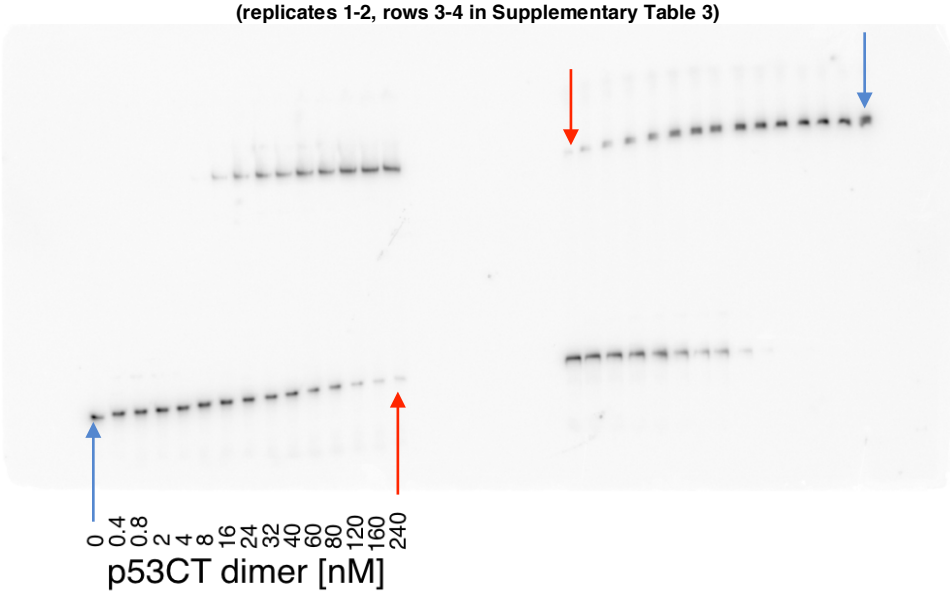
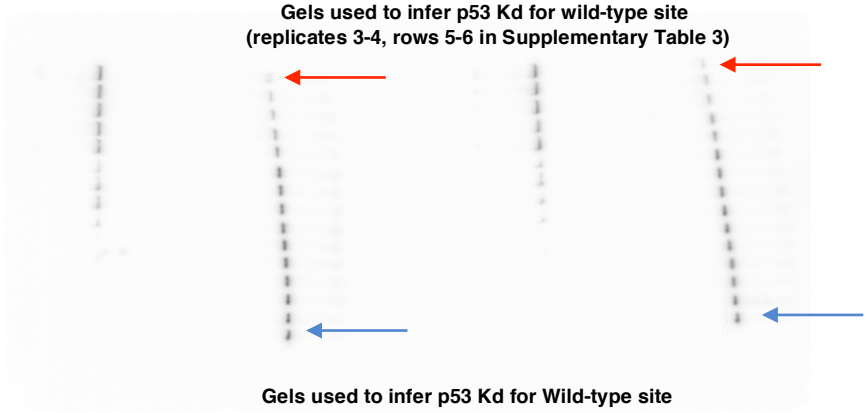
Supplementary Figure 1. Uncropped EMSA gel data

All copies of the original high-resolution image scans are available at:
https://figshare.com/projects/DNA_mismatches_reveal_conformational_penalties_in_protein-DNA_recognition/83663
Computed Kd values for all replicates are available in **Supplementary Table 3.** Average Kd values and standard deviations are available in **Supplementary Table 3** and shown in **Figure 1f** (p53 and GR EMSA) and **Extended Data Figure 3e** (p53 and GR EMSA).

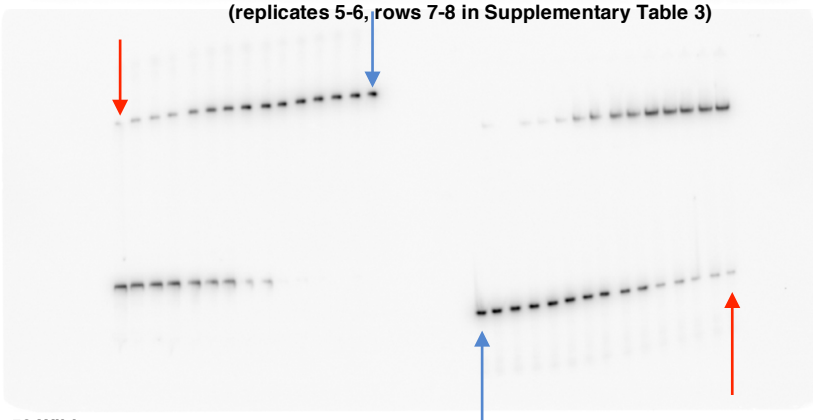
Gels used to infer p53 Kd for wild-type site
(replicates 1-2, rows 3-4 in Supplementary Table 3)



Gels used to infer p53 Kd for wild-type site
(replicates 3-4, rows 5-6 in Supplementary Table 3)

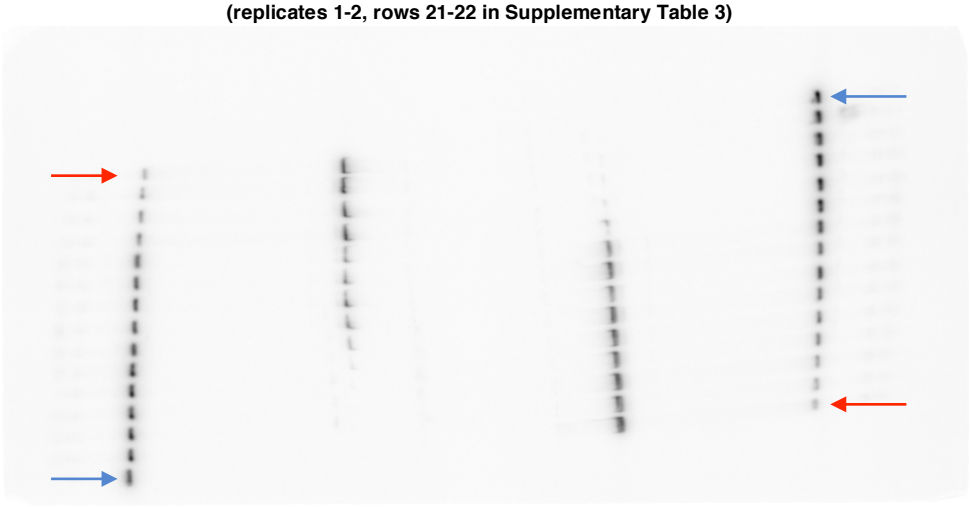


Gels used to infer p53 Kd for Wild-type site
(replicates 5-6, rows 7-8 in Supplementary Table 3)

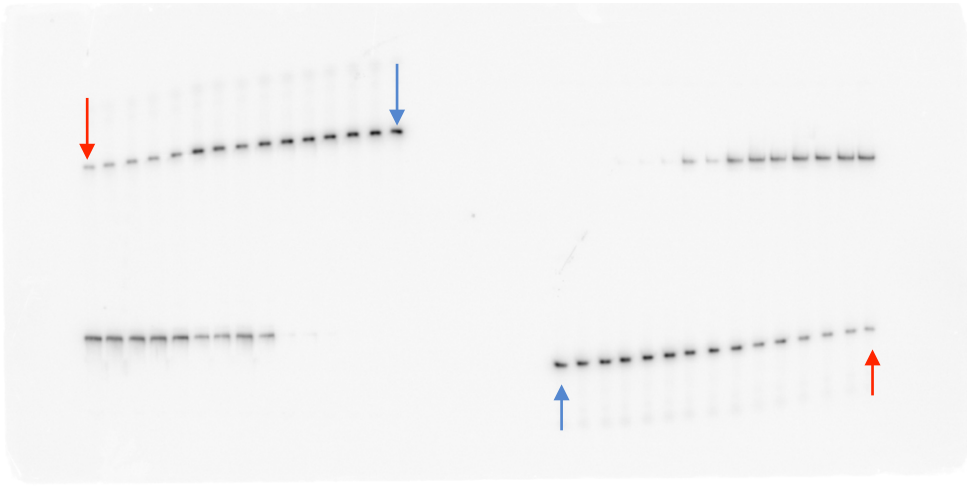


p53 Wild type sequence.
Protein concentration: 0,0.4,0.8,2,4,8,16,24,32,40,60,80,120,160,240 [nM] (Blue arrow 0nM, and Red arrow 240nM)
Original high quality image scans:
https://figshare.com/articles/figure/p53_EMSA_gels/12568691
File names: WT_CT20-1_b1e1_8.4.19.tif , WT_CT20-2_b1e2_11.4.19.tif , WT_CT20-2_b1e3_14.4.19.tif

