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Biodistribution of the boron carriers boronophenylalanine (BPA) and/or decahydrodecaborate (GB-10) for Boron Neutron Capture Therapy (BNCT) in an experimental model of lung metastases

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HIGHLIGHTS

- We performed experimental boron biodistribution studies for lung metastases.
- 3 protocols employing BPA and GB-10 would be therapeutically useful.
- BNCT at RA-3 would be potentially therapeutic for experimental lung metastases.

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ABSTRACT

BNCT was proposed for the treatment of diffuse, non-resectable tumors in the lung. We performed boron biodistribution studies with 5 administration protocols employing the boron carriers BPA and/or GB-10 in an experimental model of disseminated lung metastases in rats. All 5 protocols were non-toxic and showed preferential tumor boron uptake versus lung. Absolute tumor boron concentration values were therapeutically useful (25–76 ppm) for 3 protocols. Dosimetric calculations indicate that BNCT at RA-3 would be potentially therapeutic without exceeding radiotolerance in the lung.

1. Introduction

Metastatic lung disease is still a leading cause of death. Surgery, radio and chemotherapy have failed to improve survival satisfactorily and the overall prognosis for these patients is poor. Within this context, the search for more selective and less toxic treatment strategies is warranted, particularly in view of the marked radiosensitivity of the healthy lung (Bakeine et al., 2009). Boron Neutron Capture Therapy (BNCT) has been proposed for the treatment of diffuse, non-resectable tumors in the lung. BNCT is a binary treatment modality that combines irradiation with a thermal or epithermal neutron beam with tumor-seeking, boron containing drugs to produce selective irradiation of tumor tissue. The high linear energy transfer (LET) alpha particles and recoiling ^{7}Li nuclei emitted after the ^{10}B(n,α)^{7}Li reaction have a range of 5–9 μm in tissue and are known to have a high relative biological effectiveness (Coderre and Morris, 1999). In this way, BNCT would potentially target the tumor selectively, minimizing damage to normal tissue. The radiation doses delivered to tumor and normal tissues during BNCT are due to energy deposition from directly ionizing radiation with different LET characteristics. In addition to the alpha and ^{7}Li high-LET products that give rise to the tumor-specific boron dose component, a non-specific background dose results from: (1) low LET gamma rays in the beam, (2) low LET gamma rays resulting from the capture of thermal neutrons by hydrogen atoms [^{1}H(n,γ)^{2}H], (3) high LET protons produced by the scattering of fast neutrons and (4) high LET protons resulting from

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the capture of thermal neutrons by nitrogen atoms [$^{14}N(n,p)^{14}C$]. The biologically effective dose will depend, in each case, on relative biological effectiveness (RBE) and compound biological effectiveness (CBE) factors (Coderre and Morris, 1999) for the different dose components in each tissue and for each end-point considered. BNCT protocols are ideally designed to maximize the boron radiation dose and to minimize the background dose (e.g. Coderre and Morris, 1999; Kreimann et al., 2001a; Trivillin et al., 2006; Molinari et al., 2011; Molinari et al., 2012). Furthermore, being a technique that is based on biological targeting rather than geometric targeting, it would be ideally suited to treat undetectable micrometastases, a major challenge in oncological therapy (e.g. Cardoso et al., 2007).

The effects of BNCT mediated by BPA on the normal lung of Fischer 344 rats have been assessed to establish relative biological effectiveness (RBE) and compound biological effectiveness (CBE) factors (Kiger et al., 2004). In addition, functional and histological changes in the normal lung of Fischer 344 rats after BNCT mediated by BPA were assessed (Kiger et al., 2008). BPA biodistribution studies were performed in an experimental model of lung metastases of colon carcinoma DHD/K12/TRb cells in syngeneic BDIX rats using neutron autoradiography and alpha spectrometry to assess boron concentration and boron microdistribution respectively (Altieri et al., 2006; Bortolussi et al., 2011). In the same model, Bakeine et al. (2009) established the feasibility of using BNCT mediated by BPA in an in vitro/in vivo system. Suzuki et al. (2007) studied the effect of BNCT mediated by BPA in an experimental model of ectopic tumors, implanted in the thoracic cavity of mice to mimic pleural mesothelioma. Suzuki et al. (2008) went on to treat 2 patients with diffuse or multiple pleural tumors. The same group then treated a patient with recurrent lung cancer in the previously irradiated chest wall with two fractions of BNCT (Suzuki et al., 2012). All of these studies have afforded encouraging results but undoubtedly leave room for improvement.

