White paper

Bladder cancer

BioXmark®

The liquid fiducial marker

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NANOVI
Diplomvej 378
DK-2800 Lyngby
+45 31 42 11 86
www.nanovi.com

1. Introduction

This white paper covers the clinical use of Nanovi's BioXmark in patients with bladder cancer. We present background knowledge on bladder cancer and the use of fiducial markers to improve radiotherapy. Furthermore, we introduce BioXmark® - the liquid fiducial marker, and the clinical evidence supporting that BioXmark® can be implanted safely in bladder cancer patients to guide high precision radiotherapy.

2. Bladder cancer background

In North America and Europe, bladder cancer ranks 5th based on incidence with approximately 294,000 new cases and 9th based on mortality with approximately 88,000 deaths in 2020. The incidence and mortality is higher for men than for women, with men accounting for approx. 3 out of 4 new cases and deaths[1].

90-95% of bladder cancers are urothelial carcinomas. Urothelial carcinomas can be categorized as low-grade or high-grade. Patients rarely die from low-grade bladder cancer, whereas almost all deaths result from high-grade disease[2]. Bladder cancer is also grouped into muscle-invasive and non muscle-invasive cancer and metastatic cancer. Each group differs in clinical behavior, primary management and outcome[3].

3. Radiation therapy background

Radiation therapy in cancer can have different aims. It may be given with curative intent in cases with localized disease. It can be given as neoadjuvant therapy for tumor shrinkage before surgery or may be used as part of adjuvant therapy, to prevent tumor recurrence after surgical resection of the primary malignant tumor. Radiation therapy is synergistic with chemotherapy. It may also be used as palliative treatment, where cure is not possible [3,4].

The total dose of radiation used in radiation therapy varies depending on the cancer type and is fractionated into smaller doses for several reasons. Fractionation allows healthy cells time to recover, while tumor cells are generally less efficient in repair between fractions. Fractionation also allows tumor cells that were in a relatively radio-resistant phase of the cell cycle during one treatment to cycle into a sensitive phase of the cycle before the next fraction is given. A type of fractionation schedule that is increasingly being used and continues to be studied is hypofractionation. This is a radiation treatment in which the total dose of radiation is divided into fewer and larger doses. This type of radiation therapy necessitates a high degree of accuracy since just a single fraction missing the target will mean a huge decrease in total amount of radiation delivered to the tumor and an equally high dose wrongly delivered to healthy tissue[3,4].

3.1 Radiotherapy for bladder cancer

The use of radiotherapy for bladder cancer depends on the staging. Patients with non muscle-invasive cancers are in most cases treated with transurethral section of the bladder (TURB) followed by intravesical chemotherapy [2]. For this group of patients, radiotherapy is rarely used. For patients with muscle-invasive cancer, treatment options can broadly be divided into treatments that remove the bladder and those that spare it. The main treatments are either neoadjuvant chemotherapy followed by radical cystectomy or radiotherapy with concomitant chemotherapy (with radical cystectomy in case of tumor recurrence). Successful bladder-preserving approaches have evolved during the past three decades [3]. Originally reserved for inoperable patients, advancements in radiotherapy technology and radio sensitizing chemotherapy gave rise to chemoradiation as equivalent treatment alternative to a radical cystectomy [5]. There are no randomized trials that directly compare the bladder-preserving chemoradiation therapy approach with radical cystectomy, thus the relative effectiveness of these two treatments is unknown [2]. However, indirect comparison based on meta-analyses data has demonstrated similar survival and loco-regional tumor control rates [5]. Thus, today radiotherapy plays an important role in the treatment of this group of patients.

4. Fiducial markers background

A fiducial marker is an object placed in the field of view of an imaging system that appears in the image produced, for use as a point of reference. Methods to secure a target reference point in radiation therapy have a long history and were initially seen in the form of a cross penciled or tattooed mark on the skin of the patient to guide the entry point of the radiation beam. Later, when Image Guided Radiation Therapy (IGRT) was introduced, bony structures in close relation to the tumor were used as landmarks on images for patient set-up at the point of treatment and as a guide for better target precision. Most of the imaging modalities available at the point of treatment are however not able to differentiate sufficiently between different soft tissues, including the tumor and the surrounding non-cancerous tissue. Furthermore, inter fractional and intra-fractional movement of the tumor target complicates the precise delivery of the radiation dose to the tumor[4,6,7].

