The transformation zone: fall of a myth? Paradigm shift?

ONGOING WORK

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Reserve Cell - Manuscript Text a | Recibidos (1.290) - desanjose.sil | Post Attendee - Zoom | Post Attendee - Zoom

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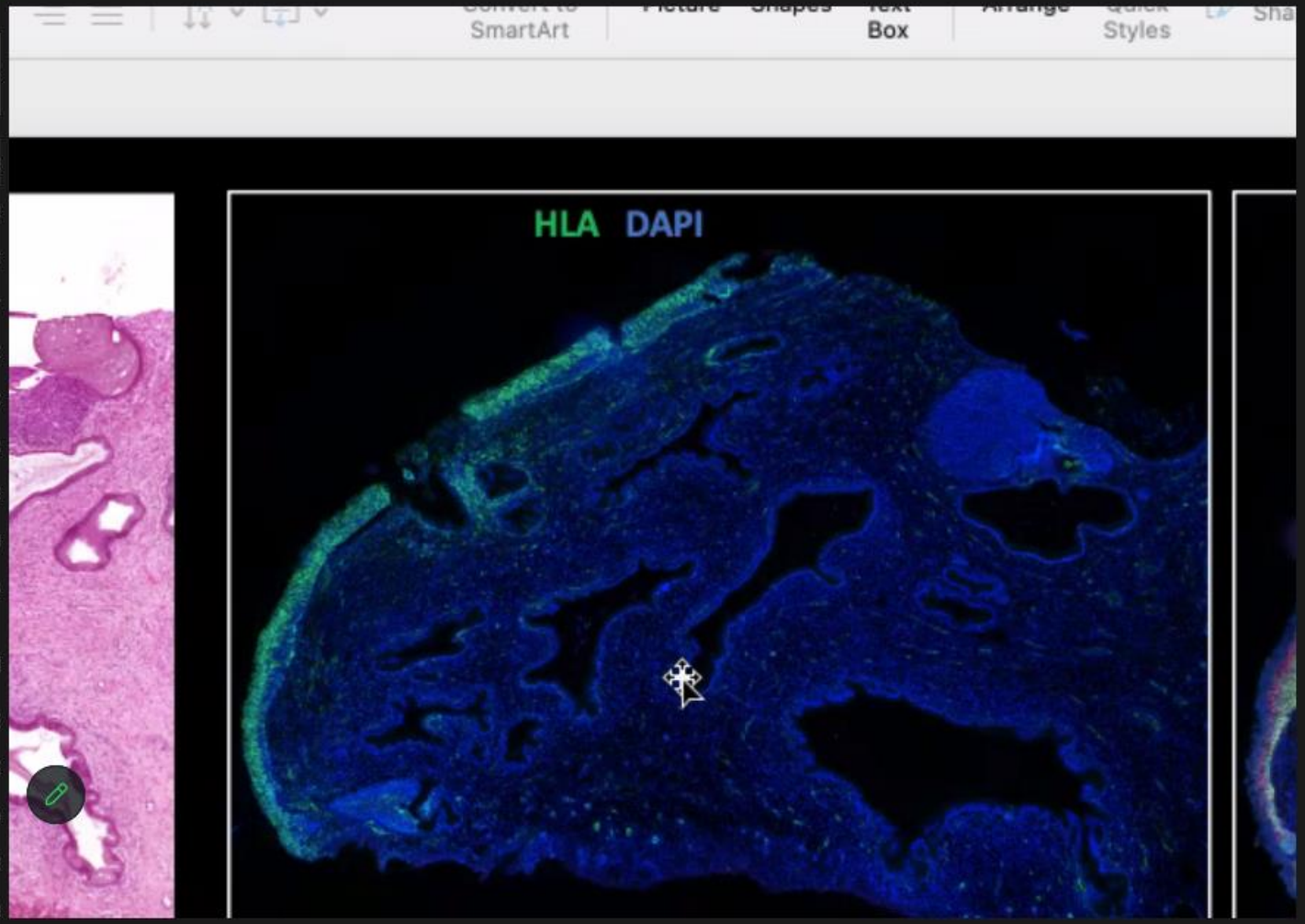
Zoom Meeting



Tanvier.Omar

Tanvier.Omar

Participants (7)

- Silvia de Sanjosé (Host, me)
- JD John Doorbar
- MS Mark Schiffman
- T Tanvier.Omar
- Chemtai Mungo
- HK Helen Kelly

Invite | Mute All | ...

Meeting Chat

can we delay 30 minutes our call for tomorrow?

Helen Kelly to You (Direct Message) 3:12 PM

HK Yes no problem !

You to Helen Kelly (Direct Message) 3:15 PM

Thanks

1 New Message

Who can see your messages?

CGR-ScreenFire
in 5 minutes
Meeting ID: 870 8449 3952

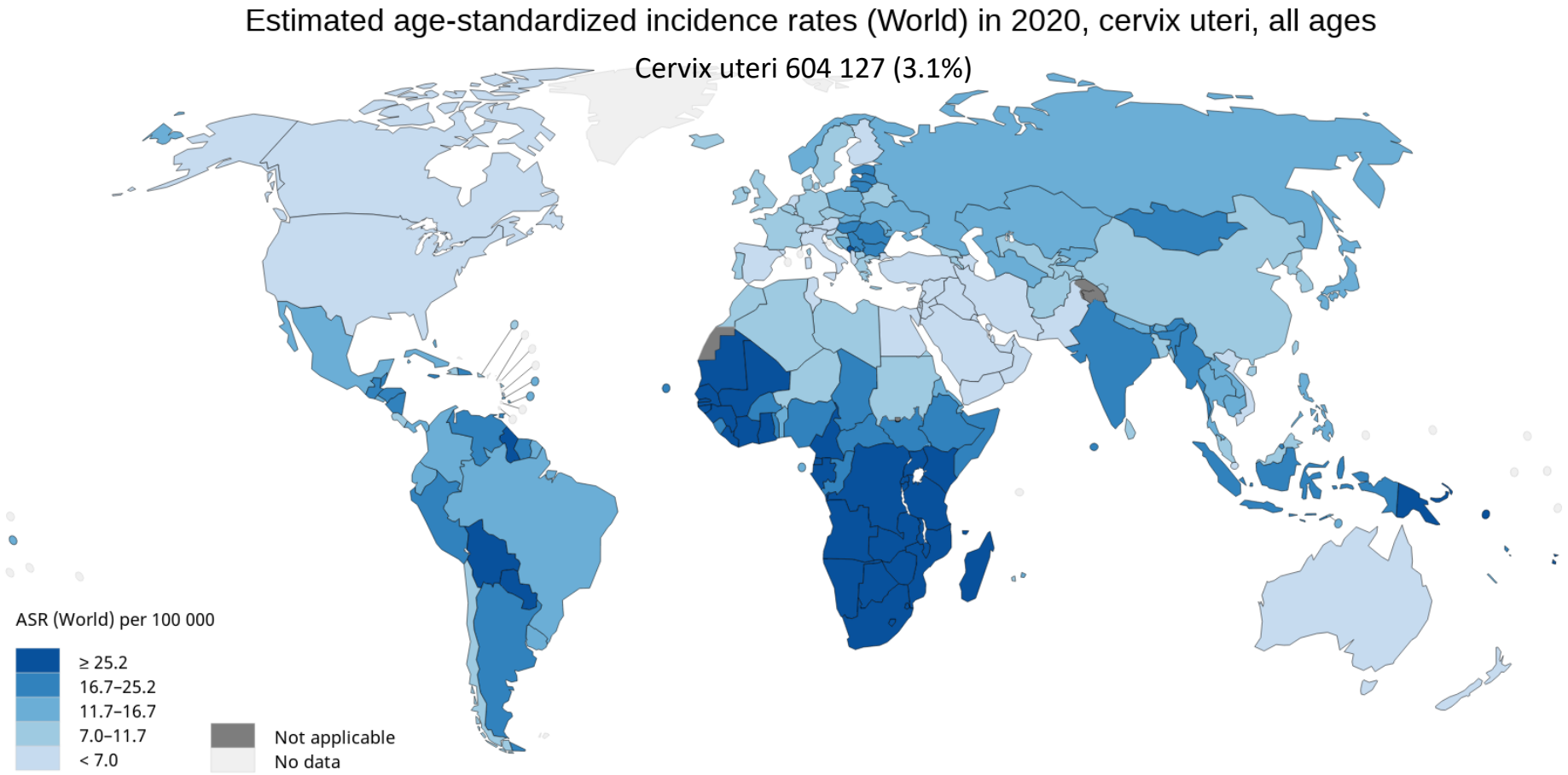
Start | Snooze

AI EMANY VII CHES MA RE: Seguiment - tractament infecció VPH - ara he quedat per a parlar amb pacho per 11:46

Recibidos ... | File Explorer | StataSE 14... | Skype [10] | Outlook | Zoom Me...

