Universitat de Girona
Facultat de Medicina

De-escalation of the adjuvant radiotherapy doses in patients with HPV-related oropharyngeal tumors with high levels of $CD8^+$ tumor infiltrating lymphocytes

A MULTICENTRE, RANDOMIZED, CONTROLLED CLINICAL TRIAL

FINAL DEGREE PROJECT

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1. ABBREVIATIONS

BOTSCC	Base of tongue squamous cell carcinoma
CT	Computed tomography
DNA	Deoxynucleic acid
E6	Early oncoprotein 6
E7	Early oncoprotein 7
EGFR	Epidermal growth factor receptor
HPV	Human papillomavirus
HNSCC	Head and neck squamous cell carcinoma
ISH	In situ hybridization
MRI	Magnetic resonance imaging
OPSCC	Oropharyngeal squamous cell carcinoma
PCR	Polymerase chain reaction
QT	Chemotherapy
Rb	Retinoblastoma
ROI	Region of interest
RT	Radiotherapy
SCC	Squamous cell carcinoma
TILs	Tumor infiltrating lymphocytes
TSCC	Tonsil squamous cell carcinoma

2. ABSTRACT

BACKGROUND: New epidemiological data indicate that the incidence of oropharyngeal squamous cell carcinomas (OPSCC) is rapidly increasing due to human papillomavirus (HPV). Patients that suffer from a HPV-related OPSCC present distinct characteristics such as younger age, early T-stage but more advanced N-stage, absence of tobacco and alcohol as risk factors, but, more importantly, a significantly better prognosis. Despite their better outcome, these patients are still being treated with the same protocols as not HPV-related tumors and thus, are exposed to the same toxicity and the potentially unnecessary side-effects should not be underestimated. Several protocols of treatment de-escalation are under study in subgroups of patients with different characteristics. In the current study, we will address patients with the best prognosis in order to limit the possibility of negatively affecting the oncologic control. For this purpose we will, incorporate $CD8^+$ tumor infiltrating lymphocytes (TILs) as an additional inclusion criteria.

OBJECTIVES: The aim of this study is to prove that de-escalation of the radiotherapeutic adjuvant treatment in patients with HPV-related OPSCC in early stages and displaying additional good prognosis factors, is non-inferior in terms of overall survival. As secondary outcomes we will address local recurrence and post-treatment swallowing dysfunction.

DESIGN AND SETTING: This is designed as a multicentre, randomized, open-labelled non-inferiority clinical trial. It will be performed among four tertiary hospitals of Catalonia.

PARTICIPANTS: patients aged between 18 and 60 years, with HPV-related OPSCC that have high numbers of $CD8^+$ TILs, are tributary to transoral robotic surgery and have a T-stage <3 and N-stage <N2c.

METHODS:

- Participants will be recruited consecutively and aim at 260 patients.
- We will assign randomly patients to two treatment groups: standard adjuvant treatment (A) and de-escalated treatment (B) while stratifying by the type of adjuvant treatment required (radio or chemo-radiotherapy) (N=65 patients in each subgroup).
- The de-escalation strategy will consist on reducing the protocol of adjuvant radiotherapy in 10 Gy. A follow-up of 5 years will be performed.
- Cox hazard ratio will be used for the statistical analyses, in which a confidence interval of 95% will be calculated and significance is defined as a bilateral $p<0.05$.

KEYWORDS: Human papillomavirus, oropharyngeal squamous cell carcinoma, de-escalated radiotherapeutic treatment, overall survival, post-radiotherapy dysphagia.

3. INTRODUCTION

3.1. BACKGROUND

3.1.1. GLOBAL BURDEN AND EPIDEMIOLOGY OF CANCER ATTRIBUITABLE TO HPV

Human papillomavirus (HPV) infection is recognized as one of the major causes of infection-related cancer in both men and women. In spite of most HPV infections not causing symptoms, likewise, resolving spontaneously, it has been demonstrated that persistent infection with high-risk types of HPV may lead to carcinogenic lesions.

Head and neck cancers (HNCs) include an heterogenous group that comprise tumors arising from the oral cavity, oropharynx, hypopharynx, larynx and sinonasal tract. These malignancies represent the seventh most common cancer worldwide. Nevertheless, the incidence varies considerable depending on the anatomic tumor site and geographic region. Histologically, 90% of the HNCs are squamous cell carcinoma (SCCs) (1)

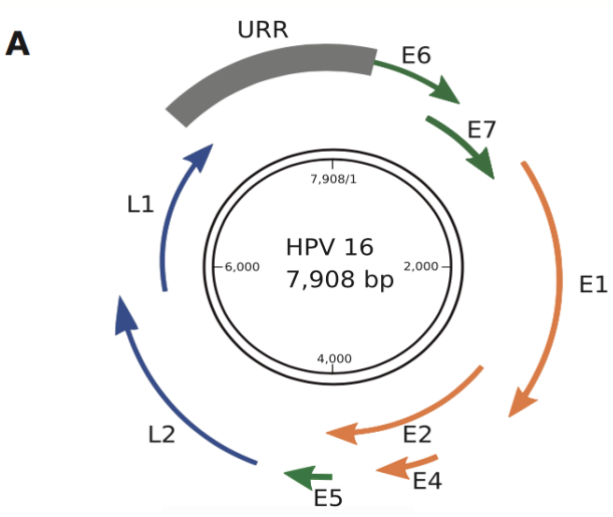
There are two major causes of HNCs: tobacco and alcohol and HPV (1,2). During the early 2000, Western countries defined and epidemic increase of the incidence of HNCs accounted mainly by the increase in oropharyngeal squamous cell carcinoma (OPSCC). This tendency was observed paradoxically while the smoking habit in these regions was declining. It was on the 2007 that HPV type 16 was recognized as a risk factor for OPSCC, besides smoking and alcohol consume. Later on, the worldwide epidemic increase on OPSCC was attributed to an increased incidence of HPV infection in the oropharyngeal anatomy. Not only HPV is and independent risk factor, but also HPV-related OPSCC are a distinct clinical-anatomopathological entity (3).

Approximately, 38.00 cases of HNCs are attributable to HPV. These cases have been described in three different anatomical sites: oral cavity, larynx, but to a greater extent oropharynx. Oropharynx tumors include tonsil, base of tongue walls of the oropharynx and soft palate. Nevertheless HPV presence is focused on tonsil (TSCC) and base of tongue (BOTSCC) (4). A not non-negligible 40%-60% of the OPSCC are attributable to HPV, depending on the geographic region (5). HPV type 16 is responsible for more than 80% of the HPV-positive OPSCC, while HPV18 is only present in 2% of the cases. The impact of the virus infection is not so big on the oral cavity and larynx, accounting for 4,400 and 3,800 cases per year respectively (6). Interestingly and opposite to cervical and other anogenital cancers, the incidence of HPV related OPSCC is greater in developed countries like North America and Europe (7).

3.1.2. BIOLOGY AND CARCINOGENESIS OF HPV

Nowadays, more than 200 genotypes of HPV have been identified. According to the International Agency of Research on Cancer (IARC) there are 12 types of HPV considered as high-risk types for their carcinogenic potential in humans, which are: types 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58 and 59 (8). A Common characteristic among all HPV types (high and low risk) is that they all have a capsid surrounding a circular double stranded DNA that can be divided in three major regions represented in **Figure 1** (3,9):

1) Early coding region that encodes up to six genes (E1, E2, E4-E7) that are responsible for the gene regulation and replication.
2) Late coding region which encodes for the L1 and L2 capsid proteins which self-assemble to form the virion.
3) Non-coding regulatory region (URR) that harbours the sites where transcription factors binds and also controls the gene expression



Locus	Function
URR	-Harbours transcription factor-binding sites -Controls gene expression
E6	-Promotes cellular immortalization by degradation of p53 -Modifies cell adhesion and differentiation by degradation of TAp63 and p73
E7	-Promotes pRb degradation, permitting cell progression to S-phase of cell cycle -Induces chromosomal instability
E5	-Induces cell proliferation -Contributes to evasion of apoptosis -Downregulates MHC expression
E1	-ATP-dependent DNA helicase activity -Role in viral DNA replication
E2	-Coactivator of viral DNA replication -Transcrition repressor of HPV E6 and E7 -Regulates cell cycle and apoptosis -Interacts with chromatin for segregation of viral genome
E4	Binds cytoskeletal proteins and disrupts cytoskeletal structure of the G2 arrested cell
L2	-Minor capsid protein -Recruits L1 protein for virus assembly
L1	-Major capsid protein -Can autoassemble into VLPs

Figure 1:

Figure 1. Schematic representation of HPV DNA genome.

On the left a schematic representation of HPV DNA genome. On the right a summary of the HPV gene functions. In green are the genes implicated in oncogenes. In orange those related to viral replication and in blue the viral capsid genes (9).

The common proteins among all HPV types are L1, L2, E1 and E2. By themselves they can fulfil the virus cycle. Nevertheless, in order to develop a carcinogenic action, presence of proteins E5, E6, and E7 is required. These proteins are considered oncogenes as they deregulate cell cycle control, as it is represented in **Figure 2**, and they are only present in the considered high-risk HPV types (3,10):

E6: several mechanisms have been demonstrated by which E6 inhibits the apoptotic response in infected cells. The most important ones are the following:

- Degradation of p53 by proteasomes. p53 regulates the expression of genes responsible for cell cycle regulation, DNA repair and apoptosis. In situations as hypoxia or DNA damage, p53 is the responsible to trigger either cycle arrest or apoptosis depending on the severity of the damage (3,10). When there is and HPV infection, E6 binds to a cellular protein called E6-associated protein (E6AP) forming a complex. The complex then binds to the central region of p53 causing its degradation. Consequently, cells with damaged or mutated DNA enter cell cycle without DNA repair resulting in the accumulation different mutations.
- Transcriptional activation of the human telomerase reverse transcriptase (*hTERT*): hTERT encodes the catalytic subunit of the telomerase complex. Its activation in the HPV16-infected cells causes a high telomerase activity, resulting in an unlimited proliferation (10).

E7: It is also implied in the deregulation of cell cycle by several mechanisms, the main one is here summarized:

- Degradation of the retinoblastoma protein (Rb). pRb function consists in the negative regulation of the activity of the E2F transcription factors, maintaining the cells in a quiescent state during the GO/G1 phase of the cell cycle.

  E7 is associated with the degradation of pRb causing the activation of E2F-regulated transcription, and therefore a progression of the cell cycle into synthesis phase (S-phase). This results in an over-expression of the cyclin-dependent kinase inhibitor $p16^{INK4a}$ (p16) that is used as a surrogate marker of the HPV infection (3,10,11).

E5: alongside E7, it downregulates de major histocompatibility (MHC) class I antigens, thus improving the virus escape form the immune recognition.

Continuous E6 and E7 expression is necessary for the maintenance of the malignant phenotype. A chronic infection by a high-risk HPV is needed for the development of malignant disease; in case of a rapid immune response and elimination of infected cells, no carcinogenic lesion would happen as there would be no time for the accumulation of chromosomal abnormalities. Beyond the action of E6 and E7 oncoproteins, other environmental factors and the host immune response also play a role in the chronicity of HPV infection (10).

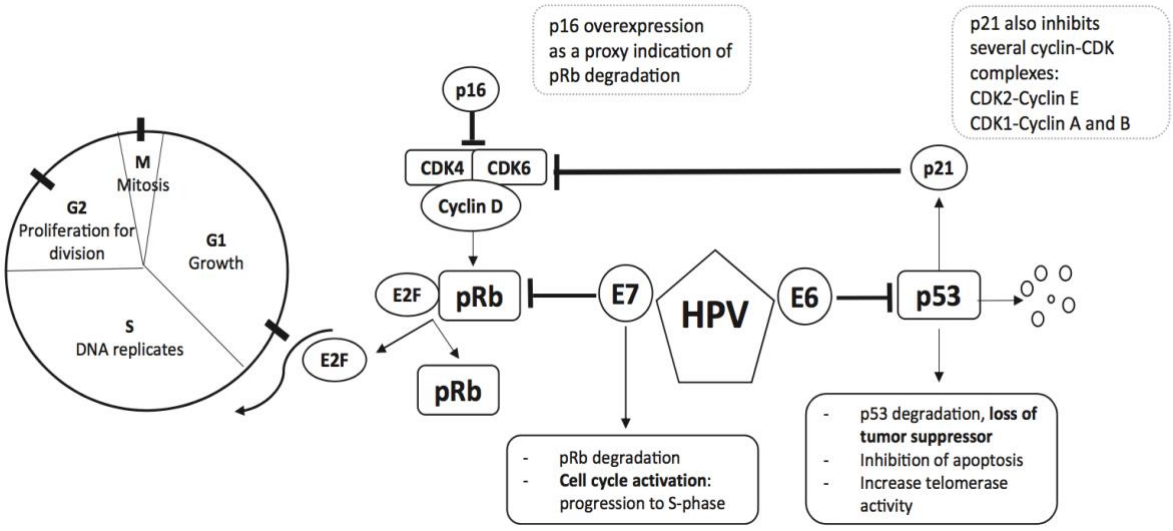


Figure 2 High risk HPV E6 and E7 oncoproteins and their role in cell cycle activation and loss of p53 tumor suppressor. (4)

3.1.3. HPV INFENCTION AND LIFE CYCLE

Most of the knowledge on the virus cycle has been obtained from studying HPV16 in the cervical cancer. HPV targets de undifferentiated keratinocytes in the basal layer of the stratified squamous cutaneous and mucosal epithelia. Entry into the epithelium can be either through micro abrasions or by entering cells of the single layered squamous cellular junction between the endo and ectocervix (9,12). The HPV replication cycle progresses intimately alongside the differentiation of this basal keratinocytes (12).

Regarding the carcinogenic cycle of HPV in oropharynx, there is still little knowledge, and it is mainly extrapolated from the cervical model; a scheme is showed in **Figure 3** (4,7). The mucosa of tongue and tonsils is invaginated forming crypts lined with reticulated epithelium. The crypts lining allows the entry of the virus and it is where HPV-positive tumors arise. On the other hand, HPV-negative tumors usually arise from the tonsil surface. A possible explanation for this anatomical differences might be that the below the continuous basal lamina of the crypts

there is a rich lymphoid stroma and an abundant intraepithelial network of blood vessels for which the papilloma virus might have some tropism (7).

