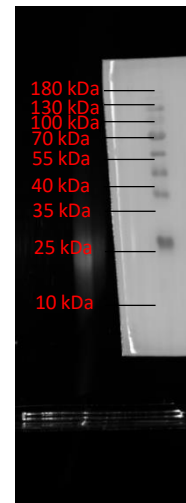
A1



A2



B1



B2

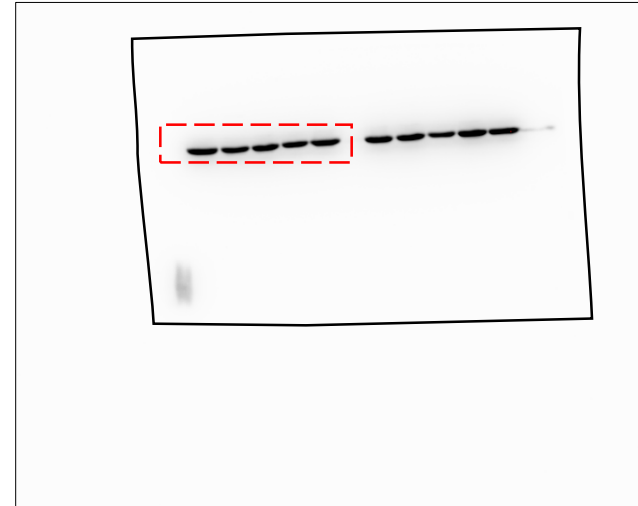


Figure S 3. **Uncropped blot images corresponding to cropped blots presented in Figure 5 E in the manuscript.** A1 – EpCAM; B1 – OPN (osteopontin). A2, B2 – loading control – β -actin. Chemiluminescence photographs with fragment of colorimetric picture showing protein ladder and the red box showing the cropping line. Visualization was performed on ChemiDoc Imaging System (Bio-Rad, Hercules, CA, USA) with exposure time estimated automatically.

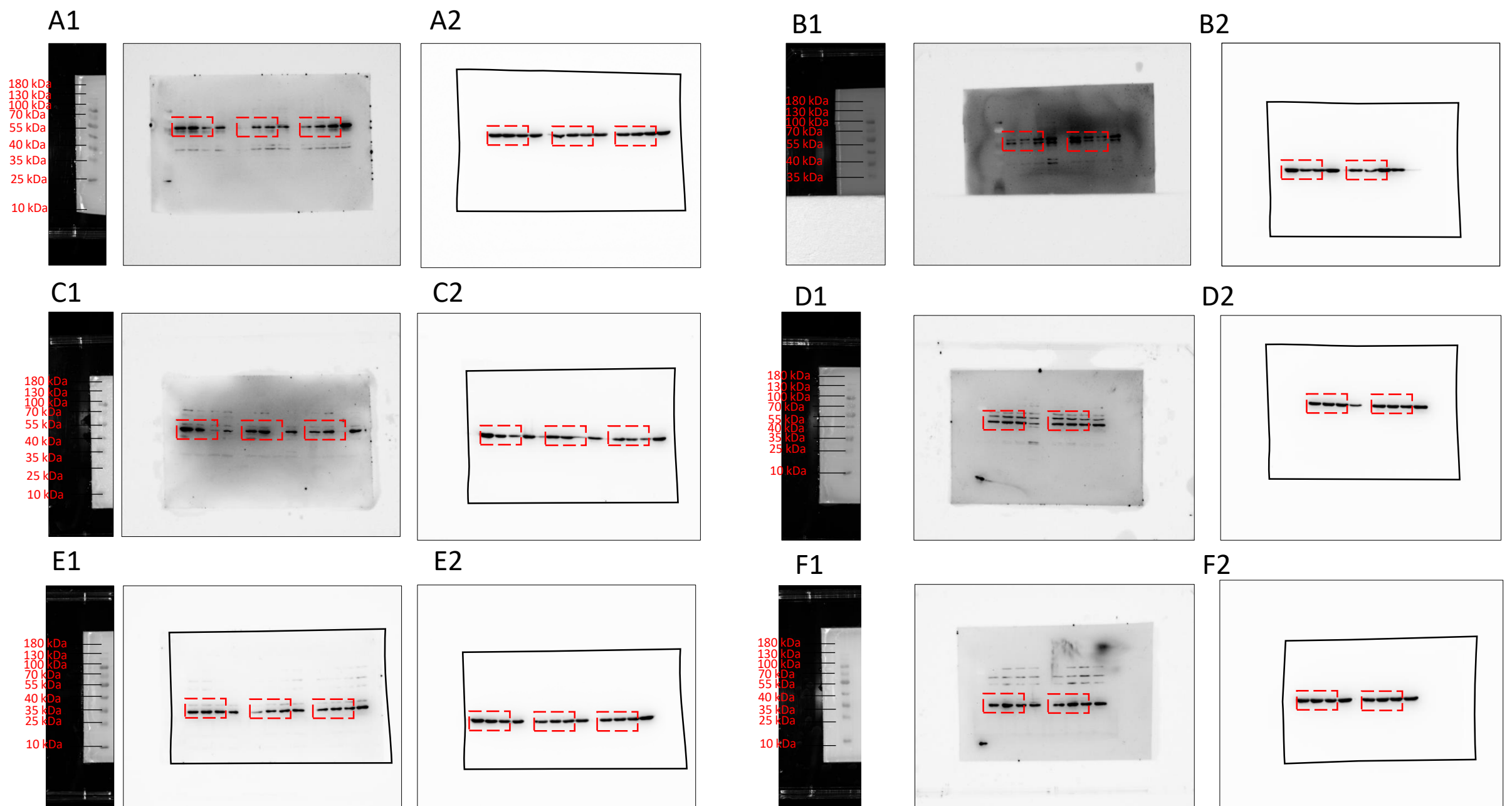


Figure S 4. **Uncropped blot images corresponding to cropped blots presented in Figure 7 E in the manuscript.** A1, B1 – IRF4; C1,D1 – MMP3; E1, F1 – TGFβ. A2, B2, C2, D2, E2, F2 – loading control – β-actin. Chemiluminescence photographs with fragment of colorimetric picture showing protein ladder and the red box showing the cropping line. Visualization was performed on ChemiDoc Imaging System (Bio-Rad, Hercules, CA, USA) with exposure time estimated automatically.

