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Title:

Treatment outcomes and survival following definitive (chemo) radiotherapy in HPV-positive oropharynx cancer: Large-scale comparison of DAHANCA vs PMH cohorts

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List of abbreviations (alphabetical):

AFRT: accelerated fractionated radiotherapy

aHR: adjusted hazard ratio

ASIR: age standardized incidence rate

BID: two times a day

CI: confidence interval

CRT: chemoradiotherapy

CSS: cause-specific survival

DAHANCA: Danish Head and Neck Cancer group

DFS: Disease free survival

DM: distant metastasis

GTV: gross tumor volume

Gy: Gray

HFCRT: hyperfractionated chemoradiotherapy

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HNSCC: squamous cell carcinoma of the head and neck

HPV: Human papillomavirus

IR: incidence rate

LF: local failure

LRF: locoregional failure

MVA: multivariable analysis

OPC: oropharynx cancer

OS: overall survival

OTT: overall treatment time

PMH: Princess Margaret Hospital Cancer Centre

PS: performance status

PTV: planning target volume

PY: pack years

QD: once a day

RF: regional failure

RT: radiotherapy

Article category: Cancer Therapy and Prevention

Novelty and Impact:

This study explores the variation in demographics, clinical behavior and outcomes in HPV+ OPC patients residing on opposite sides of the Atlantic Ocean, comparing two large-scale cohorts treated contemporaneously with primary RT/CRT. The data are informative in emphasizing the excellent outcomes from modern RT, and the impact of concurrent chemotherapy even in patients with a favorable prognosis. Our findings underscore the need for a very careful approach at efforts to de-intensify treatment for this disease.

Abstract

We compare outcomes in two large-scale contemporaneously-treated HPV-positive (HPV+) oropharynx cancer (OPC) cohorts treated with definitive radiotherapy/chemo-radiotherapy (RT/CRT). p16-confirmed HPV+ OPC treated between 2007-2015 at PMH and DAHANCA were identified. Locoregional failure (LRF), distant metastasis (DM), and overall survival (OS) were compared. Multivariable analysis (MVA) calculated adjusted-hazard-ratio (aHR) with 95% confidence interval (95% CI), adjusting for cohort, age, gender**,** performance status, smoking pack-years, T- and N-category and chemotherapy.

Compared to PMH (n=701), DAHANCA (n=1174) contained lower TNM-8 T-categories (T1-2: 77% vs 56%), N-categories (N0-N1: 77% vs 67%), and stages (stage I: 63% vs 44% (all p<0.001). PMH used standard-fractionation CRT in 69% (481) while 31% (220) received hypo-fractionated or moderately-accelerated RT-alone. All DAHANCA patients were treated with moderately-accelerated RT; 96% (1129) received Nimorazole (NIM) and 73% (856) concurrent weekly Cisplatin. DAHANCA had shorter overall-treatment-time (p<0.001), lower gross tumor (66-68 vs 70 Gy) and elective neck (50 vs 56 Gy) doses. Median follow-up was 4.8 years. DAHANCA had higher 5-year LRF (13% vs 7%, aHR=0.47 [0.34-0.67]), comparable DM (7% vs 12%, aHR=1.32 [0.95-1.82]), but better OS (85% vs 80%, aHR=1.30 [1.01-1.68]). CRT patients had a lower risk of LRF (aHR 0.56 [0.39-0.82]), DM (aHR 0.70 [0.50-1.00]) and death (aHR 0.39 [0.29-0.52]) versus RT-alone.

We observed exemplary outcomes for two large-scale trans-Atlantic HPV+ OPC cohorts treated in a similar manner. Concurrent chemotherapy was a strong, independent prognostic factor for all endpoints. Our findings underscore the need for a very careful approach to de-intensification of treatment for this disease.

Introduction:

The emergence of human papillomavirus-related (HPV+) oropharyngeal cancer (OPC) has changed the landscape and conventional understanding of the clinical behavior of squamous cell carcinoma of the head and neck (HNSCC)[1–3]. Compared to traditional HPV-negative HNSCC, HPV+ OPC represents a unique subgroup with very different epidemiology and patient risk-profile, molecular biology[4,5], and response to radiotherapy/chemo-radiotherapy (RT/CRT)[6,7]. The observed favorable prognosis of HPV+ OPC has led to the development of a separate staging criteria in the 8th edition TNM (TNM-8) to depict the prognosis of the disease more accurately[8]. Although variation exists regarding RT fractionation schedule (conventional 5 fractions/week vs moderately accelerated fractionation with 6 fractions/week) and concurrent chemotherapy schedule (weekly low-dose vs tri-weekly high-dose cisplatin), current standard of care for patients with node-positive HPV+ OPC is still concurrent CRT[9]. Excellent disease control but high toxicity burden with CRT has stimulated the design of new clinical trials to explore the possibility of deintensification for low-risk patients and more intensified treatment for higher risk populations. However, the first putative 'deintensification' strategy substituting cisplatin with EGFR-inhibitor failed to show improvement in toxicity and disappointingly showed higher locoregional failure (LRF) and worse survival in two phase III randomized trials[10,11]. Hence, caution is needed when selecting patients for treatment deintensification, and CRT remains the standard of care for many HPV+ OPC patients.

The global variation in the incidence of HPV+ OPC is considerable, and countries with high age standardized incidence rates of the disease are mainly located in North America and Europe[12]. Denmark and Canada represent jurisdictions with a high HPV prevalence among OPC, and both countries have shown a significant increase in incidence over the last

40 years[2,13,14]. People in Denmark and Canada have comparable access to healthcare and patients are treated within the frames of well-organized public health care systems. Moreover, the respective treatment guidelines are similar where RT/CRT is the main treatment for HPV+ OPC, albeit minor variations in RT schedule and doses, as well as usage of nimorazole (NIM), a hypoxic radiosensitizer.

These similarities in the treatment philosophy provide the opportunity to examine common features and differences in patient and tumor characteristics, and outcomes in two population-based cohorts of HPV+ OPC patients. This study aims to explore our understanding of variation in demographics, clinical behavior, and outcomes in HPV+ OPC patients residing on opposite sides of the Atlantic Ocean.

Material and methods

Patients and tissues

The Princess Margaret Hospital Cancer Centre (PMH) is the largest quaternary cancer center in Ontario, Canada, and represents one of only two cancer centers in the Greater Toronto Area (GTA) within a population of approximately 6 million inhabitants. PMH receives approximately 75-80% of OPC referrals in the region[14]. All PMH head and neck cancer (HNC) patients are managed according to institutional guidelines. All are registered in an institutional prospective Head and Neck Anthology of Outcomes database[15] where clinical information including outcomes is collected at point-of-care.

