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***Observational Study***

**Intensified intensity modulated radiotherapy in anal cancer with prevalent HPV p16 positivity**

Belgioia L *et al*. Anal cancer SIB-IMRT: outcomes and toxicity

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**Abstract**

**AIM:** To investigate toxicity and response of intensity modulated radiotherapy schedule intensified with a simultaneous integrated boost in anal canal cancer.

**METHODS:** From March 2009 to March 2014 we analyzed retrospectively 41 consecutive patients treated with intensity modulated radiotherapy (IMRT) and concurrent chemotherapy for anal canal squamous cell carcinoma in our Institute. Radiotherapy was delivered with a simultaneous integrated boost (SIB) technique by helical tomotherapy and doses were adapted to two clinical target volumes according to the disease TNM stage: 50.6 Gy and 41.4 Gy in 23 fractions in T1N0, 52.8 Gy and 43.2 Gy in 24 fractions in T2N0 and 55 Gy and 45 Gy in 25 fractions in all patients with N positive and/or ≥T3, respectively to planning target volume 1 and 2. The most common chemotherapy regimen was 5-fluorouracil and mitomycin based. HPV p16 expression was performed by immunohistochemistry and evaluated in the majority of patients. Acute and late toxicity was scored according to CTCAe v 3.0 and RTOG scales.

**RESULTS:** The median follow up was 30 mo (range: 12-71). Median age was 63 years (range 32-84). The stage of disease was: stage I in 2 pts, stage II in 13 pts, stage IIIA in 12 pts and stage IIIB in 14 pts, respectively. Two patients were known to be HIV positive (4.9%). HPV p16 expression status was positive in 29/34 (85.3%) patients. The 4**-**year progression free survival and overall survival in HPV positive patients were 78% and 92%, respectively. Acute grade 3 skin and grade 3 gastrointestinal toxicity was reported in 5% and 7.3% of patients, respectively; patients’ compliance to the treatment was good due to a low occurrence of severe acute toxicity, treatment interruptions due to toxicity were required in 7.3% of patients. At 6 mo from treatment ending 36/40 (90%) patients obtained a complete response; during follow up 5 (13.8%) patients presented disease progression (local or systemic).

**CONCLUSION:** In our experience**,** an intensified SIB-IMRT with chemotherapy is well feasible in clinical practice with excellent results in terms of overall survival and local control.

**Key words:** Anal canal cancer; Intensity modulated radiotherapy; Simultaneous integrated boost; Helical tomotherapy; HPV

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**Core tip:** This study evaluated intensity modulated radiotherapy with simultaneous integrated boost in anal canal cancer in prevalent HPV positive patients. The results show excellent outcomes in HPV positive tumors and suggest that an intensified radiotherapy schedule associated to chemotherapy is safe and it allows to obtain oncologic results comparable to standard schedule without an increase in acute toxicity.

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**INTRODUCTION**

Anal canal cancer is a rare pathology accounting for 2.4% of digestive system cancer[1]. The standard procedure consists of a combined radiation-chemotherapy treatment, which allows the achievement ofhigh rates of local control, overall survival (OS) and disease free survival (DFS)[2]. However, due to the high toxicity that could lead to treatment interruptions, different chemotherapy regimens and schedule of RT have been tentatively explored to improve compliance to treatment[3,4]. Over the last few years, the introduction of intensity modulated radiotherapy (IMRT) has resulted in being attractive because it has shown to maintain radiation dose within clinical target volumes, while reducing average and threshold doses to Organs-at-Risk (OARs) such as genitals, perineum, small bowel and bladder compared with conventional 3D-Conformal RT[5]. IMRT with a Simultaneous Integrated Boost of Dose (SIB) could be devised as an innovative intensified strategy to escalate radiation doses to sites of macroscopic disease. This approach can be used with some advantages compared to a sequential dosage increment: the possibility to deliver different fraction doses to different volumes, to shorten overall treatment time (OTT) and to guarantee a better coverage of Gross Tumor Volume (GTV) with non-target tissue sparing. SIB use has been investigated also in other diseases than anal canal cancer, such as head and neck and cervical cancer, and it demonstrated to be feasible and reduce acute toxicity[6-8]. Here we presented the results of a study carried out with an intensified radiotherapy regimen associated with chemotherapy applied in order to improve oncologic efficacy and reduce overall acute toxicity in patients with squamous cell carcinoma of the anal canal treated in our Institute.

**MATERIALs AND METHODS**

***Population***

We analyzed retrospectively 41 consecutive patients with histologically documented anal canal squamous carcinoma, clinical stage T1-4, N0-3, treated between March 2009 and March 2014 in our Institute, a public university hospital. All tumors originated from anal canal and could extended to perianal region in advanced disease.

Pretreatment evaluation included complete history, physical, blood chemistry, chest radiogram or chest/abdominal Computed Tomography (CT), Magnetic Resonance Imaging (MRI) of pelvis and/or FDG Positron Emission Tomography (PET-TC), sigmoidoscopy or transrectal ultrasound to assess the disease stage. Patients were excluded if they had metastatic disease or prior pelvic radiotherapy.