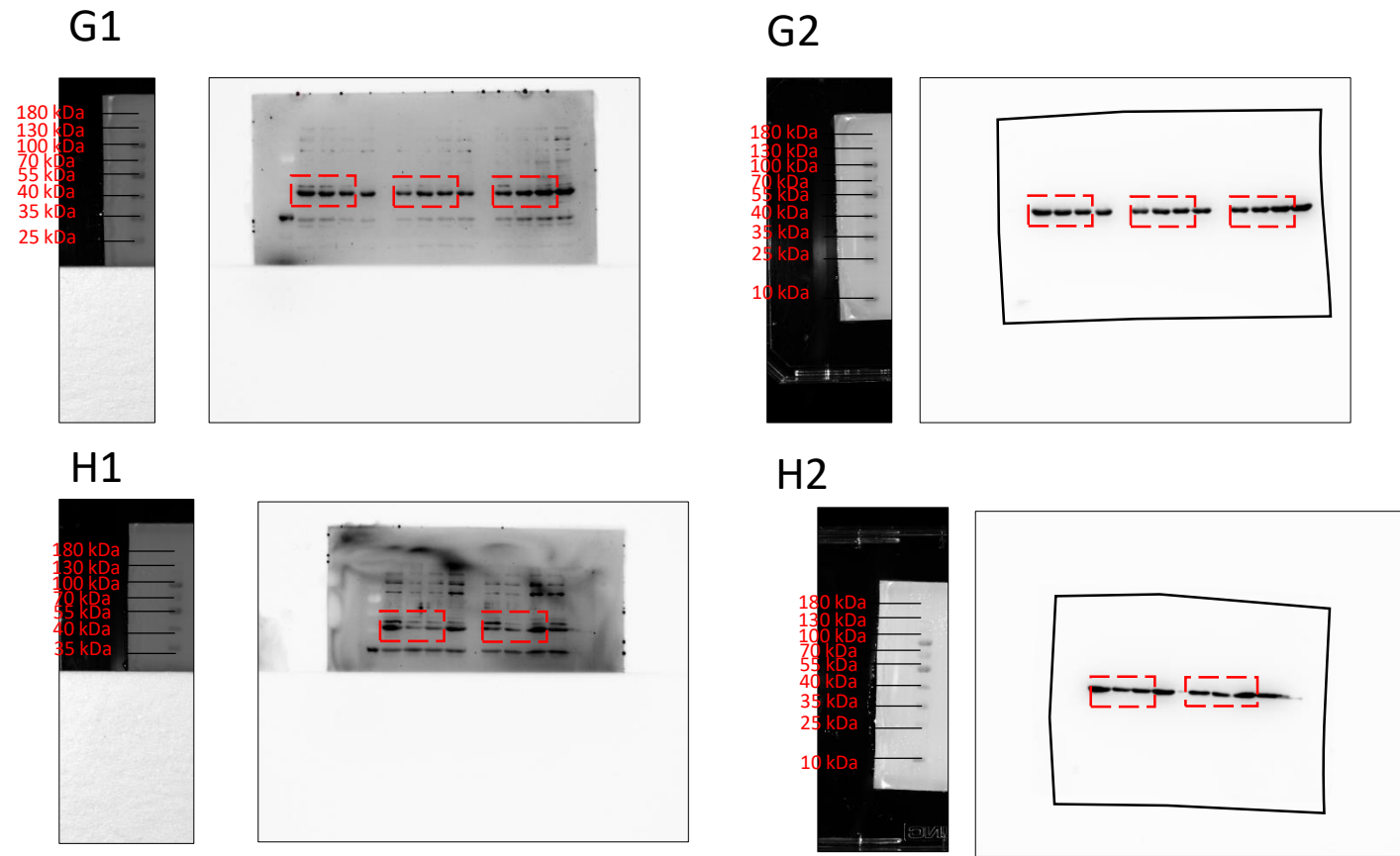


Figure S 4 c.d. **Uncropped blot images corresponding to cropped blots presented in Figure 7 E in the manuscript. G1,H1 – EpCAM; G2, H2 loading control – β -actin.** Chemiluminescence photographs with fragment of colorimetric picture showing protein ladder and the red box showing the cropping line. Visualization was performed on ChemiDoc Imaging System (Bio-Rad, Hercules, CA, USA) with exposure time estimated automatically.

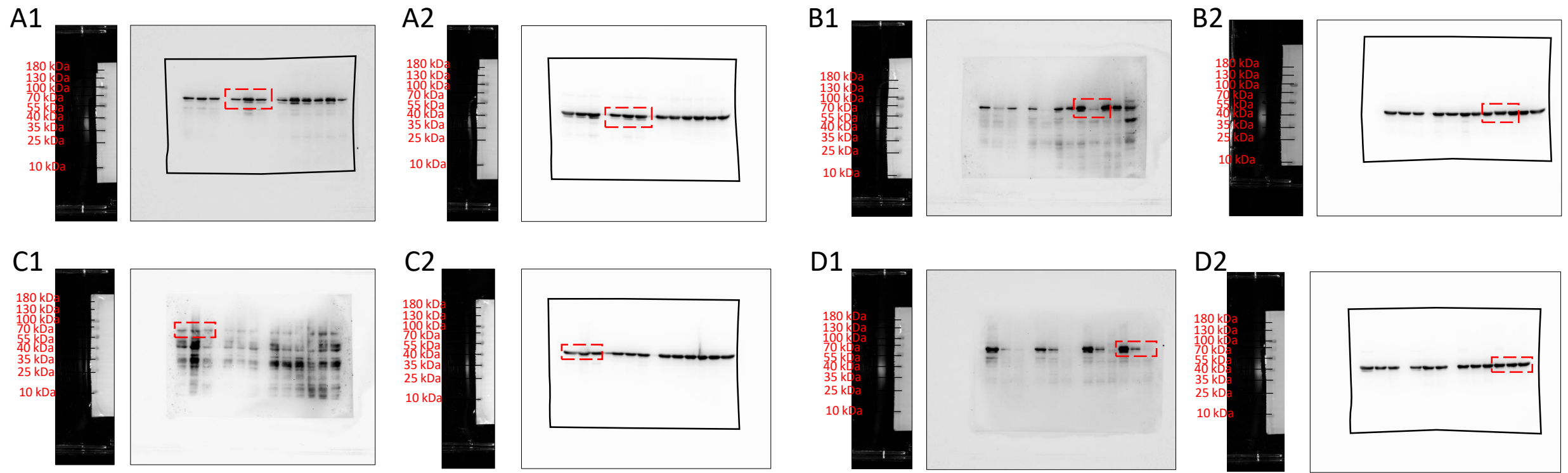


Figure S 5. **Uncropped blot images corresponding to cropped blots presented in Figure 8 C (A-C) and D (D) in the manuscript.** A – 4T1 cell line, B – 67NR cell line, C – E0771 cell line, D – comparison of all tested cell lines. A1, B1, C1, D1 – COX2 (cyclooxygenase 2); A2, B2, C2, D2 – loading control- β -actin. Chemiluminescence photographs with fragment of colorimetric picture showing protein ladder and the red box showing the cropping line. Visualization was performed on ChemiDoc Imaging System (Bio-Rad, Hercules, CA, USA) with exposure time estimated automatically.

