Chapter 13

Gene-Category Analysis

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Abstract

Gene-category analysis is one important knowledge integration approach in biomedical sciences that combines knowledge bases such as Gene Ontology with lists of genes or their products, which are often the result of *high-throughput* experiments, gained from either wet-lab or synthetic experiments. In this chapter, we will motivate this class of analyses and describe an often used variant that is based on Fisher's exact test. We show that this approach has some problems in the context of Gene Ontology of which users should be aware. We then describe some more recent algorithms that try to address some of the shortcomings of the standard approach.

Key words Enrichment, Overrepresentation, Knowledge integration, Fisher's exact text, Gene propagation problem

1 Introduction

The result of biological *high-throughput* methods is often a list consisting of several hundreds of biological entities, which are in case of gene expression profiling experiments identifiers of genes or their products. As a biological entity may have different context-specific functions, it is difficult for humans to interpret the outcome of an experiment on the basis of such a list. Computational approaches to access the biological knowledge about features of biological entities therefore play an important part in the successful realization of research based on high-throughput experiments. A practical way to address the question of *what is going on?* is to perform a gene-category analysis, i.e., to ask whether these responder genes share some biological features that distinguish them among the set of all genes tested in the experiment.

First of all, gene-category analysis involves a list of gene categories, in which genes with similar features are grouped together. The exact definition of the attribute *similar* depends on the provider of the categories. For instance, if Gene Ontology is the choice, then genes usually are grouped according to the terms, to which they are annotated. Another scheme is the KEGG database [1],

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in which genes are grouped according to the pathways in which they are involved. The second ingredient is a statistical method for identifying the really interesting categories.

In this chapter, we introduce some commonly used approaches for gene-category analysis. Throughout the remainder of this chapter, we refer to the set of items, which a study could possibly select, as the *population set*. We denote this set by the uppercase letter M while the size of the set, or its *cardinality*, is identified by its lowercase variant m. If, for example, a microarray experiment is conducted, the population set will comprise all genes whose expression can be measured with the microarray chip. The actual outcome of the study is referred to as the *study set*. It is denoted by N and has the cardinality n. In the microarray scenario the study set could consist of all genes that were detected to be differentially expressed.

2 Fisher's Exact Test

One approach for gene-category analysis is to cast the problem as a statistical test. For this purpose, the study set is assumed to be a random sample that is obtained by drawing n items without replacement from the population. The population is dichotomic as the items can be characterized according to whether they are annotated to term t or not. In particular, the set M_t with cardinality m_t constitutes all items that are annotated to t. Denote the random variable that describes the number of items of the study set that are annotated to t in this random sample as X_t. The hypergeometric distribution applies to X_t, and the probably of observing exactly k items annotated to t, i.e., $P(X_t = k)$ is specified by

$$X_t \sim h(k|m;\, mt;\, n) := P\left(X_t = k\right) = \frac{\binom{m_t}{k}\binom{m - m_t}{n - k}}{\binom{m}{n}}.$$

# of ways of choosing k items among all items annotated to t

# of ways of choosing the remaining $n - k$ items that are not annotated to t

# of ways of choosing n items among m

Furthermore, the set of items that are annotated to t and members of the study set are denoted by N_t with cardinality n_t. The objective is to assess whether the study set is enriched for term t, i.e., whether the observed n^t is higher than one would expect. This forms the alternative hypothesis H_1 of the statistical test. The null hypothesis H_0 in this case is that there is no positive association between the observed occurrence of the items in the study set and the annotations of the items to the term t. Thus, the proportion of

items annotated to term t is approximatively identical for the study set and the population set. In order to be able to reject H_0 in support of H_1 we conduct a one-tailed test, in which we ask for the probability of the event that we see n_t or more annotated items given that H_0 is true:

$$p_t^{\text{tft}} = P(X_t \geq n_t \mid H_0) = \sum_{k=n_t}^{\min(m_t,m)} \frac{\binom{m_t}{k}\binom{m-m_t}{n-k}}{\binom{m}{n}}. \quad (1)$$

If the probability obtained by this equation[1] is below a certain significance level α, e.g., $\alpha < 0.05$, we reject H_0 in favor of H_1. In that case, the tested term t is regarded as an interesting term that contributes to the characterization of the study set.

