

Nintedanib inhibits growth of human prostate carcinoma cells by modulating both cell cycle and angiogenesis regulators

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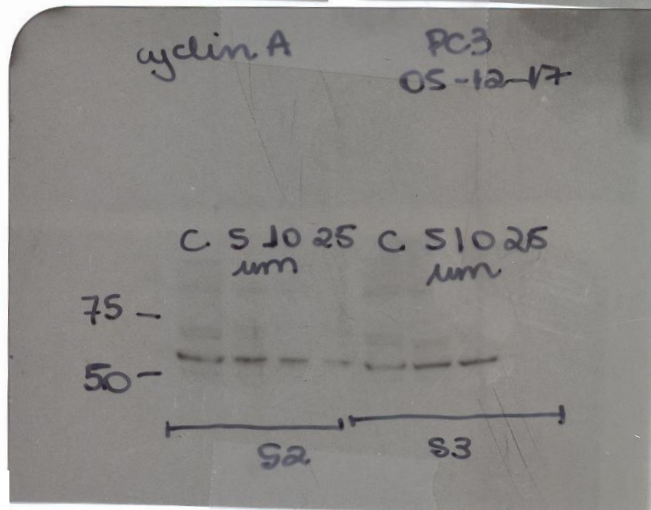
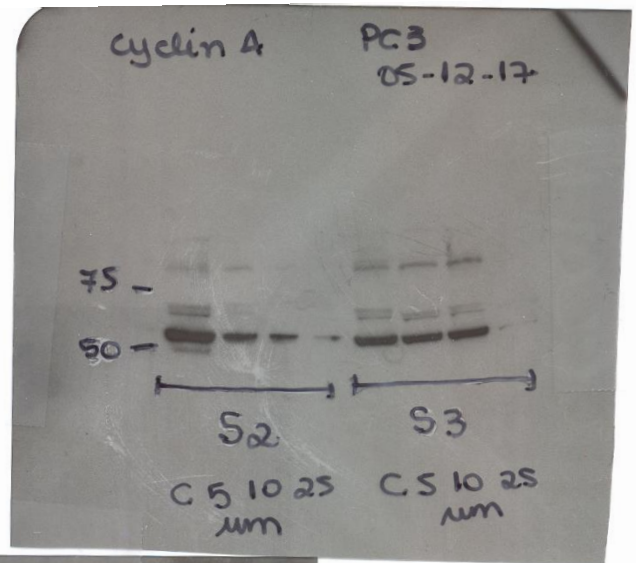
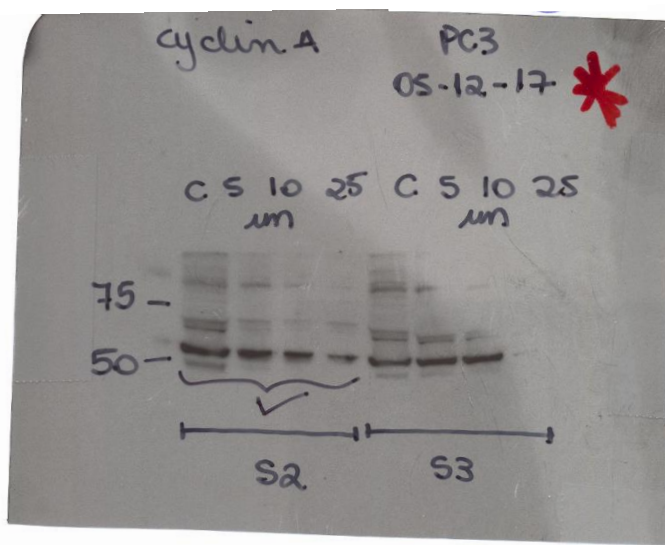
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Running title: Nintedanib is a novel anti-prostate cancer agent.

