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# New Approach to the Safety Evaluation of Textile Goods. Part I. Bioindicative Measuring Method for Formaldehyde Content in Textiles

**Abstract**
*This article presents research results of the adaptation of the bioinducative measuring method to evaluate the toxicity of textiles. In the experiment, Tetrahymena pyriformis as a bioindicator and formaldehyde, a well known toxic substance used in some finishing processes, were used as a sample chemical substance. To examine the usefulness of the bioindicative method in the safety evaluation of textiles, formaldehyde harmfulness within the range 1.5 – 50 mg/L of the solution and extracts of textiles with an analytically determined content of formaldehyde were analysed in the presence of a bioindicator. Changes in cell development in formaldehyde solutions, extracts as well as in group control, were registered by directly measuring the optical density over time (0 - 24 h period), and microscopic observations and absorbance measurements were made in the presence of the alamarBlue® cell viability reagent. Empirical data enabled to fit logistic curves, on the basis of which the level of formaldehyde toxicity in dependence on time was estimated. The experiment performed showed a high toxicity of formaldehyde as well as the possibility of the application of the bioindicative measuring method based on ciliate to estimate very small amounts of formaldehyde in water solutions and textile extracts.*

**Key words:** *safety of textiles, formaldehyde, bioindicative method, Tetrahymena pyriformis.*

## Introduction

To assess the safety of textile products, many laboratory tests that enable to detect different harmful substances have to be performed. They require special preparation of samples and various expensive equipment. For example, the content of formaldehyde is evaluated colorimetrically by means of absorbance measurements of water extracts of materials in the presence of Nash's reagent, which forms a colour complex with free formaldehyde. Azo dyes and amines are detected by means of liquid chromatography using a diode array detector (DAD). Pesticides and pentachlorophenol are measured and quantified by means of gas chromatography through detection using an electron capture detector (ECD), where heavy metal detection needs the use of Atomic Absorption Spectrometry (AAS) [1]. Although these methods are certainly very precise, they are not sufficient to provide information on the harmful effect of chemicals analysed on a human, nor concerning unknown chemicals that could exist in textiles. Such information could be provided only from toxicological methods, which could enable not only to simplify the estimation of hazardous substances in products but could simultaneously show the real risk [2, 3] connected with extraction of these substances from textiles. The advantage of toxicological assays is their ability to react or detect only the available fraction of potentially hazardous substances, including their synergic effect, whereas common analytical methods are not able to distinguish between fractions of substances that are available or non-available to biological systems.

The aim of this paper is to present the results of research on the application of a bioindicative measuring method in the safety evaluation of textile products in the context of hazards related to the presence of harmful substances. Since the scope of this topic is very extensive, the first part of this paper focuses on the presentation of two analytical issues:

- determination of the toxicity level for formaldehyde using *Tetrahymena pyriformis* as a bioindicator, and
- safety evaluation of textile extracts using the bioindicative method.

## Formaldehyde as a harmful substance

The choice of formaldehyde as a sampling test substance results from its usage universality and well known toxicity for human and other organisms [4]. Formaldehyde is a compound that has been used for many years in the textile industry as a constituent of methylated or propylene urea – formaldehyde resins, e.g. in some finishing processes such as resist-dyeing, where it performs the role of a fixing agent, improving the resistance of dyeing, or in the finishing of creaseproof woven fabrics made from cellulose fibres as a cross-linking agent [5 - 7]. The research studies proved that this compound is hazardous to animals and humans. For example, for formaldehyde, $LD_{50}$ (skin, rabbit) was fixed at a level of 270 mg/kg of the body mass, while $LD_{50}$ (oral, rat) was 100 mg/kg of the body mass. Although the results of epidemiological tests of its carcinogenic activity in humans bring no equivocal conclusions, it is considered that formaldehyde may cause cancer. It was classified into Class 3 of carcinogenic substances. It is thought that it is even more harmful in such effects as this substance can be released both in the fixation process and during the use of a finished product. Depending on its concentration, this may cause irritation in the conjunctiva and mucous membranes of the upper respiratory tract (this happens more often due to gaseous formaldehyde release in the work environment of finishing rooms) and may be allergic to skin (during permanent use of clothes containing formaldehyde, which could be extracted from textiles by sweat) [8].

