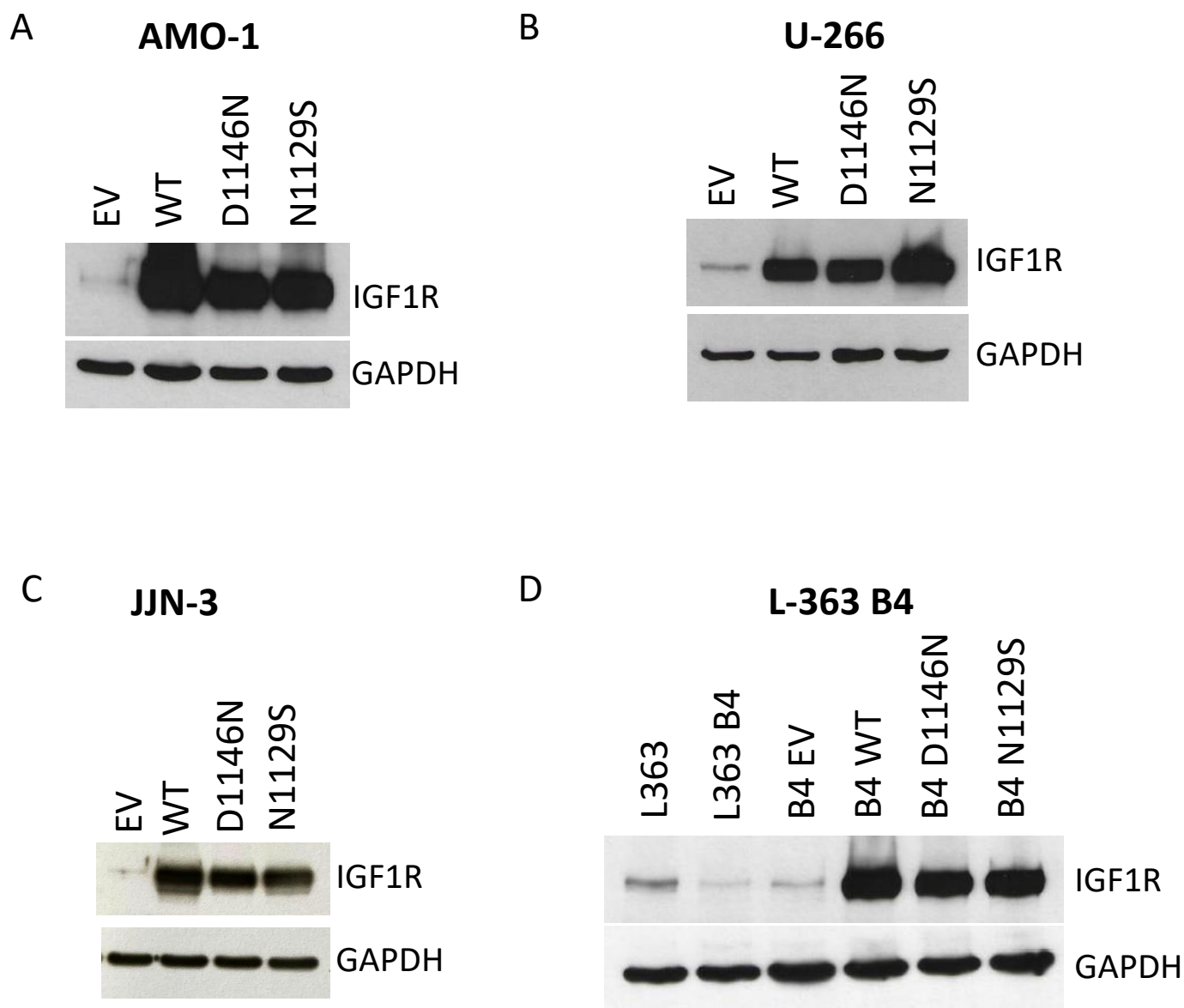
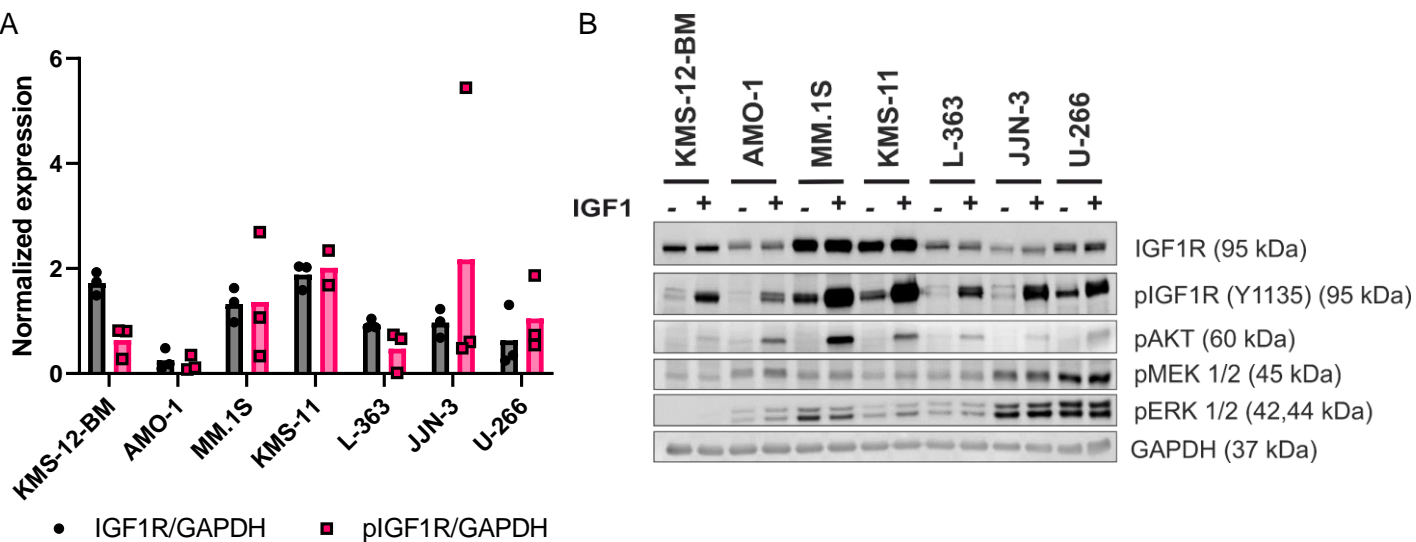


