**Venn diagrams and probability in clinical research**

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**Abstract**

Statistics is the science dealing with the collecting, summarizing and interpreting of associations in research data, and has a leading role in medical research. This article is an introductory publication in a series devoted to biomedical statistics. The aim of this article is to acquaint the readers with the basic concepts of Venn diagrams, probability and set theory, which are required to further understand descriptive and inferential statistics. First, we discuss the applications of Venn diagrams in current clinical research. Then we discuss the definitions of sample space, events, basic set operations (union and intersection) and their implementation in the classical approach to probability theory. All examples are introduced with Venn diagrams to illustrate the cases.

**Keywords:** statistics; set theory; probability; Venn diagram

**MeSH terms:**

CLINICAL STUDIES AS TOPIC  
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Применение диаграмм Венна и теории вероятностей в клинических исследованиях

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Аннотация
В настоящее время статистика играет ключевую роль при проведении медицинских исследований, объектом ее изучения являются накопление и обобщение данных, а также интерпретация взаимосвязей между экспериментальными данными. Данная статья открывает вводный цикл, посвященный проблемам биомедицинской статистики, и знакомит читателей с диаграммами Венна, а также с базовыми концепциями теории вероятностей и теории множеств, понимание которых необходимо для освоения принципов описательной статистики и статистики вывода.

В статье представлено применение диаграмм Венна в современных клинических исследованиях, а также определены пространство элементарных событий, базовые операции с множествами (объединение, пересечение) и их применение в контексте классической теории вероятностей. Все примеры проиллюстрированы диаграммами Венна.

Ключевые слова: статистика; теория множеств; вероятность; диаграмма Венна

Рубрики MeSH:
КЛИНИЧЕСКИЕ ИССЛЕДОВАНИЯ КАК ТЕМА
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Statistics is the science of the collecting, summarizing, presenting and interpreting of data; using them to estimate the magnitude of associations and test hypotheses [1]. Today statistics has a leading role in medical and biological research. Therefore, it is crucial for all doctors and researchers to understand and be able to apply at least the basic concepts of data analysis to their studies. However, most healthcare providers in Russia have almost no training in mathematics and a very limited understanding of statistics resulting in the misinterpretation and uncritical acceptance of published medical data, as well as an inability to organize, perform and analyze their own research properly. For this reason, we decided to publish this introductory course on biomedical statistics in a series of articles.

This course was originally designed for Master and PhD students at University College London and was later adapted for students and postgraduate trainees at Sechenov University. The main idea of the course is to take a practical approach, and this is why the current and further articles will focus on the most important concepts, techniques and statistical tests required for the critical understanding of state-of-art clinical research and the preparation of manuscripts for submission to high-ranking medical journals.

We encourage the readers to learn to present data adequately, apply the basic statistical tests properly and interpret their results without help from a professional statistician. Nevertheless, we have also included in this course some advanced statistical methods such as basic machine-learning algorithms in order to acquaint the reader with their possible applications. Another aim of the course is to achieve efficient cooperation between doctors and statisticians in terms of creating rational clinical research study designs, formulating realistic inquiries, choosing optimal approaches and interpreting the results of the research. All of the discussed methods can be universally applied in any statistical software, both commercial (SPSS, Stata, SAS, MatLab, etc.) and freeware (R).

The material is designed to be understandable by the readers who are not mathematical specialists. However, some mathematical concepts and expressions will be introduced throughout the course in a simplified form since the authors believe it is the most adequate and accurate way to present the underlying ideas of statistical techniques. It was decided to present the course both in English and Russian to familiarize readers with the terms frequently used in scientific publications.

The first article will discuss the concepts of the set theory and probability theory with some examples illustrated by Venn diagrams.

### VENN DIAGRAMS

Venn diagrams (also called Set diagrams) are widely used in clinical and population health research reports to represent the logical relationships between two or more sets of data. A Venn Diagram involves overlapping circles or other closed curves, which present how things are organized, including their common and different sets. Venn diagrams were developed in 1880 by John Venn, lecturer in Moral Science at Caius College, Cambridge University in a paper entitled, “On the diagrammatic and mechanical representation of propositions and reasonings” [2]. It is noteworthy that the use of diagrams in formal logic is easily traced historically, and the diagrams normally associated with Venn, probably originated much earlier [3]. More information on the history of Venn diagrams can be found in other publications [4].