***HPV detection***

Evaluation of p16 expression status, as surrogate of Human Papilloma Virus (HPV) infection, was performed by immunohistochemistry using the automated immunostainer Ventana BenchMark® XT platform (Ventana Medical Systems, Arizona, United States) and anti-p16INK4a antibodies (Ventana Medical Systems, Arizona, United States). p16 is a cyclin dependent kinase inhibitor with an important role in cell cycle and cellular differentiation. Overexpression of p16 is found in neoplastic cells and is strongly associated with the molecular expression of the E7 oncoprotein of HPV.

***Radiotherapy***

CT-based planning with a slice thickness ≤ 5 mm from the upper lumbar spine to the midfemur was performed. Oral contrast was recommended to allow better visualization of the bowel; all patients had a full bladder and a radio-opaque vaginal marker was placed in female patients to assist in target delineation. Patients were scanned in supine position and immobilized with a thermoplastic mask from lung bases to mid femur with All-In-One (AIO) solution or COMBIFIXTM for pelvic district. The images were transferred to the Eclipse treatment planning system (Varian Medical Systems) for contouring. The Gross Tumor Volume (GTV), Clinical Tumor Volume (CTV), Planning Target Volume (PTV), and avoidance structures including the bowel bag, bladder, external genitalia, femoral heads, pelvic bones and intergluteal sulcus were contoured. Data was sent to Tomotherapy planning station for treatment planning. The GTV, including the primary tumor and macroscopically involved lymph nodes, was identified using examination, imaging, and endoscopy findings. Two clinical target volumes were delineated: CTV1 and CTV2. The CTV1 consisted of the GTV plus 5 mm expansion, CTV2 included elective nodal stations (perirectal, internal iliac, external iliac, obturator, presacral and inguinal nodes); an isotropic margin of 5 mm was added to generate Planning Tumor Volumes (PTV1 and PTV2). The treatment consisted of IMRT with SIB delivered by Helical Tomotherapy (6 MV). Dose prescription for target volumes varied according to the clinical stage disease: 50.6 Gy and 41.4 Gy in 23 fractions in T1N0, 52.8 Gy and 43.2 Gy in 24 fractions in T2N0 and 55 and 45 Gy in 25 fractions in all patients with N+ and/or ≥T3 prescribed to PTV1 and PTV2, respectively. The plan was accepted if: (1) at least 97% of the volume was covered by 95% of the dose; (2) at least 99% of the volume was covered by 90% of the dose both for PTV1 and PTV2; (3) no more than 1% received more than 5% of the prescription dose for PTV1; and (4) no more than 5% received more than 5% of the prescription dose for PTV2. MVCT scans before each fraction was performed to verify patients’ positioning; image matching was executed using automatic bone algorithm supervised by experienced medical staff. Normal tissue dose constraints are listed in table 1.

***Chemotherapy***

All patients were evaluated for chemotherapy (CHT) by a medical oncologist; most patients received concurrent chemotherapy with 5-FU continuous venous infusion 1000 mg/m2 over 96 h and mitomycin-C (MMC) 10 mg/m2 during first and last week of radiation therapy; in case of contraindication to polychemotherapy**,** patients received capecitabine (825 mg/m2 bid/die) concomitant with RT or underwent exclusive radiotherapy.

***Toxicity and follow up***

Toxicities were graded weekly during chemo-radiation and in follow-up (every week in the first month after combined treatment and then at 8 wk post-treatment). Patients underwent rectal examination at 6-8 wk after the completion of radiotherapy course, then at 3 mo from treatment they underwent pelvic MRI and rectoscopy; most patients also underwent an endorectal ultrasound; at 4 mo a PET/CT was performedto evaluate metabolic response. In patients with complete remission, follow-up investigations were carried out at 3 mo-intervals in the first year, then every 6 mo for 5 years by MRI or endorectal ultrasound alternated with PET/CT. In cases of incomplete response, the clinical and radiological evaluation was repeated every 4 weeks until the complete remission was recorded[7]. In cases of evidence of progression or recurrence**,** surgery was recommended. Post-treatment biopsies were not routinely performed. Local regional recurrence was defined as recurrence of thedisease within the pelvis. All failures were documented by a biopsy. Distant failure was defined as thedevelopment of thedisease outside the pelvis or inguinal nodes. Acute and late adverse events were measured according to the Common Toxicity Criteria for Adverse Events scale v3.0 and RTOG criteria, respectively[9,10]

***Statistical analysis***

The primary endpoint of this study were disease free survival (DFS) and local control rate (LC), secondary end points were overall survival (OS), progression free survival (PFS), acute and late toxicity. Statistical analysis was performed with JMP v 10.0 (SAS institute Inc. Cary, NC) and DFS, LC, OS rate were calculated with Kaplan Meier non parametric estimation from treatment start.

**RESULTS**

From March 2009 to March 2014 we treated 41 patients, the characteristics of patients are summarized in table 2. The median follow up was 30 mo (range 12-71); all but one patient had a follow up of more than 12 mo. The stage of disease was: stage I in 2 pts, stage II in 13 pts, stage IIIA in 12 pts and stage IIIB in 14 pts respectively. Evaluation of p16 expression status was positive in 29/34 (85.3%) patients and it was not available in 6/41 patients; two patients were known to be HIV positive (4.9%).

