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Section II
– MEDICAL ISSUES -

25 th September, 2003 **11^{00} - 13^{00}**

25 th September, 2003 **15^{00} - 19^{00}**

"ALEXANDRU SIMIONESCU" CITY HOSPITAL'S AMPHITHEATHER - HUNEDOARA

Mircea Ioan LAZAR – President & Chairman
Alexandra ENACHE – Member
Jecu AVRAM - Member

	Name of the Authors	Title of Presented Papers
0201.	*Brigitha VLAICU, Cristina PETRESCU, Salomeia PUTNOKY, Corneluţa Fira-MLĂDINESCU – ROMANIA*	*Adolescence and a High Risk Behaviour: Drugs*
0202.	*Krisztina MÁGORI, Beáta HAVASI, Anita TÓTH, L. KISS - HUNGARY*	*Fatal Suicide Cases from 1991 to 2000 in Szeged, Hungary*
0203.	*Corina SEIMAN, R. NUŢIU, C. CIUBOTARIU, D. CIUBOTARIU, D. DUDA – ROMANIA*	*A Qsar Study Upon Thymine Derivatives with Anti-Hiv Activity*
0204.	*SZABÓ, A., PAPP, A., NAGYMAJTÉNYI, L. - HUNGARY*	*Functional Neurotoxic Effects in Rats Elicited by 3-Nitropropionic Acid in Acute and Subacute Administration*

0205. *PECZE, L., PAPP, A.* - *HUNGARY* — *Neurotoxicity of Lead and Mercury in Acute Exposure*

0206. *Cristina PETRESCU, Gh. MOISE, Sorina DOROFTEI, Brigitha VLAICU* – *ROMANIA* — *Teenagers Eating Behaviour - A Comparative Study Between Two Schools in Arad and Turnu Severin, Romania*

0207. *Alexandra ENACHE, Florin ENACHE, Gelu CĂDARIU, F. CHATSINIKOLAOU* – *ROMANIA* — *Household Cadmium Poisoning*

0208. *C. JIANU, Adelina ADĂMESCU* – *ROMANIA* — *Study of Synthesis and Characterisation Possibilities of B-2 Ethylhexyl (EH) Polyethylenoxy ($\bar{n}$ = 0-20) Propionic Acid Amides*

0209. *V. HERMAN, C. PASCU, L. COSTINAR* – *ROMANIA* — *The Dynamics of the Veterinarian Personnel in Romania, Hungary, Serbia and Montenegro in 1997-2001 Periods*

0210. *Biljana F. ABRAMOVIĆ, Vesna B. ANDERLUH, Anđelka S. TOPALOV, Ferenc F. GAÁL* - *SERBIA & MONTENEGRO* — *Direct Photolysis of 2-amino-5-chloropyridine*

0211. *M. KATA,* - *HUNGARY* *Á. GYÉRESI* – *ROMANIA* — *Trilateral Cooperation in the Fields of University Activities*

0212. *M. MATAVULJ, V. RAJKOVIĆ, B. LAŽETIĆ* - *SERBIA & MONTENEGRO* — *Effect of Extremely Low Frequency Electromagnetic Field on Blood Vessels of Adenohipohysis in Rat*

0213. *J. CSANÁDI, A. JÁVOR, J. FENYVESSY, G. SZABÓ, F. ESZES, I. BAJÚSZ* - *HUNGARY* — *Changes In The D-Amino Acid Content Of Sheep Milk Related Technologies*

0214. F. CĂDARIU, I. O. AVRAM, Al. ENACHE, G. CĂDARIU, F. ENACHE, M. MURARIU, A. GLĂVAN, S. GRAURE – ROMANIA — *The Frequency of Varicose Disease in Specialized Ambulatory Unit and Family Doctors Practice*

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0219. AVRAM J., MANCIU S., AVRAM I.O., GRAURE S., GLĂVAN A., MURARIU M., RÂMNEANŢU D. – ROMANIA — *The Natural History and the Evolution of the Treated and Not Treated Varicose Disease*

0220. NAGYMAJTÉNYI, L. - HUNGARY — *Pesticides and Health - Abstract*

0221. Gyula SZABÓ - HUNGARY — *Pathophysiology of Alcoholism - Abstract*

0222. *VEZÉR, T., PAPP, A. NAGYMAJTÉNYI, L.* - *HUNGARY* — *Ill Effects of Inorganic Metal Pollutants*

0223. *Emil TÎRZIU* – *ROMANIA* — *The Stimulation of Immune Reactivity in Poultry as Consequence to the Administration of Probiotics*

ADOLESCENCE AND A HIGH RISK BEHAVIOUR: DRUGS

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Abstract

Adolescence is defined by psychical fragility and exposure, most frequently voluntar, to terribilism and excesses. The relative psychological balance requires a rational knowledge of exogenous factors involved in the harmonious development of the teenager's personality. The present paper focusses on some aspects of drug consumption in teenagers from 2 cities in the South-West of Romania. The Western border is an entrance and exit gate for drugs which cross this part of Europe. We instrumented an individual anonimus questionaire in high-school teenagers in four classes. A 25% (28) of the 17-18 year old pupils had already consumed drugs once or twice, 2.7% (3) 3-10 times, 1.8% (2) over 10 times. The age of the first drug intake is between 16-18 years. The used drugs are canabis, psychotropical drugs associated with alcohol, ecstasy. In the peer groups of the teenager, 20% (23) of the frends and 26% (29) of aquaintances offer the example of drug consumption. Health promotion programs must take into account the main information source indicated by interviewed subjects: mass media.

Keywords:

adolescence, psychical lability, drugs, health promotion

1. Introduction

Studies on drug consumption in teenagers from Romania (NP 8) show an increase of the number of consumers over the last years and with age. Toxicophiliacs create a frendly circle where the consumption is spread and a trend for abandoning the family and quitting school emerges. Due to the fact that drug consumption is illegal, users are involved in a network of delinquency and potential violence. Many youngsters use prostitution, crimes in order to aquire drugs.

