Clinical and Translational Radiation Oncology 34 (2022) 112–119

Contents lists available at ScienceDirect

Clinical and Translational Radiation Oncology

journal homepage: www.sciencedirect.com/journal/clinical-and-translational-radiation-oncology

A scoping review of small animal image-guided radiotherapy research: Advances, impact and future opportunities in translational radiobiology

Kathryn H. Brown [a,*], Mihaela Ghita [a], Ludwig J. Dubois [b], Dirk de Ruysscher [c], Kevin M. Prise [a], Frank Verhaegen [c], Karl T. Butterworth [a]

[a] Patrick G. Johnston Centre for Cancer Research, Queen's University Belfast, Belfast, United Kingdom
[b] The M-Lab, Department of Precision Medicine, GROW – School for Oncology, Maastricht University, Maastricht, The Netherlands
[c] Department of Radiation Oncology (MAASTRO), GROW – School for Oncology and Developmental Biology, Maastricht University Medical Centre, Maastricht, The Netherlands

ARTICLE INFO

Keywords:
Radiotherapy
Preclinical models
Preclinical radiotherapy
Small animal irradiators

ABSTRACT

Background and purpose: To provide a scoping review of published studies using small animal irradiators and highlight the progress in preclinical radiotherapy (RT) studies enabled by these platforms since their development and commercialization in 2007.
Materials and methods: PubMed searches and manufacturer records were used to identify 907 studies that were screened with 359 small animal RT studies included in the analyses. These articles were classified as biology or physics contributions and into subgroups based on research aims, experimental models and other parameters to identify trends in the preclinical RT research landscape.
Results: From 2007 to 2021, most published articles were biology contributions (62%) whilst physics contributions accounted for 38% of the publications. The main research areas of physics articles were in dosimetry and calibration (24%), treatment planning and simulation (22%), and imaging (22%) and the studies predominantly used phantoms (41%) or *in vivo* models (34%). The majority of biology contributions were tumor studies (69%) with brain being the most commonly investigated site. The most frequently investigated areas of tumor biology were evaluating radiosensitizers (33%), model development (30%) and imaging (21%) with cell-line derived xenografts the most common model (82%). 31% of studies focused on normal tissue radiobiology and the lung was the most investigated site.
Conclusions: This study captures the trends in preclinical RT research using small animal irradiators from 2007 to 2021. Our data show the increased uptake and outputs from preclinical RT studies in important areas of biology and physics research that could inform translation to clinical trials.

Introduction

Radiotherapy (RT) is a highly effective cancer treatment that is delivered to around 50% of all cancer patients [1]. RT continues to evolve, largely driven by advances in technology yet advanced preclinical studies using small animal irradiators are enabling more sophisticated studies to be undertaken leading to an increased understanding of radiobiological responses at the cell, tissue and whole-organism levels.

Small animal models of RT response are important tools in bridging the gap between *in vitro* concepts and translation to the clinic [2]. Clinical translation is often viewed as a 2-step process: the translation of *in vitro* data to preclinical animal models, and the transfer of knowledge gained from preclinical animal models to clinical practice [3]. However, an important question is how well preclinical models reflect human disease phenotypes and responses to treatment.

Conventional radiobiology studies have been commonly performed using broad fields delivered from fixed sources with lead shielding for beam targeting. These approaches lack image guidance or treatment planning systems (TPS) and had limited dosimetry and quality assurance

* Corresponding author at: Patrick G. Johnston Centre for Cancer Research, Queen's University Belfast, 97 Lisburn Road, Belfast BT9 7AE, United Kingdom.
E-mail addresses: kathryn.brown@qub.ac.uk (K.H. Brown), m.ghita@qub.ac.uk (M. Ghita), ludwig.dubois@maastrichtuniversity.nl (L.J. Dubois), dirk.deruysscher@maastro.nl (D. de Ruysscher), k.prise@qub.ac.uk (K.M. Prise), frank.verhaegen@maastro.nl (F. Verhaegen), k.butterworth@qub.ac.uk (K.T. Butterworth).

https://doi.org/10.1016/j.ctro.2022.04.004
Received 25 March 2022; Accepted 4 April 2022
Available online 6 April 2022

[4–5]. Small animal irradiators have largely addressed these limitations by downscaling beam geometries and incorporating cone beam CT image guidance and dedicated TPSs [2,4,6]. Several small animal irradiators have been developed consisting of rotating or fixed gantries with CBCT detectors or conversions of micro-CT devices [5]. Two systems have been commercialized and widely established into research laboratories around the world; the small animal radiotherapy research platform (SARRP, Xstrahl Life Sciences) originally developed at Johns Hopkins University [7], and the X-Rad small animal radiotherapy (SmART) system from Precision X-ray Inc originally developed at Princess Margaret Hospital [8]. The principles and development of small animal irradiators has been discussed previously in several reviews [2,4,9]. In this study, we aimed to provide a scoping review of the published literature in preclinical RT research using small animal irradiators and to describe the trends in research from 2007 to 2021.

Materials and methods

Search strategy

This review was restricted to peer-review research articles presenting novel experimental findings using small animal image-guided irradiators. Articles were first identified from PubMed searches using search criteria of "small animal image-guided radiotherapy", "small animal irradiator", "preclinical image-guided radiotherapy", "tumor radiotherapy preclinical model" and "normal tissue radiotherapy preclinical model". Articles were then cross referenced with manufacturer records requested from Xstrahl and Precision X-ray Inc databases to add papers which were missed in the initial search. A total of 907 studies were identified.

Exclusion criteria

Article screening and exclusions are displayed in Fig. 1A. This review was restricted to articles published from 2007 to December 2021. 848 studies were assessed for relevance with their title, abstract and methods manually screened. Review articles, poster/conference abstracts, technical notes, studies using companion animals, studies using internal sources of radiation (brachytherapy) and studies that did not use small animal image-guided sources (including clinical sources and preclinical non-image guided studies) were excluded from the analysis (489) (Fig. 1A). A total of 359 studies were identified for further review.

Article screening and classification

The abstract and methods (and further if required) of the identified articles were carefully read. The aim of each article was used to classify the article as a biology (221) or physics (138) contribution and are presented by year in Fig. 1B. The articles were sub-classified based on the main research focus, experimental models used and study design.