For a fiducial marker to be a relevant tool through all phases of radiation therapy the following features are needed:

- Feasible to implant with low risk of procedure related complications
- Visible on relevant imaging modalities
- Positional stable throughout the entire treatment course and through follow-up

Advantages of using fiducial markers:

- Accurate identification of tumor target location for better treatment planning, treatment, and follow-up

- Maximization of radiation to the tumor target and minimization of radiation to healthy surrounding tissue
- Makes it possible to locate the tumor target despite day-to-day variation on the treatment unit and help overcome the challenge of inter-fractional target movement
- Makes it possible to live monitor tumor motion during a fraction of radiation treatment and help overcome the challenge of intra-fractional target movement
- Allowing accurate re-identification of the tumor target in the time of follow-up

4.1 Fiducial markers for bladder cancer

Delivering precision radiotherapy to maximize radiation delivered to the tumor and minimize radiation to healthy surrounding tissue is challenging in bladder cancer since the bladder is a mobile, deformable structure and bladder volume can vary markedly during a course of radiotherapy and motion can be dependent on the location of the tumor in the bladder wall and can vary up to 3 cm [8,9]. This means that large margins are needed to ensure tumor tissue is targeted thereby increasing radiation to healthy tissue[5].

To assist with tumor delineation and daily planning CT fiducial markers are utilized in bladder cancer. Currently applied fiducial markers include golds seeds, titanium clips, hydrogel or Lipiodol[8].

The safety and feasibility of placing gold seed markers in the bladder has been evaluated in several studies and an increase in target accuracy has been demonstrated[10–12]. In a study with 20 patients, Biancia *et al.* finds a significant advantage in using intravesical fiducial markers to determine daily translational corrections[12], and in a study with 18 patients, Garcia *et al.* concludes that bladder fiducial markers improve targeting accuracy, and may increase treatment efficacy and reduce morbidity from collateral radiation[11]. A challenge with gold seed markers can be the positional stability and a review article by Nolan and Forde[8], describes marker loss during treatment ranging from 2% to 41%, when assessed on various imaging modalities[10–12]. Another challenge is that these marker need to implemented by a rigid cystoscope, which is less convenient for the patient and certain locations in the bladder can be difficult to reach [5].

The safety and feasibility of using titanium clips has also been evaluated[13,14]. Hulshof *et al.* demonstrated how titanium clips could help with target delineation in a study with 19 patients[13]. The positional stability of the clips was a challenge for the reliability for positional verification on the cone beam scan, with half of the clips lost after 1 week. However, the study concludes that when combined with positional verification during an adaptive margin strategy for focal bladder boost irradiation, the method signifies a major improvement in the accuracy of external beam radiotherapy for bladder cancer.

In a retrospective study Shahbaz *et al*. showed how titanium endoclips can be used as fiducial markers in trimodality bladder preserving therapy on 15 patients with muscle invasive bladder carcinoma[14]. 49 endoclips were applied and all clips remained through the course of the radiotherapy and were removed thereafter. It is concluded that the patient cohort showed favorable outcomes with the use of endoclips.

Wortel *et al*. performed a study that evaluated the feasibility and technical performance of cystoscopy-guided placement of hydrogel markers in patients with bladder cancer referred for radiotherapy [15]. A total of 107 hydrogel spots were implanted in 32 patients with bladder cancer. Their results showed that "*on the simulation CT scan 76.6% of the implanted markers were sufficient for tumor delineation. At start of treatment 52.3% were visible on the CBCT and adequate for positional verification on a routine basis. The washout rate during treatment was 9.3%. At the end of treatment 46.7% of implanted spots were visible on CBCT scan and adequate for routine positional verification. At patient level, in 31.2% of cases, use of hydrogel fiducials showed adequate performance throughout the whole course*". Wortel *et al*. concludes that the results leave room for improvement and states that "*the current results call for a search for a better liquid fiducial in bladder cancer treatment, with improvement of visibility on CT and CBCT scan (higher density) and less blurring, migration, and wash out*".

5. BioXmark® - the liquid fiducial marker

BioXmark® is a unique carbohydrate/iodine-based liquid low density fiducial marker. The liquid nature of BioXmark® enables implantation of multiple size-adaptable markers in the same uninterrupted procedure. BioXmark® can be implanted with thin needles and flexible scopes guided visually, by fluoroscopy and/or ultrasound. Upon injection of the BioXmark® liquid into soft tissue, efflux of ethanol leads to the *in-vivo* formation of a radiopaque and gel-like fiducial marker.