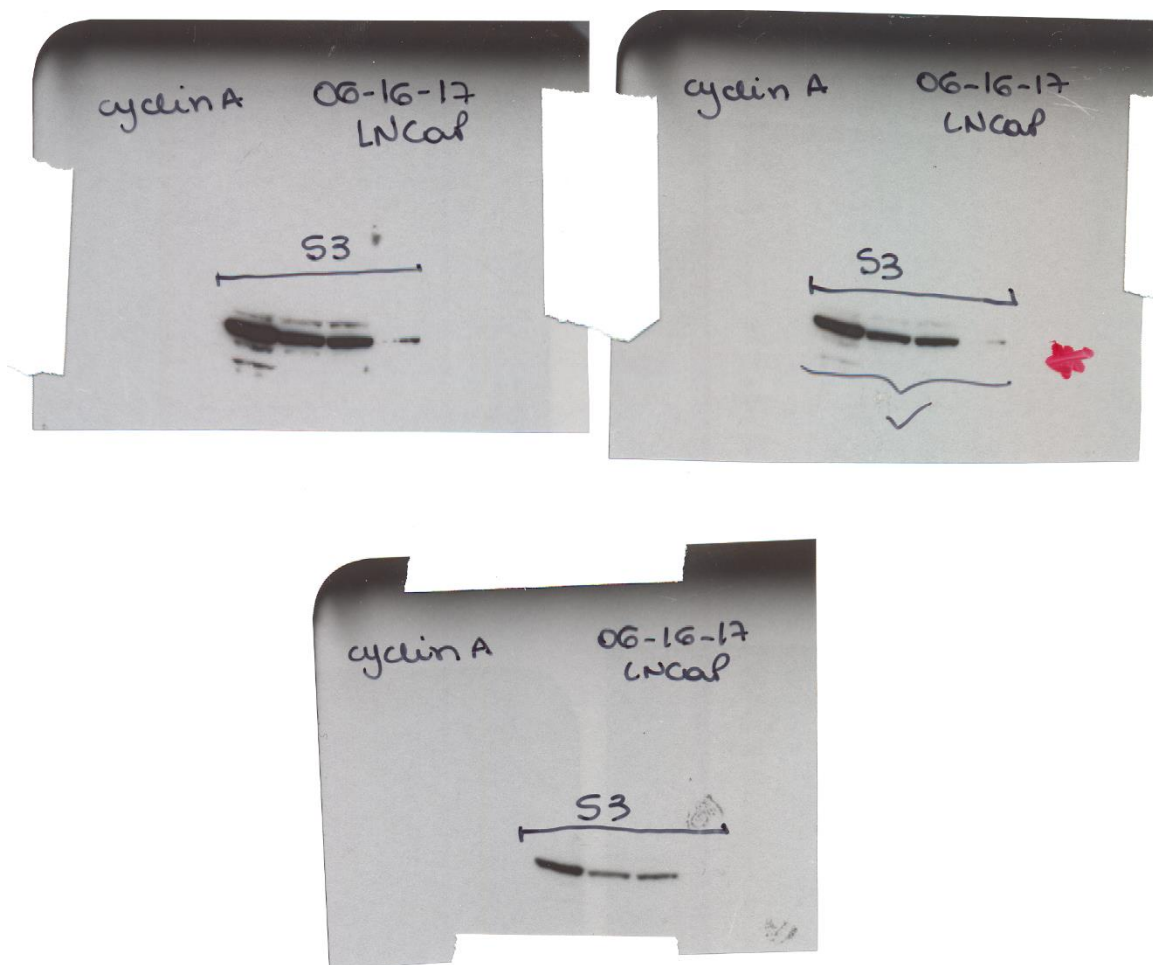
Keywords: Prostate cancer; Nintedanib; Angiogenesis, Cell Cycle; EMT

The sequence of samples in a lane is as- C, 5, 10, 25 μ M NT
 'S' represents different sample sets



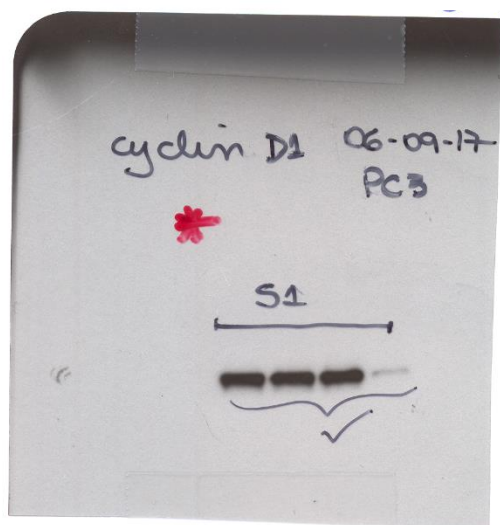
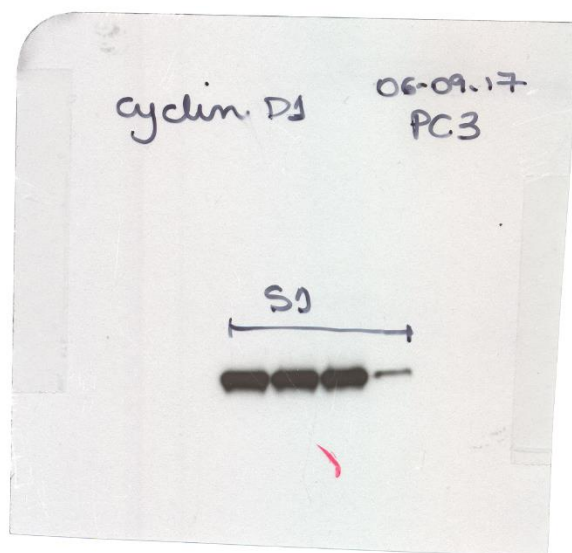
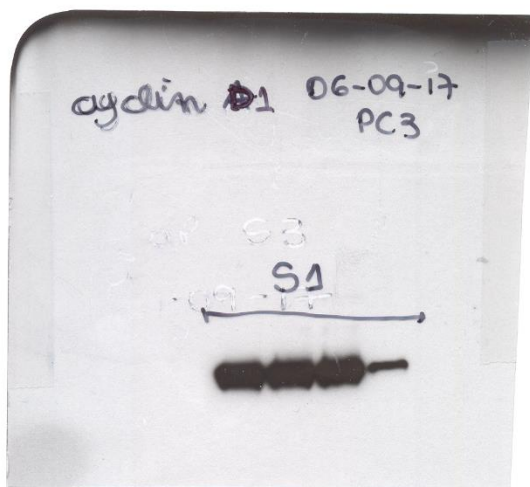
Supplementary information for Figure 2e **PC3 cells- Cyclin A (54 KDa)**
 Blots marked with asterisk (red) were used in main manuscript file