## Figure S1

Verification of IGF1R<sup>WT</sup> and IGF1R<sup>mut</sup> (D1146N and N1129S) overexpression in stably transfected AMO-1 (A), U-266 (B), JJN-3 (C) and L-363 B4 (D). GAPDH was detected on the same blot as IGF1R. EV: empty vector transfected control.



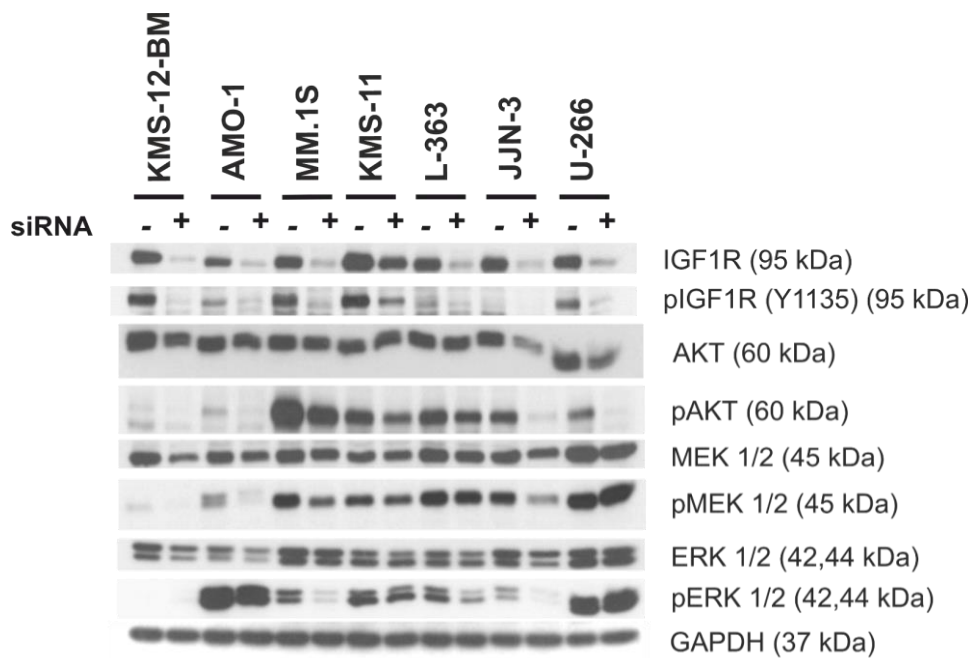
**(A)** Investigation of the expression and activation status of IGF1R under normal culturing conditions. Intensities revealed with the Fiji tool were calculated for each marker by normalizing it to those of the corresponding GAPDH bands. Data shown is mean of three independent experiments. **(B)** Expression and activation status of IGF1R and activation status of AKT, MEK and ERK before (serum-reduced (0.5% FBS)) and after stimulation with IGF1. Western blot shown is representative for three independent experiments.