It is suggested that likewise in the cervical cycle, the persistence of HPV infection can lead to a precancerous lesion that could eventually progress to invasive OPSCC. The estimated time between the infection and the development of invasive cancers in oropharynx is within 10 years, much shorter than in cervical cancer. It is also important to note that unlike cervix cancer, the precancerous lesion in OPSCC has not been yet identified (4).

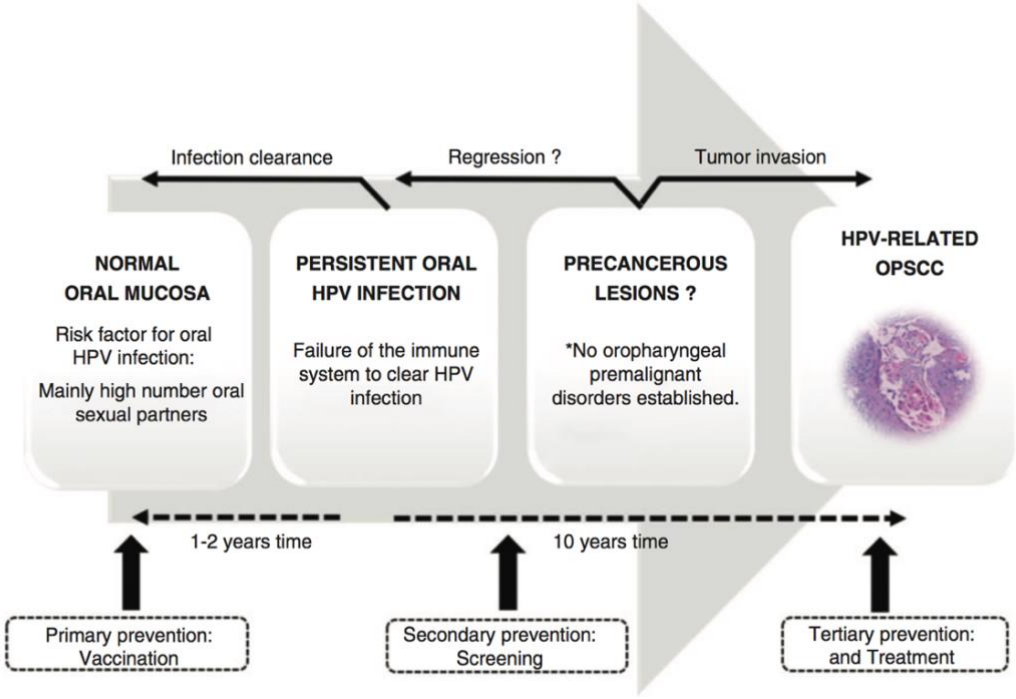


Figure 3: Natural history of HPV oral infection and the carcinogenesis process (4)

It is clear though, that immune response plays a role in the outcome of OPSCC. Nearby lymphoid tissue must lead to the presentation of HPV antigens to the immune cells, as there have been some cases of detection of antibodies against HPV oncoprotein E6 up to 10 years before diagnosis. Furthermore, cases with high numbers of tumor-infiltrating lymphocytes (TILs) have a better prognosis, representing a strong host immune response against the malignant lesion.

3.1.4. CLINICOANATOMOPATHOLOGIC DIFFERENCES OF THE HPV+ OROPHARYNX CANCERS

HPV infection has caused a complete shift of the head and neck neoplasia's paradigm, as patients that have an HPV-related tumor have several distinct characteristics versus those that have an HPV-negative OPSCC. These are detailed below and summarized in **Table 1**:

- Demographics: Both HPV-positive and HPV-negative HNSCCs have a higher incidence in men than women, but the ratio is even higher in HPV positive HNSCC (1). This is probably because of differences in the HPV oral infection between genders. *D'Souza et al* observed that men had a higher prevalence of oral HPV infection versus women, not explainable by the number of sex partners. Furthermore men also have a higher risk of persistence of HPV oral infection than women due to less clearance (13).

  Incidence is also directly related with age in both scenarios, but HPV-positive patients tend to be younger, up to 4 to 10 years. Also, and oppositely to HPV-negative, patients with HPV-positive HNSCC tend to be white and with a high socioeconomic status (SES). A possible explanation for this race difference might be the variance in smoking and sexual habits between races (1).
- Alcohol and Tobacco: They are recognized as the main risk factors for HNSCC. Tobacco consumption increases the risk of suffering from a HNSCC up to 5-10 times, depending on the anatomical site. Its carcinogenic effect was proved to be dose-dependent, being related to the duration and intensity of the smoking. Alcohol has an important role as a synergic cofactor when there is a combined consume of alcohol and tobacco. Most of the patients had history of use of either of both of them, but the increase of incidence of HPV has supposed a complete change.

  Nowadays, almost half of the patients with HPV-positive OPSCC have no history of alcohol or tobacco use. Nevertheless, tobacco still plays a role on HPV positive OPSCC as it has been seen that smoking habit is associated with a high oral HPV prevalence and persistence. Also patients HPV-positive who have smoking history have a poorer prognosis that those who do not (1).
- Sexual behaviour: Certain sexual behaviours are the main way of exposure to any type of HPV infection, and therefore it is an important risk factor for HPV-positive OPSCC but not for HPV-negative. Some of these behaviours are: high number of oral sex partners, high number of vaginal sex partners, younger age at sexual debut and history of genital warts (1,14,15).

- Anatomical site and clinical stage at presentation: Not only HPV-related head and neck cancers are more frequent in oropharynx, but rise in incidence of OPSCC is due to an increase of tumors in specific subsites such as tonsils and base of tongue (3,16).
  Characteristically HPV-related OPSCC usually have less advanced T-stage and more advanced N stages at presentation than HPV-negative tumors (17).
- Biological profiles: Oppositely to HPV-nonrelated HNSCC, HPV-positive OPSCC usually have not mutated p53, but deregulated or inhibited. They also have overexpression of $p16^{INK4A}$ and a low expression of EGFR (3,18,19).
- Prognosis: HPV is probably the most determinant prognostic factor for oropharyngeal cancer. In spite of patients with HPV-related OPSCC having higher N-stage, it has been seen that they have a better response to treatment, locoregional control and overall survival than patients with HPV-non related OPSCC (1,20,21). HPV-positive tumors are more sensitive to radiation thanks to p53 not being mutated, and thus maintaining its mediated apoptosis response in spite of the E6 oncoprotein actions (22).
  This better outcome is also due to the fact that HNSCC caused by tobacco and/or alcohol have a high rate of second primary, recurrent or synchronous tumors attributable to the carcinogenic mechanisms of these risk factors. (1)

Table 1: HPV negative compared with HPV positive head and neck cancers.

Adapted from Retting et al (1)

PARAMETER	HPV-NEGATIVE	HPV-POSITIVE
GENDER	Men > women ratio 2-3:1	Men > Women ratio 4-5:1
AGE	Median age late 50s-60s	Median age early mid 50s
RACE	Worse prognosis in blacks	High incidence in whites
SMOKING	>90% have smoking history; risk increases with increasing tobacco use	50-65% have smoking history
ALCOHOL USE	Synergistic with tobacco in increasing risk	Not a significant risk factor
SES	Lower	Higher
SEXUAL HISTORY	Not a significant risk factor	Number of oral sex partners is a strong risk factor
SITE	Larynx and oral cavity most common. Others include: oropharynx, hypopharynx, nasopharynx, nasal cavity and paranasal sinuses	Oropharynx, especially lymphoid tissue of tonsils or base of tongue. < 20% HPV-positivity at other HNC sites
PRESENTATION	Varies	Early T-stage, advanced N-stage Others: oropharyngeal pain, dysphagia and referred otalgia
INCIDENCE TRENDS	Decreasing	Increasing
MOLECULAR CHANGES	TP53 mutated High EGFR expression	TP53 wild type No overexpression of EGFR p16 positive
SECOND PRIMARY TUMORS	Less common	More common
PROGNOSIS	Oropharynx: - 5-year survival 30-35% - 5-year recurrence: 50%	Oropharynx: - 5-year survival: 60-90% - 5-year recurrence: 10-15%

3.1.5. DIAGNOSIS OF HPV RELATED TUMOR

It is important to have a reliable method not only to detect HPV in OPSCCs but also to discern if it is a transitory infection or a carcinogenic factor. It is not only relevant to its prognostic implications, but also for future perspectives seeking the introduction of de-escalation therapies for patients with good prognostic characteristics, such as HPV status. When studies of HPV prevalence in head and neck cancers show different results, it is partially due to the material and methodology used (3). The main difference between strategies are their detection targets, which include a wide range: HPV DNA, HPV RNA and viral oncoproteins (11). These are explained below and summarized in **Table 2** (4,11,23).

I. Routine microscopic evaluation: HPV-related HNSCCs have some distinctive microscopic characteristics that if detected can facilitate and/or guide the interpretation of the HPV detection assays. HPV-related tumors consistently arise from tonsillar crypts and tumor nests are often surrounded by lymphoid cells.

II. $p16^{INK4a}$: it is the most widely employed technique in the clinical practice. $p16^{INK4a}$ has been used as a surrogate marker of HPV involvement, when compared to the gold-standard it has a sensitivity of > 90% and a specificity around the 80%. On the other hand, frequently in HPV-negative HNSCC have a loss of p16 expression through an homozygous deletion or mutation.

It is technically easy and the interobserver variability is low. To consider this test positive it is required that >70% of tumor cells show nuclear and cytoplasmatic immunostaining.

Using this method alone to diagnose HPV-related HNSCCs is debatable as some of the not related HPV tumors confirmed by other techniques as mRNA and HPV-DNA, still show diffuse staining for $p16^{INK4a}$. The rate of discordant results between $p16^{INK4a}$ and E6/E7 mRNA is around 25%. Thus, $p16^{INK4a}$ is not entirely specific of HPV activity. As a result, if we were solely based on this, we would classify and stage differently some patients who in fact are HPV-negative and therefore have an intrinsic poorer prognosis.

III. HPV-DNA polymerase chain reaction (PCR): it uses a wide variety of HPV type-specific primers. It is a method with a high sensitivity for the presence detection of HPV. However, it does not discern HPV "passage" infections from those causatives of OPSCC. Thus, its specificity is low.

IV. HPV-DNA detected by in-situ hybridization (ISH): It allows the visualization of DNA within the tumor cells. A punctate nuclear pattern is specific for integration of viral DNA in the host genome, making this a very specific technique. It might be preferable to DNA PCR detection, as ISH can be performed in formalin fixed and paraffin embedded (FFPE) tissues. On the other hand, sensitivity is lower as it is limited by the viral copy numbers, which in some cases can be low.

V. E6/E7 HPV-mRNA detection by PCR: It proves that HPV is not only present but also transcriptionally active for its main oncogenes E6 and E7. Thus, it proves HPV causality and therefore it is considered the gold standard. Unfortunately, it is a technically challenging technique that requires the extraction of RNA from fresh or frozen tissues followed by polymerase chain reaction (PCR) amplification of the viral RNA.

VI. E6/E7 HPV-mRNA evaluation by ISH: it is a technique recently developed, that allows the detection of E6/E7 mRNA transcripts in FFPE tumor tissues. In comparison to HPV-DNA

ISH it allows to detect transcriptionally active form of the HR-HPV, and it also has a higher sensitivity. Unfortunately, it has not been approved for clinical use yet.

VII. Multi-modality HPV analysis: Is based on performing a more HPV-specific detection assay (such as HPV-DNA ISH or PCR) in those patients with a previous $p16^{INK4a}$ positive result. This would correct the false positives included in the positive results of the $p16^{INK4a}$ immunostaining. Its relevance is also based in the fact that some studies (24) showed significant differences on survival between patients with discordant results ($p16^{INK4a}$ positive but HPV-DNA negative) and those with positive results in both tests, having the last group a significant better survival. When it comes to possible de-escalation strategies, no error can be made when it comes to detection of real HPV-related neoplasia.

Nowadays the current practice in some of Catalunya hospitals is that if $p16^{INK4a}$ has an intermediate result (50-70% stained cells) they proceed to a HPV-DNA PCR test.

Table 2: Comparison between the different HPV detection assays in HNSCC.

Adapted from (4,23,25)

	DESCRIPTION	ADVANTAGES	DRAWBACKS
MICROSCOPIC STUDY	Study of the tumor tissue on the microscopic	Inexpensive Serves as a base for interpretation of further HPV tests	Not sufficiently specific to be used alone
P16 IMMUNOSTAINING	Surrogate marker for E7 HPV	Easy and most implemented technique on clinical setting Available commercial kit with CE certification High sensitivity, adequate for screening. Available for FFPE tissues	Moderate specificity. No exclusive link between HPV infection, p16 overexpression and carcinogenesis.
HPV-DNA PCR	Amplification of conserved regions of the HPV genome	PCR technique used is available on multiple hospitals HPV type specific with high sensitivity Feasible on FFPE material	Low specificity Requires DNA extraction Easy contamination
HPV-DNA ISH	Detection and location of HPV in tissue or cells	High specificity Available commercial kit with CE certification Provides visual detection of infected cells Feasible on FFPE material	Low sensitivity when low viral load Technically difficult interpretation
HPV E6/E7 MRNA PCR	Detection of messenger RNA E6/E7 HPV HPV type-specific	Considered Gold-standard High sensitivity and specificity Detects active HPV oncogene transcription	Technically challenging Difficult to reproduce on a clinical setting Requires fresh or frozen tissue.
HPV E6/E7 MRNA ISH	Detection of messenger RNA E6/E7 HPV HPV type-specific	High sensitivity and specificity despite quantity of viral load Feasible in FFPE material	Novel technique, limited available data Not approved for clinical use
MULTIMODAL ANALYSIS	p16 and another method	Allows for combining the high sensitivity of p16 with a more specific method	Increased cost

3.1.6. MANAGEMENT: PRESENT INTERVENTION AND FUTURE DE-ESCALATION?

CHANGE IN STAGING

In oncology an important indicator for choosing the treatment is the TNM staging system. The increase in incidence of HPV-related OPSCC and these cases having a better response to treatment with an improved prognosis has led to reassess the staging of these patients and evaluate new treatment approaches.