It is well known that one of the most efficient ways to optimize BNCT is to improve the delivery of boron carriers in terms of tumor absolute boron content, selective tumor uptake and tumor targeting homogeneity. Within this context, the aim of the present study was to tailor the coloncarcinoma diffuse lung metastases model in BDIX rats for biodistribution studies and perform boron biodistribution studies with different administration protocols employing the boron compounds BPA and GB-10, administered alone or in combination. Based on these biodistribution data and dosimetric considerations, the feasibility of BNCT for lung metastases in this model at the RA-3 Nuclear Reactor (Ezeiza Atomic Center, National Atomic Energy Commission) was assessed.

2. Materials and methods

2.1. Animal model

Based on the work of Bortolussi et al. (2011), we adapted the model of disseminated lung metastases in BDIX rats. Different experimental conditions, i.e. tumor cell load and time post-administration, were assayed to optimize the model for biodistribution studies. We sought to maximize tumor yield for boron measurements while preserving enough healthy lung for evaluation. DHD/K12/TRb colon-carcinoma cells were maintained in vitro in F10-DMEM culture medium supplemented with 10% fetal calf serum. $1–6 \times 10^6$ DHD/K12/TRb colon-carcinoma cells in 0.5 ml F10-DMEM culture medium were injected in the jugular vein of syngeneic BDIX rats under ketamine (140 mg/kg bw)–xylazine (21 mg/kg bw) anesthesia, using a 27 gauge needle. The selected cell load of 3×10^6 guaranteed acceptably reproducible, controlled tumor cell growth. At different times post-injection of the tumor cells (2, 3, 4 and 5 weeks) the animals were killed and their lungs were removed and fixed in Bouin's solution. Macroscopically visible metastases were counted under a dissection microscope. Three to five weeks was the time post-injection that guaranteed reproducible development of abundant vascularized lung metastases while preserving enough healthy lung for evaluation. National and institutional guidelines for the use and care of laboratory animals were followed throughout.

2.2. Biodistribution studies

Subsequent biodistribution studies were performed employing the established conditions to prepare the animals. Five boron compound administration protocols were assessed in 3–5 rats per group, 4 weeks post-injection of 3×10^6 tumor cells: 1. BPA, 18 mg ^{10}B/kg, intravenous (iv); 2. BPA, 46.5 mg ^{10}B/kg, intraperitoneal (ip); 3. GB-10, 50 mg ^{10}B/kg, iv; 4. BPA, 46.5 mg ^{10}B/kg, ip+GB-10, 20 mg ^{10}B/kg, iv; 5. BPA, 31 mg ^{10}B/kg, ip+GB-10, 34.5 mg ^{10}B/kg, iv. Three hours post-administration of the boron compounds, samples of blood were taken from the surgically exposed jugular vein under ketamine (140 mg/kg bw)—xylazine (21 mg/kg bw) anesthesia. The animals were then sacrificed by overdose of anesthesia immediately prior to tissue sampling. Samples of the target tissue, i.e. lung metastases, samples of organs/tissues that would lie within the treatment volume, i.e. lung, skin, xiphoid cartilage, spinal cord marrow, thymus, heart, diaphragm, and intercostal muscle and reference organs/tissues, i.e. kidney, spleen, intestine, stomach, and liver were taken. The samples were processed as previously described (e.g., Molinari et al., 2011) for boron measurement by ICP-OES. Particular attention was given to normal tissues that would lay within the treatment volume in the case of in-situ lung BNCT, i.e. normal lung, spinal cord marrow, skin, heart, thymus, diaphragm, and intercostal muscle. Based on previous BNCT radiobiological studies in the hamster cheek pouch oral cancer model with other boron compounds (e.g., Kreimann et al., 2001b; Trivillin et al., 2006, Molinari et al., 2011), we previously defined the following guidelines to establish the potential therapeutic value of the boron carriers, the administration protocols and time-points post administration (Garabalino et al., 2011):

- No manifest toxicity.
- Absolute boron concentration in tumor $\geq$ 20 ppm.
- Boron concentration ratio tumor/normal tissue $\geq$ 1.
- Boron concentration ratio tumor/blood $\geq$ 1.