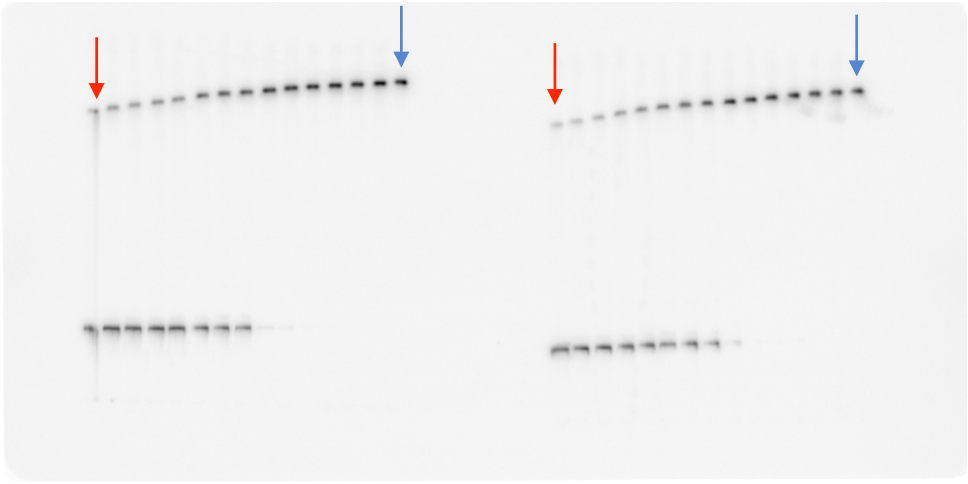
Gels used to infer p53 Kd for the T-A variant site
(replicates 1-2, rows 21-22 in Supplementary Table 3)



Gels used to infer p53 Kd for the T-A variant site
(replicates 3-4, rows 23-24 in Supplementary Table 3)

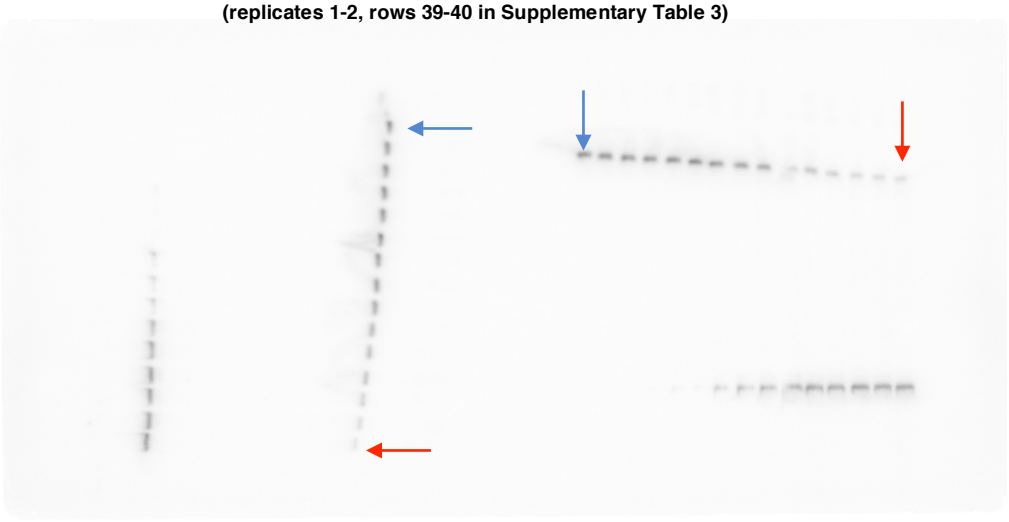


Gels used to infer p53 Kd for the T-A variant site
(replicates 5-6, rows 25-26 in Supplementary Table 3)

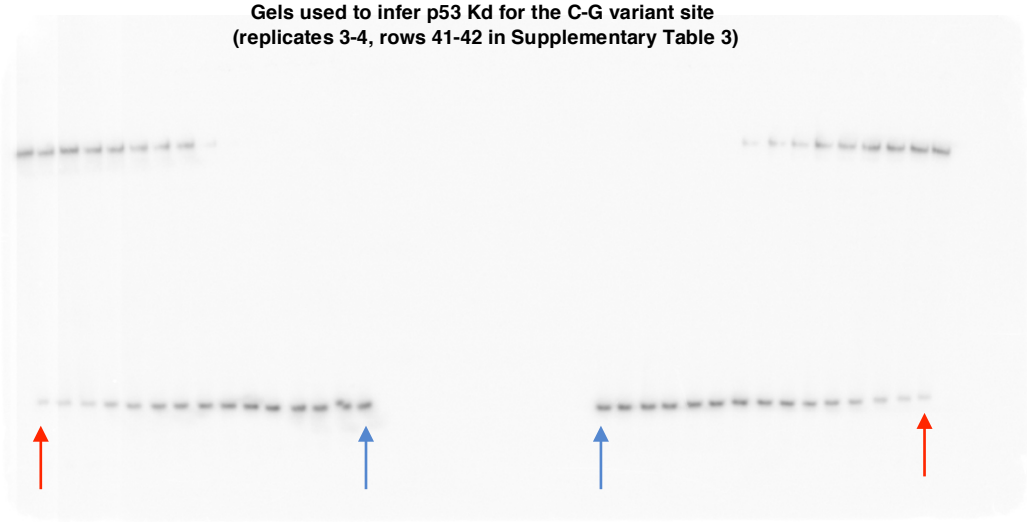


p53 T-A variation.
Protein concentration: 0,0.4,0.8,2,4,8,16,24,32,40,60,80,120,160,240 [nM] (Blue arrow 0nM, and Red arrow 240nM)
Original high quality image scans:
https://figshare.com/articles/figure/p53_EMSA_gels/12568691
File names: Var_T-A_CT20-2_b1e1_17.4.19.tif , Var_T-A_CT20-2_b1e2_21.4.19.tif , Var_T-A_CT20-2_b1e3_23.4.19.tif

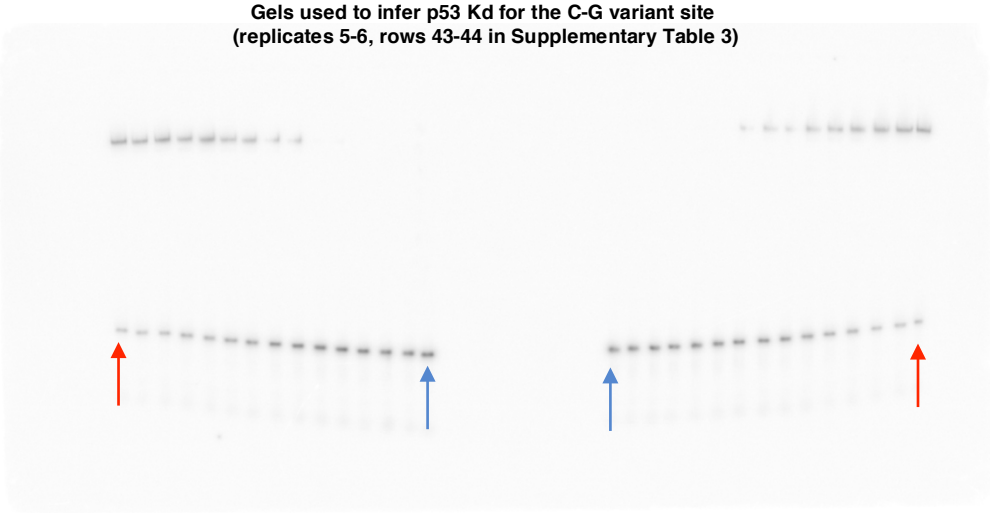
Gels used to infer p53 Kd for the C-G variant site
(replicates 1-2, rows 39-40 in Supplementary Table 3)



Gels used to infer p53 Kd for the C-G variant site
(replicates 3-4, rows 41-42 in Supplementary Table 3)