ESP 3:55 PM 10/5/2023

CERVICAL CANCER REMAINS THE CANCER OF THE GLOBAL INEQUALITY

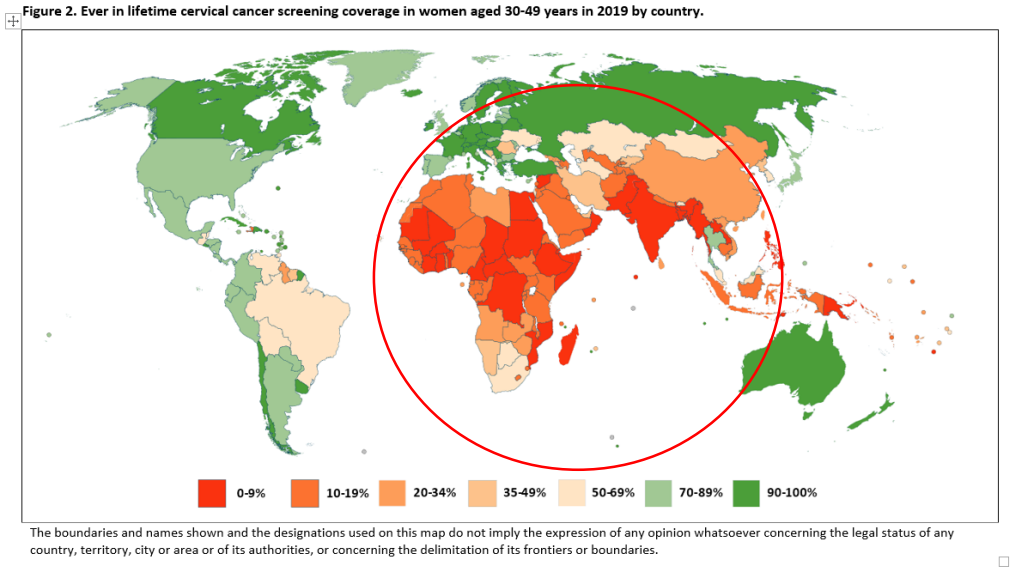


Data source: GLOBOCAN 2020
Graph production: IARC (http://gco.iarc.fr/today)
World Health Organization

WHO in 2030 to reach in **2100 Incidence 4x100,000**
Vaccinate 90%, Screen 70%, Treat 90%

Vaccination

Screening



Treatment

?

Still 245M of girls 9-14 are not receiving HPV vaccination

Many parts of the world have less than 10% of the population screened with limited efficacious tests

No global compliance parameters

WHO elimination campain

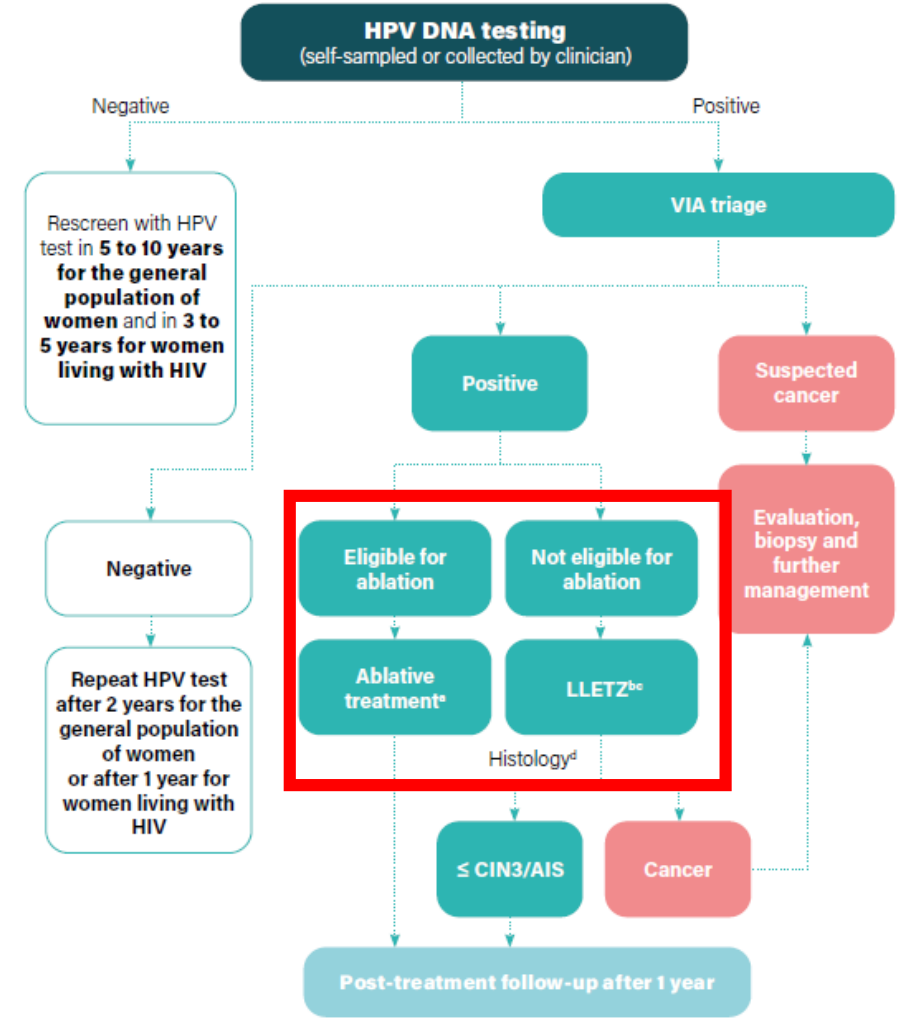
in 2030 to reach in **2100 Incidence 4x100,000**

Vaccinate 90%, Screen 70%, Treat 90%

Thermal ablation may be the only feasible approach to reach high coverage

ALGORITHM 5. PRIMARY HPV DNA SCREENING AND VIA TRIAGE (SCREEN, TRIAGE AND TREAT APPROACH)

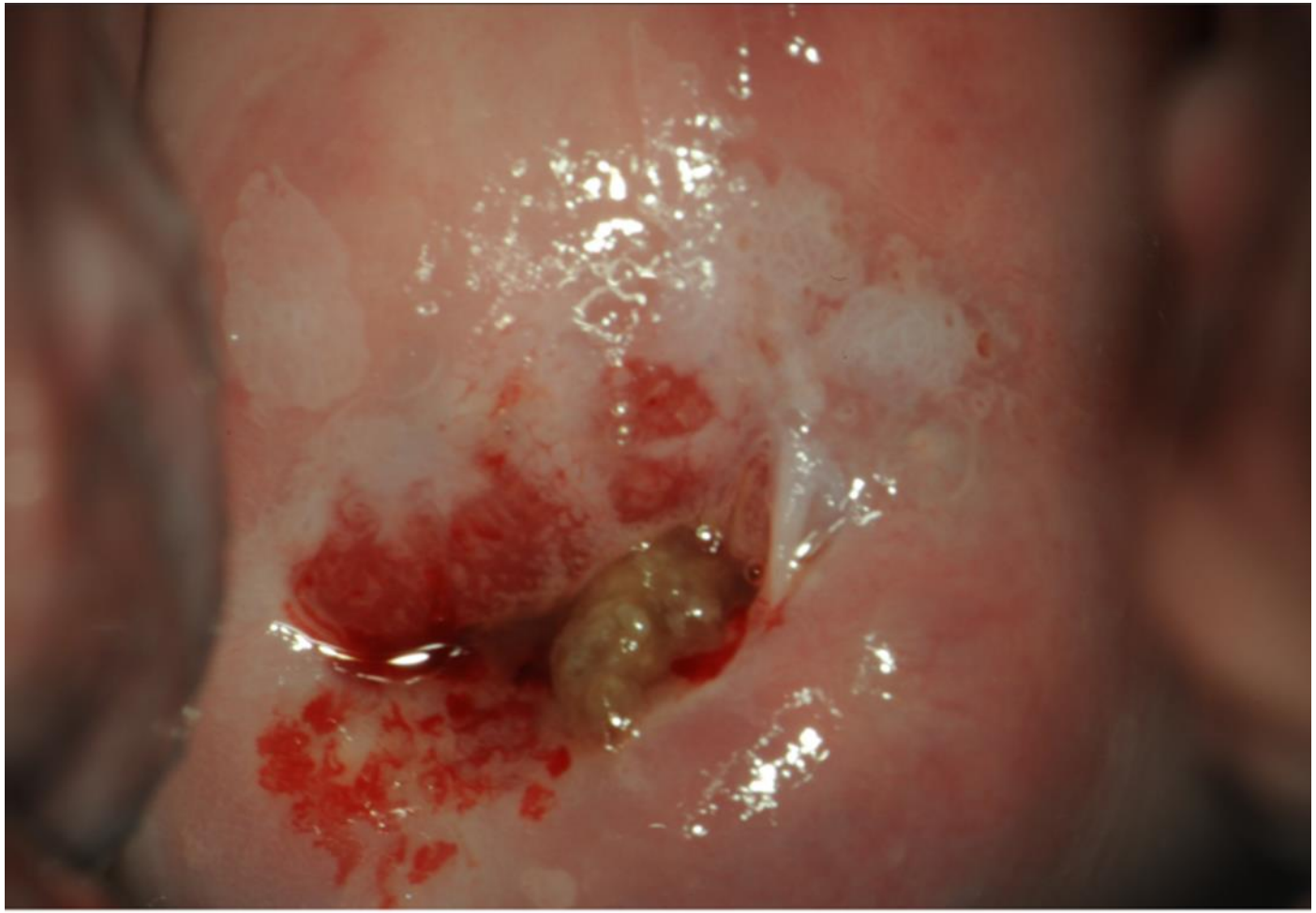
For both the general population of women and women living with HIV