***Chemo-radiotherapy***

The prescription dose of PTV1 was 55 Gy in 29 patients, 52.8 Gy in 10 patients and 50.6 Gy in 2 patients; the median dose of PTV1 and PTV2 was 55 Gy (range, 50.6-55) and 45 Gy (range, 41.4-45), respectively. All patients completed radiotherapy treatment except one who interrupted treatment, on his own accord, at 50.6 Gy of 55 Gy planned and refused the second chemotherapy course. Treatment breaks due to toxicity have occurred in 3 patients (7.3%) (median 1 d – range 1-3 d) and due to other causes (holidays or broken machine) in 27 patients (65.8%) (median 1 d- range 1-5 d). The median OTT was 35 d (range 30-40). OARs optimization constraints were respected. Concomitant CHT was delivered in 39/41 patients, 2 patients did not receive CHT for comorbidity. 3/39 patients did not complete chemotherapy courses as planned mainly for haematological toxicity (thrombocytopenia and anaemia) and 1 patient presented cardiovascular disease (angina pectoris) after first CHT course.

***Toxicity***

All patients were evaluated for acute toxicity. Grade 2 (11 patients, of whom 10 had diarrhea and 1 had both nausea and diarrhea) and grade 3 (diarrhea) gastrointestinal toxicity was reported, respectively, in 26.8% and 7.3% of patients. 29.4% of patients developed grade 2 pain and 24/41 (58.5%) had to be administered major analgesic therapy; only 4 patients were receiving analgesic therapy before treatment start. Grade 3 skin toxicity occurred in 2 patients. 2 patient**s** developed a recto-vaginal fistula during RT (at 33 Gy and 19.3 Gy respectively), both of them were T4 stage, one healed on its own after 7 mo from CHT/RT and the other one underwent temporary colostomy 2 mo after treatment end. Late toxicity was assessed in 40/41 (97.5%) patients: no patient received a colostomy for stricture. One patient died for chemotherapy related toxicity with pancytopenia and sepsis one week after treatment end. Acute and late toxicity are shown in tables 3 and 4.

***Outcomes***

40/41 patients were evaluable as 1 patient died one week after treatment end. The first imaging evaluation showed a complete response (CR) in 25/40 (62%) patients and a partial response (PR) in 15/40 (38%) patients. At 6 mo, 11 of the 15 partial responders had achieved a CR. Among the 4 patients with residual tumor shown radiologically at 6 mo, 2 underwent biopsy and were confirmed to have loco-regional persistence, 1 presented systemic and local progression and 1 systemic progression; as of today two of them are alive and under chemotherapy, the other two patients (both T4 stage) died at 28 and 24 mo from treatment end. During follow-up 5/36 patients in complete response presented a progression disease: 1 patient had a local recurrence 11 mo after treatment end and after undergoing abdominal-perineal resection and today is disease free. One patient developed lung metastasis after 18 mo and was treated with SBRT (48 Gy/4 fx - 12 Gy/fraction), and has, as of today, obtained a metabolic complete response; three patients developed metastatic disease - liver metastasis (2 pts) and bone metastasis (1pt): they underwent to second line chemotherapy and at last follow up presented stable disease. Among the 9 patients that presented an event (persistence or progression disease) 6 were p16 positive and 3 were p16 negative. The PFS and OS in HPV positive patients were 78% and 92% at 4 years, respectively. The DFS, CFS, LC and OS were 78.3%, 94%, 92% and 93% at 2 years, respectively (Figures 1 and 2).

**DISCUSSION**

Over the past 30 years, concurrent FU/MMC based chemo-radiotherapy has been the standard of care in anal carcinoma. This approach enables sphincter preservation in most patients without compromising rates of cure. However**,** CHT/RT is associated with significant acute gastrointestinal, genitourinary, dermatological and hematological toxicities when conventional radiation therapy techniques are used. As a consequence, prolonged treatment breaks are usually necessary and have been shown to negatively affect local control[11] as highlighted by several studies (v RTOG 9208)[12]. Different trials have demonstrated that IMRT may decrease the incidence of acute toxicity rates, with similar outcomes compared with previous clinical trials. The RTOG 0529 trial[13], a phase II study, demonstrated a reduction in grade 3 gastrointestinal, grade 3 dermatologic and grade 2 hematologic toxicity using IMRT when compared with 3D-CRT. Moreover**,** the RTOG 0529 is the only phase II prospective trial, to our knowledge, in which RT is delivered with a simultaneous integrated boost (SIB)[13].

***Statement of principal findings***

In our study we used an IMRT-SIB technique intensifying treatment with a modest accelerated hypofractionated schedule. This schedule allows us to shorten the radiotherapy overall treatment time**;** our median OTT was 35 d *vs* 43 and 49 d reported respectively in RTOG 0529 and RTOG 98-11; furthermore**,** we reported very few breaks due to toxicity (7.3% *vs* 49% when compared with RTOG 0529 trial rate). This data appears to be very interesting and may be explained by the fact that the major toxicities are likely to occur when the radiotherapy course is concluded, thus they do not affect the treatment course that could be administered more easily**.** We reported a relatively high rate of patients (58.5%) that received major analgesics during treatment even if not encountering grade 3 pain, this is probably explained by the fact that patients presented a basal pain well controlled with minor analgesics but also a breakthrough pain (e.g. during defecation) that required major analgesics. Our acutetoxicity rate is however comparable with those reported in literature (table 5).