As our knowledge of the complexity of physiological and molecular functions expands, the study of multi-omics and Big Data become more and more important. There is an increasing need for novel methods to study multiple datasets [5], for even such a classical method of visualization as the Venn diagram has undergone further development. Venn diagrams are currently widely used in clinical research, for example, to study a core microbiome [6], or in obstructive lung diseases [7], and even genetic studies often include them. For example, one of the most effective ways of examining the overlap of resulting gene lists is a Venn diagram [8]. The graphic way to represent interactions can be read easily because it shows all the logical relations and overlaps between the sets.

Numerous online Venn programs are currently available to work with up to four independent datasets; with examples being Panglos Venn diagram generator or Venny². These programs provide a convenient resource but only generate images without the relevant output information. BioVenn [9] is another available program allowing the generation of area-proportional Venn set analyses. Programs such as GeneVenn¹ and VennMaster⁴ have the additional feature of linking genes within each group to related information in the NCBI Entrez Nucleotide database or the Gene Ontology database. However, these programs are still limited to the analysis of two or three gene lists [8, 10].

For microarray datasets GeneSpring and SilicoCyte can be used, both of which are specifically designed for microarray data with several visualizations, including Venn diagrams. For more than three data sets, the intersections can start to become problematic in terms of visualization as the representation of all overlapping

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¹ [http://www.pangloss.com/seidel/Protocols/venn.cgi](http://www.pangloss.com/seidel/Protocols/venn.cgi)
² [https://bioinfogp.cnb.csic.es/tools/venny/](https://bioinfogp.cnb.csic.es/tools/venny/)
³ [http://genevenn.sourceforge.net/](http://genevenn.sourceforge.net/)
⁴ [https://sysbio.uni-ulm.de/?Software:VennMaster](https://sysbio.uni-ulm.de/?Software:VennMaster)
regions and symmetry can be lost. Venn himself performed visualization using additional ellipses that intersect with the primary circles [2]. A.W.F. Edwards also developed an elegant method of displaying the diagram composed of higher number of sets that feature some interesting symmetries [11]. VENNTURE⁵ [5] is a novel freely available Venn diagram-generating tool, and is a good example of an Edwards—Venn diagram, which can easily be performed with the program.

Using the VENNTURE program can allow us to generate the Edward—Venn diagram for an abstract example: Let us assume that there are 6 pharmacies in the city that can sell 26 types of drugs and let us designate these drugs with the letters of the English alphabet. However, each pharmacy can only accommodate 20 different types of drugs due to storage restrictions. If the drugs were randomly distributed between pharmacies, then one could use the Venn diagrams to analyze how the different drugs were distributed between the pharmacies. This is seen in the Fig. 1 for 2 (A), 3 (B), 5 (C) and 6 (D) datasets representing current list of letters-drugs in the corresponding pharmacies. From Fig. 1A we can see that 15 drugs are offered in both pharmacies because they are at the intersection of the datasets corresponding to the first (red) and second (blue) pharmacy. Five drugs can only be found in one of the two pharmacies. Suggested visualization method enables representation for up to 6 datasets. For example, Fig. 1B shows that drug “O” can only be found in the first pharmacy, whereas 10 drugs are being sold in all three pharmacies under analysis. One can also see that only 6 drugs can be found in 5 pharmacies (Fig. 1C) and 5 drugs are sold on all 6 pharmacies (Fig. 1D) because only the letters “DFGSZ” are at the intersection area of all 6 datasets.

**TUTORIAL: SET THEORY AND VENN DIAGRAMS**

Venn diagrams can be best understood if we review the concept of probability and the link between the theory of sets and Venn diagrams. Probability is

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⁵ https://www.nia.nih.gov/research/resource/vennture

**FIG. 1.** Edwards—Venn diagram representing overlaps of 2 (A), 3 (B), 5 (C) and 6 (D) datasets in a symmetric way.

**РИС. 1.** Диаграммы Эдвардса — Венна, симметрично изображающие пересечения 2 (A), 3 (B), 5 (C) и 6 (D) наборов данных.

Note. The symbols, which are common in the corresponding sets, are displayed at the intersections between these datasets.