[a] Ablative treatment includes cryotherapy and thermal ablation.
[b] Cold knife conization (CKC) if LLETZ not available.
[c] LLETZ and LEEP (loop electrosurgical excision procedure) indicate the same procedure.
[d] Histology may not be available in certain settings; women should be advised to attend follow-up after 1 year or to report earlier, if they have any of the symptoms of cervical cancer.
AIS: adenocarcinoma in situ; CIN: cervical intraepithelial neoplasia; HPV: human papillomavirus; LLETZ: large-loop excision of the transformation zone; VIA: visual inspection with acetic acid.

Loose indications based on the transformation zone concept

Management of precancerous lesions relies on how we define what we see

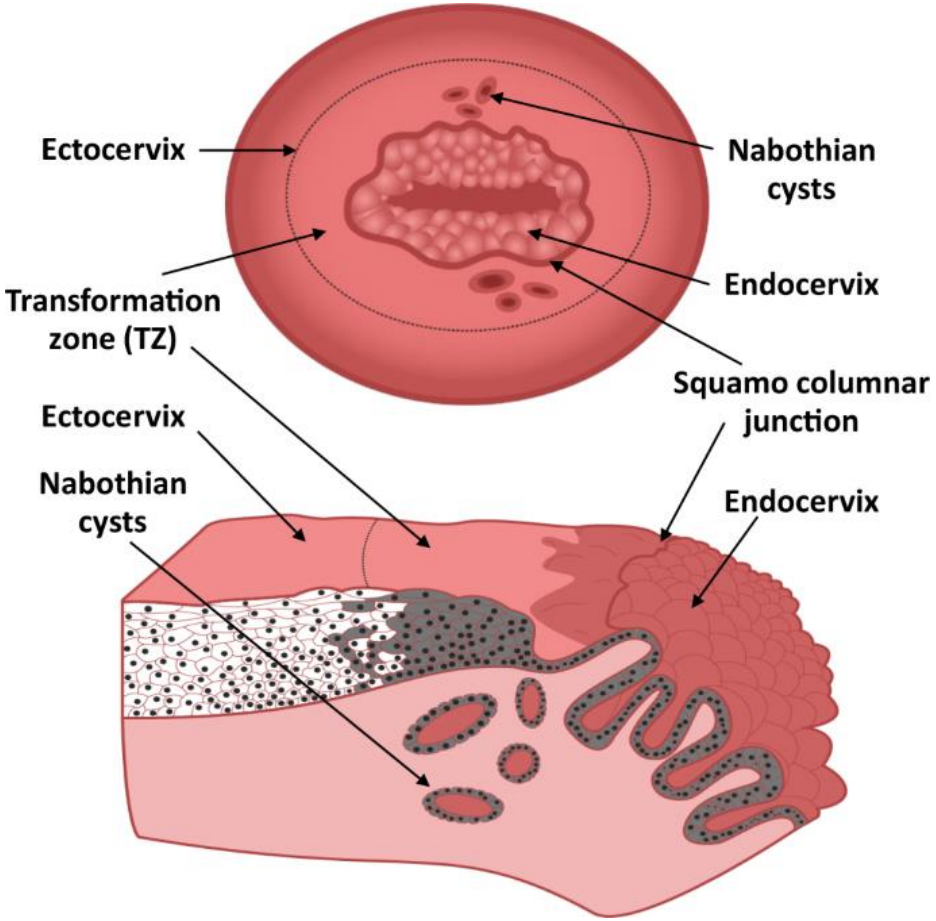


Is the cervix evaluable for treatability?

- ○ FULLY EVALUABLE – Current SCJ (and lesions) fully visible
- ○ EVALUABLE WITH CAVEAT – Current SCJ (and/or lesions) extends into canal and partly visible
- ◉ NOT EVALUABLE – Current SCJ is endocervical and/or completely not visible

Cervical precancerous lesions

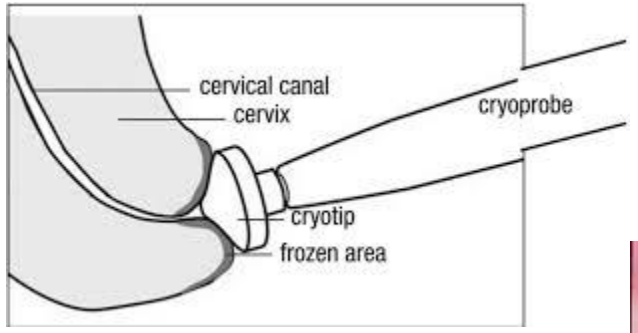
- The invasive potential of cervical precancer involve an accumulation of somatic mutations in cells at the junction of metaplastic squamous and glandular epithelium (SCJ).
- Cervical transformation zone (TZ),where metaplasia occurs*, is where we detect >90% of precancer
- Loose definition of what is TZ, Squamous columnar junction (SCJ) is more clearly defined

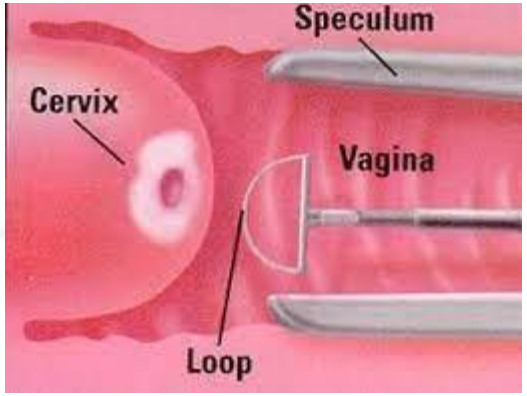


* Classical definition

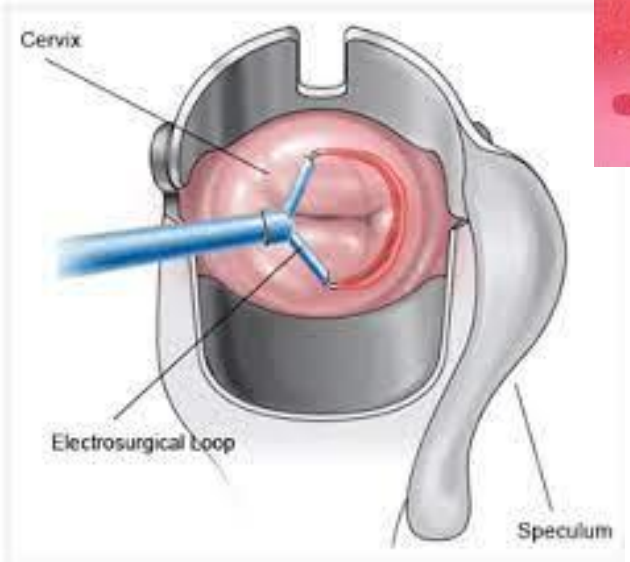
Management of cervical precancer lesions. The aim is minimal tissue removal to minimize adverse events.