Research areas for physics studies were outlined as dosimetry & calibration, treatment planning and simulation, imaging, platform development, novel detectors, phantom development and *in vivo* dosimetry. The reported irradiation protocols and methods were screened to record the irradiator manufacturer, treatment schedule (single or fractionated dose schedule), collimator size, imaging modality and model used. The research model used was recorded as *in vivo* (small animal model), *in silico* (computational simulations) or phantom (in-house and commercial).

Biology studies were classified as normal tissue (69) or tumor focused studies (152). Both tumor and normal tissue types were

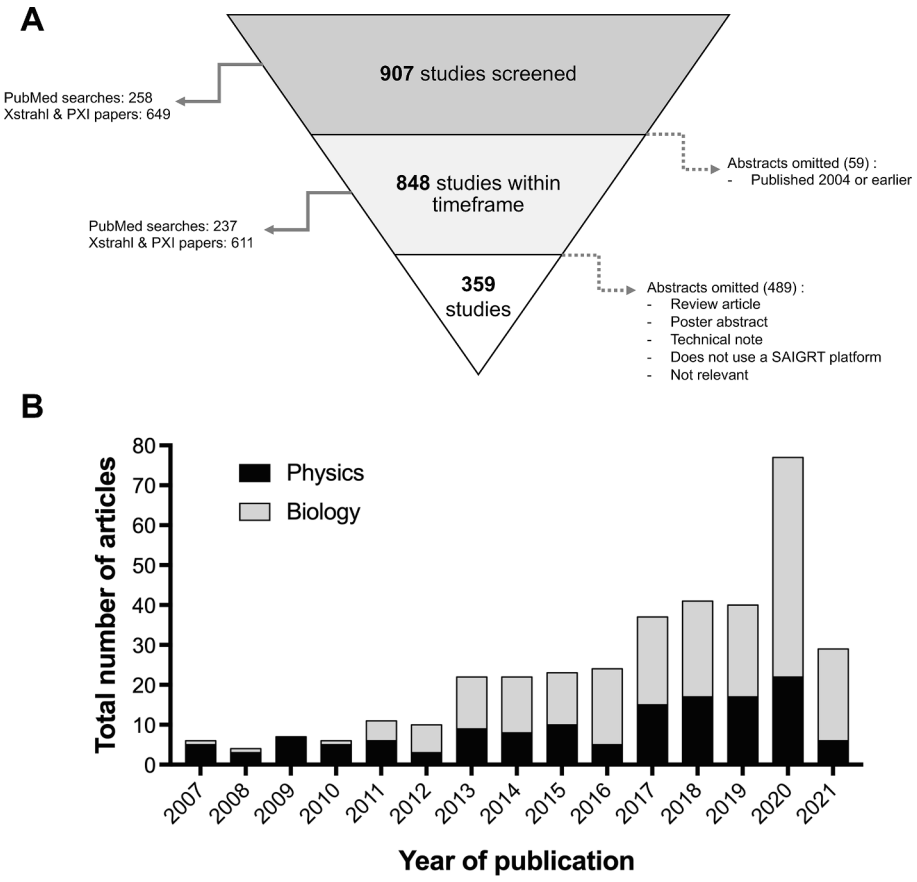

Fig. 1. Overview of article screening and the analysis of small animal image-guided radiotherapy publications by year according to study area. Panel A: A flow chart of the article process used to identify a total of 359 articles from 2007 to 2021 and those that were excluded in the analysis; Panel B: Classification of articles in areas of physics & biology research from 2007 to 2021. In total there are 38% physics contributions and 62% biology contributions.

classified by target tissue or tumor model detailed in the methods. Studies which used multiple tumor types within the same study were added to an additional subgroup titled 'multiple'. The biology contributions were also subdivided into 6 main research areas of radiosensitizers, model development, imaging, delivery modality, radioprotectors and fiducial marker. Details of all drug + RT combinations, imaging contrast agents, fiducial markers or study specific information was recorded during screening. Tumor models were classified as cell-line derived, patient derived xenograft (PDX), environmentally induced or genetically engineered mouse model (GEMM).

Results

We analyzed the outputs from 359 articles that reported the use of small animal irradiators from 2007 to 2021. Studies published from 2007 to 2010 were largely physics contributions (Fig. 1B). Following commercialization and the early uptake of small animal irradiators in 2011, a steady growth in the physics and biology publications was observed, and since 2016 there has been an increasing contribution from biology focused publications (Fig. 1B). The exponential growth of studies published by year did not continue in 2021 as a significant decrease in the number of published articles was observed in 2021 (Fig. 1B). These data could potentially be due to the impact of the coronavirus pandemic on research activities in this area.

Physics contributions

From a total of 138 physics papers, we identified 7 main research areas reported in the literature with the majority of studies focused on dosimetry & calibration (24%), treatment planning & simulation (22%) and imaging (22%) (Fig. 2A). These research areas are essential for the development and quality control of preclinical RT set-ups and have enabled the exponential growth of biology contributions (Fig. 1B). Novel detectors, phantom development and *in vivo* dosimetry represent challenging aspects of preclinical RT requiring further attention due to the very small dimensions of the target regions relative to the established methods in the clinic. These 3 areas only accounted for 18% of the total physics contributions (Fig. 2A).

Experimental models from physics contributions

Physics studies used 3 main experimental approaches of *in silico*, phantom and *in vivo* models with the majority of studies conducted using phantoms (41%) (Fig. 2B). The three largest research areas of dosimetry & calibration (24%), treatment planning & simulations (22%) and imaging (22%) required the use of all 3 types of models (Fig. 2C).

Phantoms were reported in the majority of research areas (41%), highlighting their importance for the development of small irradiators (Fig. 2C). *In vivo* models were most frequently used in imaging studies (12%) and for platform development (9%). Multi-tissue density phantoms were used in 6 out of 7 of the main research areas. *In silico* models were used in 25% of physics studies (Fig. 2B) and mainly applied to treatment planning & simulations (13%), and dosimetry & calibration studies (9%) (Fig. 2C).