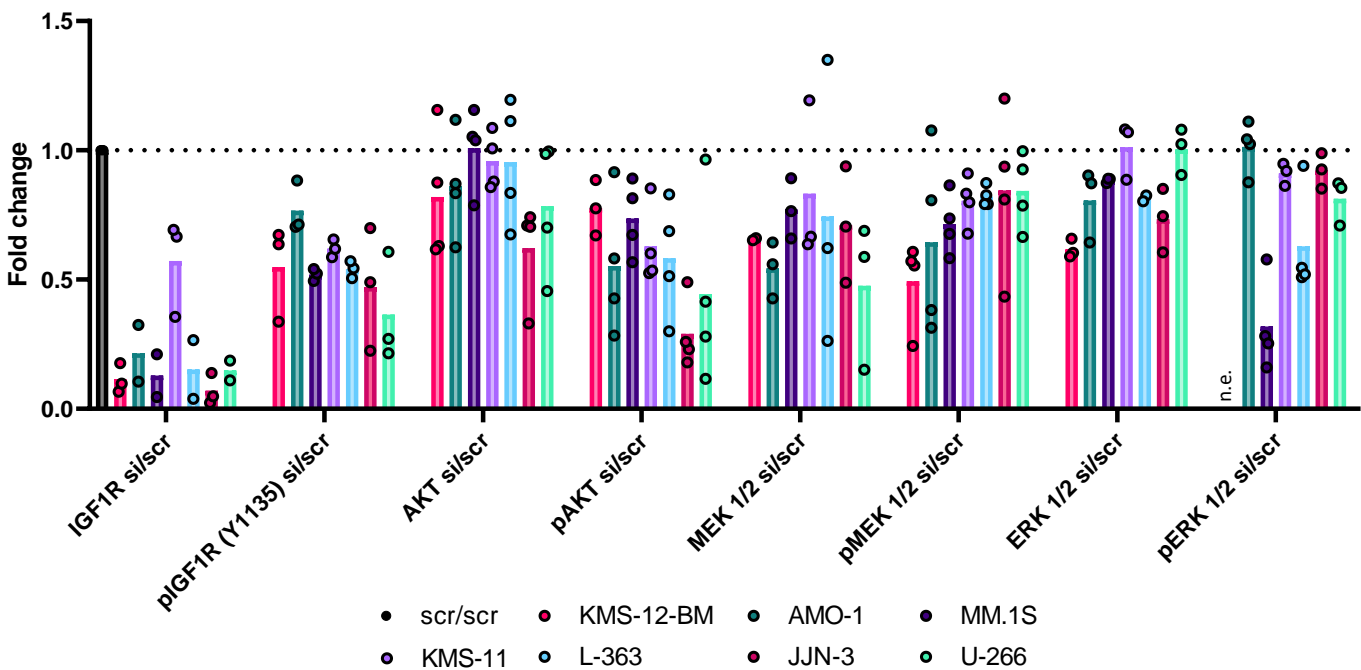
**Figure S3**

Investigation of the expression and activation status of potential IGF1R-effectors by Western analysis in 7 HMCLs after siRNA-mediated IGF1R-knockdown. **(A)** The blot shown is representative for three independent experiments. **(B)** In addition, the signal intensities as revealed by Western blot analyses in the siRNA knockdown experiments, were calculated for all repetitions using Fiji ("Gels" tool) and depicted as a bar diagram. Intensities revealed with the Fiji tool were calculated for each marker by normalizing it to those of the corresponding GAPDH bands and the intensities of the siRNA-treated samples were subsequently normalized to those of the corresponding scrRNA-treated samples. Cells transfected with scrambled siRNA (scrRNA) served as negative controls. Knockdowns were performed in at least three independent experiments. Each data point represents one independent experiment. The signal intensities for pERK in KMS-12-BM were too low for calculation with Fiji (n.e.).