The Danish Head and Neck Cancer (DAHANCA) database is a nationwide clinical quality database that contains prospectively collected data since the early 1970s, and the registration of patients with HNC constitutes the backbone of the DAHANCA structure[13]. The population of Denmark is 5.8 million people and all HNC patients treated in Denmark are registered in the database. A specific comparison to the Danish Cancer Registry is performed at least annually, and the concordance between the two has been shown to be almost complete. This demonstrates that the DAHANCA database consistently maintains nationwide coverage of Danish HNC patients.

We identified patients with newly diagnosed HPV+ OPC treated with definitive RT/CRT between 2007-2015 **(Figure 1)**. Exclusion criteria were: p16-negative or p16-status unknown OPC, distant metastatic disease at presentation, primary surgery, non-curative RT (<60Gy), incomplete RT or follow-up information. To minimize variation in treatment regimens, we also excluded patients treated with epidermal growth factor receptor inhibitors and/or hyperfractionation RT**.** All patients were re-staged according to TNM-8 for this study, and TNM-8 classification is used throughout the study unless otherwise specified.

HPV-association was determined by p16 immunohistochemistry staining, an established surrogate for tumor HPV in OPC. Tumors were classified as p16-positive based on strong and diffuse nuclear or cytoplasmatic staining in >70% of tumor cells[16,17]. Smoking history was prospectively recorded by attending clinicians at first patient consultation in a similar manner at both PMH and DAHANCA. Smoking habits were defined as: (1) Never-smoker, (2) Former smoker (ceased smoking prior to the time of the first consultation), and (3) Current smoker (still smoking at time of the first consultation). Quantification of cumulative tobacco exposure (smoking pack-years) was based on patient reported number of cigarettes/day, years since smoking cessation and number of years that the patient had smoked. One pack-year (PY) was defined as years of smoking 20 cigarettes/day [7,18].

Treatment

The RT/CRT schedules and dose to various clinical targets for the PMH and DAHANCA cohorts are listed in **Table 1**. PMH patients were treated according to institutional guidelines as described previously[19]. TNM-7 stage I-II patients were treated with RT-alone. Stage III disease was treated with either RT-alone or CRT. Stage IV was treated with CRT. Where chemotherapy was otherwise indicated, RT-alone was reserved for the following situations: age above 70 years, frailty other than age, co-morbidity including renal/ hepatic/ cardiac/ hearing/ neural impairment, patient's choice and, minimal nodal disease (≤ 3 overt lymph nodes all under 3 cm in size) by physicians' choice. Predominantly, RT-alone comprised moderately accelerated fractionation (70 Gy in 35 fractions over 6 weeks, 2.0 Gy per fraction, 6 fractions per week) but a few patients (N=42) were treated with a hypofractionated schedule (60 Gy in 25 fractions over 5 weeks, 2.4 Gy per fraction, 5 fractions per week). Concurrent CRT included tri-weekly (100 mg/m^2) (preferred) or weekly (40 mg/m^2) cisplatin (when concerned about chemotherapy tolerance), delivered concurrently with 5-fraction-per-week RT to a total dose of minimum 70 Gy in 35 fractions over 7 weeks. No planned neck dissections were performed. Doses were delivered as simultaneous integrated boost where doses to elective neck were 56 Gy and 50 Gy for 70 Gy/35 fractions and 60 Gy/25 fractions regimens, respectively (**Table 1**). During the study period, the clinical goals during optimization were 95% of the PTV receiving 100% prescription dose and 100% of the PTV receiving 95% of the prescription dose, whenever achievable.

In the DAHANCA cohort, RT was applied according to the 2004 and 2013 DAHANCA radiotherapy guidelines[20], using moderately accelerated fractioned RT[21]. Accordingly, a tumor dose of 66–68 Gy (2 Gy per fraction, 6 fractions per week) was prescribed dependent

on tumor size, with larger tumors receiving the larger dose. Thus, patients who had a primary tumor and/or lymph node with a diameter of 4 cm or less, were given 66 Gy, and for tumors and/or nodes larger than 4 cm, a mean dose of 68 Gy was prescribed. Dose to elective neck was 50 Gy in 33-34 fractions, 6 fractions per week (**Table 1**). The primary tumor and lymph node volumes were delineated according to consensus guidelines[20], and dose prescription followed ICRU recommendations, where prescription dose is the mean dose to CTV1 with a 95% iso-dose coverage to 98% of the PTV[20]. To further intensify treatment for patients with nodal involvement, concurrent cisplatin was administered using a weekly (40 mg/m^2) schedule[9]. Chronological age was not a criterion for omitting chemotherapy according to DAHANCA guidelines; rather the decision was made based on evaluation of the performance status, frailty and co-morbidity (including impaired renal function) of the patients. A few DAHANCA patients (N=18) received concurrent carboplatin due to impaired renal function. Since completion of the DAHANCA 5 trial, hypoxic modification with nimorazole (NIM) has been standard of care for Danish OPC patients treated with primary RT/CRT[22], and accordingly almost all patients in the present DAHANCA cohort received NIM. No planned neck dissections were performed.

Follow-up

In both cohorts, patients were followed in multidisciplinary settings according to institutional protocols[23]. Treatment response was assessed at 3 months following RT/CRT. The frequency of regular follow-up was as follows: 3-monthly interval in the first year; 3-4-monthly in the 2nd year, 4-6 monthly in the 3rd year, and 6-monthly in the 4th and 5th year. Surgical salvage (primary tumor resection and/or neck dissection) in the case of biopsy-proven T-site and/or N-site failure was applied in a similar manner for both cohorts. Salvage

surgery was performed where feasible, but generally not undertaken in cases deemed unresectable, if synchronous distant metastases (DM) were present, or if the patient was medically unfit.

Statistical analysis

Actuarial rates of local failure (LF), regional failure (RF), locoregional failure (LRF), ultimate-LRF, distant metastasis (DM), and cause-specific survival (CSS) were calculated using competing risk methods where death without an event of interest was considered a competing risk. Disease free survival (DFS) and overall survival (OS) were calculated using the Kaplan-Meier method. Outcomes were measured from start of RT to the date of the event of interest. Events considered for LRF were local failure, regional failure or synchronous local and regional failure. Ultimate-LRF included initial LRF for cases without salvage and subsequent LRF after salvage; successfully salvaged initial LRF was not considered as an event in the ultimate-LRF estimation. Patients alive without the events of interest were censored at the date of last follow-up. DFS was defined as time to the first failure (ultimate-LRF or DM) or death from any cause. OS was defined as death from any cause. All outcomes were truncated at 5 years while events occurring after 5 years were censored at 5 years. Multivariable analysis (MVA) with Cox Proportional-Hazards regression was used to calculate adjusted hazard ratio (aHR) with 95% confidence interval (95% CI), adjusting for cohort, and covariates with accepted significance from clinical practice and previous trials , such as age, gender**,** performance status (PS), smoking pack-years, T- and N-category and chemotherapy. The Fine-Gray competing-risk regression was used for the MVA on LRF, ultimate-LRF and DM. All tests were two-sided and results were considered significant at levels less than 5%.