As to mid-term late toxicity we did not observe any grade 4 late toxicity and no patient underwent surgery for fecal incontinence or stricture. According to these preliminary results,we believe that severe late effects should not affect our patients even if an intensified schedule of radiotherapy with single SIB dose of 2,2 Gy is adopted in a volume including anal sphincter and muscles involved in anal function; we also have to considered that the analysis of treatment related toxicity is commonly difficult after radio-chemotherapy for anal cancer; late effects were not specifically reported in any trial, since the current RTOG late effects instruments are not sufficiently specific, and itshould be considered that quite a few patients appear to adapt even to animpaired function of pelvic organs.

***Strengths and weakness of the study and in relation to other studies***

An important aspect to analyze is the possibility that the use ofa SIB technique gives to deliver different doses to different volumes, therefore to increasedose to GTV. Although the role of dose escalation is still controversial in anal cancer, multivariate analysis of data from RTOG 98-11 trial showed that positive lymph nodes and tumor size greater than 5 cm were an independently prognostic factor for worse OS**;** tumor diameter could also be prognostic for colostomy rate and time to colostomy[14]; similarly**,** recent multivariate analysis of data from the ACT1 trial also showed that positive lymph nodes is a prognostic indicator for higher local regional failure, anal cancer death and lower OS[15]. The final analysis of UNICANCER ACCORD 03 trial has not demonstrated to benefit from high dose radiotherapy, even if it reported a small increase in CFS and LC that has not reached statistical significance but leads to further investigation on the role of dosege escalation[16]. It should also be considered that radiation boost in ACCORD 03 trial was delivered 3 wk after treatment end, and this gap could have contributed to reducing a possible positive effect of high dose radiotherapy. On the basis of this data and considering that local relapse remains the main site of failure especially in locally advanced disease, a dose escalation might be reasonable.

In our casuistic 83% of patients were HPV p16 positive and higher disease control was expected for these patients, in fact our results in this subgroup of patients are excellent (OS and PFS were 92% and 78% at 4 years, respectively) and comparable with that reported in literature[17]. A preliminary analysis on HPV positive *vs* HPV negative patients seems to show a trend in favor of the first group even if patients number is too small. In literature other experiences reported that HPV negative tumors were linked to a worse prognosis[17-19]. Another consideration is that in our study we detected p16 expression by immunohistochemistry but we have not performed HPV DNA testing. In literature some data reported that p16 is not a perfect surrogate marker for tumor HPV status, its specificity might be insufficient and the use of HPV DNA could still be required if patients were to be stratified based on HPV status[19], this aspect is particularly interesting and it has been demonstrated also in diseases different from anal cancer. In oropharyngeal tumors it is possible to identify a subgroup of patients p16 positive but HPV DNA negative whose survival is significantly different compared with p16+ and HPV DNA positive patients, the survival curve of this group converged on the survival curve of HPV negative patients[20]. Based on these observations it is possible that we registered some HPV false positive tumors and this probably represents another limit of our study.

In the past years there appears to be on the increase in the incidence of anal canal cancer, probably related to HPV infection, we have considered 41 patients treated at a single institution with a relatively short, median follow**-**up (30 mo) and analyzed them retrospectively; these factors represent the main limit of this study, but as of today several casuistics reported in literature refer to small groups of patients (18-36 patients) with a short follow up (14-32 mo)[21-27]; in accordance to this, our study aligns with other experiences, moreover our preliminary results in terms of LC and OS are encouraging and in line with those reported in literature (Table 6).

Our study is distinct from other studies in terms of radiotherapy dose, fractionation. Further investigations on HPV status are necessary in order to understand if different schedules of radiotherapy (intensified or not) should be delivered according to viral related biology of anal cancer. Our next project is to isolate HPV DNA in all patients to better investigate this aspect and to clarify the crucial aspects of anal function in surviving patients by periodically administering questionnaires on thequality of life at least 6 mo after treatment end.

**COMMENTS**

***Background***

Despite different strategies have been adopted to minimize local recurrence, in anal canal cancer it remains a major issue. Recently the importance of biological factor (HPV) has been proven as a predictor of the prognosis.

***Research frontiers***

HPV could be used for the individualization of treatment. Moreover the introduction of intensity modulated radiotherapy and integrated boost (SIB-IMRT) technique allowed the reduction toxicity related treatment.

***Innovations and breakthroughs***

SIB allowed to easily modify the radiotherapy schedule, increasing effective biological dose without increasing the overall treatment time. This aspect could be particularly interesting in HPV negative patients.

***Applications***

The results of the study show excellent outcomes in HPV positive patients and highlighted the good tolerance of a moderately accelerated SIB-IMRT radiotherapy schedule.

***Terminology***

IMRT is a radiotherapy technique that allows to obtain a better target coverage, a better dose distribution and a reduction of the dose received by normal tissue compared to the previous technique (3D conformal radiotherapy). SIB allowed the treatment, at the same time, of different volumes at different doses and allowed an increase of the dose prescribed to the target safely.

***Peer-review***

This is a good study in which the authors evaluated the prognostic significance of HPV status and the feasibility and outcomes of an accelerated intensity modulated radiotherapy in anal canal cancer patients. The study is well structured and the subject is clear and interesting. The manuscript is correctly written and the conclusions are justified by the results found in the study.