Example 2.1. *Suppose that we are given a population of $m = 18$ genes, of which $m_t = 4$ genes are annotated to a term t. The outcome of an experiment yields a study set of 5 differentially expressed genes. A total of $n_t = 3$ genes from the genes of the study set are annotated to t. Figure 1 illustrates the participating sets and how they are related to one another in that particular situation.*

In order to check whether term t can be used to characterize the experiment, we ask whether term t is overrepresented in the study set. The application of Eq. 1 yields a p-value for t

$$p_t^{\text{tft}} = P(X_t \geq 3 \mid H_0) = \frac{\binom{4}{3}\binom{14}{2}}{\binom{18}{5}} + \frac{\binom{4}{4}\binom{14}{1}}{\binom{18}{5}} = 0.044.$$

Thus, the null hypothesis is rejected and the term is said to be overrepresented among the differentially expressed genes and is thus likely to reflect an association between the term and the experiment.

3 Multiple Testing Problem

In hypothesis-generating studies it is a priori not clear, which terms should be tested. Therefore, the procedure is not only conducted using a single term but also applied to many, often all terms that Gene Ontology provides and to which at least one gene is annotated. The result of the entire analysis is then a list of terms that were found to be significant. This, however, implies that the number of false-positive terms is high.

[1] The superscript *tft* in p_t^{tft} stands for *term-for-term*. It allows to distinguish this *p*-value with other measures that are described later.

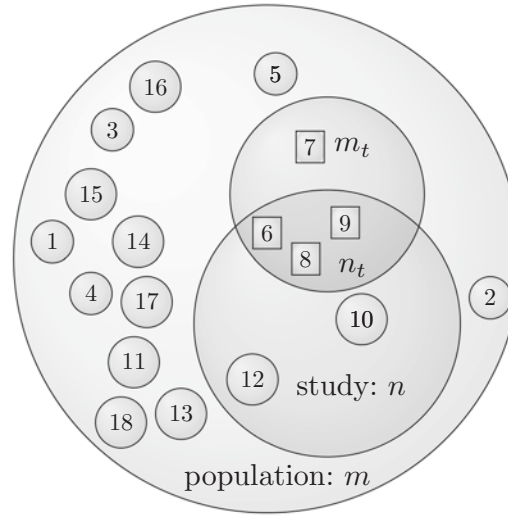


Fig. 1 Sets and their relations in the standard approach. In this example the population consists of $m=18$ genes and $n=5$ of them are part of the study set. Exactly $m_t=4$ genes of the population are annotated to term t. This term has $n_t=3$ genes in common with the study set. The null hypothesis of the standard approach (*term-for-term*) is that there is no association between the number of genes that are in the study set and the number of genes that are annotated to the term t, i.e., the study set is a random sample of the population set. We therefore would expect that it contains the same proportion of annotated terms as the population set does. The probability under the null hypothesis of the event to see at least nt genes can be assessed via Eq. 1.

To see this, suppose that there are T tests to be performed. We assume that the null hypothesis is true for all of those tests. Before its actual determination, any p-value can be considered as a random variable as well, for which $P(p \leq \alpha \mid H_0) \leq \alpha$ holds [2]. This implies that it can be expected that $\alpha \times T$ tests lead to the rejection of a null hypothesis although it is true.

Example 3.1. *If there are 10,000 null hypotheses that are true and all of them are tested, then we expect that we reject the null hypotheses for about 500 tests. Obviously, describing the result of experiment with 500 random terms is not useful.*

Therefore, the result of a term enrichment analysis shall be further subjected to a multiple test correction. The most simple is the Bonferroni correction [3]. Here, each p-value is simply multiplied by the number of tests saturated at a value of 1.0. Bonferroni controls the so-called family-wise error rate, which is the probability of making one or more false discoveries. It is a very conservative approach because it handles all p-values as independent. But as we see later, this is not a typical case of gene-category analysis, so this approach often goes along with a reduced statistical power.

