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Photodynamic Therapy in Spinal Metastases: A Qualitative Analysis of Published Results

Hai-tao Fan, Lei Wang, Ping Zhang, Shu-bin Liu

Department of Orthopedics, The PLA 307 Hospital, Beijing, China

The current study was to perform qualitative comparison of photodynamic therapy (PDT), based on previously published articles on spinal disease distribution status before and after treatment. Spinal metastasis, the migration of primary cancer cells and establishment of secondary tumors in the spine. We electronically searched CENTRAL (The Cochrane Library 2012), MEDLINE, EMBASE, CINAHL and AMED (from their beginning to December 31, 2012) to identify published studies assessing the effectiveness of PDT in spinal metastases. Our inclusion criteria resulted in only 4 articles, all in mice models. Due to study limitations and sparse data, the quality of evidence for all outcomes was low. Our analyses shows that effects on stereological and mechanical properties observed at the 1-week time point post-PDT are maintained at a longer 6-week time point, with combined PDT + bisphosphonate treatment being the most beneficial in terms of bone enhancement. Additionally, the combination of PDT + radiation therapy also demonstrated significant increases in stereological parameters, suggesting that previous radiation therapy treatment does not preclude the bone-enhancing effects of PDT and in fact may be synergistic in the longer term. The bone-enhancing effects of PDT in combination with conventional treatments, and its ability to destroy metastatic human breast cancer cells within bone, present PDT as an attractive novel treatment for spinal metastasis. The positive results from these preclinical studies might motivate future clinical translation of PDT for spinal metastasis.

Key words: Spinal metastases – Photodynamic therapy – Metastatic human breast cancer – Photosensitizer

Spinal metastasis, the migration of primary cancer cells and establishment of secondary tumors in the spine, can occur in 30 to 50% of all cancer patients,[1–3] with the vertebral column being the most frequent site of secondary skeletal tumor formation.[4] Metastasis is associated with advanced

Corresponding author: Haitao Fan, MD, Department of Orthopedics, The PLA 307 Hospital, No. 8 East Street, Fengtai District, Beijing 100071, China.
E-mail: fan_hait@163.com

stages of cancer and prognosis is poor for these patients. Over 80% of patients with advanced breast cancer develop spinal metastasis,[5] and while the 5-year survival rate for localized or regional breast cancer is greater than 80%, it is less than 25% for distant-stage disease.[6] Furthermore, up to 90% of all cancer patients have metastatic lesions in the spine at the time of death,[7] suggestive that spinal metastasis is a common progression of many cancers. Unfortunately, it is believed that while advances in cancer treatment improve the survival of patients, it also provides more opportunity and time for metastases to develop.[8] Thus, the incidence and prevalence of spinal metastasis has potential to increase.

The high incidence of metastasis to the spine is in part due to a fertile microenvironment for tumors to develop. The bone matrix contains many growth factors, as well as bone-resorbing osteoclasts and bone-forming osteoblasts.[9] Spinal metastasis primarily establishes within the vertebral body centrum with secondary involvement of the posterior elements. The vertebral body bears 80% of the mechanical loads sustained by the spine.[10] Metastatic disease, whether osteolytic, osteoblastic, or mixed osteolytic/osteoblastic, can compromise the mechanical integrity of the spine. This can lead to skeletal-related events (SREs), such as pathologic fracture and spinal cord compression, resulting in debilitating neurological dysfunction and pain.[11] Spinal metastasis has considerable consequences for patients in terms of both morbidity and mortality, adversely affecting patient quality of life.[8]

While current treatment strategies are mostly palliative, they also may be aimed at reducing tumor burden and restoring stability in the spinal column.[12] Current treatment for spinal metastasis involves a multimodal approach, including bisphosphonates and radiation therapy.[12] Although a multitude of treatment options are available to patients with spinal metastasis, tumor response still remains variable. Furthermore, the recurrence of spinal metastasis may necessitate additional treatment options as toxicity accumulation may prohibit repeated use of single methods over time. The lack of a treatment or combination of treatments that is able to achieve a comprehensive effect on spinal metastasis provides an impetus to investigate other treatment options.

