

1 R programming (6900) Basics of NGS and Micro-array analysis

Course BIOL 6930,
Location Langdale Hall 405,
CRN is 94748,
Timings: MW 8-9.45 AM.
Credits 4.

Instructor

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Proposed Topics

1. Introduction to NGS, Microarrays, Databases (DAVID, KEEG, BIOCARTA etc.).
2. Introduction to R environment and Bioconductor packages.
3. Unix/Linux operating system basics, Installing R and packages. Getting familiar with R commands.
4. Introduction on operating GSU server and HPC cluster
5. Start-up R exercise for sample NGS data.
6. Follow-up on Start-up R exercise for sample NGS data.
7. Start-up R exercise for sample Microarray data.
8. Follow-up on Start-up R exercise for sample Microarray data.
9. State of art algorithms used in NGS and Microarray analysis.
10. Application of Bio-conductor analysis packages-NGS, Microarrays.
11. Gene expression analysis packages-NGS, Data visualization: Heatmaps, Pie-charts, Venn diagrams, Pathway analysis- GSEA etc.

What is Bioinformatics?

- Bioinformatics is an interdisciplinary field that develops methods and software tools for understanding biological data. As an interdisciplinary field of science, bioinformatics combines biology, computer science, mathematics and statistics to analyze and interpret biological data. Bioinformatics has been used for in silico analyses of biological queries using mathematical and statistical techniques.

What is Bioinformatics?



https://www.youtube.com/watch?v=v1cTNhiZ2_c

Structural Bioinformatics:

- Protein structure prediction is another important application of bioinformatics. The amino acid sequence of a protein, the so-called primary structure, can be easily determined from the sequence on the gene that codes for it.
- <http://asterix.cs.gsu.edu/~weber/>

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Irene Weber
Regent's Professor

Education
Ph.D. Molecular Biophysics,
Oxford University, England, 1978

Specializations
Molecular Genetics and Biochemistry Research Group
Neurobiology and Behavior Research Group
Molecular Genetics Cell Physiology

Biography
Crossing paths and Bioinformatics: Neurobiology & Molecular Genetics
The major research theme of my laboratory is to understand the molecular basis of disease and how to develop new treatment strategies. We study key regulatory proteins at the molecular level by applying the techniques of bioinformatics, biochemistry and crystallography to study the structure and function of protein-ligand complexes. Current research projects include:
1. Studies of drug resistance in HIV and comparative studies of other retroviruses, HIV and other retroviruses cause AIDS and cancer in humans and other animals. We are studying the crystal structures and enzymatic activities of drug resistant mutants of HIV proteases in order to understand the molecular basis for drug resistance. New protease inhibitors are studied as potential drug candidates to overcome resistance. Comparative studies on structure and substrate specificity in the human and non-human retroviral protease domain.

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Robert Harrison
Professor Computer Science

Education
B.S. Biophysics, Pennsylvania State University, 1979
Ph.D. Molecular Biophysics, Yale University, 1983

Specializations
Computational chemistry and bioinformatics, numerical analysis and algorithms design, randomized and parallel approaches to difficult problems in scientific computing

Biography

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Network and Systems Biology

- Network analysis seeks to understand the relationships within biological networks such as metabolic or protein–protein interaction networks.
- Systems biology involves the use of computer simulations of cellular subsystems (such as the networks of metabolites and enzymes that comprise metabolism, signal transduction pathways and gene regulatory networks) to both analyze and visualize the complex connections of these cellular processes.
- <http://alan.cs.gsu.edu/NGS/?q=cscazz>

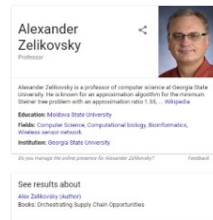
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Alexander Zelikovsky
Professor



Alexander Zelikovsky is a professor of computer science at Georgia State University. He is known for an approximation algorithm for the maximum k-clique problem with an approximation ratio of 1.55. [info@zelik](#)