Although the actual usefulness of a particular boron carrier and protocol can only be determined by in vivo radiobiological BNCT studies, our previous studies in the hamster cheek pouch oral cancer model with other boron compounds suggest that the protocols that meet the above requirements are potentially therapeutic and warrant radiobiological assessment.

Within the context of contributing to the assessment of ex-situ BNCT for lung metastases, and based on the fact that autograft techniques involve organ perfusion with a preservation solution, we performed a complementary biodistribution study to assess the potential effect of perfusion on boron concentration in metastases and the lung tissue. We proceeded as described above, using administration protocol 2. Three hours post-administration of BPA, 1000 units of heparin (Veinfar, Argentina) were injected ip in each animal. The animals were sacrificed 5 min later by overdose of anesthesia and the lungs were perfused with Ringer´s solution (Roux-OCEFA, Argentina). Samples of perfused lung and metastases were taken and processed for boron measurement by ICP-OES.

3. Results

No toxic effects were observed for any of the boron compound administration protocols. Table 1 shows the absolute boron concentration [B] values and the metastases/lung and metastases/blood boron concentration ratios for each of the protocols. All 5 protocols exhibited preferential boron uptake by tumor tissue versus the healthy lung ([B] ratio metastases/normal lung: 1.2–1.9). [B] values in tumor for protocols **2** (BPA, 46.5 mg ^{10}B/kg, ip), **4** (BPA, 46.5 mg ^{10}B/kg, ip+GB-10, 20 mg ^{10}B/kg, iv) and **5** (BPA, 31 mg ^{10}B/kg, ip+GB-10, 34.5 mg ^{10}B/kg, iv) ranged between 23 and 76 ppm. Tumor boron values for protocols 1 and 3 exhibited sub-optimum values of approximately 13 ppm. The blood boron values corresponding to the protocols that included GB-10 were equal to, or higher than, tumor values. Boron concentration in healthy tissues that would lay in the treatment volume in the case of in-situ lung BNCT (normal lung, spinal cord marrow, skin, heart, thymus, diaphragm, and intercostal muscle) were overall equal to, or lower than, tumor values.

Protocols **2** (BPA, 46.5 mg ^{10}B/kg, ip), **4** (BPA, 46.5 mg ^{10}B/kg, ip+GB-10, 20 mg ^{10}B/kg, iv) and **5** (BPA, 31 mg ^{10}B/kg, ip+GB-10, 34.5 mg ^{10}B/kg, iv) warrant radiobiological evaluation in actual BNCT studies.

Table 2 shows the boron concentration values for samples of perfused lung and metastases, compared to the values for the corresponding samples of non-perfused tissue. Perfusion induced a statistically significant (Student's t test, $p \leq 0.0006$) reduction in boron concentration in metastases and the lung tissue and induced a rise in metastases/lung boron concentration ratio. Perfusion in a small organ involves technical restrictions that might cause the organ to retain excess fluid, artificially enhancing the reduction in boron concentration measured herein. Thus, these results are not conclusive but do suggest that potential changes in boron concentration in perfused organs must be considered when performing dose calculations for ex-situ BNCT.

4. Discussion

The present study explores the potential therapeutic value of different boron compound administration protocols. Three of the five protocols evaluated herein exhibited preferential tumor uptake and absolute tumor boron values within a potentially therapeutic range and warrant radiobiological evaluation in in vivo BNCT studies. Both GB-10 and BPA are approved for use in human subjects by the United States Food and Drug Administration and are thus particularly interesting to assess (e.g., Molinari et al., 2011; Kankaanranta et al., 2012). Despite the fact that BSH has a long history of use in humans (e.g., Ono et al., 1999) and would be of interest to test in the future, our vast experience with GB-10 in the hamster cheek pouch model, its proven therapeutic efficacy based on a selective effect on aberrant tumor blood vessels rather than selective tumor uptake, its lack of toxicity at high doses and ease to handle (e.g., Trivillin et al., 2006) led us to perform the present experiments with GB-10.