In contrast, the Westfall–Young [4] procedure also takes dependencies into account. This correction, however, is computationally more costly as it is based on resampling schemes. In particular in the gene category setting, this scheme involves randomly sampling study sets of the same size as the original study set from the population. Each set is subjected to the test procedure yielding a set of p-values for each term, also referred to as the *null distribution* of that term. By relating the original p-value to the null distribution, an adjusted p-value is derived. There are other types of multiple test corrections that do not aim to control the family-wise error rate. For instance, the Benjamini–Hochberg [5] approach controls the expected false discovery rate (FDR), which is the proportion of false discoveries among all rejected null hypotheses. This has a positive effect on the statistical power at the expense of having less strict control over false discoveries. Controlling the FDR is considered by the American Physiological Society as "the best practical solution to the problem of multiple comparisons" [6].

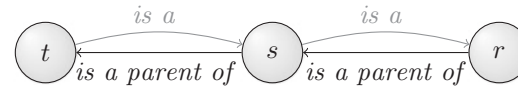
Note that less conservative corrections usually yield a higher amount of significant terms, which may be not desirable after all. In the following section, we further explore the structural origin of the correlations of the p-values in the setting of enrichment tests for ontology terms.

4 Gene Propagation

While the application of multiple testing correction aims to reduce the number of false-positives in a rather universal manner, one can also try to tackle the problem at a more basic level. The root of the problem is that if a term shares genes with a second term, and one of the terms is overrepresented, then it is not too surprising that the other term is also detected as overrepresented.

That the gene sharing of terms of an ontology is more a rule than an exception can be deduced from the principles of how ontologies are designed. Within an ontology, terms describe concepts of a domain that can be related to other terms by various types of relationships. The most prominent relationship thereby is the *is a* relationship, which effectively propagates the membership of the subject (source) of the relationship to the object (destination). That means, if a term T_1 is related to a term T_2 by the *is a* relationship, and a gene is annotated to T_1, then it is implicitly annotated also to term T_2 (*see* Chap. 1 [7]). In the context of GO overrepresentation analysis, we refer to this as the *gene propagation problem*.[2]

[2] Note that in addition to this gene sharing that is due to the graph structure of the ontology, also unrelated terms can be annotated to similar sets of genes, for instance, if the same gene plays a role in distinct biological processes.



$$\begin{array}{lll} m_t = 4 & m_s = 6 & m = 18 \\ n_t = 3 & n_s = 4 & n = 5 \\ p_t = 0.044 & p_s = 0.022 & \end{array}$$

Fig. 2 Extended example with three terms. This depicts the situation of Example 2.1 with two more terms. Term *t is a s* and therefore *s is a parent of t*. Term *r* is the root of the ontology. It is the only parent of *s*. As indicated in the last row, the procedure based on Fisher's exact test determines a *p*-value below 0.05 for both terms. Thus, both terms will be considered as a meaningful summary of the underlying experiment.

Example 4.1 (Continuation of Example 2.1). *There is another term s, which is the only parent of t. For s we know that* $m_s = 6$ *and* $n_s = 4$. *Figure 2 shows this structure graphically. There, it is also indicated that the p-values of terms t and s are 0.044 and 0.022, respectively, which means that both terms are considered as significant for* $\alpha < 0.05$ *if no multiple test correction is performed. Obviously, both terms share the majority of items that are also part of the study set. One can argue that the fact that term t is identified as overrepresented is a consequence of the fact that s is overrepresented.*

A simple synthetic experiment, in which a term will be artificially overrepresented, demonstrates the extent of the problem. Let's select the term *localization* for this purpose. We create a study set that consists of all genes that are annotated to that term with probability 0.8. This corresponds to false-negative rate $\beta = 0.2$. Furthermore, to introduce some background noise, each gene that is not annotated to the term is added to that study set with a false-positive rate of $\alpha = 0.1$. In this example, the procedure yields a set of 1542 genes. For each considered term, this set is subjected to Fisher's exact test resulting in a list of 4549 *p*-values[3]. Finally, the *p*-values are adjusted using the Bonferroni correction.

