INTRODUCTION

Toxicological-pharmacological experiments result in a great number of data. For the sake of their easier observation, interpretation, comparison and quantification of regularities, they should be processed and shown in a suitable manner. To this aim, tables showing mean values and percentages have been most commonly used and comparisons have been made using Gosset's (Student) - t-test or the analysis of variance, depending on the type of results.

In this paper, the application of the following methods is demonstrated:

1) regression analysis: non-linear (polynomial) function and linear (probit) analysis for the determination of LD₅₀-value,
2) analysis of variance (two- and three-way),
3) space histogram for result presentation, and
4) space fitting (three-dimensional non-linear bivariate fitting).

They will also show the problems encountered in space fitting.

The following examples have been used:

1) the effects of heavy metals (Cu, Zn and Pb), phenol and Na-salts on root length of onion (Allium caepa L.) and on the survival rate of water flea (Daphnia magna St.), which have been processed using: a) non-linear (polynomial) regression analysis, b) linear (probit) analysis;

* Correspondence
2) the interaction of Amikacin and Complamin (generic name: xanthinol nicotinate), processed by: a) space histogram, b) analysis of variance (two- and three-way);

3) the disintegration of pesticides under the influence of microorganisms using: a) space histogram, b) analysis of variance (two- and three-way);

4) the effects of different NaCl concentrations on the survival time and survival rate of fertilized egg cells of pond snail (Lymnaea stagnalis L.) using: a) space histogram, b) space fitting (three-dimensional non-linear bivariate fitting).

METHODS AND RESULTS

The effects of heavy metals, phenol and Na–salts on root length and survival rate

In the study of the effects of different heavy metal concentrations (Cu, Zn and Pb), phenol and Na-salts on root length of onion (Allium caepa L.) and on the survival rate of water flea (Daphnia magna St.) it was found that both the root length and the survival rate of the animal were in inverse proportion to the concentrations of tested materials. This applied to sublethal concentrations only while the lethal ones that completely inhibited root growth and the survival rate of the animals were 0% (1).

The results are presented in Fig. 1, where the concentrations (mmol l⁻¹, log scale) and the mean values for root length (expressed as a % of the control – 100%) and survival rate of the animals (as a % of the total number of animals subjected to a certain concentration – 100%) are plotted on the abscissa and ordinate, respectively.

![Graph](image)

Fig. 1. Comparison of the influences of heavy metal salts, phenol, and Na-salts on the root length of onion (Allium caepa L.) and the survival rate of water flea (Daphnia magna St.). Approximation curves result from polynomial regression analysis.
Determination of LD$_{50}$-values, as a measure of toxicity of the tested substances, can be done by determining the analytical expression in the first place, which best approximates the experimental results. In our case, the determination of the most applicable analytical expression was performed using the method of least squares, while the size of square error was the criterion of the suitability of the mathematical model (analytical expression) (2). A sufficiently precise approximation of the results, having 5–8 data points, is obtained by the application of the third- and fourth-order polynomial functions ($Y = a + bx + cx^2 + dx^3$) and ($Y = a + bx + cx^2 + dx^3 + ex^4$), respectively. The application of a higher order polynomial function (e.g. fifth-, sixth- or seventh-) is, however, more suitable because of the lower square error, but the final results (LD$_{50}$-values) do not essentially differ from those obtained by the application of third- and fourth-order polynomial functions. With the known analytical expression and the values for the parameters which determine it, it has not been difficult to determine the value for independent variable $X$ (LD$_{50}$) when the value for the dependent variable $Y$ is 50%. This can be done graphically or mathematically.

The determination of LD$_{50}$ using polynomial regression has the disadvantage that the LD$_{50}$-value is dependable on the values for the parameters that define the polynomial expression and that standard deviation (S.D.) of LD$_{50}$ depends on the standard error size of the regression model approximating experimental results. The higher the standard error of the analytical expression, the larger the standard deviation of the LD$_{50}$-value.

This disadvantage could be avoided by using probit analysis which means linearization of experimental data by log determination of the values for independent variable ($X$), while the values for dependent variable ($Y$) (in %) could be transformed in probit values (3). Fig. 2 shows these results in linear form.