Education: Middle Tennessee State University

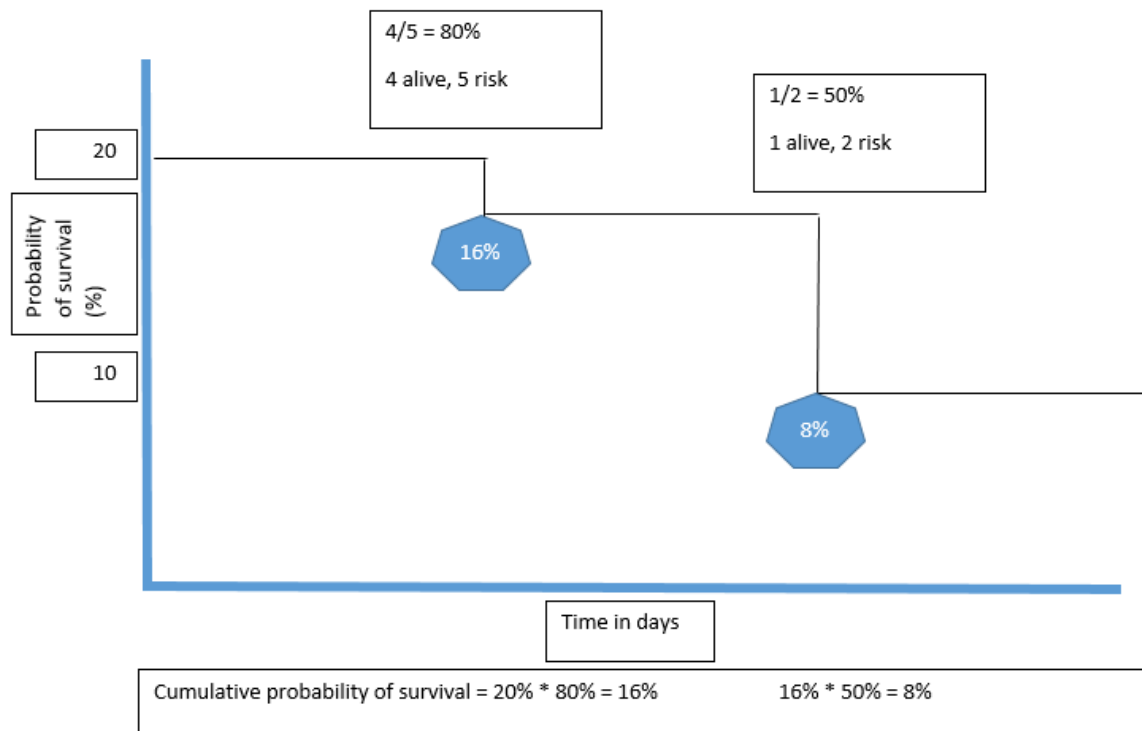
Fields: Computer Science, Computational Biology, Bioinformatics, Protein-protein network

Institution: Georgia State University

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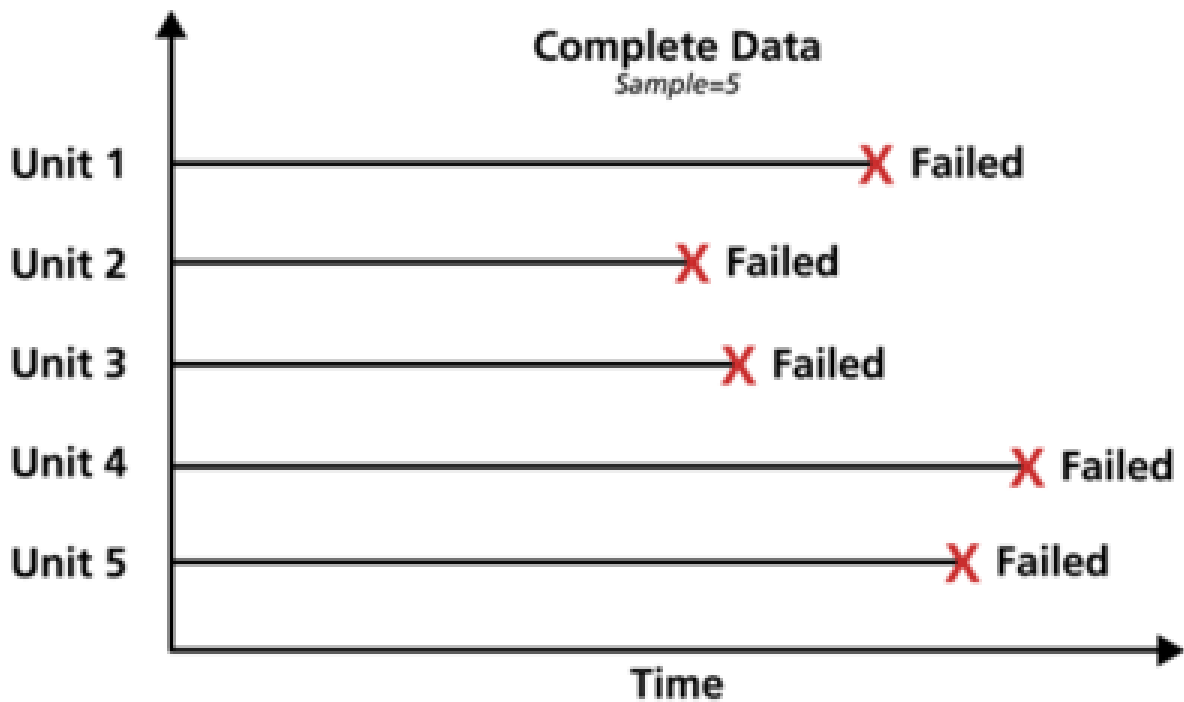
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1.1 Survival Plots: Kaplan Meier Analysis