The American Joint Committee on Cancer (AJCC)/Union for International Cancer Control (UICC) developed two TNM staging systems depending on the HPV tumor status. In **Annex 1** is described the TNM for OPSCC HPV-related determined by p16 immunostaining. In **Table 3** below it is shown a comparison between the TNM staging in the last UICC edition (8th) for HPV-related OPSCC and the previous edition where there was no stratification based on the HPV status.

Table 3: Main differences between the 7th and the new 8th edition for TNM staging system classification for HPV-related OPSCC developed by the UICC (4)

CHARACTERISTICS	7TH EDITION TNM	8TH EDITION TNM ICON-S
STAGE CLASSIFICATIONS	**Stage I** (T1N0) **Stage II** (T2N0) **Stage III** (T3N0 or T1-T3N1) **Stage IVa** (T4aN0-1 or T1-T4aN2) **Stage IVb** (T4b or T1-T4bN3) **Stage IVc** (M1)	**Stage I** (T1-T2 N0-N1) **Stage II** (T1-T2N2 or T3N0-N2) **Stage III** (T4 or N3) **Stage IV** (M1)
MAIN N (LYMPH NODES) DIFFERENCES	**N1**: metastasis in a single ipsilateral lymph node, <3cm **N2a**: metastasis in a single ipsilateral lymph node > 3cm but < 6 cm **N2b**: metastasis in multiple ipsilateral lymph nodes, < 6 cm **N2c**: metastasis in bilateral or contralateral lymph nodes < 6 cm	**N1**: ipsilateral metastasis in lymph node(s), < 6 cm **N2**: bilateral or contralateral metastasis in lymph node(s), < 6 cm*
MAIN T (TUMOR) DIFFERENCES	**T4a**: tumor invades larynx, extrinsic muscle of tongue, medial pterygoid, hard palate or mandible **T4b**: tumor invades lateral pterygoid muscle, pterygoid plates, lateral nasopharynx, skull base or encases carotid artery	**T4**: tumor invades any of the following: larynx, deep/extrinsic muscle of tongue, medial pterygoid, hard palate, mandible, lateral pterygoid muscle, pterygoid plates, lateral nasopharynx, skull base or encases carotid artery **

*Because 5-year OS was similar among N1, N2a, and N2b, they re-termed the N categories
**Because 5-yers OS was similar among T4a and T4b, they were no longer subdivided and it was re-termed as T4

ACTUAL MANAGEMENT AND TREATMENT OF HPV-RELATED OPSCC

Nowadays when an oropharyngeal neoplasia is detected, the following complementary test need to be performed (26):

- Tumor human papillomavirus testing by p16 immunohistochemistry (IHC)
- Complete head and neck exam: mirror and fibreoptic examination as clinically indicated
- Biopsy of primary site or fine-needle aspiration of the neck
- CT with contrast and/or MRI with contrast of primary and neck
- As clinically indicated:
    - Preanesthetic studies
    - FDG-PET/CT
    - Chest CT is recommended for advanced nodal disease to screen for distant metastases, and for select patient who smoke to screen for lung disease
    - Nutrition, speech and swallowing evaluation

If p16 immunochemistry has a positive result, results from clinical and imaging exploration will be classified to the corresponding TNM staging for HPV+ oropharyngeal neoplasia explained above. The initial standard treatment for OPSCCs at present is mainly dependent on the stage of the disease and also patient/clinician preferences. Below are developed the standard treatment possibilities for early tumor stages, that are the ones included in this study (26).

- For early stages such **as T1-T2 N0-N1** (if single node < 3 cm) a single-modality treatment will be performed. This can be:
    - Definitive fractionated radiotherapy (RT) from 66 Gy (2.2 Gy/fraction) to 70 Gy (2.0 Gy/fraction) daily in 6-7 weeks. Alternatives for definitive RT is a concomitant boost accelerated RT or an hyper fractionated RT.
    - Resection of primary tumor with either ipsilateral or bilateral neck dissection if considered necessary. If anatomopathological exam after surgery shows any adverse features adjuvant treatment is indicated. The preferred interval between resection and postoperative radiotherapy is less than 6 weeks.

      Adjuvant RT is indicated if anatomopathological features are any of the following: pT3 o pT4 primary, one positive node > 3 cm, perineural invasion, vascular invasion, lymphatic invasion

      Adjuvant systemic therapy with concurrent RT is indicated if extranodal extension or positive margins (<1mm) are present and re-resection is not

feasible. The chemotherapeutical agent used is cisplatin, with RT at doses of 60-66 Gy (2 Gy/fraction) in 6 weeks.

- **For T1-T2 N1** (if single node > 3 cm, or 2 or more ipsilateral nodes < 6 cm) or **T1-T2 N2** or **T3 N0-N2** there are also several available options
    - Concurrent systemic therapy and RT: there are many treatment patterns, but the one usually used is cycles of cisplatin and 5-FU with concurrent definitive radiotherapy of 70 Gy (2 Gy/fraction) in 6-7 weeks
    - Resection of primary and ipsilateral or bilateral neck dissection. Site of neck dissection (unilateral or bilateral) will depend on the clinical nodal stage.

      Adjuvant sole radiotherapy should be considered if other risk features are present such as neural, vascular and lymphatic invasion.

      Adjuvant systemic therapy and radiotherapy should be considered if extranodal extension and/or positive margin are present
    - Induction chemotherapy followed by RT or systemic therapy/RT

DE-INTENSIFICATION STRATEGIES

An evident problematic arises: as discussed, patients with HPV-related OPSCC have a better outcome in comparison to those who have an OPSCC caused by tobacco and alcohol. Nevertheless, they still receive the same treatment protocols, and therefore they are susceptible to the same side effects. Most likely they could receive less intensive treatment, in order to develop fewer toxic effects without a detriment of the oncological outcomes.

There are several ongoing clinical trials, within them there are many de-escalation options being evaluated in different TNM OPSCC stages. Some of these are listed below (27), but given the aims and objectives of this work, we will focus on commenting on the last one:

1) **Radiation combined with cetuximab instead of cisplatin**
2) **Induction chemotherapy followed with decreased radiation doses and/or volumes for good responders**
3) **Radiation alone instead of chemoradiation**
4) **Transoral surgery followed by less intensive adjuvant protocols:**

   In 2009 the FDA approved the use of transoral robotic surgery for T1 and T2 oropharynx tumors. Its introduction was an opportunity to spare patients from radical high-dose radiation or concurrent chemoradiation.

   It seems logical that if HPV-related OPSCC are more sensitive to radiotherapy, then less doses should be necessary to obtain the same control. Another ongoing studied option is to remove chemotherapy from the adjuvant treatment.

   There are some ongoing trials studying these types of de-escalation (28–30).

3.1.7. INFILTRATING CD8⁺ LYMPHOCYTES: A POTENTIAL ADDITIONAL PRONOSTIC BIOMARKER FOR FURTHER STRATIFICATION AND DE-ESCALATION

When investigation on de-escalation possibilities started, many people were reasonably concerned by the potential risks of oncologic failure. The most important ethic principle is to first not harm the patient; it is therefore completely necessary from our point of view to select the lowest risk patients in order to limit as much as possible the possibility of negatively affecting the oncologic control.

There has been a lot of investigation in search of additional relevant prognostic factors besides the HPV status in order to stratify further more these patients in terms of prognosis. An interesting one, the one that will be included in our study as an inclusion criteria, are tumor infiltrating lymphocytes (TILs), specifically $CD8^+$. Number and types of tumor infiltrating lymphocytes have previously been shown to be a favourable prognostic factor in other cancer patients such as: colon lymphoma and breast (31,32). High TILs represent the organism immune response combating the neoplasia.

Nordfors et al (33) studied the infiltration of $CD8^+$ and $CD4^+$ TILs in the pre-treatment biopsies of 385 patients who had been previously diagnosed and treated for TSCC or BOTSCC of whom a total of 220 were HPV+ by $p16^{INK4A}$ and HPV-DNA PCR test; TILs could only be evaluated in only 216. They observed that HPV-related OPSCC had significantly higher numbers of $CD8^+$ TILs than HPV-negative tumors. In addition, they divided patients into quartiles according to the number of $CD8^+$ TILs and saw that there was a significant difference between the three groups of higher TILs (values higher than 13.2 per ROI [$3.5*10^5$ mm^2]) with the lowest quartile group regarding 3-year overall survival (approximately 90% vs 65% respectively) and 3-year disease free survival (approximately 85% vs 70% respectively). This was further investigated in other studies that showed similar results (34–36).

These studies show that $CD8^+$ TILs are a strong prognostic factor that can potentially be used as a stratification criteria in order to identify patients with a better initial prognosis, which is what we will realize in this study.

In addition to this supplementary prognostic factor, it is crucial to correctly identify HPV-related patients. As explained before, p16 immunohistochemistry is not a completely specific marker. This is the reason why in our study we will only de-escalate treatment on those patients with p16 positive immunostaining confirmed by an additional HPV-DNA test.

3.1.8. COMPLICATIONS RELATED TO ADJUVANT TREATMENT: DYSPHAGIA

DEFINITION

The difficulty in the transport of endogenous secretion our any aliment is the symptom known as dysphagia. In the oropharynx, the upper respiratory and gastrointestinal tract are in intimate contact. When any substances entry the airway instead of the upper gastrointestinal tract is called aspiration, which is a sever and not so uncommon complication of the dysphagia.

To understand the physiopathology of the dysphagia it is important to mention some basic notions of the normal swallowing process. It is composed of 3 phases depending on the anatomic regions.

- In the **oral phase** the preparation of the alimentary bolus happens. This mainly depends on a correct salivation and muscle coordination for its propulsion. This phase is under voluntary control.
- The **pharyngeal phase** is composed by all the processes required for the bolus arriving from the isthmus to the superior oesophageal sphincter. It is the most delicate phase as it is where the aerodigestive crossroads happens.
- Lastly, **oesophageal phase** finishes when the bolus reaches the stomach through oesophageal peristalsis.

All this process is under a complex neurologic regulation: the afferents travel through the glossopharyngeal (IX) and the superior laryngeal (X) nerves. Efferent pathways go through motor branch of the trigeminus (V), facial (VII), hyoglossus (XII) and some motor fibbers from the ambiguous nucleus (IX, X and XI)(37).

EVALUATION

The method used on the clinical practice to evaluate the dysphagia is the **swallowing video endoscopy,** which is considered one of the gold-standard (38)**.** It is and easy procedure, well tolerated with low risk of complications. Firstly, an anatomic evaluation is performed, evaluating respiration, phonation and the protection of the airway through a series of techniques. Then they proceed to evaluate the swallowing function by administrating to the patient a set of different foods with different consistencies and volumes coloured by methylene blue. This way it is possible observe if either aspiration or food retention happens and in what swallowing phase happens (37).

A COMPLICATION FROM ADJUVANT TREATMENTS

Many factors can alter the correct swallowing function. A determining one in oncologic patients is the treatment with either radiotherapy and chemotherapy or both. Nowadays, adjuvant protocols for adjuvant treatment in HNSCC often require aggressive schemes.

Specifically, for radiotherapy, several factors can trigger a damaging response such as: volume and total dose administered, dose fractionation and overall time of treatment. If radiation injuries are clinically classified we can see two major group: early mucosal injuries and deeper tissue responses (39).

- **Early mucosal injures** can be acute (< 3 months) or subacute (>3 and < 6 months). The changes are evident in the epidermis and mucosa and are a direct consequence of cell death, inflammation and hypoplasia that lead to mucositis, edema and erythema. Clinically they are presented as erosive and/or ulcerative lesions that cause moderate-severe pain (40). The damage is caused by an excessive production of reactive oxygen and nitrogen species (ROS), resulting from the inflammatory reaction, that trigger cell dysfunction and apoptosis.

  Frequently, the duration of these lesions is brief, and its severity does not correlate with chronic radiation-induced injuries. (39).
- **Deep tissue responses** are chronic consequences (>6 months) attributed to a vascular and surrounding tissue damage. These can happen independently from early injuries, and it is mainly related by the treatment fractionation. They occur when there is a failure to down-regulate fibrotic deposition and thus, a sequence of events takes place: fiber disorganization, altered microvasculature and production of pro-fibrotic growth factors. Other important structures for the swallowing process are also damaged by radiation such as: muscle tissue and salivary tissue, specially the submandibular grand (39).

The difficulty in swallowing as a result of chronic radiotherapy effects are known as late radiation-associated dysphagia (RAD), which has a prevalence of 38%. In order to reduce late RAD suggestions are to minimize the volume of irradiation to pharyngeal constrictors and larynx to a maximum of 50-60 Gy if it is possible to not compromise target doses. A dose-effect relationship has been established: with every 10 additional Gy there is an increase of the probability of RAD of 19% (38).

Chemotherapy, frequently necessary in adjuvant treatments of head and neck cancers, is also a common cause of oral mucositis. Cisplatin and 5-fluoracil (5-FU) protocols are particularly related to s this type of toxicity, and with every additional cycle, toxicity is worsened (40).

3.2. JUSTIFICATION

An epidemic increase of HPV-related OPSCC is taking place. Patients suffering from it are usually men without risk factors such as alcohol and tobacco and younger (mid 50s) than the average age at presentation of non-HPV related OPSCC. HPV carcinogenic potential has been demonstrated, but no efficient preventive methods are available. Vaccines against different HPV serotypes do seem very promising, but population coverage is still low, and in most countries, men are not tributary to it.

HPV-positive OPSCC are a distinct clinicopathologic entity, with HPV being a very favourable prognostic factor. Especially when patients are non-smokers, they have a better disease-free recurrence and overall survival. Nonetheless, these patients still receive the same treatments and at the same doses that patients with HPV-negative OPSCC.

Chemotherapy and radiotherapy benefits in oncologic control are evident, but they are also followed by toxicities. When it was observed that patients with HPV-related OPSCC had such a better prognosis, the intention to de-escalate their treatment rapidly emerged. The optimal treatment would be one that would provide them the optimal oncological control with as minimum toxicity as possible.

Several ongoing studies aim to find the best de-escalation strategy. From our point of view, it is crucial not only the type of de-escalation, but also that initial trials address patients with HPV-related OPSCC that have a good prognosis. However, most studies exploring de-escalation are only considering HPV status. High tumor infiltrating lymphocytes (TILs) have been demonstrated to be another relevant favourable prognostic factor (patients with high TILs levels show better 5 year overall survival) and we have included this feature among inclusion criteria.