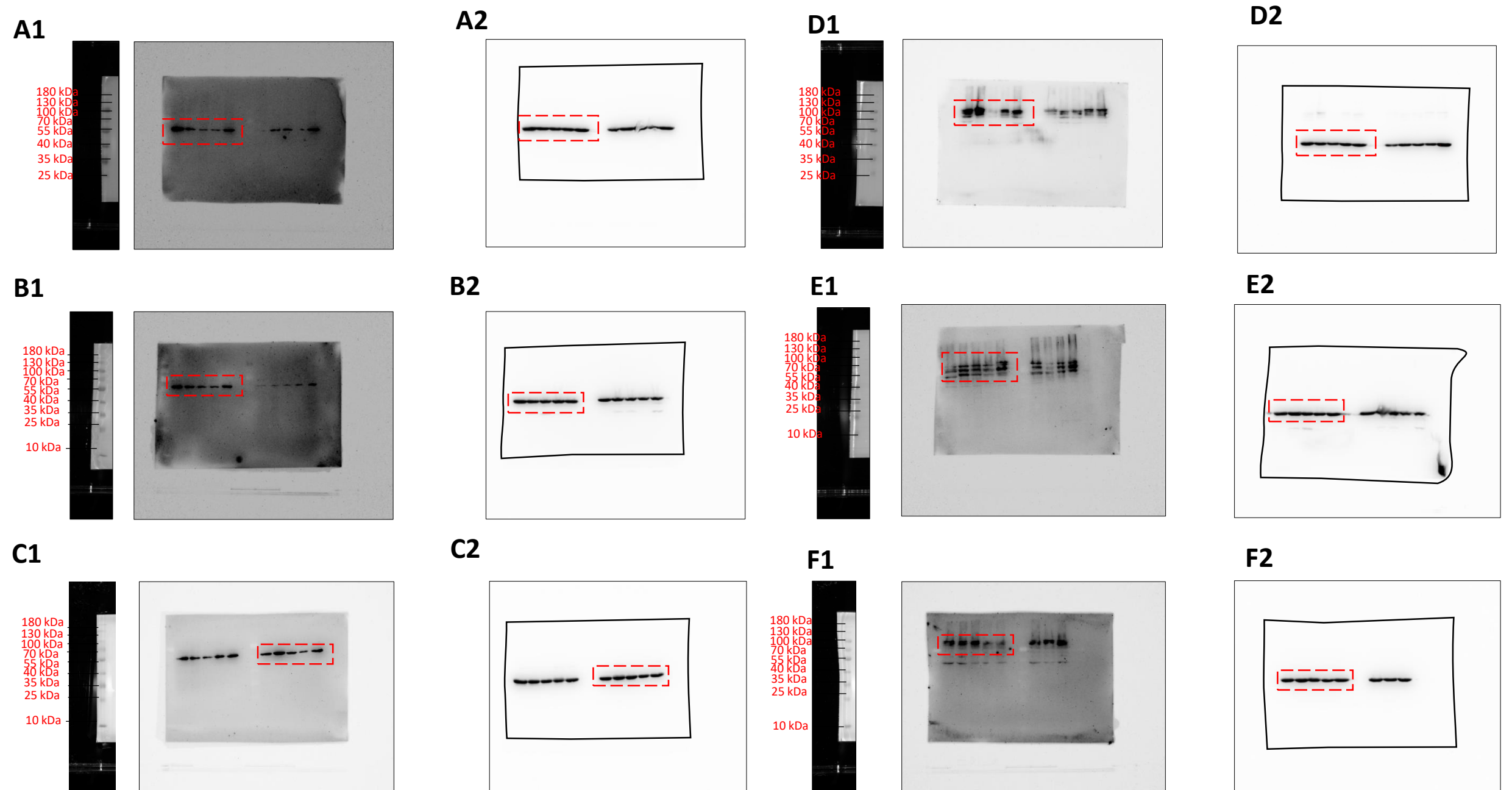


Figure S 6. Uncropped blot images corresponding to cropped blots presented in Figure S 1 B in the supplementary file 1. A1, B1, C1 - PDIA3; D1, E1, F1 - β -catenin. A2, B2, C2, D2, E2, F2 – loading control – β -actin. A,D - 4T1, B,E - 67NR, C, F - E0771. Chemiluminescence photographs with fragment of colorimetric picture showing protein ladder and the red box showing the cropping line. Visualization was performed on ChemiDoc Imaging System (Bio-Rad, Hercules, CA, USA) with exposure time estimated automatically.

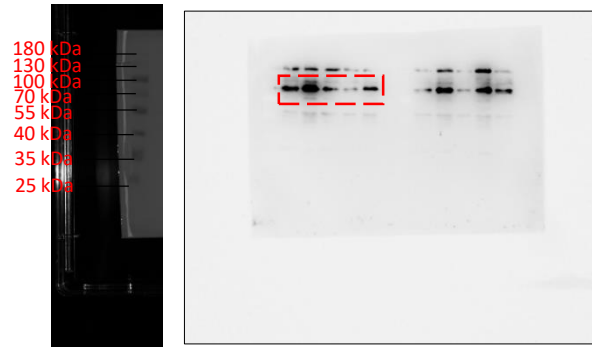
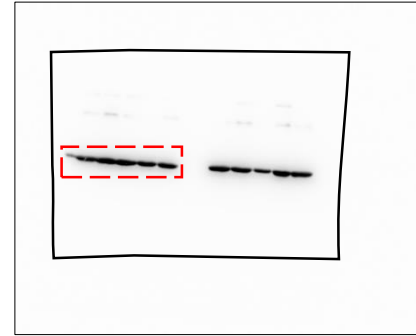
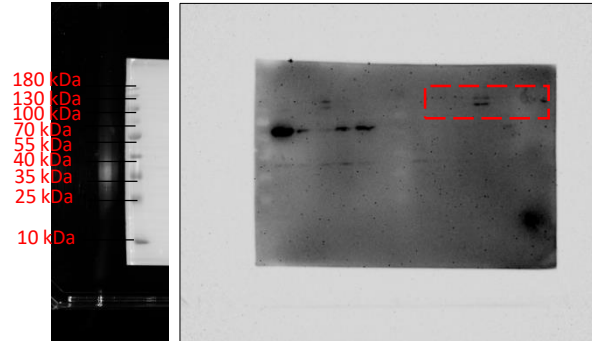
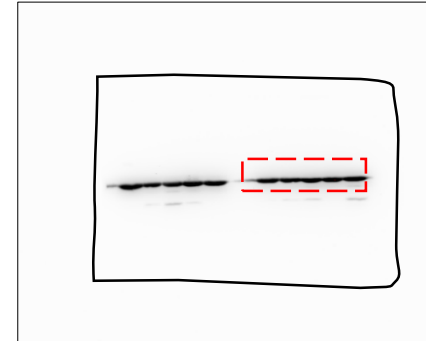
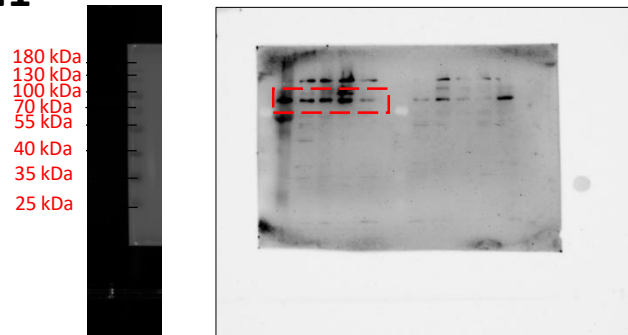
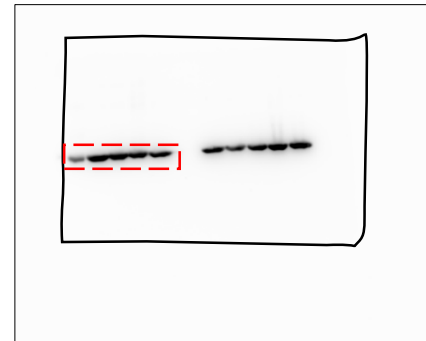
G1**G2****H1****H2****I1****I2**

Figure S 6 c.d.. **Uncropped blot images corresponding to cropped blots presented in Figure S 1 B in the supplementary file 1. G1, H1, I1 - E-cadherin.; G2, H2, I2 - loading control – β -actin. G - 4T1, H - 67NR, I - E0771.** Chemiluminescence photographs with fragment of colorimetric picture showing protein ladder and the red box showing the cropping line. Visualization was performed on ChemiDoc Imaging System (Bio-Rad, Hercules, CA, USA) with exposure time estimated automatically.