Results:

A total of 1875 patients were eligible including 1174 from DAHANCA and 701 from PMH. The cohort derivation is shown in **Figure 1**.

Baseline characteristics and treatment parameters stratified by cohort are shown (**Table 2**). No difference in median age was observed. Subdividing patients according to age categories revealed an almost identical age distribution between the cohorts: about 75% of the patients were diagnosed at age 51-70 years. Compared to the PMH cohort, the DAHANCA cohort had more women (24% vs 16%, p<0.001), better performance status (PS) (0: 81% vs 68%, p<0.001), and more heavy smokers (median smoking pack-years: 15 vs 10, p<0.001; >30 pack-years: 33% vs 21%, p<0.001).

The DAHANCA cohort contained a significantly lower proportion of T3-T4 (23% vs 44%, p<0.001) or N2-N3 (23% vs 34%, p<0.001) disease compared to the PMH cohort, ultimately yielding significant differences in stage II-III (39% vs 53%, p<0.001).

As all DAHANCA patients were treated with moderately accelerated RT (6 fractions/week) compared to 23% of PMH patients, overall treatment time (OTT) was significantly shorter in the DAHANCA cohort (median 38 days vs 46 days, p<0.001). Dose to gross tumor volume (GTV) was lower in DAHANCA (median 66.9 Gy vs 69.2 Gy, p<0.001) and only DAHANCA patients received NIM. Concurrent chemotherapy was used in approximately 70% of patients in both cohorts and mainly restricted to TNM-7 stage III-IV disease. Chemotherapy was omitted in 231 of 1083 (21%) TNM-7 stage III-IV DAHANCA patients (mainly due to frailty), and in 185 of 666 (28%) patients in the PMH cohort (mainly due to elderly age [n=55] and frailty [n=53]). Nineteen DAHANCA patients with node-negative disease received chemotherapy.

The median age was significantly lower in patients treated with CRT compared to RT-alone and only 25% of elderly (>70 years) patients were treated with CRT. In addition, performance status was significantly better in patients treated with CRT than RT-alone (**Supplementary table**).

Median follow-up in the total cohort was 4.8 years (0.2-5.0). At the time of evaluation, 181 patients experienced LRF: 134 (11%) from DAHANCA vs 47 (6%) from PMH (p<0.001), (**Figure 2A**); the difference remained significant in MVA (aHR 0.47 [0.34-0.67]), (**Table 3**). The frequency of LF was very low in both DAHANCA (4.5%) and PMH (2.6%) patients, yielding a 5-year LF rate of 5% and 3% (aHR=0.48 [0.28-0.81]), respectively. RFs were slightly more frequent and occurred in 7% and 5% of DAHANCA and PMH patients, respectively, with corresponding 5-year RF rate of 8% and 5%, respectively (aHR=0.51 [0.34-0.77]). Risk of LRF was associated with high T- and N-category **(Table 3)**. Only 4 patients with cN0 neck at baseline experienced a RF.

In the DAHANCA cohort, salvage surgery was successful in 54 patients with isolated N-site failure, 11 with T-site failure alone, and 2 patients with combined T- and N-site failure, yielding an ultimate-LRF of 6% at 5-years (64 ultimate failures) (**Figure 2B**). In the PMH cohort, salvage procedures were successful in 17 N-site failures, but in none of the isolated T-site failures or combined T- and N-site failures. The 5-year ultimate-LRF rate was 4% (28 ultimate failures) (**Figure 2B**). The ultimate-LRF remained higher in the DAHANCA vs PMH cohort in MVA (PMH vs DAHAHCA: aHR=0.53 [0.33-0.83]) after adjusting for T-, N-, PS, age, gender, smoking pack-years, and chemotherapy usage (**Table 3**).

In univariable analysis, DAHANCA cohort had a lower risk of DM compared to PMH (7% vs 12%, p<0.001), (**Figure 2C**), but no significant difference in DM rates could be

detected between the two cohorts after adjusting for confounders (aHR=1.32 [0.95-1.82]), (**Table 3)**.

Death occurred in 271 patients including 141 (12%) from the DAHANCA and 130 (18.5%) from the PMH cohorts (p<0.001). Causes of death were similar (p=0.31) for the DAHANCA and PMH patients: index cancer death: 86 (61%) vs 78 (60%); death from other cancers (mostly lung primaries): 16% vs 11%; death from other causes: 17% vs 26%; "unknown" cause of death: 6% vs 5% (**Figure 2D**). The 5-year disease-free survival rates were 81% in DAHANCA and 77% in PMH (p=0.003, **Figure 2E**) but DFS did not differ significantly between the two cohorts in MVA (aHR=1.19 [0.94-1.49], **Table 3**). The 5-year OS was 85% in DAHANCA compared to 80% in PMH (p<0.001, **Figure 2F**), which retained statistical significance in MVA (aHR=1.30 [1.01-1.68], **Table 3**).

Multivariable analysis revealed that patients receiving concurrent chemotherapy had significantly better prognosis than those treated with RT alone. The independent impact of chemotherapy was seen for all endpoints examined and resulted in a marked risk reduction for LRF (aHR=0.56 [0.39-0.82]), cancer-specific death (0.42 [0.33-0.54]) and overall death (0.39 [0.29-0.52], **Table 3**).

T- and N-category retained independent prognostic impact for all outcomes, and heavy smoking (>30 PY) increased the risk of LRF, cancer-specific death and overall mortality but did not affect risk of distant metastasis (**Table 3**).

To account for inherent selection bias where chemotherapy was often withheld for frail and/or elderly patients (PMH cohort), sensitivity analyses were performed confining to the subgroup (n=1478) of younger patients (≤70 years) with good performance status (Zubrod PS 0-1). Chemotherapy remained a significant prognostic factor for reducing risk of LRF (aHR=0.59 [0.39-0.89], p=0.012), ultimate LRF (aHR=0.45 [0.25-0.82], p<0.001), and

overall mortality (aHR=0.48 [0.34-0.68], p<0.001), but nonsignificant for cancer-specific death (aHR=0.70 [0.42-1.18], p=0.180) and DM (aHR=1.35 [0.77-2.36], p=0.29). T- and N-category remained significant for all endpoints (all p<0.01).

Discussion

This comparison of two large contemporary cohorts of patients with HPV-associated p16+ OPC treated with curative intent confirms the very favorable prognosis of this disease globally. Observable, but small differences in both LRF (favouring PMH) and OS (favouring DAHANCA) were shown between cohorts. Yet the effect sizes based on the absolute numbers of events were low, yielding a general impression of very comparable and acceptable results on both sides of the Atlantic Ocean. The observed differences are probably multifactorial including inherent differences in demographics, stage distribution at presentation, selection bias and modest variations in treatment and follow up strategies.

