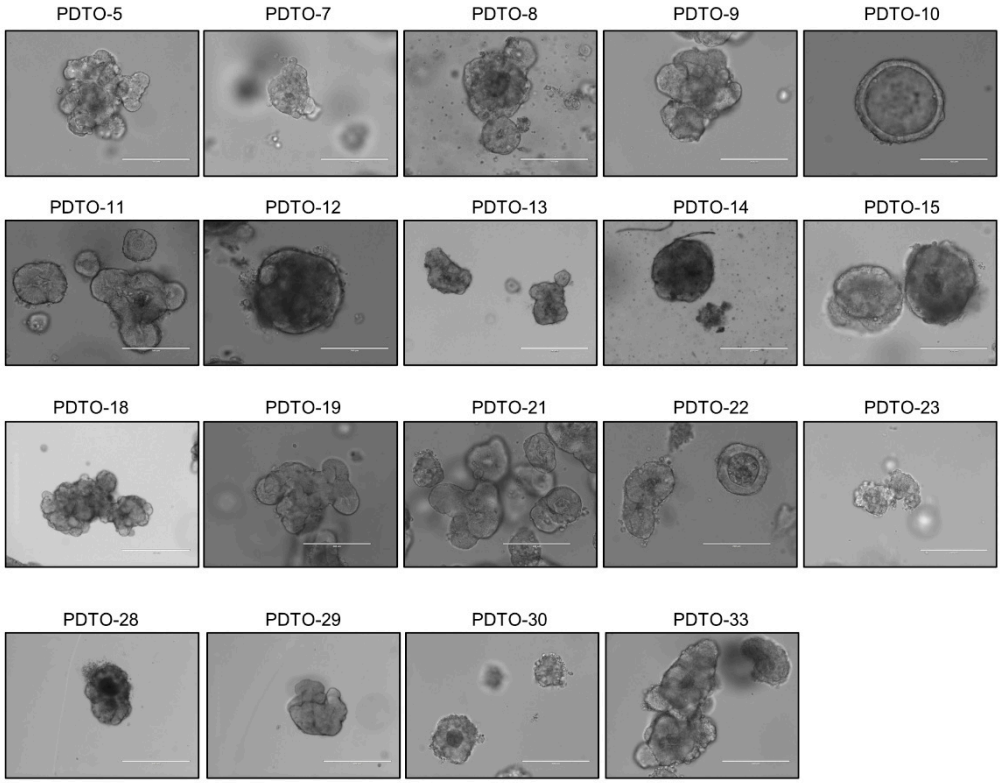


**Figure S1.** Observed morphologies of 19 PDTOs. PDTO, patient-derived tumour organoid.

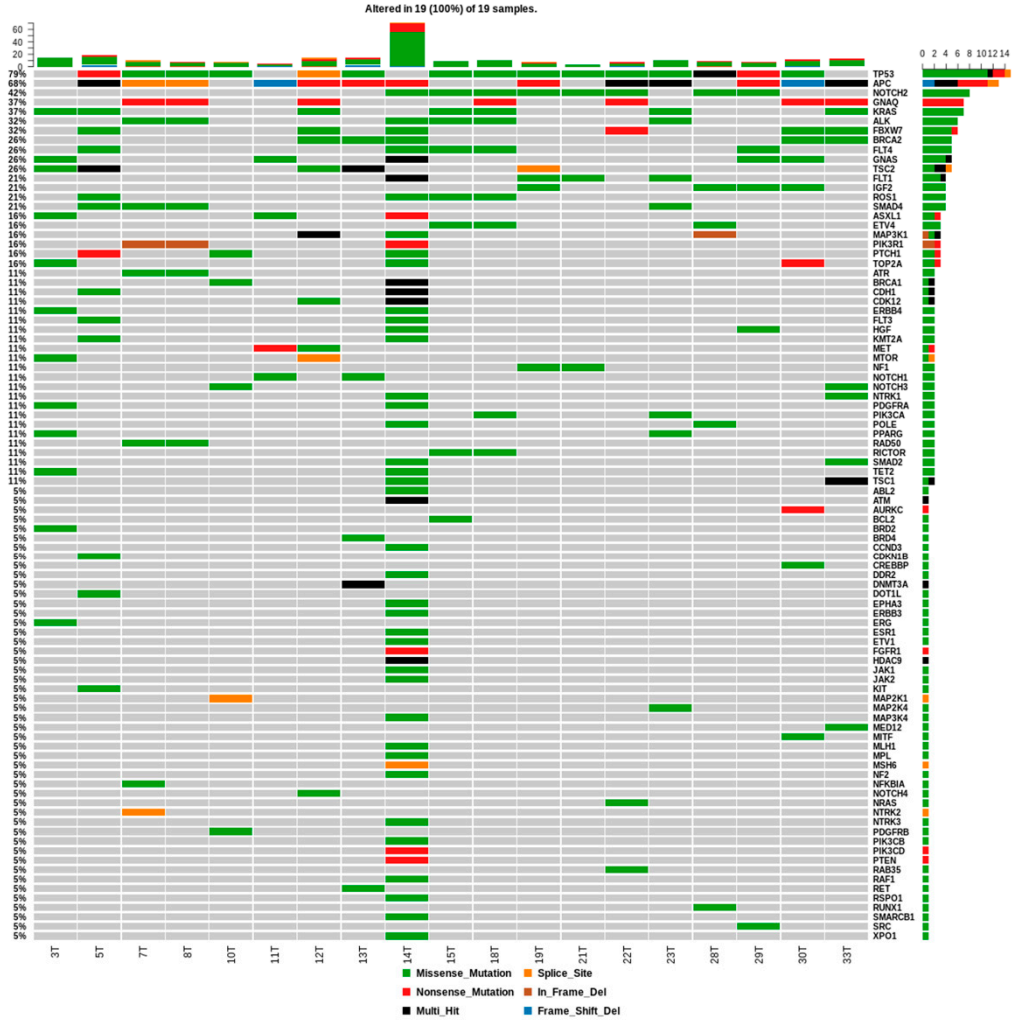


**Figure S2.** The mutation of 19 PDTOs for all alterations is displayed. The frequency of alteration is noted along with the type of genetic alteration relative to frame-shift, in-frame, missense, nonsense, or splice-site alterations (as indicated by the colour code). PDTO, patient-derived tumour organoid.