Populational studies which offer information on drug consumption in teenagers are necessary for designing programs of health promotion in schools. The research which is the basis of the present paper is included in this context.

2. Methodology

The descriptive epidemiological inquest based on anonimous questionaires included 112 teenagers aged between 17 and 18 years from 2 high school classes in Timişoara and Arad, respectively. The age groups and the two genders had a balanced repartition in this case study. The individual results were statistically processed.

3. Results and Discussions

- The perception of the self-image in teenagers and causes that may lead to drugconsumption (Table 1, 2).

Table 1. Perception of the self-image

How would you describe yourself?	Number of cases	Prevalence %
good realtions with the environment	35	31.3
happy	15	13.4
nervous	4	3.6
depressed	2	1.8
agressive	7	6.3
tired	9	8.0
euphorical	1	0.9
sleepless	26	23.2

The positive perception of the self-image is present in 56.3% (63) of the teenagers, only 6.3% higher than the negative perception. The most important causes of self-discontency are insufficient sleep, tiredness, aggressively, nervosity.

Curiosity is the most frequently involved reason for drug consumption: 42.9% (48). It is followed, in decreasing order of prevalence, by the need of new experiences and even the offer of drugs in 17.8% (20), then peer group example in 13.4% (15).

Table 2. Motivations leading to drug consumption

What is the reason for drug consumption?	Number of cases	Prevalence %
curiosity	48	42.9
need for new experiences	20	17.8
the offer of drugs	20	17.8
peer group example	15	13.4
fear of responsability	6	5.4
family conflicts	3	2.7

- Family climate and drug consumption in the family
- The optimal family climate offered by an organised family is mentioned in 83.9% of cases (94).
- Harmonious family relationships are predominant in 62.5% (70) of the teenagers. In the same time, tensioned or indiferent relationships are frequently indicated in 26.8% (30) and 10.7% (12) of cases, respectively.
- In the families of interviewed teenagers, drug consumption among brothers is present in 2 cases (1.8%).
- Extrafamilal groups and drug consumptions
- A percent of 26 (29 cases) of teenagers declare that they know persons who consume drugs
- In peer groups, 20% (23) consume drugs
- Sources of information about drugs (Table 3)

Table 3. Sources of information about drugs

Where did you first find out about drugs?	Number of cases	Prevalence %
movies	60	53.6
TV, radio shows	19	17.0
friends	12	10.7
News-papers, journals	11	9.8
Health education classes	10	8.9
family	2	1.8

Sources of information represented by movies, TV and radio shows, journals and news-papers represent 80.4% (90). Health education classes in schools offer information in 8.9% (10) of cases. The role of the family in the information about high risk behaviour of drug consumption is dangerously low.

- Peculiarities of drug consumption in teenagers
- The reality of drug consumption (Table 4)

Table 4. Drug consumption in teenagers aged between 17 and 18 years

Do you consume drugs? How many times have you been under the influence?	Number of cases	Prevalence %
never	79	70.5
Yes, once	6	5.4
Yes, twice	22	19.6
Yes, 3-10 times	3	2.7
Yes, more than 10 times	2	1.8

Non-consumers are predominant – 70.5% (79).

Out of the 29.5% teenagers who admit consumption, only 5.4% (6) used a drug only once. Most of them admit two occasions in which they consumed drugs, 19.6% (22); 3-10 occasions, 27% (3) and over 10 times 1.8% (2).

- Used drugs (Table 5).

Table 5. Types of drugs consumed

Consumed drugs	Number of cases	Prevalence %
Only canabis	15	45.5
Only medicines with alcohol	7	21.2
Only ecstasy	3	9.1
Canabis and medicines with alcohol	5	15.2
Canabis and ecstasy	3	9.1

A percent of 75.8 (25 cases) consumed only one type of drug. In decreasing order of prevalence, the list of drugs is: canabis, with a high prevalence as compared to the other types – 45.5%; medicines with alcohol, 21.2% (7); ecstasy, 9.1% (3).

Experimenting more drugs is present in 8 cases, 24.3%; more frequently, canabis and medicines with alcohol, less frequently, canabis and ecstasy.

- The first used drug (Table 6)

On the first place is canabis, as the first used drug, 69.7% (23). On the second place are the combination of medicines and alcohol, and on the third place ecstasy.

Table 6. First used drug

First used drug	Number of cases	Prevalence %
canabis	23	69.7
Medicines with alcohol	7	21.2
ecstasy	3	9.1

Taking into account statistical data from other countries, our study also points out the more frequent use of canabis and its "quality" of first used drug. Although there is no such a rule, canabis is considered to be a drug, which opens the way to other drugs.

- The administration route is smoking of canabis cigarettes, 69.7% (23), and the oral administration for the other used drugs.
- The age of the first consumption is situated between 16 and 18 years, more than half of the consumers – 54.5% (18) - declaring the age of 17.
- The answers to questions about the way of acquisition show that school surroundings and even schools are used by drug dealers in order to offer drugs for consumption in 97.0% of the situations. Consumer friends are also an important source, being also dealers: 57.6% (19) of the cases of consumption.
- The opinions of teenagers about the effects of drug consumption on human health (Table 7)

Table 7. Effects of drug consumption indicated by teenagers

Which of the effects of drugs do you consider to be more important?	Number of cases	Prevalence, %
They give you self confidence.	21	18.8
They comfort you.	2	1.8
They cause addiction.	60	53.6
They induce aggressive behaviour	17	15.2
They lead to antisocial acts	12	10.7

4. Conclusions

- Drug consumption is a reality in the interviewed teenagers. Most of the consumers used drugs at least 2 times. The first consumed drug and also the most frequently used is canabis. Thus, literature data which state that canabis is a possible intermediate stage on the road to a stronger drug: heroine, cocaine, are confirmed.
- Acquisition of drugs especially in the surroundings of schools and even in schools is one of the causes of drug consumption
- The drug offer must be in the attention of authorities. Detection of dealers and consumers networks and respecting the legislation are imperative.
- Informations on drugs rarely received in school or in the family, together with the important informative role of the media are major medical, educational, psychological and social problems of the Romanian society.
- A positive aspect is represented by the level of information in teenagers with respect to the negative effects of drug consumption. Over 50% (60) mention addiction, this being in truth one of the first serious health consequences: psychical and physical addiction, tolerance. The level of information should be an important cathalist in choosing not to expriment this behaviour.