Photodynamic therapy (PDT) has been shown to be successful in destroying vertebral osteolytic tumors and enhancing vertebral structure, particularly in combination with bisphosphonates.[13] Photodynamic therapy is a minimally invasive technique that utilizes a drug (called a photosensitizer) activated by light of a specific non-thermal wavelength to locally destroy cells. Photodynamic therapy is used in the treatment of lung,[14] esophageal, intraperitoneal,[15] and prostate cancers.[16] It also is approved for the treatment of wet macular degeneration,[17] and has potential application for the treatment of psoriasis[18] and severe acne.[19] In photodynamic therapy for cancer, the photosensitizer is administered into the circulation, where it is preferentially taken up by tumor cells. When the excitation light is introduced, the photosensitizers produce reactive oxygen species (ROS), known as singlet oxygen, which react with essential biomolecules in the tumor cells, eventually causing cell toxicity and death. In addition to depleting oxygen to create singlet oxygen, the activation of photosensitizers in the blood stream can also damage blood vessels that provide nutrients to tumors, effectively starving them. The damage caused by PDT also activates the innate immune system, recruiting it to further attack the tumor cells.

Given the high cost-of-burden of spinal metastases and the public health relevance, it is highly imperative to systematically analyze the actual pre-/clinical potential of PDT in comparison with conventional management strategies for treatment of spinal metastases.

Materials and Methods

Search strategy

We electronically searched CENTRAL, MEDLINE, EMBASE CINAHL and AMED (from their beginning to December 2012). The goal of the search strategy was to identify articles that assessed the effectiveness of photodynamic therapy (PDT) in the treatment of spinal metastases. The search strategy employed an electronic database search and examination of bibliographies of relevant review papers. The following MeSH terms, alone or in various combinations were used: "spine," "metastases," "spinal primary tumor," "surgery," "treatment," "cancer," "photodynamic therapy," and "PDT."

Inclusion criteria, definition, endpoints, and data extraction

All published studies that investigated the effect of PDT alone or in combination with palliative or surgical treatment on spinal metastases—irrespective of blinding, animal model (mice or human), or

language—were identified. Randomized crossover trials were included only if the trial reported a washout period. Patients in the included studies had to be aged 18 to 65 years with duration of symptoms longer than 6 months, regardless of previous treatment, with a pathologically confirmed diagnosis of spinal metastases. Studies in mice models of spinal metastases needed to have confirmed identity of spinal metastases induction. The endpoints considered were spinal function, paralysis, and vertebral structure.

The data were independently extracted and recorded by 2 review authors (H-TF, LW) on a data extraction form. Consensus among the authors was used to make decisions. Intention-to-treat (ITT) analysis was not performed in any of the studies that met our inclusion criteria. For binary outcomes, we recorded the number of participants experiencing the event in each group of the trial. For continuous outcomes, we extracted the arithmetic means and standard deviations for each group. If the data were reported using geometric means, we extracted the standard deviations on the log scale. Medians and ranges were extracted and reported if available. Risk bias was not calculated because of the heterogeneity of the search criteria and ensuing search results.

Assessment of heterogeneity

Descriptive statistics were used to provide a summary description of the groups, interventions, and outcomes. All results reported were based on a relatively small sample size. Potential publication bias was not tested with a funnel plot or other corrective analytical methods as the number of studies included in the review is small.[20] There were insufficient data available to use quantitative analyses to summarize the data. However, we assessed the overall quality of the evidence for each outcome using an adapted GRADE approach.

Results

Search results

We identified and screened 21 citations. After reviewing the titles and the abstracts, 14 reports were excluded as they were not related to spinal metastases and the therapeutic role of PDT. Three appeared to be potentially relevant but were excluded on closer examination because they were review articles. Four studies[21–24] that met the inclusion criteria were extracted for more detailed evaluation. All 4 included studies were conducted in mice models. Not even one study could be found where the effect of PDT in human patients with spinal metastases was evaluated.

Risk of bias in included clinical trials

The 12 criteria recommended by the Cochrane Back Review Group[25] could not be used to assess the risk of bias as the studies were not randomized or reporting on human subjects. However, none of the included studies had either an adequate description of withdrawals and dropouts or appeared to use an intention-to-treat analysis.