The analysis correctly identifies the term *localization* as significantly enriched. In addition to that, it identifies 275 other terms as significantly enriched. In particular, 6 of the 6 children, to which at least one gene is annotated, are significant. Among the 681 possible descendants of *localization*, we find 172 significant ones. These figures suggest that descendants come up only because their annotations converge in the term *localization*. Although, in the statistical sense, this is a correct result, it is not desirable to use that huge amount of terms to characterize the study set, especially as it is sufficient to use the term *localization* for this purpose, and what is

[3] This corresponds to the number of terms from the *biological process* subontology that are annotated by at least one gene.

more, the result suggests a specificity that we did not put in there. It makes sense to consider each of the additional 275 significant terms as a false-positive and in the next sections we will briefly describe methods that attempt to reduce that number.

5 Parent–Child Approach

The *parent–child* approach [8] is still based on Fisher's exact test, but the probability of t being overrepresented is conditioned on properties of the parental terms. In the following, let $\mathrm{pa}(t)$ be the set of parents of term t, which are, for instance, those terms, to which t is connected by a *is a* relation. In order to introduce the principal ideas of the *parent–child* approaches, we initially assume that there is only a single parent of t, i.e., $\mathrm{pa}(t) = \{s\}$.

Instead of drawing the items from the population M, items will be drawn just from the set of items that are annotated to the parent of t, which is written as $M_{\mathrm{pa}(t)}$ and whose size is $m_{\mathrm{pa}(t)}$. This consideration yields the following equation:

$$P(X_t = k \mid \mathrm{pa}(t)) = \frac{\binom{m_t}{k}\binom{m_{\mathrm{pa}(t)} - m_t}{n_{\mathrm{pa}(t)} - k}}{\binom{m_{\mathrm{pa(t)}}}{n_{\mathrm{pa}(t)}}}. \tag{2}$$

The right part of Fig. 3 shows the setting of the *parent–child* approaches. Effectively, in the *parent–child* approaches, we change the population that underlies Fisher's exact test to the items annotated to the parents. Obviously, this also alters the involved sets for the study set. As previously, we ask for the probability of seeing the observed number of items or a more extreme event:

$$p_t^{\mathrm{pc}} = P(X_t \geq n_t \mid H_0) = \sum_{k=n_t}^{\min(m_t, m_{\mathrm{pa}(t)})} \frac{\binom{m_t}{k}\binom{m_{\mathrm{pa}(t)} - m_t}{n_{\mathrm{pa}(t)} - k}}{\binom{m_{\mathrm{pa}(t)}}{n_{\mathrm{pa}(t)}}}. \tag{3}$$

Example 5.1 (Continuation of Example 4.1). *As shown in Fig. 2, the parent of term s is the root r of the ontology, which is always annotated to all genes of the population. Therefore, the p-value for s is the same for previous approach and for* parent–child *approach, i.e.,* $p_s^{\mathrm{pc}} = p_s = 0.22$ *However, for term t, Eq. 3 yields*

$$p_t^{\mathrm{pc}} = P(X_t \geq n_t \mid H_0) = \frac{\binom{4}{3}\binom{2}{1}}{\binom{6}{4}} + \frac{\binom{4}{4}\binom{2}{0}}{\binom{6}{4}} = 0.6.$$