It has been postulated (e.g., Ono et al., 1999; Trivillin et al., 2006) that the combined administration of different boron compounds with different properties and complementary uptake mechanisms may enhance the therapeutic advantage of BNCT. We previously demonstrated that the combined administration of BPA and GB-10 in the hamster cheek pouch oral cancer model contributes to homogeneous boron targeting in heterogeneous tumors (Heber et al., 2006). This is undoubtedly an asset in BNCT because it minimizes the occurrence of tumor cell populations that are refractory to treatment. Within this context, the protocols that involve the combined administration of BPA and GB-10 would confer a potential advantage. The radiobiological role of the high blood boron values in the administration protocols that include GB-10 can only be assessed in actual BNCT studies. They may pose a risk in terms of vascular damage, but they could also confer an advantage in terms of the previously described selective damage to aberrant tumor blood vessels induced by GB-10-BNCT in the hamster cheek pouch oral cancer model (Trivillin et al., 2006). In addition, the relative biological effectiveness (RBE) and compound

Table 2
Boron concentration (mean ± standard deviation) (ppm) in blood and tissue samples.

Tissues	BPA (46.5 mg ^{10}B/Kg) ip	BPA (46.5 mg ^{10}B/Kg) iv Local Perfusion
Blood	13.7 ± 2.3 (n=5)	11.7 ± 1.9 (n=3)
Metastases	22.9 ± 7.2 (n=48)*	11.2 ± 2.3 (n=23)*
Lung	12.2 ± 7.2 (n=9)*	3.7 ± 3.1 (n=15)*
Metastases/lung	1.9	3.0

n: number of samples.
* Student´s t test, $p \leq 0.0006$.

Table1
Boron concentration (mean ± standard deviation) (ppm) in blood and tissue samples for the different administration protocols.

Tissues	BPA (18 mg ^{10}B/Kg) iv	BPA (46.5 mg ^{10}B /Kg) ip	GB-10 (50 mg ^{10}B/Kg) iv	BPA (46.5 mg ^{10}B/Kg) ip+GB-10 (20 mg ^{10}B/Kg) iv	BPA (31 mg ^{10}B/Kg) ip+GB-10 (34.5 mg ^{10}B/Kg) iv
Blood	6.1 ± 2.4 (n=5)	13.7 ± 2.3 (n=5)	26.8 ± 14.2 (n=5)	77.6 ± 14.7 (n=4)	31.9 ± 5.9 (n=4)
Metastases	12.0 ± 2.3 (n=17)	22.9 ± 7.2 (n=48)	12.8 ± 4.1 (n=5)	75.7 ± 11.4 (n=18)	32.8 ± 8.7 (n=32)
Lung	8.1 ± 3.6 (n=5)	12.2 ± 7.2 (n=9)	10.3 ± 2.4 (n=5)	63.8 ± 8.1 (n=4)	28.3 ± 5.6 (n=6)
Skin	5.9 ± 1.7 (n=5)	18.5 ± 7.9 (n=4)	16.1 ± 8.7 (n=5)	45.0 ± 11.2 (n=4)	17.4 ± 3.9 (n=3)
Costal muscle	10.8 ± 2.3 (n=5)	25.0 ± 2.6 (n=4)		35.5 ± 5.8 (n=4)	19.5 ± 1.1 (n=3)
Diaphragm	9.5 ± 2.0 (n=5)	25.9 ± 7.7 (n=4)	4.8 ± 2.2 (n=4)	47.2 ± 7.6 (n=4)	34.2 ± 5.3 (n=3)
Xiphoid	6.8 ± 2.2 (n=5)		9.8 ± 3.7 (n=5)	40.0 ± 11.1 (n=4)	22.0 (n=1)
Heart	6.8 ± 2.6 (n=5)	14.9 ± 3.7 (n=4)	7.2 ± 1.4 (n=5)	36.4 ± 8.9 (n=4)	18.9 ± 3.0 (n=3)
Thymus	7.5 ± 1.7 (n=5)	19.2 ± 4.7 (n=4)	4.9 ± 1.7 (n=4)	24.5 ± 3.9 (n=4)	20.7 ± 6.0 (n=3)
Spinal cord marrow	4.3 ± 1.8 (n=5)	5.5 ± 2.4 (n=8)	1.6 ± 0.8 (n=5)	14.7 ± 4.2 (n=3)	4.7 ± 2.0 (n=5)
Liver	7.5 ± 3.6 (n=5)	15.1 ± 6.5 (n=10)	7.2 ± 2.8 (n=5)	52.0 ± 11.0 (n=4)	21.8 ± 4.5 (n=3)
Kidney	21.2 ± 6.3 (n=5)	69.0 ± 32.1 (n=10)	27.3 ± 7.6 (n=5)	158.6 ± 32.3 (n=4)	126.5 ± 25.0 (n=6)
Spleen	6.0 ± 2.2 (n=4)	20.1 ± 8.6 (n=10)	8.7 ± 1.8 (n=5)	53.5 ± 16.8 (n=4)	31.6 ± 9.9 (n=6)
Intestine	7.5 ± 2.3 (n=5)	15.1 ± 8.2 (n=10)	5.0 ± 2.3 (n=5)	39.1 ± 10.6 (n=4)	16.4 ± 7.4 (n=3)
Stomach	9.6 ± 3.7 (n=5)	18.7 ± 11.0 (n=10)	5.6 ± 0.9 (n=5)	43.9 ± 13.3 (n=4)	20.7 ± 4.8 (n=3)
Metastases/lung	1.5	1.9	1.2	1.2	1.2
Metastases/blood	2.0	1.7	0.5	1.0	1.0