Due to the recognised toxicity of formaldehyde, its use has for some time come under closer scrutiny, leading to zero formaldehyde finishes, e.g. polycarboxy-

lic acids, being developed. However, the performance of such finishes is generally either poorer and/or more expensive to achieve [6], hence formaldehyde is still commonly used in the textile industry. To reduce the application of formaldehyde in the textile industry, thus assuring safe use of products, the obligatory [9] and facultative (ecolabel, GPP, oecotex) requirements establishing the limits for permissible concentrations of this substance in products are set forth. However, when analysing these requirements, some doubts arise about the basis on which these requirements are set down, and thus also about the limits specified. Moreover, as results from the reports published by the Monitoring System for Dangerous Products RAPEX [10] and the Trade Inspection, the exceedance of the permissible formaldehyde concentration is one of the problems that occurs more often in the safety assurance of textile products offered on the Polish and other EU markets. In particular, this applies to children's clothes, in which the maximum allowable concentration of formaldehyde in accordance with applicable regulations [9] should not exceed 20 mg/kg of the product.

## Bioindicative methods in toxicometric studies

To gain knowledge of real hazards created by substances present in products, it is necessary to carry out toxicometric studies to find the functionally or morphologically harmful activity of the substance under investigation and to derive qualitative and quantitative characteristics of such activity [3]. Obviously the most reliable information might be gathered from studies carried out directly on humans. However, this is impossible for ethical reasons. Therefore, studies are conducted by employing *in vivo* methods, i.e. on whole organisms (e.g. on such animals as rats or mice) or their organs, and more often with *in vitro* methods, i.e. by using cell cultures as a kind of bioindicator. Literature on the subject gives numerous examples where bioindicative methods are used in water and sewage quality tests and for determining the toxicity level of various substances or products [11]. It follows from these studies that the harmfulness level for formaldehyde cannot be established unequivocally, varying depending on the test organism and method used [12 - 14].

## Procedure of the method

In this study, the strain *Tetrahymena pyriformis* (Ehrenberg) Wolff 1947 of reference number CCAP 1630/1W, originating from the Culture Collection of Algae and Protozoa, Ambleside UK, was used. These organisms were selected due to the fact that *Tetrahymena pyriformis* meets most requirements for test organisms and is one of the bioindicators commonly used in laboratory tests. *Tetrahymena* belonging to protozoans is an unicellular organism with a cell membrane of quite different structure than those of bacteria, yeasts or algae that form a barrier to toxic compounds. In the case of *ciliate sp.,* the interior of the cell is separated from its environment by a thin cell membrane only, thus causing that ciliates are very sensitive, even to traces of toxic compounds in their environment. In addition, in terms of vital functions, cellular structures and gene functionality, *Tetrahymena* cells are closer to human cells than other model microorganisms [15 - 18].

Ciliates were prepared as axenic cultures in 18 × 1.2 cm test-tubes containing 10 mL of standard agar PPY (Difco). The cultures grew in Sanyo MLR-350 climate chambers and a MEMMERT INB 400 incubator at 20 °C in total darkness. Every week new cultures were inoculated in sterile conditions by placing 200 μL of inoculum in a new test-tube containing 10 mL of sterile PPY agar. In such conditions the growth of *T. pyriformis* cultures was consistent with the theory [19 - 21], verified by own experiment. For toxicometric purposes, culture incubated for 48 hours, being in the 'log' phase, i.e. at the intensive growth stage, was used.

To determine formaldehyde harmfulness, the optical density of cell culture [13, 18, 22, 23] in formaldehyde solutions was directly measured, and absorbance measurements in the presence of the alamarBlue® cell viability reagent [24] were made. Formaldehyde solutions of 6 concentration levels within the range 1.5 – 50 mg/L (the choice of concentrations levels was proceeded with preliminary experiments on concentrations within the range 1.5 – 150 mg/L) were placed in 1.5 mL microcuvettes, and then test microorganisms suspended in agar were added, thus obtaining a cell density of approx. 5 000 cells/mL. The samples prepared were incubated at 28 °C. Changes observed in the samples analysed were assessed based on:

- turbidimetric measurements of direct readings of solution absorbance (optical density) measured at 330 nm after 4, 6, 8 and 24 hours;
- microscope observations;
- organoleptic assessment of colour changes for the cell vitality indicator;
- colorimetric measurements of absorbance at 570 nm, using 600 nm as the reference wavelength, in the presence of almarBlue® added after the incubation of *Tetrahymena pyriformis* for 4 hours in formaldehyde solutions. In this method, measurements were made 2, 4 and 20 hours after the indicator was added.