Effects of intervention (summarized in Fig. 1)

The first proof-of-concept adaptation of PDT to treat spinal metastasis was reported in 2005 by Burch *et al*.[21] They developed an approach taken from vertebroplasty/kyphoplasty to utilize PDT for spinal metastasis and had successfully shown that a single PDT treatment in this configuration was able to ablate tumor cells in a rat metastatic breast cancer model.[21] The effect of PDT on tumor cells was comparable in both thoracic and lumbar vertebrae, indicating that PDT can be adapted to various regions of the spine. However, it is important to note that due to the variation proximity of the spinal cord to the bone structure, the dosage of photosensitizer and light energy should be adjusted corresponding to vertebral level. Burch *et al* also successfully used a cannulated approach to administer PDT to a single vertebral body in a porcine model, demonstrating the feasibility of applying such a technique to humans.[26] Photodynamic therapy was found to destroy bone marrow cells, adversely affecting osteoclasts. This may be clinically advantageous, as osteoclasts have a symbiotic relationship with tumor cells in skeletal resorption. Yet, the bone tissue itself was found to be highly resilient against photodynamic damage, a positive finding as further damage to the bone would result in an elevated risk of skeletal-related events.

Important parameters in determining the treatment protocol of PDT are the drug-light interval, drug, and light dose. A study by Akens *et al*[22] examined varying light and drug doses, as well as the therapeutic window at which PDT would maximize treatment effect and minimize side effects in a rat model of metastatic breast cancer. It was determined that a benzoporphyrin derivative monoacid dose of 0.5 mg/kg and a light dose of 50 J for

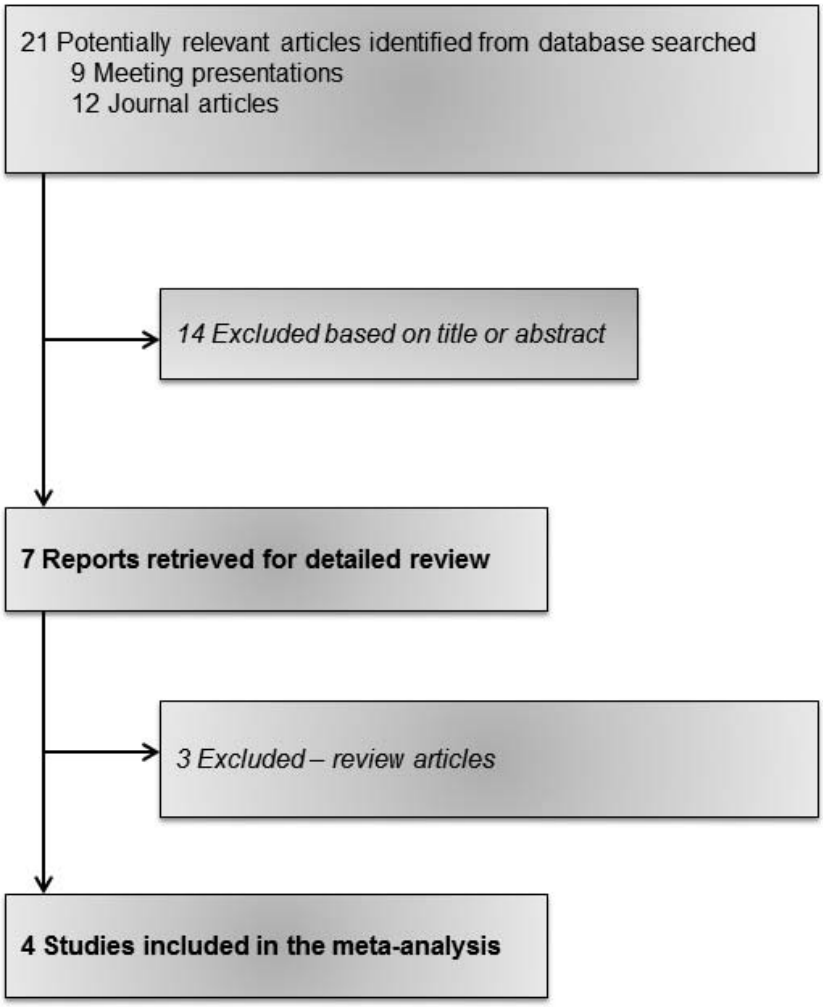

Fig. 1 Quality of reporting of meta-analyses (QUOROM) flow diagram.