Biology contributions

The majority of published articles were biology contributions (62%). Over two thirds of these articles focused on evaluating tumor responses (69%) while 31% focused on responses in normal tissues. Fig. 3A & B

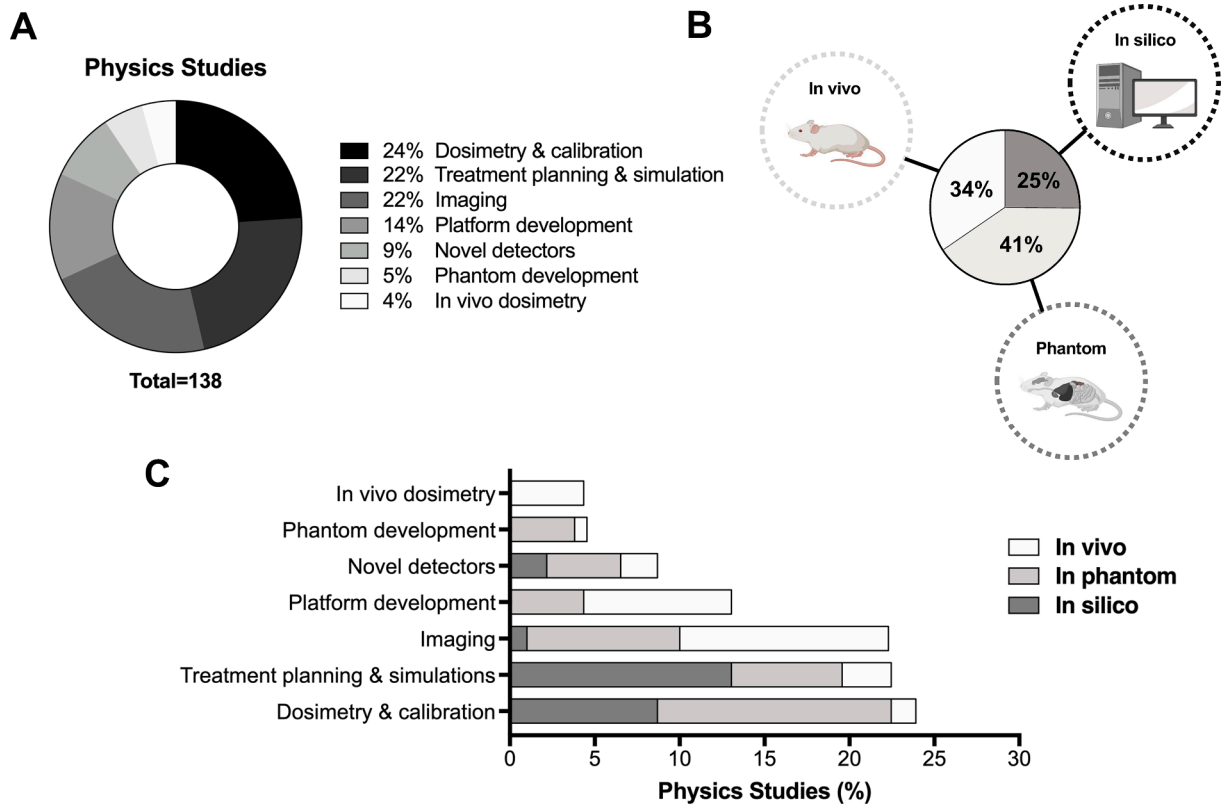

Fig. 2. Analysis of physics papers according to the main research areas and experimental models reported from 2007 to 2021. Panel A: Breakdown of physics studies into main research aims or area of interest of the published studies. Panel B: The percentage contribution of different experimental approaches used in physics articles. Studies reported the use of phantom (41%), *in vivo* (34%) and *in silico* (25%) approaches. Panel C: The distribution of the experimental approaches according to the main areas of physics research. From a total of 138 papers, we identified 7 main research areas reported in the literature with 3 main research focuses of dosimetry & calibration (24%), treatment planning & simulations (22%) and imaging (22%).

