Contents at a Glance

Introduction ................................................................................................................. 1

Part I: Getting Started in Bioinformatics ................................................................. 7
Chapter 1: Finding Out What Bioinformatics Can Do for You ..................................... 9
Chapter 2: How Most People Use Bioinformatics .......................................................... 29

Part II: A Survival Guide to Bioinformatics .............................................................. 67
Chapter 3: Using Nucleotide Sequence Databases ..................................................... 69
Chapter 4: Using Protein and Specialized Sequence Databases .................................... 105
Chapter 5: Working with a Single DNA Sequence ..................................................... 129
Chapter 6: Working with a Single Protein Sequence .................................................. 159

Part III: Becoming a Pro in Sequence Analysis ......................................................... 197
Chapter 7: Similarity Searches on Sequence Databases ............................................. 199
Chapter 8: Comparing Two Sequences ..................................................................... 235
Chapter 9: Building a Multiple Sequence Alignment ............................................... 265
Chapter 10: Editing and Publishing Alignments ....................................................... 303

Part IV: Becoming a Specialist: Advanced Bioinformatics Techniques ......................... 327
Chapter 11: Working with Protein 3-D Structures ....................................................... 329
Chapter 12: Working with RNA ............................................................................... 353
Chapter 13: Building Phylogenetic Trees ................................................................... 371

Part V: The Part of Tens ............................................................................................ 403
Chapter 14: The Ten (Okay, Twelve) Commandments for Using Servers ................. 405
Chapter 15: Some Useful Bioinformatics Resources .................................................. 411

Index ............................................................................................................................ 417
Table of Contents

Introduction ........................................................................................................ 1
What This Book Does for You...........................................................................1
Foolish Assumptions .......................................................................................2
How This Book Is Organized .........................................................................2
  Part I: Getting Started in Bioinformatics .................................................3
  Part II: A Survival Guide to Bioinformatics ............................................3
  Part III: Becoming a Pro in Sequence Analysis .....................................3
  Part IV: Becoming a Specialist: Advanced Bioinformatics Techniques ..........3
  Part V: The Part of Tens .........................................................................4
Icons Used in This Book..............................................................................4
Where to Go from Here..................................................................................4

Part I: Getting Started in Bioinformatics .................................................7

Chapter 1: Finding Out What Bioinformatics Can Do for You ............... 9
  What Is Bioinformatics? ...........................................................................9
  Analyzing Protein Sequences .................................................................10
    A brief history of sequence analysis......................................................12
    Reading protein sequences from N to C .............................................13
    Working with protein 3-D structures ..................................................14
    Protein bioinformatics covered in this book .....................................16
  Analyzing DNA Sequences .....................................................................17
    Reading DNA sequences the right way................................................17
    The two sides of a DNA sequence ......................................................18
    Palindromes in DNA sequences ........................................................20
  Analyzing RNA Sequences .....................................................................21
    RNA structures: Playing with sticky strands ....................................22
    More on nucleic acid nomenclature ....................................................23
  DNA Coding Regions: Pretending to Work with Protein Sequences ......23
    Turning DNA into proteins: The genetic code ...................................24
    More with coding DNA sequences .....................................................25
    DNA/RNA bioinformatics covered in this book ...............................26
  Working with Entire Genomes ...............................................................26
    Genomics: Getting all the genes at once ............................................27
    Genome bioinformatics covered in this book ....................................28
Chapter 2: How Most People Use Bioinformatics ................. 29
  Becoming an Instant Expert with PubMed/Medline ...................... 29
  Finding out about a protein by its name .................................. 30
  Searching PubMed using author’s names ................................... 32
  Searching PubMed using fields ............................................. 35
  Searching PubMed using limits .............................................. 38
  A few more tips about PubMed ............................................. 41
  Retrieving Protein Sequences .............................................. 42
    ExPASy: A prime Internet site for protein information ............... 42
    More advanced ways to retrieve protein sequences .................. 45
    Retrieving a list of related protein sequences ....................... 48
  Retrieving DNA Sequences .................................................. 51
    Not all DNA is coding for protein ....................................... 51
    Going from protein sequences to DNA sequences .................... 52
    Retrieving the DNA sequence relevant to my protein ............... 53
  Using BLAST to Compare My Protein Sequence
    to Other Protein Sequences ............................................... 57
  Making a Multiple Protein Sequence Alignment with ClustalW ...... 62