The aim of this study is to find a proper balance between the therapeutic effectiveness and iatrogenic harm. We consider that including TILs levels as an inclusion criteria is a way to increase the security of the de-escalated treatment. Also, based on bibliography, we believe that by reducing 10 Gy would not harm disease control, but would significantly improve patient quality of life in relation of a reduction of the prevalence and/or intensity of the dysphagia.

4. HYPOTHESES

4.1. MAIN HYPOTHESIS

The de-escalation treatment based on a reduced dose of adjuvant radiotherapy treatment of 50 Gy instead 60 Gy after transoral tumor resection in patients with oropharyngeal squamous cell carcinoma (OPSCC) caused by human papillomavirus (HPV) will show no inferiority in terms of **overall survival** against the standard protocols of adjuvant treatment.

4.2. SECONDARY HYPOTHESES

- The de-escalation treatment based on a reduced dose of adjuvant radiotherapy treatment of 50 Gy instead 60 Gy after transoral tumor resection in HPV-related OPSCC will show no inferiority in terms of **local recurrence** versus the standard adjuvant protocols.
- By reducing the intensity of adjuvant radiotherapy after transoral tumor resection in patients with HPV related oropharyngeal squamous cell, there will be an improve of the **swallowing function**.

5. OBJECTIVES

5.1. MAIN OBJECTIVE

To confirm de non-inferiority on **overall survival** in patients with low stage (T1-T2 N1-N2) HPV-related oropharyngeal squamous cell carcinoma treated with in the less aggressive adjuvant radiotherapeutic protocols based on a 10 Gy reduction protocols after radical transoral resection.

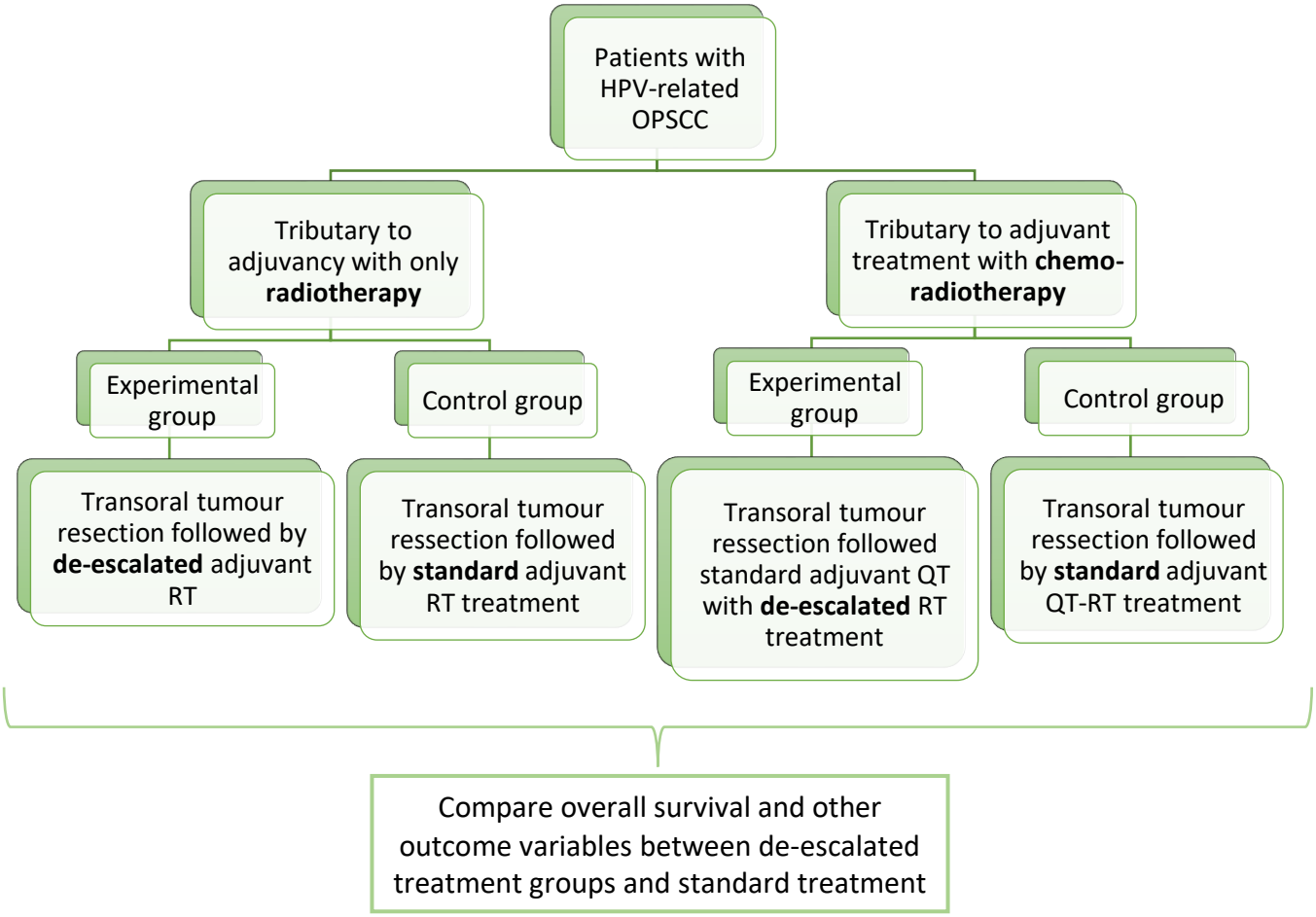
5.2. SECONDARY OBJECTIVES

- To confirm de non-inferiority on **local recurrence** in patients with low stage HPV-related oropharyngeal squamous cell carcinoma treated with less aggressive adjuvant radiotherapeutic protocols based on a 10 Gy reduction protocols after radical transoral resection.
- To assess whether the **swallowing function** is improved by reducing 10 Gy the intensity of adjuvant radiotherapy protocols after radical transoral resection in patients with low stage HPV related oropharyngeal squamous cell carcinoma.

6. MATERIAL AND METHODS

6.1. STUDY DESIGN

This protocol is designed as a multi-center non-inferiority, open-labelled, randomized, controlled clinical trial with four parallel groups.



In spite of four independent groups existing, the statistical analyses would be between two groups: de-escalated treatment group and standard treatment group. This will be further developed in the statistical analysis section.

6.2. SITUATIONS TO STOP THE TRIAL

- An increased rate of local recurrence in any of the groups.
- A significant decrease in overall survival in any of the groups.

6.3. STUDY SETTING

This study is designed to be multicenter. It will be set among the following hospitals from Catalunya that perform transoral surgery and are part of the national public health system:

- *Hospital Universitari de Girona Doctor Josep Trueta*, Girona (inhabitants of 800.000 as a population of reference)
- *Hospital de Bellvitge*, Barcelona (inhabitants of 343.172 as a population of reference)
- *Hospital de Germans Trias i Pujol*, Badalona (inhabitants of 800.000 as a population of reference)
- *Hospital Joan XXIII,* Tarragona (inhabitants 1.200.000 as a population of reference)

The population of reference stated is not the one from primary attention, but the one estimated considering the reference population of tertiary attention in the otorhinolaryngology field. Patients affected by a head and neck tumor are medically assisted only in tertiary centers.

The reference center in this trial will be *Hospital Universitari de Girona Doctor Josep Trueta*. One researcher will be assigned as the representant of each of the hospitals mentioned, in order to obtain a good communication between all the centers.

6.4. STUDY POPULATION

The study population will include adult patients diagnosed with oropharyngeal cancer caused by HPV in either *Hospital Josep Trueta*, *Hospital de Bellvitge*, *Hospital de Germans Trias i Pujol* and *Hospital Joan XXIII*. Neoplasia need confirmation of being HPV-related by p16 immunohistochemistry and with positive result in HPV-DNA PCR test.

Our target population will only include new oncologic diagnoses that have not been treated yet since the beginning of our recruitment. There will be no use of previous data collected in any of the Hospitals mentioned.

6.5. SUBJECTS SELECTION

INCLUSION CRITERIA

- Aged over 18 and under 60.
- Histologically confirmed diagnosis of squamous cell carcinoma of oropharynx.
- HPV positive by $p16^{INK4a}$ immunohistochemistry (70% cutoff) or intermediate $p16^{INK4a}$ (50-70%) with additional positive result in HPV-DNA test by polymerase chain reaction (PCR) (23).
- Neoplasia with tumor T-stage < 3 with $CD8^+$ TILs above 13.2 (33).
- Cases where transoral resection and neck dissection is considered to be the primary treatment modality.
- Signature of written informed consent provided.

EXCLUSION CRITERIA

- T3-T4 tumors, or T1-T2 that transoral surgery is considered not feasible.
- N2c-N3 nodal disease.
- Positive (< 1mm) margins around the primary tumor after resection.
- Distant metastatic disease determined by pre-operative image staging investigations (thorax and upper abdomen CT and/or PET CT).
- Prior radiation above the clavicles.
- Current smokers with N2b disease, including those smokers up to 6 months before diagnosis.
- History of preexisting swallowing dysfunction prior to the diagnosis of oropharyngeal cancer and /or any preexisting medical condition likely to impair swallowing function.
- History of infiltrating neoplasia in the 5 years prior to the OPSCC diagnosis.

6.6. SAMPLING

6.6.1. SAMPLE SELECTION

The sample selection will be executed in the four centers mentioned based on a convenience criteria. These centers have been chosen because they use the same treatment protocol when it comes to transoral surgery, therefore the inter-surgeon variability will be reduced. Furthermore, they also use the same schemes of adjuvant treatment.

Sampling system will be consecutive and therefore it will not be probabilistic. Patients that attend any of the hospitals mentioned that further meet inclusion and not exclusion criteria will be enrolled in this clinical trial.

As mentioned, statistical analysis will be between the de-escalated treatment group and standard treatment group. This decision based on clinical practice judgement: we consider that this study is too relevant to exclude a significant group of patients, either those initially tributary to only adjuvant radiotherapy or the group initially tributary to adjuvant concurrent chemo-radiotherapy.

Importantly, in order to guarantee the randomization, the sample will be balanced by stratifying for treatment type meaning that each type of treatment group will be composed by the same number of individuals.

6.6.2. SAMPLE SIZE

In a bilateral contrast with a level of signification (α) of 5%, with a potency (1-β) of 80, with a 15% of maximum difference for non-inferiority on an actual 90% of overall survival equivalent to a Cohen's d equal to 0.2, i.e. moderate effect) we will need a sample of 196 people.

Assuming that the percentage of drop out was 5%, then 206 people will be required.

Computations were carried out with the Professor Dr. Marc Saez' software based on the package "pwr" pf the free statistical environment R (version 3.6.2).

We expect that only 75% of the patients recruited will meet our inclusion criteria of TILs (33). Therefore, in order to compensate it, we will extend our sample size to 1.25*206 resulting on a total of **260**, with **65 patients in each parallel group**.

6.6.3. ESTIMATED TIME OF RECRUITMENT

There is no available data about the number of patients who meet the inclusion criteria and seek medical attention in the participant hospitals. Nevertheless, taking into account that the study is designed as multicentric and the clinical estimation of the incidence, the expected time of recruitment for enrolling the required number of participants (***260***) is approximately three years.

If the sample size is not achieved in this period, the recruitment time will be extended.

6.7. VARIABLES

6.7.1. OUTCOME VARIABLES

Main outcome

- **Overall survival**: evaluated in the 5 following years after the initial diagnosis.

Secondary outcome

- **Local recurrence with histopathological and image techniques confirmation:** it will be evaluated in the 3 following years after the initial diagnosis by imaging techniques.
- **Presence of dysphagia:** determined by swallowing video endoscopy. It will be considered that dysphagia is present if any of these clinical conditions exist: (i) residue of food remaining in the mouth or pharynx structures, (ii) penetration of food or liquid entering the airway entrance and (iii) aspiration for food or liquid entering the trachea.

6.7.2. INDEPENDENT VARIABLES:

Type of adjuvant treatment. They are dichotomic qualitative variables:

⇒ In those patients who would be initially tributary to adjuvant radiotherapy:

- **Control group** ($\mathbf{A_1}$): postoperative radiotherapy at a dose of 60 Gy in 30 fractions over 6 weeks
- **Experimental group** ($\mathbf{B_1}$): postoperative radiotherapy at a dose of 50 Gy in 25 fractions over 5 weeks

⇒ In those patients who would be tributary to adjuvant treatment with chemo-radiotherapy .

- **Control group** (A_2): postoperative radiotherapy at a dose of 60 Gy in 30 fractions over 6 weeks with concurrent Cisplatin, 3 cycles at 100 mg/m^2 every 21 days.
- **Experimental group** (B_2): Postoperative radiotherapy at a dose of 50 Gy in 25 fractions over 5 weeks with concurrent Cisplatin 3 cycles at 100 mg/m^2 every 21 days.

6.7.3. COVARIABLES

- **Age:** expressed in years at the moment of diagnosis, measured in one year-intervals since the date of birth. The age from of the patient will be consulted from the ID card or any other official document facilitated to the investigator.
- **Gender:** Male/female. Data will be consulted from the ID card or any other official document facilitated to the investigator.
- **Socioeconomic status:** Social class I to V. Constructed with education level and occupation following Domingo et al. (41,42).
- **Ethnicity:** four groups will be considered (Caucasian, African, Asian and Latin-American). Classification will be done by self-referred.
- **Smoking:** three groups will be made (i) non-smokers, (ii) smokers (currently, or in the last 6 months prior to diagnosis) or (iii) ex-smokers.
- **Alcohol consumption:** separated in three groups (i) non consumers (ii) moderate consume for women if consume per day is between 20 to 40 grams and for men if 40 to 60 grams (iii) high consume defined as a consume higher than 40 grams in women and 60 grams in men.
- **Primary site of the tumor**: considering all the possible subsites of the oropharynx: tonsillar, base of tongue, uvula, soft palate, posterior pharyngeal wall and lateral pharyngeal wall. The location will be assigned by the investi1gators given the physical examination also taking into account the video-endoscopy and imaging techniques.
- **T-stage at enrollment:** based on imaging techniques.
- **N stage at enrollment:** based on imaging techniques.

