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Queen's University

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The purpose of this Standard Operating Procedure (SOP) is to describe common techniques used for estrus monitoring in rats.

1. Introduction and Definitions: Microscopic evaluation of the types of cells present in vaginal smears has long been used to document the stages of the

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Large Nucleated Epithelial Cells: These cells are larger with moderate to abundant amounts of blue to sky blue cytoplasm and a lower nuclear to cytoplasmic ratio in comparison to small epithelial cells. They are roud to polygonal and may have irregular, jagged or angular borders. Large epithelial cells can have some degree of keratinization and possess nuclei that may be intact, degenerate, or pyknotic. Occasionally, large nucleated epithelial cells will contain finely stippled cytoplasmic granules.

<u>Anucleated Keratinized Epithelial Cells:</u> Anucleated epithelial cells are also known as sqaumes or "cornflakes". They are aged cells and are characterized by an abundant blue to sky blue cytoplasm with jagged or angular edges. They lack nuclei

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Stages of the Estrous Cycle

The stages of the estrous cycle are identified by the absence, presence, or proportion of the described four basic cell types as well as by the cell density and arrangement of the cells on the slide. Conventionally, the cycle is divided into the four stages of proestrus, estrus, metestrus, and diestrus, however some researchers condense or subdivide the stages.

One general approach to staging vaginal smears is to first assess for the presence of neutrophils. If neutrophils are a dominant feature, or consistently observed,

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<u>Metestrus</u>

Metestrus is a short stage of 6–8hr in rats. Metestrus is characterized by a combination of anucleated keratinized epithelial cells and neutrophils. In rats, the small and large nucleated cells of late estrus are present in low to moderate numbers throughout the stage.

The separation between where metestrus ends and diestrus begins is not always obvious, as they (i.e., the end of metestrus and start of diestrus) are very similar in appearance and are defined by the same cell types.

Early and mid metestrus, however, are easily identified if and when sample collection happens to occur at these times.

<u>Diestrus</u> Diestrus is the longest stage of the estrus cycle with

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Mouse Estrous Cycle Identification Tool and Images. Shannon L. Byers1, Michael V. Wiles2, Sadie L. Dunn1, Robert A. Taft11 R.(u)-5(b)-5(j)4(ec)-2(t:)-3()]TJETQ335.11 700.54 241.34 27.264 reW*