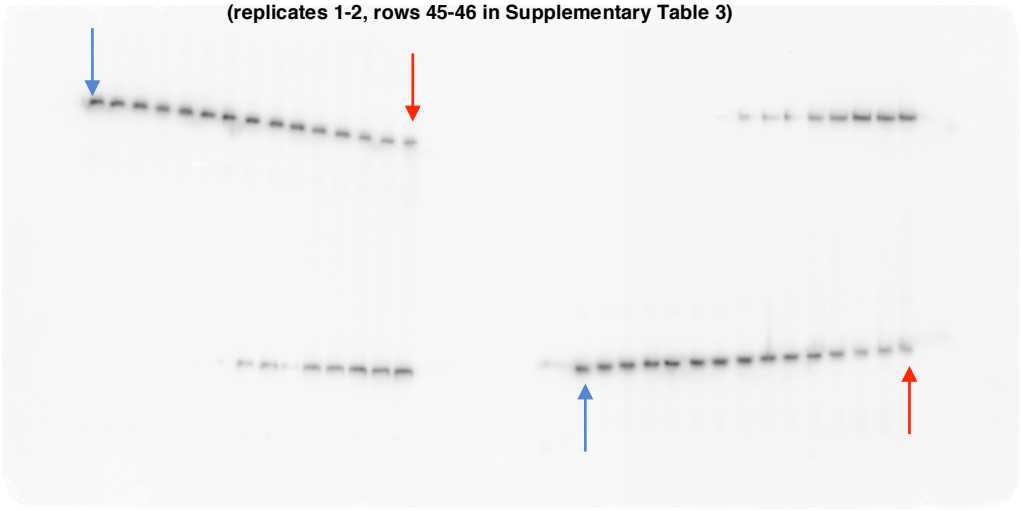


Gels used to infer p53 Kd for the C-G variant site
(replicates 5-6, rows 43-44 in Supplementary Table 3)

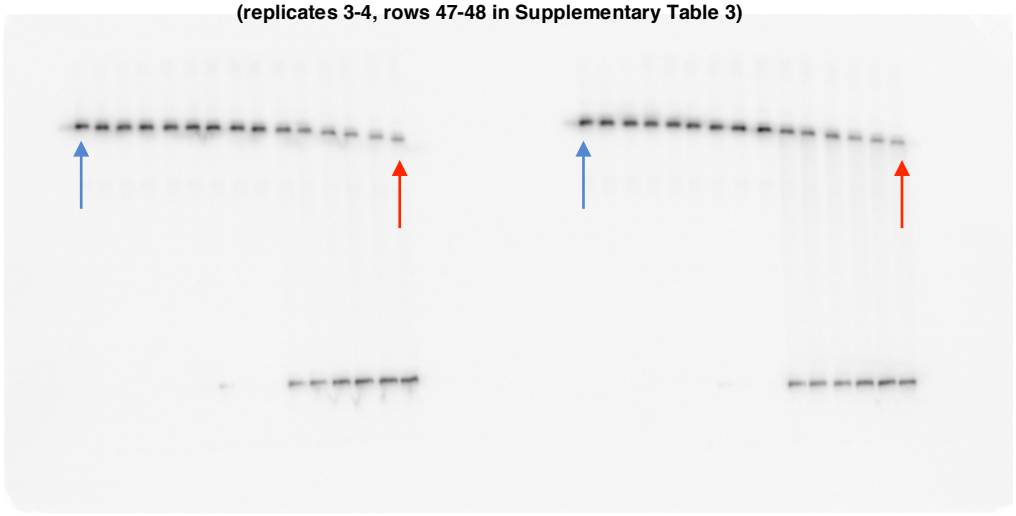


p53 C-G variation.
Protein concentration: 0,0.4,0.8,2,4,8,16,24,32,40,60,80,120,160,240 [nM] (Blue arrow 0nM, and Red arrow 240nM)
Original high quality image scans:
https://figshare.com/articles/figure/p53_EMSA_gels/12568691
File names: Var_C-G_CT20-2_b1e1_18.4.19.tif , Var_C-G_CT20-2_b1e2_22.4.19.tif , Var_C-G_CT20-2_b1e3_28.4.19 .tif

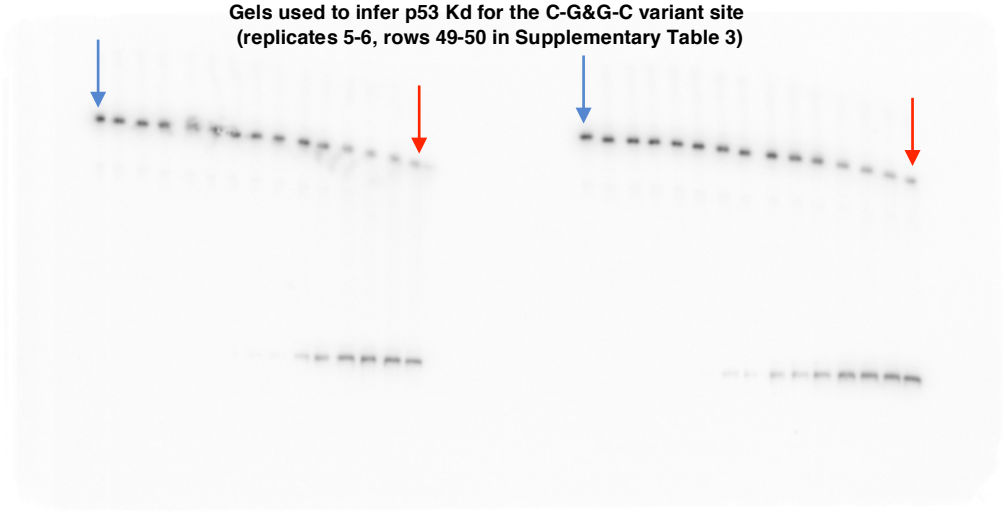
Gels used to infer p53 Kd for the C-G&G-C variant site (replicates 1-2, rows 45-46 in Supplementary Table 3)



Gels used to infer p53 Kd for the C-G&G-C variant site (replicates 3-4, rows 47-48 in Supplementary Table 3)

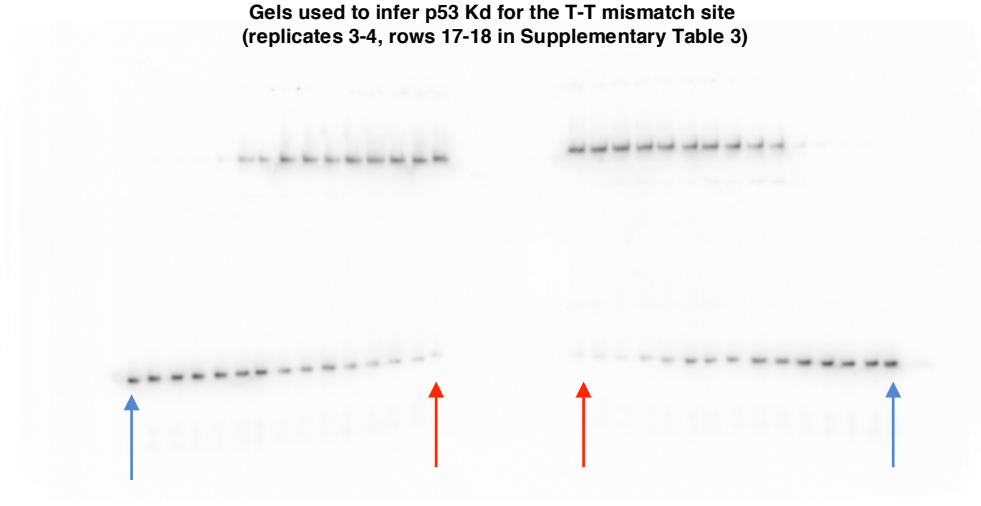


Gels used to infer p53 Kd for the C-G&G-C variant site (replicates 5-6, rows 49-50 in Supplementary Table 3)