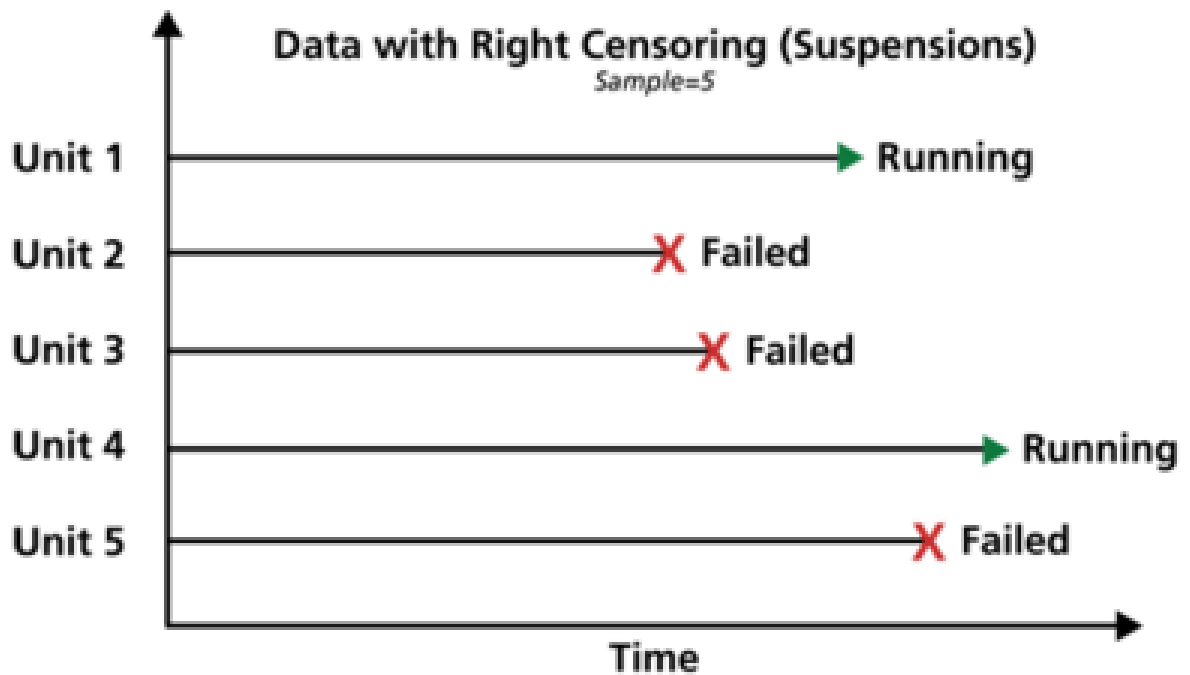


The first thing to do is to use function `Surv()` to build the standard survival object. The variable `t1` records the time to death or the censored time. A plus sign after the time in the print out indicates censoring. The formula instructs the `survfit()` function to fit a model with intercept only.

