

## HYGIENIC INVESTIGATION OF THE TISZA-WATER, AND THE DRINKING-WATER MADE OF THAT, WITH THE MODIFIED DAPHNIA TEST

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### Abstract

The author carried out investigations, with Daphnia-test procedure, to form a judgment of the toxicity of the water of the Tisza and of the drinking-water, obtained from this. 54.4 per cent of the altogether 546 Tisza-water samples, investigated in the period from February 1974 till October 1979, were positive ( $Lc 50 < 48^h$ ). A close connection could be established between the values of water-level and Daphnia-positivity. The positivity belonging to the single water-level values is the greatest at  $\pm 100$  cm water-levels, close to 0-level: 62.1 per cent (average value) of the samples, investigated in this province. A rising water-level enables a decrease in the toxic material concentration, which supposedly moves at an approximately standing value. This had no more any harmful effect on Daphniae, at a determined cc.m/s, within a 48-hour exposition. Corresponding to this, according to our investigations, the water of the Tisza only ceased to be toxic to Daphniae in the days of great floods, above Szolnok at a high water of about 900 cm, which means an eightfold increase in the water output at 0-level. Positivity occurred not only when a micropolluter was present in a lethal concentration to Daphniae, but often in cases, as well, when the single micropolluters were present separately in sublethal quantities. The perdition of Daphniae may supposedly be explained with the synergetic effect, which is rather dominant in the interactions of the micropolluting materials.

It follows unambiguously from the results of the Daphnia-test investigations of drinking-water that the degree of the purifying efficiency of the town surface water-work must be further improved because 49.6 per cent of the altogether 546 samples proved to be positive.

### Introduction

In the special literature several data may be found, which more and more emphasize the importance of making inquiries in the interest of protecting the purity of waters and water life. Recently it became unquestionable that a number of the necessary answers are given by the investigations into toxicity (TARZWELL 1962, 1966). Though these investigations are mostly non specific, nevertheless they enable a qualitative evaluation in respect of toxicity.

Without striving after completeness, we are mentioning that more than one publication dealt with the problem of the biological investigation and qualification of the water of the Tisza (MEGYERI 1970, 1971, UHERKOVICH 1971), with the changes induced by the Kisköre river barrage (ÁDÁMOSI—BANCSI—HAMAR—KATONA—B. TÓTH—VÉGVÁRI 1974, CSUKA 1970). The presence, investigation or indication of the toxic and cumulated matters, getting into the Tisza, was not treated by any of these papers.

Since 1974, our laboratory has dealt with the indication of the toxicity of waters. On the first occasion in 1974, then following this, in 1976, we reported on our inves-

tigations in connection with this (PALICKA—CSÉPAI—HÓGYE 1974, 1976). We are convinced that the matters of foreign origin, which can be demonstrated from the water of the Tisza, endanger the living world of the Tisza and have an importance in the relation of public health, as well, that cannot be left out of consideration.

The disturbances emerging in connection with the drinking-water supply of the town Szolnok and environs are also first of all of qualitative character: the taste and smell complaints, lodged by the population frequently, are also to be brought into a close connection with the pollution of the Tisza and the not satisfying degree of clearing by the water-work built on the river.

These facts have justified the elaboration and routine application of a testing process, which — despite its simplicity — makes possible the systematic, fast, and informative estimation of the toxicity of the Tisza and of the drinking-water, made of its water. Earlier, we already discussed in detail the laboratory breeding and preserving of *Daphniae*, used as test-organisms for this purpose (CSÉPAI 1975). In the following we are reporting on our test procedure and the results of our investigation.

## Materials and Methods

From February 1974 till October 1979 we investigated, with a regularity of about every four days, into 546 samples, from drinking-water sterilized with chlorine similarly into 546 samples.

The sampling site of the Tisza-water samples is the Tisza above the surface water-work of the town Szolnok.

The sampling place of the drinking-water, sterilized with chlorine, is the Station of Public Hygiene and Epidemics of County Szolnok, the wall-tap of the central dining-hall, in Szolnok.