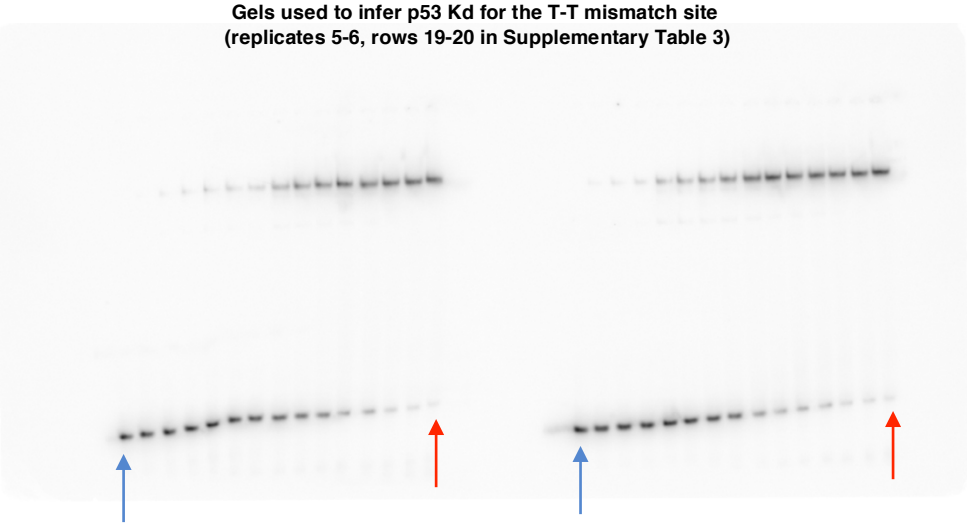


p53 C-G + G-C variation.
Protein concentration: 0,0.4,0.8,2,4,8,16,24,32,40,60,80,120,160,240 [nM] (Blue arrow 0nM, and Red arrow 240nM)
Original high quality image scans:
https://figshare.com/articles/figure/p53_EMSA_gels/12568691
File names: Var_C-G G-C_CT20-4_b2e1_15.7.19.tif , Var_C-G G-C_CT20-4_b2e2_16.7.19.tif , Var_C-G G-C_CT20-4_b2e3_17.719.tif

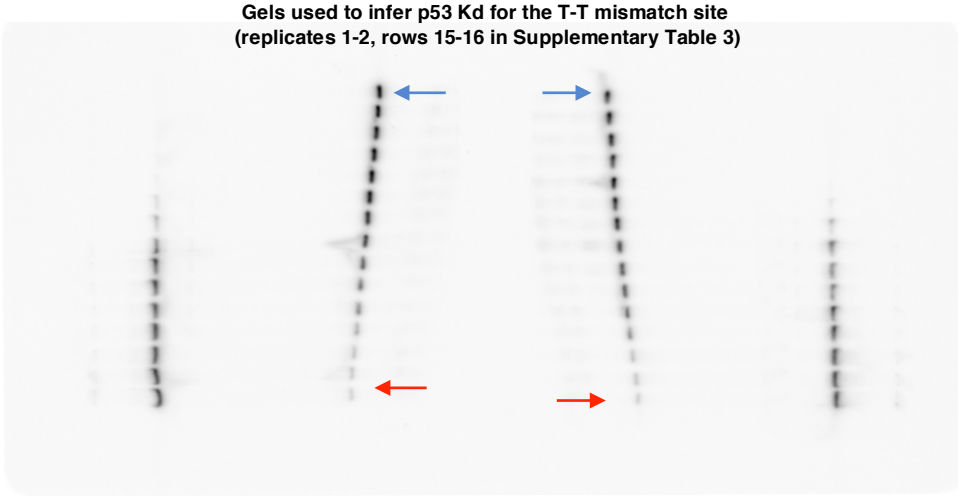
Gels used to infer p53 Kd for the T-T mismatch site
(replicates 3-4, rows 17-18 in Supplementary Table 3)



Gels used to infer p53 Kd for the T-T mismatch site
(replicates 5-6, rows 19-20 in Supplementary Table 3)

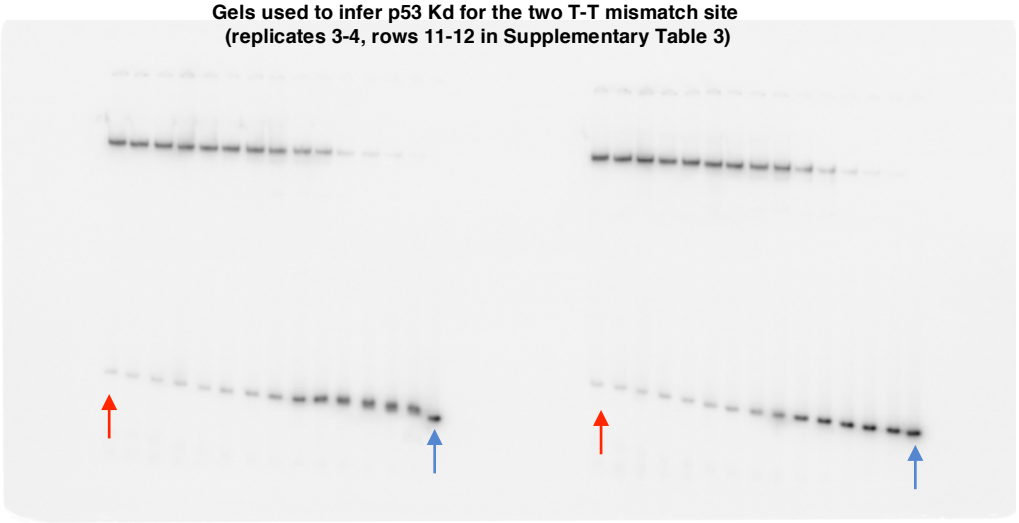


Gels used to infer p53 Kd for the T-T mismatch site
(replicates 1-2, rows 15-16 in Supplementary Table 3)

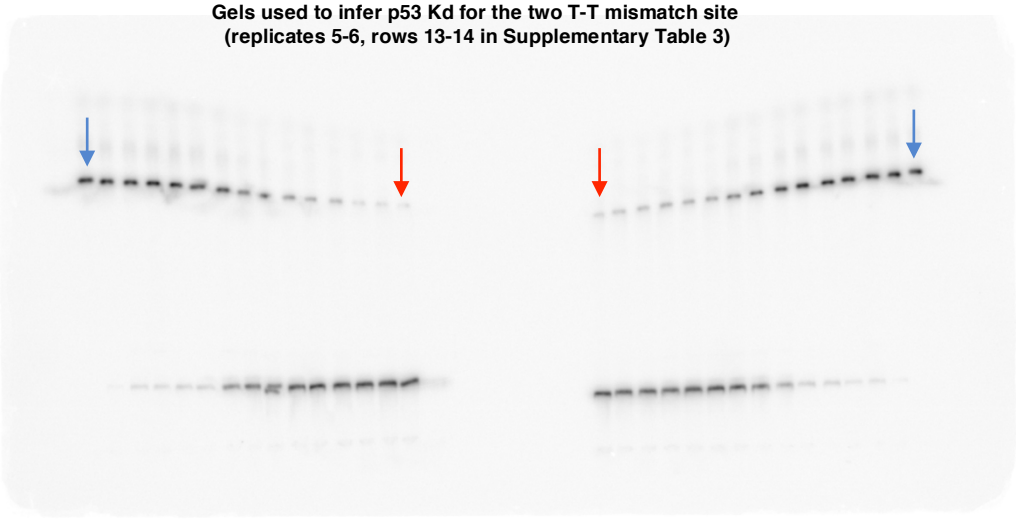


p53 T-T mismatch
Protein concentration: 0,0.4,0.8,2,4,8,16,24,32,40,60,80,120,160,240 [nM] (Blue arrow 0nM, and Red arrow 240nM)
Original high quality image scans:
https://figshare.com/articles/figure/p53_EMSA_gels/12568691
File names: MM_T.T_CT20-2_b1e3_14.4.19.tif , MM_T.T_CT20-2_b1e4_16.4.19.tif , MM-T.T_CT20-1_b1e2_9.4.19.tif

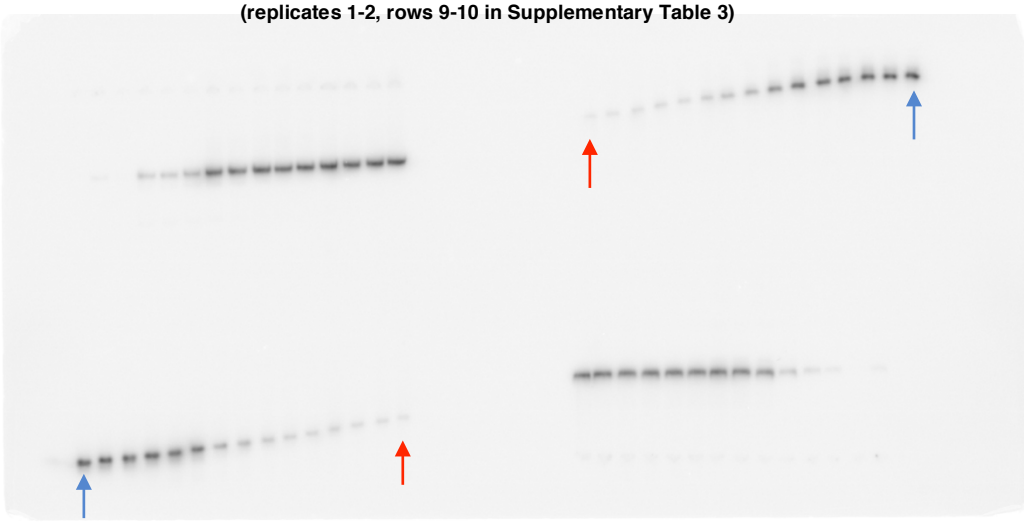
Gels used to infer p53 Kd for the two T-T mismatch site (replicates 3-4, rows 11-12 in Supplementary Table 3)