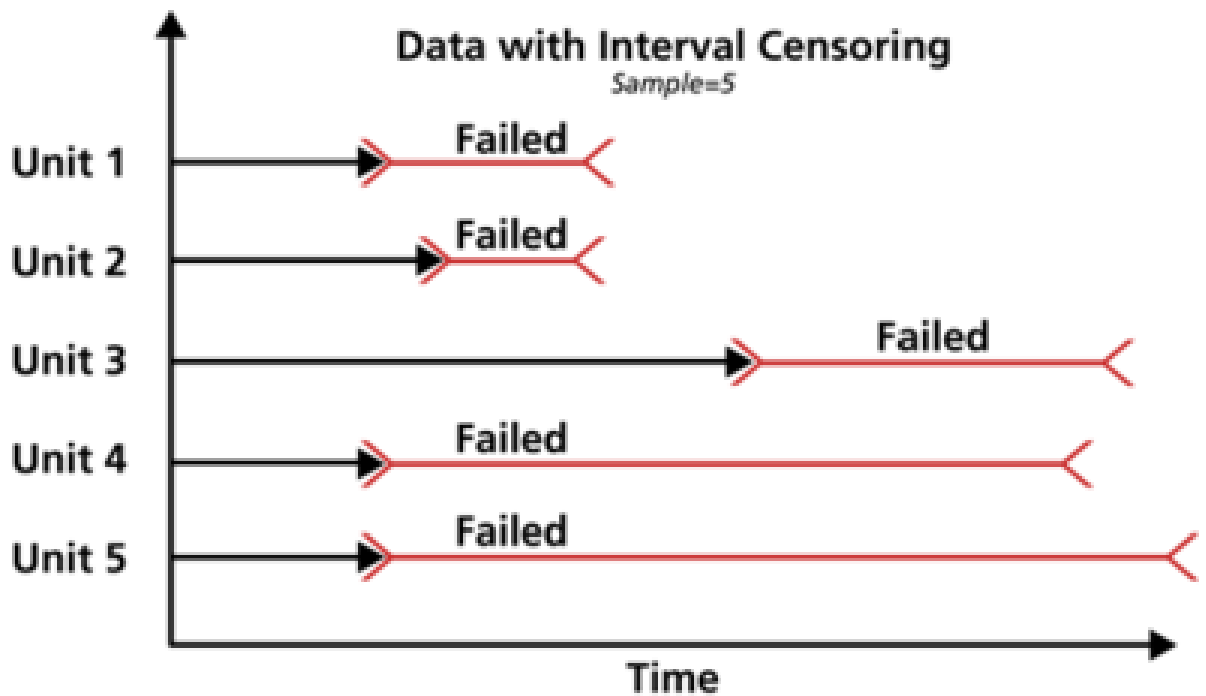
1.2 What is censoring? Understanding your data is extremely important