n: number of samples.

biological effectiveness (CBE) factors for each Reactor/boron compound administration protocol/tissue/end-point will condition outcome and must be assessed in appropriately designed experiments (Hopewell et al., 2011).

To date only BPA has been explored as a boron carrier for BNCT of diffuse lung metastases. Within the characteristic spread in boron concentration values, the values reported herein for protocol 1 (BPA at 18 mg B/kg iv) fall within the range of values previously reported for BPA assuming a linear correction for differences in administered boron dose between studies (Suzuki et al., 2007; Bortolussi et al., 2011). Thus, the present study explores, for the first time, the therapeutic potential of GB-10 and GB-10+BPA as the boron carriers for future studies of BNCT in an experimental model of diffuse lung metastases and reports the potential therapeutic value of 3 protocols, i.e. Protocols **2** (BPA, 46.5 mg ^{10}B/kg, ip), **4** (BPA, 46.5 mg ^{10}B/kg, ip+GB-10, 20 mg ^{10}B/kg, iv) and **5** (BPA, 31 mg ^{10}B/kg, ip+GB-10, 34.5 mg ^{10}B/kg, iv). Complementary boron microdistribution studies by neutron autoradiography would be necessary to assess boron targeting homogeneity and localization and establish the potential correlation between boron content and tumor invasion in lung samples (e.g., Bortolussi et al., 2011).

Given that the current state-of-the-art treatment strategies have failed to improve survival as hoped for, BNCT emerges as a treatment modality worthy of evaluation. In conventional external radiotherapy, it is difficult to deliver curative doses to malignant cells without causing radiation pneumonitis in the healthy lung (Suzuki et al., 2007). BNCT can offer a dose gradient between tumor and normal cells if ^{10}B atoms accumulate preferentially in tumor cells. Furthermore, the fact that BNCT is based on biological rather than geometric targeting makes it ideally suited to treat undetectable micrometastases and infiltrating tumor cells (e.g., Bortolussi et al., 2011; Pozzi et al., 2012). In addition, with BNCT it is unnecessary to adjust for breathing motions (Bortolussi et al., 2011).

When dealing with a large cell load of metastatic spread coupled to significant tumor control following BNCT, an issue that must not be overlooked in terms of toxicity is the acute inflammatory reactions potentially induced in lung by the debris of dead cells and associated breakdown products. Within this context, radiation pneumonitis might be more severe than anticipated based on the dose delivered (Bakeine et al., 2009). This phenomenon was observed in liver metastases patients treated with ex-situ BNCT (Zonta et al., 2006) and must be considered within the complexities of dose prescription in BNCT. However, this issue has not been addressed in due depth to date.

Based on the biodistribution data that proved potentially useful, we assessed the feasibility of actual treatments using the same experimental model at the thermal nuclear facility at RA-3 (Miller et al., 2009). For irradiation in the thermal column at RA-3 it was necessary to design, build and characterize an adequate shielding device built from lithium-6 carbonate to protect the body of the animal while exposing the lung area through a collimated aperture (Razetti et al., 2012). Dosimetric calculations were based on biodistribution data for protocol 2 (BPA at 46.5 mg ^{10}B/kg, ip) taken as an example, and thermal neutron flux data for the shielding device (Bortolussi et al., 2011). The dose constraints imposed on the healthy lung were that the maximum dose should be $<18.6\,Gy_w$, that the mean dose should be $<9.1\,Gy_w$ and that less than 30% of the lung volume should receive 9.1 Gy_w. Dose constraints were also imposed on the most sensitive organs exposed through the collimated aperture, i.e. heart (maximum dose $<10.4\,Gy_w$) and spinal cord (maximum dose $<13\,Gy_w$). The dose constraints adopted for the organs at risk were taken from QUANTEC report for conventional photon therapy (Bentzen et al., 2010) and when necessary, properly translated into single fraction doses using the LQ model and alpha/beta ratios found in the same report.

