
Contents

| | |
|---|-----------|
| <i>Preface</i> | <i>v</i> |
| <i>Contributors</i> | <i>ix</i> |
| PART I SETTING AND PERFORMING ELISPOT ASSAY | |
| 1 Unique Strengths of ELISPOT for T Cell Diagnostics <i>Paul V. Lehmann and Wenji Zhang</i> | 3 |
| 2 The Impact of Harmonization on ELISPOT Assay Performance <i>Sylvia Janetzki and Cedrik M. Britten</i> | 25 |
| PART II ELISPOT FOR VETERINARY RESEARCH | |
| 3 Equine ELISPOT Assay to Study Secretion of IFN γ and IL-4 from Peripheral Blood Mononuclear Cells <i>Jodi Hagen, Chris Hartnett, Jeffrey P. Houchins, Steeve Giguère, and Alexander E. Kalyuzhny</i> | 39 |
| 4 Utilization of Feline ELISPOT for Mapping Vaccine Epitopes <i>Jeffrey R. Abbott, Ruiyu Pu, James K. Coleman, and Janet K. Yamamoto</i> | 47 |
| 5 Analyzing Cellular Immunity to AAV in a Canine Model Using ELISPOT Assay <i>Zejing Wang, Rainer Storb, Stephen J. Tapscott, and Stanley Riddell</i> | 65 |
| PART III ADVANCED ELISPOT TECHNIQUES | |
| 6 Dual- and Triple-Color Fluorospot <i>Niklas Ahlborg and Bernt Axelsson</i> | 77 |
| 7 ELISPOT Assay as a Tool to Study Oxidative Stress in Lymphocytes <i>Jodi Hagen, Jeffrey P. Houchins, and Alexander E. Kalyuzhny</i> | 87 |
| 8 ELISPOT Assay for Neuroscience Research: Studying TNF α Secretion from Microglial Cells <i>Jodi Hagen, Jeffrey P. Houchins, and Alexander E. Kalyuzhny</i> | 97 |
| 9 ELISPOT Assay as a Tool to Study the Effects of Stem Cells on Cytokine Secretion <i>Jun-Seop Shin and Chung-Gyu Park</i> | 105 |
| 10 Combining ELISPOT and ELISA to Measure Amounts of Cytokines Secreted by a Single Cell <i>Jodi Hagen, Jeffrey P. Houchins, and Alexander E. Kalyuzhny</i> | 115 |

PART IV IMAGE AND DATA ANALYSIS

- 11 How ELISPOT Morphology Reflects on the Productivity and Kinetics
of Cells' Secretory Activity 125
Alexey Y. Karulin and Paul V. Lehmann
- 12 Mathematical Algorithms for Automatic Search, Recognition,
and Detection of Spots in ELISPOT Assay 145
Sergey S. Zadorozhny and Nikolai N. Martynov
- 13 Objective, User-Independent ELISPOT Data Analysis Based
on Scientifically Validated Principles 155
Wenji Zhang and Paul V. Lehmann
- 14 Statistical Analysis of ELISPOT Assays 173
Marcus Dittrich and Paul V. Lehmann
- 15 Response Determination Criteria for ELISPOT: Toward a Standard
that Can Be Applied Across Laboratories 185
Zoe Moodie, Leah Price, Sylvia Janetzki, and Cedrik M. Britten

PART V ELISPOT ASSAY FOR VACCINE DEVELOPMENT AND DIAGNOSTICS

- 16 Detection of Vaccinia Virus-Specific IFN γ and IL-10 Secretion
from Human PBMCs and CD8⁺ T Cells by ELISPOT 199
*Benjamin J. Umlauf, Norman A. Pinsky, Inna G. Ovsyannikova,
and Gregory A. Poland*
- 17 ELISPOT Assays to Enumerate Bovine IFN- γ -Secreting Cells
for the Development of Novel Vaccines Against Bovine Tuberculosis 219
Martin Vordermeier and Adam O. Whelan
- 18 IL-7 Addition Increases Spot Size and Number as Measured
by T-SPOT.TB[®] 229
*Marsha L. Feske, Miguel Medina, Edward A. Graviss,
and Dorothy E. Lewis*
- 19 Overview of Membranes and Membrane Plates Used in Research
and Diagnostic ELISPOT Assays 243
Alan J. Weiss
- Index* 257

Handbook of ELISPOT

Methods and Protocols

Kalyuzhny, A.E. (Ed.)

2012, X, 261 p. 59 illus., 13 illus. in color., Hardcover

ISBN: 978-1-61779-324-0

A product of Humana Press