5.1 BioXmark® - Indications for use

5.1.1 Europe

BioXmark® is indicated for use to radiographically mark soft tissue. BioXmark® is intended to mark tissue for at least 2 months after implantation.

5.1.2 United States

BioXmark® has De Novo clearance from the US FDA with an indication for use to radiographically mark lung, bladder and lymph nodes in adult patients for whom it has been determined that radiographical marking of tissue for radiation treatment is indicated for their cancer treatment.

BioXmark® is implanted via image-guided injection into tissue relevant for radiotherapy planning at a healthcare facility. BioXmark® can be implanted in the tumor, lymph nodes or tissue adjacent to the tumor subject to irradiation or in healthy tissue which should not be irradiated.

BioXmark® is intended to mark tissue for at least 3 months after implantation.

5.2 Positional stability and long-term visibility

BioXmark® is positional stable and visible on CT and MRI during treatment planning, treatment, and follow-up. Long-term visibility has been demonstrated up to 6 years[a].

5.3 Low level of artifact and MR safe

Streaking and shadowing artifacts are commonly encountered in CT with currently used metal-based markers. These artifacts are problematic since they induce a loss of clarity and increase inaccuracy in dose calculation during tumor target delineation in treatment planning and in the patient positioning during treatment[17].

Fiducial markers creating a lower level of artifacts allows for better dose calculation accuracy due to better image quality, including the area around the marker, than for markers with higher level of artifacts.

Due to its non-metallic composition BioXmark® has been found to generate a low level of artifacts in CT. This has been demonstrated in a study by Scherman *et al.* using a water phantom in a clinical diagnostic CT-scanner using various tube voltages from 80kV to 140kV in 20kV steps (Figure 1)[18] and has been confirmed by clinical investigations in bladder and lung[5,19]

The non-metallic composition is also an advantage in MR since there are no displacements of BioXmark®. The product is labelled MR safe according to ASTM F2503-13.

[a] Additional follow up on patients from clinical investigation by de Blanck *et al*. [16]

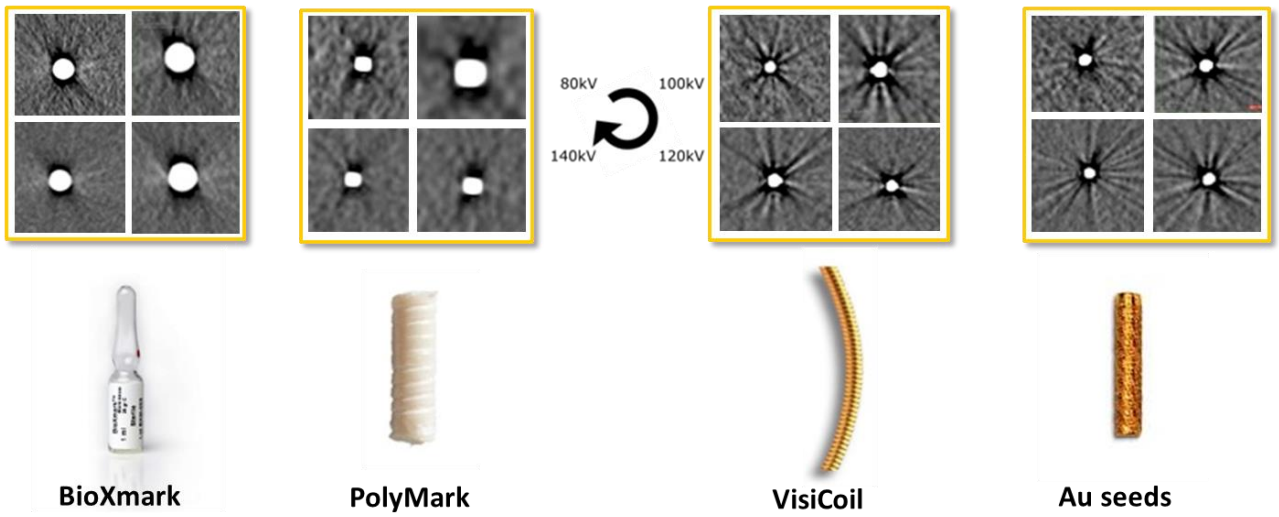

Figure 1. Artifacts of different markers on CT images at different tube voltages.