All the variables are summarized in **Table 4**, found below:

Table 4: Variables summary

	VARIABLE	DESCRIPTION	METHOD OF MEASUREMENT
OUTCOME VARIABLE	**Dysphagia**	Nominal dichotomous qualitative	Swallowing Video endoscopy
	Local recurrence	Nominal polytomic qualitative	Image techniques, anatomopathological exam
	Overall survival	Discrete quantitative	Measured by the investigator from diagnosis until confirmed decease
COVARIATES	**Age**	Discrete quantitative	Data collection sheet (***Annex 2***)
	Gender	Nominal dichotomous qualitative	Data collection sheet (***Annex 2***)
	Socioeconomic status	Ordinal polytomic qualitative	Data collection sheet (***Annex 2***)
	Ethnicity	Nominal polytomic qualitative	Data collection sheet (***Annex 2***)
	Tobacco	Nominal polytomic qualitative	Data collection sheet (***Annex 2***)
	Alcohol	Nominal polytomic qualitative	Data collection sheet (***Annex 2***)
	Location of the primary site	Nominal qualitative	Direct visualization and image techniques (CT and MRI)
	T-stage	Ordinal qualitative	Image techniques, anatomopathological exam
	N Stage	Ordinal qualitative	Image techniques, anatomopathological exam

6.8. STUDY CIRCUIT AND DATA COLLECTION

6.8.1. STUDY CIRCUIT

A *quick diagnosis* circuit exists for when any patient is suspected to suffer from a tumor in the head and neck area. This circuit ends up in a committee known as *Head and Neck Functional Unit,* where a multidisciplinary team discusses and decides which is the following tests that need to be performed.

A head and neck CT scan is requested on every patient in order to establish the TNM stadium of cancerous lesion; if margins are not clear a MRI will be also requested. A chest CT will be also requested in order to assess presence or absence of distant metastases.

On the next appointment the physician will realize a video-endoscopy in order to perform a biopsy of the suspicious area under local anesthesia. The biopsy sample is conducted to the Pathology Department of the Hospital where HPV status will be determined. An alternative way to obtain the HPV status is by analyzing a sample obtained from fine needle puncture-aspiration assessment (PAAF).

In the course of 2 to 3 weeks after the first visit, every case is displayed again in a meeting of the *Head and Neck Committee*. In this visit, with the information of HPV status and the imaging techniques, the committee decides the best treatment for each case. Patients who are HPV-positive and meet the TNM inclusion criteria, and furthermore the committee consider that they are tributary to a tumor transoral resection will be considered as potential candidates for the clinical trials but will need to meet the other inclusion criteria such as TILs.

The decision of the committee will be presented on the subsequent visit, and if he/she comply with it, surgery will take place on the following weeks. In this same visit, the study will be explained to them: all the criteria our study will be presented by an information document (***Annex 3***) making sure that they understand the terms and agreements of the investigation. Then we would proceed to ask for a written consent (***Annex 4***), only patients that sign will be considered to entry the study.

In order to homogenize the intervention, all surgeons participating on this study will undergo a preparation before the start of the study and will follow a protocol when it comes operate. After surgery the tumor piece will be sent for when the anatomopathological exam to estimate the TILs and see if they finally meet all the inclusion criteria.

If patients meet the final additional TILs inclusion criteria, patients will definitely by recruited for the study if he or she has previously signed the informed consent document. This will be notified to them in the post-surgery visit. Then the type of adjuvant treatment (standard

or de-escalated) will be randomly assigned as mentioned before. Also, a detailed history taking will be performed in order to obtain all the information presented in the Data collection sheet (***Annex 2***).

After adjuvant treatment is finalized clinical follow up visits will take place: on the first two years post-treatment every 3-4 months, from the 2nd year every 6 months, and on the 4th and 5th year visits will be annual. On these visits, exhaustive anamnesis, neck physical examination and video-endoscopy will be performed.

Control imaging techniques as CT or MRI will also be performed. The first one will be after 3 months after adjuvance has ended, and then every 6 months during the first two years. After the first two years of control, imaging techniques will be performed annually.

The data analysis phase will begin when the data from the 260 patients included in our sample is collected (in an estimated time of three years). Nevertheless, preliminary analysis will be performed when half of the sample has already been collected. We are aware that the potency of the results with half of the sample will not be as strong. Nonetheless in case we obtain clear detrimental results from the de-escalated therapies study will be stopped for ethical reasons.

6.8.2. DATA COLLECTION

History taking

- A detailed history taking and physical examination will be performed on the first visit when a suspicious cancer of the otorhinolaryngological area is suspected.
    - History taking will be addressed to find out about the time of progression of lesion/symptoms and some preliminary data related to the covariates that will be collected hereafter
    - Physical examination will punt emphasis on the palpation of possible lymphadenopathies
    - Patient information such as smoking habits, oral sex history and other epidemiologic information will be obtained with a data collection form (***Annex 2***) only after he or she have signed the informed consent.

Determination of HPV status

- HPV status will be determined by p16 immunochemistry on paraffin-embedded biopsies obtained from either video-endoscopy or PAAF. It will be considered positive if there is a strong and diffuse staining of the nucleus and cytoplasm in ≥ 70% of the tumor specimen (4).
- HPV-DNA will be extracted from paraffin-embedded biopsies obtained from video-endoscopies and will be determined by linear array LINEAR ARRAY HPV Genotyping Test (*Roche Diagnostics*) (25).

Tumor staging

- TNM stage will be obtained through the imaging techniques (see **Annex 1**).

TILs determination

- $CD8^+$ TILs will be visualized by standard streptavidin-biotin peroxidase method with mouse monoclonal antibodies anti-CD8 performed on the tumor mas extracted from surgery (33).

Dysphagia evaluation

- Dysphagia presence/absence will be evaluated through swallowing video-endoscopy on the 6, 12 and 24 months follow-up visits after surgery and adjuvant treatment have finalized.

6.9. STUDY INTERVENTIONS

6.9.1. RANDOMIZATION

The patients included in this study will be randomly distributed in two groups (de-escalated or standard adjuvant radiotherapy protocol). A stratified random sample will be done, so the population will be split in two groups according to what adjuvant treatment they are initially tributary to: radiotherapy or concurrent chemo-radiotherapy. This way, four subgroups are formed, listed below:

- **Group A (control group):** this group will receive the standard adjuvant treatment at its standard doses
    - **A_1:** only radiotherapy as adjuvant treatment
    - **A_2:** chemoradiotherapy as adjuvant treatment

- **Group B (experimental group):** this group will receive a de-escalated radiotherapeutic adjuvant treatment.
    - **B_1:** only de-escalated radiotherapy as adjuvant treatment
    - **B_2:** chemotherapy and de-escalated radiotherapy as adjuvant treatment

Randomization of treatments will be performed according to a randomization list generated by nQuert Advisor 7.0 (Statiscal Solutions Ltd., Cork, Ireland) in which patients remain into 4 blocks with 65 subjects each, balanced for what adjuvant treatment where they initially tributary. By this, we will ensure the comparability of survival between groups, by balancing their intrinsic baseline characteristics (such as TNM stage and anatomopathological features) that imply different initial prognosis and therefore determine their adjuvant treatment.

The radiotherapist will be the one responsible for dispensing the intervention according to the randomization list above mentioned.

6.9.2. STUDY INTERVENTION

First step when a patient enrolls the trial, he or she will be introduced on the database. Investigators will make sure to collect history of each subject as stated in the data collection sheet (***see Annex 2***). This database will be shared with the rest of participant hospitals. Each patient will be identified with a subject identification number.

All patients will undergo transoral resection. According the tumor staging and anatomopathological features patients will be tributary to either radiotherapy or chemo-radiotherapy as adjuvant treatment. Study interventions will be done depending on the randomly assigned group explained before, from which the patient belongs to.

- **Group A (control group):** depending on the anatomopathological features:
    - **Postoperative radiotherapy (A_1)** at 60 Gy in 30 fractions in 6 weeks if they show any of the following characteristics: T1-T2 tumors that have additional risk factors such as pN2a or pN2b, evidence of perineural and/or vascular invasion and histologically normal tissue margin around the primary tumor of 1-5 mm.
    - **Concurrent cisplatin chemotherapy (**3 cycles at 100 mg/m^2 every 21 days) **with radiotherapy (A_2)** at 60 Gy in 30 fractions in 6 weeks if there is evidence of cervical lymph node extracapsular spread

- **Group B (experimental group):** this group will receive a de-escalated radiotherapeutic adjuvant treatment with or without concurrent chemotherapy based on the same anatomopathological features explained in the control group (A)
    - **Postoperative radiotherapy (B_1)** at 50 Gy in 25 fractions over 5 weeks if they show any of the following characteristics: T1-T2 tumors that have additional risk factors such as pN2a or pN2b, evidence of perineural and/or vascular invasion and histologically normal tissue margin around the primary tumor of 1-5 mm.
    - **Concurrent cisplatin (**3 cycles at 100 mg/m^2 every 21 days) **with radiotherapy (B_2)** at 50 Gy in 25 fractions over 5 weeks indicated by the evidence of cervical lymph node extracapsular spread.

All patients, either control or experimental group, will undergo the same pattern of follow-up visits explained in the data collection section.

6.9.3. SAFETY

Transoral resection has been proved to be as effective conventional surgery. It usually allows an *en bloc* tumor resections enabling a more accurate intra-operative evaluation of the resection margins (43). Therefore, it does not affect the overall safety of the procedure.

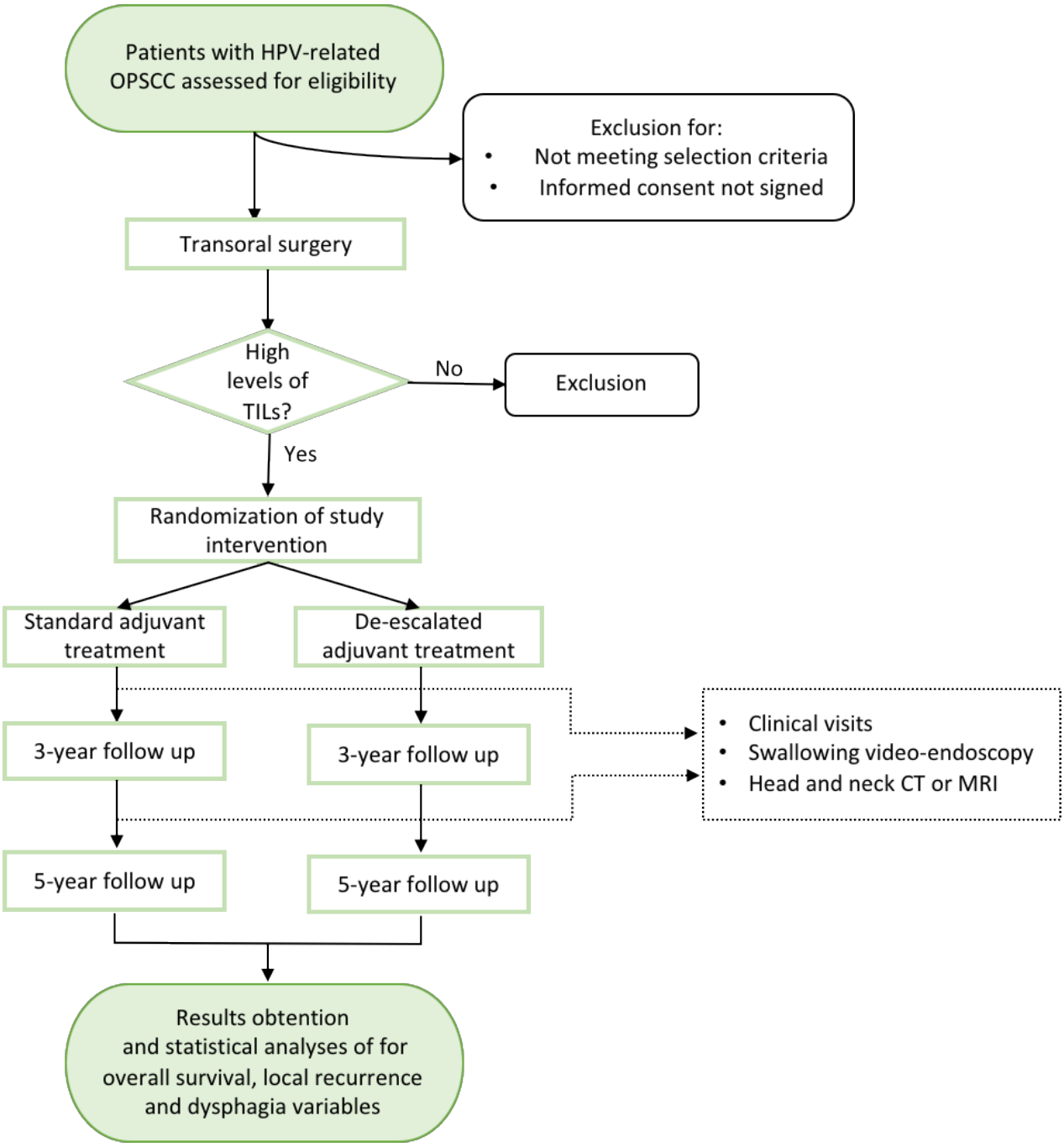
Some might consider that the decrease on **adjuvant radiotherapeutic doses** could be a reason to worry about the oncologic control. Nevertheless, it has been long proved that HPV-relayed OPSCC tumors are more radio-sensible and have a better response to radiotherapy. Therefore, we consider that it will not affect oncologic control. Furthermore, we expect that patients treated with de-escalated protocols will have an improve in functional outcomes, especially on swallowing function.

6.9.4. DEGREE OF BLINDING

This study is designed as an open-labelled clinical trial. Patients will know what type of treatment they are receiving, and researchers will have the information about what treatment group are subjects assigned to.

It is not possible to mask the intervention to patients, as reducing the total dose of radiotherapy implies having less cycles with a shorter duration of time. Nevertheless, what we can ensure is that the statistician who will do the final analysis will be blinded.

6.10. FLOW CHART



6.11. STATISTICAL ANALYSIS

The statistical analysis will be done using the Statistical Package for the social Sciences (SPSS Windows®) and the responsible statistician who will do it will be blinded to the study groups.

Results will be considered as statistically significant at a value of $p<0.05$ defining a confidence interval of 95%.

6.11.1. DESCRIPTIVE ANALYSIS

The time of overall survival of the groups of intervention **A** and **B** will be summarized using median and interquartile range. The curves of survival will be estimated and represented with the Kaplan-Meier method.