As pointed out in the introduction, Denmark and Canada represent areas with a high proportion (>70%) of p16-positivity/HPV-association among OPC[2,14]. However, the incidence rate (IR) of OPC is somewhat higher in Denmark[24] (Age Standardized Incidence Rate (ASIR) 2015: men: 8; women: 2)[25], compared to Canada (ASIR 2012: men 6.4; women:1.4) [26]. Moreover, in Ontario between 1993-2010 the IR of OPC is reported to be 4.3 (both genders)[27] and data based on OPC treated at PMH between 2000-2012 shows an IR of 3.1[14]. Consequently, this yields a basic difference between the two cohorts as DAHANCA represents a nationwide cohort, whereas the PMH cohort reflects the referral pattern in Toronto receiving 75-80% of referral in this metropolitan region. To some extent, this difference can also be deduced from the consort diagram in Figure 1, where the incidence of OPC in Denmark apparently is more than twice the incidence observed in the PMH-cohort.

Moreover, as we have restricted our analyses to the group of patients treated with curative intent, complete data on treatment and follow-up and known p16-status, we cannot ascribe the observed demographic differences between our cohorts to epidemiological causalities. Nevertheless, the demographic profile for DAHANCA and PMH exhibit some similarities and differences worthy of discussion.

Age distribution was close to identical in the two cohorts. Interestingly, the age group >70 was almost as large as the group of patients under 50 years, supporting previous observations that HPV+ OPC is also a relatively frequent disease of the elderly[28].

PMH patients were diagnosed with higher T- and N-category and consequently higher disease stage compared to DAHANCA patients. This raises the possibility of longer interval to diagnosis; however, our data are not suitable to interrogate this hypothesis. Delayed onset of RT negatively impacts outcomes[29,30]; documentation of tumor growth in a Danish population of HNSCC patients waiting for treatment[31] led to the implementation of a "fast track cancer referral program" in Denmark that has been mandatory since 2009. These "Cancer Patients Pathways" have demonstrated accelerated diagnostic processes, a reduction of waiting times from diagnosis to treatment, and less advanced disease[32]. While the Canadian Partnership Against Cancer, including the Canadian Association of Radiation Oncology (CARO), has established target wait times for radiotherapy as "within 14 days for >90% of patients"[33] since the 1990's and the province of Ontario has publicly posted real-time radiotherapy wait times, these initiatives have not addressed the lead-time to diagnosis, in contrast with the Danish mandate. We are not able to elaborate further on whether the observed differences in disease stage at diagnosis could be explained by this Danish initiative.

DAHANCA patients were heavier smokers[34], and heavy smoking (>30 PY) independently impacted DFS and OS in a negative way, which is in accordance with other reports for HPV+ OPC[7,35]. Whether the heavier smoking history in the DAHANCA cohort contributes to the observed LRF differences is uncertain. Previous studies on head and neck cancer have shown that current smoking may reduce radiotherapy efficacy by introducing hypoxia[18]. However, data on whether heavy tobacco usage alters HPV+ tumor biology is warranted to facilitate management of this population.

Differences in practice pattern between DAHANCA and PMH included treatment parameters, such as dose (including dose-prescription) to GTVs and dose and fraction size to elective volumes, hypoxic modification and fractionation schedule, all of which could also potentially contribute to differences in LRF. It is important to note that most LRFs were salvageable, especially in the neck, with a 6% difference in 5-year LRF (13% vs 7%) translating to only a 2% difference in ultimate-LRF (6% vs 4%) when factoring in salvage surgery. Compared to DAHANCA, PMH cohort may have approximately 8-10% higher dose to GTVs due to higher prescription dose and difference in prescription method. In addition, dose and fraction size to elective volumes were also higher in PMH, and investigation of the exact pattern of failures would be an obvious area of future research in order to shed light on the correlation between dose and anatomical/geographical location of recurrences[36]. In that context, it would also be valuable to examine potential differences in acute- and long-term morbidity between these cohorts, as higher doses are associated with increased risk of dysphagia, xerostomia and fibrosis[37].

Tumor cell hypoxia is a well-known adverse prognostic factor for HNSCC resulting in increased radio-resistance and impaired loco-regional control, and several attempts have been made over the years to modify hypoxia during radiotherapy in order to improve

outcome[38]. Since completion of the DAHANCA 5 trial hypoxic modification with Nimorazole has been standard of care for all Danish OPC patients treated with primary radiotherapy. Although p16+ tumors are known to harbor hypoxia[39,40] re-analysis of the trial revealed no apparent benefit in HPV+ tumors[22] and similar observations have been made for the hypoxic cell cytotoxin tirapazamine[41]. The lack of benefit from hypoxic modification in HPV+ OPC is probably attributable to the higher radiosensitivity[42], offsetting the effect of hypoxic modification. Whether integration of a hypoxic gene expression classifier[39] will enable prediction of outcome after hypoxic modification for both HPV+ and HPV- HNSCC is currently addressed in the ongoing DAHANCA 30 trial (NCT02661152).

Reducing overall treatment time by use of accelerated radiotherapy, thereby leaving tumor stem-cells with a shorter time interval for repopulation, is associated with improved overall and progression-free survival when compared to standard fractionation RT[43]. Analysis of the DAHANCA 6&7 trial using p16 as a stratification parameter demonstrated that the benefit of moderately accelerated RT (AFRT) seems to be independent of p16-status[21]. However, a subgroup analysis of the MARCH metaanalysis restricted to p16+ OPC with known smoking status did not indicate a differential response to AFRT relative to p16-status[35]. In the present study, all DAHANCA patients were treated with AFRT (6Fx/week) compared to only 23% of PMH patients, which resulted in a significantly different OTT between the cohorts. However, the shorter OTT did not translate into a benefit in favor of DAHANCA patients. This observation should be viewed in context, since most patients in our study received concurrent chemotherapy, which retained a very strong independent prognostic impact for all endpoints. Another important observation is that the improvement obtained with AFRT is predominantly a T-site effect with limited influence on nodal

control[43]. Since nodal involvement is a prominent feature in HPV+ OPC, this probably also contributes to a dilution of the acceleration effect[21].