### (a) The sample preparation of the Tisza-water

From the filtered Tisza-water, 2×200 ml are parallel measured out into 250 ml tumblers then 2 times diluted samples were also made, similarly in a 2×200 ml total volume. The earlier described solution (CSÉPAI 1975) was used as diluent. Before putting in the test organisms, the undiluted samples and the twice diluted ones were placed into the semi-dark part of an indicated room, which is free from chemical vapours, where the temperature is set at about 20° C (±3° C) for 2 to 3 hours. Then we put in each vessel twelve *Daphniae magna* in the third grade of development, from our breeding stock and leave the samples without mixing and ventilating for 48 hours.

### (b) Preparation of the chlorinated drinking-water sample

In the drinking-water, the concentration of free chlorine and chloroamines, which endanger *Daphniae*, fluctuates — according to our investigations — within wide limits. The first important step is, to neutralize and reliably inactivate these.

For this purpose Na-thiosulphate was applied. It may be considered as a lucky circumstance that sodium thiosulphate does not mean any danger to *Daphniae* from physiological point of view. Thus, in the routine work we could avoid to determine the total chlorine content before testing the drinking-water in order to establishing the quantity of sodium thiosulphate, necessary to inactivation.

The minimum safety value was determined so that the maximum value was selected from the results of our total-chlorine investigations of drinking-water performed in the preceding five years and the quantity of sodium thiosulphate, equivalent to this, was added to the water samples in a model experiment. In these samples *Daphniae* did not perish. The additional experiments aimed at investigating into the consequences of a possible overdosage of thiosulphate. We have established that even a fiftyfold (!) quantity of the theoretically calculated thiosulphate has no harmful or destructive effect on *Daphniae*. For security reasons, however, the quintuple of the absolutely necessary value has been applied and this is given in ml/l in the practical work. It follows, therefore, from the foregoing that 0.5 ml Na-thiosulphate/litre drinking-water, sterilized with chlorine, proved to be reliable for the "overensuring" physiological neutralization of the total chlorine content, which has so far been highest, according to our measuring.

The 1 litre drinking-water, prepared in this way, is left in room-temperature for a few hours. Further on, we have acted as described in the above point.

### (c) Control water sample

For control and diluting water sample, we use drinking-water, perfused through an active carbon column (CSÉPAI 1975), controlling with it the faultless quality of the Daphnia material and the diluting water. In 200 ml of this, we put, like before, 12 Daphniae.

It is to be noted concerning all the samples that, during the investigation, the test organisms do not get any food. If Daphniae perish in the control vessel, the test cannot be appreciated.

### (d) Appreciation of tests

The evaluation of tests takes place after 48 hours, resp. within this, when — if possible — we follow with attention and record the changes in the akinesia of Daphniae. We had to decide in the formal relations of giving the results. It turned out from the special literature at our disposal that in the water toxicological works no uniform terminology has taken shape, as yet. In selecting from possibilities, we have primarily relied on Sprague's comprehensive critical review (1975) and on the French Water-biological STANDARD (1974). On this basis, we identify the expression "medium lethal level" with the symbol LC 50, completing this designation by indicating the length of exposition, because time and concentration are inseparably connected in case of the tests carried out on water organisms. We have set up as a criterion of the water acceptable from hygienic point of view that the water of the Tisza, resp. the drinking-water samples, may not contain any higher value than that of the concentration of a toxic matter, corresponding to a 48-hour long medium survival, because if they do contain that then:  $LC\ 50 < 48^h$  and we regard the sample as positive. In order to study the effect of the diluting water, we set always in double-diluted samples, as well.

Taking into consideration the facts mentioned above, we have summarized the method of giving the results of Daphnia tests in the following:

In case of a negative result (medium survival):

- 1) The undiluted water sample is negative after  $LC > 50\ 48^h$ .
- 2) The water sample of double dilution is negative after  $LC > 48^h$ .

In case of a positive result:

- 1) The undiluted water sample is positive after LC 50 (e.g.)  $32^h$ .
- 2) (a) The water sample of double dilution is positive after LC 50 (e.g.)  $32^h$ .  
(b) The water sample of double dilution is positive after  $LC < 50\ 48^h$ .

## Results

The results of the Daphnia tests of the Tisza water and the chlorinated drinking-water, in the period between 1974—1979, are summarized in Table 1. It turns out of this, besides other data, that 52.4 per cent of the undiluted Tisza-water samples and 49.6 per cent of the undiluted drinking-water samples proved to be positive.