**REFERENCES**

1 **Siegel R**, Naishadham D, Jemal A. Cancer statistics, 2013. *CA Cancer J Clin* 2013; **63**: 11-30 [PMID: 23335087 DOI: 10.3322/caac.21166]

2 **Northover J**, Glynne-Jones R, Sebag-Montefiore D, James R, Meadows H, Wan S, Jitlal M, Ledermann J. Chemoradiation for the treatment of epidermoid anal cancer: 13-year follow-up of the first randomised UKCCCR Anal Cancer Trial (ACT I). *Br J Cancer* 2010; **102**: 1123-1128 [PMID: 20354531 DOI: 10.1038/sj.bjc.6605605]

3 **Ajani JA**, Winter KA, Gunderson LL, Pedersen J, Benson AB, Thomas CR, Mayer RJ, Haddock MG, Rich TA, Willett C. Fluorouracil, mitomycin, and radiotherapy vs fluorouracil, cisplatin, and radiotherapy for carcinoma of the anal canal: a randomized controlled trial. *JAMA* 2008; **299**: 1914-1921 [PMID: 18430910 DOI: 10.1001/jama.299.16.1914]

4 **Gunderson LL**, Winter KA, Ajani JA, Pedersen JE, Moughan J, Benson AB, Thomas CR, Mayer RJ, Haddock MG, Rich TA, Willett CG. Long-term update of US GI intergroup RTOG 98-11 phase III trial for anal carcinoma: survival, relapse, and colostomy failure with concurrent chemoradiation involving fluorouracil/mitomycin *vs* fluorouracil/cisplatin. *J Clin Oncol* 2012; **30**: 4344-4351 [PMID: 23150707 DOI: 10.1200/JCO.2012.43.8085]

5 **Kachnic LA**, Tsai HK, Coen JJ, Blaszkowsky LS, Hartshorn K, Kwak EL, Willins JD, Ryan DP, Hong TS. Dose-painted intensity-modulated radiation therapy for anal cancer: a multi-institutional report of acute toxicity and response to therapy. *Int J Radiat Oncol Biol Phys* 2012; **82**: 153-158 [PMID: 21095071 DOI: 10.1016/j.ijrobp.2010.09.030]

6 **Stromberger C**, Jann D, Becker ET, Raguse JD, Tinhofer I, Marnitz S, Budach V. Adjuvant simultaneous integrated boost IMRT for patients with intermediate- and high-risk head and neck cancer: outcome, toxicities and patterns of failure. *Oral Oncol* 2014; **50**: 1114-1121 [PMID: 25204515 DOI: 10.1016/j.oraloncology.2014.08.006]

7 **Yi J**, Huang X, Gao L, Luo J, Zhang S, Wang K, Qu Y, Xiao J, Xu G. Intensity-modulated radiotherapy with simultaneous integrated boost for locoregionally advanced nasopharyngeal carcinoma. *Radiat Oncol* 2014; **9**: 56 [PMID: 24533569 DOI: 10.1186/1748-717X-9-56]

8 **Herrera FG**, Callaway S, Delikgoz-Soykut E, Coskun M, Porta L, Meuwly JY, Soares-Rodrigues J, Heym L, Moeckli R, Ozsahin M. Retrospective feasibility study of simultaneous integrated boost in cervical cancer using Tomotherapy: the impact of organ motion and tumor regression. *Radiat Oncol* 2013; **8**: 5 [PMID: 23286694 DOI: 10.1186/1748-717X-8-5]

9 CTEP: Cancer therapy evaluation program. Available from: URL: http: //ctep.cancer.gov/protocolDevelopment/electronic\_applications/docs/ctcaev3.pdf

10 RTOG: radiation therapy oncology group. Available from: URL: http: //www.rtog.org/ResearchAssociates/AdverseEventReporting/RTOGEORTCLateRadiationMorbidityScoringSchema.aspx

11 **DeFoe SG**, Beriwal S, Jones H, Rakfal S, Heron DE, Kabolizadeh P, Smith RP, Lalonde R. Concurrent chemotherapy and intensity-modulated radiation therapy for anal carcinoma--clinical outcomes in a large National Cancer Institute-designated integrated cancer centre network. *Clin Oncol* (R Coll Radiol) 2012; **24**: 424-431 [PMID: 22075444 DOI: 10.1016/j.clon.2011.09.014]

12 **Konski A**, Garcia M, John M, Krieg R, Pinover W, Myerson R, Willett C. Evaluation of planned treatment breaks during radiation therapy for anal cancer: update of RTOG 92-08. *Int J Radiat Oncol Biol Phys* 2008; **72**: 114-118 [PMID: 18472363 DOI: 10.1016/j.ijrobp.2007.12.027]

13 **Kachnic LA**, Winter K, Myerson RJ, Goodyear MD, Willins J, Esthappan J, Haddock MG, Rotman M, Parikh PJ, Safran H, Willett CG. RTOG 0529: a phase 2 evaluation of dose-painted intensity modulated radiation therapy in combination with 5-fluorouracil and mitomycin-C for the reduction of acute morbidity in carcinoma of the anal canal. *Int J Radiat Oncol Biol Phys* 2013; **86**: 27-33 [PMID: 23154075 DOI: 10.1016/j.ijrobp.2012.09.023]

14 **Ajani JA**, Winter KA, Gunderson LL, Pedersen J, Benson AB, Thomas CR, Mayer RJ, Haddock MG, Rich TA, Willett CG. US intergroup anal carcinoma trial: tumor diameter predicts for colostomy. *J Clin Oncol* 2009; **27**: 1116-1121 [PMID: 19139424 DOI: 10.1200/JCO.2008.19.6857]