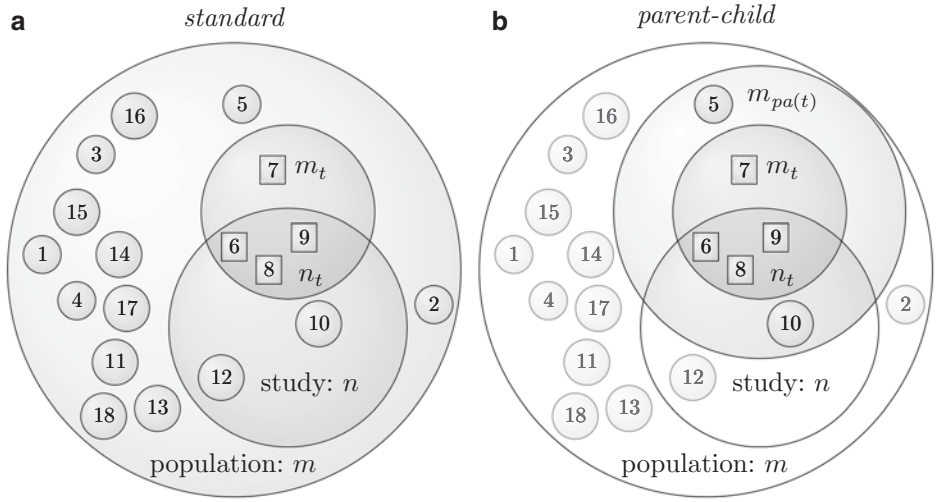


Fig. 3 Sets and their relations in the *parent–child* approaches. Part (**a**) depicts the model of the *term-for-term* approach as it was shown in Fig. 1. This is contrasted in part (**b**) with the model of the *parent–child* approaches. In this approach, we shift the focus to a smaller set of genes, for instance to the genes that are annotated to at least one of the parents of term t. In this particular situation it is the set whose size is $m_{\mathrm{pa}(t)} = 6$ with $\mathrm{pa}(t) = \{s\}$ following Example 4.1. Genes that are not part of this set do not contribute to the calculation. This has an effect on the involved proportions, and thus on the outcome of the test. Effectively, for each term, we alter the population of the association test. Eq. 2 quantifies the probability.

Thus, the null hypothesis for term t is not rejected, which is in contrast to the result of the previous approach. Given the initial observations that the study set is already skewed to the parent s of t makes the enrichment of term t less surprising, which the parent–child *approaches reflect by returning a higher p-value.*

If term t has more than one parent term, then it is not immediately apparent how to calculate $m_{\mathrm{pa}}(t)$ and the observation $n_{\mathrm{pa}}(t)$ in Eqs. 2 and 3. In Grossmann et al. [8] we examined two variants in detail, the union and the intersection of genes that are annotated to each of the parents.

6 Topology-Based Algorithms

Alexa et al. devised another method to address the *gene propagation problem*. The authors propose calculating a score for the term that depends on the relevance of the children of the term [9]. They argue that capturing the meaning in that way is biologically more interesting as the definitions of children are more specific. Following this argumentation, the authors formulated two concrete algorithms that try to provide a more suitable, i.e., less correlated, distribution of terms that get flagged as important. While the first approach which they called the *elim*-algorithm strictly favors significance of the most specific levels of the GO graph, their second

algorithm called *weight* relaxes this restriction such that terms that are most significant are favored.

As before, we understand the top of the graph as the root of the ontology, while the bottom of the graph consists of the most specific terms. The idea of the *elim* algorithm is to traverse the graph representation of the ontology in bottom-up fashion, which, for instance, can be accomplished by utilizing the backtrack phase of a depth-first search (DFS) [10].

The *elim* procedure awaits a term t as a variable parameter and returns a set of flagged genes. On its initial invocation, it begins with the root of the ontology. For the current term t, we apply Fisher's exact test in order to relate the genes of the study set to the genes of the population with respect to the genes that are annotated to term t. As in the *parent–child* approaches, not all genes of the study set contribute to the calculation. For *elim*, a set of previously determined genes is subtracted from the set of the study set before the calculation for p_t is carried out. This set is constructed by recursively applying the *elim* procedure for all children of t and taking the union of the result. If p_t is significant, we add all genes of t to the set of flagged genes. Finally, we return the set of flagged genes to the caller. Note that when the DFS reaches a leaf node of the ontology, Fisher's exact test is performed exactly as in the standard approach.

Obviously, the complexity of the algorithm is the same as the complexity of a depth-search algorithm if we assume that the number of genes that are annotated to a term is constant. Note in the original publication of the *elim*, the algorithm was based on an iteration over the levels of the GO DAG, which partitions the nodes according to their longest distance to the root. The algorithm as outlined here yields an equivalent result without the need to explicitly keep track of the DAG levels.