5. References

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FATAL SUICIDE CASES FROM 1991 TO 2000 IN SZEGED, HUNGARY

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ABSTRACT:

The authors analysed the fatal suicide cases occuring between 1991 and 2000 in the Department of Forensic Medicine, University of Szeged, Hungary.

There was a fall in the number of suicides both nationwide and more significantly in Csongrád county during the investigated period. The ratio of male to female remained unchanged. The mean age of the victims: in men reached its peak in 1996, while in women following an increase remained constant from 1996. Amongst the methods of commitment the violent ones became dominant (e.g. hanging). The proportion of alcoholic influenced victims is still high.

Keywords:
Suicide, Hungary, Szeged

1. INTRODUCTION

Both the number and ratio of suicidal attempts and completed suicidal cases are traditionally high in Hungary. The occurrence frequency in the early 1990s was continuously increasing and was the highest in the world for decades. Recently the Baltic States and Sri Lanka have reported higher occurrence frequency. Previous epidemiological tests indicated significant geographic differences within the country and there were differences in the distribution of suicidal cases in the capital city and the country. Since the change in the political regimes from 1990 the overall occurrence frequency figures have decreased by 20%, although, no overall epidemiological test has been performed to investigate the current situation.

In this paper we wish to analyze the changes occurring in 1991-2000 on the basis of the fatal suicidal cases on which post mortem were carried out in the University of Szeged, Department of Forensic Medicine

2. MATERIAL AND METHOD

County Csongrád is situated in the south-eastern part of Hungary. Its population is about 460 thousand people. The seat of the County is in Szeged. The total population figure of Szeged on 1st January 2000 was 158.158 people. In accordance with the law all the victims who suffered violent deaths including those committing suicides undergo a post mortem and the material available in the University of Szeged, Department of Forensic Medicine includes all the fatal suicidal cases occurring in the county seat.

During the post mortem apart from macroscopic and microscopic tests blood alcohol and toxicological tests were carried out in each case where death occurred in the site of the suicide or within six hours after admission to hospital. The determination of blood and urine alcohol content was carried out with a head-space gas-chromatographic method. For the screening toxicological test the immunological test of the urine and the thin-layer chromatography test of the urine and the liver were carried out. The confirmation tests are usually performed in the National Forensic Toxicological Institute with GC/MS tests.

3. RESULTS

The number of suicides in County Csongrád corresponds to the national trend and shows an approximate decrease of 20% in the examined time period. Considering the annual trends it can be stated that the decrease stopped in about 1995/1996 and the number and ratio of fatal suicidal cases have been stagnating since then. The analyzed material is characteristic of the City of Szeged and represents approximately one-third of the deaths in the county. The decreasing tendency could also be detected in our material as well (Fig. 1).

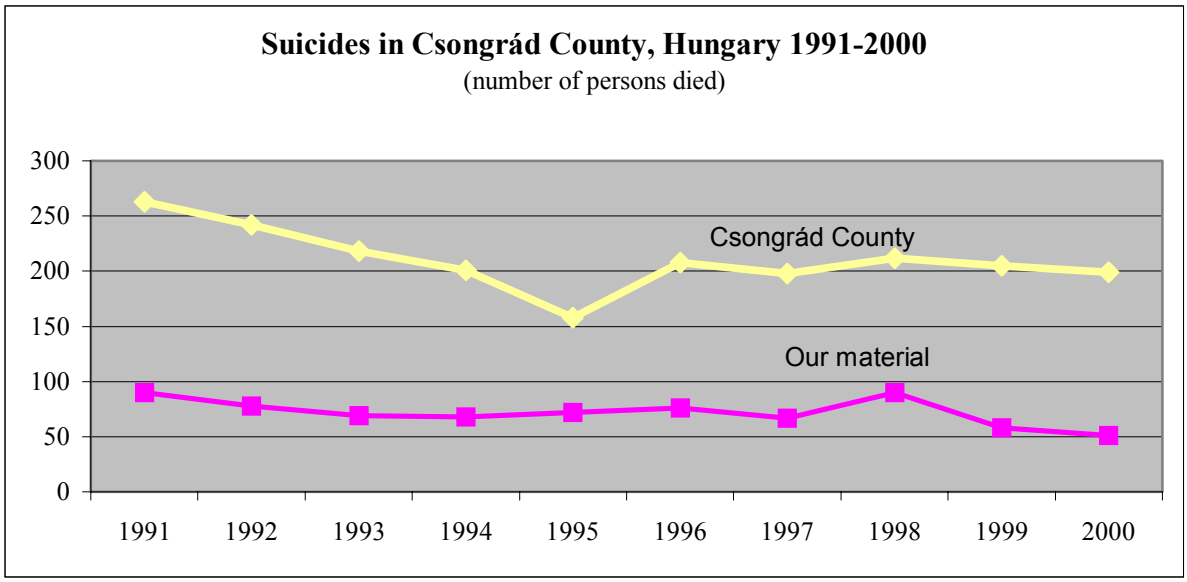

Fig. 1. *Suicide in Csongrád County*

From among 719 deaths there were 501 men and 218 women, and the female-male ratio was 2.5:1. In the examined period it showed the same distribution apart from a minimum variation. The age distribution differs significantly in the male and the female group. The greatest number of male suicide cases happened in the male age group of 41-50, whereas in the female age group it was only between 71 and 80. By standardizing the values, it can be stated that the occurrence frequency begins to rise at the age of 30-40 for males and the same trend can only be noticed after the age of 70 for females (Figure 2). The average male age is 51.12 years and the average female age is 58.29 years. Within the analyzed time period the female average age was gradually increasing, whereas the average male age showed an increase until 1996 only and started to decrease gradually afterwards (Figure 3).