- Cryotherapy (cold)
- Thermal ablation (heat)





- Loop electrosurgical excision procedure (LEEP) or Large loop excision of the transformation zone (LLETZ)
- Laser conization



High recurrences after treatment in WLWH

In the general population <5% at 5 years follow up

In women living with HIV

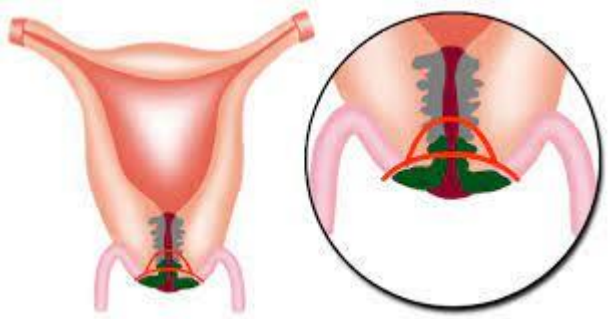
Type-specific persistence at 6 months associated with CIN2+ recurrence		
Overall	**Cryotherapy**	**LEEP**
73/196 (37%)	45/107 (42%)	28/89 (31%)

- Recurrent precancer
  - True recurrence of a clonal expansion from the original cell that generated the lesion -> initial failure of treatment, continued growth of the original clonal lesion, increased risk of secondary mutational events, and risk of invasion
  - Development of a new lesion -> implies much lower risk

Bogani et al. 2020; Fernandez et al. 2019; Chung M et al. JAMA 2021; Green et al

TopHat conization

- WLWH, over 2-year follow-up,
- Recurrences of CIN2+ after TopHat conization
  - inadequate colposcope exam (endocervical lesions) 35 (29%)
  - ectocervical lesions 19 (24%)
- Hazard ratio 1.32; 95% confidence interval 0.75–2.31; P = 0.338



Lower recurrences

Int J Gynecol Obstet. 2021;152:118–124.

But, why recurrences?

The team of the ´Recurrence’ project

- Epidemiology: Mark Schiffman, Kanan Desai, Helen Kelly, Silvia de Sanjosé at NCI, US
- Molecular Evaluation: John Doorbar, Ademola Aiyenuro, Heather Griffin, Konstanze Schichl, at Cambridge, UK
- Case selection/Pathology:
  - Tanvier Omar, South Africa
  - Jaume Ordi & Marta del Pino, Spain
  - Michael Chung, Kenya
  - Chemtai Mungo, Malawi

Overall objectives of the ´Recurrence´ project

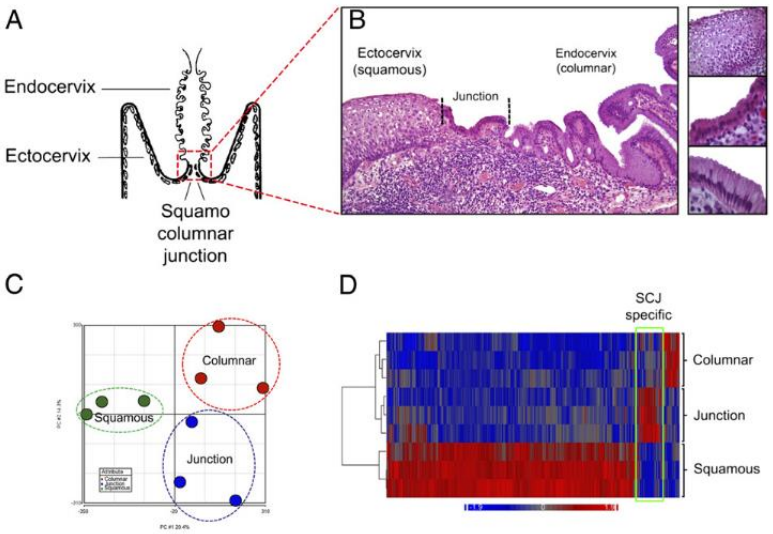
CLARIFYING THE MALIGNANT POTENTIAL OF NEW AND "RECURRENT" ANOGENITAL PRECANCERS IN HIV-INFECTED INDIVIDUALS TO GUIDE OPTIMAL MANAGEMENT OF HPV-HIV DUAL INFECTION

1. Describe the cellular composition of the transformation zone and confirm distribution and function of reserve cells (RC)
2. Evaluate impact of SCJ removal in the cervix after 1 year in patients with recurrences
3. Confirm patterns of recurrences in a long follow up cohort in Kenya
4. Provide implications for treatment of precancerous lesions

Funded by NCI & Cambridge University

- What is the impact of removing TZ/SCJ?

The junction cells hypothesis



…our findings suggest that carcinogenic HPV-related CINs and cervical cancers are linked to a small, discrete cell population that localizes to the SC junction of the cervix, expresses a unique gene expression signature, and is not regenerated after excision. Herfs et al. 2012 PNAS, Herfs & Crum Nature 2015

Krt7 are identified as the key cells in the junction

The Herfs theory

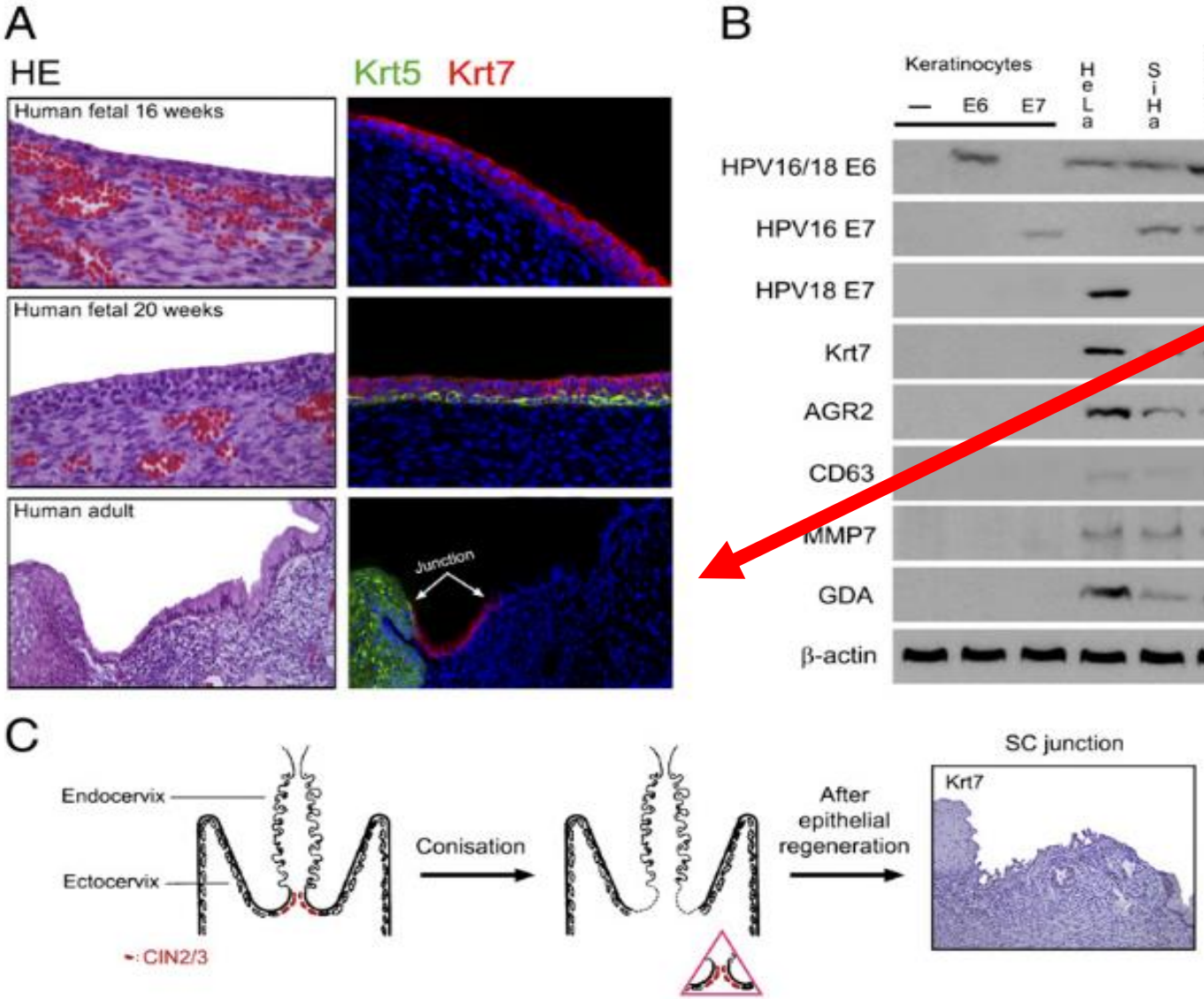
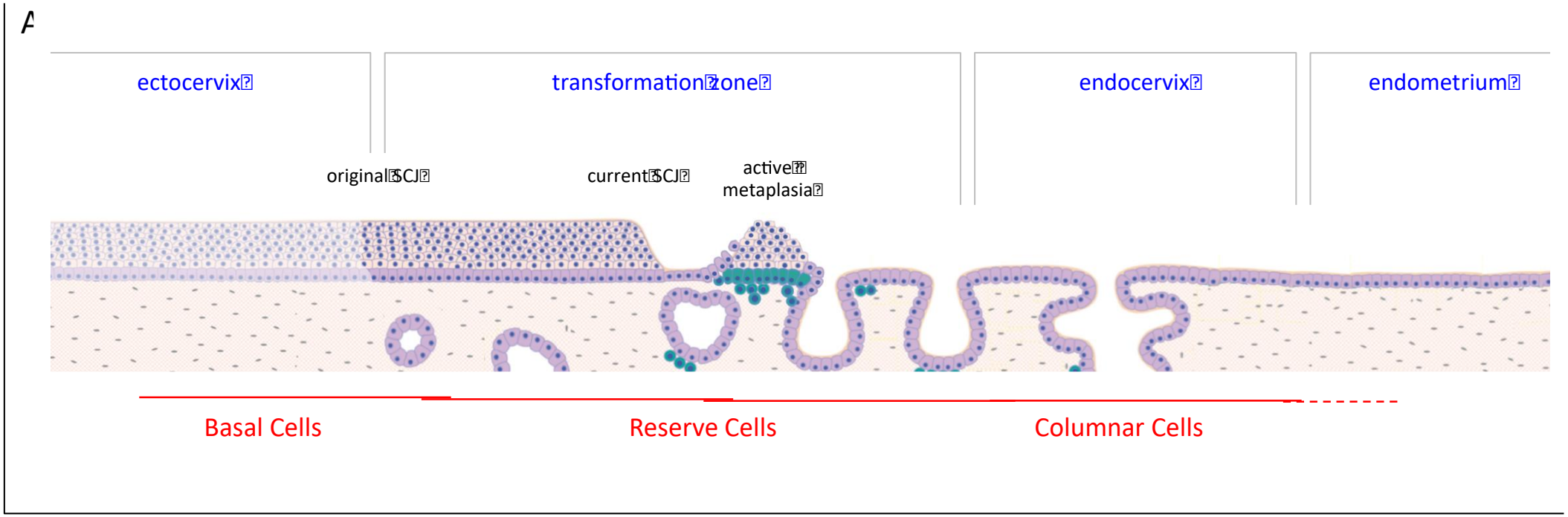