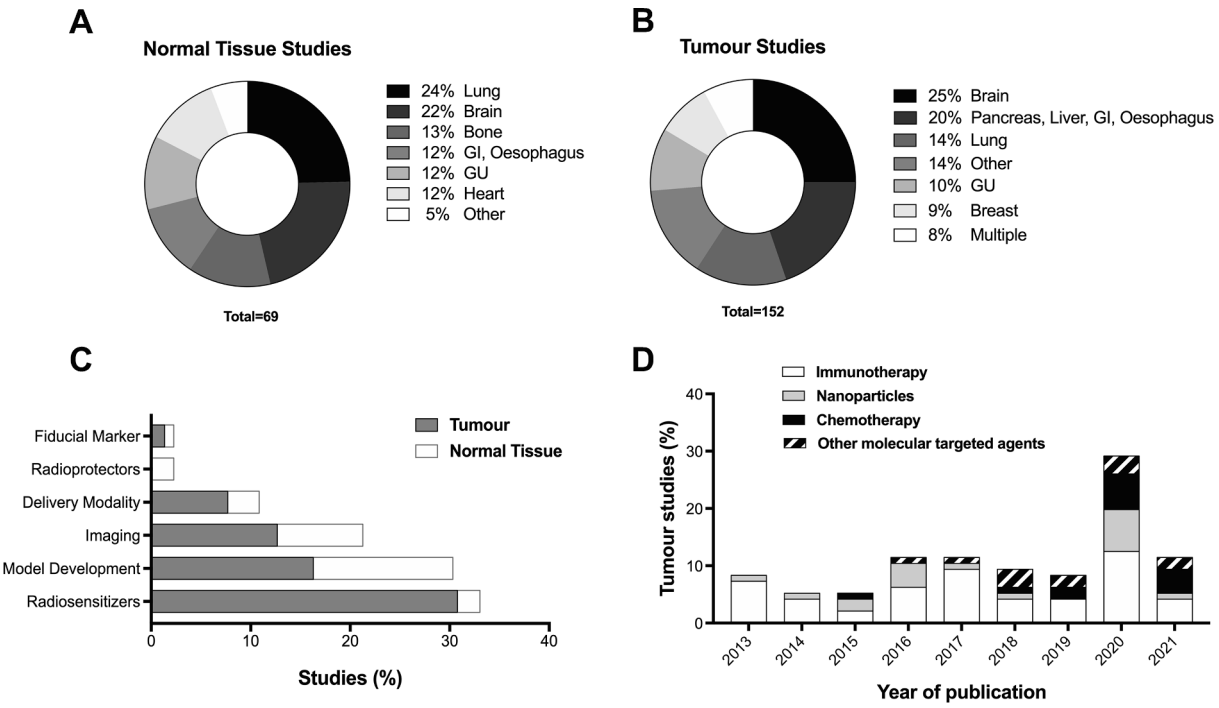

Fig. 3. Classification of biology papers based on normal tissue site and tumor indications from 2007 to 2021. Panel A: Percentage breakdown of the tissue types reported from a total of 69 normal tissue articles. Panel B: Distribution of tumor models reported from a total of 152 studies. Multiple includes studies which reported more than 1 tumor model. Panel C: Analysis of biology papers based on the main research areas. From a total of 221 papers, we identified 6 main research areas reported in the literature with 3 main research focuses of radiosensitizers (33%), model development (30%) and imaging (21%).Panel D: The percentage of tumor studies which combine RT with therapeutic agents reported from 2013 to 2021. We identified 4 main groups of immunotherapy, nanoparticles, chemotherapy, and other molecular targeted agents from a total of 96 tumours studies. Other molecular targeted agents encompass hypoxic agents, viral agents and cannabinoids.

shows the distribution of the main normal tissue and tumor sites reported in the literature. The majority of normal tissue studies were focused on the lung (24%) and brain (22%) with a small number of studies reported in the GU, GI, oesophagus and heart (Fig. 3A).

In tumor studies, 92% of the studies focused on a single tumor site whilst the remaining 8% investigated responses in multiple tumor sites. Tumor model studies were classified by tumor type, yet these were not always at the origin organ site (i.e. orthotopically) and were frequently subcutaneous flank models. Brain tumors were most commonly explored and accounted for 25% of the published studies (Fig. 3B). This was followed by tumor studies in the pancreas, liver, GU and oesophagus that accounted for 20% of the combined studies. Only 8% investigated responses in more than one tumor model (Fig. 3B) that commonly assessed drug + RT combinations.

From a total of 221 biology papers, we identified the 6 main research areas as evaluation of novel radiosensitizers (33%), model development (31%), imaging (21%), novel delivery modality (11%), evaluation radioprotectors (2%) and assessment of fiducial markers (2%) (Fig. 3C).

Articles published on radiosensitizers include all drug + RT combinations with 93% of these assessed using tumor studies (Fig. 3C). Studies involving radiosensitizers are classified as immunotherapies (54%), nanoparticles (19%), chemotherapy (15%) or other molecular targeted (12%) agents. The distribution of articles in these areas is shown in Fig. 3D. Over half of research in therapeutic agents from 2013 to 2021 has focused on targeting immune modulators or immunotherapy agents (54%) (Fig. 3D).

Studies focusing on model development included the use of orthotopic, GEMMs, tissue targeting and dose schedules. The distribution of these articles was relatively equal across tumor (16%) and normal tissue (14%) studies (Fig. 3C). Also, preclinical models have trialed different imaging methods (21%) and delivery modalities (11%) to achieve more complex delivery schedules. Studies focused on radioprotectors, and fiducial markers were by far the least reported only accounting for 4% of biology contributions.

Experimental tumor models within biology contributions

The distribution of different tumor sites and models is shown in Fig. 4. Due to their simplicity and adaptability allograft and xenograft cell-line derived tumor models account for the largest percentage of tumor studies (82%) and are reported across all tumor types (Fig. 4B & C). These included the implantation of murine tumor cells into syngeneic immunocompetent mice or human tumor cells into immunocompromised mice. In contrast, GEMM accounted for 8% of tumor models mainly involving lung and abdominal tumors. PDXs were most frequently used in brain and abdominal tumor studies. Environmentally induced models accounted for < 1% of the studies (Fig. 4B & C).

Preclinical RT set-up and dose scheduling

Irradiation field sizes were recorded for studies targeting orthoptic tumors and shown in Fig. 5A. Subcutaneous tumor models were not included in the analysis as these often require larger field sizes for targeting and have no proximal surrounding organs at risk. The measured radiochromic film (RCF) output for circular collimators is shown in Fig. 5B. 18% of articles reviewed used collimators < 5 mm and are therefore at risk of underdosing if no output correction was performed (Fig. 5A). 12% used the less severely impacted 3 mm collimator whereas the other 6% are at risk of severe dose overestimation, which could have great impact on the study reliability and reproducibility. No studies used the 0.5 mm collimator in orthotopic tumor models potentially reflecting the high level of uncertainty in dosimetry associated with fields of this size.

Within the tumor biology contributions, only 36% of studies reported fractionated treatment schedules. The number of studies using fractionated protocols is shown in Fig. 6. In 2020, an increased number