Moreover, measurements were also made for *Tetrahymena pyriformis* culture solutions in spring water. These measurements were treated as the so-called control group.

The usefulness of the bioindicative method in the evaluation of textile safety was verified by taking, as an example, three 100% cotton fabrics whose formaldehyde content exceeded the limits allowed. The amount of free and hydrolysed formaldehyde within the samples tested, determined in accordance with Standard PN-EN ISO 14184-1:2001 [25], was as follows: 32 mg/kg within sample No. 1 (bedding textile for children), 50 mg/kg within sample No. 2 (bedding textile for children) and 2 200 mg/kg within sample No. 3 (clothes textile). The bioindicative method based on *Tetrahymena pyriformis* was performed in 10% water extracts of the samples, prepared on the basis of Standard PN-EN ISO 14184-1:2001. The proliferation of *Tetrahymena pyriformis* culture in the extracts was analysed by means of turbidimetric measurements and colorimetric measurements in the presence of the alamarBlue® cell viability reagent for an incubation time of 6 and 24 hours.

## Analysis of the optical density of *Tetrahymena pyriformis* culture in formaldehyde solutions

When analysing the effect of *Tetrahymena pyriformis* culture incubation time on the optical density (absorbance) values, distinct changes in absorbance were observed for measurements made after the incubation of *Tetrahymena pyriformis* both in spring water and formaldehyde

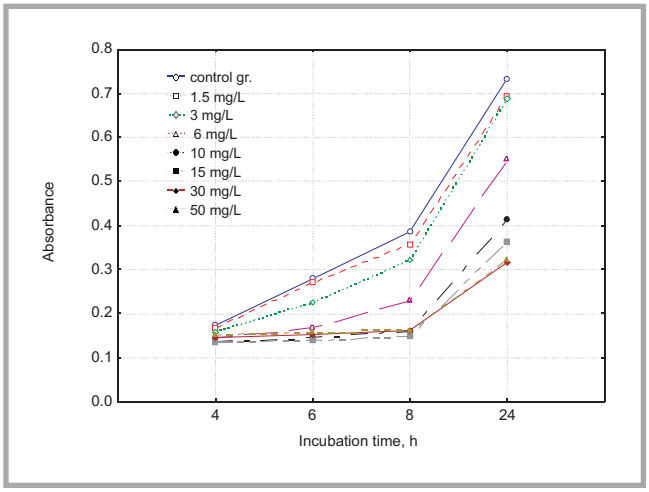


***Figure 1.*** *Effect of incubation time on absorbance values of formaldehyde solutions in relation to the concentration level.*

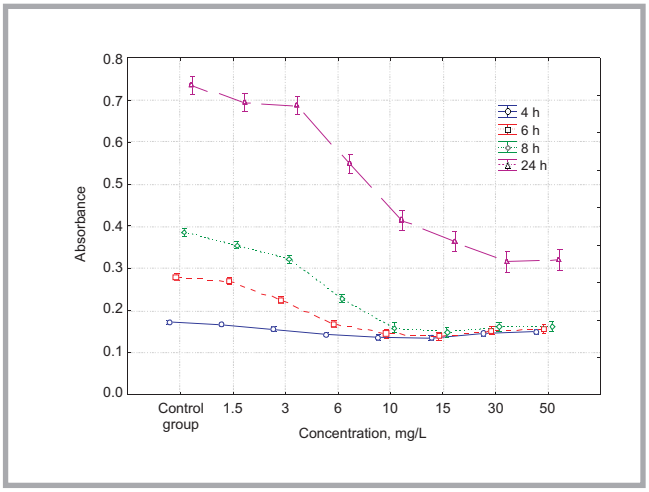


***Figure 2.*** *Effect of formaldehyde concentration on solution absorbance values in time in relation to 95% confidence intervals for individual absorbance means.*

solutions for at least 6 hours, as shown in ***Figure 1***.

