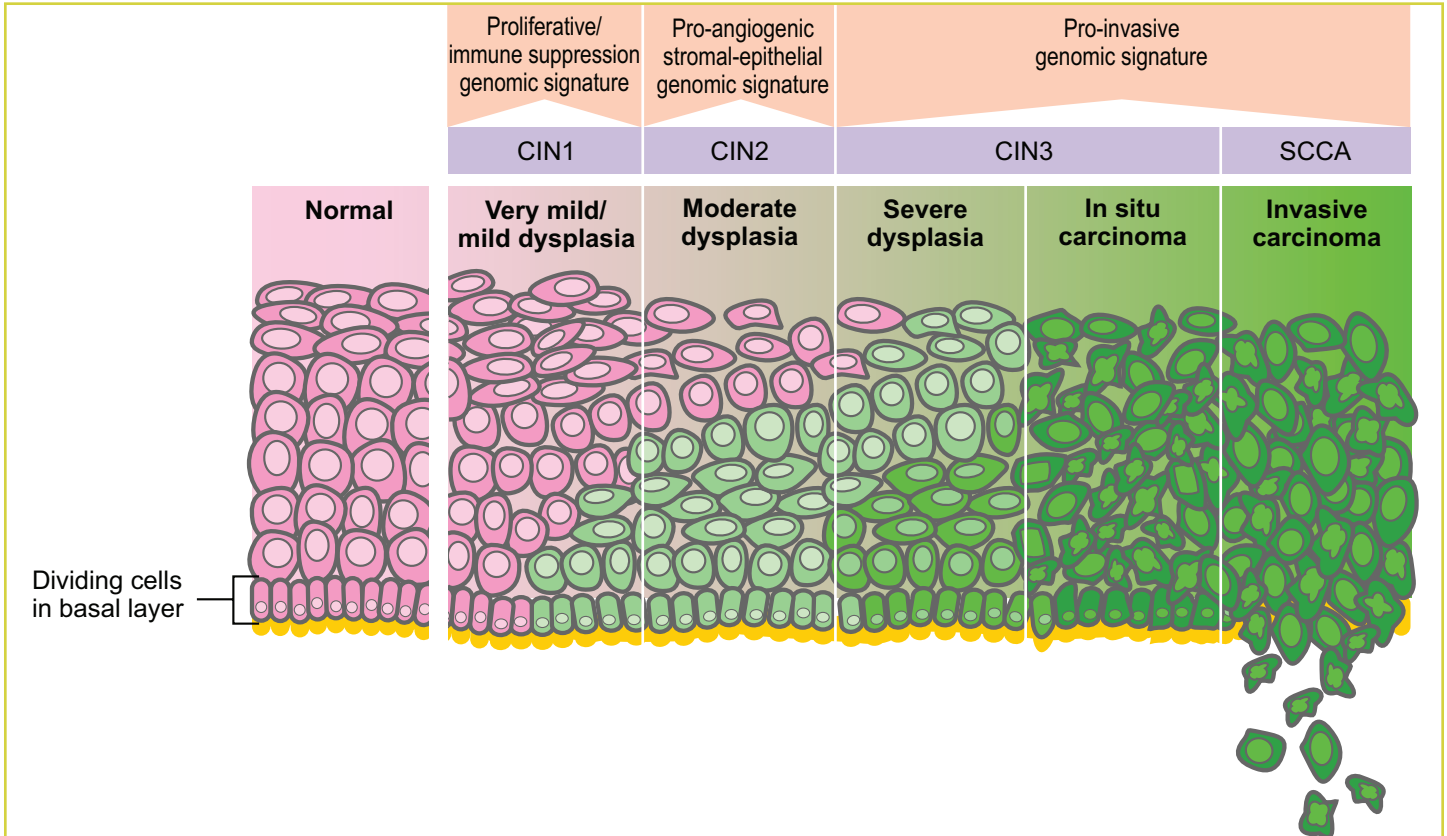


# Identifying Molecular Culprits of Cervical Cancer Progression

Human papillomavirus (HPV) DNA is found in 99.7% of invasive cervical carcinomas, providing strong evidence that the virus is a causative agent in the development of this disease. However, most women who become infected with HPV do not develop invasive cervical lesions, indicating that additional exogenous or genetic factors may determine whether HPV preclinical