5.4 Low dose perturbation

For the use of a fiducial marker to be beneficial, an improved positioning accuracy must not be offset by marker-induced dose distortion. This constitutes a negligible challenge in photon therapy, but is a significant consideration in proton therapy, where fiducials can cause severe perturbations of the proton dose and lead to cold-spots downstream the marker, where the tissue will not receive the intended radiation dose. This interaction is described as the Relative Stopping Power (RSP), which is high in metals.

The ideal fiducial marker for proton therapy combines a low RSP value with good visibility on 2D X-ray and CBCT with a low level of artifacts.

BioXmark®'s non-metallic composition gives a low RSP, compared to metal, which ensures low dose perturbation in proton radiation therapy combined with the low levels of artifacts described above.

The RSP of BioXmark® has been calculated to be 1.174 and measured to be 1.164 by Troost et al. in a phantom model[20]. Furthermore, the BioXmark® markers were evaluated after being exposed to normofractionated and extremely hypofractionated proton therapy and no chemical degradation was observed[20].

Rydhög and colleagues has, in collaboration with Professor Lomax from the Paul Scherrer Institute, performed a gelatin phantom study where BioXmark® markers of 0.01-0.1 ml were investigated for dose perturbation in proton therapy. The largest of the BioXmark marker (0.1 ml) perturbed the proton beam in a spread-out Bragg Peak with a maximum of 4.8% as measured in the film placed the furthest from the phantom meant to capture downstream shadowing effects. The dose perturbation shall be taken into account when planning treatment doses in proton therapy in accordance with local procedures and national guidelines[21].

5.5 Injectable with thin needles

Injection of BioXmark® is possible with percutaneous and endoscopic needles. The liquid formulation can be injected using thin needles up to 25G. The use of thin needles gives lower risk of procedure related complications such as bleedings and pneumothorax.

5.6 Endoscopic implantation

BioXmark® can be implanted using flexibles scopes, making it possible to access tumors located at anatomical locations not accessible with rigid scopes or percutaneously.

The possibility of implanting BioXmark® endoscopically has been evaluated in several different types of endoscopes, e.g., flexcystoscopy[[5], endoscopic ultrasound, endobronchial ultrasound and video bronchoscope[16].

5.7 Implantation of multiple size-adaptable markers in the same procedure

BioXmark® enables the implantation of multiple markers in the same uninterrupted endoscopic or percutaneous procedure, with no need for retraction of endoscope and/or needle for reloading. This has been demonstrated by de Blanck S. *et al.* concluding: *"The liquid formulation also allows for the placement of several markers in one session without needing to reload the endoscopy needle between each implantation […]"*[16]. Fewer injections are associated with less risk of procedure related complications.

The optimal injection volume depends on the intended target site, planned treatment, and the applied image modality as well as desired visibility and artifact level. In general, both visibility and artifacts increase with larger injection volumes[17]. The volume of each BioXmark® marker can be determined prior to, or adapted during, the implantation procedure.

5.8 Implantation guided by ultrasound and fluoroscopy

During the marker implantation procedure, the location of the needle and BioXmark® marker can be visualized and guided by fluoroscopy and/or ultrasound, ensuring precision and safety during marker placement and verification of marker location. The feasibility of guiding BioXmark® implantation by fluoroscopy and/or ultrasound has been demonstrated, incl. clinical investigation in lung and bladder cancer[5,16].

5.9 Biocompatible

BioXmark has been biologically evaluated and tested in compliance with ISO standards and FDA guidance related to the biocompatibility of medical devices. It was found to be safe and biocompatible within the intended use.

6. Clinical use of BioXmark® in bladder cancer

In a prospective, single center study, de Ridder *et al.* evaluated the performance of BioXmark® in IGRT for muscle-invasive bladder cancer[5].

In the study, 20 patients with muscle-invasive bladder cancer had BioXmark® injected around the tumor using a flexible cystoscope. Minimum number of markers implanted were 3 and maximum 5 with a median of 4 markers. Markers were implanted in an outpatient setting with a flexible cystoscope.