Gels used to infer p53 Kd for the two T-T mismatch site (replicates 5-6, rows 13-14 in Supplementary Table 3)

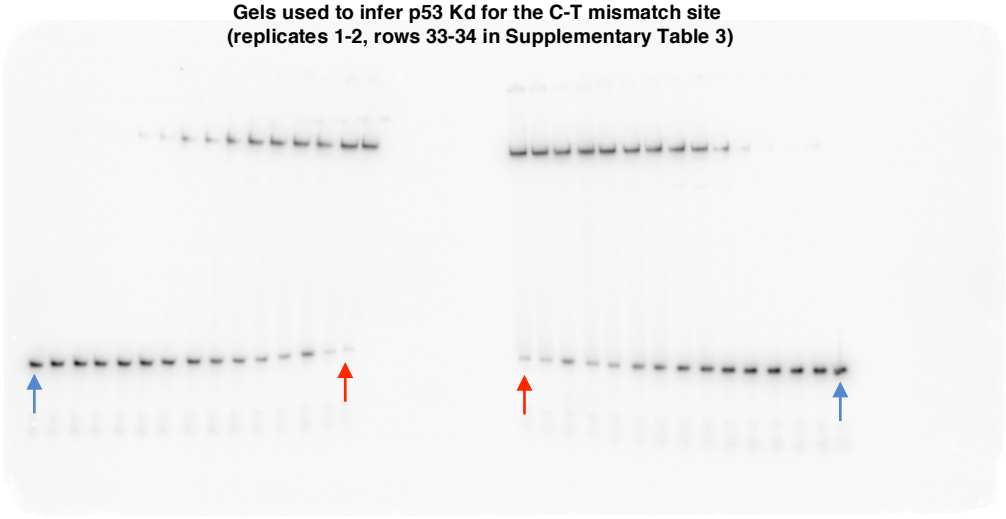


Gels used to infer p53 Kd for the two T-T mismatch site (replicates 1-2, rows 9-10 in Supplementary Table 3)

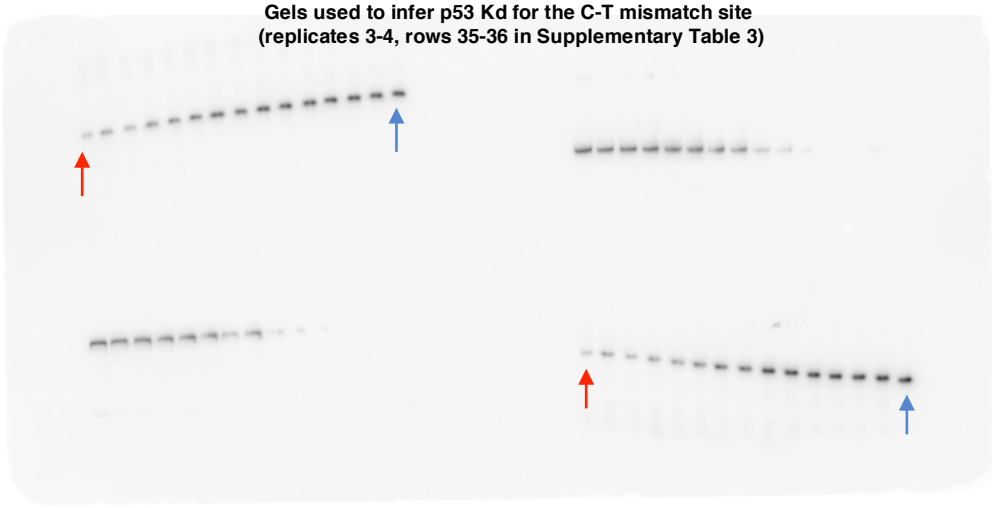


p53 two T-T mismatch.
Protein concentration: 0,0.4,0.8,2,4,8,16,24,32,40,60,80,120,160,240 [nM] (Blue arrow 0nM, and Red arrow 240nM)
Original high quality image scans:
https://figshare.com/articles/figure/p53_EMSA_gels/12568691
File names: MM_2xT.T_CT20-2_b1e2_10.6.19.tif , MM_2xT.T_CT20-4_b2e1_14.7.19.tif , MM_2xT.T&CT20-2_b1e1_4.6.19.tif

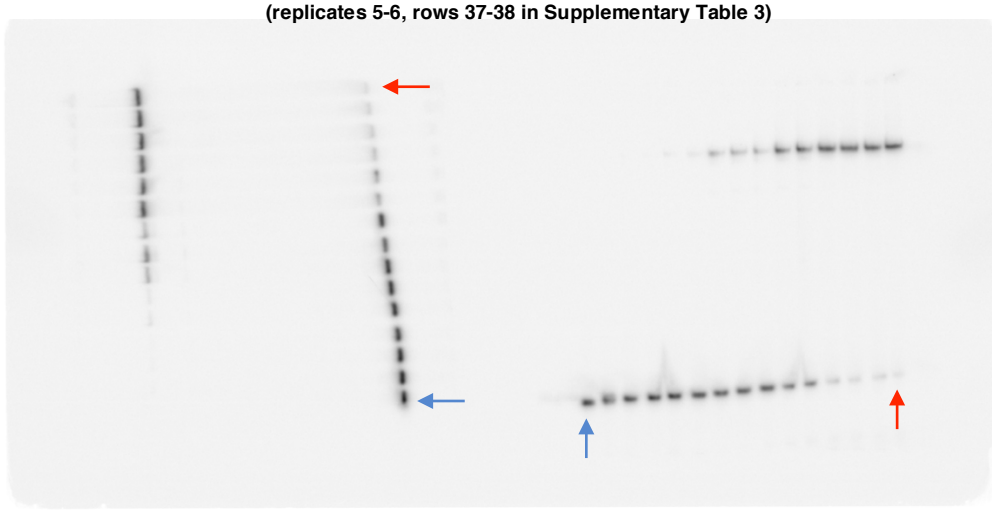
Gels used to infer p53 Kd for the C-T mismatch site
(replicates 1-2, rows 33-34 in Supplementary Table 3)



Gels used to infer p53 Kd for the C-T mismatch site
(replicates 3-4, rows 35-36 in Supplementary Table 3)

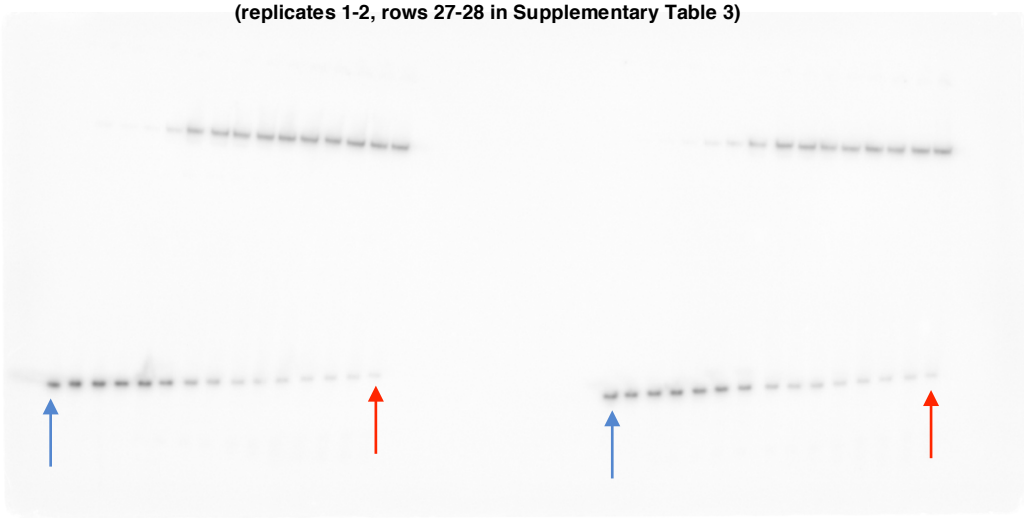


Gels used to infer p53 Kd for the C-T mismatch site
(replicates 5-6, rows 37-38 in Supplementary Table 3)