For the variables local recurrence and dysphagia, we will use proportions for the groups **A** and **B** of intervention represented in a contingency table.

These analyses will be stratified for the different covariables. Age will be categorized with quartiles.

For covariables below, the ones that are qualitative will be summarized using proportions, age will be summarized using mean and standard deviation, stratifying by groups of intervention.

- Age
- Gender
- Socioeconomic status
- Ethnicity

6.11.2. BIVARIATE INFERENCE

The difference between the overall survival between the intervention groups (**A** and **B**) will be compared with the contrast log-rank (log-rank test).

The difference of proportions between treatment groups **A** and **B** on the variables local recurrence and dysphagia will be analyzed using Chi-square. We will use the exact Fisher's test when the expected number of counts in any cell of the table of contingency will be lower than 5.

These analyses will be stratified for the covariables. Age will be categorized in quartiles.

6.11.3. MULTIVARIATE ANALYSIS

Because randomization, we expect our groups to be balanced in terms of age, gender, smoking and alcohol habits and therefore there should be no cofounding. Nevertheless, when it comes statistical analysis if cofounding is suspected, we will adjust the associations between the outcome and the independent variables controlling for the confounders.

The effect of the intervention on the overall survival will be assessed in a Cox regression, where the dependent variable will be overall survival and the independent the group of intervention (**A** or **B**), controlling for covariables.

In the Cox regression we will also evaluate the possible interaction between the intervention and sex, age and staging.

For local recurrence and dysphagia (dependent variables) the effect of the intervention (independent variable) will be assessed with logistic regression controlling for covariables. Again, age will be categorized, and we will take into consideration possible interactions.

7. ETHICAL CONSIDERATIONS

This trial is proposed according the basic ethical principles. **Patient autonomy** will be in all cases respected, participation will be completely voluntary and only those who sign the informed consent will entry the trial. The main objective of this study is to seek for the **patient's beneficence** by reducing the adjuvant treatment's toxicity. **No maleficence** is expected to happen to patients. Based on the bibliographic research and the inclusion of additional favorable prognostic factors (TILs) in the inclusion criteria, risk of oncologic control being damaged is very low. Benefits are superior to risks. Every person that meets the inclusion criteria and not the exclusion criteria will be proposed to entry the trial, without discrimination. Thus, there will be **justice.**

This clinical trial will respect the medical ethic principles described in the World Medical Association Declaration of Helsinki (1964, last reviewed on 2013) about the ethical principles for medical research involving human subjects. This protocol has also been developed according to what establishes the *Real Decreto 1090/2015, de 4 de diciembre*.

The study protocol will be firstly submitted and introduced to the *European Clinical Trials Database* in order to get a registration number. Then, it will be submitted to the Clinical Research Ethics Committee (CEIC, "*Comitè Ètic de Investigació Clínica"*) and to the Medicament Research Ethics Committee (CEIM, "*Comitè Ètic de Investigació amb Medicaments*") of the Hospital Universitari de Girona Doctor Josep Trueta for its review. After the approval from the ethic committees, the protocol will be sent to directors of each of the participant hospitals requiring for each of their approval. Finally, it will be sent to the Spanish Agency of Drugs and Sanitary Products (AEMPS*, "Agència Espanyola de Medicament i Productes Sanitaris")* for the final authorization.

Only after approval from all of these entities will this study be carried out. Furthermore, any suggestions given by the CEIC will be taken into account and introduced.

All the members of the research team of all the participant hospitals must sign a statement indicating that they have read and approved the final protocol and agree with the ethics aspects of research. Data will be published with transparency and will not exclude unfavorable events.

Patients will be informed about all the details in the protocol (***Annex 3***) and a *sine qua non* condition for entering the trial is that they have signed the informed consent (***Annex 4***) as stated on the "*Ley 41/2002 Básica reguladora de la autonomía del paciente y de derechos y obligaciones en materia de información y documentación clínica"*.

ETHICAL CONSIDERATIONS

According to the "*Ley Orgánica 3/2018, de 5 de diciembre, de Protección de datos personales y garantía de los derechos digitales"*, all information obtained from patients will be completely confidential, being used only for the purpose of the research. Only the ones in charge of the investigation will have access to the data.

Being aware that our study could be potentially be considered as an invasive procedure by the CEIC, we will also follow the "*Ley 14/2007, de 3 de julio, de investigación biomédica*" in order to seek prevailing the health and comfort of the patients that participate.

No conflicts of interests are declared by the investigators in charge of this study.

8. LIMITATIONS AND STRENGTHS

8.1. LIMITATIONS

On one hand, this study has several limitations that need to be taken into account:

- Being a multicentric trial, in which surgery (operator-dependent) plays an important role into the final success of the intervention, **variability** between hospitals and surgeons when it comes to the intervention could happen.

  In order to prevent this, all surgeons that participate in this study will undergo a preparation before the study starts and will operate following the same protocol.
- In a prospective study, as long as this one is expected to be, there is always a risk of **withdrawals**. Nevertheless, in our study we expect it to be low, as patients' disease is severe, and they are aware of the importance of compliance. Nevertheless, if withdrawals take place, the medical and research team will try to find out the reasons behind them.

  A percentage of expected losses has been anticipated and also been taken into account when calculating the sample size.
- To prevent the **compliance bias** generated by patients preferring one type of treatment rather than the other, it will be properly explained to participants that treatment will be randomly assigned without possibility of choosing. It will also be clarified that there is strong evidence that suggests that de-escalated treatment will cause no detriment in their oncologic control.
- In spite of being a clinical trial, it is **open labelled** for both patients and investigators as it is not possible to mask de-escalated radiotherapy treatment as it requires a smaller number of cycles.

  Nevertheless, the statistician that will do the data will be blinded.
- This protocol has been designed with a consecutive non-probabilistic sampling method due to the population of study's characteristics, which could potentially produce a **selection bias,** in order to avoid it, treatment groups will be assigned randomly making the groups equivalent.
- Although the study design, sample size and time of recruitment of this study make this an **expensive** study. These are required to accomplish the objectives and draw significant conclusions.

8.2. STRENGTHS

On the other hand, our study also has several strengths that have to be taken into consideration:

- This is one of the first studies, if not the first, to incorporate TILs besides HPV status as an inclusion criteria. If results are significant, it would confirm that TILs are a good prognostic factor to further stratify patients in terms of prognosis.
- By incorporating TILs as an additional inclusion criteria, this study becomes safer for participant patients in terms of oncologic risk.
- HPV-related OPSCC's incidence is increasing rapidly. If the results of this study prove that de-escalated radiotherapy is non-inferior, a growing number of patients could be treated with another effective treatment that moreover would spare them several adverse effects that would affect significantly their quality of life.
- If de-escalated treatment is proved to be non-inferior and is then incorporated in the future medical practice, it will reduce expenses in terms of less radiotherapy and fewer hospitalizations due to possible complications from intensive radiotherapeutic treatment as dysphagia and aspiration.
- De-escalated treatment in this study is only considered for patients with HPV-related OPSCC in early stages. If it is proved to be non-inferior than standard adjuvant protocols, it would be set a basis so that future trials could study this same de-escalation in patients affected by a more advanced HPV-related OPSCC.
- Being a multicentric study composed by tertiary hospitals from different provinces of Catalunya implies that if significant, the study results could be more easily generalized.

9. WORK PLAN AND CHRONOGRAM

The activities developed will happen in the following sequence:

STAGE 0: STUDY DESIGN (November 2019 – January 2020)

- Activity 1: Bibliographic research about HPV etiopathogenetic role in OPSCC and its impact in overall survival and also research on options of de-escalation has been performed. The research has been performed in PubMed and Clinical Trials databases.
- Activity 2: Protocol elaboration: objectives, hypothesis, variables and methodology.

STAGE 1: EHTICAL EVALUATION AND STUDY APPROBATION (February - March 2020)

- Activity 3: the protocol will be sent to get a registration number to the European Union Drug Regulating Authorities Clinical Trials (EudraCT).
- Activity 4: The current protocol will be presented to the CEIm and CEIC of the *Hospital Universitari de Girona Josep Trueta.*

  Once it gets the approbation, the protocol will be shared with the other services and direction of the other participant hospitals in order to get their approval.
- Activity 5: The protocol will be sent to the *Agencia Española de Medicamentos y* Productos (AEMPS) for its authorization
- Activity 6: Contracting a liability insurance.

STAGE 2: COORDINATION (March 2020 – April 2020)

- Activity 7 (March 2020): The research team of each hospital will have to select a representant that will perform the coordination measures with the other centers during the full duration of the study recruitment. This activity also includes the organization of tasks.
- Activity 8 (March 2020): Hospitals' representants will have a first meeting in *Hospital Universitari de Girona Doctor Josep Trueta* to solve any problems of the protocol and discuss their organization.

  From here, representants of each hospital will meet annually during the recruitment process with the purpose to evaluate if the protocol is being followed correctly.
- Activity 9 (April 2020): The otorhinolaryngologic specialists of the hospitals involved in the study will meet in *Hospital Universitari de Girona Doctor Josep Trueta* to participate

in a practical workshop with an expert of robotic transoral surgery that will train them to do this procedure following the same protocol.

- Activity 10 (April 2020): The otorhinolaryngologic specialists that participate in this trial will take part in a workshop with an expert in evaluation of swallowing function by video-endoscopy. This will take place in *Hospital Universitari de Girona Doctor Josep Trueta.*

Activity 9 and 10 are necessary in order to avoid variability between specialists and hospitals when diagnosing and treating. This will safeguard the homogeneity required to obtain more representative results and therefore conclusions.

STAGE 3: DATA COLLLECTION (May 2020 – May 2028)

- Activity 11 (May 2020 - May 2023): Patient recruitment will be done by a consecutive sampling in the 4 hospitals participants in the trial. Only patients that meet the inclusion and not the exclusion criteria and that have signed the informed consent will be included in the sample. Then, surgery intervention will be performed.

  Patients will be distributed in the treatment groups randomly. The correspondent adjuvant treatment will be performed depending on what group they are assigned to.
- Activity 12 (May 2020 – May 2028)**:** Follow up visits will be performed periodically. Clinical follow up visits will take place every 3-4 months on the first two years post-treatment, every 6 months from the 2nd year, and on the 4th and 5th year visits will be annual. On these visits, exhaustive anamnesis, neck physical examination and video-endoscopy will be performed.

  First control imaging technique (CT or MRI) will be performed after 3 months of the finalization of the adjuvance. Then, every 6 months during the first two years and after the first two years of control, imaging techniques will be performed annually.

  The complete duration of the follow up will be 3 years since they entered the study in order to establish local oncologic control, and 5 years for overall survival.
- Activity 13 (May 2020 – May 2028)**:** The specialists will record the information collected of the different variables in every visit in the database.

STAGE 4: DATA ANALYSIS AND INTERPRETATION (May 2026 - June 2028)

- Activity 14 (May 2026 – May 2028): statistical analysis will be performed by a subcontracted statistician that will be masked for the treatment groups. He or she will perform an intermediate analysis once half of the sample's data has been collected (May 2026) to see if any situation to stop the trial exists. When all the data has been collected, a final statistical analysis will be performed (May 2028).
- Activity 15 (June 2028): The statistician will draw conclusions from the previous analyses.

STAGE 5: PUBLICATION AND DISSEMINATION OF THE RESEARCH FINDINGS (July – September 2028)

- Activity 16: presentation of the results on the *Societat Catalana de Otorinolaringologia* (SCORL).
- Activity 17: presentation of the results on the European Academy of Otorhinolaryngology, Head and Neck Surgery (EAROL-HNS).
- Activity 18: publication of the results on scientific journals.

	TASK	2019	2020				2021	2022	2023	2024 - 2027	2028			
		Nov - Dec	Jan	Feb	Mar	Apr	May - Dec	Jan - Dec	Jan - Dec	Jan - Dec		Jan - Apr	May	Jun	Jul - Sep
STAGE 0	***A1:*** Bibliographic research														
	A2: Protocol elaboration														
STAGE 1	***A3-5:*** CEIC, CEIm and AEMPS														
	A6: Insurance contracting														
STAGE 2	***A7:*** Selection of the representants														
	A8: Representants' annual coordination meeting														
	A9: Surgery workshop														
	A10: Swallowing evaluation workshop														
STAGE 3	***A11:*** Patient recruitment and intervention														
	A12-13: Follow up visits and data collection														
STAGE 4	***A14:*** Statistical analyses														
	A15: Statistical interpretation														
STAGE 5	***A16-17:*** Dissemination at SCORL and EAROL-HNS														
	A18: Publication														

10. FEASABILITY AND MEANS AVAILABLE

On one hand, the medical team in this study will be composed by otorhinolaryngologic specialists, pathologists and radiotherapists experienced enough in order to attend the necessities of the included patients and to execute the procedures of this study. All these professionals are currently employed by the Hospitals included in this study; thus, no additional medical employment will be necessary. On the other hand, a statistical analyzer will be hired in order to process all the data an interpret the results from his analyses.

The procedures that are part of the study (transoral robotic surgery and swallowing videoendoscopic evaluation) are part of the usual clinical practice in these settings as well, so no extra material and intensive preparation will be required. Only a practical workshop for each procedure seeking to homogenize how all the participant clinicians realize them.

When it comes to the determination of the tumor's HPV status by p16 immunostaining it is part of the clinical practice in all the integrant Hospitals. It will be performed by the pathology service of the hospital where each patient is assisted. For the determination of HPV-DNA PCR, samples will be sent to the hospital of Bellvitge where its determination is already part of their current clinical practice, and thus they have the means and experience to perform it.

The participant hospitals also dispose from the necessary means to realize all the follow-ups, so they are all able to participate in all the phases of the study.

The patients required for study sums up a total of 260, 65 per group. It is an ambitious sample, but we consider possible to collect it within 3 years as it is a multicentric study composed by reference hospitals.

11. BUDGET

PERSONNEL EXPENSES

The research team are employed by the Hospitals included in the study. Therefore, no additional clinicians are required, and this does not suppose and additional cost.

LIABILITY INSURANCE

In case of being considered by the CEIC as an invasive clinical trial, we will contract an insurance to cover any possible adverse effect that included patients suffer attributable to the participation in the study. The estimated cost is 78.000 € (about 300 per patient); the precise cost will be confirmed at the time of the study kick-off.