The sequence of samples in a lane is as- C, 5, 10, 25 μ M NT
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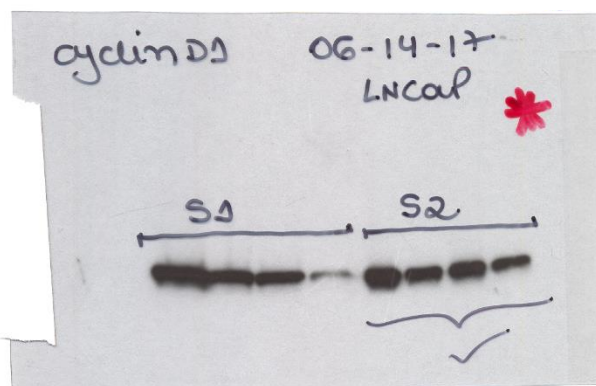
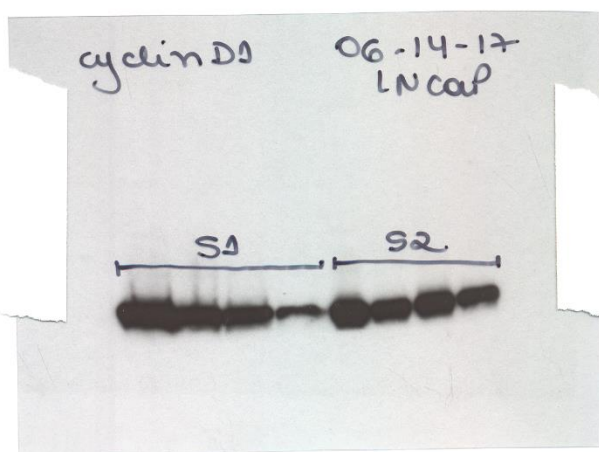
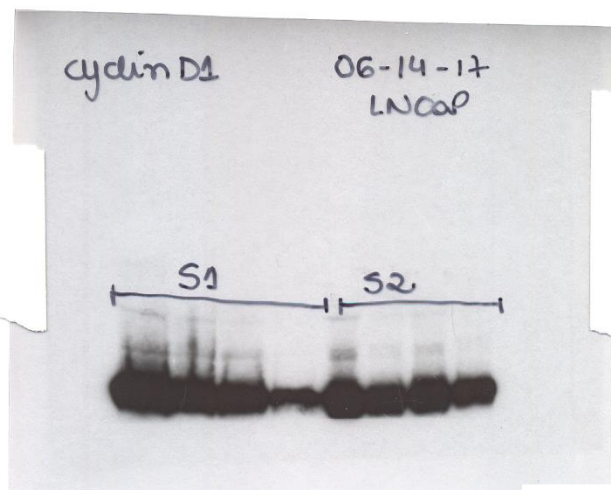
Supplementary information for Figure 2f **LNCaP cells- Cyclin A (54 KDa)**
Blots marked with asterisk (red) and checked were used in main manuscript file

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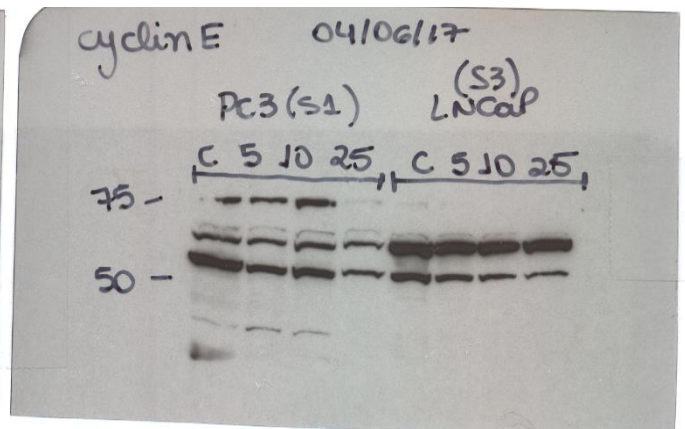
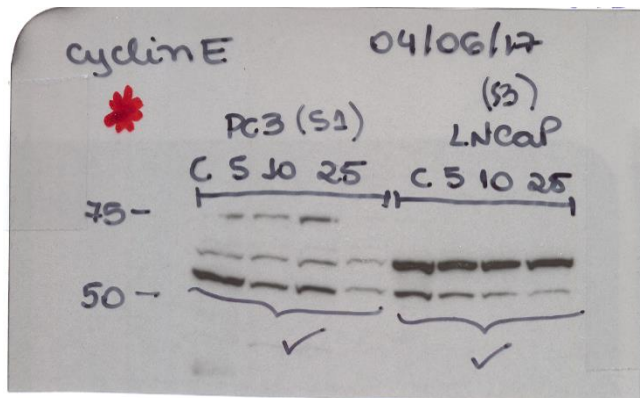
Supplementary information for Figure 2e **PC3** cells- **Cyclin D1** (31 KDa)
Blots marked with asterisk (red) and checked were used in main manuscript file

