24h

150

100

2.5

2.0

E

GI50= 14.76nM

1.5

1.0

A

MiaPaCa2

\*\*\*\*

\*\*\*\* \*\*\*\* \*\*\*\* \*\*\*\* \*\*\*\*

% Viability

Abs 450nm

50

0.5

0 0.0

10-1

100

Dose CuB nM

101

## 48h

150

100

Dose CuB nM

4

B

\*\*\*\*

\*\*\*\*

\*\*\*\*

\*\*\*\* \*\*\*\* \*\*\*\* \*\*\*\* \*\*\*\*

M

F

GI50= 39.22 n

3

Abs 450nm

2

50 1

% Viability

0

0 10-1 100 101

Dose CuB nM

## 72h

150

100

% Viability

50

Dose CuB nM

4G

3 GI50= 35.21 nM

Abs 450nm

2

1

0 0

C

\*\*\*\* \*\*\*\*

\*\*\*\*

\*\*\*\* \*\*\*\* \*\*\*\* \*\*\*\* \*\*\*\*

10-1 100 101

Dose CuB nM

## 96h

150 5

4

D

Dose CuB nM

\*\*\*\*

\*\*\*\* \*\*\*\* \*\*\*\* \*\*\*\*

\*\*\*\*

H

GI50= 66.69 nM

100

Abs 450nm

3

2

% Viability

50

1

0 0

10-1

100

Dose CuB nM

101

Dose CuB nM

Supplementary Figure 5 Viability of MiaPaCa2 cells treated with Cucurbitacin B (CuB) ranging from 0-1000 nM and Gemcitabine 50nM as determined by CCK8 colourimetric assay *MiaPaCa2 cells treated with CuB for A: 24 hours, B: 48 hours, C: 72 hours, D: 96 hours. GI50 values of MiaPaCa2 cells E: 14.76 nM for 24 hours F: 39.22 nM for 48 hours G: 35.21 nM for 72 hours and H: 66.69 nM for 96 hours. Significance between treatment groups and the negative control calculated using one-way ANOVA represented over treatment columns by ‘\*’ = p<0.05, ‘\*\*’ = p<0.01, ‘\*\*\*’ = p<0.005, ‘\*\*\*\*’ = p<0.001.*