15 **Glynne-Jones R**, Sebag-Montefiore D, Adams R, Gollins S, Harrison M, Meadows HM, Jitlal M; United Kingdom Coordinating Committee on Cancer Research Anal Cancer Trial Working Party. Prognostic factors for recurrence and survival in anal cancer: generating hypotheses from the mature outcomes of the first United Kingdom Coordinating Committee on Cancer Research Anal Cancer Trial (ACT I). *Cancer* 2013; **119**: 748-755 [PMID: 23011911 DOI: 10.1002/cncr.27825]

16 **Peiffert D**, Tournier-Rangeard L, Gérard JP, Lemanski C, François E, Giovannini M, Cvitkovic F, Mirabel X, Bouché O, Luporsi E, Conroy T, Montoto-Grillot C, Mornex F, Lusinchi A, Hannoun-Lévi JM, Seitz JF, Adenis A, Hennequin C, Denis B, Ducreux M. Induction chemotherapy and dose intensification of the radiation boost in locally advanced anal canal carcinoma: final analysis of the randomized UNICANCER ACCORD 03 trial. *J Clin Oncol* 2012; **30**: 1941-1948 [PMID: 22529257 DOI: 10.1200/JCO.2011.35.4837]

17 **Yhim HY**, Lee NR, Song EK, Kwak JY, Lee ST, Kim JH, Kim JS, Park HS, Chung IJ, Shim HJ, Hwang JE, Kim HR, Nam TK, Park MR, Shim H, Park HS, Kim HS, Yim CY. The prognostic significance of tumor human papillomavirus status for patients with anal squamous cell carcinoma treated with combined chemoradiotherapy. *Int J Cancer* 2011; **129**: 1752-1760 [PMID: 21128253]

18 **Ravenda PS**, Magni E, Botteri E, Manzotti M, Barberis M, Vacirca D, Trovato CM, Dell'Acqua V, Leonardi MC, Sideri M, Fazio N, Zampino MG. Prognostic value of human papillomavirus in anal squamous cell carcinoma. *Cancer Chemother Pharmacol* 2014; **74**: 1033-1038 [PMID: 25209946 DOI: 10.1007/s00280-014-2582-x]

19 **Meulendijks D**, Tomasoa NB, Dewit L, Smits PH, Bakker R, van Velthuysen ML, Rosenberg EH, Beijnen JH, Schellens JH, Cats A. HPV-negative squamous cell carcinoma of the anal canal is unresponsive to standard treatment and frequently carries disruptive mutations in TP53. *Br J Cancer* 2015; **112**: 1358-1366 [PMID: 25871546 DOI: 10.1038/bjc.2015.20]

20 **Rietbergen MM**, Brakenhoff RH, Bloemena E, Witte BI, Snijders PJ, Heideman DA, Boon D, Koljenovic S, Baatenburg-de Jong RJ, Leemans CR. Human papillomavirus detection and comorbidity: critical issues in selection of patients with oropharyngeal cancer for treatment De-escalation trials. *Ann Oncol* 2013; **24**: 2740-2745 [PMID: 23946330 DOI: 10.1093/annonc/mdt319]

21 **Deenen MJ**, Dewit L, Boot H, Beijnen JH, Schellens JH, Cats A. Simultaneous integrated boost-intensity modulated radiation therapy with concomitant capecitabine and mitomycin C for locally advanced anal carcinoma: a phase 1 study. *Int J Radiat Oncol Biol Phys* 2013; **85**: e201-e207 [PMID: 23517808 DOI: 10.1016/j.ijrobp.2012.12.008]

22 **Tozzi A**, Cozzi L, Iftode C, Ascolese A, Campisi MC, Clerici E, Comito T, De Rose F, Fogliata A, Franzese C, Mancosu P, Navarria P, Tomatis S, Villa E, Scorsetti M. Radiation therapy of anal canal cancer: from conformal therapy to volumetric modulated arc therapy. *BMC Cancer* 2014; **14**: 833 [PMID: 25403766 DOI: 10.1186/1471-2407-14-833]

23 **Janssen S**, Glanzmann C, Bauerfeind P, Stieb S, Studer G, Brown M, Riesterer O. Clinical experience of SIB-IMRT in anal cancer and selective literature review. *Radiat Oncol* 2014; **9**: 199 [PMID: 25199879 DOI: 10.1186/1748-717X-9-199]

24 **Bazan JG**, Hara W, Hsu A, Kunz PA, Ford J, Fisher GA, Welton ML, Shelton A, Kapp DS, Koong AC, Goodman KA, Chang DT. Intensity-modulated radiation therapy *vs* conventional radiation therapy for squamous cell carcinoma of the anal canal. *Cancer* 2011; **117**: 3342-3351 [PMID: 21287530 DOI: 10.1002/cncr.25901]

25 **Kim KH**, Chang JS, Keum KC, Ahn JB, Lee CG, Koom WS. Chemoradiotherapy in squamous cell carcinoma of the anal canal: a single institution experience. *Radiat Oncol J* 2013; **31**: 25-33 [PMID: 23620866 DOI: 10.3857/roj.2013.31.1.25]

