CHAPTER 2

Methods of Histology

Technological progress in various scientific disciplines affords histologists many ways to study cells and tissues. The information desired usually dictates the use of one approach over another or the use of combined approaches. The maximal information attainable with any technique requires familiarity with the technique, an understanding of information output, and an appreciation of its advantages and disadvantages. The following will acquaint you with diverse techniques available to examine living cells, dead cells, and cell components. Histologists use these methods to solve histological peoblems.

Cell culture permits direct observation of living cells. It is routine because of the use of antibiotics. Cell culture also allows the continual observation, manipulation, and testing of explanted cells without donor jeopardy (Figs. 2-1 and 2-2). It facilitates studying cell differentiation, cell transformations, cytogenetics, cellular metabolism, cell-to-cell interactions, host-parasite relationships, and other biological processes. Cell culture is indispensable in diagnostic virology (Figs. 2-3 and 2-4), vaccine development, and vaccine production.

Implantation of transparent viewing chambers and the exteriorization and transillumination of organs and rissues complement direct observation. Although exteriorization techniques limit observation time, insights about microcirculation and the response of tissues and organs to chemotherapeutical agents have been obtained. Transparent viewing chambers extend the observation period. Neovascularization, cellular differentiation and movement, and other vital processes are studied by these techniques. The anterior chamber of the eye is a naturally occurring viewing chamber used in this approach.

The selective uptake of vital and supravital stains has added to our understanding of cell function, cell structure, and extracellular matrix materials. These stains have low toxicity; they may be injected into a living organism (vital) or used on living cells removed from the organism (supravital).

Oxytetracycline, a broad-spectrum antibiotic, is also a vital stain. At therapeutic dosages, this drug selectively stains all surfaces where bone, cementum, and dentin form (Figs. 2-5 and 2-6). Insights

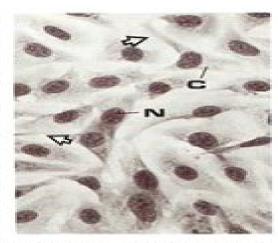


Figure 2-2. Medium-power micrograph of a morollayer of normal bovine field spiece cells. The muclei (N) are vesicular and the fourny and granular cytoplasm (C) has processes (open arrows) impairing a stellate or spindle appearance. (X100, Germa stain.) (Courters H. Storr.)

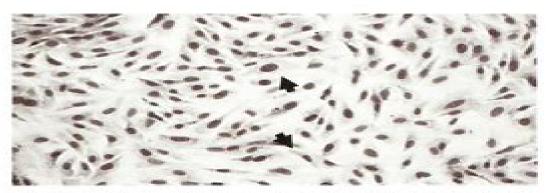


Figure 2-1. Low-power micrograph of a monolayer of normal bovine fetal spleen cells. Contact inhibition results in the formation of a monolayer of stellate- and spindle-shaped cells (acrows). (X-40, Glema stain.) (Courtey) H. Stort.)

concerning rates of bone formation, turnover, and maintenance have been obtained through carefully timed, spaced, serial administration of the drug.

Lithium carmine and trypan blue are vital dyes for studying phagocytosis. Janus green B and neutral red are supravital stains used to study mitochondria and besoomes.

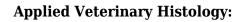
The separation and purification of cell fractions through differential centrifugation and density gradient centrifugation are aids for studying biochemical and metabolic phenomena.

The simplest method for the direct observation of living cells is to remove them from the organism, place them on a slide, and examine them with a phase-contrast microscope (Fig. 3-4) or dark-field mi-

Examining living cells and tissues is difficult microscopically, because they are

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- 1. Understanding the eBook Applied Veterinary Histology
 - The Rise of Digital Reading Applied Veterinary Histology
 - Advantages of eBooks Over Traditional Books
- 2. Identifying Applied Veterinary Histology
 - Exploring Different Genres
 - Considering Fiction vs. Non-Fiction
 - Determining Your Reading Goals
- 3. Choosing the Right eBook Platform
 - Popular eBook Platforms
 - Features to Look for in an Applied Veterinary Histology
 - User-Friendly Interface
- 4. Exploring eBook Recommendations from Applied Veterinary Histology
 - Personalized Recommendations
 - Applied Veterinary Histology User Reviews and Ratings
 - Applied Veterinary Histology and Bestseller Lists
- 5. Accessing Applied Veterinary Histology Free and Paid eBooks
 - Applied Veterinary Histology Public Domain eBooks
 - Applied Veterinary Histology eBook Subscription Services
 - Applied Veterinary Histology Budget-Friendly Options
- 6. Navigating Applied Veterinary Histology eBook Formats
 - o ePub, PDF, MOBI, and More
 - Applied Veterinary Histology Compatibility with Devices
 - Applied Veterinary Histology Enhanced eBook Features
- 7. Enhancing Your Reading Experience
 - Adjustable Fonts and Text Sizes of Applied Veterinary Histology
 - Highlighting and Note-Taking Applied Veterinary Histology
 - Interactive Elements Applied Veterinary Histology
- 8. Staying Engaged with Applied Veterinary Histology

- Joining Online Reading Communities
- Participating in Virtual Book Clubs
- Following Authors and Publishers Applied Veterinary Histology
- 9. Balancing eBooks and Physical Books Applied Veterinary Histology
 - Benefits of a Digital Library
 - Creating a Diverse Reading Collection Applied Veterinary Histology
- 10. Overcoming Reading Challenges
 - Dealing with Digital Eye Strain
 - Minimizing Distractions
 - Managing Screen Time
- 11. Cultivating a Reading Routine Applied Veterinary Histology
 - Setting Reading Goals Applied Veterinary Histology
 - Carving Out Dedicated Reading Time
- 12. Sourcing Reliable Information of Applied Veterinary Histology
 - Fact-Checking eBook Content of Applied Veterinary Histology
 - Distinguishing Credible Sources
- 13. Promoting Lifelong Learning
 - Utilizing eBooks for Skill Development
 - Exploring Educational eBooks
- 14. Embracing eBook Trends
 - Integration of Multimedia Elements
 - Interactive and Gamified eBooks

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