Statistical analysis of empirical data by employing post hoc tests (Tukey's test) confirmed the existence of statistically significant differences between the absorbance measurements within the range 6 – 24 h and lack of them for measurements made at the beginning of the experiment and after 4 hours of incubation (***Table 1***).

The results of direct measurements (***Figure 2***) indicate that even very low formaldehyde concentrations in the range 1.5 – 6 mg/L cause a slower absorbance increase compared to those of the control group. For example, after incubation for 6 hours the absorbance values of the solutions analysed were at the following levels: 79.6% of the control for a concentration of 1.5 mg/L, 51.5% - for 3 mg/L and 23.6% - for solutions of 6 mg/L concentration (***Table 2***).

The post-hoc analysis (***Table 1***) showed that there are statistically significant differences between absorbance values for concentrations in the range 1.5 – 6 mg/L measured after 6 h, 8 h and 24 h. This analysis also indicates a lack of significant differences between the control and formaldehyde concentration of 1.5 mg/L. Parallel microscopic observations showed that formaldehyde concentrations at a level of 1.5 – 6 mg/L cause no noticeable lethality of test cells. The microscopic image revealed quite numerous and mobile cells that showed proliferation ability, causing a noticeable increase in optical density after 8 and 24 hours of incubation. At higher formaldehyde concentrations in the solution, the absorbance value increments were very small - at a level of approx. 13% of the control for a formaldehyde concentration of 10 mg/L after 6 and 8 hours of incubation and below 10% for formaldehyde solutions of concentrations above 15 mg/L. The post-hoc tests confirmed that these differences had no statistical significance. Microscopic observations indicated that the ratio of dead to live cells in the solution increases with an increasing formaldehyde concentration, and at a concentration of 50 mg/L no live cells were observed after 4 hours of incubation. Moreover, microscopic observations showed behavioural and morphological changes in *Tetrahymena pyriformis*. At a formaldehyde concentration range of 15 – 30 mg/L, there were a few live cells of small mobility, rounded shape and considerably enlarged vacuoles.

***Table 1.*** *Results of post-hoc analysis of absorbance measurements of formaldehyde solutions of various concentrations in relation to the incubation time. The values marked with an asterisk show statistically significant differences in the average absorbance level between 2 groups defined at the intersection of an appropriate row and column (a dash indicates no significant differences). Within each table cell the first item indicates the measurement made after 4 hours, the second after 6 h, the third after 8 h and the fourth after 24 h.*

| | Control group | 1.5 mg/L | 3 mg/L | 6 mg/L | 10 mg/L | 15 mg/L | 30 mg/L | 50 mg/L |
|---|---|---|---|---|---|---|---|---|
| Control group | | - / - / - / * | - / * / * / * | - / * / * / * | - / * / * / * | * / * / * / * | - / * / * / * | - / * / * / * |
| 1.5 mg/L | - / - / - / * | | - / * / * / - | - / * / * / * | - / * / * / * | - / * / * / * | - / * / * / * | - / * / * / * |
| 3 mg/L | - / * / * / * | - / * / * / - | | - / * / * / * | - / * / * / * | - / * / * / * | - / * / * / * | - / * / * / * |
| 6 mg/L | - / * / * / * | - / * / * / * | - / * / * / * | | - / - / * / * | - / - / * / * | - / - / * / * | - / - / * / * |
| 10 mg/L | - / * / * / * | - / * / * / * | - / * / * / * | - / - / * / * | | - / - / - / * | - / - / - / * | - / - / - / * |
| 15 mg/L | * / * / * / * | - / * / * / * | - / * / * / * | - / - / * / * | - / - / - / * | | - / - / - / * | - / - / - / * |
| 30 mg/L | - / * / * / * | - / * / * / * | - / * / * / * | - / - / * / * | - / - / - / * | - / - / - / * | | - / - / - / - |
| 50 mg/L | - / * / * / * | - / * / * / * | - / * / * / * | - / - / * / * | - / - / - / * | - / - / - / * | - / - / - / - | |