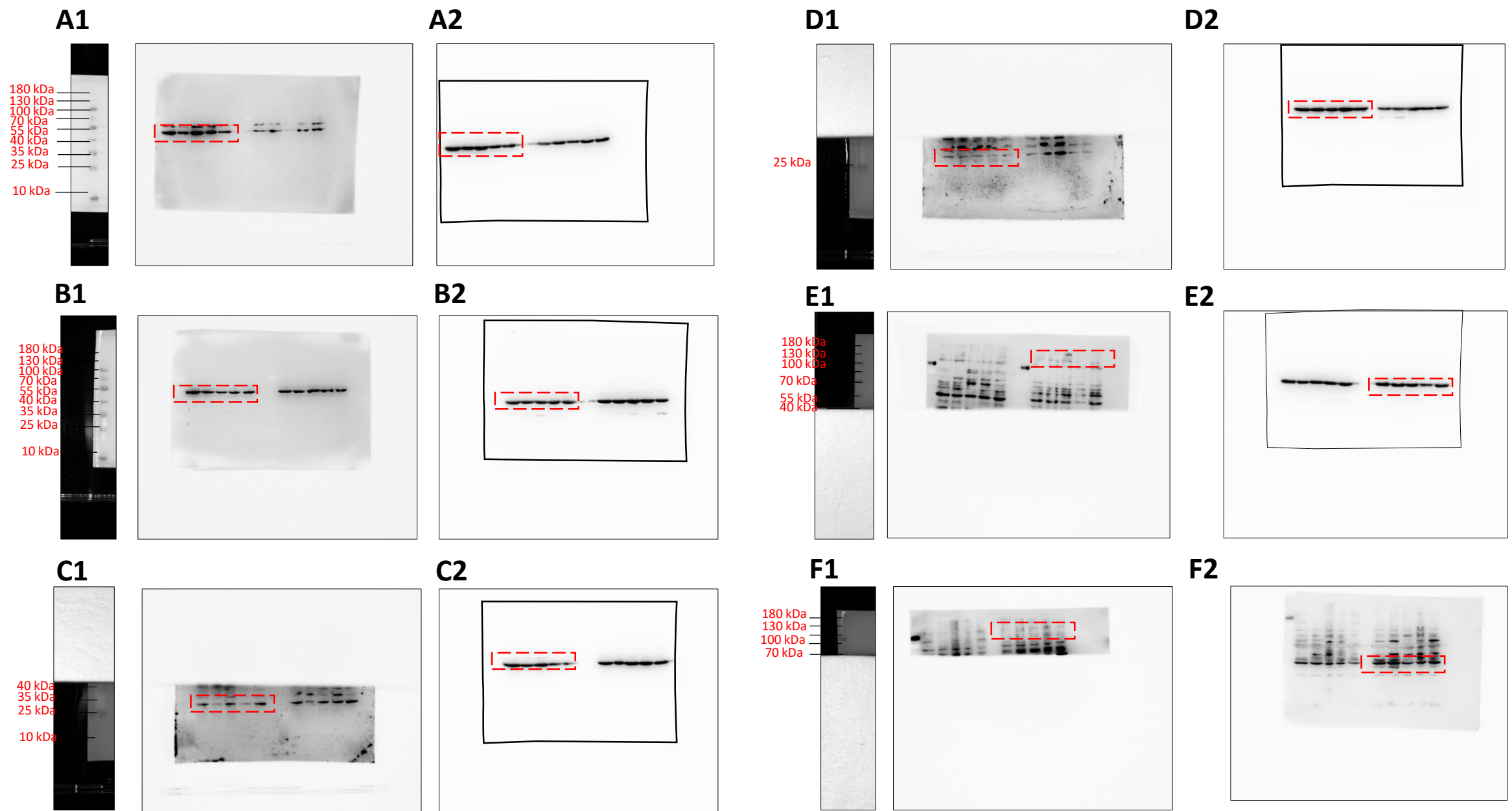


Figure S 6. **Uncropped blot images corresponding to cropped blots presented in Figure S 1 C in the supplementary file 1.** A1, B1 - MMP3; C1, D1 – PTHLH; E1, F1 – DIS3. A2, B2, C2, D2, E2, F2 - loading control – β -actin. A, C, E - 4T1; B, D, F - 67NR. Chemiluminescence photographs with fragment of colorimetric picture showing protein ladder and the red box showing the cropping line. Visualization was performed on ChemiDoc Imaging System (Bio-Rad, Hercules, CA, USA) with exposure time estimated automatically.

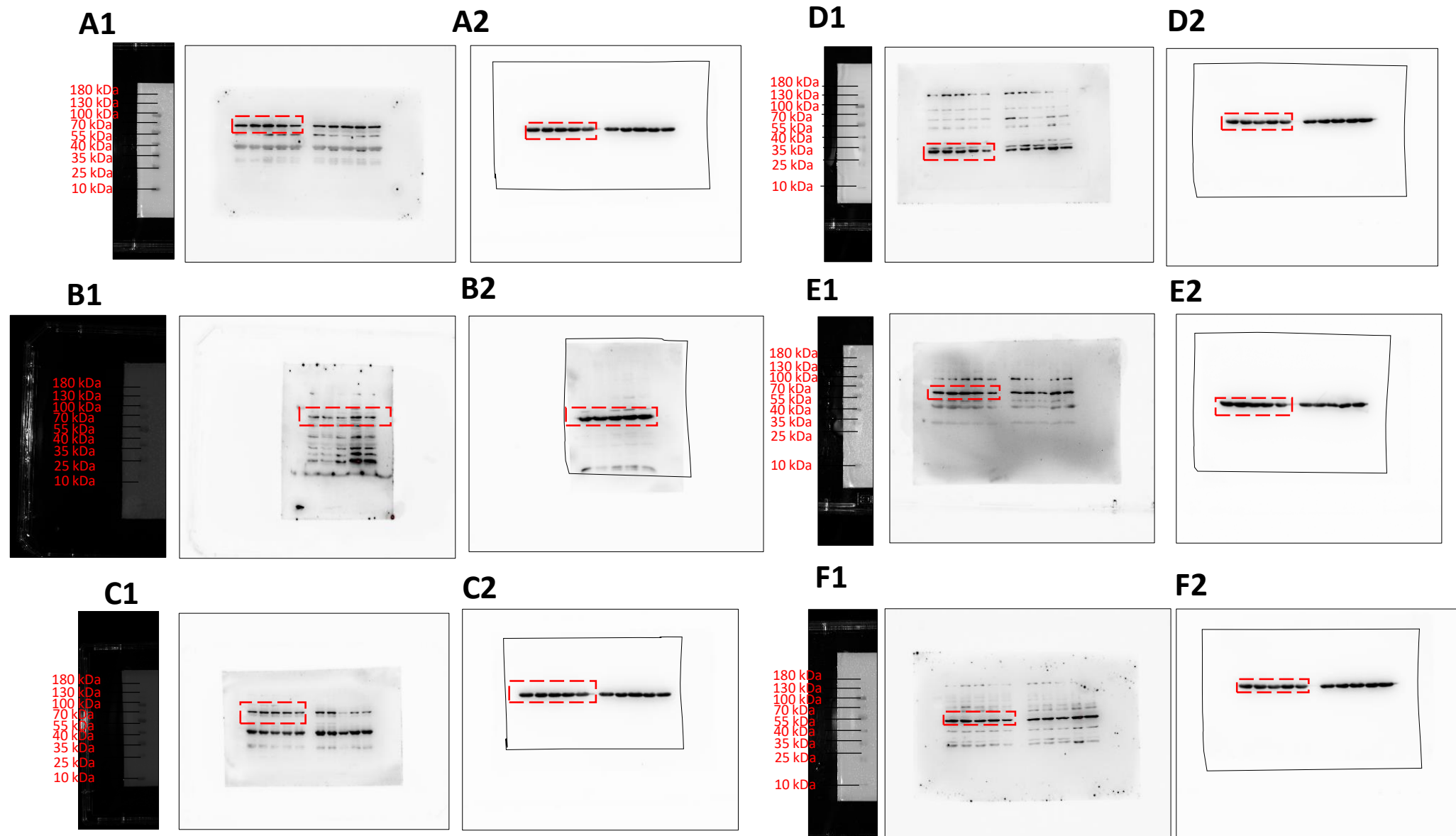


Figure S 7. **Uncropped blot images corresponding to cropped blots presented in Figure S 5 B in the supplementary file 1.** A1. CYP27B1, B1 VDR, C1 OPN, D1 TGFβ, E1 PDIA3, F1 MMP3; A2, B2, C2, D2, E2, F2 - loading control – β-actin. A,C, E - 4T1; B, D, F - 67NR. Chemiluminescence photographs with fragment of colorimetric picture showing protein ladder and the red box showing the cropping line. Visualization was performed on ChemiDoc Imaging System (Bio-Rad, Hercules, CA, USA) with exposure time estimated automatically.