The weighted doses were calculated employing previously reported RBE and BPA CBE factors, i.e. CBE factor of 1.4 for lung (Kiger et al., 2008), CBE factor of 3.8 for tumor (Coderre et al., 1993), CBE factor of 2.5 for skin (Gonzalez et al., 2004), and an RBE factor of 3 for all tissues (Kiger et al., 2008). Based on these dosimetric considerations and an average irradiation time of 20 min, the lung would receive a mean dose of $6.7 \pm 1.7\,Gy_w$ and a maximum dose of $9.5 \pm 2.6\,Gy_w$ and the Planning Treatment Volume (considering the whole lung as metastases) would receive a mean dose of $18.6 \pm 4.6\,Gy_w$ and a maximum dose $27.7 \pm 7.1\,Gy_w$. Within this context, BNCT at RA-3 for experimental lung metastases would be feasible and potentially therapeutic without exceeding radiotolerance in the healthy lung. Another issue that must be considered is the degree to which the colon is included in the radiation field. Radiation colitis is a potential acute adverse effect that must be cautiously monitored (Suzuki et al., 2012).

Ex-situ BNCT based on extracorporeal irradiation and autotransplantation would be therapeutically advantageous in terms of minimizing adverse effects and optimizing dose distribution (Zonta et al., 2006; Wortmann and Knorr, 2012) and warrants further study. Within this context, the biodistribution data reported herein for a perfused organ suggests a potential reduction in boron concentration and must be considered when performing dose calculations for ex-situ BNCT for diffuse lung metastases.

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References

Altieri, S., Bortolussi, S., Bruschi, P., Fossati, F., Vittor, K., Nano, R., Facoetti, A., Chiari, P., Bakeine, J., Clerici, A., Ferrari, C., Salvucci, O., 2006. Boron absorption imaging in rat lung colon adenocarcinoma metastases. J. Instrum. 41, 123–126.

Bakeine, G.J., Di Salvo, M., Bortolussi, S., Stella, S., Bruschi, P., Bertolotti, A., Nano, R., Clerici, A., Ferrari, C., Zonta, C., Marchetti, A., Altieri, S., 2009. Feasibility study on the utilization of boron neutron capture therapy (BNCT) in a rat model of diffuse lung metastases. Appl. Radiat. Isot. 67, S332–S335.

Bentzen, S.M., Constine, L.S., Deasy, J.O., Eisbruch, A., Jackson, A., Marks, L.B., Ten Haken, R.K., Yorke, E.D., 2010. Quantitative analyses of normal tissue effects in the clinic (QUANTEC): an introduction to the scientific issues. Int. J. Radiat. Oncol. Biol. Phys. 76 (3), S3–S9.

Bortolussi, S., Bakeine, J.G., Ballarini, F., Bruschi, P., Gadan, M.A., Protti, N., Stella, S., Clerici, A., Ferrari, C., Cansolino, L., Zonta, C., Zonta, A., Nano, R., Altieri, S., 2011. Boron uptake measurements in a rat model for boron neutron capture therapy of lung tumours. Appl. Radiat. Isot. 69 (2), 394–398.

Cardoso, J.E., Trivillin, V.A., Heber, E.M., Nigg, D.W., Calzetta, O., Blaumann, H., Longhino, J., Itoiz, M.E., Bumaschny, E., Pozzi, E.C.C., Schwint, A.E., 2007. Effect of boron neutron capture therapy (BNCT) on normal liver regeneration: towards a novel therapy for liver metastases. Int. J. Radiat. Biol. 83 (10), 699–706.

Coderre, J.A., Makar, M.S., Micca, P.L., Nawrocky, M.M., Liu, H.B., Joel, D.D., Slatkin, D. N., Amols, H.I., 1993. Derivations of relative biological effectiveness for the high-let radiations produced during boron neutron capture irradiations of the 9 L rat gliosarcoma in vitro and in vivo. Int. J. Radiat. Oncol. Biol. Phys. 27 (5), 1121–1129.