thoracic vertebrae and 75 J for lumbar vertebrae was most effective in ablating tumor tissue while minimizing damage to the spinal cord.[22,26]

Won *et al*[23] published a study demonstrating a bone-enhancing effect provided by PDT on healthy vertebrae. In this study, the effects of PDT on the structural integrity and strength of the spine was evaluated in healthy Wistar rats both 1 and 6 weeks after treatment. It was found that there was a trend toward enhancements in both bone architecture and strength 1 week after PDT treatment, which became statistically significant at 6 weeks posttreatment.

Bisphosphonates (BPs) are a clinical standard of care for patients with spinal metastasis that have been shown to reduce the risk of skeletal-related events. Radiation therapy (RT), like BP, is a standard in care for spinal metastasis patients, and is the cornerstone noninvasive treatment to reduce pain. As such, it is clinically relevant to understand the interaction of PDT with previously administered BP and RT treatment on healthy and metastatically involved vertebrae. Recently, Lo *et al*[24] evaluated combined PDT + BP and PDT + RT to determine if PDT could be a potential adjuvant to treat tumors untreatable by BP or RT alone. An athymic nude rat model was used in this study in order to be able to establish metastases secondary to injection of MT-1 human breast cancer cells. Stereological and mechanical properties were determined using μCT imaging and compression testing. Histological evaluation was also performed using hematoxylin and eosin (H&E), Goldner's trichrome, tartrate-resistant acid phosphatase (TRAP), and human epidermal growth factor receptor (hEGFr) sections.

In healthy vertebrae, at a 1-week time point, PDT, BP, or RT alone did not demonstrate significant differences in stereology compared with untreated controls.[23] Combined PDT + RT[24] and PDT + BP[23] increased bone volume fraction and trabecular thickness compared with untreated controls. Com-

bined PDT + RT also showed an increase in trabecular number with increasing trends in bone volume fraction compared with RT alone, indicating structural benefits associated with PDT. While the benefits of combined PDT + BP treatment did not statistically differ from individual BP or PDT treatment, combined PDT + BP therapy demonstrated an increase in bone mass whereas individual treatments did not.[23] This suggests that a combinatory benefit exists in PDT + BP treatment not seen in either BP or PDT alone.[23]

At this 1-week time point, no differences in mechanical properties were found in treatment groups compared with untreated healthy vertebrae.[23] Since only half of the samples were tested, this may have resulted in a lack of statistical power to detect differences compared with controls. Yet, Pearson correlation analyses demonstrated that improvements in stereological parameters were associated with increases in stiffness and strength.[23] Surprisingly, PDT + RT demonstrated a reduction in ultimate stress compared with alone, yet had no significant effect on ultimate force or stiffness values. Vertebrae treated with PDT + RT, however, showed no differences in mechanical properties compared with untreated controls[23] at the 1 week time point.

In metastatically involved vertebrae, at a 1 week time point, stereological analysis determined that PDT reduced the amount of tumor-mediated osteolysis, resulting in increased bone volume fraction and trabecular number compared with untreated controls.[23] Not surprisingly, BP increased bone volume fraction, trabecular number and thickness as well.[23] While treatment with BP did not demonstrate tumor cell death, the work of Lo *et al* showed that it inhibited metastatic progression into the vertebrae by up to 40%. Combined PDT + BP demonstrated even greater benefits than either PDT or BP alone, by increasing bone volume fraction, trabecular number and thickness.[23] Similar to results in healthy vertebrae, combination PDT + BP appears to be more beneficial than PDT or BP alone. A radiation therapy dose of 4Gy at day 7 unexpectedly destroyed the tumor cells in all treated vertebrae and prevented further metastasis, as supported by hEGFr sections. This resulted in a negative bias when comparing RT and PDT + RT–treated vertebrae to other groups. Therefore, both RT and PDT + RT groups significantly increased bone volume fraction, trabecular thickness and number, and reduced trabecular separation compared with untreated controls. Even though PDT alone induced tumor destruction and showed improvement over untreated controls, the protective effects of early RT did not allow for consideration of the potential effects of combined PDT with RT on the tumor in this study.[24]

Mechanically, in metastatically involved vertebrae, significant increases in stiffness and strength were found in the BP treatment group, with similar trends in the PDT alone and combined PDT + BP treatment groups compared with controls.[23] Not surprisingly, mechanical properties of RT and PDT + RT–treated vertebrae were also improved over untreated controls and PDT-only treated vertebrae. However, no differences in mechanical properties were found between RT alone and combined PDT + RT–treated vertebrae, in contrast to what was observed in healthy vertebrae.