![Graph](image-url)
Table I shows comparative LD$_{50}$-values and LD$_{50}$ ± S.D.-values obtained by non-linear (polynomial) and by linear (probit analysis) regressions, respectively.

Table I. Comparative presentation of LD$_{50}$-values obtained by non-linear and linear regression analyses
(in mmol l$^{-1}$)

<table>
<thead>
<tr>
<th></th>
<th>Non-linear (polynomial) regression analysis</th>
<th>Linear regression (probit) analysis</th>
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<tr>
<td></td>
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<td></td>
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<tr>
<td><strong>Allium caepa L</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CuSO$_4$</td>
<td>0.00112</td>
<td>0.00057 ± 0.00019</td>
</tr>
<tr>
<td>Pb(NO$_3$)$_2$</td>
<td>0.03976</td>
<td>0.02851 ± 0.00273</td>
</tr>
<tr>
<td>ZnSO$_4$</td>
<td>0.39916</td>
<td>0.33989 ± 0.01612</td>
</tr>
<tr>
<td>Phenol</td>
<td>3.02166</td>
<td>3.6559 ± 0.22159</td>
</tr>
<tr>
<td>Na$_2$SO$_4$</td>
<td>54.76006</td>
<td>54.68130 ± 4.06648</td>
</tr>
<tr>
<td>NaCl</td>
<td>191.73991</td>
<td>195.94100 ± 9.13503</td>
</tr>
<tr>
<td><strong>Daphnia magna St.</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CuSO$_4$</td>
<td>0.00115</td>
<td>0.00138 ± 0.00005</td>
</tr>
<tr>
<td>Pb(NO$_3$)$_2$</td>
<td>0.26854</td>
<td>0.18699 ± 0.01794</td>
</tr>
<tr>
<td>ZnSO$_4$</td>
<td>0.01151</td>
<td>0.01213 ± 0.00092</td>
</tr>
<tr>
<td>Phenol</td>
<td>0.10192</td>
<td>0.08494 ± 0.00840</td>
</tr>
<tr>
<td>Na$_2$SO$_4$</td>
<td>64.25605</td>
<td>60.28680 ± 1.14702</td>
</tr>
<tr>
<td>NaCl</td>
<td>81.19799</td>
<td>81.33070 ± 2.79145</td>
</tr>
</tbody>
</table>

The interaction of Amikacin and Complamin (generic name: xanthinol nicotinate)

Interaction of the antibiotic Amikacin and Complamin was monitored indirectly through the behaviour of animals (white laboratory mice bred at the biofarm of ICN - GALENKA – Institute), that is, through the ability of animals to swim or float. It was found that at lower concentrations of the antibiotic, animals could swim normally, while higher concentrations induced their floating on the surface. As the result of Amikacin + Complamin interaction, the floating reaction was observed at lower Amikacin concentrations (4).

In the quoted paper, the results were presented in tabular form, although the form of space histogram could be applied as well (Fig. 3, A and B) where the floating reaction (as % of the total number of animals subjected to a certain concentration — 100%) was plotted on the applicable axis, dependent variable ($Y$), as the function of concentration, on the abscissa (independent variable $X_1$) and as the function of time (duration of experiment, independent variable $X_2$), on the ordinate. The results presented either in tabular form or in the form of space histogram could be processed using two-way analysis of variance: a) the effect of Amikacin (control II), b) Amikacin + Complamin interaction, and c) three-way analysis of variance which is used for a comparison of Amikacin (control II) and Amikacin + Complamin interaction. The analysis of variance can also be used for the processing of results since you compare the number of animals which showed a certain reaction of changed behaviour (floating on the surface) to the total number of animals. The analysis of variance can be carried out according to the algorithm of Plohinskij (5, 6).
Fig. 3. Comparative space presentation of the influence of Amikacin (A) and Amikacin + Complanin (generic name: xanthinol nicotinate) (B) on the behaviour of animals. Figures in histograms represent numbers of animals (in % of the total number of animals subjected to a certain concentration) with which floating reaction was recorded.

a) Two-way analysis of variance: the effect of Amikacin (Fig. 3, A). — Factor A = time (5 grades: 1st, 7th, 14th, 21st and 28th day), factor B = concentration (7 grades: 0.9% NaCl (control), 9.5, 19.0, 37.5, 75.0, 150.0, and 300.0 mg kg$^{-1}$ of Amikacin.)
The results of two-way analysis of variance are quantified values for each factor (A, B) as well as for the effects of their 1st order interaction (AB). The importance of the influence of each factor and that of their interaction determines the values for Fisher's coefficient (F) for each factor and their interaction, for a certain number of degrees of freedom (d.f.).