Concurrent CRT is standard of care for advanced stage HNSCC and meta-analyses have shown that the addition of chemotherapy leads to a superior outcome relative to RT-alone[9]. A recent network meta-analysis based on individual patient data evaluating the role of chemotherapy (MACH-NC) and altered fractionation radiotherapy (MARCH) suggests hyperfrationated RT combined with chemotherapy (HFCRT) may be superior for the treatment of locally advanced HNSCC[44]. However, tumor HPV-status was unavailable for these analyses. We have observed a strong impact of concurrent chemotherapy on LRF risk reduction in the entire cohort as well as in the subgroup of young patients with good performance status, indicating that cisplatin is a potent radiation sensitizer. Thus, cisplatin-based CRT should remain standard of care in loco-regionally advanced HPV+ OPC. For patients with advanced disease stage who are not fit for chemotherapy, the use of hyperfractionated accelerated radiotherapy represents a treatment option, used by both PMH and DAHANCA. At PMH the schedule is applied as 64 Gy/40f/4w 10f/w BID, 6 hours apart (HARDWINS)[45]. In DAHANCA, the practice is to use moderately accelerated, dose escalated hyperfractionation 76Gy/56f/5,6w, 10f/w BID, 6 hours apart, and application of this schedule in loco-regionally advanced HPV+ OPC has demonstrated a favorable outcome (3-year LRF-rate of 7%) and no increased risk of morbidity compared to (C)RT[46]. These findings suggest hyperfractionation with dose escalation to be an attractive approach also for patients with HPV+ OPC, where intensification of treatment cannot be done by use of concurrent chemotherapy.

This manuscript reports a comparison between contemporaneous data from HPV+ OPC patients residing on opposite sides of the Atlantic Ocean. Inevitably, differences

between health system, patient stage, risk factors and comorbidity, and treatment factors limit the interpretation of our findings. Additionally, data describing the interval from first patient symptom to diagnosis, and about smoking during therapy were not retrieved. HPV status was based on p16 staining, although potential differences in HPV genotype distribution between both cohorts is unknown. Despite these limitations, these data are informative in emphasizing the excellent outcomes from modern RT/CRT, and the role of chemotherapy even in patients with a favorable prognosis.

In conclusion, we observed similar exemplary outcomes for these large population-based cohorts of patients with p16+ OPC treated with primary RT/CRT and contemporary RT techniques and doses. Concurrent chemotherapy was a strong, independent prognostic factor, and our findings underscore the need for a careful approach at efforts to de-intensify treatment for this disease.

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Conflict of Interest

Scott V. Bratman declares ownership and leadership of Adela and patents/licensing with Roche and Adela.

Anna Spreafico declares grant funding paid to the institution by Novartis, BMS, Symphogen, Merck, AstraZeneca/Medimmune, Surface Oncology, Bayer, Northern Biologics, Pfizer, Treadwell Therapeutics, GSK, Janssen Oncology, Roche, Regeneron, Array Biopharma.

Claus Andrup Kristensen declares being on the advisory board of MSK (outside the submitted work).

All other authors declare no conflict of interest.

Ethics Statement

Data on PMH patients was retrieved from the PMH institutional prospective Head and Neck Anthology of Outcomes database with PMH institutional board approval. In a similar manner, data was retrieved from the Danish Head and Neck Cancer Group (DAHANCA) database on Danish patients, with approval from relevant national Danish authorities.

Data Availability Statement

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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Figure legends

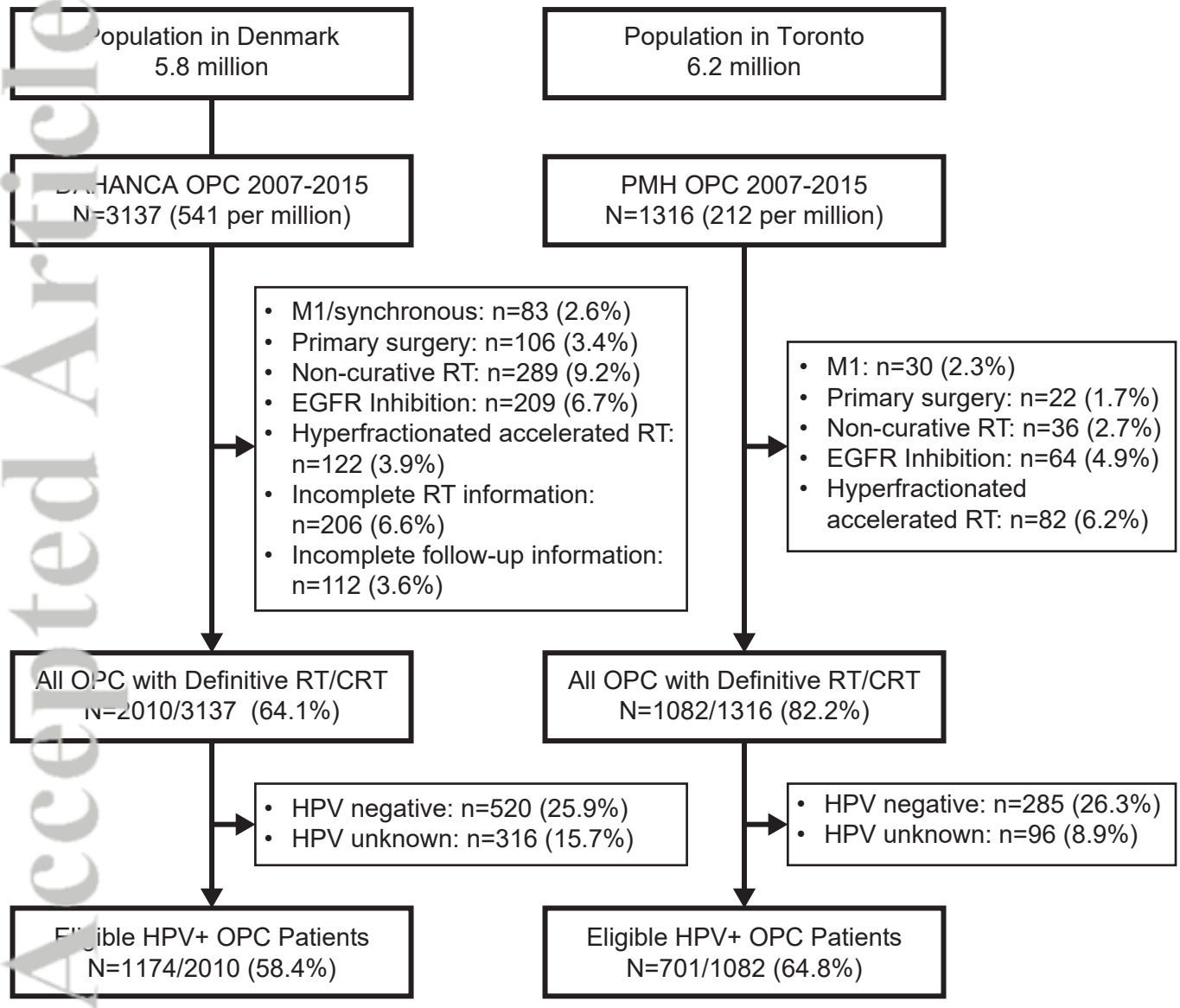
Figure 1: Consort diagram for OPC treated between 2007 to 2015