26 **Pepek JM**, Willett CG, Wu QJ, Yoo S, Clough RW, Czito BG. Intensity-modulated radiation therapy for anal malignancies: a preliminary toxicity and disease outcomes analysis. *Int J Radiat Oncol Biol Phys* 2010; **78**: 1413-1419 [PMID: 20231064 DOI: 10.1016/j.ijrobp.2009.09.046]

27 **Bentzen AG**, Guren MG, Vonen B, Wanderås EH, Frykholm G, Wilsgaard T, Dahl O, Balteskard L. Faecal incontinence after chemoradiotherapy in anal cancer survivors: long-term results of a national cohort. *Radiother Oncol* 2013; **108**: 55-60 [PMID: 23891095 DOI: 10.1016/j.radonc.2013.05.037]

28 **Vieillot S**, Fenoglietto P, Lemanski C, Moscardo CL, Gourgou S, Dubois JB, Aillères N, Azria D. IMRT for locally advanced anal cancer: clinical experience of the Montpellier Cancer Center. *Radiat Oncol* 2012; **7**: 45 [PMID: 22445137 DOI: 10.1186/1748-717X-7-45]

29 **Chuong MD**, Freilich JM, Hoffe SE, Fulp W, Weber JM, Almhanna K, Dinwoodie W, Rao N, Meredith KL, Shridhar R. Intensity-Modulated Radiation Therapy vs. 3D Conformal Radiation Therapy for Squamous Cell Carcinoma of the Anal Canal. *Gastrointest Cancer Res* 2013; **6**: 39-45 [PMID: 23745158]

30 **Koerber SA**, Slynko A, Haefner MF, Krug D, Schoneweg C, Kessel K, Kopp-Schneider A, Herfarth K, Debus J, Sterzing F. Efficacy and toxicity of chemoradiation in patients with anal cancer--a retrospective analysis. *Radiat Oncol* 2014; **9**: 113 [PMID: 24886574 DOI: 10.1186/1748-717X-9-113]

31 **Dasgupta T**, Rothenstein D, Chou JF, Zhang Z, Wright JL, Saltz LB, Temple LK, Paty PB, Weiser MR, Guillem JG, Nash GM, Goodman KA. Intensity-modulated radiotherapy vs. conventional radiotherapy in the treatment of anal squamous cell carcinoma: a propensity score analysis. *Radiother Oncol* 2013; **107**: 189-194 [PMID: 23692961 DOI: 10.1016/j.radonc.2013.03.012]

32 **Mitchell MP**, Abboud M, Eng C, Beddar AS, Krishnan S, Delclos ME, Crane CH, Das P. Intensity-modulated radiation therapy with concurrent chemotherapy for anal cancer: outcomes and toxicity. *Am J Clin Oncol* 2014; **37**: 461-466 [PMID: 23466576 DOI: 10.1097/COC.0b013e31827e52a3]

**P-Reviewer:** Francois E, Ong JJ, Zilli T **S-Editor:** Ma YJ **L-Editor:** **E-Editor:**

**Table 1 Constraints for normal tissue**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Organ** | **Constraints** | | | |
| Bladder | Dmedian < 30 Gy | V45 < 35% |  |  |
| Bowel bag | Dmedian < 20 Gy | V40 < 30% | V45 < 195cc |  |
| Femoral heads | V35 < 15% | V20 < 55% |  |  |
| Pelvic bones | V10 < 90% | V20 < 75% | V30 < 50% | V40 < 37% |
| External genitalia | Dmedian < 15 Gy |  |  |  |

**Table 2 Patients characteristics *n* (%)**

|  |  |
| --- | --- |
| age  Median  Range | 63  32-84 |
| Gender  Male  Female | 3 (7.3)  38 (92.7) |
| Tumor stage  T1  T2  T3  T4 | 3 (7.3)  19 (46.3)  10 (24.4)  9 (22) |
| Nodal stage  N0  N1  N2  N3 | 16 (39)  18 (44)  6 (14.6)  1 (2.4) |
| Staging  I  II  IIIA  IIIB | 2 (4.8)  13 (31.8)  12 (29.3)  14 (34.1) |
| Smoke  Yes  No  Unknown | 18 (44)  7 (17)  16 (39) |
| Histology  SCC | 41 (100) |
| Chemotherapy  MMC + 5FU  MMC + Capecitabine  Capecitabine  None | 33 (80.7)  4 (9.7)  2 (4.8)  2 (4.8) |

SCC: squamous cell cancer; MMC: mitomycin–C; 5-FU: 5 fluorouracil.

**Table 3 Acute toxicity (41 patients analyzed) *n* (%)**

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
|  | **G0** | **G1** | **G2** | **G3** | **G4** | **G5** |
| Gu | 26 (63.4) | 11(26.8) | 4 (9.8) | / | / | / |
| Skin | 0 (0) | 10(24.4) | 29(70.7) | 2 (4.9) | / | / |
| Gi | 10 (24.4) | 17(41.5) | 11(26.8) | 3 (7.3) | / | / |
| Pain | 7 (17) | 22(53.6) | 12(29.4) | / | / | / |
| Hemato- logical | 31 (75.6) | 4 (9.8) | 3 (7.3) | 2 (4.9) | / | 1 (2.4) |

GU: Genitourinary; GI: Gastrointestinal.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Table 4 Late toxicity (40 patients analyzed) *n* (%)** | | | | |
|  | **G0** | **G1** | **G2** | **G3** |
| GU  (dysuria) | 38 | 0 | 1 (3.8) | 1 (3.8) |
| GI  (incontinence) | 29 | 6 (23) | 4 (15.4) | 1 |
| Genital tract female (atrophy of vaginal mucosa) | 37 | 3 (11.5) | 0 | 0 |

GU: Genitourinary; GI: Gastrointestinal.