Results: factor A: F = 7.793 (d.f. 4, 34); factor B: F = 14.854 (d.f. 6, 34); interaction AB: F = 2.030 (d.f. 24, 34); residual variance = 0.06508.

Comparison of the obtained and the tabulated values for F for each factor, as well as for their interaction, serves to determine whether the tested factor exerts either a significant or probably significant effect on the reaction monitored.

Interpretation of results: factor A: P < 0.0005, F exp. = 7.793 > F₀.₀₀₀₅ (d.f. 4, 34) = 6.82; factor B: P << 0.0005, F exp. = 14.854 >> F₀.₀₀₀₅ (d.f. 6, 34) = 5.66; interaction AB: P < 0.05, F exp. = 2.030 > F₀.₀₅ (d.f. 24, 34) = 1.93.

Factors A (time) and B (concentration) exert a statistically significant influence on the reaction monitored (P < 0.0005), while their 1st order interaction AB has a probable statistically significant influence (P < 0.05).

The experimental value for Fisher's coefficient (F) is the quotient of factor (organized) and residual (unexpected, internal) variances. The value for the residual variance is important for the determination of the significance of differences between individual experimentally obtained frequencies. The differences between certain frequencies can be compared using Scheffe's method (7) as confirmed by the examples from this paper.

Comparison of the differences between certain frequencies showed that the statistically significant difference (P < 0.01) between control I (0.9% NaCl) and animal reaction (in %) was obtained only when the floating reaction was recorded in 30% or more animals. This statistically significant difference in reactions occurred on the 21st and 28th days of the experiment at Amikacin concentrations of 150.0 and 300.0 mg kg⁻¹.

b) Two-way analysis of variance: Amikacin + Complamin (generic name: xanthinol nicotinate) interaction (Fig. 3, B). — Factor A = time (5 grades: 1st, 7th, 14th, 21st and 28th day), factor B = concentration [7 grades: 0.9% NaCl (control I), 9.5, 19.0, 37.5, 75.0, 150.0 and 300.0 mg kg⁻¹ of Amikacin]. Complamin was administrated in a single dose of 250.0 mg kg⁻¹.

Results: factor A: F = 7.769 (d.f. 4, 34); factor B: F = 10.809 (d.f. 6, 34); interaction AB: F = 1.037 (d.f. 24, 34); residual variance = 0.10794.

Interpretation of results: factor A: P < 0.0005, F exp. = 7.769 > F₀.₀₀₀₅ (d.f. 4, 34) = 6.82; factor B: P << 0.0005, F exp. = 10.809 >> F₀.₀₀₀₅ (d.f. 6, 34) = 5.66; interaction AB: P > 0.05, F exp. = 1.037 < F₀.₀₅ (d.f. 24, 34) = 1.93.

Each individual factor A (time) and B (concentration) exerts statistically very significant effect (P < 0.0005) on the reaction, which is not the case of their 1st order interaction (AB) (P > 0.05).

Comparison of the differences between certain frequencies showed that the statistically significant difference (P < 0.01) between control I (0.9% NaCl) and animal reaction (in %) was obtained only when the floating reaction was recorded in 30% of animals and even more. This statistically significant difference in reaction occurred on the 7th, 14th, 21st and 28th days of treatment at Amikacin concentrations of 37.5, 75.0, 150.0 and 300.0 mg kg⁻¹.
c) Three-way analysis of variance: comparison of the influence of Amikacin (control II) and Amikacin + Complainin (generic name: xanthinol nicotinate) interactions (Fig. 3, A and B). — The results of three-way analysis of variance are, besides quantified values for each individual factor (A, B and C), the influences of their 1st order (AB, AC and BC) as well as their 2nd order interactions (ABC).