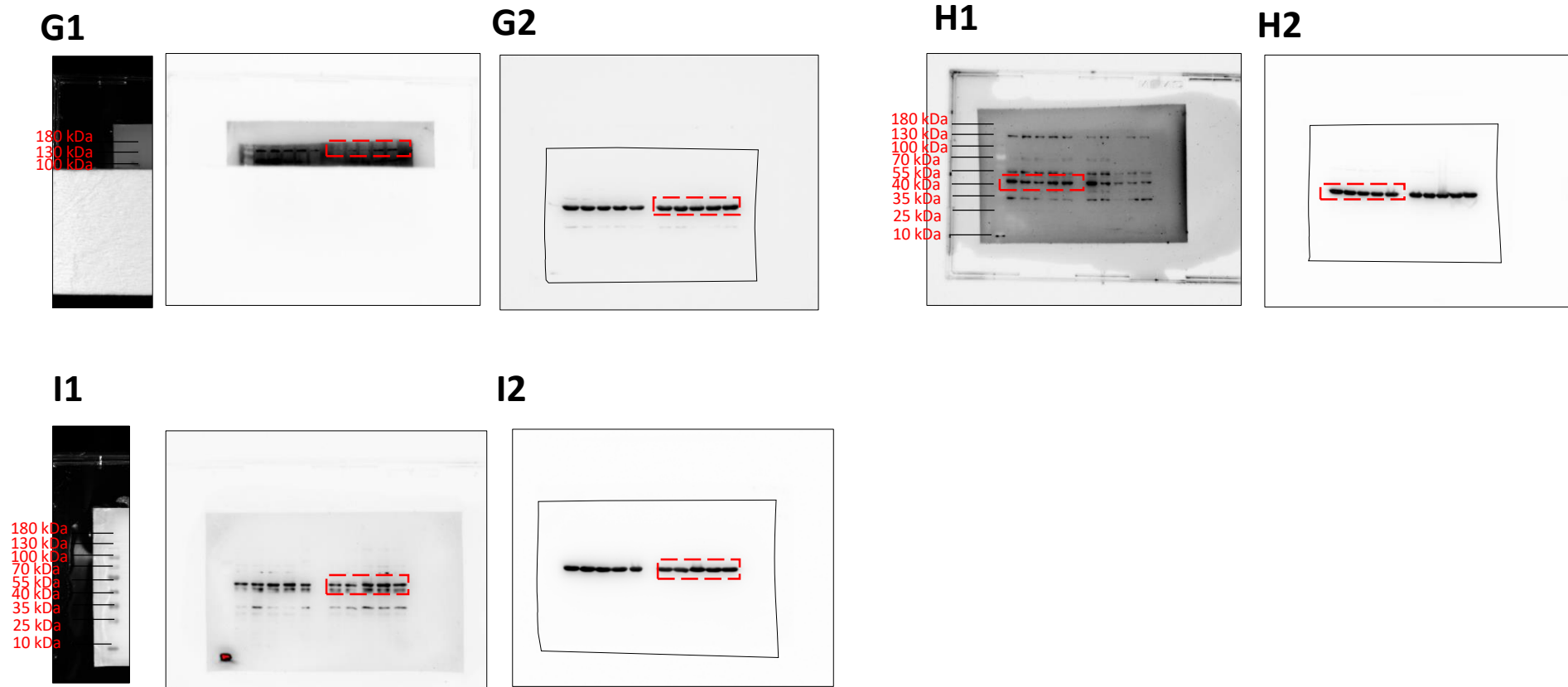


Figure S 7. **Uncropped blot images corresponding to cropped blots presented in Figure S 5 B in the supplementary file 1.** G1 Zeb1, H1 AP-1, I1 IRF4; G2, H2, I2 - loading control – β -actin. Chemiluminescence photographs with fragment of colorimetric picture showing protein ladder and the red box showing the cropping line. Visualization was performed on ChemiDoc Imaging System (Bio-Rad, Hercules, CA, USA) with exposure time estimated automatically.

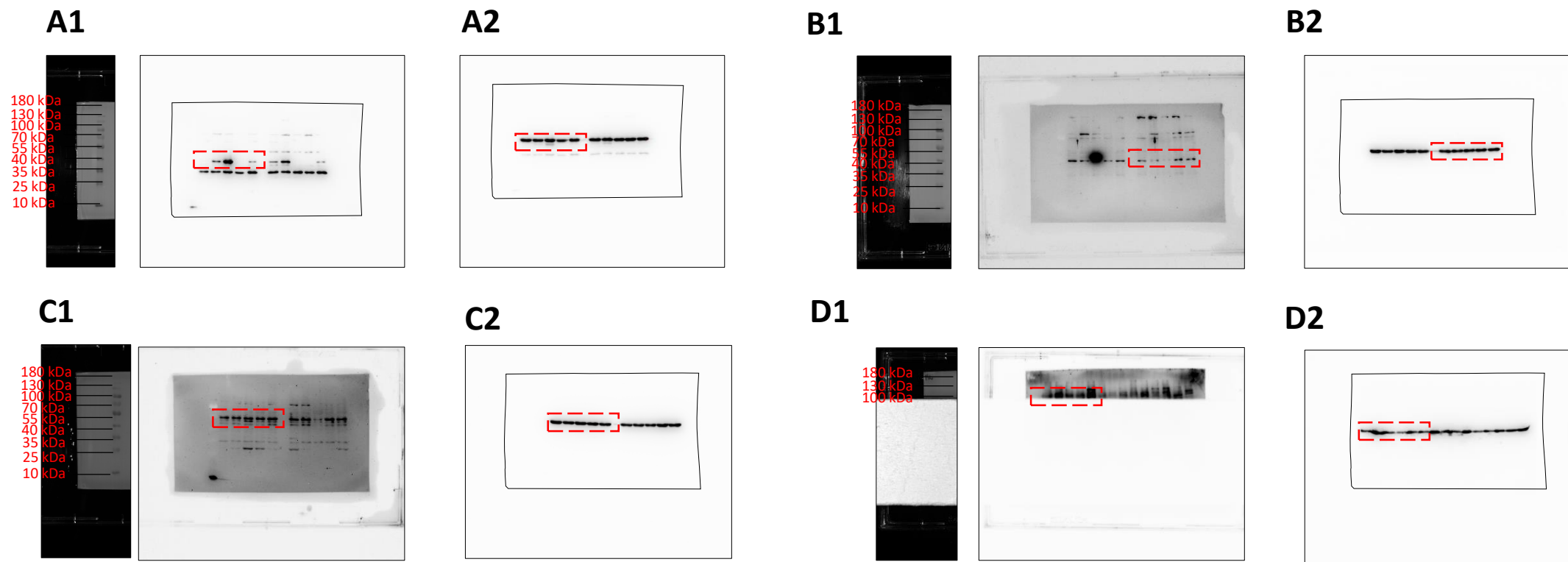


Figure S 7. **Uncropped blot images corresponding to cropped blots presented in Figure S 5 C in the supplementary file 1.** A1 TGFβ, B1, AP-1, C1 IRF4, D1 Zeb-1; A2,B2,C2,D2 -- loading control – β-actin. Chemiluminescence photographs with fragment of colorimetric picture showing protein ladder and the red box showing the cropping line. Visualization was performed on ChemiDoc Imaging System (Bio-Rad, Hercules, CA, USA) with exposure time estimated automatically.