Fig. 4. Topographic specificity of the SC junction immunophenotype. (*A*) Fluorescence micrograph of human cervix at 16 wk (*Top*) showing diffuse Krt7 immunopositivity. At 20 wk of gestation, basal Krt5 expression emerges (*Middle*). In the adult cervix (*Bottom*), the Krt7 staining is limited to the SC junction. For each case, a corresponding histology image [hematoxylin–eosin staining (HE)] is shown. (*B*) Western blots of lysates of control (–), HPV16 E6- or E7-expressing primary human keratinocyte cultures, cervical adenocarcinoma (HeLa), and squamous carcinoma (SiHa, CaSki) reacted with antibodies specific for the SC junctional cells. Only cervix-derived tumor cells (HeLa, SiHa, and CaSki) score positive. (*C*) Schematic illustration of the squamocolumnar junction before and after LEEP (*Left* and *Center*). Absence of Krt7 staining in a "new" SC junction following LEEP (*Right*).

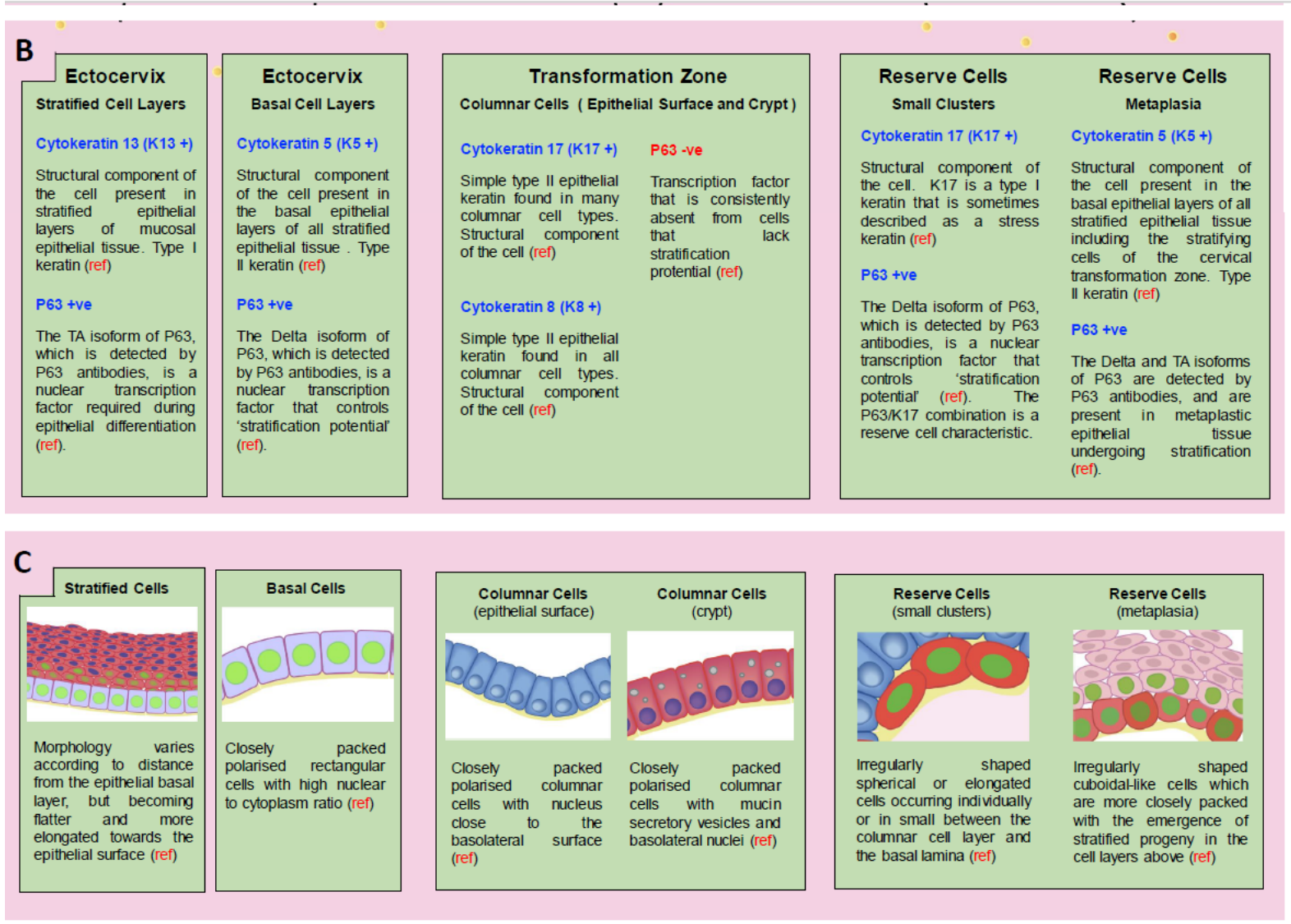
Current Model of HPV Disease