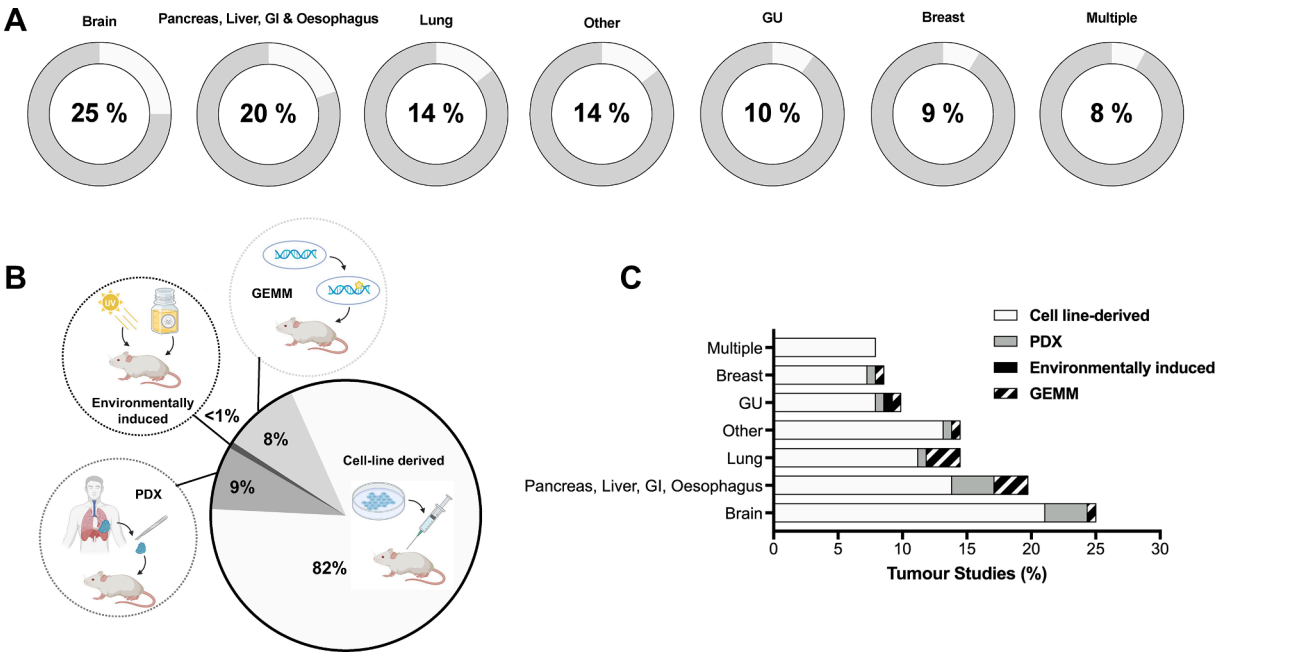

Fig. 4. Further analysis of tumor radiobiology papers based on the main tumor sites and experimental models reported in studies from 2007 to 2021. Panel A: Percentage of different tumor types in all analysed tumor studies. From a total of 152 papers, we identified the main tumor sites reported in the literature in the brain, pancreas, liver, GI & oesophagus and lung. Panel B: Schematic representing the percentage of tumor experimental models reported in biology contributions using small animal irradiators. 4 main approaches of cell-line derived (82.2%), and genetical engineered mouse models (GEMM) (7.9%), patient-derived xenograft (PDX) (9.2%) and environmentally induced models (0.7%) were reported. Panel C: The relative distribution of experimental tumor models reported for each tumour type.

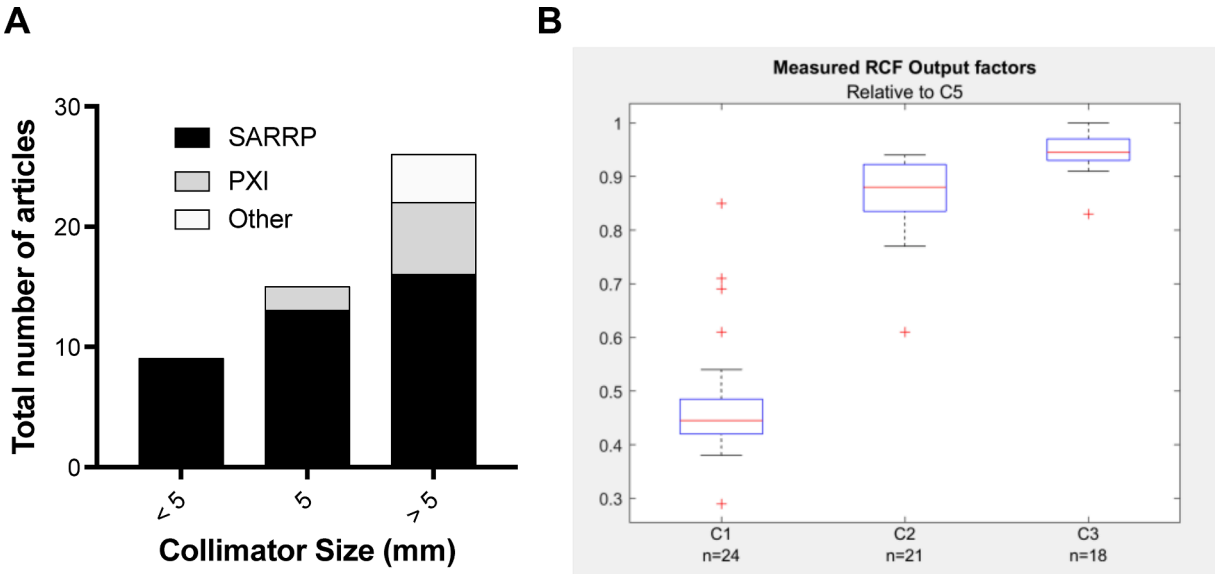

Fig. 5. Overview of treatment delivery methods reported in biology studies. Panel A: Collimator sizes reported for targeting orthotopic tumor studies (n = 50) on the SARRP, XRAD-SmART or other small animal irradiator platforms. From a total of 152 tumor studies, 75 were conducted in orthotopic models (49%) of which 25 studies did not report the treatment field sizes used or the details could not be accessed. 82% of orthotopic tumor models were treated using collimators ≥ 5 mm. Panel B: The measured radiochromic film (RCF) output for circular collimators of 1 mm (C1), 2 mm (C2) and 3 mm (C3) diameter. Results shown are in comparison to a 5 mm diameter field using a Precision X-ray Inc platform. Collimators with a size of<5 mm are at risk of dose overestimation (therefore, of underdosing) which could impact study performance.

of studies used fractionated deliveries accounting for over half (59%) of the tumor studies in that year. Around half (49%) of these tumor studies reported treatment delivery in 3 fractions of doses between 2 and 20 Gy. Only 4 tumor studies (7%) reported the use of ≥ 10 fractions delivered in 2 Gy doses. Only 12% of normal tissue set-ups reported the use of fractionation (Fig. 6). In these studies, RT was delivered in either 2, 3 or 5 fractions of doses ranging from 2 to 10 Gy. However, a more recent study from Lee *et al* used a clinically relevant protocol of 50 Gy in 2 Gy fractions to characterize cardiovascular injury [41].

Discussion

Preclinical models are critical tools in RT research that aim to provide important data to support translation to early phase clinical trials.