Part II: A Survival Guide to Bioinformatics ....................... 67

Chapter 3: Using Nucleotide Sequence Databases ............... 69
  Reading into Genes and Genomes ......................................... 70
  Prokaryotes: Small bugs, simple genes .................................. 70
  Eukaryotes: Bigger bugs, complex genes ................................ 72
  Making Use (and Sense) of GenBank ...................................... 73
    Making sense of the GenBank entry of a prokaryotic gene ......... 73
    Making sense of the GenBank entry of an eukaryotic mRNA ...... 78
    Making sense of a GenBank eukaryotic genomic entry .......... 79
  Working with related GenBank entries .................................. 84
  Retrieving GenBank entries without accession numbers ............ 85
  Using a Gene-Centric Database ........................................... 86
  Working with Whole-Genome Databases ................................ 88
    Working with complete viral genomes .................................. 89
    Working with complete bacterial genomes ............................. 92
    More bacterial genomics at TIGR .................................... 94
    Microbes from the environment at DoE ................................. 96
  Exploring the Human Genome ............................................. 97
    Finding out about the Ensembl project ................................. 98

Chapter 4: Using Protein and Specialized Sequence Databases . . . 105
  From Translated ORFs to Mature Proteins ............................... 107
  ORFs: What you see is NOT what you get ................................ 107
  A personal final destination for each protein ......................... 109
  A combinatorial diversity of folds and functions ..................... 109
Chapter 5: Working with a Single DNA Sequence ......................... 129
  Catching Errors Before It’s Too Late................................................ 130
  Removing vector sequences .......................................................... 130
  Cases when you shouldn’t discard your sequence ...................... 133
  Computing/Verifying a Restriction Map ........................................ 134
  Designing PCR Primers ................................................................ 135
  Analyzing DNA Composition....................................................... 138
    Establishing the G+C content of your sequence ....................... 138
    Counting words in DNA sequences ............................................ 139
    Counting long words in DNA sequences .................................... 140
    Experimenting with other DNA composition analyses ............ 142
    Finding internal repeats in your sequence ............................... 142
    Identifying genome-specific repeats in your sequence ............ 145
  Finding Protein-Coding Regions .................................................. 145
    ORFing your DNA sequence ..................................................... 146
    Analyzing your DNA sequence with GeneMark ....................... 148
    Finding internal exons in vertebrate genomic sequences ........ 149
    Complete gene parsing for eukaryotic genomes ...................... 151
    Analyzing your sequence with GenomeScan ............................ 151
  Assembling Sequence Fragments............................................... 153
    Managing large sequencing projects with public software ....... 154
    Assembling your sequences with CAP3 .................................... 155
  Beyond This Chapter ................................................................... 157

Chapter 6: Working with a Single Protein Sequence ................... 159
  Doing Biochemistry on a Computer ............................................. 160
    Predicting the main physico-chemical properties of a protein ... 161
    Interpreting ProtParam results .................................................. 164
    Digesting a protein in a computer ............................................ 166
Doing Primary Structure Analysis ..............................................................166
Looking for transmembrane segments .............................................168
Looking for coiled-coil regions .........................................................174
Predicting Post-Translational Modifications in Your Protein ..............174
Looking for PROSITE patterns ..........................................................175
Interpreting ScanProsite results .......................................................177
Finding Known Domains in Your Protein ..................................................180
Choosing the right collection of domains .......................................182
Finding domains with InterProScan .................................................183
Interpreting InterProScan results .....................................................185
Finding domains with the CD server ...............................................187
Interpreting and understanding CD server results ........................189
Finding domains with Motif Scan .....................................................190
Discovering New Domains in Your Proteins .............................................194
More Protein Analysis for Free over the Internet ..........................................................194