***Table 2.*** *Changes in the optical density of T. pyriformis culture in formaldehyde solutions.*

| Incubation time, h | Normalised relative increase in the optical density of *T. pyriformis* culture in formaldehyde solutions in comparison to the group control, % | | | | | | |
|---|---|---|---|---|---|---|---|
| | Concentration of formaldehyde solutions, mg/L | | | | | | |
| | 1.5 | 3 | 6 | 10 | 15 | 30 | 50 |
| 4 | 61.61 | 38.43 | 29.22 | 25.82 | 15.25 | 18.99 | 30.59 |
| 6 | 79.64 | 51.51 | 23.58 | 13.46 | 7.40 | 10.45 | 13.41 |
| 8 | 77.84 | 64.37 | 36.37 | 13.62 | 7.43 | 9.00 | 10.13 |
| 24 | 84.11 | 81.86 | 65.22 | 46.83 | 37.08 | 26.88 | 28.01 |

## Analysis of absorbance measurements of *Tetrahymena pyriformis* in formaldehyde solutions in the presence of alamarBlue®

The use of the alamarBlue® cell viability reagent allows both qualitative and quantitative harmfulness assessment of the

substance under investigation, as shown below. Colour changes of alamarBlue® in formaldehyde solutions with *Tetrahymena sp.* after 6 and 24 hours of incubation are illustrated in ***Figure 3***.

The pink colour characteristic of control and formaldehyde concentrations of 3 and 6 mg/L indicates a large amount and high vitality of test cells that completely reduced alamarBlue® after 24 hours of incubation. For the solution at a concentration of 10 mg/L the colour violet can be observed, thus leading to the conclusion that the solution contains far fewer live cells of considerably lower vitality than those of the control. In solutions at concentrations above 15 mg/L the colour blue predominates. This colour characteristic of a non-reduced form indicates a lack of live cells or loss of their metabolic abilities. Colour changes visible to the naked eye are accompanied by absorbance changes. The values of the vitality index presented in ***Table 3*** explain the behaviour of *Tetrahymena pyriformis* cells in formaldehyde solutions and confirm the conclusions derived from the results of optical density direct measurements.

Formaldehyde at concentrations ranging from 1.5 to 6 mg/L impedes the growth rate of ciliates, while at concentrations ≥ 15 mg/L it causes the dieback of cells, as evidenced by values obtained at various times during absorbance measurements after alamarBlue® was added. At lower formaldehyde concentrations, the percentage of alamarBlue® reduction increases with the incubation time, while at higher concentrations this increase is inconsiderable (15 mg/L after 24 hours of incubation) or practically unnoticeable (≥ 30 mg/L). The cell vitality indices computed from absorbance measurements made after 24 hours of incubation and the photograph presented in ***Figure 3*** indicate that *Tetrahymena* cells present in formaldehyde solutions at concentrations below 15 mg/L retain their metabolic abilities for a long time.

## Estimation of the formaldehyde toxicity level based on the regression of logistic function

To assess the formaldehyde toxicity level, the data obtained with the two methods were transformed to a form enabling the percentage absorbance increase suppression coefficient in relation to the control

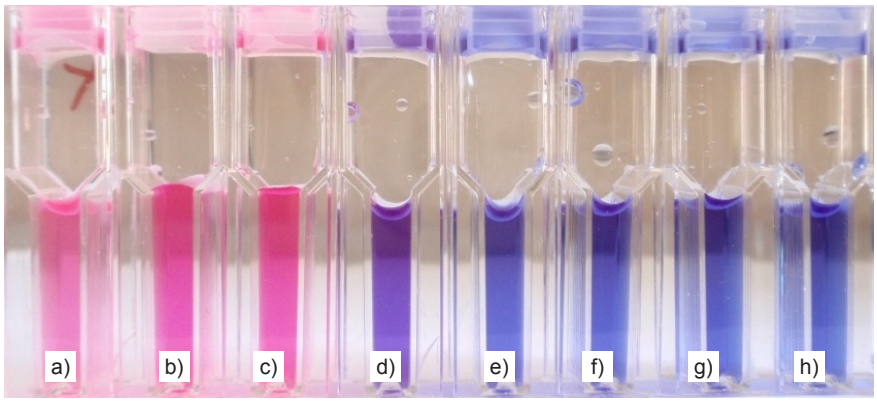


***Figure 3.*** *Colour changes of alamarBlue® in solutions of Tetrahymena pyriformis cultures incubated in formaldehyde for 24 hours; a) control, b) 3 mg/L, c) 6 mg/L, d) 10 mg/L, e) 15 mg/L, f) 30 mg/L, g) 50mg/L, h) blank.*