Primary endpoints of the study were "*safety of the marker implantation procedure, marker visibility and positional stability of the fiducial markers over time*". The visibility was evaluated on planning CT and weekly CBCT during treatment. "*Accepted criteria for performance and clinical applicability were that 75% of the markers had to remain visible and positionally stable from the CT acquisition for RT planning to the last CBCT without causing grade three toxicity (CTCAE v4.0)*". Secondary endpoints were "*the appearance of blurring (i.e. diffuse spread out of the liquid through the bladder wall instead of a single dot), time needed for implantation and possibility for automatic online matching for IGRT (XVI Elekta)*".

The results showed that of the 76 implanted markers, 60 markers (79% CI 70-88%) were visible on treatment planning CT. All visible markers on the planning CT continued to be visible in the same position during the treatment course to the last CBCT. All separate spots were continuously classified as clearly visible without artifacts, no migration occurred, and the markers did not fade. No blurring occurred and all present markers could be used for delineation, patient setup and for automatic marker matching. Figure 2, 3 and 4 shows the same BioXmark® marker in a bladder patient during planning, treatment and follow-up scans.

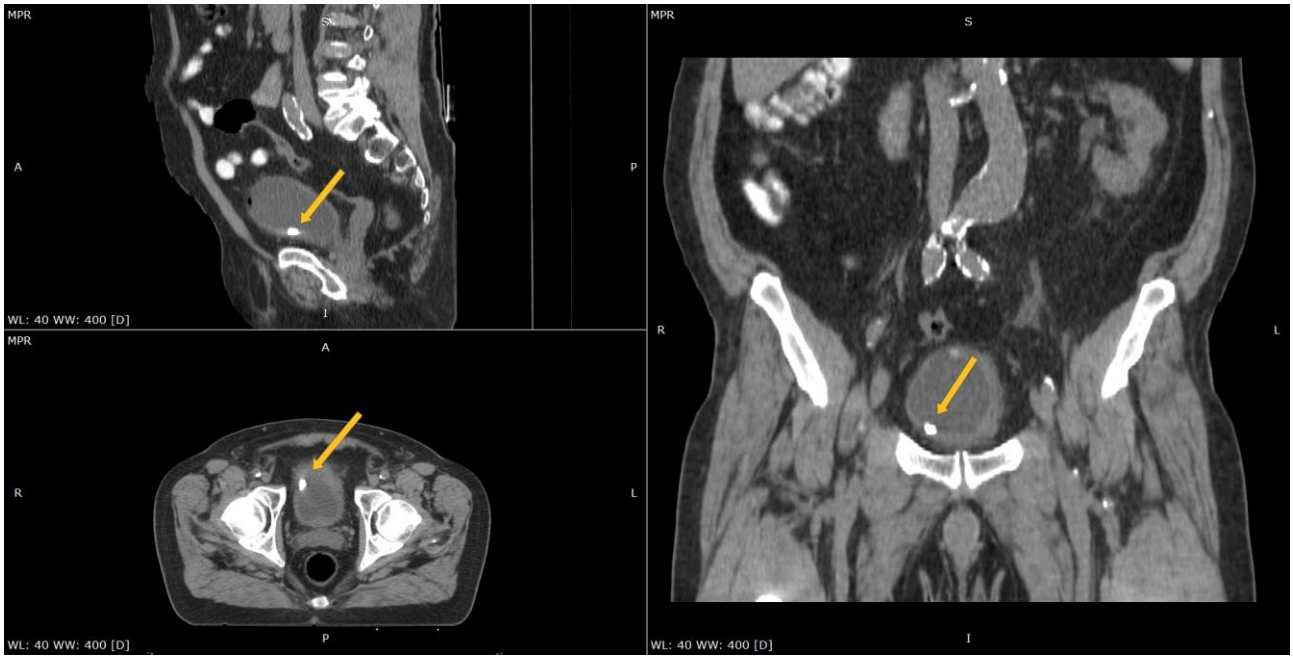

Figure 2: BioXmark® in bladder, Planning CT Scan

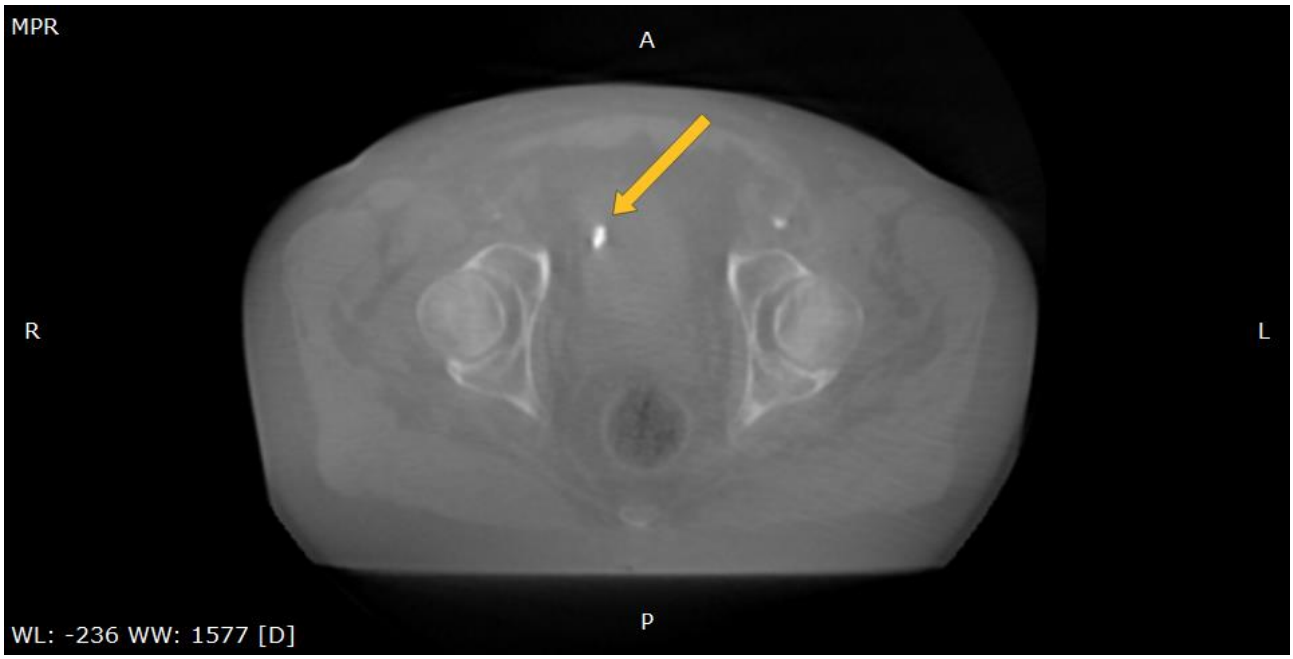

Figure 3: BioXmark® in bladder, CBCT scan

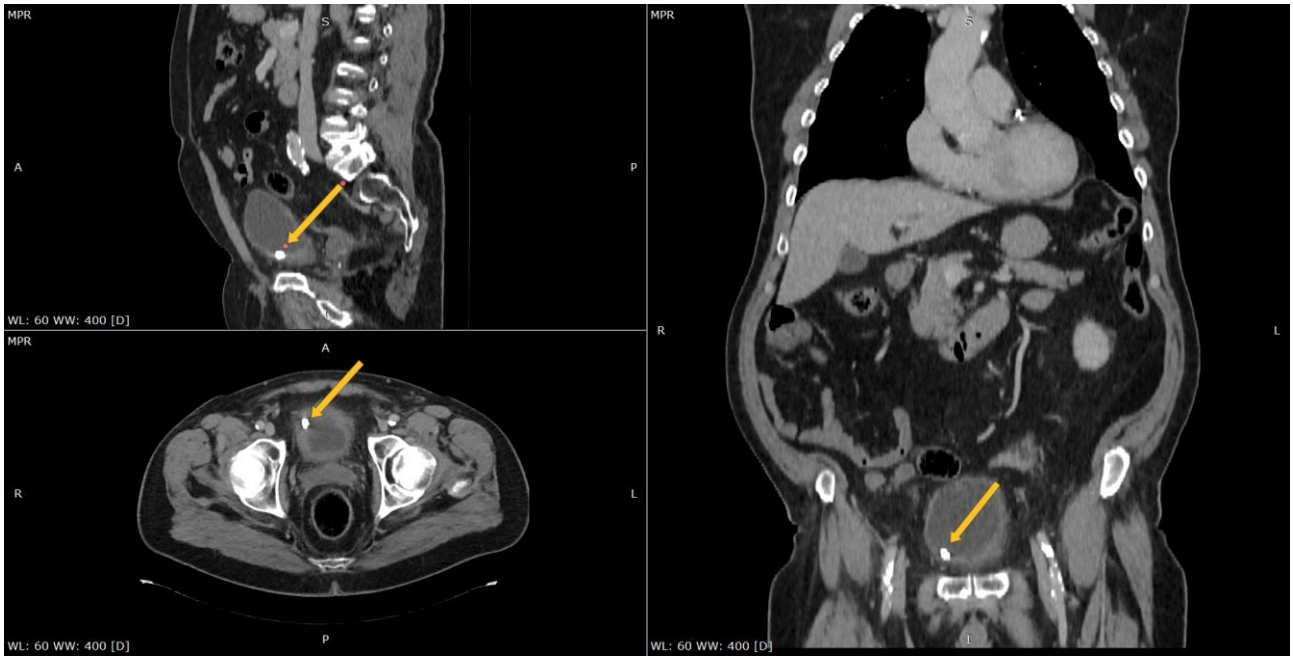

Figure 4: BioXmark® in a bladder, 9 months follow up CT scan.