A



B



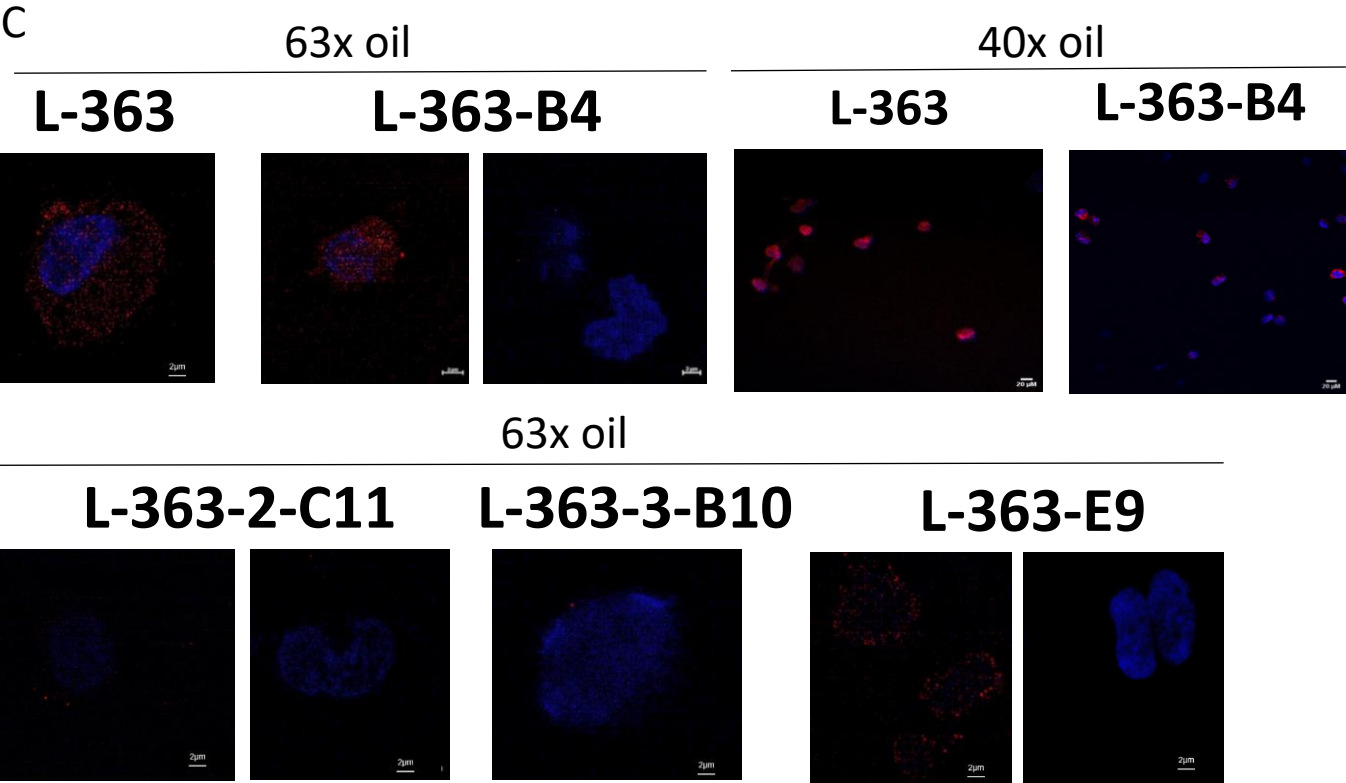


**Figure S4 Part I** Overview of L-363 CRISPR-Cas9 IGF1R knockout clones. **(A)** Comparison of IGF1R sequence in IGF1R KO clones at the sgRNA target site (Exon 18 KO). Figure adapted from Ref<sup>45</sup>.