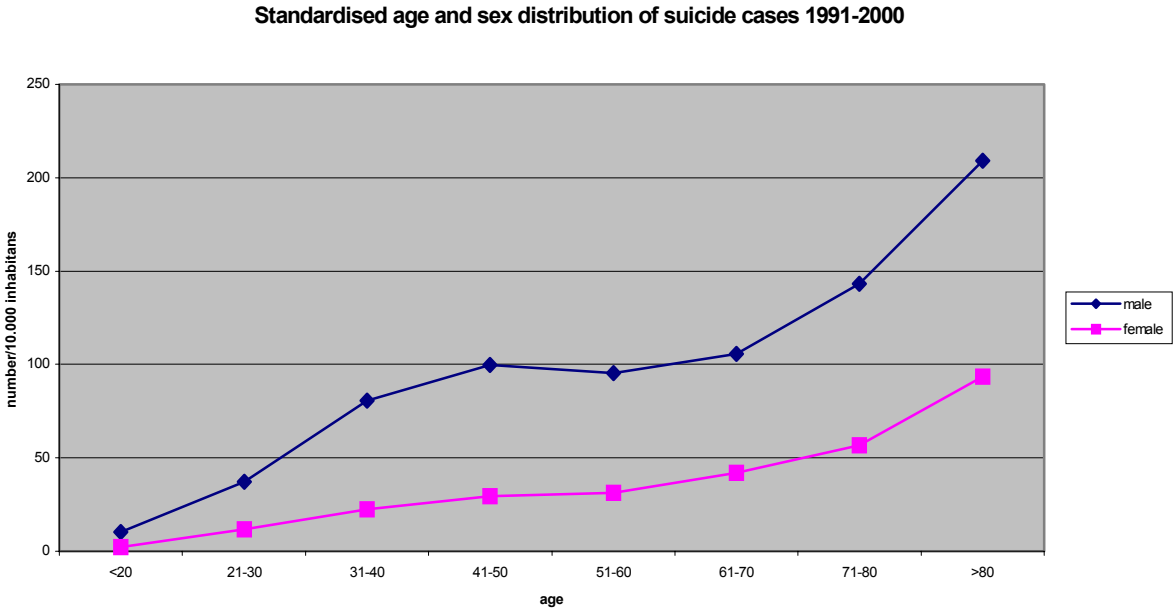

Fig. 2. *Age specific mortality*

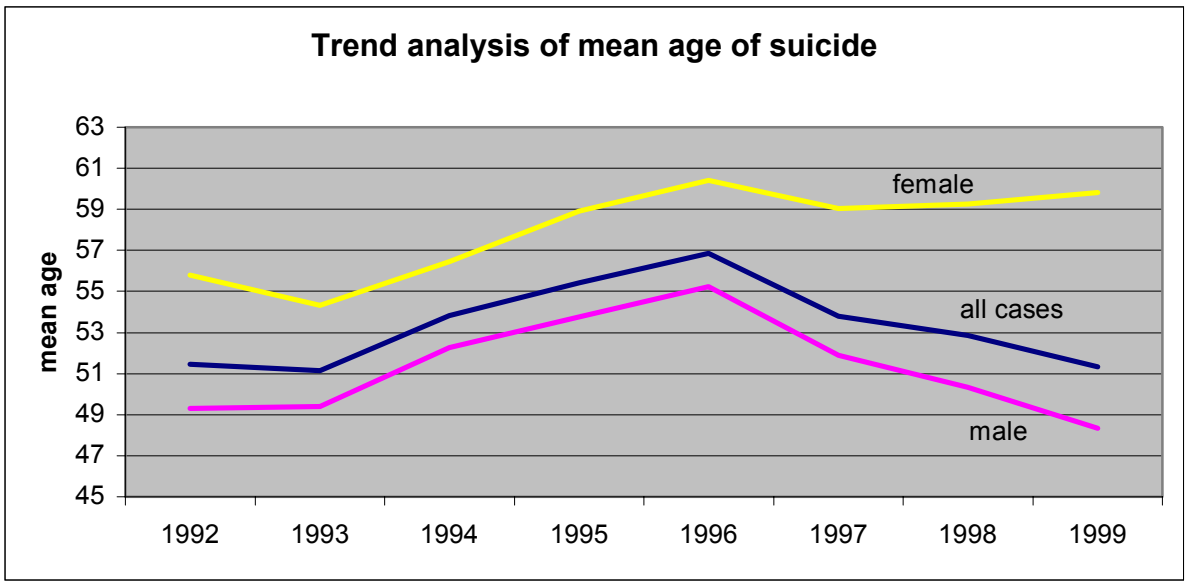

Fig. 3. *Trend analysis of mean age*

As to the methods of committing a suicide, the most frequent method is hanging in both the male and the female cases and the occurrence frequency gradually increased in the examined time period (44% in 1991 and 57% in 2000). The number and ratio of suicide cases committed with drugs show a slight but gradual decrease (22% in 1991 and 14% in 2000). The other methods are rare and their number and ratio did not change considerably in the examined time period. For men hanging and suicide with pesticides are the most frequent, whereas suicide with drugs is characteristic of females mainly.