EXECUTION EXPENSES

Material for the bibliography research, such as articles, has not represented an additional expense. The determination of p16 status is part of the clinical practice, so its cost is not included in the study budget.

The execution expenses are composed firstly by the printing of all the protocol information papers (**Annex 3**) and informed consents (**Annex 4**) to hand the included patients. Considering that 5 sheet copies will be handed to each patient, of a total of 260 patients, the cost will be 39 €. Nevertheless, this is not included in this budget.

Secondly, each determination of the HPV-DNA determination by PCR has a cost of 85 €. This determination is already part of the habitual clinical practice in the participant hospitals *Bellvitge* and *Germans Trias i Pujol*. Therefore, the cost from this determination will only be budgeted for the other hospitals *Joan XXIII* and *Doctor Josep Trueta* that do not perform it the regular practice. Based on the reference population of each hospitals, these last ones represent approximately 60% of the sample, thus 0,6*260 = 156 patients. Therefore, the cost of the HPV-DNA determination by PCR will be 13.260 €.

Given that this is a multicenter study, in order to ensure the validity of the data collection, an investigation assistant will be subcontracted at partial time. It will take approximately 160 h, paying 15 €/h it sums up a total of 2.400 €.

For the statistical analysis an expert statistician will be subcontracted at partial time. We estimate that 60 hours of work will be necessary, they will be distributed within the recruitment and follow up data collection period. This will include constructing and the maintenance of the database and also all the statistical analysis required. We will pay 40€/h, therefore we calculate a cost of 2.400 €.

Finally, we will like to highlight that it will be not necessary to pay for the AEMPS fees as this study is classifies as a clinical investigation without commercial intentions that will be promoted by the hospital.

TRAVEL AND COORDINATION EXPENSES

In total, 4 coordination meetings will take place, one before the start of the recruitment, and another one annually during the three years of recruitment. The assistants of these coordination meetings will be the representative researchers of each of the four participant hospitals. We estimate a mean expense of 100 € per researcher per meeting that will include travel costs, accommodation and diets. Thus, we calculate a subtotal of 1.600 €

In order to homogenize the way surgical intervention is performed and the swallowing videoendoscopic evaluation, a practical workshop for each activity will be organized in the *Hospital Universitari de Girona Doctor Josep Trueta*. Two otorhinolaryngology experts from each of the four participant hospitals will assist. The reference investigators from the hosting hospital will be the one imparting it. We estimate a cost of 100 € per researcher in terms of travel and diets. Thus, the cost of the workshops will be approximately 800 € per workshop.

CONFERENCES EXPENSES

In order to disseminate the results to the rest of the scientific community we will attend national and international congresses. Two researchers will attend the national congress (SCORL) in two occasions: the first one with the preliminary results when half of the data has been already collected, and the second with the final analyses. The admission fee estimated on 500 € per person and per congress. Travel and accommodation cost will be approximately 500€ per person and per congress. Therefore, we calculate a global cost of 4.000 €.

Scientific results will also be presented in an international congress (EAROL-HNS), by two researchers only when final results and analyses are obtained. The admission fee is approximately 800 € per person and the costs of travel and accommodation are estimated to be 1.000 € per person. Thus, this will be budgeted on 3.600 €.

PUBLICATION EXPENSES

After the finalization of the study and the extraction and interpretation of results, we will publish it as a journal article. Taking into account the English correction (500 €) and preparation of the open access (1.800 €), the estimated subtotal of the publication costs are budgeted on 2.300 €.

The different expenses are summarized in **Table 5** below:

Table 5: Budget summary

ITEM	COST	SUBTOTAL
PERSONNEL EXPENSES		
Staff and research team	0 €	**0 €**
LIABILITY INSURANCE		
Policy insurance cost*	78.000 €	**78.000 €**
EXECUTION		
p16 determination	0 €	**18.060 €**
HPV DNA determination by PCR	13.260 €	
Investigation assistant	2.400 €	
Statistician	2.400 €	
TRAVEL AND COORDINATION		
Coordination meetings of the different hospitals' representatives (*travels and diets)*	1.600 €	**3.200 €**
Transoral surgery practical workshop *(travel and diets)*	800 €	
Video-endoscopic Swallowing evaluation practical workshop *(travel and diets)*	800 €	
CONFERENCES EXPENSES		
SCORL *(in two occasions, for two researchers including admission fee and travel and accommodation)*	4.000 €	**7.600 €**
EAROL-HNS *(in one occasion, for two researchers including admission fee and travel and accommodation)*	3.600 €	
PUBLICATION EXPENSES		
Article publication fee	2.300 €	**2.300** €
TOTAL: 109.160 €		

* the precise cost will be confirmed at the time of the study kick-off.

12. PROJECT IMPACT ON THE NATIONAL HEALTH SYSTEM

The incidence of HPV-related OPSCC has increased significantly over the last years, becoming a worrying issue and also causing a shift of the paradigm of the type of patients that have a head and neck tumor. These patients are much younger, and usually without additional risk factors such as alcohol and tobacco. In addition, they have a much better prognosis but are still treated with the same protocols as those with non-HPV related tumors, treatments that are associated with high toxicity and impairing of the swallowing function.

If the hypothesis of this study is proved to be true, it could translate into an important change in clinical practice. Treatment de-escalation in patients with HPV-related OPSCC would imply less dysphagia and secondary aspirations in these patients, a better life quality and a more optimal balance between effectiveness and iatrogenic harm.

Last but not least, a de-escalated treatment would imply less expenses in terms of radiotherapy cycles (equipment and health care professional time). It will also reduce costs when it comes to assessment and treatment required for dysphagia-related complications (aspiration and respiratory infections).

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14. ANNEXES

14.1. ANNEX 1: TNM FOR HPV RELATED OPSCC

CHARACTERISTIC	CATEGORY	CRITERIA
T-STAGE	T0	No primary tumor identified
	T1	Tumor 2 cm or smaller in greatest dimension
	T2	Tumor > 2 cm but < than 4 cm
	T3	Tumor > 4 cm in greatest dimension or extension to lingual surface of epiglottis
	T4	Moderately advanced local disease; tumor invades the larynx, extrinsic muscle of tongue, medial pterygoid, hard palate, or mandible or beyond
CLINICAL N-STAGE	NX	Regional lymph nodes cannot be assessed
	N0	No regional lymph node metastasis
	N1	One or more ipsilateral lymph nodes, none larger than 6 cm
	N2	Contralateral or bilateral lymph nodes, none larger than 6 cm
	N3	Lymph node (s) larger than > 6 cm
PATHOLOGICAL N-STAGE	NX	Regional lymph nodes cannot be assessed
	pN0	No regional lymph node metastasis
	pN1	Metastasis in 4 or fewer lymph nodes
	pN2	Metastasis in more than 4 lymph nodes
M-STAGE	M0	No distant lesion
	M1	Distant lesion

14.2. ANNEX 2: DATA COLLECTION SHEET

Hoja de recogida de datos de las variables demográficas y epidemiológicas en los pacientes participantes del estudio. Marcar con una cruz la opción que mejor se adecue.

1) **Código numérico asignado:** _____
2) **Fecha de nacimiento (día/mes/año):** ____/____/____
3) **Sexo:**
   - ☐ Mujer
   - ☐ Hombre
4) **Etnia**
   - ☐ Caucásica
   - ☐ Africana
   - ☐ Latinoamericana
   - ☐ Asiática
5) **Status Socioeconómico**
   - ☐ Clase I
   - ☐ Clase IV
   - ☐ Clase II
   - ☐ Clase V
   - ☐ Clase III
6) **Tabaquismo**
   - ☐ No fumador
   - ☐ Exfumador
   - ☐ Fumador (en el momento actual, o en los 6 meses previos al diagnóstico)
7) **Alcohol:**
   - ☐ No consumidor
   - ☐ Consumo moderado (en mujeres 20-40 g/día; en hombres 40-60 g/día)
   - ☐ Consumo elevado (en mujeres > 40 g/día; en hombres > 60 g/día)
8) **Estadio T al comienzo del estudio**
   - ☐ T1
   - ☐ T2
   - ☐ T3
9) **Estadio N al comienzo del estudio**
   - ☐ N0
   - ☐ N2a
   - ☐ N1
   - ☐ N2b

14.3. ANNEX 3: PROTOCOL INFORMATION DOCUMENT

HOJA DE INFORMACIÓN SOBRE EL ENSAYO CLÍNICO

Nombre del estudio: *Desescalada en las dosis de radioterapia adyuvante en pacientes afectados por tumores de orofaringe relacionados con el VPH con una cifra elevada de linfocitos CD8⁺ infiltrantes en el tumor.*

Centro asistencial:

Investigador/a principal:

Bienvenido/a

Nos dirigimos a usted para invitarle a participar en un estudio de investigación, llevado a cabo por los Servicios de Otorrinolaringología de varios hospitales de referencia en Cataluña. Este estudio ha sido aprobado por el Comité de Ética e Investigación Clínica de el l'Hospital Universitari Doctor Josep Trueta y por la Agencia Española del Medicamento y Productos Sanitarios

Nuestra intención es que usted comprenda el motivo por el cuál se está realizando este estudio i qué implica formar parte de él para que así, pueda decidir de forma voluntaria si finalmente desea participar o no. Es por eso que le rogamos que se tome el tiempo necesario para leer detenidamente y comprender este resumen informativo sobre nuestro estudio. No tiene por qué decidir hoy acerca de su participación; cualquier duda que surgiese, nuestro equipo le responderá y pondrá a su disposición toda la información necesaria.

DESCRIPCIÓN Y OBJETIVO DEL ESTUDIO

Clásicamente, los tumores de orofaringe se habían relacionado con consumos elevados de tabaco y /o alcohol. Sin embargo, la frecuencia de los tumores de orofaringe va en aumento, y esto se debe a un nuevo agente causal: el virus del papiloma humano (VPH).

Se ha observado que los pacientes cuyo tumor se relaciona con el VPH suelen ser más jóvenes y normalmente no han tenido consumos elevado de tóxicos como factor de riesgo. En los últimos años además, se ha visto que el curso de la enfermedad en estos pacientes es significativamente mejor que en los que su tumor no está causado por el virus, teniendo menos

recurrencias y una supervivencia más prolongada. Esto se atribuye a que dichos tumores son más sensibles a la radioterapia. Entre este grupo de pacientes, aquellos en que en el estudio microscópico se observan más células inmunitarias tipo $CD8^+$ tienen todavía una mejor evolución.

A pesar de estas observaciones, se sigue aplicando el mismo tratamiento a todo paciente con tumor de orofaringe, independientemente de la causa. Se trata de un tratamiento eficaz pero agresivo, que suele componerse por una intervención quirúrgica inicial seguida de radioterapia y en ocasiones de quimioterapia, según las características de cada tumor.

Sin embargo, este tratamiento no está exento de efectos indeseados, uno de los principales es la disfagia, o dificultad en la deglución. Además de molesta, puede llegar a ser grave asociándose con otras complicaciones que pueden llegar a requerir asistencia médica adicional. Su aparición está directamente relacionada con la dosis administrada de radioterapia, y sucede en alrededor de un 30% de los pacientes que la reciben.

Este estudio va dirigido a pacientes con tumores de orofaringe causados por el virus del papiloma humano, y que además tengan como factor pronóstico favorable adicional cifras elevadas de linfocitos $CD8^+$ en el estudio microscópico.

La motivación detrás de este proyecto de investigación es demostrar que la reducción en las dosis de la radioterapia no supone un perjuicio en el control oncológico y además supondría evitar parte de las sus complicaciones asociadas y/o reducir su gravedad. Se propone un nuevo protocolo de tratamiento, en el que se reducirán las dosis de radioterapia administrada: 10 Gy menos en comparación al tratamiento estándar.

METODOLOGÍA E INTERVENCIÓN

En este estudio participarán un total de 260 pacientes; a todos ellos se les operará con cirugía robótica. El total de pacientes será distribuido aleatoriamente en dos grupos (**A** y **B**) de igual tamaño que estarán estratificados por el tipo de tratamiento adyuvante indicado según las características de su tumor (radioterapia o quimioradioterapia) formando dos subgrupos también igual en tamaño dentro de los grupos principales.

- El **grupo A** recibirá la radioterapia a las dosis del tratamiento estándar que corresponde a 60 Gy en 30 ciclos distribuidos en 6 semanas. Se asociará quimioterapia según las características de la lesión.
- El **grupo B** recibirá radioterapia a dosis menores que el tratamiento estándar, que corresponderán a 50 Gy en 25 ciclos a lo largo de 5 semanas. La asociación de quimioterapia se hará según las características de la lesión.

Debido a las diferencias en número de ciclos y duración total del tratamiento radioterapéutico, tanto el paciente como el médico sabrán a cuál de los grupos pertenece el paciente.

Todos los participantes, independientemente del grupo de tratamiento al que pertenezcan, serán seguidos de la misma forma que en la práctica clínica habitual. Este seguimiento consistirá en visitas clínicas, pruebas de imagen y evaluación de la deglución.

BENEFICIOS Y RIESGOS DEL ESTUDIO

El principal beneficio que se espera para los participantes de este estudio, es reducir las complicaciones que puedan padecer como consecuencia del tratamiento radioterapéutico. Dichas complicaciones pueden ser desde una simple molestia al tragar hasta una grave secuela que impida deglutir correctamente, puesto que el alimento pueda pasar a la vía aérea generando nuevas complicaciones.

Estas complicaciones derivan del tratamiento actual estándar, que es considerado como el más efectivo en términos generales. Sin embargo, visto que los pacientes con un tumor relacionado con el VPH tienen un mejor pronóstico, se considera que se les está administrando actualmente un tratamiento más agresivo de lo necesario para su control oncológico que asocia además, unos efectos indeseados potencialmente evitables. Es por eso que en este estudio se propone reducir las dosis de radioterapia, considerando que el riesgo de que esto suponga un peor control de la enfermedad es mínimo. Además, para limitar el posible perjuicio en la salud, decidimos realizar esta pauta en un grupo muy seleccionado de personas, siguiendo unos criterios de inclusión y exclusión estrictos que permiten identificar a los individuos tienden a tener una evolución más favorable.

ALTERNATIVAS AL PROCEDIMIENTO

La alternativa al tratamiento de estudio considerada como de elección es recibir un tratamiento adyuvante a dosis completas de 60 Gy.