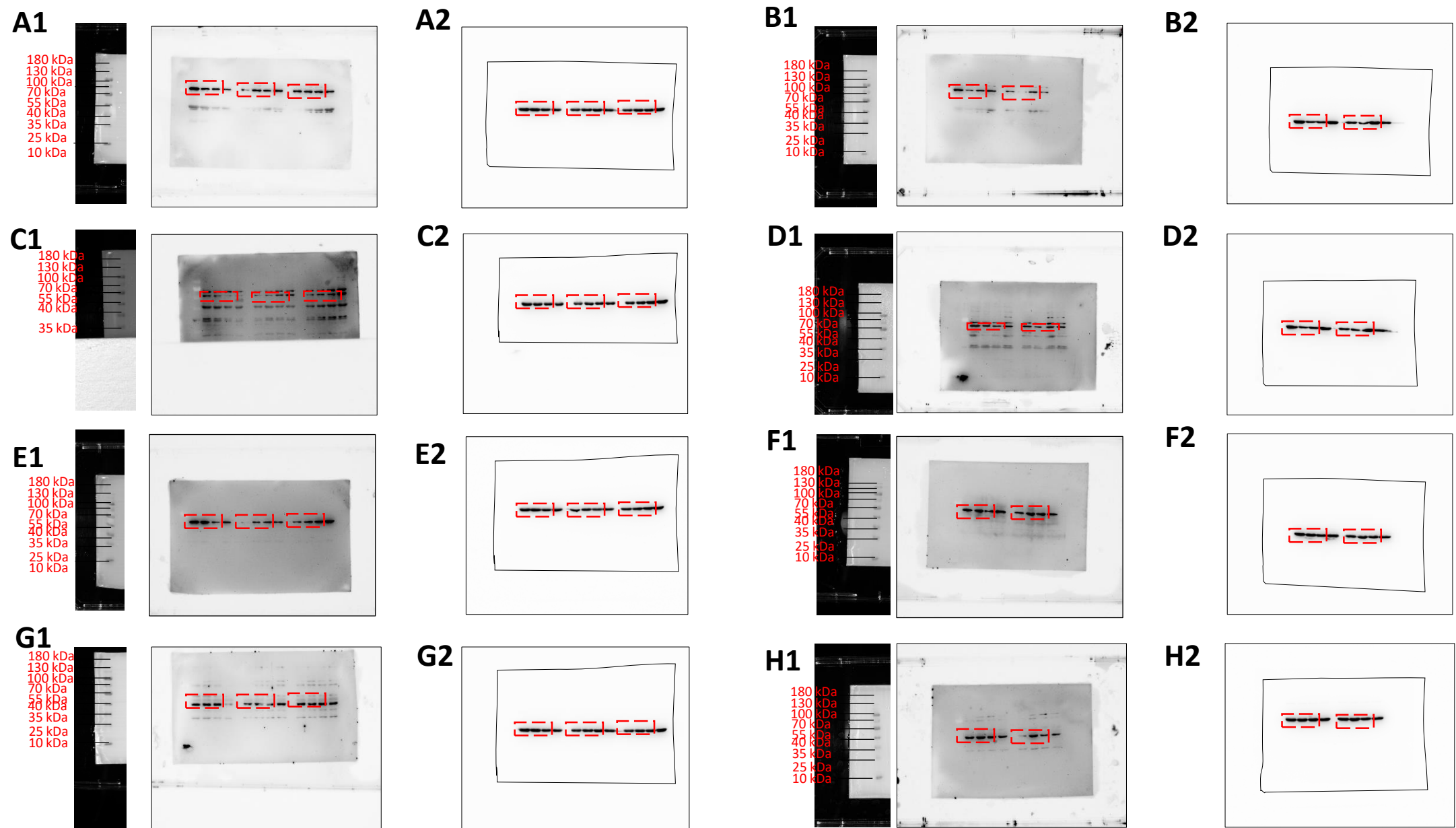


Figure S 7. **Uncropped blot images corresponding to cropped blots presented in Figure S 6 I-L in the supplementary file 1. A1, B1 OPN, C1, D1 CYP27B1, E1, F1 PDIA3, G1, H1 AP-1. A2, B2, C2, D2, E2, F2, G2, H2 - loading control – β -actin.** Chemiluminescence photographs with fragment of colorimetric picture showing protein ladder and the red box showing the cropping line. Visualization was performed on ChemiDoc Imaging System (Bio-Rad, Hercules, CA, USA) with exposure time estimated automatically.