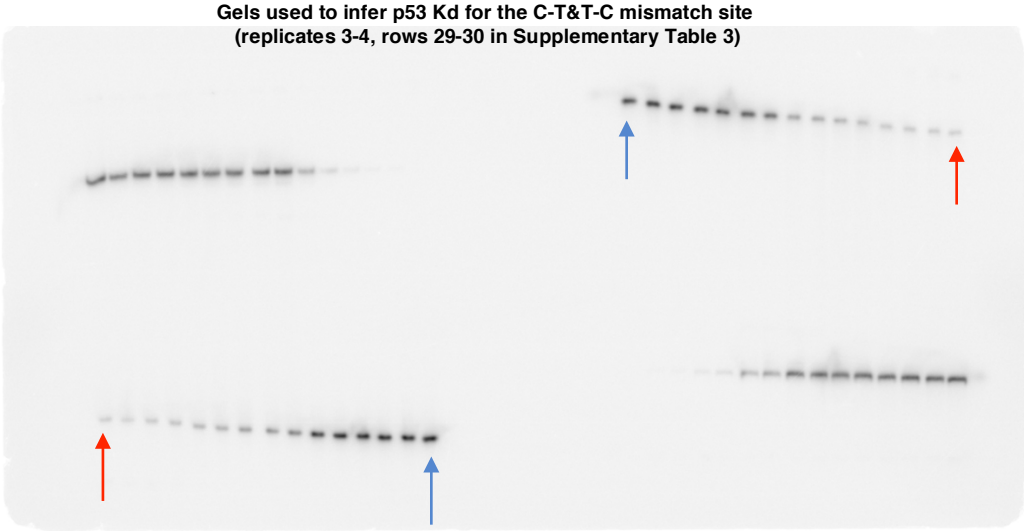


p53 C-T mismatch.
Protein concentration: 0,0.4,0.8,2,4,8,16,24,32,40,60,80,120,160,240 [nM] (Blue arrow 0nM, and Red arrow 240nM)
Original high quality image scans:
https://figshare.com/articles/figure/p53_EMSA_gels/12568691
File names: MM_C.T_CT20-2_b1e1_18.4.19.tif , MM_C.T_CT20-2_b1e2_22.4.19.tif , MM_C.T_CT20-2_b1e3_23.4.19.tif

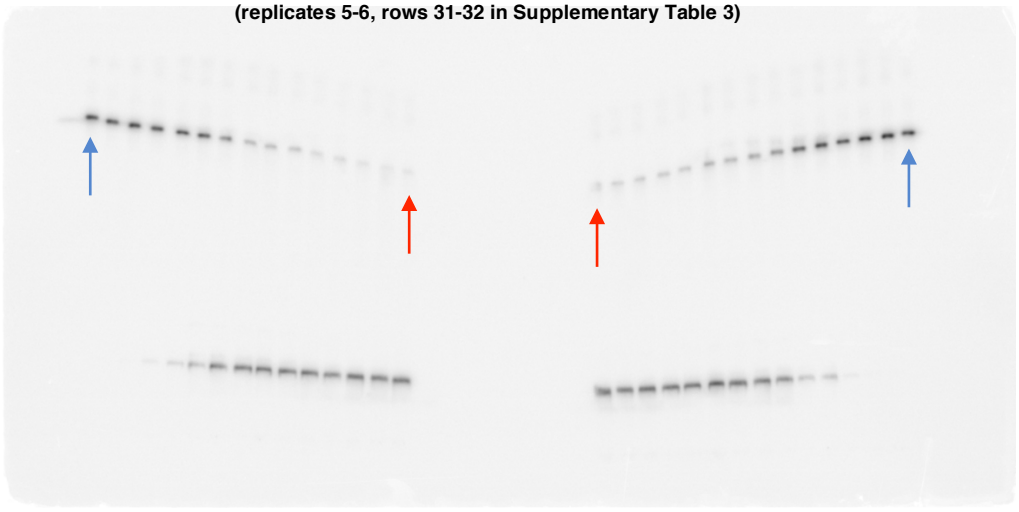
Gels used to infer p53 Kd for the C-T&T-C mismatch site (replicates 1-2, rows 27-28 in Supplementary Table 3)



Gels used to infer p53 Kd for the C-T&T-C mismatch site (replicates 3-4, rows 29-30 in Supplementary Table 3)

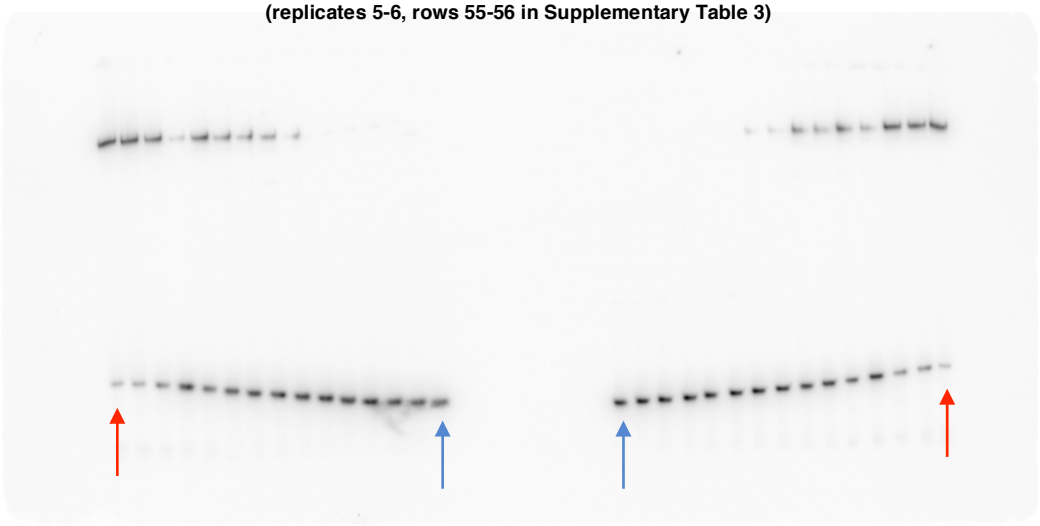


Gels used to infer p53 Kd for the C-T&T-C mismatch site (replicates 5-6, rows 31-32 in Supplementary Table 3)

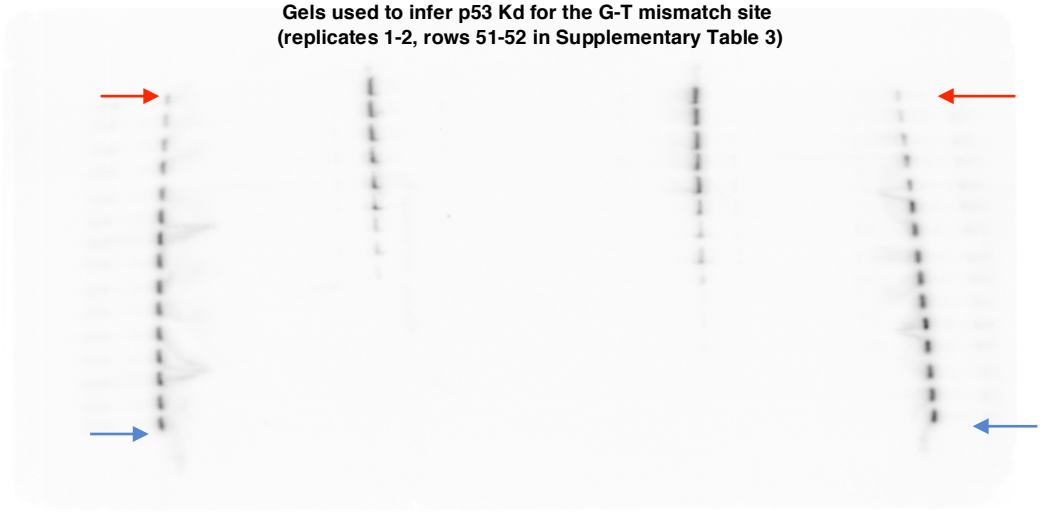


p53 C-T & T-C mismatches.
Protein concentration: 0,0.4,0.8,2,4,8,16,24,32,40,60,80,120,160,240 [nM] (Blue arrow 0nM, and Red arrow 240nM)
Original high quality image scans:
https://figshare.com/articles/figure/p53_EMSA_gels/12568691
File names: MM_C.T T.C_CT20-2_b1e1_6.6.19.tif , MM_C.T T.C_CT20-2_b1e2_11.6.19.tif , MM_C.T T.C_CT20-2_b1e4_19.6.19.tif