Example 6.1 (Continuation of Example 5.1). *The p-value of term t matches the p-value of term t of the standard approach, i.e.,* $p_t^{\text{elim}} = p_t^{\text{tft}} = 0.044$ *. As this is a significant result, at least, if correction for multiple testing is omitted, all four genes that are annotated to t are removed in the consideration of upper terms, i.e., we assume that those four genes are not annotated to them. This leaves two genes for the computation of term s, of which only one is member of the study set (Fig. 3b). With* $m_s = 2$, $n_s = 1$, *and the rest as before, Eq. 1 yields*

$$p_s^{\text{elim}} = P(X_s \geq 1 \mid H_0) = \frac{\binom{2}{1}\binom{16}{4}}{\binom{18}{5}} + \frac{\binom{2}{2}\binom{16}{3}}{\binom{18}{5}} = 0.49.$$

Hence, the elim *method doesn't report term s as important.*

An equivalent characterization of the *elim* method is the following: If a term t is identified as significant, all genes that are annotated to t are no longer considered in the computation of the relevance of the ancestors of t. As it was discussed in Example 2.1 at page 2.1 and as can also be seen in Fig. 2, the *term-for-term* approach assigns term s a lower p-value than it does for term t. One may conclude that it is more appropriate to take term s than to take term t in order to provide a compact description of the study set. However, in Example 13.6.1 we saw that the application of the *elim* method results in usage of term t to describe the outcome, which is contrary to that conclusion.

This concern is addressed by *weight* method. It compares significance scores of a family terms (a parent and its child) to identify the locally most significant terms and down-weight genes in less significant neighbors. This effectively decorrelates the p-values of the related terms such that their differences are enforced while the existence of the most significant terms is still maintained.

7 Model-Based Approaches

The previously described procedures that address gene propagation problem have in common that they successively test overrepresentation for each of the terms. They all use some form of the Fisher's exact test. In contrast to this, model-based gene set analysis (MGSA) models the gene response in a genome-wide experiment as the result of an activation of a number of terms [11].[4]

The approach is based on a model that can nicely be expressed using a Bayesian network with three layers of Boolean random variables. The *term layer* consists of m Boolean nodes corresponding to m terms of the ontology. A term can be *active* or *inactive*. A parameter p, usually much less than 0.5, represents the prior probability of a term being active. The hidden layer contains n Boolean nodes representing the n hidden state of the genes. The hidden state of a gene is a consequence of the states the terms to which the gene is annotated: The gene is *on* if and only if at least one term to which the gene is annotated is active, otherwise it is *off*. The third layer, the *observed layer*, contains Boolean nodes reflecting the experimentally observed state of all genes. For instance, in the setting of a microarray experiment, the *on* state would correspond to differential expression, and the *off* state would correspond to a lack of differential expression of a gene. The observed gene state depends on the corresponding hidden gene state in a one-to-one fashion with a false-positive (α) and false-negative rates (β) that is identical and independent for all genes. A simple instance of the model is depicted in Fig. 4.

[4] We use the word *term* here because we primarily work with GO, but the method can be applied to any other structured or unstructured vocabulary.

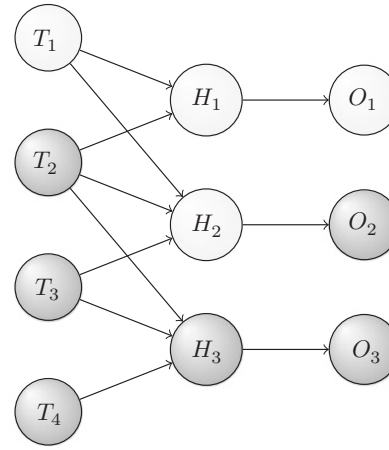


Fig. 4 The graphical representation of an MGSA network. An example structure for four terms and three genes with a possible realizations is displayed. Terms (T_i) that constitute the first layer can be either *active* (*light*) or *inactive* (*dark*). Terms that are *active* enable the hidden state (H_j) of all genes annotated to them, the other genes remaining *off*. The observed states (O_j) of the genes are noisy observations of their true hidden state. In this example, the observed states for gene 1 and 3 match the hidden state while for some unknown reasons the measurement of gene 2 doesn't correspond to the hidden state. It's a false-negative.