INTERRUPCIÓN DEL ESTUDIO

Si durante la realización del estudio se observara un detrimento en el control oncológico o un aumento de efectos indeseados no esperados en alguno o ambos grupos, el estudio se interrumpiría.

CONFIDENCIALIDAD

Desde el comienzo de su participación en este estudio, todos los datos personales que se recojan serán gestionadas y almacenadas con total confidencialidad ajustándose a la actual legislación de la *Ley Orgánica 3/2018, de 5 de diciembre, de Protección de datos personales y garantía de los derechos digitales*. Dicha información será identificada con un número y será usada tan sólo con fines de investigación.

El acceso a ella estará sólo a disponibilidad a investigadores y otras autoridades sanitarias. Usted dispone el derecho de poder consultar la información recopilada y corregirla en caso de error. Le garantizamos que ninguna información personal será publicada.

DIFUSIÓN DE LOS RESULTADOS

Cuando haya finalizado el estudio y se hayan extraído conclusiones, la intención es publicar dichos resultados en revistas científicas. De esta forma, otros centros asistenciales y pacientes con la misma afección podrán beneficiarse.

Como se ha comentado anteriormente, en dichas publicaciones no aparecerá ningún dato personal.

PARTICIPACIÓN Y COMPENSACIÓN ECONÓMICA

Los investigadores que participan en este estudio no obtienen beneficio económico. Además, debe comprender que su participación en este estudio es estrictamente voluntaria. Por lo tanto, si usted decide participar no recibirá ningún tipo de compensación económica, pero tampoco les supondrá ningún gasto. En el caso contrario, tampoco supondrá un cambio en cuanto a su atención médica por el equipo de especialistas.

Si usted decide participar, deberá firmar la hoja de consentimiento informado conforme da su aprobación. Es también su derecho poder salir del estudio si en algún momento de su transcurso así lo decide; dicho hecho tampoco alterará su atención médica, aunque le rogamos que lo comunique a alguno de los profesionales del Servicio de Otorrinolaringología de su hospital.

Antes de decidir sobre su participación, usted es libre de pedir una segunda opinión a otros profesionales médicos si así lo requiere.

RESPONSABILIDAD Y ASEGURANZA

Los promotores de este estudio tienen contractada una póliza de seguro para su realización, tal como se establece en la legislación. En caso de perjuicio o de detrimento de su salud en consecuencia a la participación de su participación en este estudio, se le proporcionará la indemnización correspondiente.

CONTACTO

En caso de cualquier duda durante la realización de este estudio, podrá ponerse en contacto siempre que lo necesite con:

_____

FULL DE INFORMACIÓ SOBRE EL ASSAIG CLÍNIC

Nom de l'estudi: *Desescalada en las dosis de radioteràpia adjuvant en pacients afectes per tumors d'orofaringe relacionats amb el VPH amb una xifra elevada de limfòcits CD8⁺ infiltrants en el tumor .*

Centre assistencial:

Investigador/a principal:

Benvingut/da,

Ens dirigim a vostè per convidar-lo a participar en un estudi de investigació, dut a terme pels Serveis d'Otorinolaringologia de diversos hospitals de referencia a Catalunya. Aquest estudi ha sigut aprovat pel Comitè d'Ètica i Investigació Clínica de l'Hospital Universitari Doctor Josep Trueta i per l'Agència Espanyola del Medicament i Productes Sanitaris.

La nostra intenció es que vostè comprengui el motiu pel qual s'està realitzant aquest estudi i què implica formar-hi part, per a que així, pugui decidir de manera voluntària si finalment desitja participar-hi o no. És per això que li preguem que es prengui el temps necessari per llegir detingudament i comprendre aquest resum informatiu sobre el nostre estudi. No té perquè decidir avui sobre la seva participació; en cas que qualsevol dubte sorgís, el nostre equip el respondrà i posarà a la seva disposició tota la informació necessària.

DESCRIPCIÓ I OBJECTIU DE L'ESTUDI

Clàssicament, els tumors d'orofaringe s'havien relacionat amb consums elevats de tabac i/o alcohol. Tot i així, la freqüència dels tumors d'orofaringe va en augment, i això es deu a un nou agent causal: el virus del papil·loma humà (VPH).

S'ha observat que els pacients el tumor dels quals està relacionat amb el VPH solen ser més joves i normalment no han tingut consums alts de tòxics com a factor de risc. En els últims anys, a més, s'ha vist que el curs de la malaltia en aquests pacients és significativament millor que en els que el tumor no és causat pel virus, tenint menys recurrències i una supervivència més perllongada. Això s'atribueix a que aquests tumors són més sensibles a la radioteràpia. Entre aquest grup de pacients, aquells en el que el estudi microscòpic s'observen més cèl3lules immunitàries tipus CD8⁺ tenen encara millor pronòstic.

Tot i aquestes observacions es segueix aplicant el mateix tractament a tot pacient amb tumor d'orofaringe, independentment de la causa. Es tracta d'un tractament eficaç però

agressiu, que sol estar format per una intervenció quirúrgica inicial, seguida de radioteràpia i a vegades de quimioteràpia, segons els característiques de cada tumor.

D'altra banda, aquest tractament no està exempt d'efectes adversos, un dels principals és la disfàgia, o la dificultat en la deglució. A més de molesta, pot arribar a ser greu associant-se a altres complicacions que poden arribar a requerir assistència mèdica addicional. La seva aparició està directament relacionada amb la dosis administrada de radioteràpia, i que succeeix en el voltant de un 30% dels pacients que la reben.

Aquest estudi va dirigit a pacients amb tumors d'orofaringe causats per el virus del papil·loma humà, i que a més tenen com factor pronòstic favorable addicional xifres elevades de limfòcits $CD8^+$ en el estudi microscòpic.

La motivació darrera d'aquest projecte de investigació és demostrar que la reducció en les dosis de la radioteràpia no suposa un perjudici en el control oncològic i a més suposaria evitar part de les seves complicacions associades i/o reduir la seva gravetat. Es proposa un nou protocol de tractament, en el que es reduiran les dosis de radioteràpia administrada, 10 Gy menys en comparació al tractament estàndard.

METODOLOGIA I INTERVENCIÓ

En aquest estudi participaran un total de 400 pacients; a tots ells se'ls operarà amb cirurgia robòtica. El total de pacients serà distribuït aleatòriament en dos grups (**A** i **B**) de la mateixa mida (200) que estaran estratificats pel tipus de tractament adjuvant indicat segons les característiques del seu tumor (radioteràpia o quimioradioterapia) formant dos subgrups també de la mateixa mida (100) dins dels grups principals.

- El **grup A** rebrà la radioteràpia a les dosis del tractament estàndard que correspon a 60 Gy en 30 cicles distribuïts en 6 setmanes. S'associarà la quimioteràpia o no segons les característiques de la lesió.
- El **grup B** rebrà radioteràpia a dosis menors que el tractament estàndard, que correspondrà a 50 Gy en 25 cicles al llarg de 5 setmanes. L'associació de quimioteràpia adjuvant es farà segons les característiques de la lesió.

Degut a les diferències en nombre de cicles i duració total del tractament radioterapèutic, tant el pacient com el metge sabran a quin dels grups pertany el pacient.

Tots els participants, independentment del grup de tractament al que pertanyin, seran seguit de la mateixa manera que en la pràctica clínica habitual. Aquest seguiment consistirà en visites clíniques, proves de imatge i avaluació de la deglució.

BENEFICIS I RISCOS DE L'ESTUDI

El principal benefici que s'espera per als participants d'aquest estudi és reduir les complicacions que puguin patir com a conseqüència del tractament radioteràpic. Aquestes complicacions poden ser des d'una simple molesta al deglutir fins a una seqüela greu que impedeixi tragar correctament, donat que l'aliment pot passar a la via respiratòria generant noves complicacions.

Les esmentades complicacions deriven del tractament actual estàndard, que és considerat com el més efectiu en termes generals. Tot i així, vist que els amb un tumor relacionat amb VPH tenen un millor pronòstic, es considera que se'ls hi està administrant actualment un tractament més agressiu del necessari per al seu control oncològic, que associa uns efectes adversos potencialment evitables. És per això que en aquest estudi es proposa reduir les dosis de radioteràpia, considerant que el risc que això suposi un pitjor control de la malaltia és mínim. A més, per assegurar no perjudicar la salut, decidim realitzar aquesta pauta en un grup molt seleccionat de individus, seguint uns criteris de inclusió i exclusió estrictes que permet identificar les persones que tendeixen a tenir una evolució més favorable.

ALTERNATIVES AL PROCEDIMENT

L'alternativa al tractament d'estudi considerada d'elecció es rebre un tractament adjuvant a dosis completes de 60 Gy.

INTERRUPCIÓ DE L'ESTUDI

Si durant la realització de l'estudi s'observés un detriment en el control oncològic o un augment d'efectes adversos no esperats en algun o ambdós grups, el estudi s'aturaria.

CONFIDENCIALITAT

Des del principi de la seva participació en aquest estudi, totes les dades personals que es recullin seran gestionades i emmagatzemades amb total confidencialitat ajustant-se a la actual legislació de la *Llei Orgànica 3/2018, de 5 de desembre, de Protecció de dades personals i garantia dels drets digitals*. Aquesta informació serà identificada amb un número i tant sòls se'n farà ús amb fins de investigació. El seu accés només estarà a disposició dels investigadors i altres autoritats sanitàries. Vostè disposa el dret de poder consultar la informació recopilada i corregir-la en cas d'error. Li garantim que ninguna informació personal serà publicada

DIFUSIÓ DELS RESULTATS

Quan hagi finalitzat l'estudi i s'hagin extret conclusions, la intenció es publicar els resultats a revistes científiques. D'aquesta manera, altres centres assistencials i pacients amb la mateixa patologia se'n podran beneficiar. Com s'ha comentat anteriorment, en aquestes publicacions no apareixerà cap dada personal.

PARTICIPACIÓ I COMPENSACIÓ ECONÒMICA

Els investigadors que participen en aquests estudi no obtenen benefici econòmic. A més, ha de comprendre que la seva participació en aquest estudi és estrictament voluntària. Per tant, si vostè decideix participar no rebrà cap tipus de compensació econòmica, però tampoc li suposarà cap despesa, En el cas contrari, tampoc implicarà un canvi en la seva atenció mèdica per part de l'equip d'especialistes.

Si vostè decideix participar, haurà de firmar la fulla de consentiment informat conforme dona la seva aprovació. Es també el seu dret poder sortir de l'estudi si en algun moment del seu transcurs així ho decideix i ho comunica; això tampoc implicarà cap canvi en l'assistència sanitària rebuda, tot i que li preguem que ho comuniqui a algun dels professionals del Servei d'Otorinolaringologia del seu hospital

RESPONSABILITAT I ASSEGURANÇA

Els promotors d'aquest estudi tenen contractada una pòlissa d'assegurança per a la seva realització, tal com s'estableix en la legislació. En cas de perjudici o de detriment de la seva salut com a conseqüència de la seva participació en aquest estudi, se li proporcionarà la indemnització corresponent.

CONTACTE

En cas de qualsevol dubte durant la realització d'aquest estudi, podrà posar-se en contacte sempre que ho necessiti amb:

14.4. ANNEX 4: INFORMED CONSENT

DOCUMENTO DE CONSENITMIENTO INFORMADO DEL PACIENTE

Yo, _____, con documento de identificación personal (DNI/NIE) _____ declaro que:

- He recibido una copia de la hoja de información para el paciente.
- He leído y comprendido toda la información que aparece en la hoja de información para el paciente.
- He podido exponer cualquier duda que me ha surgido, y me la han resuelto adecuadamente.
- Estoy conforme con la cantidad de información que me ha sido proporcionada.
- Comprendo que mi participación es voluntaria y no remunerada.
- Entiendo los potenciales riesgos y beneficios derivados de participar en este estudio.
- Comprendo que mis datos y pruebas serán confidenciales.

Acepto que los investigadores del proyecto puedan ponerse en contacto conmigo en un futuro si se considera oportuno

☐ Sí ☐ No

Además, comprendo que aún y haber firmado el consentimiento informado, puedo revocarlo en cualquier momento y que esto no supondrá un perjuicio en mi tratamiento y asistencia sanitaria.

Firma del paciente **Firma del investigador**

Lugar y fecha: _____, _____ de _____ del año _____

REVOCACIÓN DEL CONSENTIMIENTO INFORMADO

Yo, _____, con documento de identificación personal (DNI/NIE) _____ revoco el consentimiento previamente firmado para la participación en el ensayo clínico: ***Desescalada en las dosis de radioterapia adyuvante en pacientes afectados por tumores de orofaringe relacionados con el VPH con una cifra elevada de CD8⁺ TILs.***

Firma del paciente: **Firma del investigador:**

Lugar y fecha: _____, _____ de _____ del año _____

DOCUMENT DE CONSENTIMENT INFORMAT DEL PACIENT

Jo, _____, amb document de identificació personal (DNI/NIE) _____ declaro que:

- He rebut una copia del full de informació per al pacient.
- He rebut i entès tota la informació que apareix en el document de informació per al pacient.
- He pogut plantejar qualsevol dubte que m'ha sorgit, i me l'han resolt adequadament.
- Estic conforme amb la quantitat de informació que se m'ha proporcionat.
- Entenc que la meva participació és voluntària i no remunerada.
- Entenc els potencials riscos i beneficis derivats de participar en aquest estudi.
- Comprenc que les meves dades i proves seran confidencials.

Accepto que els investigadors del projecte puguin posar-se en contacte amb mi en un futur si es considera oportú.

☐ Sí ☐ No

A més, comprenc que tot i haver firmat el consentiment informat, puc revocar-lo en qualsevol moment i que això no suposarà un perjudici en el meu tractament i assistència sanitària.

Firma del pacient **Firma de l'investigador**

Lloc i data _____, _____ de _____ de l'any_____

REVOCACIÓ DEL CONSENTIMENT INFORMAT

Jo, _____, amb document de identificació personal (DNI/NIE) _____, revoco el consentiment prèviament firmat per a la participació en el assaig clínic: ***Desescalada en les dosis de radioteràpia adjuvant en pacients afectes per tumors d'orofaringe relacionats amb el VPH amb una xifra elevada de CD8⁺ TILs***

Signatura del pacient: **Signatura de l'investigador:**

Lloc i data: _____, _____ de _____ de l'any _____