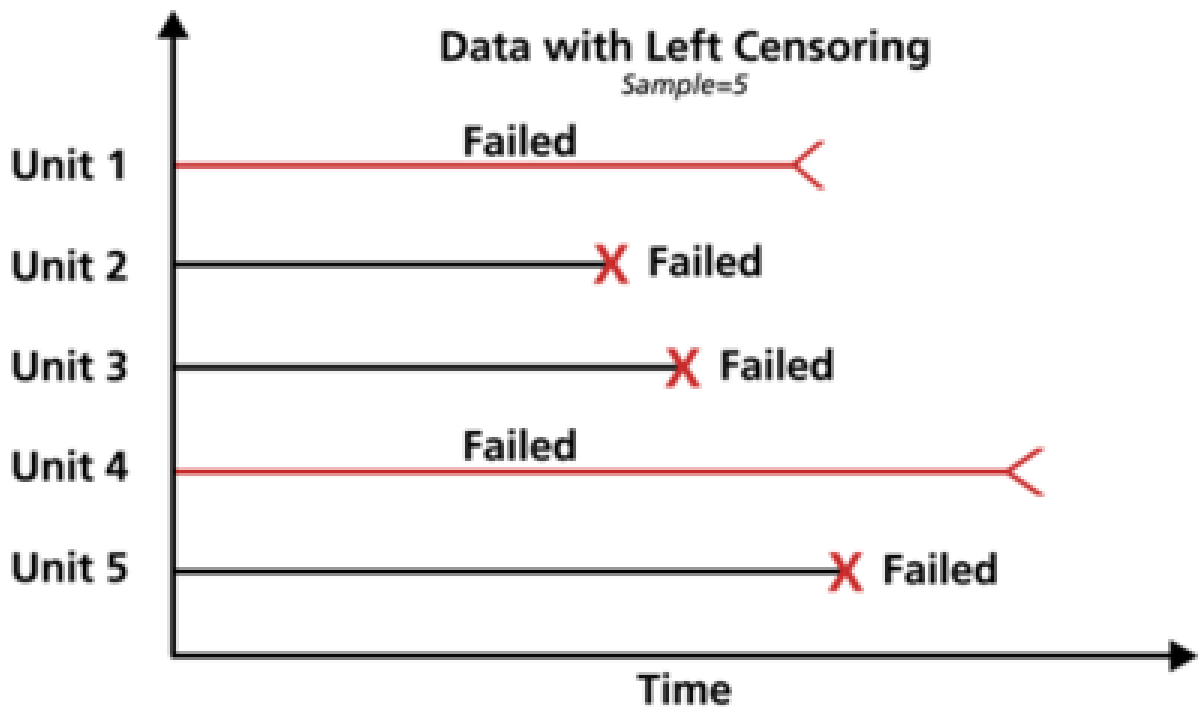
Complete data means that the value of each sample unit is observed or known. For example, if we had to compute the average test score for a sample of ten students, complete data would consist of the known score for each student. Likewise in the case of life data analysis, our data set (if complete) would be composed of the times-to-failure of all units in our sample. For example, if we tested five units and they all failed (and their times-to-failure were recorded), we would then have complete information as to the time of each failure in the sample.



The most common case of censoring is what is referred to as right censored data, or suspended data. In the case of life data, these data sets are composed of units that did not fail. For example, if we tested five units and only three had failed by the end of the test, we would have right censored data (or suspension data) for the two units that did not fail. The term right censored implies that the event of interest (i.e., the time-to-failure) is to the right of our data point. In other words, if the units were to keep on operating, the failure would occur at some time after our data point (or to the right on the time scale).