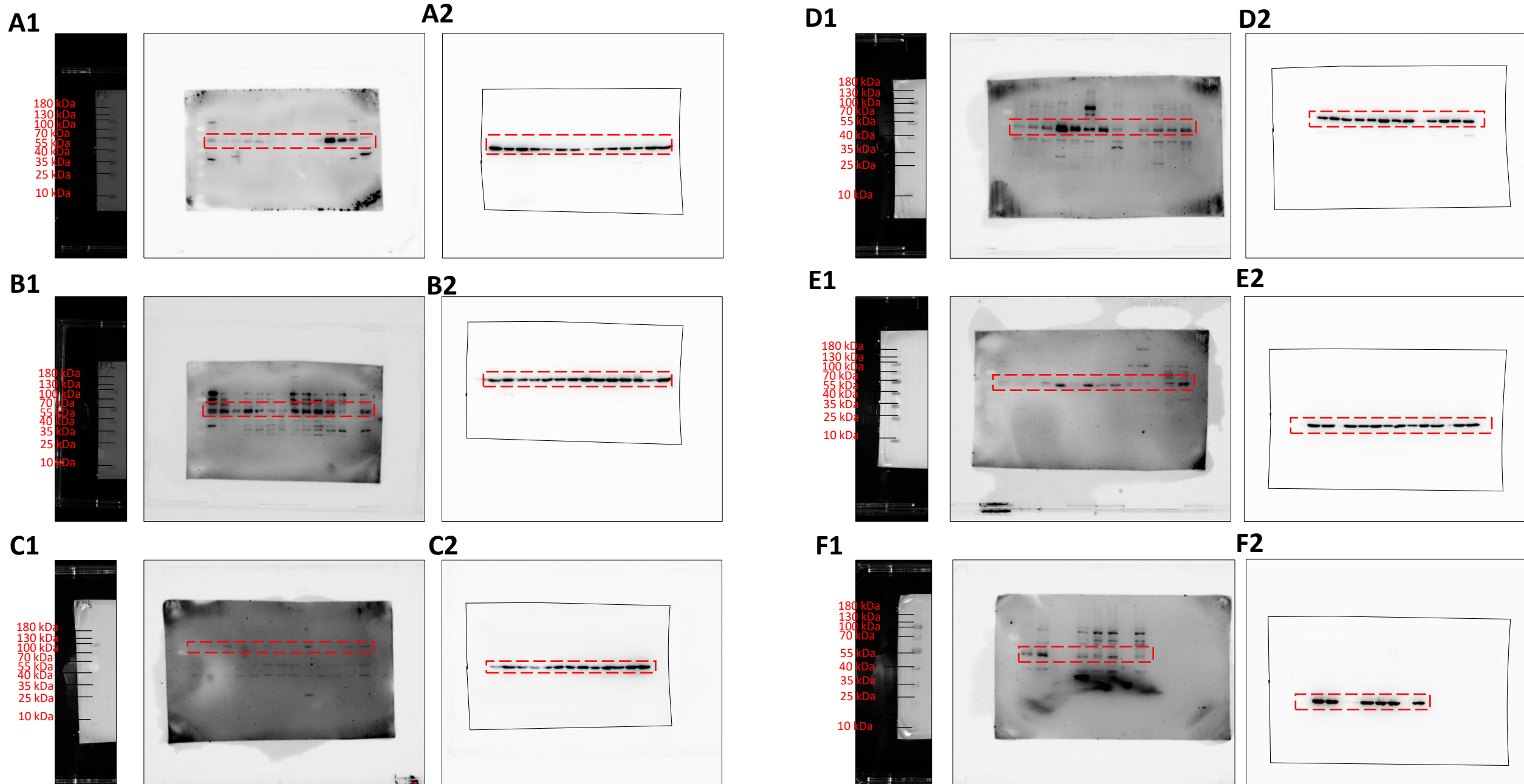


Figure S 8. **Uncropped blot images corresponding to cropped blots presented in the order shown in Figure S 9C in the supplementary file 1. A1, B1, C1, D1, E1, F1 – COX-2. A2, B2, C2, D2, E2, F2- loading control – β -actin.** Chemiluminescence photographs with fragment of colorimetric picture showing protein ladder and the red box showing the cropping line. Visualization was performed on ChemiDoc Imaging System (Bio-Rad, Hercules, CA, USA) with exposure time estimated automatically.

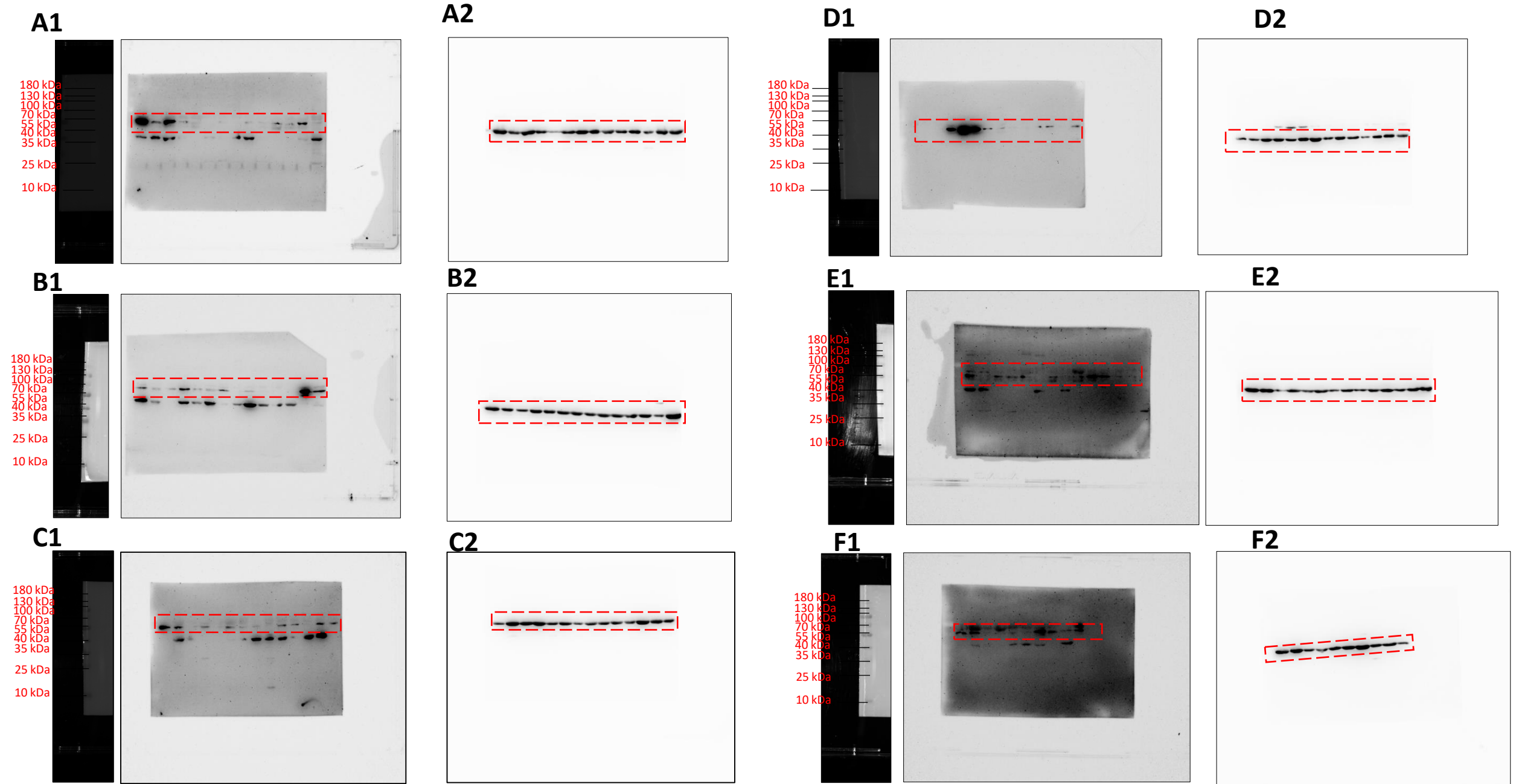


Figure S 9. **Uncropped blot images corresponding to cropped blots presented in the order shown in Figure S 9C in the supplementary file 1. A1, B1, C1, D1, E1, F1 – VDR. A2, B2, C2, D2, E2, F2- loading control – β -actin.** Chemiluminescence photographs with fragment of colorimetric picture showing protein ladder and the red box showing the cropping line. Visualization was performed on ChemiDoc Imaging System (Bio-Rad, Hercules, CA, USA) with exposure time estimated automatically.