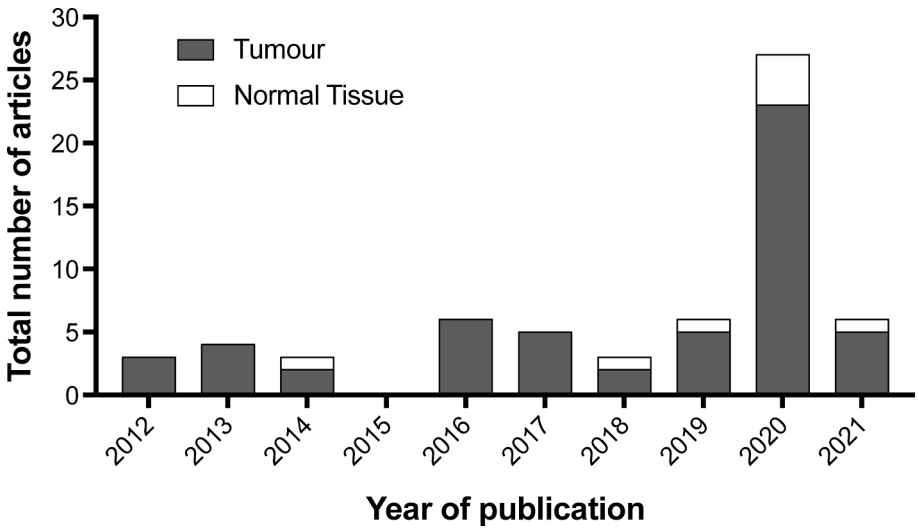

Fig. 6. Tumor and normal tissue studies reporting the use of fractionation. The total number of biology studies per year which reported the use of a fractionated dose delivery for both tumor and normal tissue studies. Fractionation was first implemented in preclinical RT tumour studies in 2012.

Small animal irradiators have provided a more clinically relevant approach to improve the translational power of preclinical models of RT response. Based on a review of all identified papers from 2007 – 2021, we assessed the main contributions of small animal irradiators to provide a broad overview of the current preclinical RT landscape.

Our data show a growth in the number of articles using small animal irradiators. From 2007 to 2010, these articles were predominantly in areas of physics research reflecting the need to establish robust dosimetry, imaging and QA. The number of biology contributions then began to increase from 2011 onwards (Fig. 1B). These changes account for the increased implementation and application of the systems during this time. This also coincides with the report from the "Lesson Learned from Radiation Oncology Trials" workshop that clearly emphasized the need for improvements in preclinical studies relating to study design, validation of models and quality assurance of treatment deliveries [10], this may have contributed to the exponential growth in biology contributions from 2011 to 2020.

Physics contributions focused on development and quality assurance of these platforms by assessing dosimetry, treatment planning and imaging and were mainly conducted using phantoms. Relatively simple phantom models have been replaced with multi-tissue density phantoms aiming to improve dose measurements for various irradiation geometries [11–12]. Dosimetry and QA are critical aspects of preclinical RT studies that are central to the poor reproducibility of studies [43]. Also, we showed orthotopic tumor studies, which reported collimator sizes, 18% used small field sizes < 5 mm (Fig. 5A). When collimating beams to smaller than approximately 5 mm diameter at isocenter, the focal spot is occluded through beam collimation [13]. As the focal spot is highly heterogeneous, this occlusion of the focal spot is challenging to simulate and should preferably be measured. If this effect has not been corrected in studies using < 5 mm collimators it may cause errors in dose calculations in both target and normal tissues that may impact on reproducibility [13].

The SARRP and PRECISION series platforms used dedicated TPSs based on the Superposition Convolution Algorithm [14] or Monte Carlo simulations [15]. A high percentage of articles focused on developing treatment planning approaches to improve tissue segmentation and dose calculations related to scattering at small field sizes and differences in the energy absorption of soft tissues for kilovoltage (kV) beams [16]. The problem arising when scaling down from megavoltage (MV) systems to kV photon energy range is the increasing importance of the photoelectric effect [17–18]. This problem is still of great interest to the community leading to a significant contribution to the literature in the field of treatment planning and simulations (Fig. 2A) [4].

Optimization of RT is based on the competing probabilities of tumor control and normal tissue complication and all biology contributions were within these areas. A small number of biology studies (2%) focused on radioprotection yet the potential of these agents in the clinic is still limited. Fractionated protocols were used in only 12% of normal tissue studies. Yet the majority of these fractionated schedules are far from clinical standards with total doses ranging from 10 to 30 Gy delivered in either 2, 3 or 5 fractions. Similar results were identified for tumor studies with the majority of fractionated studies (93%) using 2 – 6 fractions and 49% reporting the use of 3 fractions. These data indicate a need to improve normal tissue and tumor radiobiology studies using more clinically relevant RT schedules.

The majority of biology contributions focused on tumor studies (69%). However, replicating tumor models within preclinical studies is more complex than assessing normal tissue as there are countless tumor subtypes, grading and experimental models available [19]. Orthotopic tumor models have a more 'natural' anatomical structure and accurate model of tumor behavior and response to RT. These models enable interaction of the surrounding tumor microenvironment which is hugely beneficial for studying treatment response for clinical translation [20]. Small animal irradiators have provided the most accurate model to date for targeting of these orthotopic tumors accounting for just over a third (36%) of tumor models used. We expect more studies will adapt an orthotopic approach due to the advantages of these models and improvements small animal irradiators provide in target alignment.

Imaging is a common area of research across physics and biology contributions. Small animal irradiators are fitted with micro-CT or CBCT imaging platforms at a lower energy than used clinically [21]. Imaging focused studies within physics contributions (22%) mainly aim to improve the spatial resolution whilst maintaining a low imaging dose. This comes with a trade-off between the voxel size and image quality. Lower imaging energies can lead to noise and artefacts which can hinder clear visualization of small animal anatomical structures [22]. Whereas biology imaging studies (21%) assessed different imaging techniques ranging from dual energy CT, bioluminescence-guided, hypoxia-guided and contrast enhanced imaging [23–27]. Some developments have been made with the implementation of iodinated and nanoparticle-based contrast agents which have dual benefits of enhancement of CT image contrast and therapeutic dose deposition [4,28]. Some studies (2%) have also shown the possibility of improved targeting accuracy of low contrast tissues using fiducial markers for treatment alignment [29].