Site of Infection affects Disease Outcome

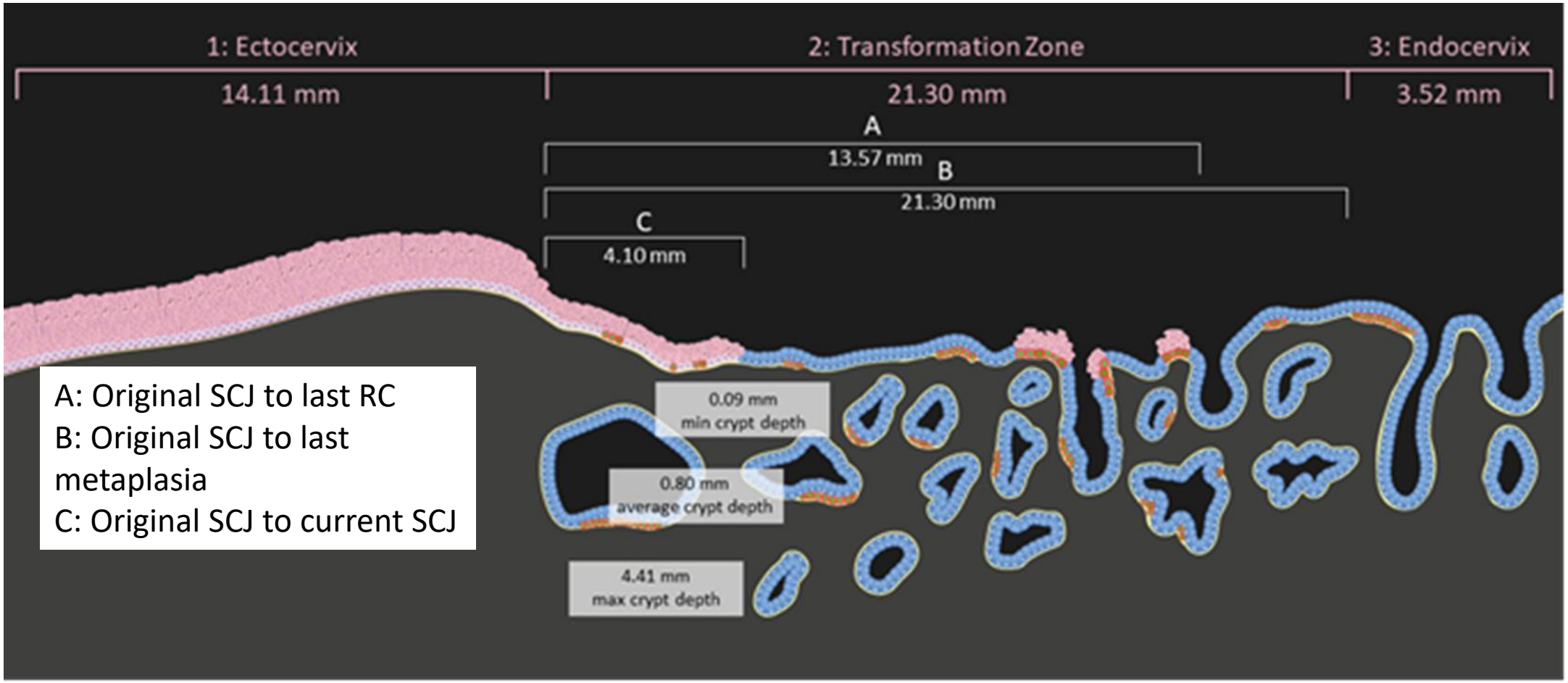
Doorbar J, Griffin H. (2019) Refining our understanding of cervical neoplasia and its cellular origins. Papillomavirus Res. 2019 Jun;7:176-179. PMID: 30974183

Five distinctive cell-types in the cervix architecture



Doorbar
et al in preparation 2023

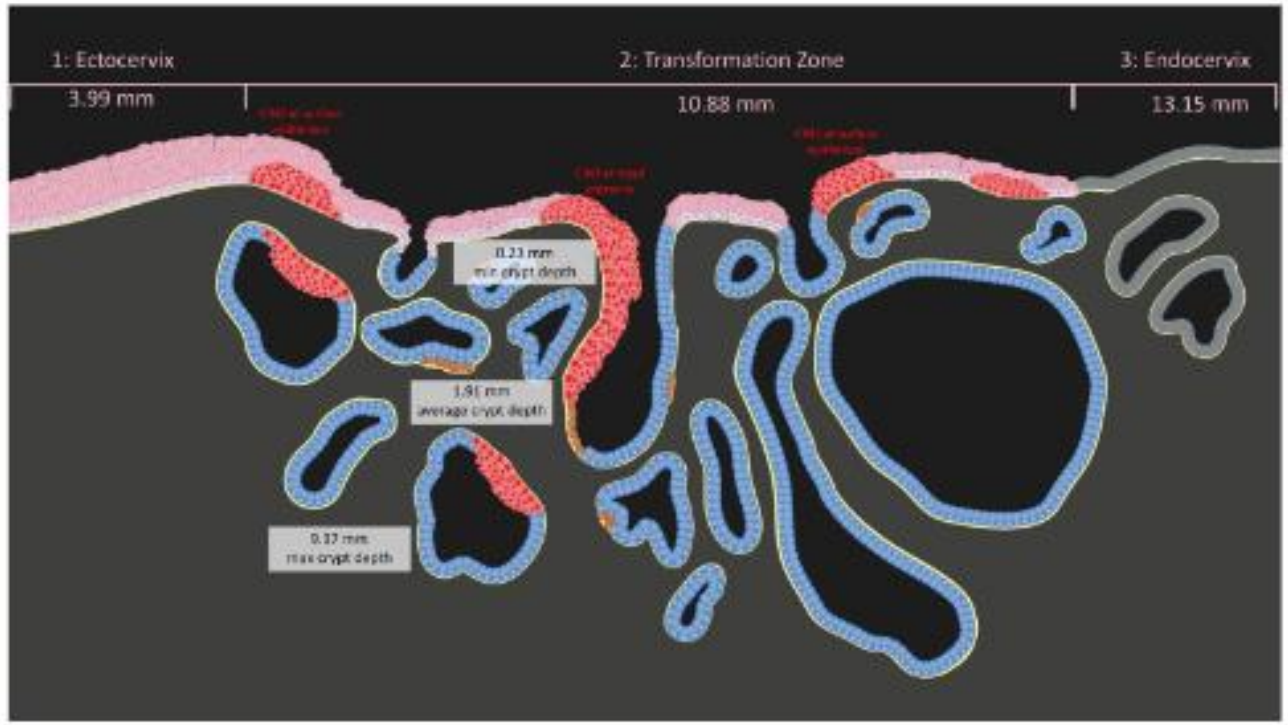
What are we calling the transformation zone ?



Measurements based on 15 hysterectomies and 15 conizations

Doorbar et al. in preparation

Table 2: **Disease types and location of crypts within the TZ**. Analysis of 15 cases treated for HPV Neoplasia to determine the average lengths of ectocervix, TZ including disease types, and endocervix, as well as the approximate location of original SCJ and last neoplasia. The average depth of crypts was also determined.



Preliminary findings

- Crypts are identified far distant form SCJ, including RC
- In conizations, crypts with RC are deeper than in the hysterectomy samples