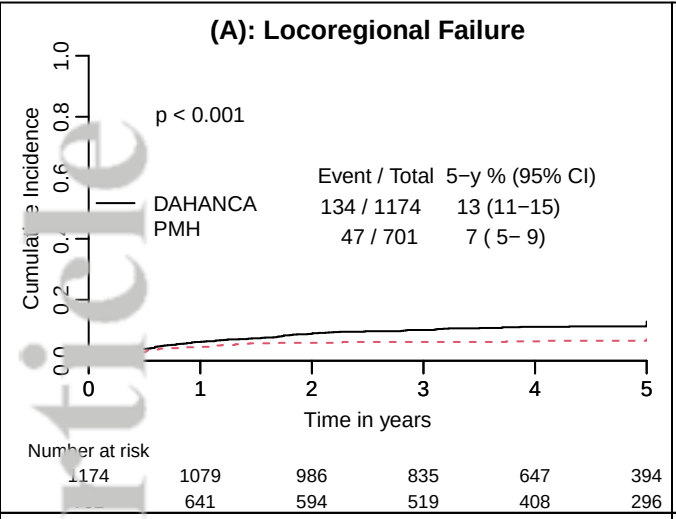
Figure 2. Outcome comparison by cohort

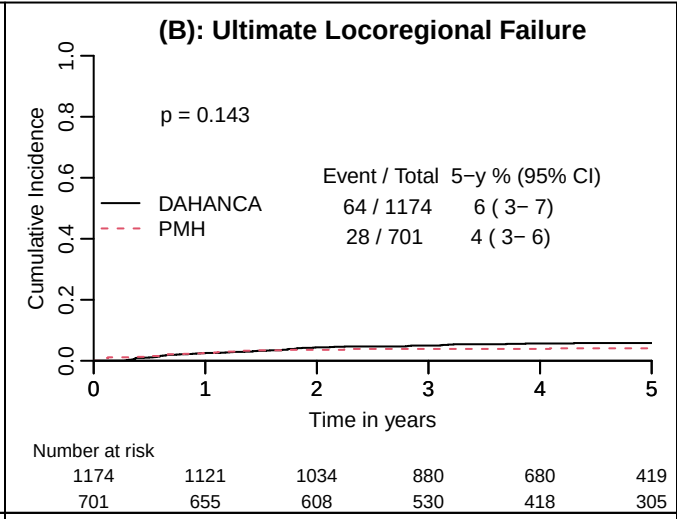
Actuarial estimated loco-regional failure (A), ultimate loco-regional failure (B), distant metastasis (C), competing mortality (death from other cancer [primarily lung], other and unknown causes) (D), disease-free survival (E), and overall survival (F)

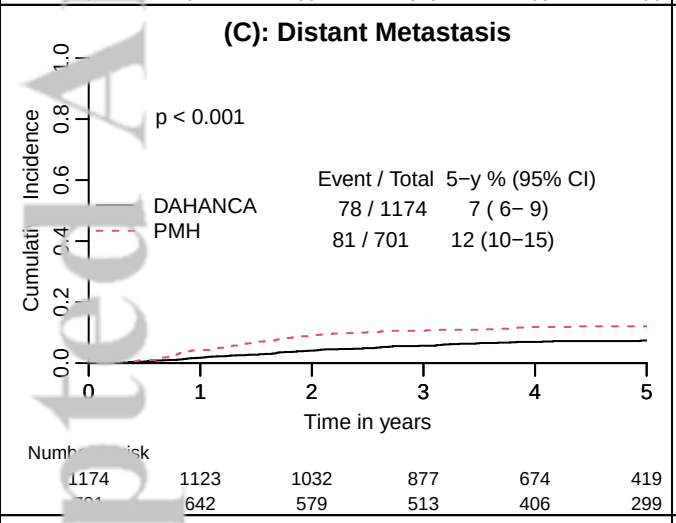
Novelty and Impact:

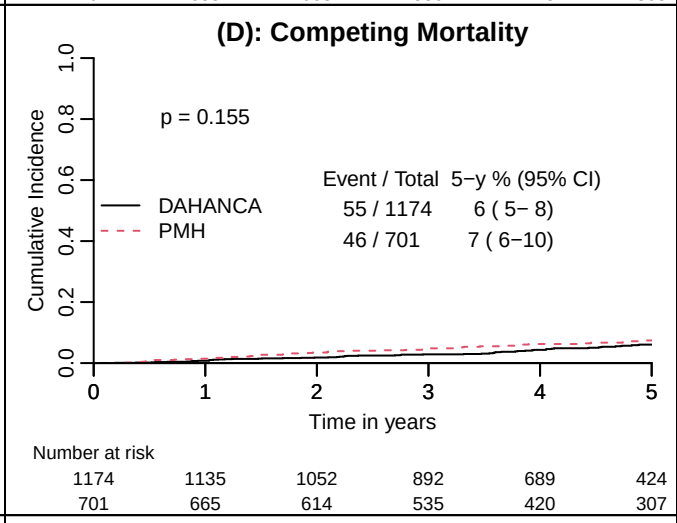
HPV-positive oropharyngeal cancer (OPC) represents a unique subgroup which has very different epidemiology, molecular biology, and response to radiotherapy/chemo-radiotherapy (RT/CRT) than standard squamous cell carcinoma of the head and neck (HNSCC). In this study, the authors compared two large cohorts, and found significantly better outcomes in the cohort that routinely received moderately-accelerated radiotherapy (RT) plus nimorazole/cisplatin chemotherapy. The authors conclude that these findings underscore the need for a cautious approach to efforts aimed at de-intensifying treatment for this disease.