Part III: Becoming a Pro in Sequence Analysis .................... 197

Chapter 7: Similarity Searches on Sequence Databases ............ 199
Understanding the Importance of Similarity ............................................200
The Most Popular Data-Mining Tool Ever: BLAST ...................................201
BLASTing protein sequences ............................................................201
Understanding your BLAST output ..................................................209
BLASTing DNA sequences .................................................................216
The BLAST way of doing things .....................................................218
Controlling BLAST: Choosing the Right Parameters ......................219
Controlling the sequence masking ...................................................220
Changing the BLAST alignment parameters ...................................223
Controlling the BLAST output ........................................................224
Making BLAST Iterative with PSI-BLAST ...................................................226
PSI-BLASTing protein sequences .....................................................226
Avoiding mistakes when running PSI-BLAST ..................................228
Discovering and using protein domains with BLAST and PSI-BLAST ..................230
Similarity Searches for Free over the Internet ........................................231

Chapter 8: Comparing Two Sequences ......................... 235
Making Sure You Have the Right Sequences and the Right Methods ...236
Choosing the right sequences ....................................................236
Choosing the right method ..........................................................237
Making a Dot Plot .................................................................................239
Choosing the right dot-plot flavor ...................................................240
Using Dotlet over the Internet ..........................................................241
Doing biological analysis with a dot plot .........................................249
<table>
<thead>
<tr>
<th>Chapter 9: Building a Multiple Sequence Alignment</th>
<th>265</th>
</tr>
</thead>
<tbody>
<tr>
<td>Finding Out if a Multiple Sequence Alignment Can Help You</td>
<td>266</td>
</tr>
<tr>
<td>Identifying situations where multiple alignments do not help</td>
<td>267</td>
</tr>
<tr>
<td>Helping your research with multiple sequence alignments</td>
<td>267</td>
</tr>
<tr>
<td>Choosing the Right Sequences</td>
<td>270</td>
</tr>
<tr>
<td>The kinds of sequences you’re looking for</td>
<td>271</td>
</tr>
<tr>
<td>Gathering your sequences with online BLAST servers</td>
<td>275</td>
</tr>
<tr>
<td>Choosing the Right Method of Multiple Sequence Alignment</td>
<td>281</td>
</tr>
<tr>
<td>Using ClustalW</td>
<td>282</td>
</tr>
<tr>
<td>Aligning sequences and structures with Tcoffee</td>
<td>287</td>
</tr>
<tr>
<td>Crunching large datasets with MUSCLE</td>
<td>291</td>
</tr>
<tr>
<td>Interpreting Your Multiple Sequence Alignment</td>
<td>291</td>
</tr>
<tr>
<td>Recognizing the good parts in a protein alignment</td>
<td>292</td>
</tr>
<tr>
<td>Taking your multiple alignment further</td>
<td>294</td>
</tr>
<tr>
<td>Comparing Sequences That You Can’t Align</td>
<td>297</td>
</tr>
<tr>
<td>Making multiple local alignments with the Gibbs sampler</td>
<td>298</td>
</tr>
<tr>
<td>Searching conserved patterns</td>
<td>299</td>
</tr>
<tr>
<td>Internet Resources for Doing Multiple Sequence Comparisons</td>
<td>299</td>
</tr>
<tr>
<td>Making multiple alignments with ClustalW around the clock</td>
<td>300</td>
</tr>
<tr>
<td>Finding your favorite alignment method</td>
<td>300</td>
</tr>
<tr>
<td>Searching for motifs or patterns</td>
<td>301</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Chapter 10: Editing and Publishing Alignments</th>
<th>303</th>
</tr>
</thead>
<tbody>
<tr>
<td>Getting Your Multiple Alignment in the Right Format</td>
<td>305</td>
</tr>
<tr>
<td>Recognizing the main formats</td>
<td>307</td>
</tr>
<tr>
<td>Working with the right format</td>
<td>307</td>
</tr>
<tr>
<td>Converting formats</td>
<td>309</td>
</tr>
<tr>
<td>Watching out for lost data</td>
<td>312</td>
</tr>
<tr>
<td>Using Jalview to Edit Your Multiple Alignment Online</td>
<td>313</td>
</tr>
<tr>
<td>Starting Jalview</td>
<td>314</td>
</tr>
<tr>
<td>Editing a group of sequences</td>
<td>316</td>
</tr>
<tr>
<td>Useful features of Jalview</td>
<td>318</td>
</tr>
<tr>
<td>Saving your alignment in Jalview</td>
<td>318</td>
</tr>
<tr>
<td>Preparing Your Multiple Alignment for Publication</td>
<td>319</td>
</tr>
<tr>
<td>Using Boxshade</td>
<td>319</td>
</tr>
<tr>
<td>Logos</td>
<td>322</td>
</tr>
</tbody>
</table>
Part IV: Becoming a Specialist: Advanced Bioinformatics Techniques ........................................327