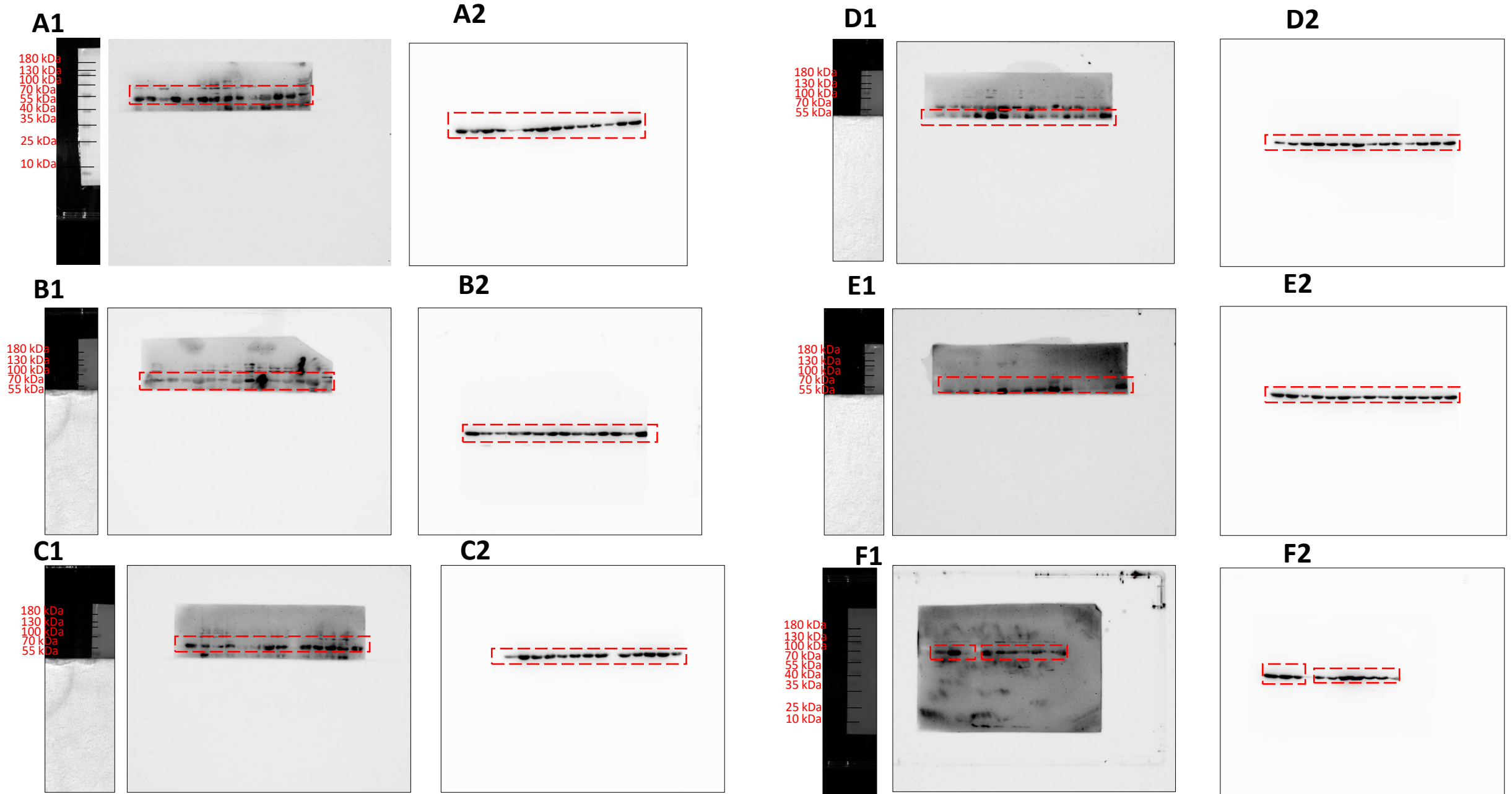


Figure S 10. **Uncropped blot images corresponding to cropped blots presented in the order shown in Figure S 9C in the supplementary file 1. A1, B1, C1, D1, E1, F1 – CYP24A1. A2, B2, C2, D2, E2, F2- loading control – β -actin.** Chemiluminescence photographs with fragment of colorimetric picture showing protein ladder and the red box showing the cropping line. Visualization was performed on ChemiDoc Imaging System (Bio-Rad, Hercules, CA, USA) with exposure time estimated automatically.

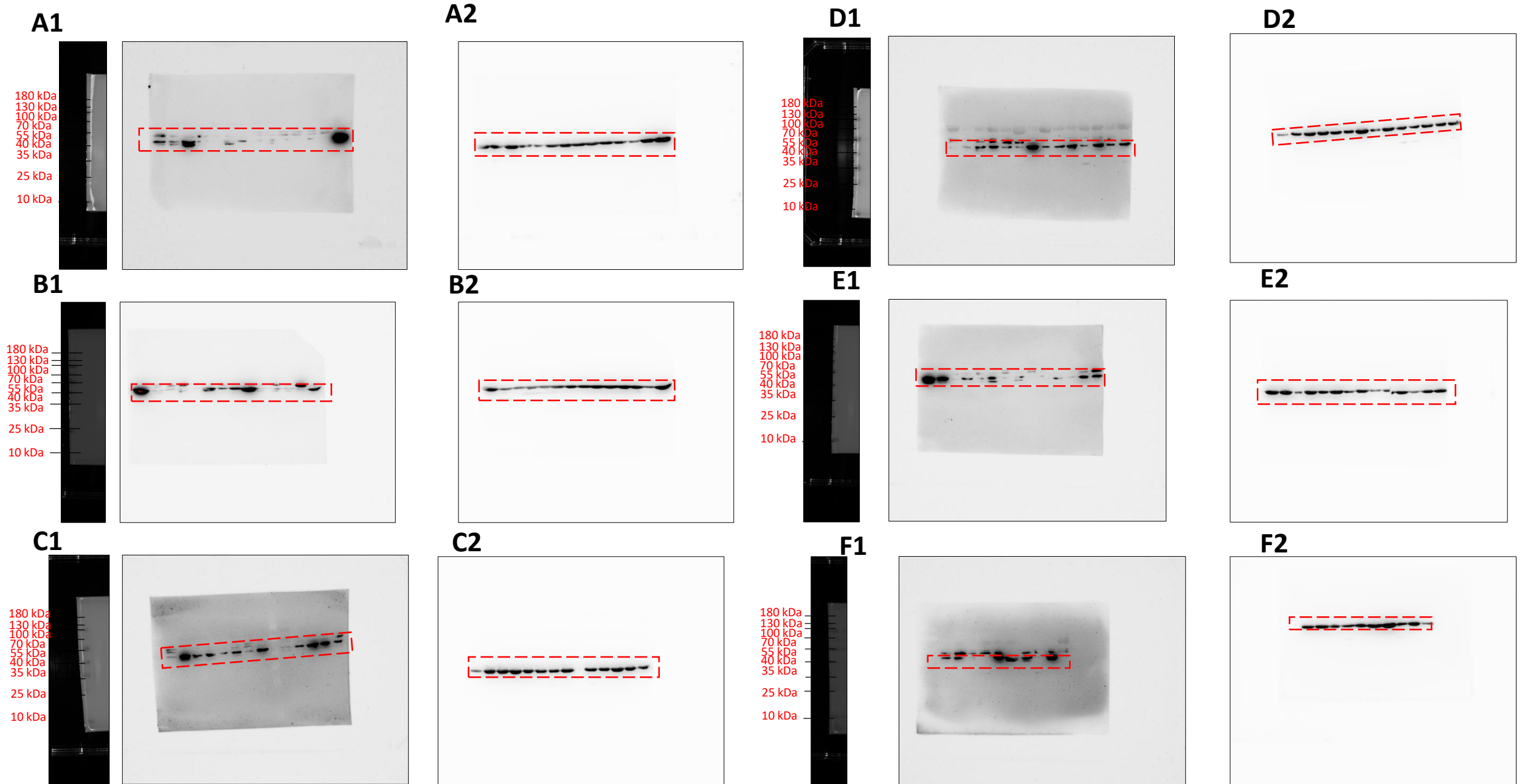


Figure S 11. **Uncropped blot images corresponding to cropped blots presented in the order shown in Figure S 9C in the supplementary file 1. A1, B1, C1, D1, E1, F1 – CYP27B1. A2, B2, C2, D2, E2, F2– loading control – β -actin.** Chemiluminescence photographs with fragment of colorimetric picture showing protein ladder and the red box showing the cropping line. Visualization was performed on ChemiDoc Imaging System (Bio-Rad, Hercules, CA, USA) with exposure time estimated automatically.