The sequence of samples in a lane is as- C, 5, 10, 25 μ M NT
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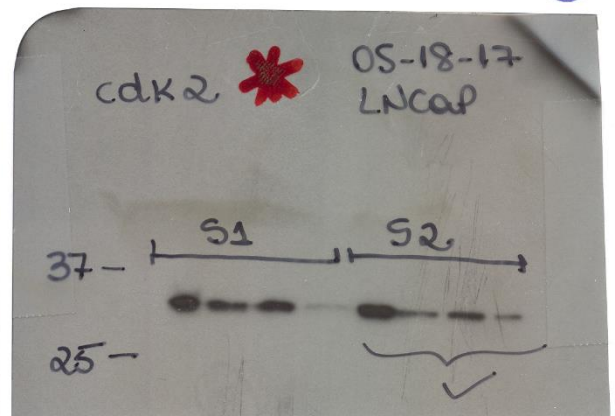
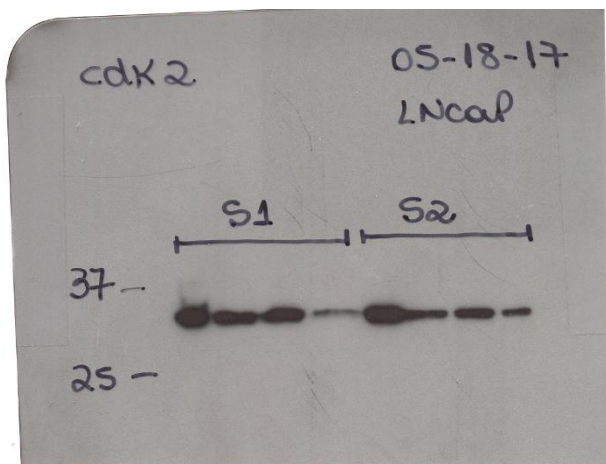
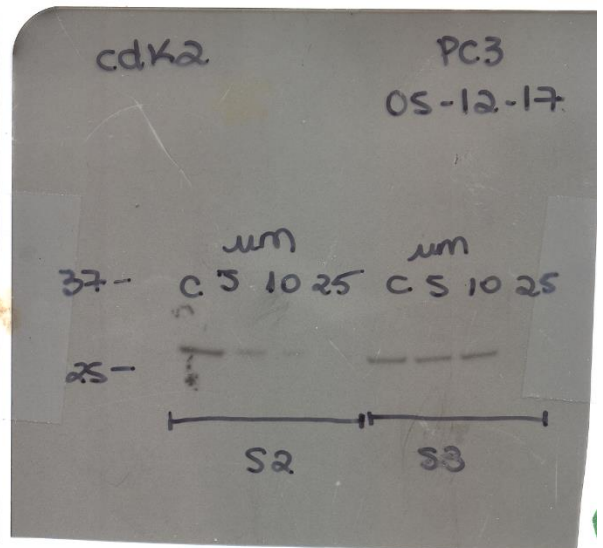
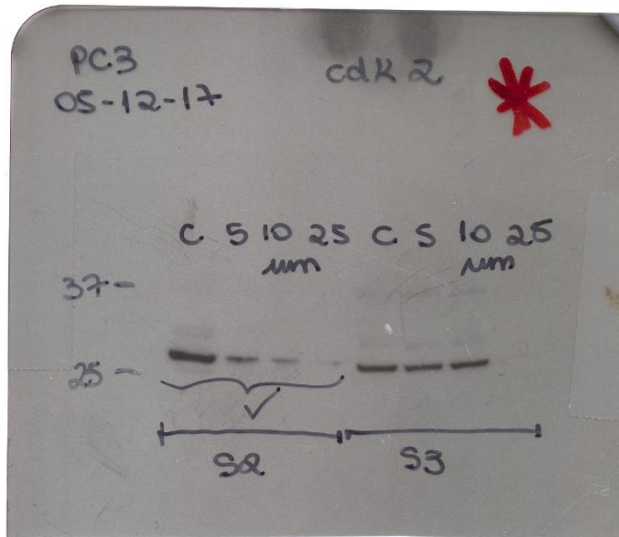
Supplementary information for Figure 2f LNCaP cells- **Cyclin D1** (31 KDa)
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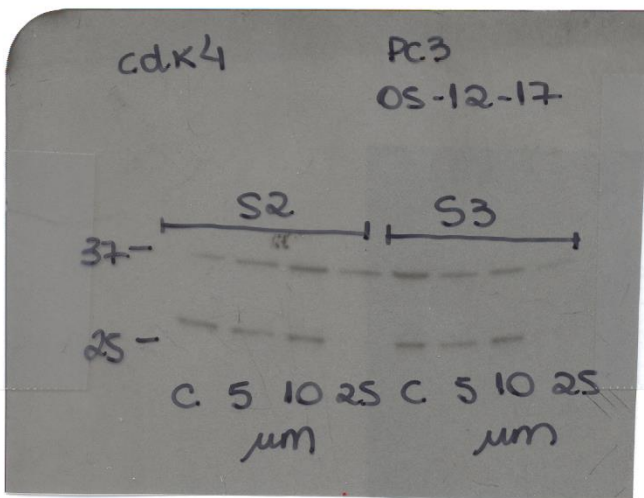
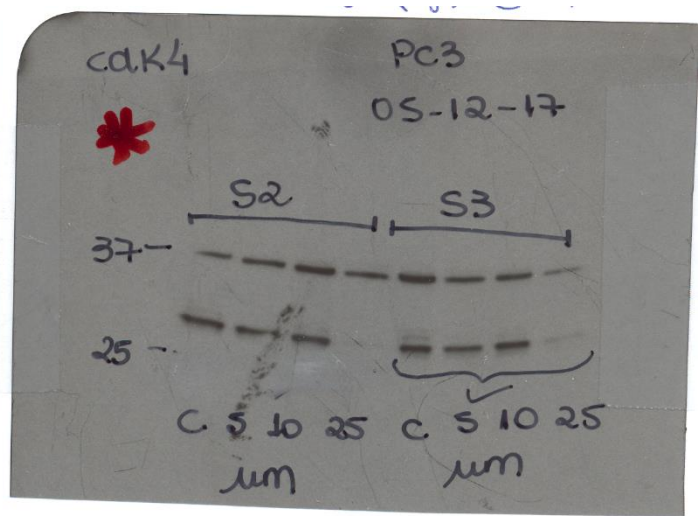
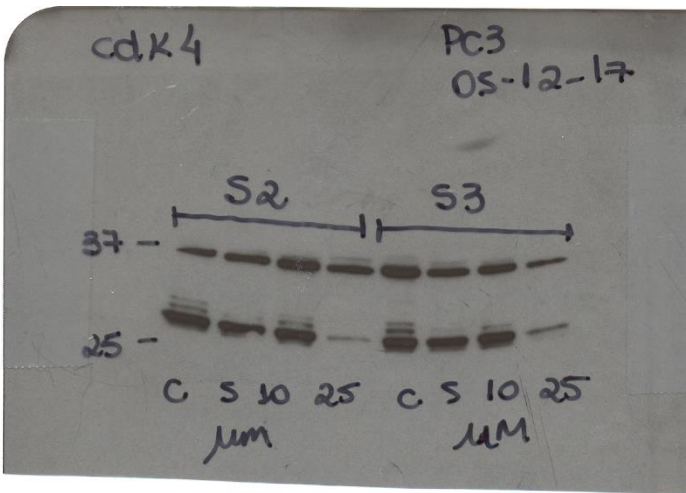
Supplementary information for Figure 2e **PC3** and Figure 2f **LNCaP** cells- **Cyclin E** (53 KDa). Blots marked with asterisk (red) and checked were used in main manuscript file