A key measure of the impact of small animal irradiators is the

number of studies that have translated to early phase trials. For example, a study from the group at Queen's University Belfast assessed the impact of AZD6738, an inhibitor of the ataxia telangiectasia related-3(ATRP) kinase in the DNA damage response, on tumor efficacy and late toxicity in the lung [30]. This study directly supported the translation of this approach to evaluate the safety of inhibitors of the DNA damage response in patients with non-small cell lung cancer through the CONDORDE trial (NCT04550104) [31–33]. Similarly, a study from Maastricht University demonstrated the efficacy of nintedanib in reducing radiation-induced lung fibrosis that subsequently led to multiple clinical trials (NCT02452463; NCT02496585) [34–35]. Another example from the University of Pennsylvania assessed the combination of immune checkpoint blockade with RT [36–38] which led to the development of clinical trials in metastatic cancers. Preclinical studies have also successfully translated in other areas including neurological diseases. Research from William Beaumont Hospital that showed the effect of low dose cranial irradiation to significantly reduced β-amyloid plaques in a murine model for Alzheimer's [39] translated to clinical trials at multiple centers around the world (NCT02359864; NCT02359864; NCT03352258). These studies demonstrate how small animal irradiators are enabling translation to the clinic, yet quantitative estimates of these successes are difficult to measure and were not captured in this study. Previously, it has been estimated that only a third of highly cited animal studies have translated to the clinic [42], yet the longer-term success of clinical trials in oncology is significantly lower at only 3.4% [40]. In addition to being unable to accurately measure the translational impact of preclinical RT studies, our data only included articles using commercial and in-house small animal irradiators but did not include articles using non-image guided cabinet or clinical sources.

Conclusions

Many radiobiology research centers have implemented small animal irradiators to refine preclinical irradiation procedures and continue to deliver innovative experimental approaches in physics and biology. Our data identifies the trends in preclinical RT research using small animal irradiators from 2007 to 2021 and highlights the major areas in which these devices have contributed to advance the field. We have identified a number of preclinical RT studies from several laboratories that have supported translation to the clinic yet more work is needed to focus on requirements in preclinical RT studies to enable clinical translation.

Funding statement

KHB is supported by a Training Fellowship from the National Centre for the Replacement Reduction and Refinement of Animal in Research (NC3Rs, NC/V002295/1). MG and KTB are supported by the Medical Research Council (MR/V009605/1).

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgements

The authors wish to acknowledge Kristie Amobie from Xstrahl Inc for providing access to publication records and Randy Jager from SmART Scientific Solutions BV for providing an overview of output factors for PXI machines.

References

[1] Delaney GP, Barton MB. Evidence-based estimates of the demand for radiotherapy. Clin Oncol (R College Radiologists (Great Britain)) 2015;27(2):70–6.
[2] Tillner F, Thute P, Bütof R, Krause M, Enghardt W. Pre-clinical research in small animals using radiotherapy technology–a bidirectional translational approach. Z Med Phys 2014;24(4):335–51.
[3] de Jong M, Essers J, van Weerden WM. Imaging preclinical tumour models: Improving translational power. Nat Rev Cancer 2014;14(7):481–93.
[4] Verhaegen F, Granton P, Tryggestad E. Small animal radiotherapy research platforms. Phys Med Biol 2011;56(12):R55–83.
[5] Ghita M, Brown KH, Kelada OJ, Graves EE, Butterworth KT. Integrating Small Animal Irradiators withFunctional Imaging for Advanced Preclinical Radiotherapy Research. Cancers 2019;11(2):170.
[6] Verhaegen F, van Hoof S, Granton PV, Trani D. A review of treatment planning for precision image-guided photon beam pre-clinical animal radiation studies. Z Med Phys 2014;24(4):323–34.
[7] Koontz BF, Verhaegen F, De Ruysscher D. Tumour and normal tissue radiobiology in mouse models: How close are mice to mini-humans? Br J Radiol 2017;90(1069): 20160441.
[8] Wong J, Armour E, Kazanzides P, Iordachita I, Tryggestad E, Deng H, et al. High-resolution, small animal radiation research platform with x-ray tomographic guidance capabilities. Int J Radiat Oncol Biol Phys 2008;71(5):1591–9.
[9] Butterworth KT, Prise KM, Verhaegen F. Small animal image-guided radiotherapy: status, considerations and potential for translational impact. The British journal of radiology 2015;88:20140634.
[10] Liu F-F, Okunieff P, Bernhard EJ, Stone HB, Yoo S, Coleman CN, et al. Lessons learned from radiation oncology clinical trials. Clin Cancer Res 2013;19(22): 6089–100.
[11] Soultanidis G, Subiel A, Renard I, Reinhart AM, Green VL, Oelfke U, et al. Development of an anatomically correct mouse phantom for dosimetry measurement in small animal radiotherapy research. Phys Med Biol 2019;64(12): 12NT02.
[12] Biglin ER, et al. Preclinical dosimetry: exploring the use of small animal phantoms. Radiat Oncol (London England) 2019;14:134.
[13] Granton PV, Verhaegen F. On the use of an analytic source model for dose calculations in precision image-guided small animal radiotherapy. Phys Med Biol 2013;58(10):3377–95.
[14] Jacques R, Wong J, Taylor R, McNutt T. Real-time dose computation: GPU-accelerated source modeling and superposition/convolution. Med Phys 2011;38 (1):294–305.
[15] van Hoof SJ, Granton PV, Verhaegen F. Development and validation of a treatment planning system for small animal radiotherapy: SmART-Plan. Radiother Oncol 2013;109(3):361–6.
[16] Verhaegen F, Dubois L, Gianolini S, Hill MA, Karger CP, Lauber K, et al. ESTRO ACROP: Technology for precision small animal radiotherapy research: Optimal use and challenges. Radiother Oncol 2018;126(3):471–8.
[17] Paget V, et al. Multiparametric radiobiological assays show that variation of X-ray energy strongly impacts relative biological effectiveness: comparison between 220 kV and 4 MV. Sci Rep 2019;9:1–10.
[18] Vaniqui A, Walters BR, Fonseca GP, Verhaegen F. Dose to water versus dose to medium from cavity theory applied to small animal irradiation with kilovolt x-rays. Phys Med Biol 2019;64(16):165001.
[19] Butterworth KT. Evolution of the Supermodel: Progress in Modelling Radiotherapy Response in Mice. Clin Oncol (R College Radiol (Great Britain)) 2019;31(5): 272–82.
[20] Gengenbacher N, Singhal M, Augustin HG. Preclinical mouse solid tumour models: status quo, challenges and perspectives. Nat Rev Cancer 2017;17:751–65.
[21] Clark DP, Badea CT. Advances in micro-CT imaging of small animals. Phys Med 2021;88:175–92.
[22] Tapiovaara MJ, Wagner RF. SNR and noise measurements for medical imaging: I. A practical approach based on statistical decision theory. Phys Med Biol 1993;38(1): 71–92.
[23] Corroyer-Dulmont A, Valable S, Fantin J, Chatre L, Toutain J, Teulier S, et al. Multimodal evaluation of hypoxia in brain metastases of lung cancer and interest of hypoxia image-guided radiotherapy. Sci Rep 2021;11(1).
[24] Tuli R, Surmak A, Reyes J, Hacker-Prietz A, Armour M, Leubner A, et al. Development of a novel preclinical pancreatic cancer research model: bioluminescence image-guided focal irradiation and tumor monitoring of orthotopic xenografts. Transl Oncol 2012;5(2):77–84.
[25] Petronek MS, et al. Assessment of Gadobutrol Safety in Combination with Ionizing Radiation Using a Preclinical MRI-Guided Radiotherapy Model. Radiat Res 2021; 195:230–4.
[26] Vaniqui A, Schyns LEJR, Almeida IP, van der Heyden B, van Hoof SJ, Verhaegen F. The impact of dual energy CT imaging on dose calculations for pre-clinical studies. Radiat Oncol (London) 2017;12(1).
[27] Schyns LEJR, Almeida IP, van Hoof SJ, Descamps B, Vanhove C, Landry G, et al. Optimizing dual energy cone beam CT protocols for preclinical imaging and radiation research. Br J Radiol 2017;90(1069):20160480.
[28] Porret E, le Guével X, Coll J-L. Gold nanoclusters for biomedical applications: toward in vivo studies. J Mater Chem B 2020;8:2216–32.
[29] Brown KH, Ghita M, Schettino G, Prise KM, Butterworth KT. Evaluation of a novel liquid fiducial marker, Bioxmark®, for small animal image-guided radiotherapy applications. Cancers 2020;12(5):1276.
[30] Dunne V, Ghita M, Small DM, Coffey CBM, Weldon S, Taggart CC, et al. Inhibition of ataxia telangiectasia related-3 (ATR) improves therapeutic index in preclinical models of non-small cell lung cancer (NSCLC) radiotherapy. Radiother Oncol : J Eur Soc Ther Radiol Oncol 2017;124(3):475–81.