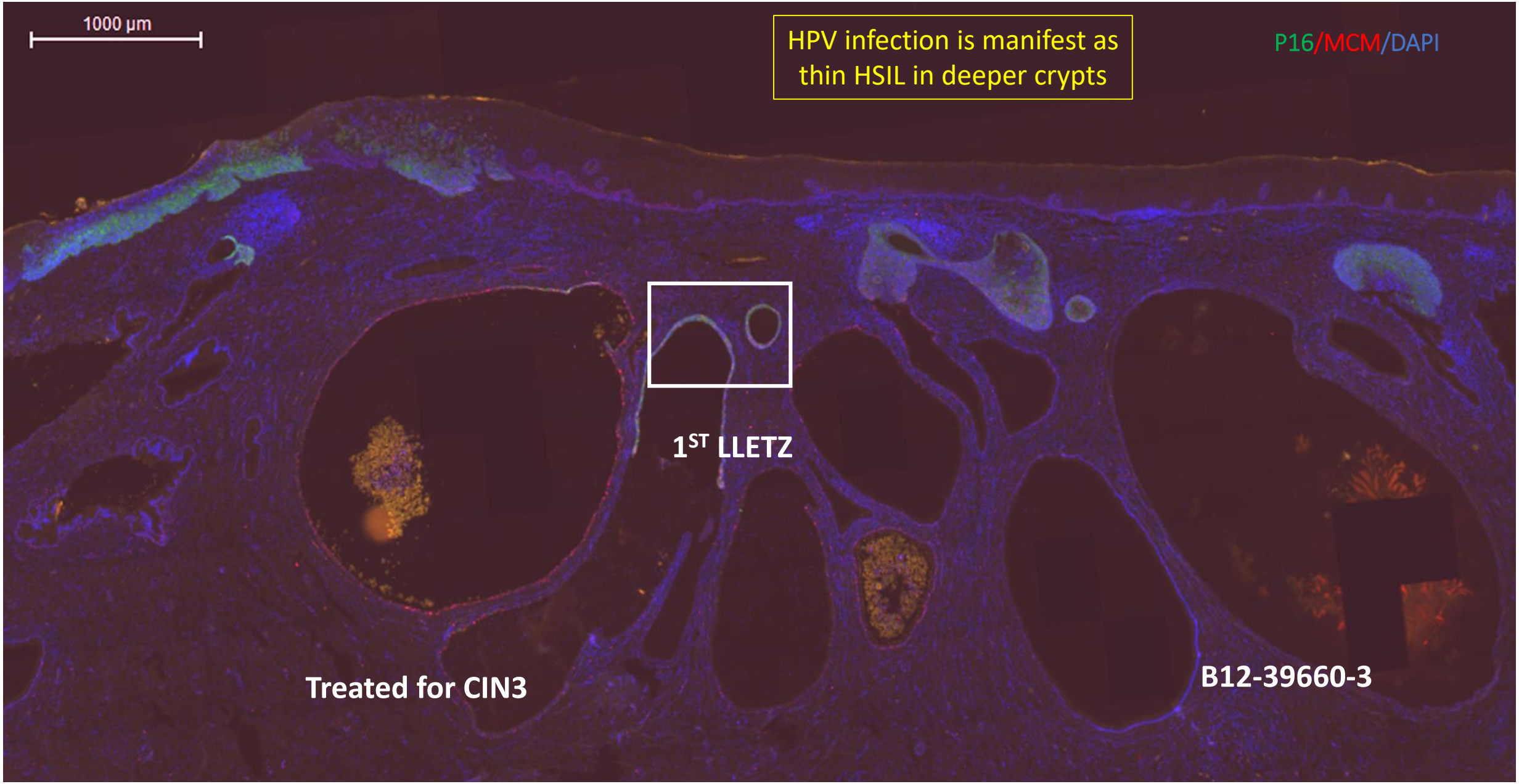
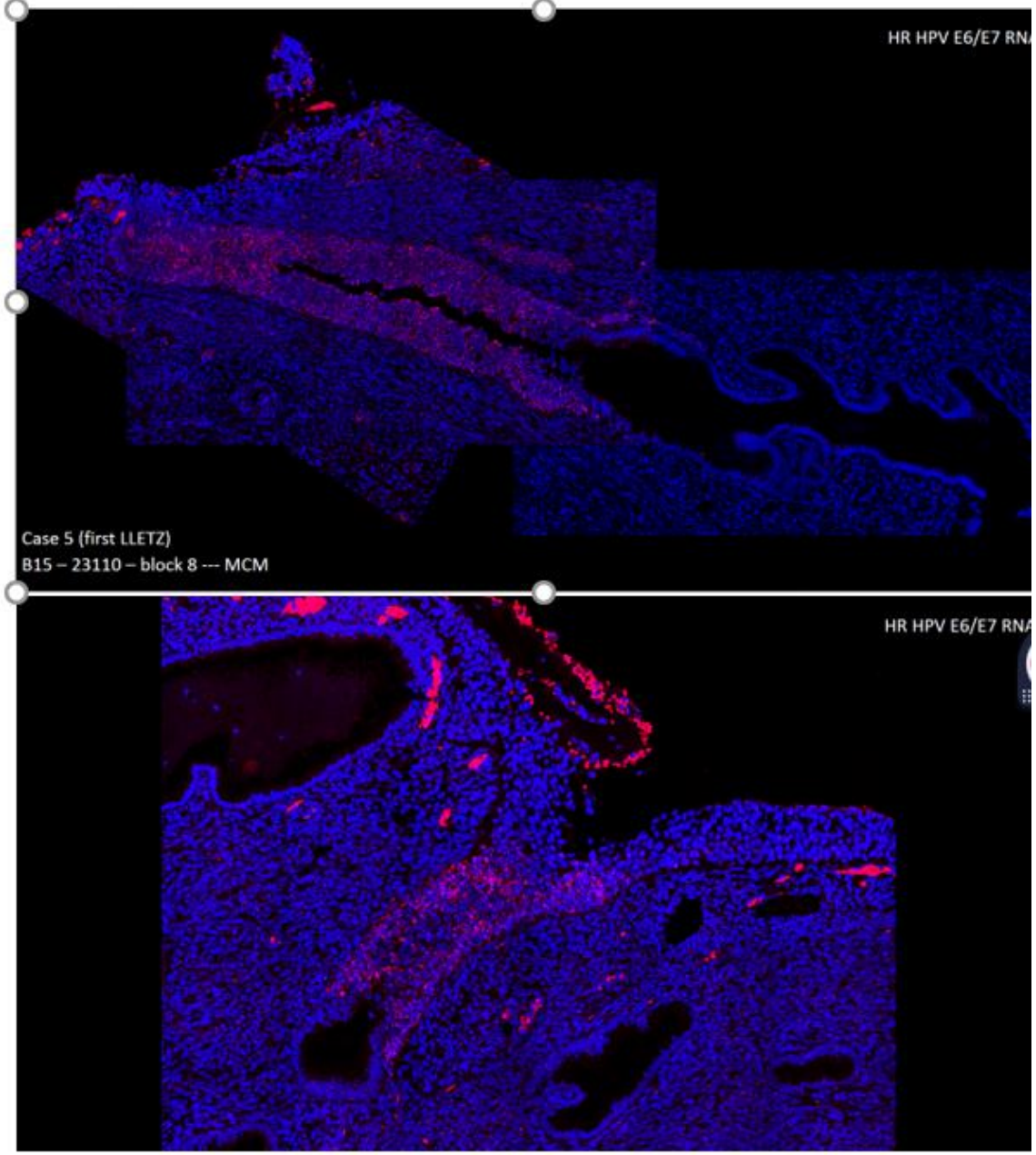
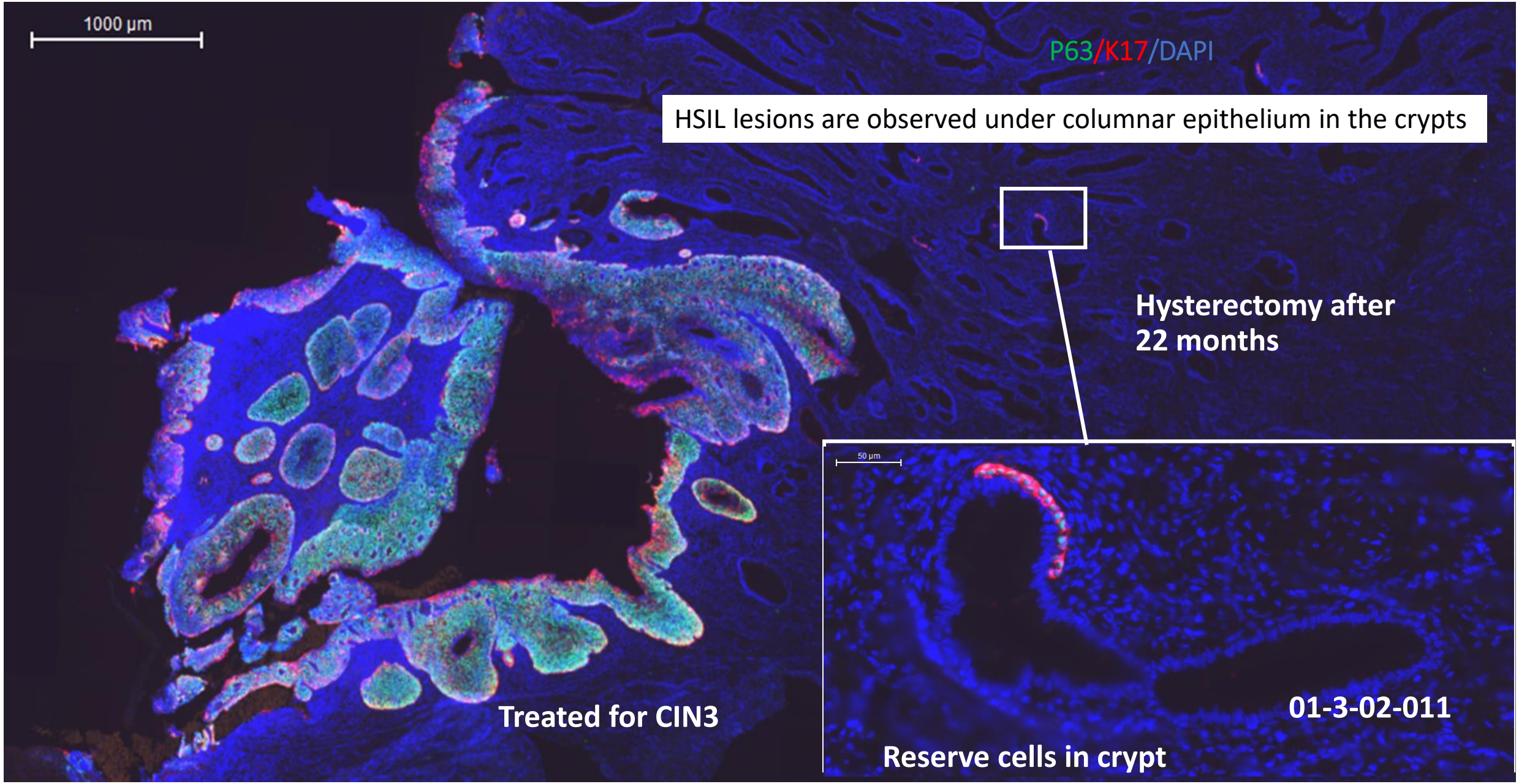