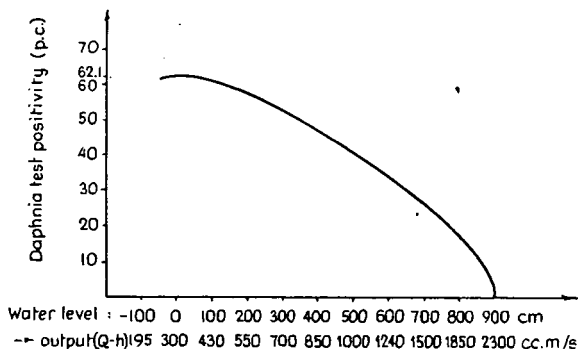


Fig. 1. The percentage distribution of 286 positive Tisza-water samples, plotted against the changes in water levels and water outputs, between February 1974 and October 1979.

Figure 1 displays a diagram about the connection between the percentile distribution of the positive Tisza-water samples and the change in water level and water output. We have indicated on the abscissa the water levels of the Tisza and the values of the Q-h curve belonging to these, expressed in cc.m/s; and on the ordinate the positive sample number compared with the total sample number of the single water-output domains, expressed in percentage. It may be read off from the Figure that 62.1 per cent (averaged value) of the contested samples fall on the water-level values close to the 0-level  $\pm 100$  cm. The gradual increase in the water output induces a decrease in the percentage of the positive water samples. Owing to the higher and higher and dilution, every sample becomes negative at last, in case of 2300 cc.m/s (+900 cm) water output, which is about eightfold of the value belonging to the 0-level.

### Discussion

Despite the high sensitivity of Daphinae to toxic materials, it is true that e.g. the fishes in the Tisza have not yet given, luckily, any immediate answer. But with the full knowledge of the data of the special literature, this establishment does not seem to be reassuring. The danger of the so-called chronic intoxications does exist. It is rightly urged that, for clearing up its causes, the sublethal action-mechanisms ought to be studied. It is said, with good reason, that the fish population is damaged by the micropolluters of lasting effect much more seriously and for a longer period than by cases of an acute poisoning.

It would be a fault to leave the possibility of this out of consideration — though without exaggerating it. In the Tisza — according to our (PALICKA—CSÉPAI—HÓGYE 1976) chemical investigations — the following micropolluters may be found, in a fluctuating concentration:

Permanently:

- Phenol (Phenol, 4-chlorophenol,  $\alpha$ -naphthol)
- Lindan,
- Detergent (anionactive),
- Oil,
- Metal-ions (Ti, Cr, Ni, Cd, Bi, Pb)

In exceptional cases:

Zn, Hg.

Some of these can get through the filtering units of the water-work into the drinking-water:

- Phenol-derivatives,
- Lindan,
- Oil,
- Metal-ions.

Though the materials, which are harmful to the above-listed living organisms, are separately at a sub-lethal level, we must conclude from the frequent positive test results some additive, and even synergetic effects, though opposite to all these, some antagonistic effects can also occur. This extremely complex problem is further

complicated by the changes in the concentration and combination of the enumerated matters, together with the rhapsodical changes in the water level of the Tisza. It is doubtless, too, that in the formation of the toxic effect, some modifying factors — like the different abiotic conditions, e.g., temperature, mineral-matter content, etc.) — have also some part.

Returning to the results of our investigation, it is to be noted, that — for reaching a harmless concentration — the samples of crude water, which were on two occasions prominently positive (LC 50 3<sup>b</sup>), were to be diluted fivefold. But the dilution of the Tisza due to flood must be of much greater degree, for creating a harmless concentration under natural conditions. The cause of this may be that newer chemicals are washed in by the river from the inundated areas and thus the level of the micropolluters being always present at normal water level, which level fluctuates around a constant concentration value, rises.

Table 1. *Tisza-water and drinking-water samples.*  
Results of *Daphnia* test between 1974—1979

Water-type	Investigated sample together	Positive samples (LC 50 < 48 <sup>b</sup> )			
		without dilution		twofold diluted	
		no.	p.c.	no.	p.c.
Tisza-water	546	286	52.4	217	39.7
Drinking water	546	271	49.6	195	35.7

The perishment of *Daphniae* only terminates if the rise of the water level becomes extremely high. On this occasion, the toxic-matter content of the Tisza water falls much more under the critical threshold, the water quality vies with the control (Fig. 1).