The sequence of samples in a lane is as- C, 5, 10, 25 μ M NT
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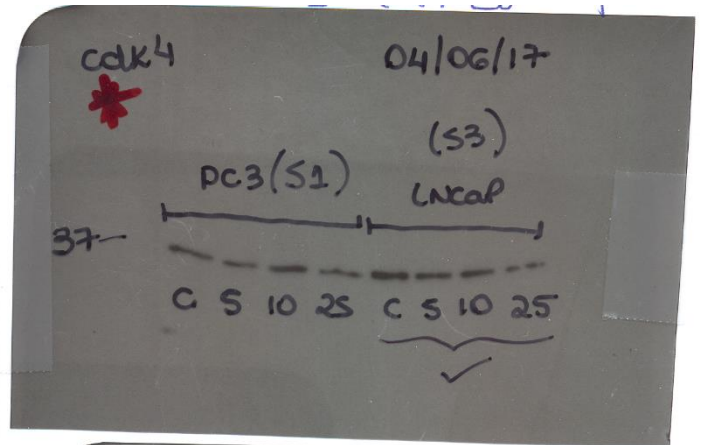
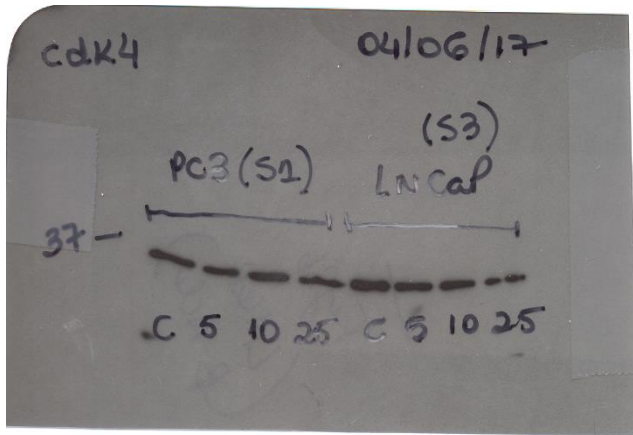
Supplementary information for Figure 2e **PC3** and Figure 2f **LNCaP** cells- **CDK2** (34 KDa). Blots marked with asterisk (red) and checked were used in main manuscript file