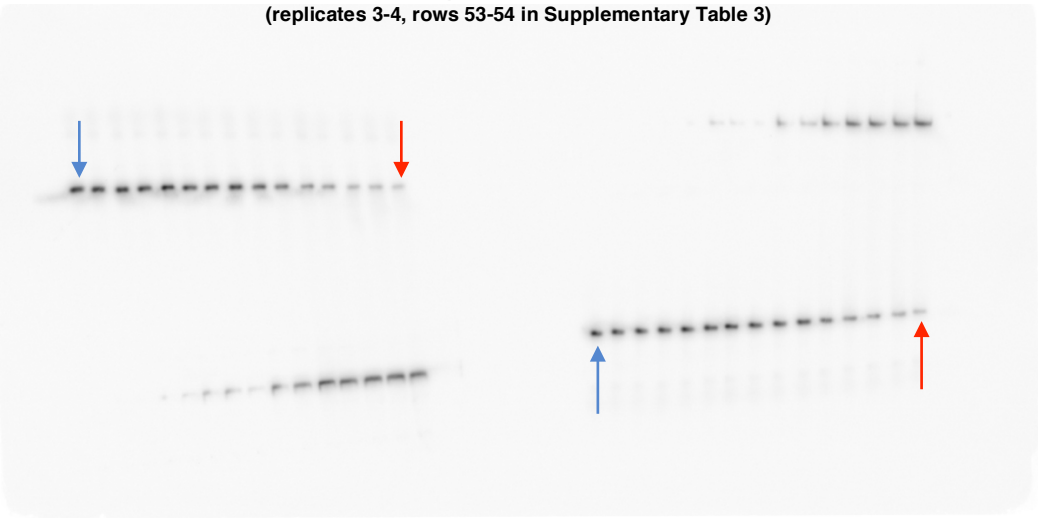
Gels used to infer p53 Kd for the G-T mismatch site
(replicates 5-6, rows 55-56 in Supplementary Table 3)



Gels used to infer p53 Kd for the G-T mismatch site
(replicates 1-2, rows 51-52 in Supplementary Table 3)

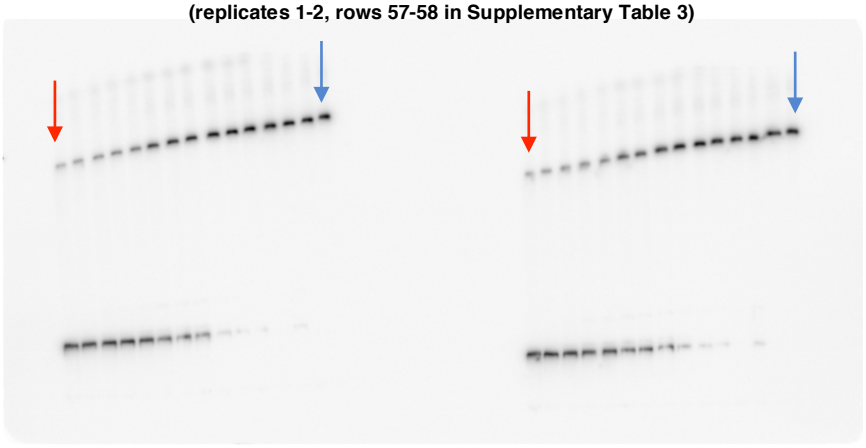


Gels used to infer p53 Kd for the G-T mismatch site
(replicates 3-4, rows 53-54 in Supplementary Table 3)

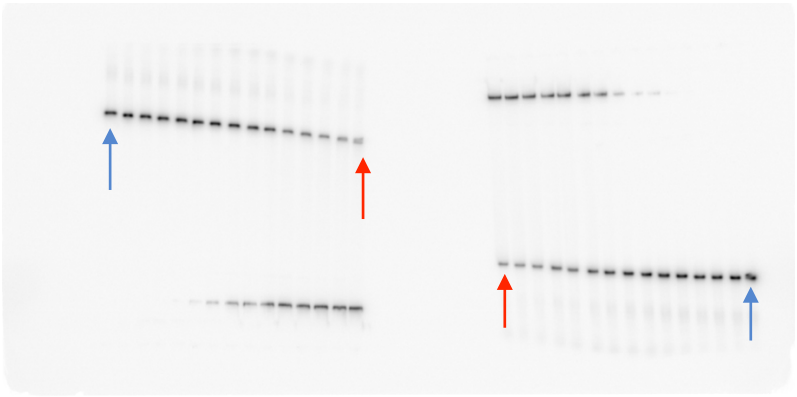


p53 G-T mismatch
Protein concentration: 0,0.4,0.8,2,4,8,16,24,32,40,60,80,120,160,240 [nM] (Blue arrow 0nM, and Red arrow 240nM)
Original high quality image scans:
https://figshare.com/articles/figure/p53_EMSA_gels/12568691
File names: MM_G.T_CT20-2_b1e3_16.4.19.tif , MM-G.T_CT20-1_b1e1_10.4.19.tif , MM-G.T_CT20-2_b1e2_11.4.19.tif

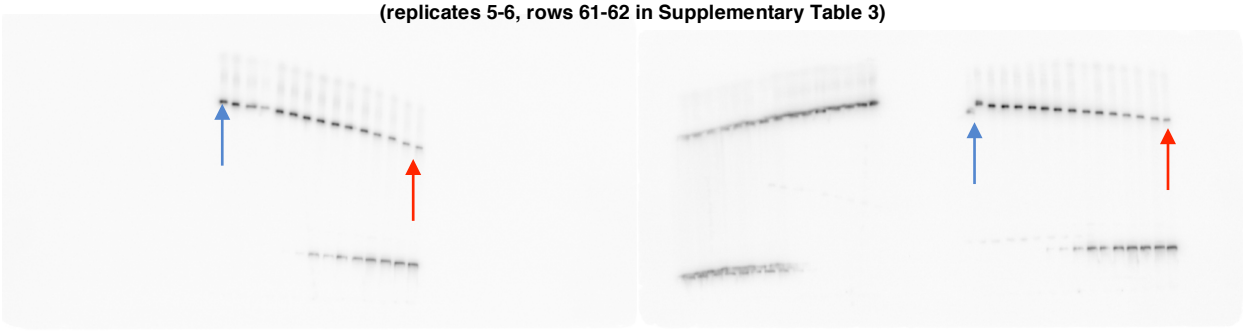
Gels used to infer p53 Kd for the G-A mismatch site
(replicates 1-2, rows 57-58 in Supplementary Table 3)



Gels used to infer p53 Kd for the G-A mismatch site
(replicates 3-4, rows 59-60 in Supplementary Table 3)

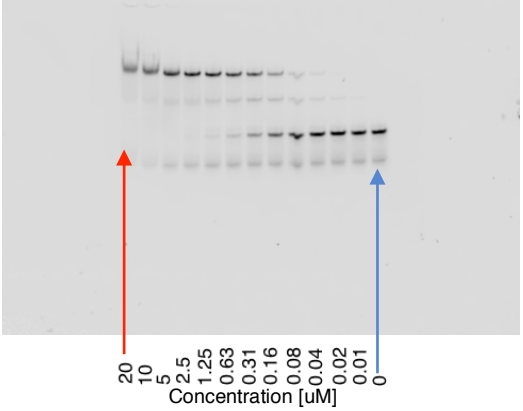


Gels used to infer p53 Kd for the G-A mismatch site
(replicates 5-6, rows 61-62 in Supplementary Table 3)

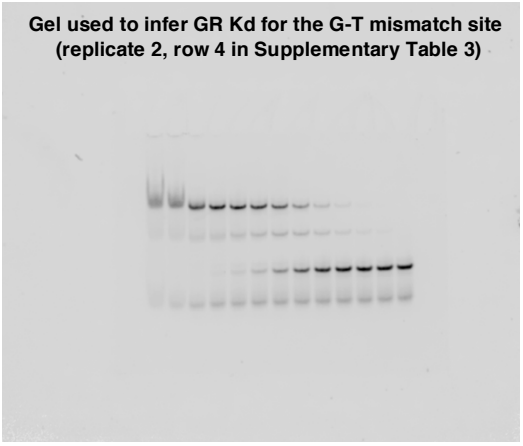


p53 G-A mismatch.
Protein concentration: 0,0.4,0.8,2,4,8,16,24,32,40,60,80,120,160,240 [nM] (Blue arrow 0nM, and Red arrow 240nM)
Original high quality image scans:
https://figshare.com/articles/figure/p53_EMSA_gels/12568691
File names: MM_G.A_CT20-4_b2e1_16.7.19.tif , MM_G.A_CT20-4_b2e2_16.7.19.tif , MM_G.A_CT20-4_b2e3_17.7.19.tif, MM_G.A_CT20-4_b2e4_17.7.19.tif