1000 µm
HPV infection is manifest as thin HSIL in deeper crypts
P16/MCM/DAPI
1ST LLETZ
Treated for CIN3
B12-39660-3

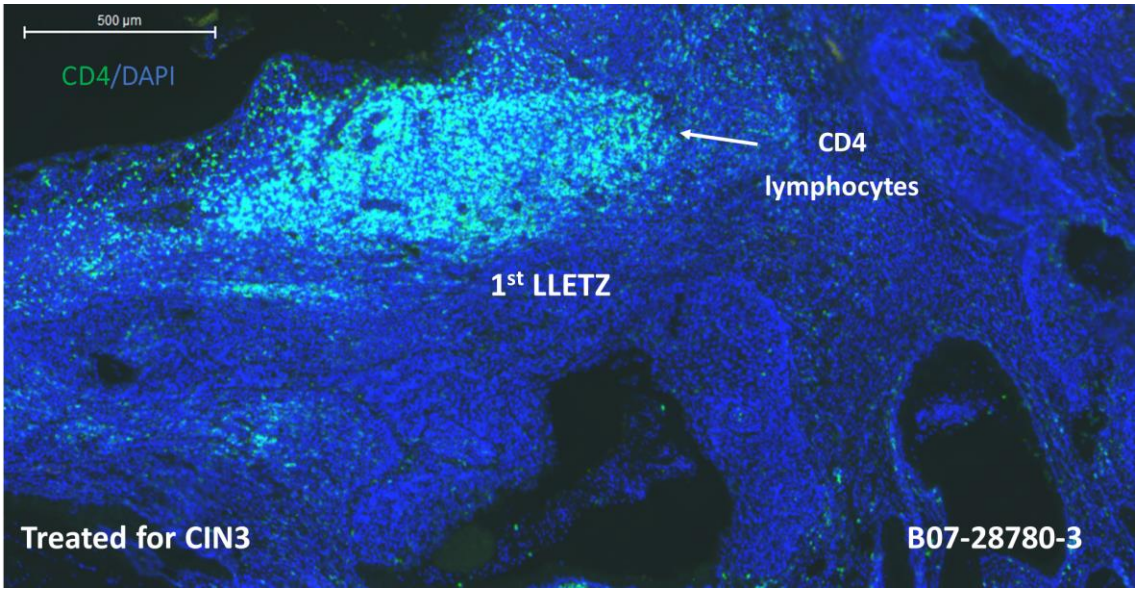
Figure 3. HR-HPV* E6/E7 positive staining (red) at crypt entrance



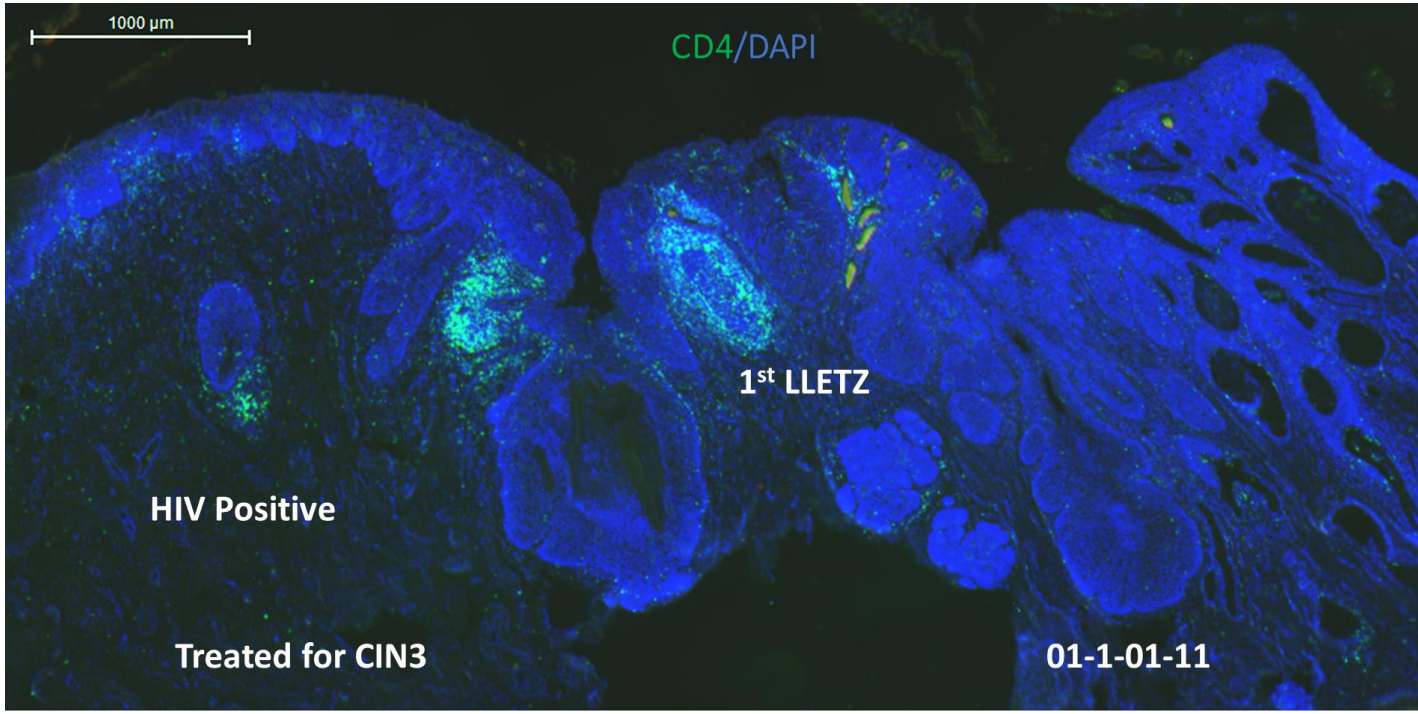
hrHPV is detected at the entry of the crypts


1000 µm
P63/K17/DAPI
HSIL lesions are observed under columnar epithelium in the crypts
Hysterectomy after
22 months
50 µm
Treated for CIN3
01-3-02-011
Reserve cells in crypt

Immune response show different patterns by HIV status



CD4 cells are more compact in non HIV
A more diffuse, and less dense accumulation of CD4 is more often identified in samples from WLWH



Discussion

- Our data suggest that RC are located under the columnar epithelium and far from SCJ
- The RC are identified in distant crypts, can be infected by HPV and can be identified as HSIL
- The impact of RC is not visible in regular colposcopy exams, but could be suspected if HPV persists after first treatment.
- To date, our data is not consistent with the existence of junctional cells.

Preliminary implications

- In settings where thermal ablation is being prompted to manage precancer, high level of recurrences, particularly in WLWH, may require a profound revision of the recommendations
- It remains unclear based on our data whether regular LEEP is sufficient in reducing recurrences in WLWH
- New treatment approaches are needed to reach deeper in the canal without affecting the reproductive outcomes.

Thank you for your attention