Factor A = time (5 grades: 1st, 7th, 14th, 21st and 28th day); factor B = concentrations [7 grades: 0.9% NaCl (control I), 9.5, 19.0, 37.5, 75.0, 150.0 and 300.0 mg kg⁻¹ of Amikacin]; factor C = substance [2 grades: Amikacin (control II) and Amikacin + Complainin]. Complainin was administrated in a single dose of 250.0 mg kg⁻¹.

Results: factor A: F = 14.796 (d.f. 4, 630); factor B: F = 24.044 (d.f. 6, 630), P << 0.0005; factor C: F = 7.283 (d.f. 1, 630); interaction AB: F = 2.514 (d.f. 24, 630), P < 0.0005; interaction AC: F = 0.761 (d.f. 4, 630); interaction BC: F = 0.616 (d.f. 6, 630); interaction ABC: F = 0.307 (d.f. 24, 630); residual variance = 0.08651.

Interpretation of results: factor A: P << 0.0005, F exp. = 14.796 >> F₀.0005 (d.f. 4, 630) = 5.00; factor B: P << 0.0005, F exp. = 24.044 >> F₀.0005 (d.f. 6, 630) = 4.02; factor C: P < 0.01, F exp. = 7.283 > F₀.01 (d.f. 1, 630) = 6.72; interaction AB: P < 0.0005, F exp. = 2.514 > F₀.0005 (d.f. 24, 630) = 2.45; interaction AC: P >> 0.05, F exp. = 0.761 << F₀.05 (d.f. 4, 630) = 2.40; interaction BC: P >> 0.05, F exp. = 0.616 << F₀.05 (d.f. 6, 630) = 2.13; interaction ABC: P >> 0.05, F exp. = 0.307 << F₀.05 (d.f. 24, 630) = 1.58.

Each factor (A, B and C) as well as their 1st order interaction (AB) showed a statistically significant influence (P < 0.01 or P << 0.0005) on the behaviour of animals. The 1st order interactions (AC) and (BC) as well as the 2nd order interaction (ABC) had no statistically significant influence (P >> 0.05).

Statistically significant difference (P < 0.01) between control II (influence of Amikacin) and Amikacin + Complainin interaction was obtained only when the differences in floating reaction were equal or over 30%. This statistically significant difference occurred on the 14th day of the treatment at Amikacin concentration of 300.0 mg kg⁻¹ when the reaction in the control group (control II – Amikacin influence) was 20%, and 50% in the group submitted to Amikacin + Complainin interaction. The differences in reactions between these experimental groups occurred on the 7th, 14th, 21st and 28th days of treatment at Amikacin concentrations of 19.0, 37.5, 75.0, 150.0 and 300.0 mg kg⁻¹ but they were not statistically significant (the differences between the reactions were 10—20%).

Disintegration of pesticides under the influence of microorganisms

The disintegration of pesticide monocrotophus under the influence of microorganisms (Degrafen® – freeze-dried composition of aerobic and anaerobic saprophyte bacteria) was monitored indirectly through toxic effects of monocrotophus on the length of onion (Allium cepa L). Owing to its solubility in water, it was found that monocrotophus was prone to self-disintegration, which was more intense under the influence of microorganisms (8).

The results can be presented in the form of space histogram (Fig. 4, A and B) where root length (dependent variable Y, in % of the control – 100%) is plotted on the applicate axis as the function of time (independent variable Xₜ, duration of biological disintegration of the pesticide) on the absissa and as the function of dilution of biologically treated
pesticide (independent variable $X_2$) on the ordinate axis. The obtained results may be processed, as in the previous example, by two-way analysis of variance: a) self-disintegration of monocrotophos (control), b) microorganism-induced disintegration of monocrotophos, and c) three-way analysis of variance: comparison of self-disintegration and microorganism-induced disintegration.

![Diagram A: Monocrotophos](image)

![Diagram B: Monocrotophos + Degrafen](image)

Fig. 4. Comparative space presentation of the influence of self-disintegration of monocrotophos (A) and Degrafen®-induced disintegration of monocrotophos (B) on the root length of onion (*Allium cepa* L.). Figures in histograms represent root length (in % of the control – 100%).

a) **Two-way analysis of variance: self-disintegration of monocrotophos (Fig. 4A).** — Factor A = time (3 grades: 4th, 8th and 12th day); factor B = dilution (3 grades: direct, 1:1 and 1:5).