In 38% (125 cases) of all the examined 474 cases alcohol could be detected in either the blood or the urine in the concentration of slight or average alcoholic impairment. Analysing the occurrence frequency of alcoholic impairment in the two sexes, it can be stated that 47% of all males and 16% of all females drank alcoholic drinks prior to the suicide (Figure 4). The occurrence and degree of alcoholic impairment is independent of the method of committing a suicide.

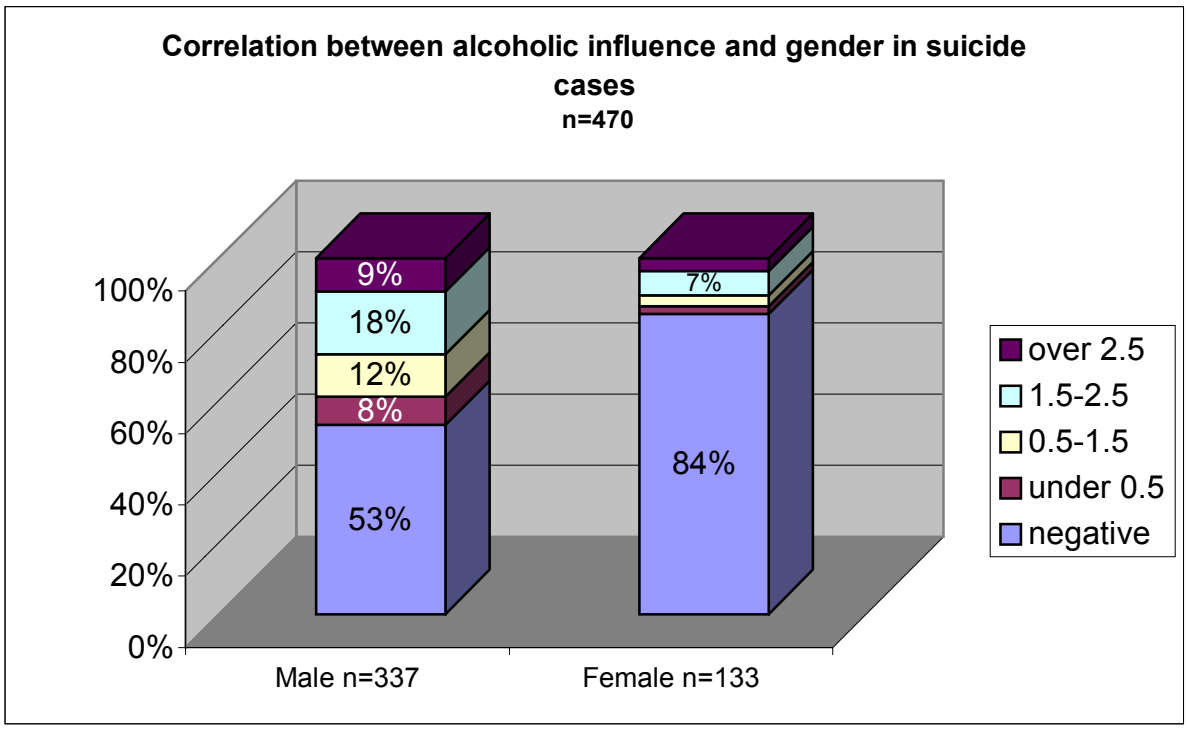

Fig. 4. *Alcoholic influence*

Table 1. *Licit drugs in not drug related deaths*

The presence of licit drugs in not drug related deaths

Drugs	No. of cases
Barbiturate	10
Benzodiazepine	51
Fenotiazine	1
Tricyclic antidepressants	6
benzodiazepine+fenotiazine	2
Glutetimid	1
Meprobamat	3
drugs of cardiovascular system	2
Total	76

In 635 of the non-drug-related suicidal cases toxicological tests were carried out, and in 76 cases (11%) the test results were positive with barbiturates (10 cases) and (51 cases) with benzodiazepines. The number of drugs that do not attack the central nervous system is negligible (Table 1).

4. DISCUSSION

The detailed analysis of the fatal suicidal cases in the population of the capital city in Hungary was carried out in 1972 and in the south-western part of Hungary in 1983-87. The epidemiological data indicate that the occurrence frequency is considerably higher in the south-eastern part of Hungary. This trend can be seen continuously in the past 50 years and basically corresponds to the occurrence frequency of chronic alcoholism. The male-female ratio is 3:1 within which however great territorial differences can be observed. During the earlier examinations 60% of all males and 18% of all females showed signs of pathological changes due to chronic alcoholism, whereas in 41% of non-drug-related suicide cases researchers detected the presence of tranquillizers or sleeping pills earlier.

The decrease in the occurrence frequency can be seen in the entire country since 1990 and can be found in both Csongrád County and our examined material. The occurrence frequency has stabilized nationwide since 1997 and in Csongrád County since 1996. In comparison to the previous years a shift can be seen in the age distribution. The difference between the average age of males and females was somewhat more than 7 years and it could be seen in the examined period continuously but with the decrease in the occurrence frequency the average age of male and female perpetrators gradually increased. An increasing tendency could be observed among the female perpetrators afterwards as well, whereas a decrease could be registered regarding the average age of male perpetrators. The standardized frequency values indicate that the number of cases increase considerably between 30 and 40 among men and after 70 among women.

The frequency of alcohol consumption before the suicide corresponds to the occurrence frequency of chronic alcoholism and is significantly higher among males. The occurrence of tranquillizers and sedatives in 11% of the non-drug-related suicide cases supports the likelihood of a previously existing depression but the occurrence frequency observed by our team is significantly lower than what was found elsewhere in the country.