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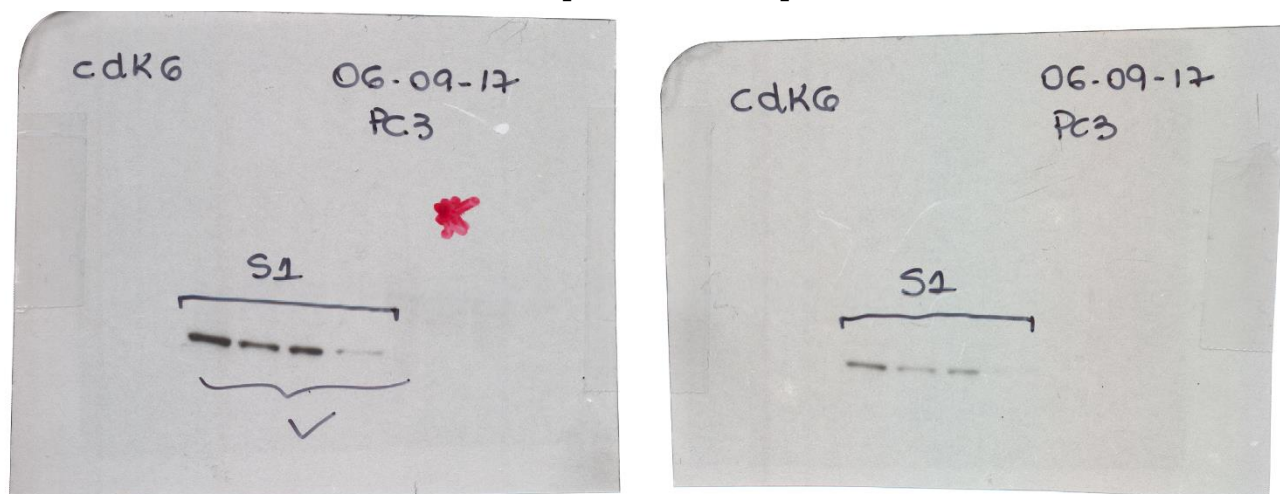
Supplementary information for Figure 2e **PC3 cells- CDK4 (34 KDa)**
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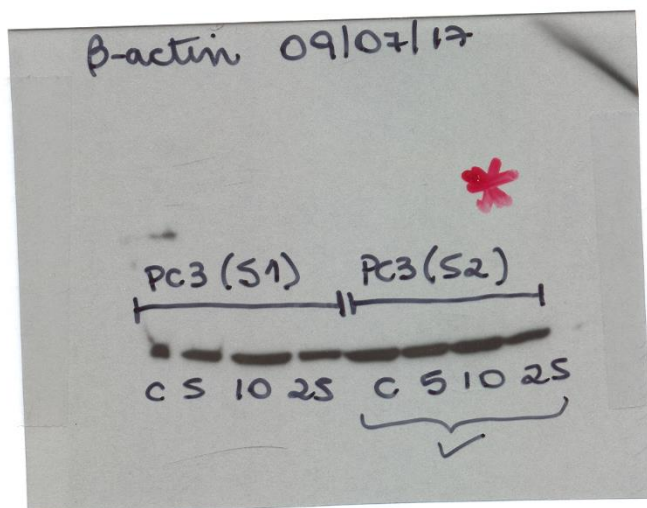
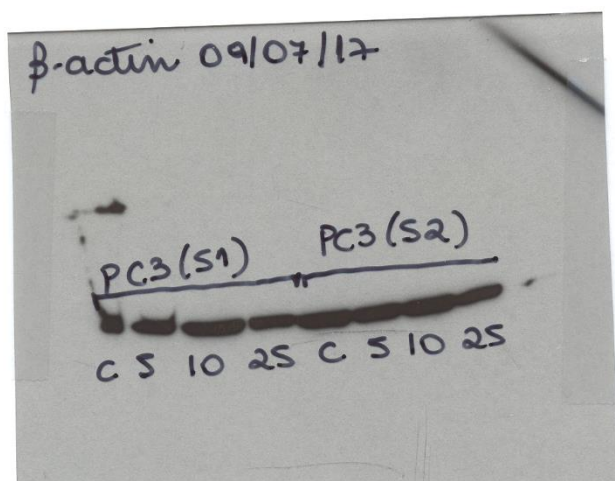
Supplementary information for Figure 2f **LNCaP** cells- **CDK4** (34 KDa)
 Blots marked with asterisk (red) and checked were used in main manuscript file

The sequence of samples in a lane is as- C, 5, 10, 25 μ M NT
'S' represents a sample set



Supplementary information for Figure 2e **PC3** cells- **CDK6** (40 KDa)
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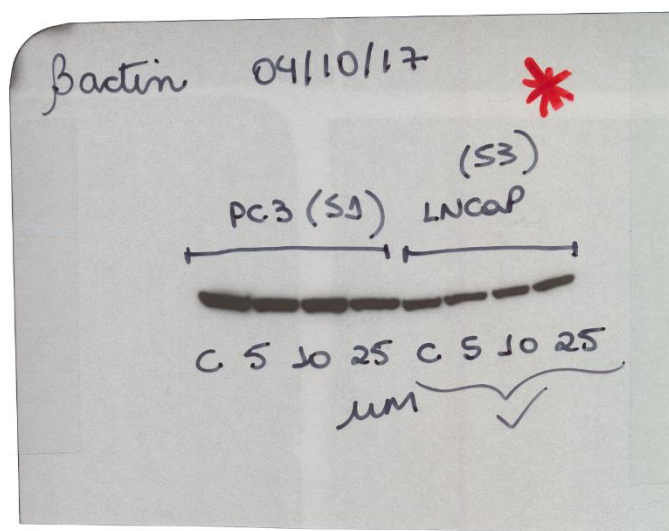
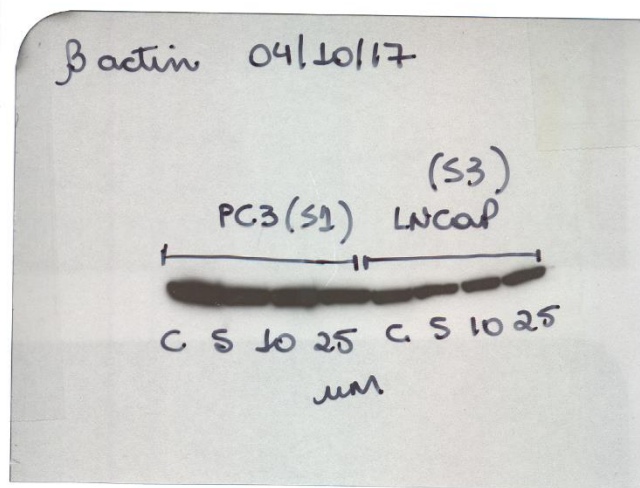
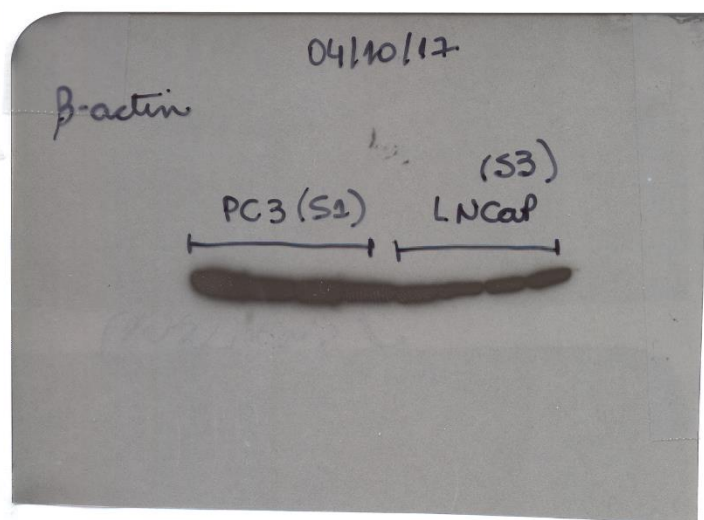
The sequence of samples in a lane is as- C, 5, 10, 25 μ M NT
'S' represents different sample sets