Примечание. Символы, которые являются общими для соответствующих наборов данных, изображены в области их пересечения.
a way of expressing knowledge or belief that some event has occurred or will occur. Any process of observing the events in the real-world setting is referred to as “experiment”. The results of experiments are the outcomes. In general, probability is required to formalize the mechanisms that generate the data we get from the observations. Any model given in terms of probability (“ideal world” of the model) can be studied through the mathematical properties which can help understand the patterns and predict some events that are likely to happen in the future or under different settings (e.g., in a clinical trial). In contrast, statistics studies the frequency of past events that have happened in real world settings (e.g., historical data or a clinical trial) to assess the probability of observed events and create or test a suitable probability model.

To define probability mathematically we need to define set $S$ consisting of all possible outcomes of the experiment. An element “$s$” of $S$ is called a sample point. A sample space $S$ is called:

- discrete if it consists of a finite number of sample points;
- countable if the elements can be placed in one-to-one correspondence with positive integers (1, 2, 3, 4, ...);
- continuous if the sample points constitute a continuum;
- empty or null set if it contains no elements; in that case this set is denoted by $\emptyset$.

Several operations can be performed with the sets of outcomes and the results can be graphically represented with the Venn diagrams (Fig. 2):

**Union of events (Fig. 2A)**

Given events $A$ and $B$ in set $S$, we can build a new larger event as their union $A \cup B$ occurs when at least one of these two events occurs:

$$A \cup B = \{ \text{all outcomes in either } A \text{ or in } B \text{ or in both} \}$$

If we have a number of events $E_i$ equal to $n$ (where $i$ is a number from 1 to $n$) their union is denoted as:

$$\bigcup_{i=1}^{n} E_i = E_1 \cup E_2 \ldots \cup E_n = \{ \text{at least one of the } E_i \text{ occurred} \}$$
The union has the following properties:
1. The union of event A with an empty set is event A (identity law):
   \[ A \cup \emptyset = A \]
2. The union of event A with itself is event A (idempotent law):
   \[ A \cup A = A \]
3. The union of event A with the sample space is the sample space, or in other words, the event is dominated by the sample space (domination law):
   \[ A \cup S = S \]
4. The union of A and B is exactly the same as the union of B and A (commutative law):
   \[ A \cup B = B \cup A \]

Examples:
1.1. If one tosses a coin the resulting events can be denoted as A corresponding to heads (\( A = \{H\} \)) and B corresponding to tails (\( B = \{T\} \)). The union of A and B will include two elements — both heads and tails (\( A \cup B = \{H, T\} \)), corresponding to the whole sample space.
1.2. If one rolls a die and denote the results 1, 3 and 4 as event E (\( E = \{1, 3, 4\} \)) and results 1, 2 and 3 as event F (\( F = \{1, 2, 3\} \)), then the union of these two event E and F will be 1, 2, 3, 4 (\( E \cup F = \{1, 2, 3, 4\} \)). Note that results 1 and 3 are presented in both sets but only included once.
1.3. If one is playing roulette, the union of all uneven results and zero is \( S = \{0\} \cup \{1, 3, \ldots, 33\} \).

**Intersection of events (Fig. 2B)**
Given events A and B, we can build a new event as their intersection \( A \cap B \), which includes all the outcomes in both A and B and occurs when both A and B occur.
\[ A \cap B = \{\text{all outcomes in both } A \text{ and } B\} \]

Note that the intersection of multiple events \( E_i \) equal to \( n \) (where \( i \) is a number from 1 to \( n \)) is denoted as:
\[
\bigcap_{i=1}^{n} E_i = E_1 \cap E_2 \ldots \cap E_n = \{\text{all of the } E_i \text{ occurred}\}
\]

**Complement and symmetric difference (Fig. 2C and 2D)**
Complement of event A is the event “not A” (\( \bar{A} \)), which includes all outcomes not present in the event A (Fig. 2C). An extension of the complement is the symmetric difference, which represents for sets A and B all outcomes in A without B and in B without A (Fig. 2D):
\[
A \Delta B = (A \backslash B) \cup (B \backslash A)
\]
The frequentist approach can be illustrated with a following example:

We repeat an experiment N number of times under exactly the same conditions. Event A may occur or may not occur on each repetition. If \( N(A) \) is a number of times A occurs in these N observations, then we would expect that the ratio of \( \frac{N(A)}{N} \) tends to probability (p) as N tends to infinity:

\[
\frac{N(A)}{N} \rightarrow p \quad \text{as} \quad N \rightarrow \infty
\]

In that case p is the constant showing the probability of event A occurrence in any particular experiment — P(A). In that case this notion has the following properties.

1. If A is equal to zero, than the event never occurs and its probability is equal to 0:

   \[ \text{if } A = \emptyset, \text{ then } N(\emptyset) = 0, \text{ and } \frac{N(\emptyset)}{N} = 0 \]

2. If A includes all sample space, then the event occurs in every experiment and its probability is equal to 1:

   \[ \text{if } A = S, \text{ then } N(S) = 1, \text{ and } \frac{N(S)}{N} = 1 \]

3. If A and B are disjoint events, then number of occasions we see their union will be equal to the sum of N(A) and N(B), so their total probability will be equal to the sum of probabilities:

   \[
   \frac{N(A \cup B)}{N} = \frac{N(A)}{N} + \frac{N(B)}{N}
   \]

To describe this experiment, we can also use the **classical** approach, where the experiment is considered as sample space S. For event A, which belongs to the sample space S, we assume there is a set probability function P(A) that assigns a weight to A. Probability function P(A) satisfies the following properties:

1. It should be between zero and 1:

   \[ 0 \leq P(A) \leq 1 \]

2. Probability of the whole sample space should be equal to 1:

   \[ P(S) = 1 \]

3. If two events A and B do not overlap (their intersection is equal to 0), then the probability of their union will be equal to the sum of probabilities of each event:

   \[ P(A \cup B) = P(A) + P(B) \]

Using these axioms, we can produce the probability function. And for each sequence of events \( A_1, A_2, \ldots \) that are mutually exclusive, the probability of the union will be equal to the sum of their individual probabilities:

\[
P \left( \bigcup_{i=1}^{\infty} A_i \right) = \sum_{i=1}^{\infty} P(A_i)
\]

One should keep in mind that probability is countably additive. It means that to measure a set we can decompose it into finitely many disjoint parts (events), measure each part separately and then add up the results. If some event A is certain (happens in all observations), then its probability is equal to 1 (100%):

\[ P(A) = 1, \text{ if } A \text{ is certain} \]

If event A is impossible (never happens), then its probability is equal to zero:

\[ P(A) = 0, \text{ if } A \text{ is impossible} \]

Using this approach, we can consider several examples.

**Probability of the complement**

Consider event A, represented by a white circle on the Venn diagram (Fig. 2C). Let us assess the probability of the complement of this event — \( \overline{A} \), which denotes that no outcome in A occurs — it is
given in blue on the Venn diagram. By using the previously discussed postulates we can conclude the following:

The probability of the total sample space is equal to 1:

\[ P(S) = 1 \]

Event A and its complement are mutually exclusive so their total probability is equal to the sum of probabilities of each event:

\[ A \cap \bar{A} = 0 \quad \Rightarrow \quad P(A \cup \bar{A}) = P(A) + P(\bar{A}) \]  

(II)

Events A and \( \bar{A} \) are mutually exclusive and exhaustive so their union is equal to the total sample space \( S \):

\[ (A \cup \bar{A}) = S(A \cup \bar{A}) = S \]  

(III)

Considering steps I–III we can conclude that since the probability of \( S \) is equal to the probability of the union of events A and \( \bar{A} \), which in itself is equal to the sum of their probabilities, then the sum of probabilities of these events is equal to 1:

\[ I = P(S) = P(A \cup \bar{A}) = P(A) + P(\bar{A}) \]

Therefore, in this setting, the probability of an event can be derived by the probability of its complement:

\[ P(A) = 1 - P(\bar{A}) \]