In the analyzed period it is possible to say that on the basis of the age distribution, the alcoholic impairment and the occurrence of tranquillizers chronic alcoholism and existential problems were the motives for males, whereas for women mainly depression, loneliness and diseases played a role.

A certain redistribution can be seen with respect to the ways suicides are committed. In comparison to the previous decades carbon-monoxide poisoning has disappeared and drug-related suicide cases have significantly increased, as well as the brutal methods, including hanging, have also been on the increase. The reason for this cannot be explained on the basis of our analysis.

A QSAR STUDY UPON THYMINE DERIVATIVES WITH ANTI-HIV ACTIVITY

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ABSTRACT :

The basic goal in developing any chemotherapeutic agent is that it produces its desired effect without compromising host functions by unacceptable reverside effects. But, in viral infections the invader becomes a component of the host cell.

Herein we describe a QSAR study of a series of HEPT compunds, in which both the influence of substitution of the terminal hydroxy group (with and without 6-phenyl-6-thiopyridyl exchange) and the more extensive alteration of the N^1 side chain have been considered. We applied in this study the MTD method. and some other structural parameters, such as ClogP – the calculated octonal/water partition coefficients of the molecules under study.

KEYWORDS :

HIV, chemotherapic agents, HEPT, MTD method

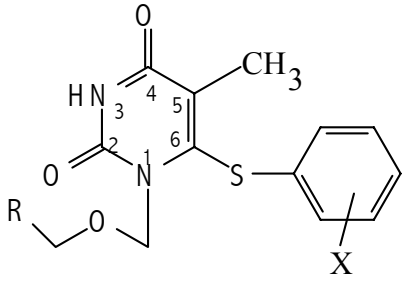
1. INTRODUCTION

Inhibition of reverse transcriptase (RT), the unique nucleic acid polymerizing enzyme – human immunodeficiency virus (HIV) – encoded polymerase which directs both RNA and DNA dependent DNA synthesis, preventing retrovirus replication, has proven to be one of the most effective ways to block the viral multiplication. There are several classes of compounds, which are specifically targeted at HIV-1 RT, from which the 1-[(2-hydroxyethoxy)methyl]-6-(phenylthio)thymine (HEPT) derivative family has been extensively investigated [1].

2. METHODS

The biological activity for a small series of N[1] side chain modified analogues of HEPT, expressed as IC_{50}, is presented in Table 1.

Table 1. *Anti- HIV-1 Activity Data of Several HEPT Derivatives (IC_{50}) and calculated hydrophobicity (Clog P)*



compd	X	R	IC_{50}(μM)[a]	A[c]	Clog P
1.	CH	NH–C_6H_5	0.055	7.26	3.62
2.	N	NH–C_6H_5	0.160	6.80	2.52
3.	N	S–Py[b]	0.440	6.36	2.28
4.	CH	Br	0.760	6.12	2.80
5.*	CH	S–Py[b]	0.830	6.08	3.38
6.*	N	S–C_6H_5	1.010	6.00	3.38
7.*	CH	I	2.200	5.66	3.19
8.	CH	N(C_6H_5)$_2$	8.750	5.06	5.89
9.	CH	NHCO(CH_2)$_2$Cl	11.400	4.94	1.93
10.	CH	N(CH_2CN)$_2$	27.500	4.56	1.44
11.	CH	NH_2	31.000	4.51	1.44

*data points not used in deriving eq. (2).

[a]Effective concentration of compound required to achieve 50% inhibition of HIV-1 multiplication in CEM-SS infected cells

[b]Py = 2-pyridyl

[c]A =log1/C (C=IC_{50})

3. MTD METHOD [2]

The minimal steric difference, MTD, is a measure for steric misfit between the molecules of a series of bioactive substances and the binding site of the biological receptor, which is represented by the hypermolecule, **H**. The hypermolecule is the result of the approximate (non-hydrogen) atom per atom superposition of the molecules i, i=1,N, in the investigated series.

The vertices j, j=1,M of the hypermolecule correspond to the positions of these atoms. Thus, the hypermolecule **H** can be considered as a topological network. If molecule "i" occupies vertex "j" from **H**, x_{ij}=1, while x_{ij}=0, if the vertex is not occupied. The minimal steric difference MTD_i of molecule "i" with respect to the receptor is calculated according to the formula (1):

$$MTD_i = S + \sum \varepsilon_j x_{ij} \quad (1)$$

with ε_j= –1, 0 or +1 for vertices attributed to the receptor cavity (beneficial), to the exterior (irrelevant) and to the receptor walls (detrimental), respectively; S is the total number of cavity vertices. Consequently, MTD_i is a measure of the steric misfit of the molecule "i" with respect to the receptor cavity and it is equal to the number of occupied wall vertices plus the number of unoccupied cavity vertices of **H** [3].

4. RESULTS AND DISCUSSIONS

The superposition procedure to construct the hypermolecule is based upon the maximal superposition of the compounds 2-11 upon the compound 1, which is the most active of this series. The entities to be superimposed are second or higher row atoms (S, Cl, Br, I).