lesions will progress to cancer. Identification of these factors would be facilitated by a deeper understanding of the cellular and molecular changes that accompany progression to malignancy. In addition, knowledge of which women are at greatest risk for disease progression would be a significant clinical advancement in the management of patients with pre-malignant cervical lesions.

The histological changes that occur as cervical tissue progresses from normal to cancerous have been well characterized. Preinvasive lesions are called cervical intraepithelial neoplasias (CINs). There are three CIN grades (CIN1–3), with CIN3 being the most advanced. Once the HPV-containing epithelial cells invade the basement membrane, the lesions become malignant and are

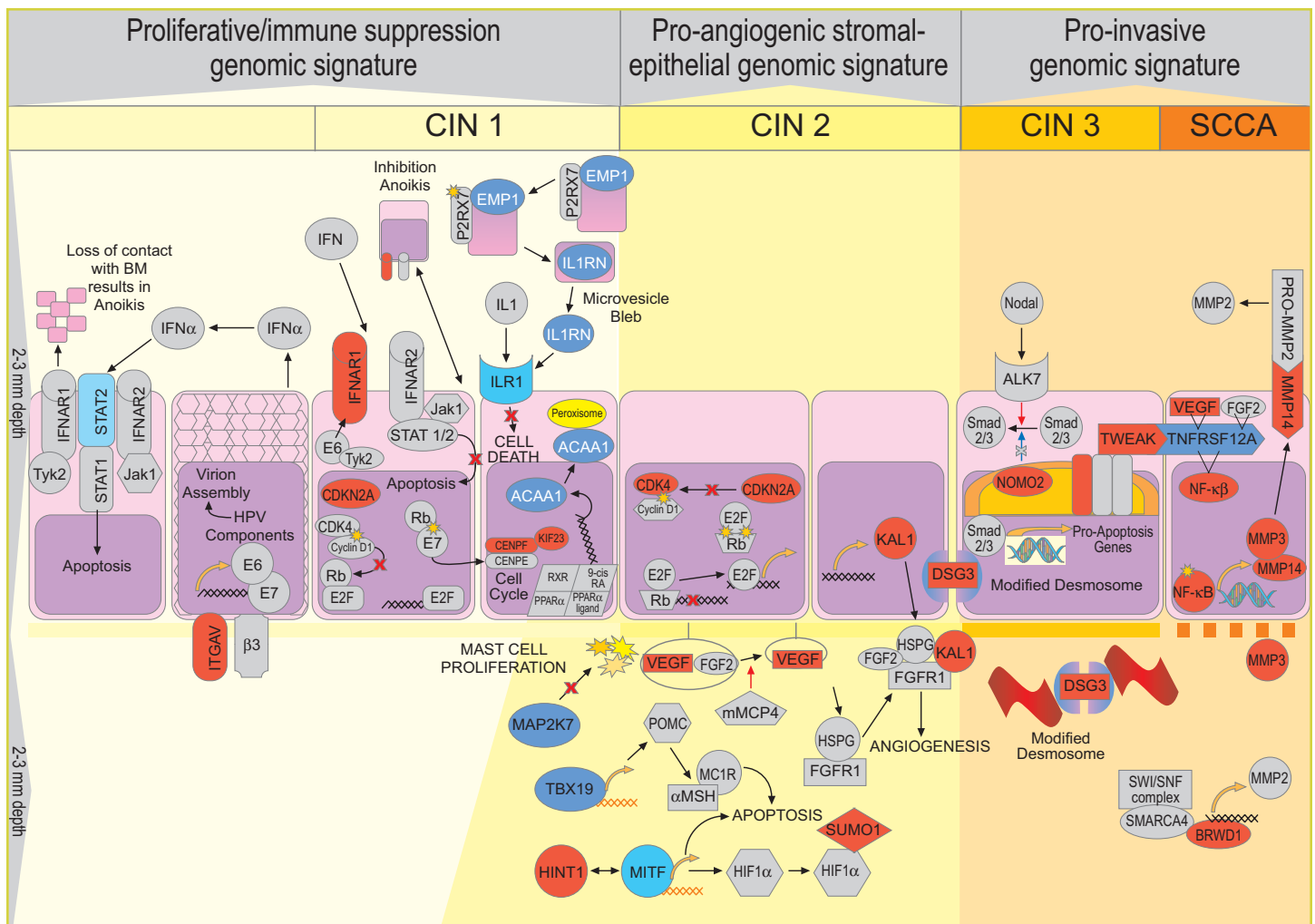


Dr. Gius and his colleagues used laser capture microdissection to analyze the gene changes that occur in individual cervical cells and the adjacent stromal cells. The researchers analyzed several precancerous stages as well as squamous cell carcinoma (SCCA) from patients' biopsy samples. See gene changes in next figure.

pathologically classified as squamous cell carcinoma. To begin to uncover molecular and genetic changes associated with these histological transitions, David Gius, M.D., Ph.D., Head of the Molecular Radiation Oncology Section in CCR's Radiation Oncology Branch, and other CCR researchers teamed up with scientists from Washington University School of Medicine. The results of their efforts were recently published in *Cancer Research*.

Dr. Gius and colleagues collected normal cervical tissue from healthy women as well as normal and abnormal cervical tissue from women with different grades of cervical disease, including CIN1, CIN2, CIN3, and cervical squamous cell carcinoma. They then used a technique called laser capture microdissection to separately collect epithelial and stromal cells from these tissues; this approach allowed them to pinpoint changes occurring within different tissue microenvironments.

RNA from the microdissected samples was used to perform gene expression profiling. Gene expression changes due to individual variation were filtered out using statistical tools, and additional samples were used to validate initial results. The subsequent gene expression patterns—or genomic signatures—generated for both epithelial and stromal cells along the continuum of cervical cancer progression were used to create the first genomically based model of cervical carcinogenesis.