Supplementary information for Figure 2e **PC3** cells- β Actin (42 KDa)
Blots marked with asterisk (red) and checked were used in main manuscript file

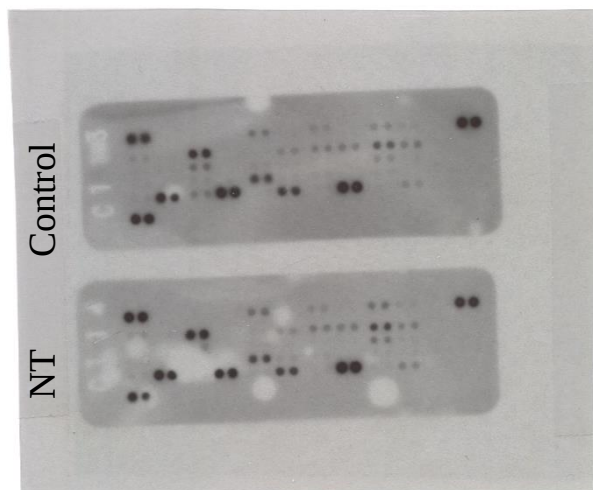
The sequence of samples in a lane is as- C, 5, 10, 25 μ M NT

'S' represents different sample sets

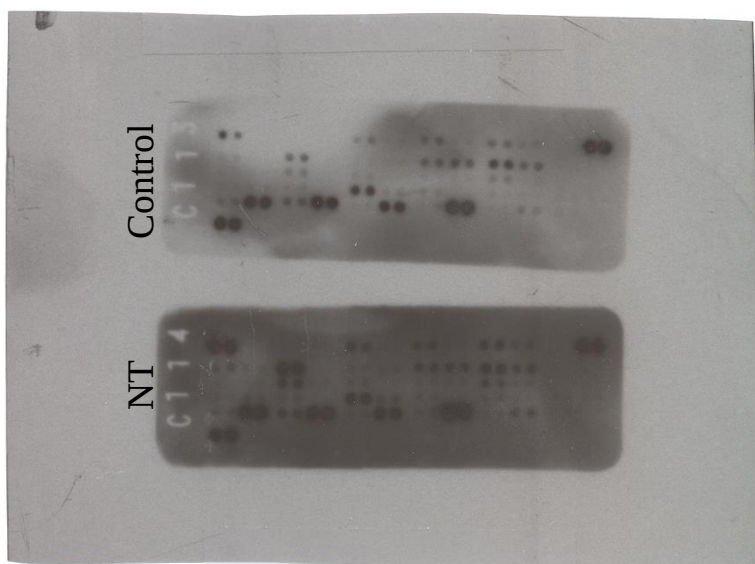


Supplementary information for Figure 2f LNCaP cells- β Actin (42 KDa)
Blots marked with asterisk (red) and checked were used in main manuscript file