Coderre, J.A., Morris, G.M., 1999. The radiation biology of boron neutron capture therapy. Radiat. Res. 151, 1–18.

Garabalino, M.A., Monti Hughes, A., Molinari, A.J., Heber, E.M., Pozzi, E.C.C., Cardoso, J.E., Colombo, L.L., Nievas, S., Nigg, D.W., Aromando, R.F., Itoiz, M.E., Trivillin, V. A., Schwint, A.E., 2011. Boron neutron capture therapy (BNCT) for the treatment of liver metastases: biodistribution studies of boron compounds in an experimental model. Radiat. Environ. Biophys. 50, 199–207.

Gonzalez, S.J., Bonomi, M.R., Santa Cruz, G.A., Blaumann, H.R., Calzetta Larrieu, O.A., Menéndez, P., Jiménez Rebagliati, R., Longhino, J., Feld, D.B., Dagrosa, M.A., Argerich, C., Castiglia, S.G., Batistoni, D.A., Liberman, S.J., Roth, B.M., 2004. First

BNCT treatment of a skin melanoma in Argentina: dosimetric analysis and clinical outcome. Appl. Radiat. Isot. 61 (5), 1101–1105.
Heber, E.M., Trivillin, V.A., Nigg, D.W., Itoiz, M.E., Gonzalez, B.N., Rebagliati, R.J., Batistoni, D., Kreimann, E.L., Schwint, A.E., 2006. Homogeneous boron targeting of heterogeneous tumors for boron neutron capture therapy (BNCT): chemical analyses in the hamster cheek pouch oral cancer model. Arch. Oral Biol. 51, 922–929.
Hopewell, J.W., Morris, G.M., Schwint, A.E., Coderre, J.A., 2011. The radiobiological principles of boron neutron capture therapy: a critical review. Appl. Radiat. Isot. 69 (12), 1756–1759.
Kankaanranta, L., Seppälä, T., Koivunoro, H., Saarilahti, K., Atula, T., Collan, J., Salli, E., Kortesniemi, M., Uusi-Simola, J., Välimäki, P., Mäkitie, A., Seppänen, M., Minn, H., Revitzer, H., Kouri, M., Kotiluoto, P., Seren, T., Auterinen, I., Savolainen, S., Joensuu, H., 2012. Boron neutron capture therapy in the treatment of locally recurred head-and-neck cancer: final analysis of a phase I/II trial. Int. J. Radiat. Oncol. Biol. Phys. 82 (1), e67–e75.
Kiger III W.S., Lu, X.Q., Harling, O.K., Riley, K.J., Binns, P.J., Kaplan, J., Patel, H., Zamenhof, R.G., Shibata, Y., Kaplan, I.D., Busse, P.M., Palmer, M.R., 2004. Preliminary treatment planning and dosimetry for a clinical trial of neutron capture therapy using fission converter epithermal neutron beam. Appl. Radiat. Isot. 61, 1075–1081.
Kiger, J.L., Kiger 3rd, W.S., Riley, K.J., Binns, P.J., Patel, H., Hopewell, J.W., Harling, O. K., Busse, P.M., Coderre, J.A., 2008. Functional and histological changes in rat lung after boron neutron capture therapy. Radiat Res. 170 (1), 60–69.
Kreimann, E.L., Itoiz, M.E., Dagrosa, A., Garavaglia, R., Farías, S., Batistoni, D., Schwint, A.E., 2001a. The hamster cheek pouch as a model of oral cancer for boron neutron capture therapy studies: selective delivery of boron by boronophenylalanine. Cancer Res. 61, 8775–8781.
Kreimann, E.L., Itoiz, M.E., Longhino, J., Blaumann, H., Calzetta, O., Schwint, A.E., 2001b. Boron neutron capture therapy for the treatment of oral cancer in the hamster cheek pouch model. Cancer Res. (Advances in Brief) 61 (24), 8638–8642.
Miller, M., Quintana, J., Ojeda, J., Langan, S., Thorp, S., Pozzi, E.C.C., Sztejnberg, M., Estryk, G., Nosal, R., Saire, E., Agrazar, H., Graiño, F., 2009. New irradiation facility for biomedical applications at the RA-3 reactor thermal column. Appl. Radiat. Isot. 67, S226–S229.