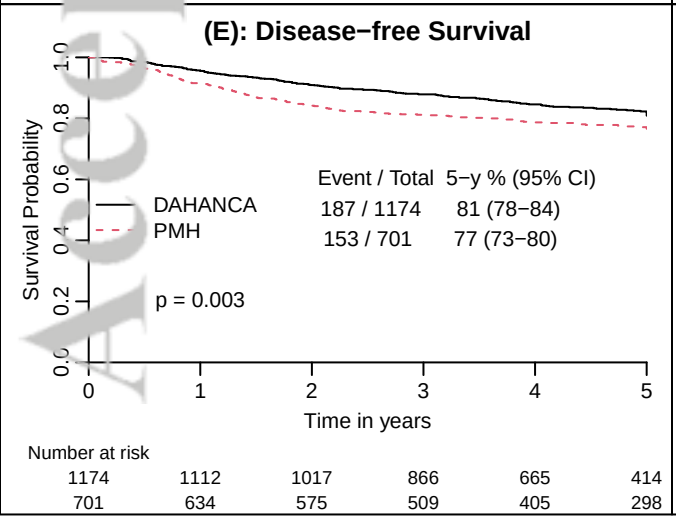












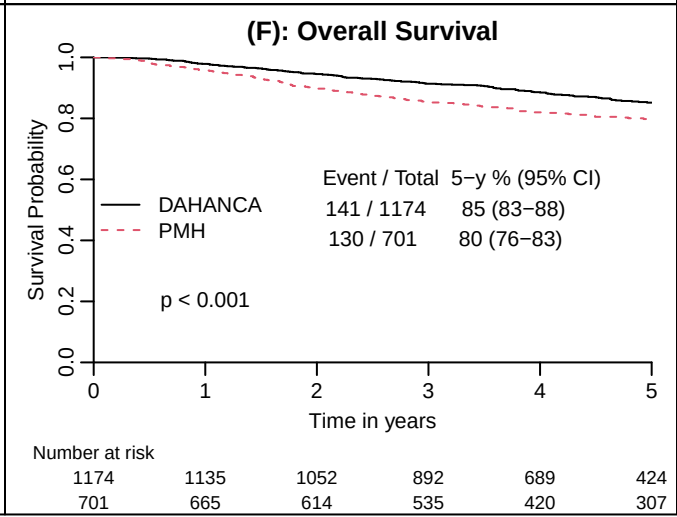


Table 1. Radiotherapy dose and fractionation schedules.

	PMH Radiotherapy Dose/Fractionation (Gy/f)		
	Hypofractionated radiotherapy alone	**Moderately accelerated radiotherapy alone**	**Chemoradiation**
RT Regimen	60 Gy/25f/5w,5f/w, QD	70 Gy/35f/6w, 6f/w, QD, BID once per week, 6 hrs apart	70 Gy/35f/7w, 5f/w, QD, with Chemotherapy
Dose to Gross Targets	60.0 Gy	70.0 Gy	70.0 Gy
Dose to Intermediate Targets*	56.0 Gy	63.0 Gy	63.0 Gy)
Dose to Elective Targets	50.0 Gy	56.0 Gy	56.0 Gy
Hypoxic Modification	-	-	-
Clinical Indication (TNM-7)	Stage I-III, or minimal nodal burden stage IV, favors in elderly patients	Stage I-III, or minimal nodal burden stage IV	Stage III-IV patients under 70 years old
	DAHANCA Radiotherapy Dose/Fractionation (Gy/f)		
	Hypofractionated radiotherapy alone	**Moderately accelerated radiotherapy alone**	**Chemoradiation**
RT Regimen	-	§66-68 Gy/33-34 f/6w, 6f/w, QD, BID once per week, 6 hrs apart	§66-68 Gy/33-34 f/6w, 6f/w, QD, BID once per week, 6 hrs apart
Dose to CTV1α	-	66-68 Gy/33-34f, 6 f/w	66-68 Gy/33-34f, 6 f/w
Dose to CTV2β	-	60.0 Gy	60.0 Gy
Dose to Elective Targets (CTV3)	-	50.0 Gy	50.0 Gy
Hypoxic Modification	-	Nimorazole	Nimorazole
Clinical Indication (TNM-7)	-	N0	N+

*Intermediate dose targets are ordinarily introduced to the treatment of radiologically suspicious lymph nodes for involvement of tumor.
** ≤ 3 malignant lymph nodes all under 3 cm in size (by physicians' choice)
§66 Gy to primary tumor and/or lymph nodes ≤ 4 cm, 68 Gy to tumors and/or nodes > 4 cm.
α Includes the primary tumor (GTV-T) and involved nodes (GTV-N) with an isotropic margin of 5 mm
β Includes the primary tumor (GTV-T) and involved nodes (GTV-N) with an isotropic margin of 10 mm

Table 2. Demographics, tumor characteristics and treatment by cohorts.

Covariate	Total cohort (n=1875)	DAHANCA (n=1174)	PMH (n=701)	p-value
Age				0.460
Mean (sd)	59.6 (8.9)	59.7 (7.7)	59.5 (9.14)	
Median (Min, Max)	59.2 (31.3,86.8)	59.5 (31.9,84.9)	58.8 (31.3,86.8)	
Age Category				0.670
<= 50	255 (14)	155 (13)	100 (14)	
51-60	749 (40)	463 (39)	286 (41)	
61-70	650 (35)	419 (36)	231 (33)	
>70	221 (12)	137 (12)	84 (12)	
Gender				**<0.001**
Female	391 (21)	279 (24)	115 (16)	
Male	1481 (79)	895 (76)	586 (84)	
Zubrod PS				**<0.001**
ECOG 0	1313 (76)	836 (81)	477 (68)	
ECOG 1	338 (20)	161 (16)	177 (25)	
ECOG 2	75 (4)	33 (3)	42 (6)	
ECOG 3	6 (0)	1 (0)	5 (1)	
Missing	143	143	0	
Smoking Status				
Never/former	567 (31)	328 (29)	239 (34)	0.099
Current	425 (23)	249 (22)	176 (25)	
Missing	30	30	0	
Smoking PY				**<0.001**
Mean (sd)	20.1 (23.6)	22.2 (25.6)	16.9 (19.8)	
Median (Min, Max)	14 (0,168)	15 (0,168)	10 (0,135)	
Missing	110	110	0	
PY-category				**<0.001**
0	567 (32)	328 (31)	239 (34)	
(0,10]	274 (16)	146 (14)	129 (18)	
(10,20]	227 (13)	120 (11)	107 (15)	
(20,30]	202 (11)	122 (11)	80 (11)	
+30	494 (28)	348 (33)	146 (21)	
Missing	110	110	0	
T-category (TNM-7)				**<0.001**
T1-T2	1295 (69)	900 (77)	395 (56)	
T3-T4	580 (31)	274 (23)	306 (44)	
N-category (TNM-7)				**<0.001**
N0	176 (9)	116 (10)	60 (9)	
N1-N2b	1194 (64)	790 (67)	404 (57)	
N2c	407 (22)	218 (19)	189 (27)	
N3	98 (5)	50 (4)	48 (7)	
Stage (TNM-7)				**<0.001**
I	31 (2)	27 (2)	4 (1)	
II	96 (5)	65 (6)	31(4)	
III	265 (14)	205 (17)	60 (9)	
IVA	1351 (72)	823 (70)	528 (75)	
IVB	132 (7)	54 (5)	78 (11)	
T-category (TNM-8)				**<0.001**