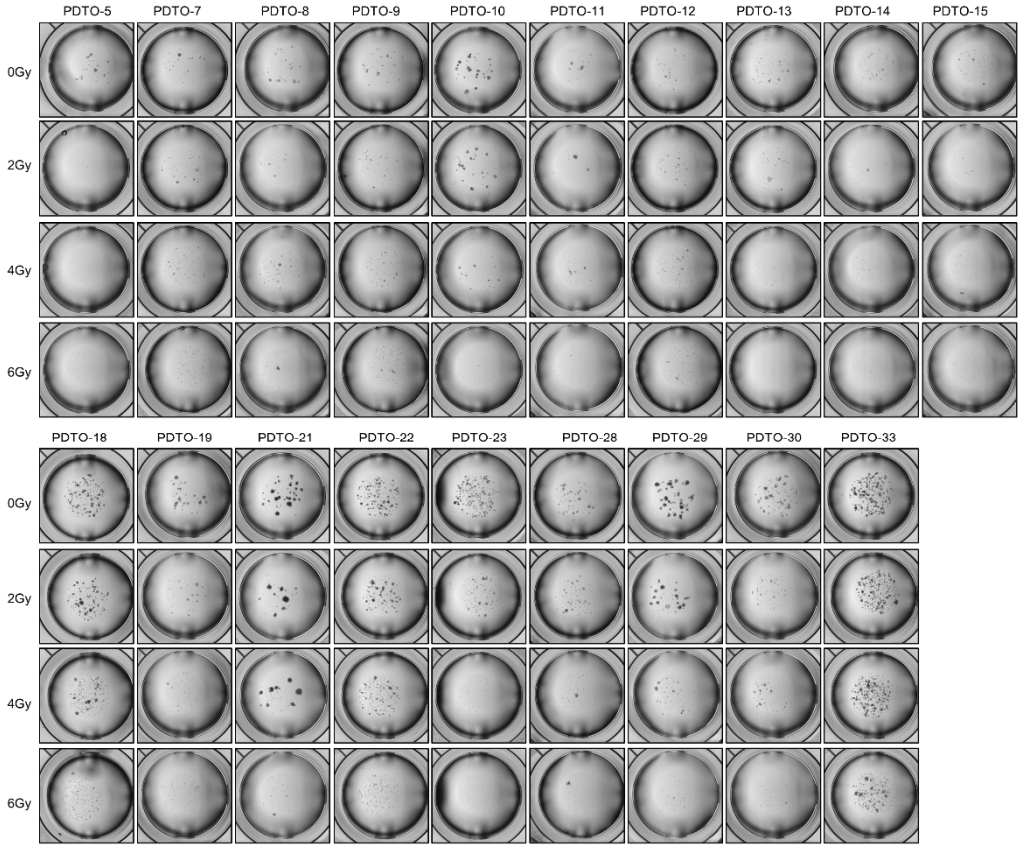


**Figure S3.** Morphologies of PDTOs after irradiation at 2 Gy, 4 Gy, and 6 Gy. PDTO, patient-derived tumour organoid.

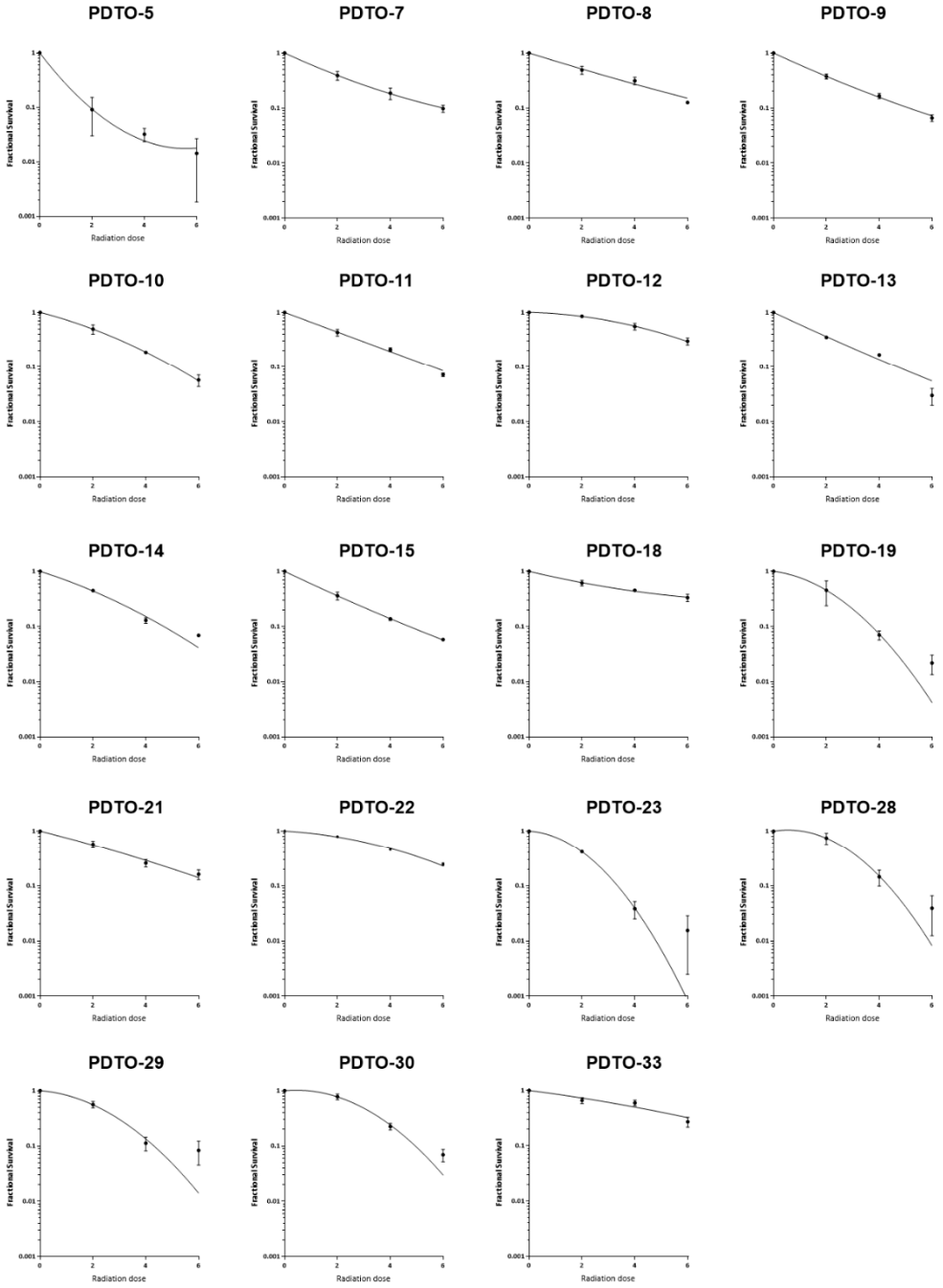


**Figure S4.** Dose-response of survival fraction in 19 PDTOs ($n$ = 4, independent experiments for each PDTO) is shown at 0 Gy, 2 Gy, 4 Gy, and 6 Gy. Data are presented as mean ± standard deviation. PDTO, patient-derived tumour organoid.