A

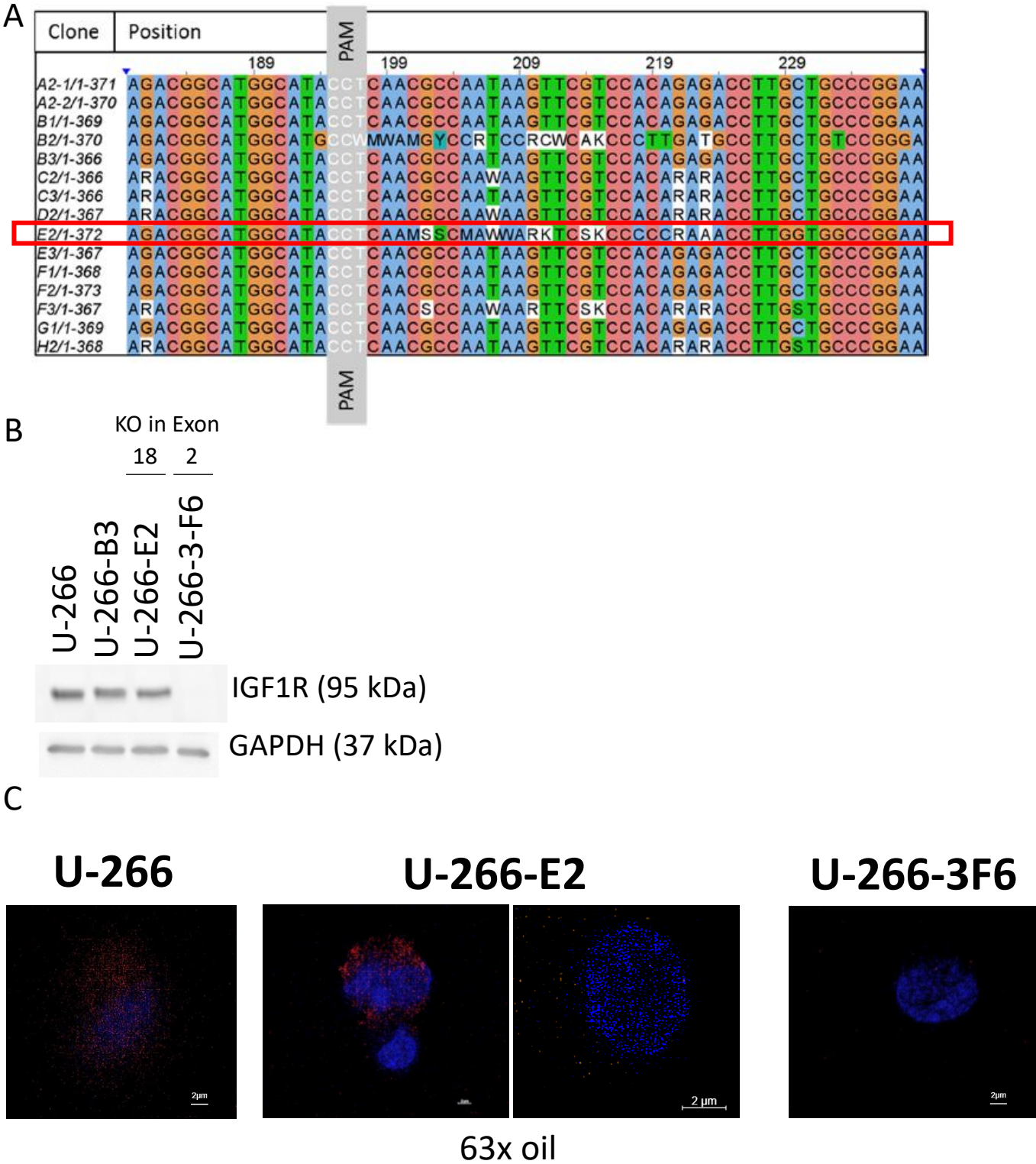
Clone	Position	PAM	173	183	193	203
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A2/1-368	AGACGGCA	TGGCMT	ACCT	CWVCGCCMA	YAARTTT	CSGCCACACASACCT
A3/1-366	AGACGGCA	TGGCMVA	ACCT	CAWCGCCMA	YAARTTT	CSGCCACAGASACCT
A4/1-375	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
A5/1-371	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
A6/1-372	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
A7/1-371	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
A8/1-370	AGACGGCA	TGGCAT	ACCT	CAACRMWAY	AASYT	CGTCCACMKAGACCT
A9/1-369	AGACGGCA	TGRCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
B1/1-372	AGACGGCA	TGGCAT	ACCT	CMA CGCCWA	TAAKYT	CSGCCACAGASACCT
B2/1-374	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
B3/1-375	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
B4/1-370	AGACGGCA	TGGCAT	ACCMWAMG	YCCRT	CCRCWCA	KCCCTTGATGCC
B5/1-369	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
B6/1-369	WSACGG	SWTGGYMT	ACCT	CAACGCCRA	TAAAGTT	CGTCCACAGAGACCT
B7/1-377	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
B8/1-369	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCAMARAGACCT
B9/1-374	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
C1/1-373	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
C2/1-372	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
C3/1-370	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
C4/1-366	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
C5/1-370	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
C6/1-366	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
C7/1-374	AGACGGCA	TGGCAT	ACCT	CMRSCCWAWA	AKYT	CSGCCACAGASACCT
C8/1-373	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
C9/1-374	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
D2/1-369	AGACGGCA	TGGCAT	ACCT	CMA CGCCMA	TAWKYT	CSGCCACASAKACCT
D3/1-373	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
D4/1-373	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
D5/1-377	AGACGGCA	TGGCAT	ACCT	CAMSSCMA	-TYAWYT	CCKCCMACCTTGCC
D6/1-370	AGACGGCA	TGGCAT	ACCT	CWACGCCMA	TAAARYT	CRKCCACWGAACCT