The model describes how the activity of terms leads to the observed stats of genes. This, however, is not the direction we are interested in. We are interested in the set of terms that explain the experimentally obtained data best, and the mathematical tool that can be applied to and such sets is probabilistic inference. The optimization problem that finds the term state configuration that explains the observed gene pattern best is NP-hard [12]. However, it is easily possible to find nearby solutions by sampling from the state space. This procedure additionally allows to determine the so-called marginal probability for each term, which is a measure how good the particular term will explain the observed genes with respect to all the other terms. The value ranges between 0 and 1 with 0 being the lowest possible support and 1 being the best possible support for a term. As all terms compete with one another, the inference takes dependencies both due to gene propagation and due to similarity of annotations into account. For example, if two unrelated terms are annotated to the same set of genes that matches the observation, the marginal probability for both terms will be 0.5. Consequently, it is advisable to run MGSA for each of the subontologies separately as they are designed to express orthogonal features.

8 Gene Set Enrichment Analysis

In addition to approaches that take a fixed subset of the population as input, procedures that take the measurements of the genes into account are also widely in use. This is attractive as it frees the investigator from the need to define a sometimes arbitrary cutoff that is used to construct the study set.

A first version of the so-called Gene Set Enrichment Analysis (GSEA) that received much attention of the scientific community was published by Mootha et al. [13]. In this approach, genes are ranked according to an interesting feature (e.g., the difference of the mean of their expression values for two experimental conditions). The null hypothesis is that the genes of the interesting set (e.g., genes annotated to a term) have no association with that list, in which case they would be randomly ordered. The alternative hypothesis is that the genes of the interesting set have an association. For instance, if the genes of the set are grouped together on the top of the list, we would tend to believe that there is such an association.

To capture the association via statistical means, the authors proposed a normalized Kolmogorov–Smirnov (KS) test statistic. Let $r_i \in M$ be the gene of the population M that has rank i in the gene list that is sorted according to the interesting gene feature. Using the previously established notation, i.e., that m is the total number of genes and N_t is the set of cardinality n_t that contains only genes that are annotated to t, the score is defined as:

$$ES(N_t) = \max_{i \in \{1,\ldots,m\}} \sum_{j=1}^{i} X_j \text{ with } X_j = \begin{cases} -\sqrt{\frac{n_t}{m-n_t}}, & \text{if } r_i \notin N_t \\ \sqrt{\frac{m-n_t}{n_t}}, & \text{otherwise} \end{cases}$$

Thus, the score is the maximum of a running sum that is increased if the gene is annotated to t and decreased if the gene is not annotated to t. In order to check if the obtained score is significant, the calculation is repeated for k randomly chosen sets $N_t^1, \ldots, N_{tk}$, which all are subsets of M with size n_t. The p-value for a term t is calculated as

$$p_t = \frac{\left|\{i \mid ES(N_t^i) \geq ES(N_t)\}\right|}{k}.$$

The GSEA method went a slight revision Subramanian et al. [14], where ad-hoc modifications are implemented that are supposed to countervail the well-known lack of sensitivity of the KS test [15, 16].

9 Software

Gene-category analysis is a very prominent use case of Gene Ontology. It shouldn't come as a surprise that users can choose among a variety of software implementations that will perform this sort of analysis. For instance, current version of the web site of Gene Ontology Consortium (geneontology.org) provides access to the method of the basic Fisher's exact test directly on the front page. There are also graphical tools that integrate into existing frameworks such as *BiNGO* [17], standalone graphical clients such as *Ontologizer*[5] [18] or packages for Bioconductor such as *topGo* [19], *mgsa* [20], or *gCMAP* [21], just to name a few of them.

10 Exercises

1. Repeat the random experiment outlined in the text that was used to show the influence of the gene propagation. When doing this in R/Bioconductor, it is advisable to use the *GO.db* and *org.Sc.sgd.db* packages that provide the structure and the annotations. The calculation involving the hypergeometric distribution can be expressed directly in R using dhyper and phyper. Now repeat this experiment with other approaches based on study sets that were outlined in this chapter and compare the results. For the topology-based algorithms the *topGo* package can be used and for the model-based approach the *mgsa* package is well suited.
2. Apply the approach now to an arbitrary example or on real world data. Compare the results.

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[5] http://ontologizer.de

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