The results of the drinking-water investigations are showing a comparatively more favourable picture than those of the Tisza-water. From the parallel performed investigations it turns out, as well, that the changes measured with *Daphnia* tests in the concentration of micropolluters in the drinking-water are not significant of the connections between the water level of the Tisza and the toxic material content, showing a certain regularity. These facts can be explained with that the oxidative and adsorptive capacity of the water-work is incalculable, accidental, and remains under the demands. As a result of this, the degree of efficiency of the clearing capacity is changing, its regulation is, in respect of the chemical polluters, not duly solved, at present.

It is shown by our above investigation series and the experiences of our connected work that the water-toxicologic investigational principles, which are valid in the non-sanitarian field, can reliably be applied — with some modifications — in the hygienic evaluation of the surface waters and the drinking-waters, obtained from these. We shall continue our *Daphnia*-test investigations, apart from introducing other biological tests, and hope to be able to contribute with our work to the satisfying functioning of the modern surface water work, the building of which is in the process of completion.

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### Tisza-víz és ebből nyert ivóvíz higiénés vizsgálata módosított Daphnia teszttel

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#### Kivonat

Szerző módosított *Daphnia* teszt eljárással vizsgálatokat végzett a Tisza-víz és az ebből nyert ivóvíz toxicitásának megítéléséhez. Az 1974—79 közötti időszakban összesen 546 Tiszavíz-minta vizsgálata szerint 52,4%-a volt pozitív (LC 50 < 48<sup>h</sup>). Ennek megfelelően a Tisza vizének toxicitása a *Daphniákra* csak nagy árvizek idején szűnik meg. A *Daphniák* pusztulása feltehetően a mikro-szennyező anyagok kölcsönhatásaiban inkább domináló színenergetikus hatással magyarázható. Az ivóvíz *Daphnia*-teszt vizsgálatai alapján kitűnt, hogy a Szolnok városi vízmű tisztítási hatásfokát tovább kell javítani, mert a megvizsgált minták 49,6%-a pozitívnak bizonyult.

# ГИГИЕНИЧЕСКИЙ АНАЛИЗ ВОДЫ РЕКИ ТИСЫ И ПОЛУЧЕННОЙ ИЗ НЕЁ ПИТЬЕВОЙ ВОДЫ С ПОМОЩЬЮ ВИДОИЗМЕНЁННОГО ТЕСТА *Daphnia*

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Сольнок

## Резюме

Автор производил исследования по определению токсикации воды р. Тисы и полученной из неё питьевой воды с помощью видоизменённого теста *Daphnia*. В период с 1974 по 1979 гг. было проведено 546 анализов, 52,4% которых оказались положительными (LC 50 < 48). В соответствии с этим, токсикация воды Тисы не наблюдалась только в период больших наводнений. Гибель *Daphnia* объясняется, вероятно, доминирующим во взаимовлиянии микрозагрязняющих материалов числом энергетическим влиянием.

На основе анализов питьевой воды с помощью теста *Daphnia* видно, что необходимо дальнейшее улучшение эффективности гидростанции Сольнока, так как исследованные образцы на 49,6% оказались положительными.

## Ispitivanje sanitarnih svojstava vode reke Tise i vode za piće dobijene iz reke modifikovanim *Daphnia* testom

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## Abstrakt

Autor je pri ispitivanju koristio modifikovan *Daphnia* test za određivanje toksikološkog dejstva vode Tise kao i vode za piće dobijene iz reke. Od ukupno 546 analiziranih uzoraka vode Tise u periodu 1974—79. 52,4% je bilo pozitivno (LC 50 < 4<sup>h</sup>). Shodno tome toksičnost vode Tise na *Daphnia* prestaje samo za vreme velikih poplava. Može se pretpostaviti tumačenje da do uginuća *Daphnia* dolazi pre svega usled uzajamnih dejstava dominantnih sinergetičnih mikrozagadivača.

Ispitivanjem uzoraka vode za piće *Daphnia* testom utvrđen je pozitivan nalaz u 49,6% analiziranih uzoraka. Na osnovu toga proizilazi potreba daljeg poboljšavanja stepena prečišćavanja gradskog vodovoda Szolnok-a.