D7/1-365	AGACGGCA	TGGCAT	ACCT	CAACGMCMAC	AAAYACS	TAGACMTAGCCCT
D8/1-371	AGACGGCA	TGGCAT	ACCAWAA	SGTCKRT	CAATTARGT	CCCTTGASACCT
D9/1-370	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
E1/1-378	AGACGGCA	TGGCAT	ACCMWAA	SGTCCAT	CAATTGK	CCCTCGATGCCA
E2/1-365	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
E3/1-369	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
E4/1-366	AGACGGCA	TGGCMT	ACCW	CAACGWCWA	CAWAKACCT	TGACCTTGGTGC
E5/1-368	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
E6/1-369	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGARACCT
E7/1-372	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
E8/1-372	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
E9/1-366	AGACGGCA	TGGCAT	ACCT	CAAMS	SCMAWWARKT	CSKCCCCRAAAACM
F1/1-384	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
F2/1-369	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
F3/1-368	AGACGGCA	TGGCAT	ACCT	CAACRCCMA	WAARWT	CKYCCACASAGACCT
F4/1-368	AGACGGCA	TGGCAT	ACCT	CMA CS	CCAA	TAAAGTTCSGCCACAGASACCT
F5/1-366	AGACGGCA	TGGCAT	ACCT	CAACRMWAY	AASYT	CGTCCACMKAGACCT
F6/1-367	AGACGGCA	TGGCAT	ACCT	CAACRMCA	AYWTCY	CSWCCACCTTGACCT
F7/1-370	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
F8/1-370	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
F9/1-367	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
G2/1-369	AAACGGCA	TGGCAT	ACCT	CAAMS	SCCAWAAS	TTGKCCCCCRAGACCT
G3/1-367	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAKT	TCGYCCACARARACCT
G4/1-367	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
G5/1-370	AGACGGCA	TGGCAT	ACCT	CMA CGCCAA	TAAAGTT	CGTCCACAGAGACCT
G6/1-371	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
G7/1-371	AGACGGCA	TGGCAT	-CCT	CAACGCCAA	TAAAGTT	CGTCCACMRARACMT
G8/1-375	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
H2/1-377	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
H4/1-370	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
H5/1-376	AGACGGCA	TGGCAT	ACCT	CAMCS	CAAWAAKTY	CGYCCMCARARACCT
H6/1-371	AGACGGCA	TGGCAT	ACCT	CAACRMWAY	AASYT	CRKCCACMKAGACCT
H7/1-370	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT
H8/1-375	AGACGGCA	TGGCAT	ACCT	CAACGCCAA	TAAAGTT	CGTCCACAGAGACCT