Results: factor A: $F = 0.100$ (d.f. 2, 8); factor B: $F = 1.235$ (d.f. 2, 8); interaction AB: $F = 0.109$ (d.f. 4, 8); residual variance = 0.13980.

Interpretation of results: factor A: $P >> 0.05$, $F_{exp.} = 0.100 << F_{0.05}$ (d.f. 2, 8) = 4.46; factor B: $P >> 0.05$, $F_{exp.} = 1.235 << F_{0.05}$ (d.f. 2, 8) = 4.46; interaction AB: $P >> 0.05$, $F_{exp.} = 0.109 << F_{0.05}$ (d.f. 4, 8) = 3.84.
Factors A (time) and B (dilution), as well as their 1st order interaction (AB), have no statistically significant influence (P > 0.05) on self-disintegration of monocrotrophos.

When none of the tested factors have statistically or probably significant influence, it is not recommended to determine the significances between some frequencies. If the difference was significant, it would manifest itself at least through probable statistically significant influence of some of the factors studied.

b) Two-way analysis of variance: microorganisms-induced disintegration of monocrotrophos (Fig. 4B). — Factor A = time (3 grades: 4th, 8th and 12th day); factor B = dilution (3 grades: direct, 1:1 and 1:5).

Results: factor A: F = 0.656 (d.f. 2, 8); factor B: F = 5.072 (d.f. 2, 8); interaction AB: F = 0.106 (d.f. 4, 8); residual variance = 0.19070.

Interpretation of results: factor A: P > 0.05, F exp. = 0.656 < F0.05 (d.f. 2, 8) = 4.46; factor B: P < 0.05, F exp. = 5.072 > F0.05 (d.f. 2, 8) = 4.46; interaction AB: P > 0.05, F exp. = 0.106 < F0.05 (d.f. 4, 8) = 3.84.

Factor B (dilution) was shown to have probable statistically significant influence (P < 0.05) on microorganism-induced disintegration of monocrotrophos, while factor A (time) and the interaction AB induced no statistically significant influence (P > 0.05).

Comparison of the differences between some frequencies has shown that the existing differences are statistically significant (P < 0.05).

c) Three-way analysis of variance: comparison of self-disintegration of monocrotrophos and microorganism-induced disintegration of monocrotrophos (Fig. 4, A and B). — Factor A = time (3 grades: 4th, 8th and 12th day); factor B = dilution (3 grades: direct, 1:1 and 1:5); factor C = substance (2 grades: monocrotrophos and monocrotrophos + Degrafen).

Results: factor A: F = 10.570 (d.f. 2, 1782); factor B: F = 100.466 (d.f. 2, 1782); factor C: F = 51.295 (d.f. 1, 1782); interaction AB: F = 2.000 (d.f. 4, 1782); interaction AC: F = 220.829 (d.f. 2, 1782); interaction BC: F = 18.705 (d.f. 2, 1782); interaction ABC: F = 107.358 (d.f. 4, 1782); residual variance = 0.00965.

Interpretation of results: factor A: P < 0.0005, F exp. = 10.570 > F0.0005 (d.f. 2, 1782) = 7.60; factor B: P < 0.0005, F exp. = 100.466 > F0.0005 (d.f. 2, 1782) = 7.60; factor C: P < 0.0005, F exp. = 51.295 > F0.0005 (d.f. 1, 1782) = 12.10; interaction AB: P > 0.05, F exp. = 2.000 < F0.05 (d.f. 4, 1782) = 2.37; interaction AC: P < 0.0005, F exp. = 220.829 > F0.0005 (d.f. 2, 1782) = 7.60; interaction BC: P < 0.0005, F exp. = 18.777 > F0.0005 (d.f. 2, 1782) = 7.60; interaction ABC: P > 0.05, F exp. = -107.358 < F0.05 (d.f. 4, 1782) = 2.37.