Experiment 1- Top panel (7 min) exposure

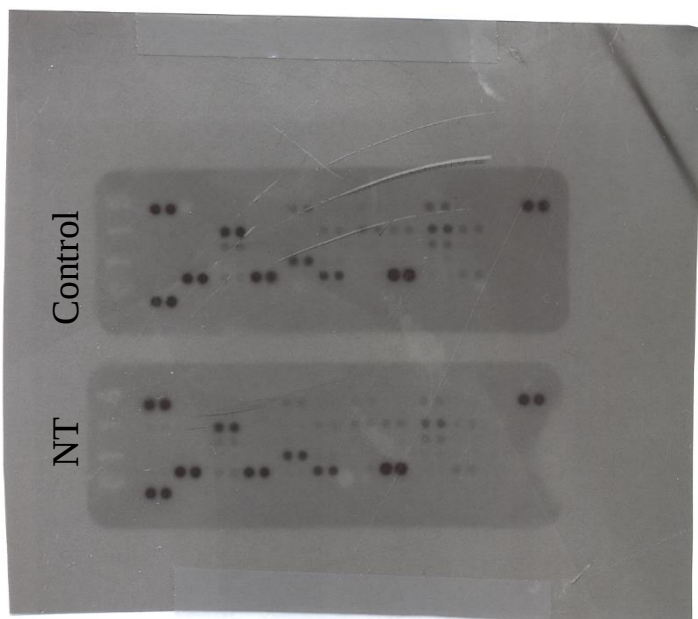


Experiment 1- Bottom panel (10 min) exposure

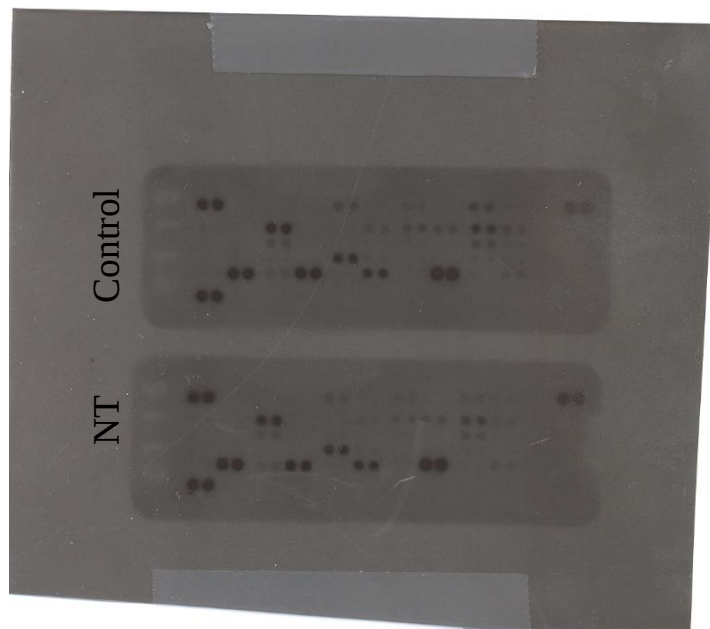


Supplementary information for **Figure 7a**

Experiment 2- Top panel (5 min) exposure



Experiment 2- Bottom panel (7 min) exposure



Supplementary information for **Figure 7b**

Antibodies used in the manuscript have been previously used by different research groups (Citation/publication details provided below)

Cyclin A (Santa Cruz - sc751):

RNF4 regulates DNA double-strand break repair in a cell cycle-dependent manner. Kuo, C-Y. *et al. Cell Cycle*. 2016;15(6):787-798. doi:10.1080/15384101.2016.1138184.

CDK2 (Santa Cruz - sc6248):

Impairment of primary cilia contributes to visceral adiposity of high fat diet-fed mice. Qiu, N. *et al. J Cell Biochem*. 2018;119: 1313–1325. <https://doi.org/10.1002/jcb.26253>.

CDK4 (Santa Cruz - sc749):

Amyloid β 1-42 (A β 1-42) Induces the CDK2-Mediated Phosphorylation of Tau through the Activation of the mTORC1 Signaling Pathway While Promoting Neuronal Cell Death. Lee, KH. *et al. Frontiers in Molecular Neuroscience*. 2017; 10:229. doi:10.3389/fnmol.2017.00229.

CDK6 (Santa Cruz - sc177):

Melatonin Suppresses the Growth of Ovarian Cancer Cell Lines (OVCAR-429 and PA-1) and Potentiates the Effect of G1 Arrest by Targeting CDKs. Srivastava SK, ed. Shen, C-J. *et al. International Journal of Molecular Sciences*. 2016;17(2):176. doi:10.3390/ijms17020176.

Cyclin E (Santa Cruz - sc198):

All-trans retinoic acid induces cell-cycle arrest in human cutaneous squamous carcinoma cells by inhibiting the mitogen-activated protein kinase-activated protein 1 pathway. Zhang, M.L. *et al*. 2014. *Clinical and experimental dermatology*. 39: 354-60; doi:10.1111/ced.12227.

Cyclin D1 (Santa Cruz (sc718) for Western Blotting and Thermo Scientific (#RM9104-S0) for IHC):

β -catenin signaling is required for RAS-driven thyroid cancer through PI3K activation. Sastre-Perona A, *et al. Oncotarget*. 2016;7 (31):49435-49449. doi:10.18632/oncotarget.10356.

Consistent Immunostaining for Cyclin D1 can be achieved on a routine basis using a newly available rabbit monoclonal antibody. Cheuk, W. *et al. Am J Surg Pathol*. 2004; 28(6): 801-807. doi:10.1097/01.pas.0000126054.95798.94

E-Cadherin (Santa Cruz - sc21791):

hTERT peptide fragment GV1001 demonstrates radioprotective and antifibrotic effects through suppression of TGF- β signaling. Chen, W. *et al. International Journal of Molecular Medicine*. 2018;41(6):3211-3220. doi:10.3892/ijmm.2018.3566.

Vimentin (Santa Cruz - sc7557):

Lentiviral vector-mediated overexpression of mutant ataxin-7 recapitulates SCA7 pathology and promotes accumulation of the FUS/TLS and MBNL1 RNA-binding proteins. Alves, S. *et al. Molecular Neurodegeneration*. 2016;11:58. doi:10.1186/s13024-016-0123-2.

Ki67 (Abcam - ab15580):

Olfactory memory is enhanced in mice exposed to extremely low-frequency electromagnetic fields via Wnt/ β -catenin dependent modulation of subventricular zone neurogenesis.

Mastrodonato, A. *et al. Scientific Reports*. 2018; 8:262.

Cleaved caspase 3 (Cell Signaling Technology #9661):

Adiponectin protects against lung ischemia-reperfusion injury in rats with type 2 diabetes mellitus. Li, D. *et al. Molecular Medicine Reports*, 2018(17), 7191-7201.

<https://doi.org/10.3892/mmr.2018.8748>.

CD31 (Abcam - ab28364):

Exosomes from human umbilical cord blood accelerate cutaneous wound healing through miR-21-3p-mediated promotion of angiogenesis and fibroblast function. Hu, Y. *et al. Theranostics*.

2018;8(1):169-184. doi:10.7150/thno.21234.

Nestin (Santa Cruz - sc33677):

Neuronal Nitric Oxide Synthase in Neural Stem Cells Induces Neuronal Fate Commitment via the Inhibition of Histone Deacetylase 2. Jin, X. *et al. Frontiers in Cellular Neuroscience*.

2017;11:66. doi:10.3389/fncel.2017.00066.

VEGF (Santa Cruz - sc152):

Interleukin-17 promotes prostate cancer via MMP7-induced epithelial-to-mesenchymal transition. Zhang, Q. *et al. Oncogene*. 2017;36(5):687-699. doi:10.1038/onc.2016.240.