**Figure S4 Part II** Overview of L-363 CRISPR-Cas9 IGF1R knockout clones. **(B)** Overview of IGF1R expression in all KO clones used for experiments (Exon 18 and Exon 2 KO clones). **(C)** Indirect immunofluorescence (IF) (not shown for B3). For IF, cells were stained with DAPI (CST (#4412); 1:1000). IGF1R [alphaR3] (Genetex mouse; 1:50) and goat/anti-mouse-Alexa-Fluor 647, (ThermoFisher (A32728), 1:400). Images were scanned using either a Zeiss Elyra S.1- structural imaging microscope (63x oil) and a S-CMOS-Camera: PCO Edge 5.5 or using a Leica SP2 confocal laser scanning microscope (40x oil). Analysis was performed using ZEN lite and Fiji.

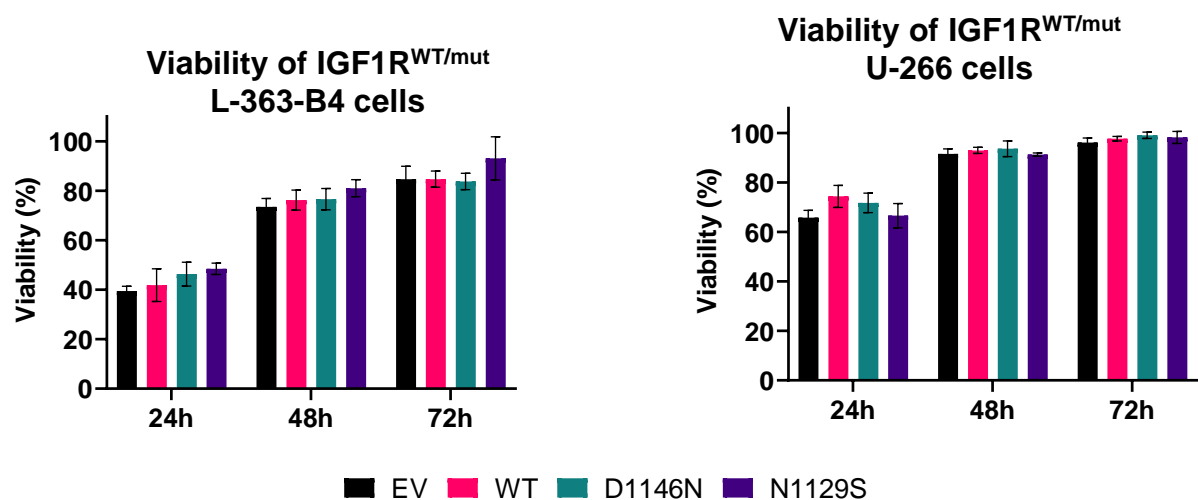




**Figure S5** Overview of U-266 CRISPR-Cas9 IGF1R knockout clones. **(A)** Comparison of IGF1R sequence in IGF1R KO clones at the sgRNA target site (Exon 18 KO). Figure adapted from Ref<sup>40</sup>. **(B)** Overview of IGF1R expression in all KO clones used for experiments (Exon 18 and Exon 2 KO clones). B3 is a WT single cell clone. **(C)** Indirect immunofluorescence (IF) (not shown for B3). For IF, cells were stained with DAPI (CST (#4412); 1:1000). IGF1R [alphaR3] (Genetex mouse; 1:50) and goat/anti-mouse-Alexa-Fluor 647, (ThermoFisher (A32728), 1:400). Images were scanned using a Zeiss Elyra S.1- structural imaging microscope (63x oil). Analysis was performed using ZEN lite and Fiji.



**Figure S6** Viability assay (AlamarBlue) of IGF1R overexpressing cells under normal culturing conditions. Results were derived from three independent experiments (mean  $\pm$  SEM).



**Figure S7** Treatment of MM cell lines with different concentrations of linsitinib and subsequent measurement of the effects on metabolism using an MTT-assay after 72h incubation (mean  $\pm$  SEM). For 48 and 96h see **Figure 5A**.