Chapter 11: Working with Protein 3-D Structures ................. 329
From Primary to Secondary Structures ..................................330
Predicting the secondary structure of a protein sequence ....330
Predicting additional structural features ................................334
From the Primary Structure to the 3-D Structure ......................336
Retrieving and displaying a 3-D structure from a PDB site ....337
Guessing the 3-D structure of your protein ..........................340
Looking at sequence features in 3-D ..................................343
Beyond This Chapter ................................................................350
Finding proteins with similar shapes .................................350
Finding other PDB viewers ................................................350
Classifying your PDB structure ..........................................351
Doing homology modeling .................................................351
Folding proteins in a computer ...........................................351
Threading sequences onto PDB structures ........................351
Looking at structures in movement ....................................352
Predicting interactions ....................................................352

Chapter 12: Working with RNA .........................................353
Predicting, Modeling and Drawing RNA Secondary Structures ....354
Using Mfold .............................................................................355
Interpreting mfold results .................................................359
Forcing interaction in mfold ..............................................361
Searching Databases and Genomes for RNA Sequences ..........362
Finding tRNAs in a genome ...............................................363
Using PatScan to look for RNA patterns ..........................363
Finding the “New” RNAs: miRNAs and siRNAs ...................367
Doing RNA Analysis for Free over the Internet .....................368
Studying evolution with ribosomal RNA ..........................369
Finding the small, non-coding RNA you need ....................369
Generic RNA resources ..................................................370
Chapter 13: Building Phylogenetic Trees ..............................371
Finding Out What Phylogenetic Trees Can Do for You..............372
Preparing Your Phylogenetic Data............................................373
Choosing the right sequences for the right tree.....................374
Preparing your multiple sequence alignment.........................380
Building the Kind of Tree You Need.....................................383
Computing your tree..............................................................383
Knowing what's what in your tree.........................................398
Displaying your phylogenetic tree........................................399
Doing Phylogeny for Free over the Internet.........................400
Finding online resources.....................................................400
Finding generic resources....................................................401
Collections of orthologous genes.........................................402

Part V: The Part of Tens.....................................................403

Chapter 14: The Ten (Okay, Twelve) Commandments for Using Servers..................................................405
Keep in Mind: Your Data Is Never Secure on the Web..............406
Remember the Server, the Database, and the Program Version You Used..................................................406
Write Down the Sequence-Identification Numbers..................407
Write Down the Program Parameters.....................................407
Save Your Internet Results the Right Way.............................407
Use E-Values........................................................................408
Make Sure You Can Trust Your Alignments............................408
Use Different Programs to Check Borderline Results...............409
Stay Away from Unpublished Methods!................................409
Databases Are Not Like Good Wine.......................................409
Just Because It Looks Free Doesn’t Mean It Is Free................410
Biting the Bullet at the Right Time........................................410

Chapter 15: Some Useful Bioinformatics Resources..............411
Ten Major Databases............................................................411
Ten Major Bioinformatics Software Programs.......................412
Ten Major Resource Locators...............................................414
Some Places to Find Out What’s Really Going On................415

Index...............................................................................417