Hematoxylin and eosin and hEGFr sections confirmed the cytotoxic effect of PDT on the MT-1 tumors within the vertebrae.[23] Bisphosphonates and PDT alone both inhibited the amount of tumor-mediated osteolysis by the 1-week time point. Photodynamic therapy offers the advantage of destroying viable tumor cells, whereas BP therapy in this study only led to inhibition of further tumor growth. Hematoxylin and eosin, hEGFr, and TRAP sections demonstrated destruction of many, but not all, osteocytes in PDT + RT–treated bone, whereas osteocytes were not impacted in the RT alone group. Viable osteocytes were seen in areas of new bone formation in PDT + RT–treated bone, both in the trabecular centrum and on the periosteal surface. Examination of Goldner's trichrome sections revealed increased osteoid volume in these areas that appeared as low-density bone on the μCT images.

Overall, the short-term results demonstrated the feasibility and benefit of administering PDT to spinal metastasis patients who have received previous BP treatment, as PDT ablates tumor and interacts with BP to yield a positive effect on bone.[23] Additionally, PDT may be administered with prior RT, but further study may be warranted to confirm the effects of mechanical properties in the short-term.[24] The combined use of BP along with PDT + RT, or other stabilizing techniques such as vertebroplasty/kyphoplasty may also serve to mitigate any potential decreases in bone strength.[24]

The long-term effects of PDT alone and combined with previous BP and RT treatment on healthy vertebrae were also investigated.[24] Stereologically, PDT alone and RT alone did not demonstrate significant improvements, similar to findings at 1 week. Bisphosphonates alone increased bone vol-

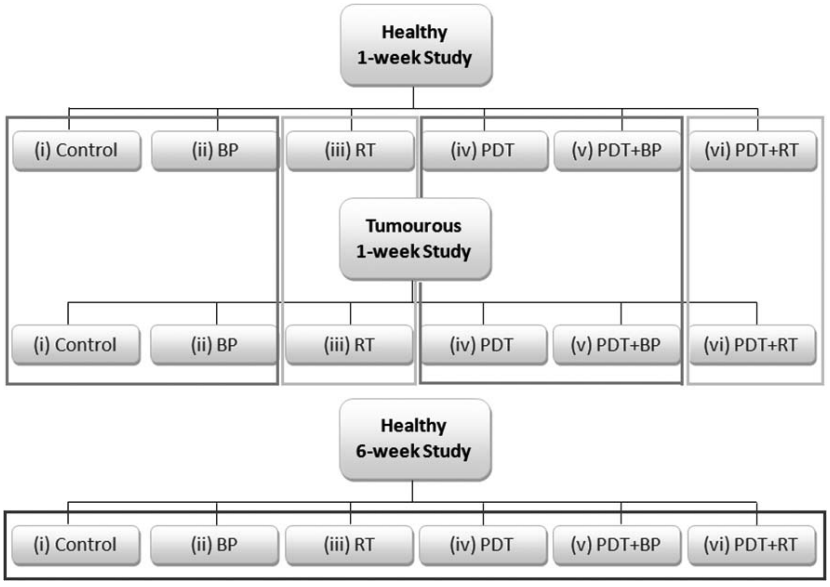

Fig. 2 Photodynamic therapy and combination treatments in healthy and metastatically involved vertebrae. Investigation into the effects of PDT and bisphosphonates or radiation therapy at 1 week (orange, 23; green, 24) and 6-weeks (purple, 24) post-PDT.

ume fraction, but not to the extent of combined treatment groups. Both combined PDT + RT and PDT + BP treatments increased bone volume fraction and decreased trabecular separation, with PDT + BP demonstrating greater improvements. Furthermore, only PDT + BP increased trabecular thickness, number, and volumetric bone mineral density (vBMD). These results reflect what is seen at the earlier time point of 1 week: that the most significant stereological improvements were observed in combination treatment groups.[24]