Comparison of the process of self-disintegration of monocrotrophos to that of microorganism-induced disintegration of monocrotrophos has shown that individual factors A, B and C and their 1st order interactions AC and BC have statistically significant influence on the processes (P < 0.005 or P < 0.0005). The 1st order interaction AB and the 2nd order one ABC have not induced statistically significant influence (P > 0.05). As it can be seen, the value for interaction coefficient F of the second order (ABC) is negative (F = -107.358).

Comparison of the differences between some frequencies observed in self-disintegration of monocrotrophos (control) and in microorganism-induced disintegration of monocrotrophos has shown that they are statistically significant only at the 1:5 dilution for all three time grades (4th, 8th and 12th day) (P < 0.001). The differences in reaction for all time-grades and for the dilution 1:1 on the 4th and 8th days are not statistically
significant ($P >> 0.05$), while the differences in reaction for the dilution 1:1 on 12th day are probably statistically significant ($0.05 > P > 0.01$).

**Effect of various NaCl concentrations on survival time and survival rate**

The study of the effects of various NaCl concentrations on the survival time of fertilized egg cells of pond snail, *Lymnaea stagnalis* L., has shown that the survival time decreases as a function of NaCl concentration. This process can be defined by double exponential function of the type: $Y = ae^{-bx} + ce^{-dx} + f$ (9). The results are shown in Fig. 5 where NaCl concentrations (in mmol l$^{-1}$) are presented on the abscissa and the mean survival time of fertilized egg cells (in days) on the ordinate.

![Fig. 5. Mean values of the survival time of fertilized egg cells of pond snail (*Lymnaea stagnalis* L.) as a function of NaCl concentration.](image)

Although the presentation of the means for the survival time of fertilized egg cells as the function of NaCl concentration was illustrative enough, the process could be anticipated in more detail using three-dimensional presentation (Fig. 6) where the kinetics of the survival rate of fertilized egg cells is presented in the form of space histogram: applicate – dependent variable $Y$ (in %), abscissa – as the function of experiment duration – independent variable $X_1$ (in days) and ordinate – as the function of NaCl concentration – independent variable $X_2$ (in mmol l$^{-1}$).

In the absence of a common mathematical model describing the function $Y = f(X_1, X_2)$, three-dimensional fitting was performed using trivariate non-linear procedure where the functions of time and concentration were fitted separately. The result of such fitting is shown in Fig. 7. However, such a procedure leads to significant deviations of a fitted form from the original one. Fig. 7 shows the deviations which are especially distinguishable while fitting the function of concentrations for days 5, 6 and 7. They consist of the following: according to Fig. 6 (space histogram), the last experimentally obtained column in the histogram for day 5 corresponds to NaCl concentration of 102.66 mmol l$^{-1}$ while according to Fig. 7 (space fitting), the last point obtained as the result of fitting corresponds to NaCl concentration as high as 154.00 mmol l$^{-1}$. The last experimentally obtained column in the histogram for day 6 corresponds to NaCl concentration of 34.22 mmol l$^{-1}$ (Fig. 6) and the last point which is the result of fitting corresponds to NaCl concentrations of 136.89 mmol l$^{-1}$ (Fig. 7). The only experimentally obtained column in the histogram for day 7 corresponds to NaCl concentration of 17.11 mmol l$^{-1}$ (Fig. 6) while the last fitting point corresponds to NaCl concentration as high as 102.66 mmol l$^{-1}$. In the fitting of time
function, the most obvious deviation appears at NaCl concentration of 85.55 mmol l⁻¹ where the last experimentally obtained column in the histogram appears on day 5 (Fig. 6) while the last fitting point at this concentration appears on day 8 (Fig. 7).

**Fig. 6.** Kinetics of the survival rate of fertilized egg cells of pond snail (*Lymnaea stagnalis* L.) as functions of NaCl concentration and time (three-dimensional space histogram).

**Fig. 7.** Kinetics of the survival rate of fertilized egg cells of pond snail (*Lymnaea stagnalis* L.) presented in the form of three-dimensional space fitting.
The deviations caused by trivariate fitting can be minimized using bivariate fitting: one independent variable is fitted (in our case it was the function of time) while the other retains its constant value. Bivariate fitting of time function is presented in Fig. 8 where para-coordinate planes have been used to present the kinetics of the survival rate of fertilized egg cells.