**Probability of the union**

Consider two possible events A and B in the set \( S \) (shown in blue on the Venn diagram, Fig. 2A). Let us assess the probability of outcome in either A or B, which obviously is the probability of their union. The problem is that an intersection between A and B exists. That is why if we just add up the probabilities of A and B we would count this section twice, which is wrong. To measure the probability of a union properly we should take into account the following:

The union of A and B is equal to the union of A and \( \bar{A} \) and B:

\[ A \cup \bar{A} = A \cup (\bar{A} \cap B) \]

And the probability of the union of A and B is equal to the sum of these probabilities:

\[ P(A \cup \bar{A}) = P(A) + P(\bar{A} \cap B) \]  

(I)

On the other hand, the event B is equal to the union of intersection of A and B and intersection of area outside A (\( \bar{A} \)) and B:

\[ B = (A \cap B) \cup (\bar{A} \cap B) \]

\[ P(B) = P(A \cap B) + P(\bar{A} \cap B) \]  

(II)

Using statements I and II, we can rearrange the equations and substitute their parts to reach the expression for the probability of the union between events, which is equal to the sum of probabilities of A and B minus probability of their intersection:

\[ P(A \cup B) = P(A) + P(B) - P(A \cap B) \]

**Repeated coin tossing**

A fair coin (probability of getting head or tail is equal to 0.5 each) is tossed repeatedly. Using the probability theory, we may show that with total certainty, i.e. with probability equal to 1, head turns up sooner or later.

First, we need to assess the probability of the complement — that no head will ever appear. It is equal to the limit of \( n \) (\( n \) tends to infinity) the probability that no head occurs in the first \( n \) tosses. The probability that we get only tails is equal to \( \frac{1}{2^n} \) times. As \( n \) tends to infinity, this equation tends to 0:

\[ P(\text{no head ever}) = \lim_{n \to \infty} P(\text{no head for the first } n \text{ tosses}) = \lim_{n \to \infty} (2^{-n}) = 0 \]

The probability that head turns up is equal to one minus its complement (no head appears), which tends to zero. Therefore, the required probability is equal to:

\[ P(\text{head turns up}) = 1 - P(\text{no head ever}) = 1 - 0 = 1 \]

Probability equal to 1 means that head will turn up with total certainty sooner or later.

**Lifetime of cells**

The last example demonstrates a simple implementation of probability theory in biomedical research. The experiment consisted of measuring the lifetime of 200 cells (table 2). The results are shown in table 2.

Our task is to compute the probability that a cell lifetime is less than or equal to 1500 hours. First, we need to define the events: \( Sh \) — short, for event lasting less than 1000 hours; \( M \) — medium, for event lasting between 1000 and 1500 hours; \( L \) — long, for events lasting more than 1500 hours.

To assess the probability of lifetime to be \( \leq 1500 \) hours we need to assess the probability of the union of events \( Sh \) and \( M \), which is equal to the sum of their probabilities, as their intersection is equal to zero (disjoint events):

\[ P(h \leq 1500) = P(Sh \cup M) = P(Sh) + P(M) = 0.225 + 0.4 = 0.625 \]

Note that since events \( Sh \), \( M \) and \( L \) are not only disjoint, but also exhaustive, they form a partition of sample space.

**CONCLUSION**

In this first introductory article, we have briefly reviewed the current applications of Venn diagrams in...
clinical research and shown that even such a classical way of visualization is still under development. We have then given several tutorials discussing the basic concepts of probability as well as its relation to the set theory and the application of the Venn diagrams for graphical representation of these concepts. We understand that this material may give the impression of being purely abstract, however we stress that it is crucial to reach a good understanding of the principles of statistical analysis since Venn diagrams are often used in research papers.

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Nikolay M. Bulanov, Alexey A. Zaikin, Oleg B. Blyuss, Daniil B. Munblit, Tatiana V. Nazarenko and Maria Yu. Nadinskaia, participated in writing the text of the manuscript. Oleg B. Blyuss, Daniil B. Munblit and Tatiana V. Nazarenko searched and analyzed the literature on the review topic. Alexey A. Zaikin and Denis V. Butnaru developed the general concept of the article and supervised its writing. All authors participated in the discussion and editing of the work. All authors approved the final version of the publication.

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