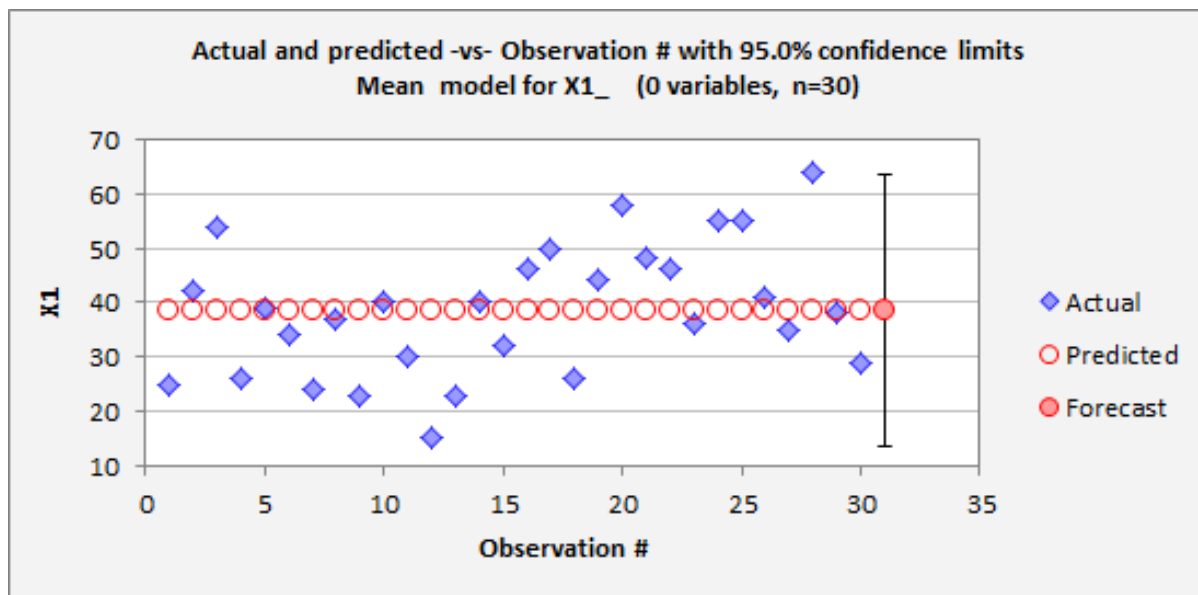


The second type of censoring is commonly called interval censored data. Interval censored data reflects uncertainty as to the exact times the units failed within an interval. This type of data frequently comes from tests or situations where the objects of interest are not constantly monitored. For example, if we are running a test on five units and inspecting them every 100 hours, we only know that a unit failed or did not fail between inspections. Specifically, if we inspect a certain unit at 100 hours and find it operating, and then perform another inspection at 200 hours to find that the unit is no longer operating, then the only information we have is that