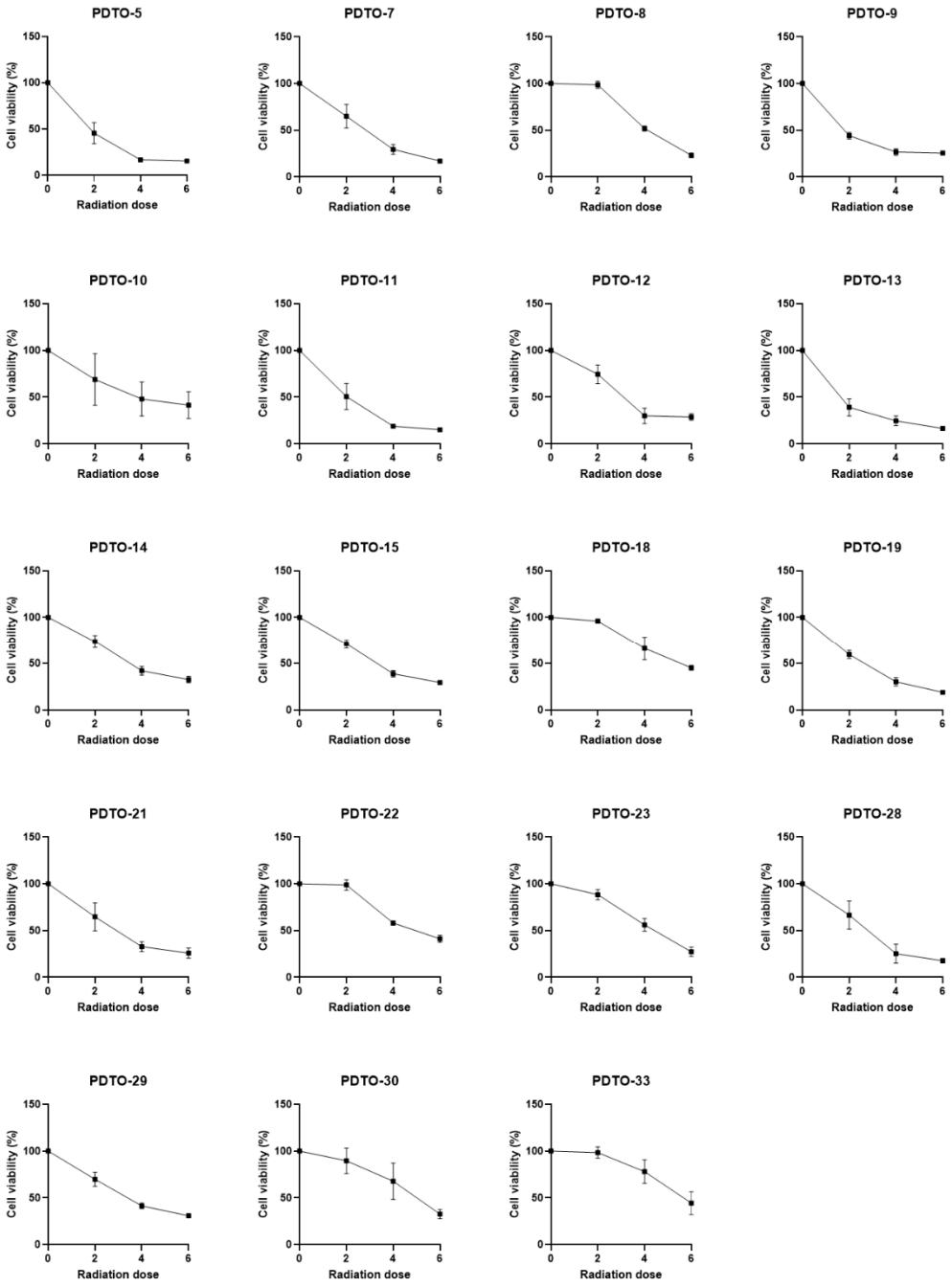


**Figure S5.** Dose-response of cell viability in 19 PDTOs ($n$ = 6, independent experiments for each PDTO) is shown at 0 Gy, 2 Gy, 4 Gy, and 6 Gy. Data are presented as mean ± standard deviation.
PDTO, patient-derived tumour organoid.