T1-T2	1295 (69)	900 (77)	395 (56)	
T3-T4	580 (31)	274 (23)	306 (44)	
N-category (TNM-8)				**<0.001**
N0	176 (9)	116 (10)	60 (9)	
N1	1194 (64)	790 (67)	404 (58)	
N2	407 (22)	218 (19)	189 (27)	
N3	98 (5)	50 (4)	48 (7)	
Stage (TNM-8)				**<0.001**
I	1041 (56)	735 (63)	306 (44)	
II	567 (30)	567 (30)	567 (30)	
III	267 (14)	105 (9)	162 (23)	
Subsite				**<0.001**
BOT	587 (31)	311 (27)	276 (39)	
Tonsil	1220 (65)	816 (70)	404 (58)	
Other	66 (4)	45 (3)	21 (3)	
Missing	2	2	0	
RT Dose/Fractionation				**<0.001**
RT alone (60 Gy/25f, 5f/w)	42 (2)	0 (0)	42 (6)	
RT alone (70 Gy/35f, 6f/w)	178 (9)	0 (0)	178 (25)	
RT alone (66-68 Gy/33-34f, 6f/w)	318 (17)	318 (27)	0 (0)	
CRT (70 Gy/35f, 5f/w)	481 (26)	0 (0)	481 (69)	
CRT (66-68 Gy/33-34f, 6f/w)	856 (46)	856 (73)	0 (0)	
Fractions per week				**<0.001**
5	542 (29)	1 (0)	541 (77)	
6	1333 (71)	1173 (100)	160 (23)	
OTT (days)				**<0.001**
Mean (sd)	40.8 (5.1)	38.1 (3.1)	45.2 (4.7)	
Median (Min, Max)	39 (23,97)	38 (28,97)	46 (23,76)	
Chemotherapy*				0.051
No	538 (29)	318 (27)	220 (31)	
Yes	1337 (70)	856 (73)	481 (69)	
Nimorazole				**<0.001**
No	746 (40)	45 (4)	701 (100)	
Yes	1129 (60)	1129 (96)	0 (0)	

Abbreviation: PS: Performance Status; PY: Pack years; OTT: overall treatment time; RT: radiotherapy; CRT: concurrent chemoradiotherapy; 60 Gy/25f, 5f/w: 60 Gy in 25 fractions, 5 fractions/week; 70 Gy/35f, 6f/w: 70 Gy in 35 fractions, 6 fractions/week; 70 Gy/35f, 5f/w: 70 Gy in 35 fractions, 5 fractions/week; 66-68 Gy/33-34f, 6f/w: 66-68 Gy in 33-34 fractions, 6 fractions/week.

* Chemoradiotherapy: In DAHANCA, it was administered as weekly low dose (40mg/ m^2) cisplatin combined with moderately accelerated radiotherapy 66-68 Gy in 33-34 fractions, 6 fractions/week; at PMH, it was generally administrated as three-weekly high-dose (100mg/m^2) cisplatin on days 1,22 and 43 (if fit) combined with conventional radiotherapy 70 Gy in 35 fractions, 5 fractions/week.
Frequency (percentage) are provided for categorical variables while mean (SD) and median (min, max) are presented for continuous variables. Non parametric Kruskal-Wallis tests are applied for comparisons of continuous variables while Chi Square tests are used for comparisons of categorical variables.

Table 3. Multivariable analysis for locoregional failure, distant metastasis, cancer specific death and overall death.

Variable	Locoregional failure HR [95% CI]	Ultimate locoregional failure HR [95% CI]	Distant failure HR [95% CI]	Cancer specific death HR [95% CI]	Overall death HR [95% CI]
Data source					
DAHANCA	Reference	Reference	Reference	Reference	Reference
PMH	**0.47 [0.34-0.67]**	**0.53 [0.33-0.83]**	1.32 [0.95-1.82]	1.19 [0.94-1.49]	**1.30 [1.01-1.68]**
Age					
<= 50	Reference	Reference	Reference	Reference	Reference
51-60	0.66 [0.42-1.03]	0.6 (0.32-1.13)	0.85 [0.51-1.42]	0.87 [0.6-1.26]	0.82 [0.54-1.24]
61-70	0.76 [0.49-1.19]	0.72 (0.39-1.33)	1.06 [0.64-1.75]	1.19 [0.82-1.72]	1.16 [0.77-1.75]
>70	**0.39 [0.20-0.77]**	0.49 (0.22-1.1)	1.48 [0.83-2.63]	1.14 [0.74-1.76]	1.06 [0.65-1.72]
Gender					
Female	Reference	Reference	Reference	Reference	Reference
Male	1.38 [0.91-2.08]	1.36 [0.76-2.44]	1.48 [0.95-2.3]	1.30 [0.97-1.73]	1.27 [0.91-1.76]
Zubrod PS					
PS: 0-1	Reference	Reference	Reference	Reference	Reference
PS: >1	1.40 [0.93-2.11]	**1.85 [1.11-3.07]**	1.40 [0.89-2.19]	**1.96 [1.46-2.64]**	**2.29 [1.66-3.15]**
Smoking PY					
0	Reference	Reference	Reference	Reference	Reference
(0, 10]	0.84 [0.48-1.46]	0.91 [0.42-1.95]	0.93 [0.55-1.56]	0.91 [0.62-1.33]	0.96 [0.62-1.50]
(10, 20]	1.02 [0.59-1.76]	0.9 [0.41-2.00]	0.98 [0.58-1.66]	0.85 [0.57-1.28]	0.99 [0.63-1.56]
(20, 30]	1.02 [0.58-1.81]	1.00 [0.45-2.24]	1.08 [0.61-1.91]	1.09 [0.73-1.64]	1.27 [0.80-2.00]
+30	**1.55 [1.05-2.29]**	1.53 [0.88-2.66]	1.27 [0.82-1.94]	**1.60 [1.20-2.12]**	**1.86 [1.34-2.59]**
T-category					
T1-T2	Reference	Reference	Reference	Reference	Reference
T3-T4	**1.89 [1.36-2.61]**	**2.45 [1.57-3.84]**	**2.25 [1.62-3.13]**	**2.67 [2.11-3.37]**	**2.81 [2.16-3.67]**
N-category					
N0	Reference	Reference	Reference	Reference	Reference
N1-N2b	1.85 [0.93-3.65]	2.55 [0.87-7.46]	2.11 [0.99-4.51]	1.34 [0.89-2.01]	1.36 [0.86-2.17]
N2c-N3	**2.85 [1.4-5.82]**	**4.66 [1.51-14.34]**	**3.16 [1.47-6.79]**	**1.86 [1.21-2.86]**	**1.91 [1.17-3.12]**
Chemotherapy					
No	Reference	Reference	Reference	Reference	Reference
Yes	**0.56 [0.39-0.82]**	**0.39 [0.25-0.62]**	**0.70 [0.50-1.00]**	**0.42 [0.33-0.54]**	**0.39 [0.29-0.52]**

Numbers in bold indicate statistical significance