the unit failed at some point in the interval between 100 and 200 hours. This type of censored data is also called inspection data by some authors.



The third type of censoring is similar to the interval censoring and is called left censored data. In left censored data, a failure time is only known to be before a certain time. For instance, we may know that a certain unit failed sometime before 100 hours but not exactly when. In other words, it could have failed any time between 0 and 100 hours. This is identical to interval censored data in which the starting time for the interval is zero.

1.3 Using Intercepts?

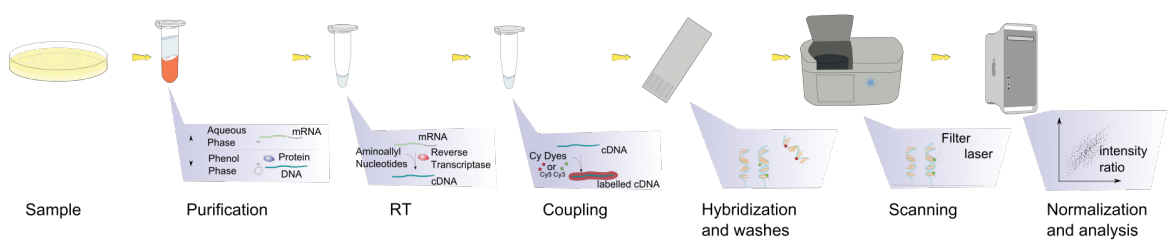
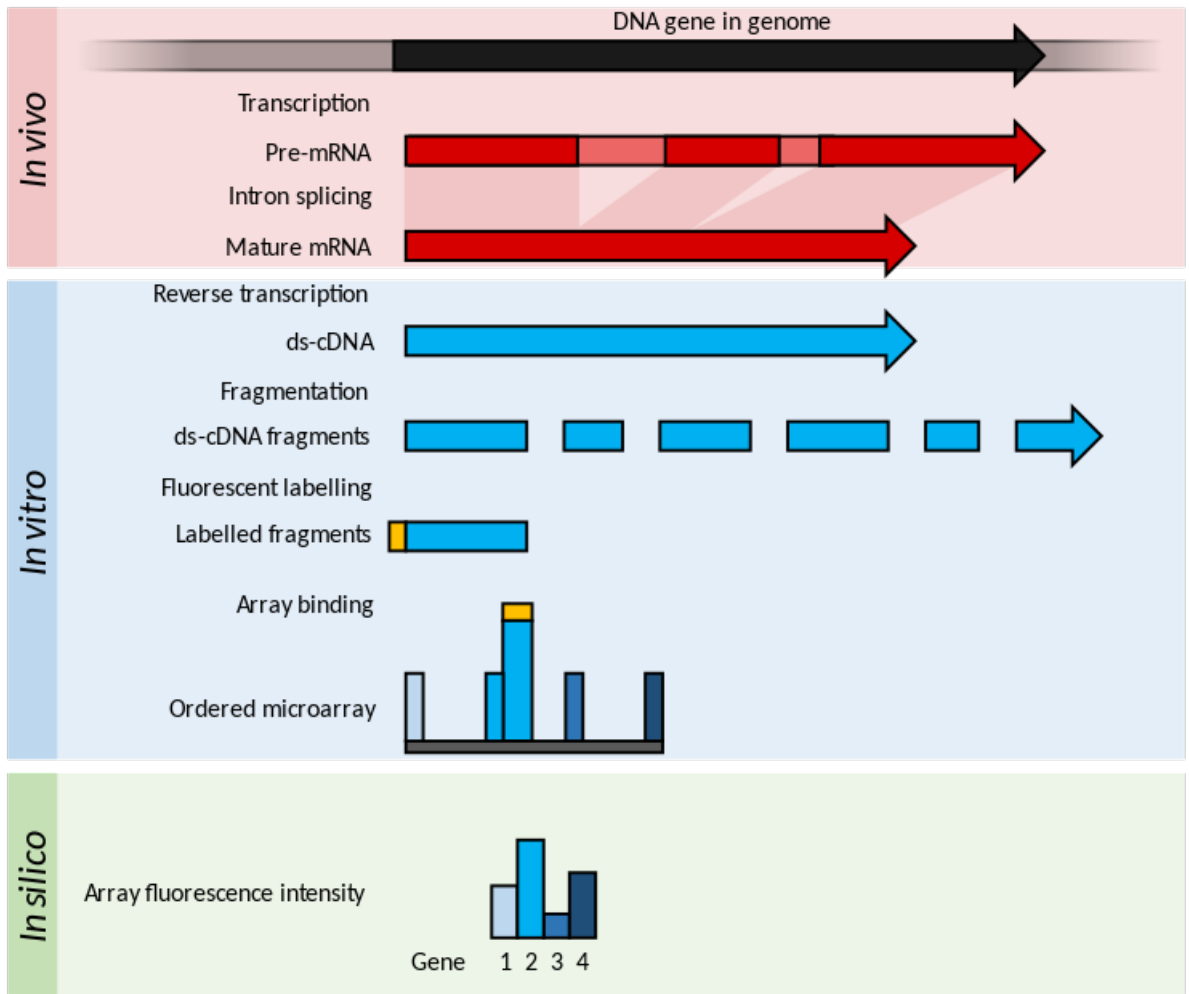