The starting map for the MTD optimized procedure were obtained by inspection of vertices found preferentially in molecules with high and low inhibition activity, respectively. For all 11 compound from Table 1, it is:

$$S_t^o = \begin{cases} j(\varepsilon_j = -1): & 1-6 \\ j(\varepsilon_j = 0): & - \\ j(\varepsilon_j = +1): & 7-12 \end{cases} \qquad r = 0.695$$

The corresponding optimized receptor map, r being the correlation coefficient, and the correlation equation (2) are:

$$S_t^* = \begin{cases} j(\varepsilon_j = -1): & 4-7 \\ j(\varepsilon_j = 0): & 2,3,9,10 \\ j(\varepsilon_j = +1): & 1,8,11,12 \end{cases}$$

$$\hat{A} = 7.77(\pm 0.50) - 0{,}63(\pm 0.15)MTD \quad (2)$$
$$(n = 11, \quad r = 0.816)$$

***Table 2**. The results of the MTD method:*

i	$j(x_{ij}=1)$[a]	MTD_i* [b]	MTD_i* [c]
1.	1–6	2	3
2.	1–6	2	3
3.	1–6	2	3
4.	–	4	5
5.	1–6	2	–
6.	1–6	2	–
7.	–	4	–
8.	1–12	4	5
9.	1–4	4	5
10.	1,2,7,8	5	6
11.	–	4	5

[a]j – vertices occupied by molecule i in the hypermolecule **H**
[b]MTD – values corresponding to the S_t* optimized receptors map
[c]MTD – values corresponding to the S_p* optimized receptors map

For the smallest series, outliers are compounds 5-7 from Table 1 – the starting map S_p^o is:

$$S_p^o = \begin{cases} j(\varepsilon_j = -1): & 1-6 \\ j(\varepsilon_j = 0): & - \qquad\qquad r = 0.749 \\ j(\varepsilon_j = +1): & 7-12 \end{cases}$$

The optimized receptor map S_p* and the corresponding correlation equation (3) A vs. MTD* are the following:

$$S_p^* = \begin{cases} j(\varepsilon_j = -1): & 2{,}4-6{,}9 \\ j(\varepsilon_j = 0): & 7{,}10 \\ j(\varepsilon_j = +1): & 1{,}3{,}8{,}11{,}12 \end{cases}$$

$$\hat{A} = 9.15(\pm 0.80) - 0{,}79(\pm 0.18)MTD^* \qquad (3)$$
$$(n = 8, \quad r = 0.876)$$

By using the ClogP values estimated by Hansch et al [4] and the MTD* values corresponding to the two optimized maps, the following correlational equations (4) and (5) have been obtained:

$$\hat{A}=4.49(\pm 1.64) + 1.54(\pm 0.74)\ Clog\ P - 0.20(\pm 0.1)(Clog\ P)^2 - \qquad (4)$$
$$- 0.38(\pm 0.18)MTD^*$$

$$(n=11,\ r=0.891,\ s=0.49,\ F=9,\ p<0.008,\ q^2=0.706)$$

$$\hat{A}=2.74(\pm 0.96) + 4.09(\pm 1.16)ClogP \qquad (5)$$

6. CONCLUSIONS

The obtained correlative results are good. However, the number of analyzed compounds is not big enough and their structural variation restricted. This hindered the performance of a detailed QSAR analysis, which would have allowed the obtaining of more accurate information upon the nature of the atomic groups in R.

The hydrophob character of those increases the anti-HIV activity by means of growing the degree of enzymatic inhibition of RT, acting upon the allosteric hydrophobic site (situated in the neighbourhood of the catalytic site of the enzyme) which imposes steric compulsions, revealed by our QSAR analysis with the MTD method.

The phenylic cycle, rigide and sufficiently hydrophil, is the best substituent for the studied series; the presence within the substrate R of some heteroatoms which decrease the hydrophobicity has a decreasing effect upon the biological activity.

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FUNCTIONAL NEUROTOXIC EFFECTS IN RATS ELICITED BY 3-NITROPROPIONIC ACID IN ACUTE AND SUBACUTE ADMINISTRATION

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Abstract: *Biochemical and morphological alterations caused by 3-nitropropionic acid in the brain of experimental animals are well described. Changes possibly induced by 3-NP in electrophysiological functional characteristics of the central nervous system are less well known. In this study ten weeks old male Wistar were subacutely and acutely treated with 3-NP. For recording, the animals' left hemisphere was exposed in urethane anesthesia. Silver electrodes were placed on the cortical (sensory foci) and tungsten needles in the subcortical (caudatum, globus pallidus) recording sites. Spontaneous electrical activity and sensory (somatosensory, visual and auditory) evoked potentials were recorded. Following subacute treatment, changes were first of all in the slowest and fastest frequencies of the spontaneous activity. The change was different in the cortical vs. subcortical sites. In the sensory evoked potentials after subacute treatment, the most characteristic change was an increase of the latency, seen in all sensory areas. In the acutely treated animals, the amplitude of the somatosensory evoked potential decreased after giving 3-NP. With double stimuli, the relation of the two responses was treatment - and interval - dependent. It needs further studies to find the possible connections between the biochemical effects of 3-NP and the functional neurotoxical changes described above.*

KEY WORDS: *3-nitropropionic acid, cortical activity, subcortical activity, rat*

1. INTRODUCTION

The substance 3-nitropropionic acid (3-NP) is naturally found in *Astragalus* species (*Leguminosae)* [7]. Human intoxication may result from infestation of foodstuffs (sugar cane, cereals etc.) with moulds of the *Anthirium* and *Aspergillus* genus producing 3-NP. Human exposure to 3-NP, even in low doses, causes acute encephalopathy followed by dystonia [9].

The morphological and functional effects of 3-NP intoxication have been replicated in animal experiments [2]. Decrease of motor performance was seen [17] with degeneration of primarily the striatum but also the hippocampus and thalamus [1,10]. At the cellular level, 3-NP inhibits succinate dehydrogenase, a key enzyme of oxidative energy production [5] which effect develops fast and is not limited to the

sites of morphological damage [3]. Beyond that, 3-NP was found to act on NMDA receptors thereby inducing excitotoxicity [12].
It seems reasonable that the latter two effects of 3-NP are reflected in the electrical activity of the brain. The aim of this work was therefore to see to what extent the neurophysiological investigation system established in our laboratory is suitable to detect functional changes caused by 3-NP administration in rats.