As a cervical cancer cell moves toward a cancerous phenotype, gene activities change. In transition toward the CIN 1 stage, gene activities involved in increasing cell proliferation and suppressing an immune response are detectable. Later, as the cervical cell moves from a CIN 1 to a CIN 2 stage, gene activities involved in corrupting the normal stromal-epithelial environment and encouraging angiogenesis are evident. Finally, gene activities involved in invasion are present in the CIN 3 stage.

The model consists of three distinct functional genomic signatures. The first signature was associated with the transition from early viral infection to CIN1. This histological change coincided with altered expression of genes associated with cellular proliferation and suppression of the immune system. The environment created by this genomic signature may allow HPV to replicate without being detected and destroyed by the immune system. Tissue that had progressed to CIN2 demonstrated a genomic signature favoring growth of new blood vessels. Interestingly, these changes occur in both epithelial and stromal cells, suggesting that communication between different cell types may be important for blood vessel expansion during CIN2.

CIN3 and squamous cell carcinoma cervical tissue exhibited a pro-invasive genomic signature, including changes in cell adhesion proteins and enzymes involved in extracellular matrix remodeling in both epithelial and stromal cells. Based on these results, the research team hypothesized that the predominant cellular stresses present at the CIN3 transition may be due to cellular overcrowding. In addition, the activation of pro-invasive genes may be critical to invasion through the basement membrane and the transition from a premalignant to malignant cervical cancer.

Expansion and modification of this genomically based model for cervical transformation through additional re-

search should help illuminate factors that facilitate progression from HPV infection to cancer. The genes and pathways identified may also contribute to the discovery of biomarkers useful for cervical cancer screening.

## Reference

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