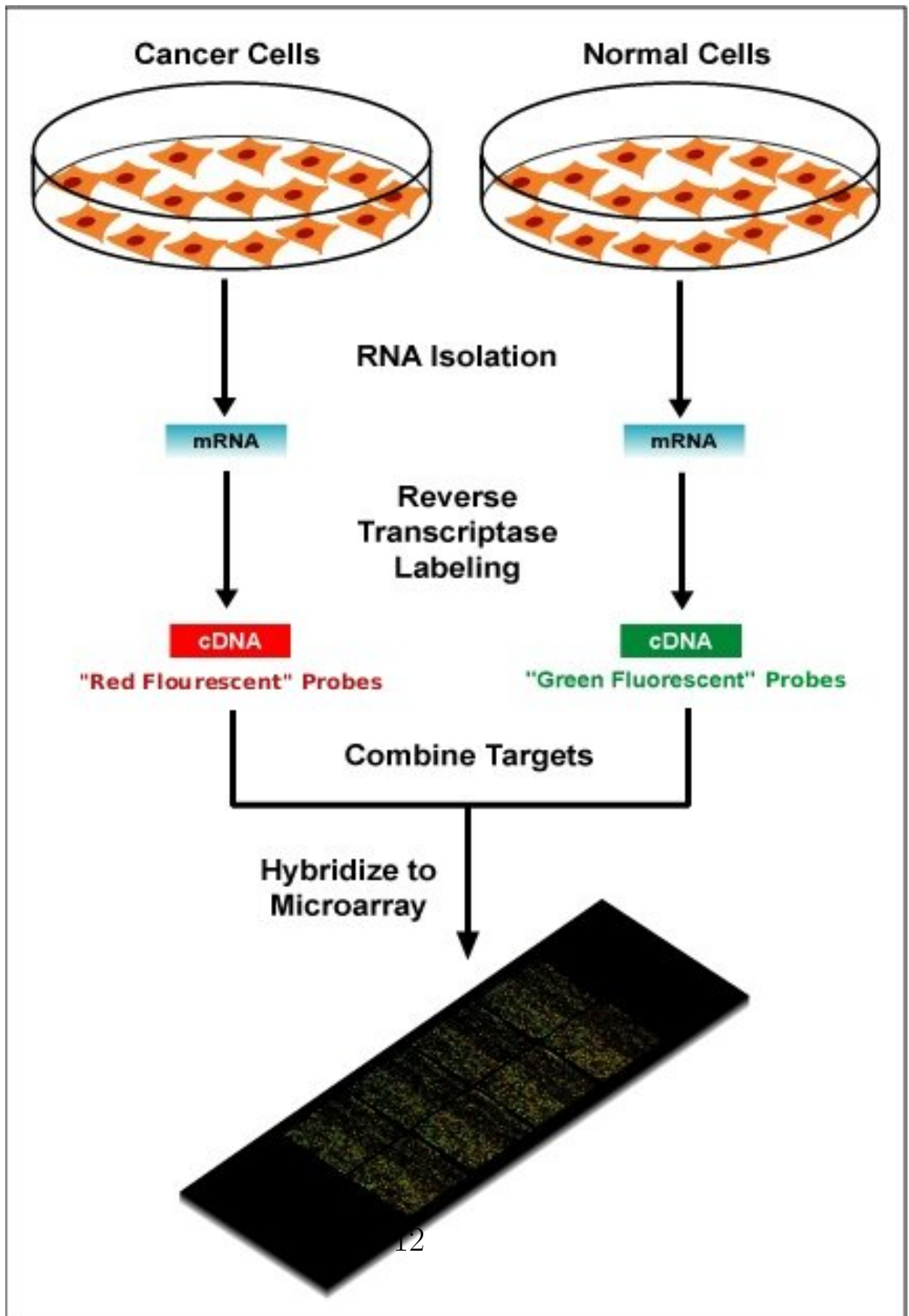
1.4 R exercise on KM plot

1.5 Microarray Technique:

A DNA microarray (biochip) is a collection of microscopic DNA spots attached to a solid surface. Scientists use DNA microarrays to measure the expression levels of large numbers of genes simultaneously or to genotype multiple regions of a genome.

General technique of performing Microarray





One-channel vs two-channel microarrays:

Two-color microarrays or two-channel microarrays are typically hybridized with cDNA prepared from two samples to be compared (e.g. diseased tissue versus healthy tissue) and that are labeled with two different fluorophores.

Fluorescent dyes commonly used for cDNA labeling include Cy3, which has a fluorescence emission wavelength of 570 nm (corresponding to the green part of the light spectrum), and Cy5 with a fluorescence emission wavelength of 670 nm (corresponding to the red part of the light spectrum). The two Cy-labeled cDNA samples are mixed and hybridized to a single microarray that is then scanned in a microarray scanner to visualize fluorescence of the two fluorophores after excitation with a laser beam of a defined wavelength

Data analysis: 1. Image analysis 2. Data processing: background subtraction (based on global or local background), determination of spot intensities and intensity ratios, visualisation of data (e.g. see MA plot), and log-transformation of ratios, global or local normalization of intensity ratios, and segmentation into different copy number regions using step detection algorithms. 3. Class discovery analysis: This analytic approach, sometimes called unsupervised classification or knowledge discovery, tries to identify whether microarrays (objects, patients, mice, etc.) or genes cluster together in groups. Identifying naturally existing groups of objects. 4. Class prediction analysis: This approach, called supervised classification, establishes the basis for developing a predictive model into which future unknown test objects can be input

in order to predict the most likely class membership of the test objects. 5. Hypothesis-driven statistical analysis: Identification of statistically significant changes in gene expression are commonly identified using the t-test, ANOVA, Bayesian method[29] MannWhitney test methods tailored to microarray data sets, which take into account multiple comparisons. 6. Network-based methods: Statistical methods that take the underlying structure of gene networks into account, representing either associative or causative interactions or dependencies among gene products.

1.6 R exercise on Microarray analysis