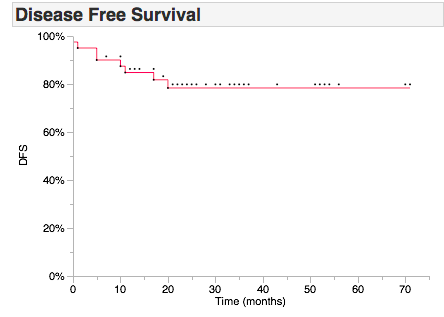
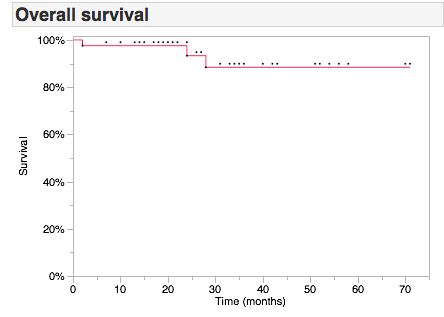
|  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Table 5 Comparison with acute toxicities data reported in literature** | | | | | | | | | | | | | |
|  | **N**° **pts** | **SIB** | **d/fx** | **CHT** | **Gastrointestinal** | | **Genitourinary** | | **Skin** | | **Hematologic** | | |
| **G2** | **G3** | **G2** | **G3** | **G2** | **G3** | **G2** | **G3** | **G4** |
| Pepek *et al*[26], 2010 | 29 | No | 1.8 | 89% | 76% | 10% | 45% | 3% | 100% | 0 | 45% | 24% | |
| Bazan *et al*[24], 2010 | 29 | Yes | 1.6-1.8 | 86% | \ | 7% | \ | \ | \ | 21% | \ | 21% | |
| Vieillot *et al*[28], 2012 | 72 | No | 1.8- 2.0 | 85% | 14% | 4% | 4% | 2% | 16% | 16% | 4% | 5% | 4% |
| DeFoe *et al*[11], 2012 | 78 | No | 1.8 | 98% | 60% | 27.7% | 18.5% | 0 | 91.3% | 29% | 51.4% | 42.9% | 12.9% |
| Kachnic *et al*[5], 2012 | 43 | Yes | 1.5-1.8 | 100% | 42% | 7% | 5% | 5% | 63% | 5% | 21% | 49% | 12% |
| Kachnic *et al*[13], 2012 | 52 | Yes | 1.5- 1.8 | 100% | 52% | 21% | 13.4% | 1.9% | 52% | 21% | 15.4% | 30% | 27% |
| Chuong *et al*[29], 2013 | 52 | Yes | 1.8- 2.0 | 100% | 39.5% | 9.6% | 26.9% | 0 | 57.7% | 11.5% | 37.8% | 28.8% | |
| Koerber *et al*[30], 2014 | 68 | Yes | 1.8-2.2 | \ | 47.1% | | 19.1% | | 63.2% | | \ | \ | \ |
| Current study | 41 | Yes | 1.8- 2.2 | 95% | 26.8% | 7.3% | 9.8% | 0 | 70.7% | 4.9% | 7.3% | 4.9% | \ |

N pts: patients number; d/fx: dose per fraction; CHT: Chemotherapy.

**Table 6 Comparison with outcome data reported in literature**

|  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
|  | **Bazan *et al*[24]** | **Chuong *et al*[29]** | **Dasgupta *et al*[31]** | **Koerber *et al*[30]** | **Mitchell *et al*[32]** | **Pepek *et al*[26]** | **Janssen *et al*[23]** | **Tozzi *et al*[22]** | **Current study** |
| N° pz | 29pz | 52 pz | 45 pz | 68 pz | 65 pz | 47pz | 25 pz | 36 pz | 41pz |
| **LC** | 92%2 | 91.3%2 | 87%1 | 75.3%2 | 90%1 | 90%1 | 92%1 | 78.1%3 | 92%1 |
| **OS** | 88%2 | 91.1%2 | 93%1 | 82.9%2 | 96%1 | 85%1 | 88%1 |  | 93%1 |

LC: Local control; OS: overall survival. 1Two years; 2Three years; 3Five years.

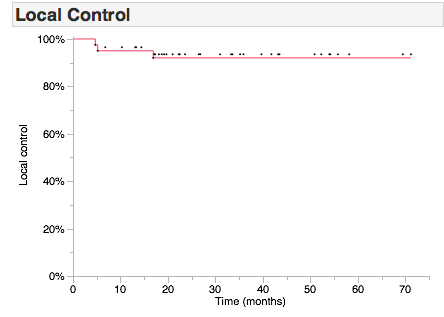
 A B

2 yy OS: 93%

4 yy OS: 88%

2 yy DFS: 78.3%

4 yy DFS: 78.3%

 C

2 yy LC: 92%

4 yy LC: 92%

**Figure 1 disease free survival (A), overall survival (B) and local control (C).**

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**A B**

**Figure 2 colostomy free survival.** A: Colostomy free survival (CFS) all disease stages; B: CFS stage I-II vs IIIA-IIIB.