Fig. 8. Kinetics of the survival rate of fertilized egg cells of pond snail (*Lymnaea stagnalis* L.) presented on para-coordinate planes (bivariate fitting of time function).

DISCUSSION

The examples used in this paper to demonstrate some mathematical-statistical methods have shown their potentials as well as their limitations.

Regression and probit analyses have an essential advantage over other methods for LD50 determination. Their advantage lies in that they can be used in situations when either the interval between the tested doses or organism count in the doses varies, that is, when both different intervals between the doses and organism counts in them vary. Similar methods are those according to Litchfield-Wilcoxon (10) and Miller-Tainter (11). In contrast, the methods according to Kärber (12), Behrens (13), Behrens-Schlosser (14) have a limitation: the same organism count in the dose and the same interval between the doses. In this paper we have presented the above methods: both the organism count and the interval between the doses have varied (effect of phenol, heavy metals and Na-salts on *Allium caepa* L.), and the constant organism count, but with varied intervals between the doses (the effect of heavy metals, phenol and Na-salts on *Daphnia magna* St.). As presented in
Table I, similar LD<sub>50</sub>-values are obtained if both regression and probit analyses are applied to the processing of the same experimental data. The application of probit analysis is, however, more suitable because LD<sub>50</sub>-values depend on the order of polynomial function, that is, on the description parameter values for the polynomial function. The differences are sometimes significant.

Presentation of the results in three dimensions is more comprehensive and informative than tabular and two-dimensional ones. The results presented in the above manner can point out the method suitable for their processing. An example to the point is the Amikacin and Complamin interaction and microorganism-induced disintegration of pesticides. The results have been processed by two- and three-way analysis of variance. This analysis gives not only the data on the presence of significant influences of the factors tested but can produce the data relative to the phase of experiment in which the influence of a factor becomes significant. Literature contains the procedure for processing results using the analysis of variance; however, the most comprehensive algorithms are contained in the publications of Plohinskij (5, 6).

Three-dimensional fitting is a logical extension of three-dimensional presentation of the results and a first step towards processes modelling, that is, towards the analytical expression that would make a space description of the function \( Y = f(X_1, X_2) \). In our example, three-dimensional fitting has been carried out using the trivariate technique by which the functions of time \( Y = f(X_1) \) and concentration \( Y = f(X_2) \) are fitted separately. Logistic function \( Y = \frac{K}{1 + e^{-a-bx}} \) (15) has been used as the mathematical model for the fitting of both functions. The result of such fitting, however, are certain deviations. The nature of the most suitable connections between logistic functions fulfilling the condition \( Y = f(X_1, X_2) \) remains to be studied.

The examples presented serve as an orientation of a possible application of the mentioned mathematical-statistical methods, which can be used for processing some different results as well. However, the choice of the method for processing depends on the type of the results.

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SAŽETAK

Prikaz nekih matematičko-statističkih metoda pogodnih za obradu rezultata toxikološko-farmakoloških eksperimenata

MILAN B. ARAMBASIQ, SVETLANA KONIDIQ, LJILJANA PTIQ I Miodrag Stojanović

U radu je na primerima: 1) uticaja soli teških metala, fenola i soli Na, na dužinu korjenića semenskog crnog luka (Allium caepa L.) i stopu preživljavanja velike vodene buve (Daphnia magna St.), 2) interakcija Amikacina i Komplamina (generičko ime: ksantinol nikotinit), 3) razgradnje pesticida monokrotofos-a pod uticajem mikroorganizama i 4) uticaja raznih koncentracija NaCl na vreme i stopu preživljavanja oplodjenih jajnih čelija barskog puža (Lymnaea stagnalis L.), prikazana primena matematičko-statističkih metoda i to: 1) regresione analize (linearne i nelincarne), 2) analize varijanse (dvo- i tro-faktorske), 3) prostornog histograma i 4) prostornog (trodimenzionalnog) fitovanja, za obradu rezultata toxikološko-farmakoloških eksperimenata. Prikazane su i diskutovane mogućnosti i ograničenja ovih metoda, kao i uslovi za njihovo korišćenje.

JCN-GALENIKA – Institut
Centar za kontrolu kvaliteta
Bioško-farmakološka služba,
Beograd, 29. Novemba 111, Jugoslavija