Molinari, A.J., Pozzi, E.C., Monti Hughes, A., Heber, E.M., Garabalino, M.A., Thorp, S.I., Miller, M., Itoiz, M.E., Aromando, R.F., Nigg, D.W., Quintana, J., Santa Cruz, G.A., Trivillin, V.A., Schwint, A.E., 2011. ‘‘Sequential’’ boron neutron capture therapy (BNCT): a novel approach to BNCT for the treatment of oral cancer in the hamster cheek pouch model. Radiat. Res. 175, 463–472.
Molinari, A.J., Pozzi, E.C., Monti Hughes, A., Heber, E.M., Garabalino, M.A., Thorp, S.I., Miller, M., Itoiz, M.E., Aromando, R.F., Nigg, D.W., Trivillin, V.A., Schwint, A.E., 2012. Tumor blood vessel "normalization" improves the therapeutic efficacy of boron neutron capture therapy (BNCT) in experimental oral cancer. Radiat. Res. 177 (1), 59–68.
Ono, K., Masunaga, S., Suzuki, M., Kinashi, Y., Takagaki, M., Akaboshi, M., 1999. The combined effect of boronophenylalanine and borocaptate in boron neutron capture therapy for SCCVII tumors in mice. Int. J. Radiat. Oncol. Biol. Phys. 43, 431–436.
Pozzi, E.C.C., Cardoso, J.E., Colombo, L.L., Thorp, S., Monti Hughes, A., Molinari, A.J., Garabalino, M.A., Heber, E.M., Miller, M., Itoiz, M.E., Aromando, R.F., Nigg, D.W., Quintana, J., Trivillin, V.A., Schwint, A.E., 2012. Boron neutron capture therapy (BNCT) for liver metastasis: therapeutic efficacy in an experimental model. Radiat. Environ. Biophys. 51 (3), 331–339.
Razetti, A., Farias, R.O., Trivillin, V.A., Pozzi, E.C.C., Thorp, S., Curotto, P., Schwint, A. E., González, S.J., 2012. Design of a neutron shield for the treatment of diffuse lung cancer in rats using BNCT. Appl. Radiat. Isot.. (this issue).
Suzuki, M., Sakurai, Y., Masunaga, S., Kinashi, Y., Nagata, K., Maruhashi, A., Ono, K., 2007. A preliminary experimental study of boron neutron capture therapy for malignant tumors spreading in thoracic cavity. Jpn. J. Clin. Oncol. 37 (4), 245–249.
Suzuki, M., Endo, K., Satoh, H., Sakurai, Y., Kumada, H., Kimura, H., Masunaga, S., Kinashi, Y., Nagata, K., Maruhashi, A., Ono, K., 2008. A novel concept of treatment of diffuse or multiple pleural tumors by boron neutron capture therapy (BNCT). Radiother. Oncol. 88 (2), 192–195.
Suzuki, M., Suzuki, O., Sakurai, Y., Tanaka, H., Kondo, N., Kinashi, Y., Masunaga, S.-I., Maruhashi, A., Ono, O., 2012. Reirradiation for locally recurrent lung cancer in the chest wall with boron neutron capture therapy (BNCT). Int. Cancer Conf. J. 1 (4), 235–238.
Trivillin, V.A., Heber, E.M., Nigg, D.W., Itoiz, M.E., Calzetta, O., Blaumann, H., Longhino, J., Schwint, A.E., 2006. Therapeutic success of boron neutron capture therapy (BNCT) mediated by a chemically non-selective boron agent in an experimental model of oral cancer: a new paradigm in BNCT radiobiology. Radiat. Res. 166, 387–396.
Wortmann, B., Knorr, J., 2012. Basic requirements and parameter optimization for boron neutron capture therapy of extracorporeal irradiated and autotransplanted organs. Appl. Radiat. Isot. 70 (8), 1709–1717.
Zonta, A., Prati, U., Roveda, L., Ferrari, C., Zonta, S., Clerici, A.M., Zonta, C., Pinelli, T., Fossati, F., Altieri, S., Bortolussi, S., Bruschi, P., Nano, R., Barni, S., Chiari, P., Mazzini, G., 2006. Clinical lessons from the first applications of BNCT on unresectable liver metastases. J. Phys.: Conf. Ser. 41, 484–495.