to be determined for solutions of the substances under investigation. To compute values of this coefficient, logistic curves expressed by ***Equation 1*** were fitted:

$$f(c) = \frac{1}{1 + \exp\{b(\ln c - \ln IC_{50})\}} \qquad (1)$$

where:

$c$ – test substance concentration,

$b$ – regression coefficient (informs about the rate at which the transition from zero to total suppression (100%) of bioindicator growth occurs. The lower this parameter, the faster the transition),

$IC_{50}$ – xenobiotic concentration at which 50% of the effect of absorbance changes occurs in relation to maximum values for the control group.

It was assumed that the inferior limit for the model is 0% (no absorbance increase suppression in relation to the control group), while the superior limit is 100% (complete suppression of absorbance increase in relation to the control group, tantamount to a halt in bioindica-

***Table 3.*** *Effect of formaldehyde concentration on the proliferation of Tetrahymena sp. culture computed from absorbance measurements in the presence of alamarBlue®.*

| Total incubation time, h | Percentage of alamarBlue® reduction in formaldehyde solutions | | | | | | |
|---|---|---|---|---|---|---|---|
| | Control group | Concentration of formaldehyde solutions, mg/L | | | | | |
| | | 1.5 | 3 | 6 | 15 | 30 | 50 |
| 6 | 52.41 | 37.32 | 20.14 | 12.53 | 7.45 | 7.53 | 8.83 |
| 8 | 98.30 | 62.99 | 38.70 | 17.40 | 7.85 | 7.85 | 8.37 |
| 24 | 100 | 97.30 | 88.72 | 92.75 | 28.12 | 9.30 | 8.37 |

***Table 4.*** *Parameters of the logistic function approximated from computed values of the percentage absorbance increase suppression coefficient for formaldehyde solutions compared to the control group based on direct measurements of absorbance. * The value $IC_{50}$ for an incubation time of 4 hours was omitted in the substantial analysis due to a low value of the coefficient of determination and lack of statistically significant differences between empirical values for various concentration levels.*

| Incubation time, h | b value | p value | $IC_{50}$ | p value | 95% confidence level for $IC_{50}$ | | $R^2$ |
|---|---|---|---|---|---|---|---|
| 4* | -0.526 | < 0.01 | 1.746 | 0.02 | 0.385 | 3.108 | 0.49 |
| 6 | -1.590 | | 3.176 | < 0.01 | 2.448 | 3.904 | 0.84 |
| 8 | -1.550 | | 3.936 | | 3.028 | 4.848 | 0.86 |
| 24 | -0.904 | | 10.706 | | 7.595 | 13.816 | 0.80 |

***Table 5.*** *Parameters of the logistic function approximated from computed values of the percentage absorbance increase suppression coefficient for formaldehyde solutions compared to the control group based on absorbance measurements in the presence of alamarBlue.*

| Incubation time, h | b value | p value | $IC_{50}$ | p value | 95% confidence level for $IC_{50}$ | | $R^2$ |
|---|---|---|---|---|---|---|---|
| 6 | -0.951 | 0.002 | 2.490 | <0.01 | 1.545 | 3.431 | 0.87 |
| 8 | -1.395 | 0.003 | 2.557 | <0.01 | 1.854 | 3.261 | 0.97 |
| 24 | -3.303 | 0.010 | 11.870 | <0.01 | 9.638 | 14.102 | 0.98 |