The clinical performance of 79% was above the preset threshold of 75%, although overlapped by the confidence intervals of the limits of significance (95% CI). However, it is noted that the bar was set high and that the results showed a learning curve for implanting the markers with the lost markers mainly being due to the learning curve of the implanting technique.

The study demonstrated that BioXmark® has advantages compared to other fiducial markers. De Ridder *et al.* states: "*Compared to other liquid markers available for endoscopic procedures, the use of BioXmark® seems to need some more training, as discussed before. But, in our view, there are many advantages of this marker compared to other markers, i.e. no blurring, fading, or migration, with all visible BioXmark® marker dots on CT acquisition remaining detectable and stable on CBCTs during the whole treatment period. Implantation in the wall of the bladder can be performed in an outpatient setting by every urologist after following a short learning curve and getting used to the specific characteristics of the liquid marker and the injection technique.*"

The study concludes that the marker showed sustained visibility and positional stability during treatment phases and appears to be safe and easy to inject. Further it is concluded that the "*novel liquid BioXmark® marker seems to be a very promising tool in daily-adaptive IGRT for bladder preserving chemoradiotherapy in muscle invasive bladder cancer*".

7. Conclusion

It is demonstrated that BioXmark® enables precision radiation therapy in patients with bladder cancer.

The use of BioXmark® in patients with bladder cancer is safe and technically feasible. BioXmark® has sustained visibility and positional stability during the entire treatment course and throughout follow-up period.

Implantation of BioXmark® in the wall of the bladder with flexible scope can be performed in an outpatient setting by urologists following a short learning curve.

8. List of references

1. GLOBOCAN 2020. Ferlay J, Ervik M, Lam F, Colombet M, Mery L, Piñeros M, Znaor A, Soerjomataram I, Bray F (2020). Global Cancer Observatory: Cancer Today. Lyon, France: International Agency for Research on Cancer. [Internet]. [cited 2022 Mar 10]. Available from: http://gco.iarc.fr/today/home

2. Bladder Cancer Treatment (PDQ®)–Health Professional Version - National Cancer Institute [Internet]. 2022 [cited 2022 Mar 30]. Available from: https://www.cancer.gov/types/bladder/hp/bladder-treatment-pdq

3. DeVita VT, Lawrence TS, Rosenberg SA, editors. DeVita, Hellman, and Rosenberg's cancer: principles & practice of oncology. 11th edition. Philadelphia: Wolters Kluwer; 2019. 2390 p.

4. Symonds P, Mills JA, Duxbury A, Walter J, Miller H, editors. Walter and Miller's textbook of radiotherapy: radiation physics, therapy and oncology. Eighth edition. Amsterdam: Elsevier; 2019. 615 p.

5. de Ridder M, Gerbrandy LC, de Reijke TM, Hinnen KA, Hulshof MCCM. BioXmark® liquid fiducial markers for image-guided radiotherapy in muscle invasive bladder cancer: a safety and performance trial. BJR. 2020 Jun 1;20200241.

6. O'Neill AGM, Jain S, Hounsell AR, O'Sullivan JM. Fiducial marker guided prostate radiotherapy: a review. BJR. 2016 Dec;89(1068):20160296.