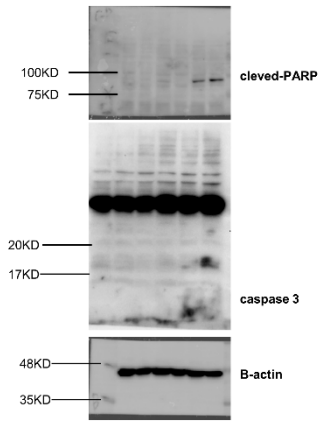


**Figure S6.** Whole blot showing all the bands with molecular weight marker.

**Table S1.** List of chemical and reagents used for studies.

| Chemical and Reagents | Source | Catalog number | Final concentration. |
|---|---|---|---|
| Matrigel | Corning | 356231 | |
| Advanced DMEM/F12 | Gibco | 12634-010 | |
| HEPES | Gibco | 15630-080 | 1x |
| Gluta max | Gibco | 35050-06 | 1x |
| Penicillin-Streptomycin | Gibco | 15140-122 | 1x |
| B27 | Gibco | 17504-044 | 1x |
| N-acetyl cysteine | United States Pharmacopeia | 1009005 | 1.25mM |
| human epidermal growth factor | BioVision | 4022-100 | 50ng/ml |
| human Noggin | Peprotech | 120-10c | 50ng/ml |
| gastrin | Sigma-Aldrich | G9145 | 10nM |
| A83-01 | BioVision | 1989-1 | 500nM |
| primocin | InvivoGen | ant-pm-1 | 100ng/ml |
| Y-27632 | BioVision | 1784-5 | 10uM |

**Table S2.** List of antibodies used for studies.

| Antibodies | Source | Catalog number | Antibody dilution |
|---|---|---|---|
| anti-Ki-67 | Abcam | ab16667 | 1:200 |
| anti-Muc2 | Abcam | ab90007 | 1:100 |
| anti-E-cadherin | BD Transduction Laboratories | 610181 | 1:200 |
| anti-VL1 | Santa Cruz Biotechnology | SC-28283 | 1:100 |
| anti-ChgA | ImmunoStar | 20086 | 1:100 |
| anti-CDX2 | Sigma-Aldrich | 235R-16 | 1:200 |
| anti-CK20 | Sigma-Aldrich | 320M-16 | 1:500 |
| anti-CK19 | Abcam | ab15463 | 1:400 |
| anti-cleaved PARP | Cell Signaling Technology | 5625 | 1:1000 |
| anti-caspase 3 | Cell Signaling Technology | 9662 | 1:1000 |
| β-actin | Sigma-Aldrich | A5441 | 1:4000 |
| goat anti-rabbit-Alexa594 | Thermo Fisher Scientific | A11012 | 1:200 |
| goat anti-mouse-Alexa488 | Thermo Fisher Scientific | A11001 | 1:200 |

**Table S3.** Results of ROC about two extreme categories. PPV; positive predictive value. NPV; negative predictive value.

| | | AUC (95% CI) | Sensitivity | Specificity | PPV | NPV |
|---|---|---|---|---|---|---|
| **TRG 0 or not** | D0 | 0.753 (0.644–0.863) | 78.3% | 69.2% | 52.9% | 87.8% |
| | Survival fraction model | 0.897 (0.83–0.965) | 95.0% | 78.6% | 61.3% | 97.8% |
| | Cell viabiiity model | 0.631 (0.525–0.737) | 89.7% | 39.3% | 33.8% | 91.7% |
| **TRG 3 or not** | D0 | 0.966 (0.926–1) | 100.0% | 93.7% | 75.0% | 100.0% |
| | Survival fraction model | 0.974 (0.941–1) | 100.0% | 93.8% | 75.0% | 100.0% |
| | Cell viabiiity model | 0.898 (0.827–0.968) | 77.8% | 92.6% | 66.7% | 100.0% |