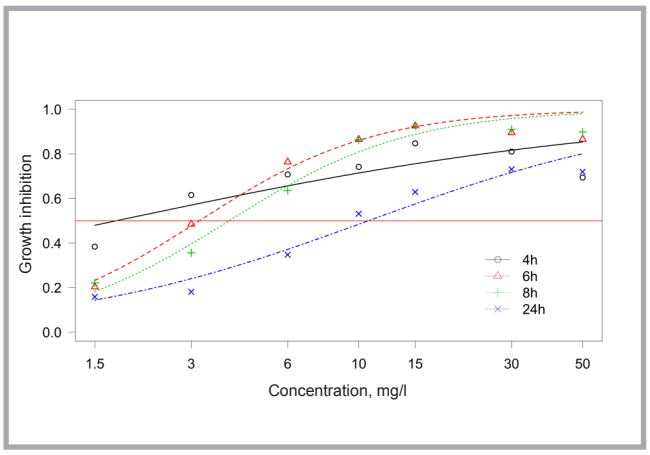


***Figure 4.*** *Effect of formaldehyde concentration on the suppression rate of bioindicator growth in relation to the incubation time based on direct measurements of absorbance.*

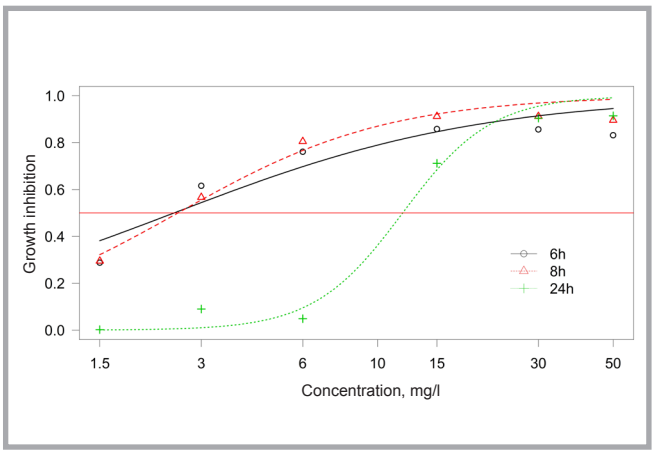


***Figure 5.*** *Effect of formaldehyde concentration on the suppression rate of bioindicator growth in relation to the incubation time based on absorbance measurements in the presence of alamarBlue®.*

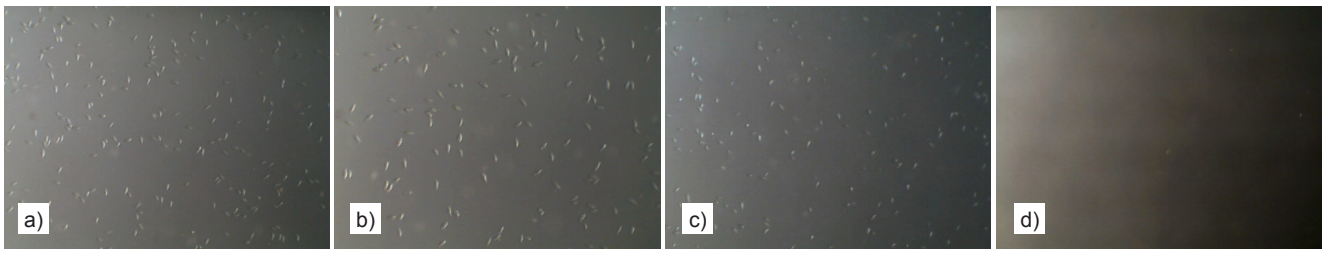


***Figure 6.*** *Photos of T. pyriformis culture incubated for 24 h in the following textile extracts: a) control group, b) sample No. 1, c) sample No. 2, d) sample No. 3.*

tor growth). The statistical environment R was used for data analysis. Graphical interpretation of the regression analysis is presented in ***Figures 4 & 5***, while the basic characteristics of approximated functions are listed in ***Tables 4 & 5*** (see page 99).

The $IC_{50}$, i.e. the toxicity index of the substance indicating a concentration that causes 50% suppression of bioindicator cell growth compared to the control group, was estimated based on the models proposed. Statistical analysis showed no statistically significant differences between $IC_{50}$ values estimated from logistic regression of data obtained using both methods. $IC_{50}$ is at the level of 2.5 – 3.9 mg/L for short-term exposure of test cells (6 – 8 h) to formaldehyde and 10.7 – 11.9 mg/L for longer exposure (24 h) of the bioindicator to formaldehyde.