7. Choi JH, Seo DW, Park DH, Lee SK, Kim MH. Fiducial Placement for Stereotactic Body Radiation Therapy under Only Endoscopic Ultrasonography Guidance in Pancreatic and Hepatic Malignancy: Practical Feasibility and Safety. Gut Liver. 2014 Jan 15;8(1):88–93.

8. Nolan CP, Forde EJ. A review of the use of fiducial markers for image-guided bladder radiotherapy. Acta Oncologica. 2016 May 3;55(5):533–8.

9. Lim Joon D, Berlangieri A, Harris B, Tacey M, O'Meara R, Pitt B, et al. Exploratory models comparing ethiodized oil-glue and gold fiducials for bladder radiotherapy image-guidance. Physics and Imaging in Radiation Oncology. 2021 Jan;17:77–83.

10. Mangar S, Thompson A, Miles E, Huddart R, Horwich A, Khoo V. A feasibility study of using gold seeds as fiducial markers for bladder localization during radical radiotherapy. BJR. 2007 Apr;80(952):279–83.

11. Garcia MM, Gottschalk AR, Brajtbord J, Konety BR, Meng MV, Roach M, et al. Endoscopic Gold Fiducial Marker Placement into the Bladder Wall to Optimize Radiotherapy Targeting for Bladder-Preserving Management of Muscle-Invasive Bladder Cancer: Feasibility and Initial Outcomes. Samant R, editor. PLoS ONE. 2014 Mar 3;9(3):e89754.

12. Biancia CD, Yorke E, Kollmeier MA. Image guided radiation therapy for bladder cancer: Assessment of bladder motion using implanted fiducial markers. Practical Radiation Oncology. 2014 Mar;4(2):108–15.

13. Hulshof MCCM, van Andel G, Bel A, Gangel P, van de Kamer JB. Intravesical markers for delineation of target volume during external focal irradiation of bladder carcinomas. Radiotherapy and Oncology. 2007 Jul;84(1):49–51.

14. Shahbaz M, Ammar A, Wang Y, Farhaj Z, Qiao L, Niu J. Endoclips as novel fiducial markers in trimodality bladder-preserving therapy of muscle-invasive bladder carcinoma: feasibility and patient outcomes. Int Braz J Urol. 2021 Feb;47(1):93–9.

15. Wortel K, Hovius MC, van Andel G, de Reijke TM, Hulshof MCCM. The Feasibility and Utility of Cystoscopy-Guided Hydrogel Marker Placement in Patients With Muscle-Invasive Bladder Cancer. Practical Radiation Oncology. 2020 May;10(3):195–201.

16. de Blanck SR, Rydhög JS, Larsen KR, Clementsen PF, Josipovic M, Aznar MC, et al. Long term safety and visibility of a novel liquid fiducial marker for use in image guided radiotherapy of non-small cell lung cancer. Clinical and Translational Radiation Oncology. 2018 Nov;13:24–8.

17. De Roover R, Crijns W, Poels K, Peeters R, Draulans C, Haustermans K, et al. Characterization of a novel liquid fiducial marker for multimodal image guidance in stereotactic body radiotherapy of prostate cancer. Med Phys. 2018 May;45(5):2205–17.

18. Rydhog Scherman J, Irming Jølck R, Andresen TL, Munck af Rosenschöld P. Quantification and comparison of visibility and image artifacts of a new liquid fiducial marker in a lung phantom for image-guided radiation therapy: A new liquid fiducial marker for IGRT. Med Phys. 2015 May 15;42(6Part1):2818–26.

19. Rydhög JS, Mortensen SR, Larsen KR, Clementsen P, Jølck RI, Josipovic M, et al. Liquid fiducial marker performance during radiotherapy of locally advanced non small cell lung cancer. Radiotherapy and Oncology. 2016 Oct;121(1):64–9.

20. Troost E, Menkel S, Enghardt W, Hytry J, Kunath D, Makocki S, et al. EP-1710: Chemical stability of BioXmark® following normofractionated and single-fraction proton beam therapy. Radiotherapy and Oncology. 2017 May 1;123:S937–8.

21. Rydhög Scherman J, Perrin R, Jølck RI, Gagnon-Moisan F, Larsen KR, Clementsen P, et al. Liquid fiducial marker applicability in proton therapy of locally advanced lung cancer. Radiotherapy and Oncology. 2017 Mar;122(3):393–9.

NANOVI
Diplomvej 378
DK-2800 Lyngby
+45 31 42 11 86
www.nanovi.com