Gel used to infer GR Kd for the G-T mismatch site (replicate 1, row 3 in Supplementary Table 3)



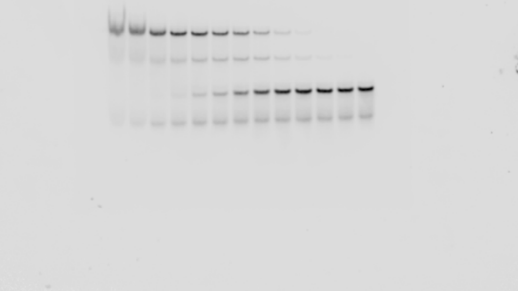
Gel used to infer GR Kd for the G-T mismatch site (replicate 2, row 4 in Supplementary Table 3)



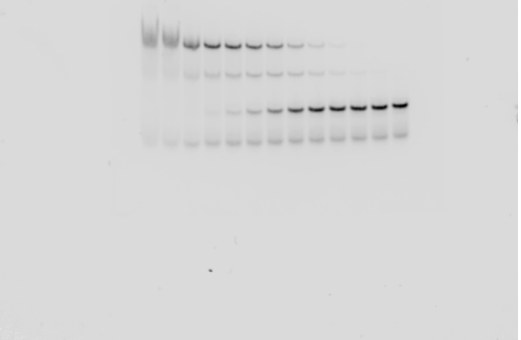
Gel used to infer GR Kd for the G-T mismatch site (replicate 3, row 5 in Supplementary Table 3)

GR MM1 (GT mismatch).
Protein concentration: 20,10,5,2.5,1.25,0.63,0.31,0.16,0.08,0.04,0.02,0.01,0 [uM]
(Blue arrow 0uM, and Red arrow 20uM), Original high quality image scans:
https://figshare.com/articles/figure/GR_EMSA_gels/12570236
File names: 07_23_19_MM1_2min_700nm.tif,07_26_19_MM1_2min_700nm.tif,07_29_19_MM1_2min_700nm.tif.

Gel used to infer GR Kd for the T-T mismatch site (replicate 1, row 6 in Supplementary Table 3)



Gel used to infer GR Kd for the T-T mismatch site (replicate 2, row 7 in Supplementary Table 3)

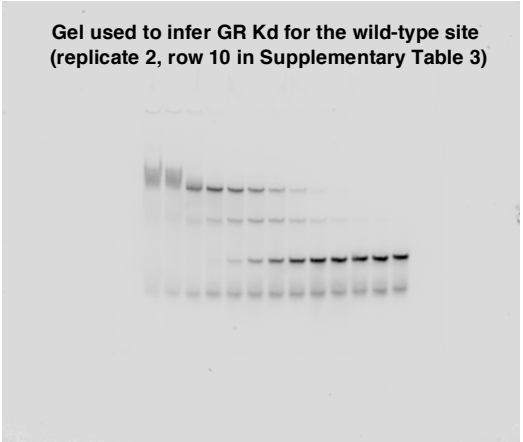


Gel used to infer GR Kd for the T-T mismatch site (replicate 3, row 8 in Supplementary Table 3)

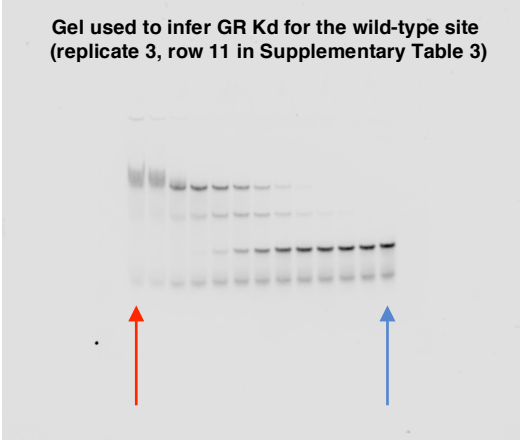
GR MM2 (TT mismatch).
Protein concentration: 20,10,5,2.5,1.25,0.63,0.31,0.16,0.08,0.04,0.02,0.01,0 [uM]
(Blue arrow 0uM, and Red arrow 20uM), Original high quality image scans:
https://figshare.com/articles/figure/GR_EMSA_gels/12570236
File names:07_23_19_MM2_2min_700nm.tif, 07_26_19_MM2_2min_700nm.tif, 07_29_19_MM2_2min_700nm.tif.

Gel used to infer GR Kd for the wild-type site (replicate 1, row 9 in Supplementary Table 3)

Gel used to infer GR Kd for the wild-type site (replicate 2, row 10 in Supplementary Table 3)

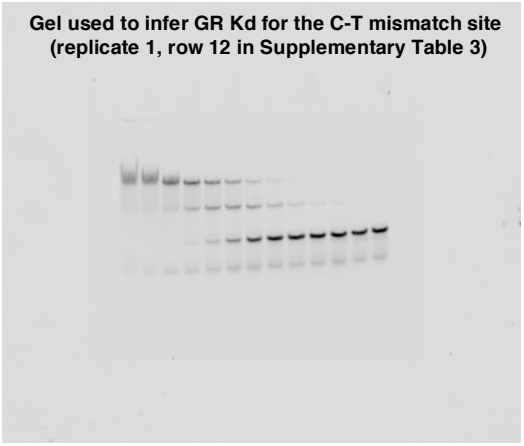


Gel used to infer GR Kd for the wild-type site (replicate 3, row 11 in Supplementary Table 3)

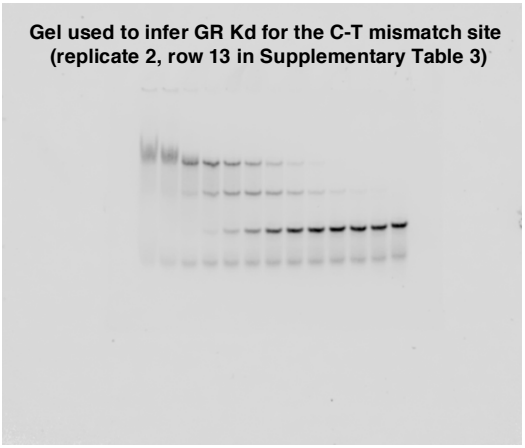


GR MM3 (Wild-type).
Protein concentration: 20,10,5,2.5,1.25,0.63,0.31,0.16,0.08,0.04,0.02,0.01,0 [uM]
(Blue arrow 0uM, and Red arrow 20uM), Original high quality image scans:
https://figshare.com/articles/figure/GR_EMSA_gels/12570236
File names: 07_23_19_MM3_2min_700nm.tif, 07_26_19_MM3_2min_700nm.tif, 07_29_19_MM3_2min_700nm.tif.

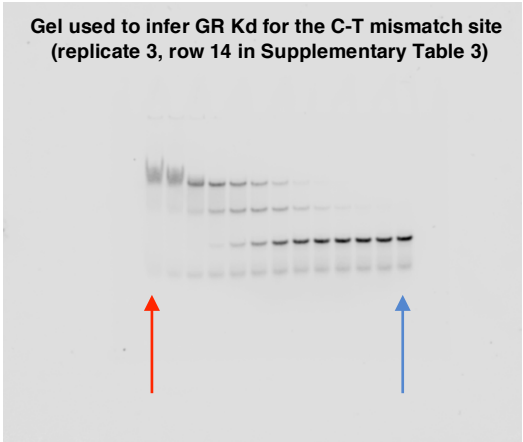
Gel used to infer GR Kd for the C-T mismatch site (replicate 1, row 12 in Supplementary Table 3)



Gel used to infer GR Kd for the C-T mismatch site (replicate 2, row 13 in Supplementary Table 3)



Gel used to infer GR Kd for the C-T mismatch site (replicate 3, row 14 in Supplementary Table 3)



GR MM4 (CT mismatch).
Protein concentration: 20,10,5,2.5,1.25,0.63,0.31,0.16,0.08,0.04,0.02,0.01,0 [uM]
(Blue arrow 0uM, and Red arrow 20uM), Original high quality image scans:
https://figshare.com/articles/figure/GR_EMSA_gels/12570236
File names: 07_23_19_MM4_2min_700nm.tif, 07_26_19_MM4_2min_700nm.tif, 07_29_19_MM4_2min_700nm.tif.