## Analysis of textile safety by means of bioindicative method

When analysing the safety of textiles by means of the bioindicative method, a decrease in the proliferation of living cells incubated in the extracts and their death were observed (***Figure 6***).

The inhibition rate of *Tetrahymena pyriformis* culture growth in the extracts computed for both the turbidimetric and colorimetric measurements is presented in ***Table 6***.

Taking up the values of *T. pyriformis* culture growth inhibition in the extracts (***Table 6***), the formaldehyde concentration (*c in mg/L*), estimated on the basis of the transformed logistic function (***Equation 2***), an attempt at computing the formaldehyde content within textiles (*C*) was made (***Equation 3***):

$$c = IC_{50}\left(\frac{1-f(c)}{f(c)}\right)^{1/b} \quad \text{in mg/L} \quad (2)$$

where:

*b* - regression coefficient of approximated function,

$IC_{50}$ - quantitative measure that indicates the concentration of a substance that is needed to inhibit cell growth by half in comparison to the control group.

$$C = c \times V / m \quad \text{in mg/kg} \quad (3)$$

where:

*V* - extract capacity in ml,

*m* - sample mass in g,

c - approximated concentration of a substance in the extract in mg/L, computed on the basis of the inhibition rate of bioindicator growth, in accordance with ***Equation 2***, resulting from the approximation of the logistic function:

Depending on the measuring method (turbidymetric or colorimetric) and du-

***Table 6.*** *Effect of the textile extracts on the inhibition rate of Tetrahymena sp. culture growth computed from turbidimetric and colorimetric measurements.*

| Sample | Inhibition of *T. pyriformis* growth in relation to the incubation time, % | | | |
|---|---|---|---|---|
| | Turbidimetric measurements | | Colorimetric measurements in the presence of alamarBlue® assay | |
| | Incubation time, h | | | |
| | 6 | 24 | 6 | 24 |
| No. 1 | 38.5 | 18.3 | 48.5 | 1.85 |
| No. 2 | 67.1 | 38.2 | 79.8 | 12.1 |
| No. 3 | 100 | 100 | 100 | 100 |

ration of the experiment (the incubation time of *T. pyriformis* in the extracts), the formaldehyde content within the samples was computed as follows:

- 20.5 - 35.7 mg/kg within sample No. 1,
- 62.3 - 68.5 mg/kg within sample No. 2 and
- 2557 - 2987 mg/kg within sample No. 3.

Therefore, the values of the formaldehyde content within the samples estimated on the basis of the bioindicative method were close to those determined by the analytical method.

## Conclusions

The researches described above have show the high suitability of the bioindicative method for formaldehyde toxicity assessment. An analysis of the results indicates a high level of toxicity of this substance, even at very low concentrations (1.5 mg/L), and short time of its impact on test organisms. In light of the results above, it seems that current requirements related to the allowable formaldehyde content in textile products are at a quite low level. It is also worthy of emphasis that the methods used in the experiment are very sensitive compared to those described in subject literature [13, 17], and using the toxicity measuring and estimating techniques independently brings similar results.

The researches presented showed that with an increasing incubation time, the formaldehyde toxicity moves towards higher concentrations, probably resulting from the adaptive ability of *Tetrahymena pyriformis* to hard conditions. This observation seems to be very important for proper determination of the toxicity of a substance. Considering 24 hours as the standard bioindicator incubation time in a toxin, it is possible to determine a toxicity level exceeding the actual one. It seems that when carrying out bioindicative researches using simple organisms, it would be more proper to determine the time of the toxin impact on the bioindicator in which the lowest toxin concentration has a noticeable effect. The studies described above proved that the use of shorter periods of bioindicator incubation in formaldehyde (6-8 hours) enables the detection of even very small doses of this substance, which pose a hazard to living organisms.

Research on the toxicity of textile extracts by means of the bioindicative method based on *Tetrahymena pyriformis,* turbidymetric and colorimetric measurements, performed with samples containing formaldehyde within them, taken as an example, showed the high efficiency of this method in the safety assessment of textile products. These results give positive encouragement to further investigations on other substances undesirable in textile products, especially those which could be extracted from textiles by sweat (water).

## Editorial note

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