[31] Chalmers AJ, Chan C, Sebag-Montefiore D, Sebag-Montefiore D, Evans M, Chalmers A, et al. CTRad 10 Years On: From 10-point Plan to Top 10 Achievements. Clinical Oncology 2020;32(1):9–12.
[32] Sharma RA, Plummer R, Stock JK, Greenhalgh TA, Ataman O, Kelly S, et al. Clinical development of new drug-radiotherapy combinations. Nat Rev Clin Oncol 2016;13 (10):627–42.
[33] Walls GM, Oughton JB, Chalmers AJ, Brown S, Collinson F, Forster MD, et al. CONCORDE: A phase I platform study of novel agents in combination with conventional radiotherapy in non-small-cell lung cancer. Clin Transl Radiat Oncol 2020;25:61–6.
[34] De Ruysscher D, Granton PV, Lieuwes NG, van Hoof S, Wollin L, Weynand B, et al. Nintedanib reduces radiation-induced microscopic lung fibrosis but this cannot be monitored by CT imaging: A preclinical study with a high precision image-guided irradiator. Radiother Oncol 2017;124(3):482–7.
[35] Dy GK, Prasad D, Kumar P, Attwood K, Adjei AA. A Phase 2 Randomized, Double-Blind, Placebo-Controlled Study Evaluating Nintedanib Versus Placebo as Prophylaxis Against Radiation Pneumonitis in Patients With Unresectable NSCLC Undergoing Chemoradiation Therapy. J Thoracic Oncol : Off Publ Int Assoc Study Lung Cancer 2021;16(3):e19–20.
[36] Twyman-Saint Victor C, Rech AJ, Maity A, Rengan R, Pauken KE, Stelekati E, et al. Radiation and dual checkpoint blockade activate non-redundant immune mechanisms in cancer. Nature 2015;520(7547):373–7.
[37] Maity A, Mick R, Huang AC, George SM, Farwell MD, Lukens JN, et al. A phase I trial of pembrolizumab with hypofractionated radiotherapy in patients with metastatic solid tumours. Br J Cancer 2018;119(10):1200–7.
[38] Maity A, Mick R, Rengan R, Mitchell TC, Amaravadi RK, Schuchter LM, et al. A stratified phase I dose escalation trial of hypofractionated radiotherapy followed by ipilimumab in metastatic melanoma: long-term follow-up and final outcomes. OncoImmunology 2021;10(1).
[39] Marples B, McGee M, Callan S, Bowen SE, Thibodeau BJ, Michael DB, et al. Cranial irradiation significantly reduces beta amyloid plaques in the brain and improves cognition in a murine model of Alzheimer's Disease (AD). Radiother Oncol : J Eur Soc Ther Radiol Oncol 2016;118(1):43–51.
[40] Wong CH, Siah KW, Lo AW. Estimation of clinical trial success rates and related parameters. Biostatistics 2019;20:273–86.
[41] Lee C, Lee J, Daniel A, et al. Characterization of cardiovascular injury in mice following partial-heart irradiation with clinically relevant dose and fractionation. Radiotherapy and Oncology 2021;157:155–62. https://doi.org/10.1016/j.radonc.2021.01.023.
[42] Hackam D, Redelmeier D. Translation of Research Evidence From Animals to Humans. JAMA 2006;296:1727–32.
[43] Desrosiers M, et al. The Importance of Dosimetry Standardization in Radiobiology. Journal of Research of the National Institute of Standards and Technology 2013; 118(403).