Mechanically the results at 6 weeks were also very similar to the short-term healthy data.[24] No significant differences were detected between the groups, despite the structural benefits observed in the μCT images. Again, statistical power was limited due to the use of only half of the samples for mechanical testing. However, as found at 1 week, ultimate force and stiffness correlated moderately well with bone volume fraction, and trabecular vBMD, indicating that increases in stereological parameters are associated with increases in mechanical properties.[24]

Histological assessment of Goldner's trichrome sections revealed a higher volume of osteoid in combination treatment groups, providing support for the increased new bone formation observed in μCT images, particularly on the periosteal surface.[24] Bone treated with PDT demonstrated incomplete osteocyte ablation maintained at 6 weeks, yet osteoclast activity was returned to physiologically normal levels at this time point.[24]

Overall, the study demonstrated that the effects on stereological and mechanical properties observed at the 1-week time point are maintained at a longer 6-week time point, with combined PDT + BP treatment being the most beneficial in terms of bone enhancement.[23,24] Additionally, the combination of PDT + RT also demonstrated significant increases in stereological parameters, suggesting that previous RT treatment does not preclude the bone-enhancing effects of PDT and in fact may be synergistic in the longer term (Fig. 2).[23,24]

Discussion

As advancements in cancer treatment prolong patient survival the opportunity for metastasis increases. Vertebral metastases commonly occur in advanced breast, prostate, and lung cancers. Current treatments remain mostly palliative and have variable responses. Thus, there is a need for new treatments or combination therapies to reduce tumor burden in the spine, maintain or restore spinal stability, and minimize risk to healthy tissue. Our analyses of 4 studies indicate the potential of photodynamic therapy to meet this clinical need.

Our analyses show that PDT with combination treatments in both healthy and metastatically involved vertebrae in the short (1 week) and longer term (6 weeks) are effective. Further study to understand the molecular, cellular, and long-term (>6 weeks) effects of PDT would provide guidance to optimize therapeutic outcomes. Particularly, *in vitro* and *in vivo* studies to examine the effects of

PDT and combination treatments on primary osteoblast and osteoclast cell cultures, to elucidate the cellular and molecular response are required. Discovery of changes in gene expression due to PDT will help to the further understanding of bone formation.

New periosteal bone formation was demonstrated within the majority of PDT-treated vertebrae.[23,24] This excess bone formation did not result in any clinical signs (no symptoms or behavioral changes). It would be of interest to examine the effects of PDT at a much longer time point (>6 weeks) to determine the fate of the vertebrae: whether this newly formed bone is maintained or is eventually resorbed.

Weaknesses of this review rest with limitations in the primary studies. We were unable to make any firm statements about the quality of the evidence, since the therapies were only reported in small samples and in animal models. The current spinal metastasis animal model limits the survival of animals to 3 weeks after tumor cell injection and can only be done in young rats. Thus, long-term studies of the effects of PDT in metastatically involved vertebrae are not possible using the current model. The development of a new larger and older animal model of spinal metastasis would be beneficial to study the effects of PDT. In particular, tumors that are more specifically targeted to the spine (metastatic or through direct injection) would allow the extension of animal survival and enable longer-term studies. Such studies would also provide insight into the potential for local reoccurrence after PDT treatment. A larger model would also allow direct placement of the laser fiber within the vertebra to better simulate the ultimate clinical utilization of PDT. Additionally, an older animal model may be more reflective of the biological bone response in a clinical setting since the spinal metastasis patient population is generally elderly. This would also generate an understanding of the effects of PDT on mature rather than rapidly growing bone.

Overall, the bone-enhancing effects of PDT in combination with conventional treatments and its ability to destroy metastatic human breast cancer cells within bone, present PDT as an attractive novel treatment for spinal metastasis. Finally, the positive results from these preclinical studies motivate studying the translation of PDT for the treatment of spinal metastases in humans. A phase I clinical trial is currently underway to study the safety of PDT, results of which are awaited. Ultimately the results from this initial safety trial will guide the future clinical translation of PDT for spinal metastasis.

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