2. METHODS

The effects of 3-NP were investigated in three different time schemes. For subacute experiments, ten weeks old male Wistar rats (10 in a group) received 10 (low dose) and 15 (high dose) mg/kg b.w. 3-NP ip. on 5 consecutive days and were kept for further 4 weeks before recording. Control animals were untreated. For acute exposure, the animals received 20 mg/kg 3-NP ip. and were kept for 24 hrs. To see immediate effects, the rats were first prepared for recording (see below) and 20 mg/kg 3-NP ip. was given to the prepared animal after a few control records. For recording, the animals were anaesthetized with urethane and were placed in a stereotaxic instrument. The left hemisphere was exposed and silver electrodes were placed on the primary somatosensory, visual and auditory areas. One steel needle electrode each was inserted in the caudato-putamen and the globus pallidus. Spontaneous electrical activity (electrocorticogram, ECoG) was recorded from these sites simultaneously for 6 min, and the relative spectral power of the frequency bands was determined.

Stimulus-evoked activity was then recorded via the surface electrodes. Somatosensory stimulation was done by a pair of needles inserted into the whiskery skin. Visual stimulation was performed by flashes (1 Hz, 60 lux) delivered by a flash generator via an optical fiber directly into the contralateral eye of the rat. For acoustic stimulation, clicks (1 Hz, 40 dB), were applied into the ear of the rat.

3. RESULTS

The most conspicuous effect in the ECoG was the increase in the delta activity seen in all cortical foci after subacute, acute and immediate 3-NP treatment (Fig. 1). In the theta and alpha bands, there was a decrease in all three treatment schemes. In the fast bands (beta2 and gamma), there was a mild increase in the subacutely treated animals and a massive decrease in the acutely treated ones. In the immediately treated rats, this alteration needed ca. an hour to appear. The difference between power spectra in the three cortical sites was insignificant. In the basal ganglia, however (recorded only in the animals with subacute treatment) the changes were opposite, i.e. the activity in the fastest bands was increasing and in the slow bands, decreasing.

Subacute treatment with 3-NP caused a general slow-down of the cortical activity, also reflected in the increased latency of the sensory evoked potentials (Fig. 2). The alteration of the duration of the responses was mild. In acute 3-NP exposure, there was no consequent effect on the latency but the duration of the potentials was increased. Double-pulse somatosensory stimulation with different inter-stimulus intervals was used to reveal any fatigue or dynamic interaction in the sensory system. The decrease in the amplitude of the second vs. first evoked potentials (Fig. 3, top) was hardly different in acutely 3-NP treated vs. control rats, but the latency increase was less in the treated animals. In immediate exposure, decrease of the

amplitude and increase of the latency appeared with a long delay. The second evoked potential seemed to be facilitated depending on the inter-stimulus interval (Fig. 3, middle and bottom).

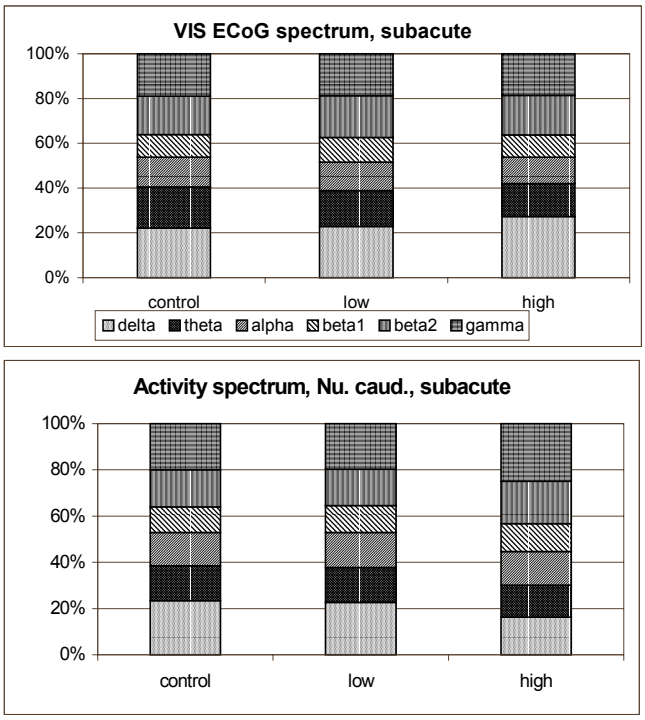


Fig. 1.
Changes of the spontaneous activity of the visual cortex (top) and the nu. caudatus (bottom) after 4 weeks exposure to 3-NP.

4. DISCUSSION

3-NP induced several alterations in the spontaneous and evoked cortical electrical activity of treated rats.

It is known that 3-NP causes energy insufficiency in the neurons by impeding mitochondrial oxidation [5]. A similar state can be induced by hypoxia. In human volunteers, breathing low-oxygen gas mixture caused an EEG shift to lower frequencies [18]. Also, in human cases of inherited or idiopathic mitchondrial dysfunction, like mitochondrial encephalomyopathy (ME), cortical functions were affected [11]. EEG abnormalities [15] and alteration of certain visual evoked potential components [6,13] were seen in ME patients. The increase of evoked potential duration was similar to what we have seen in the subacutely treated rats. In the EEG, the main abnormality of ME patients was slowed activity [14]. In the 3-NP treated rats, however, the low-frequency activity was decreased. The results obtained with double-pulse somatosensory stimulation reflect probably a kind of disinhibition, similary to what was found in the somatosensory and motor cortex of humans [8].
Another known effect of 3-NP, inhibition of glutamate uptake [16] may lead to imbalance between excitation and inhibition, producing the disinhibition mentioned above, and finally to excitotoxicity, in which long-term potentiation of NMDA-mediated excitation has probably a key role [4].

The overall effect of 3-NP on the cortical activity is complex, involving elements of depression and excitation. Further studies are needed to reveal which of the known effects of 3-NP is specifically responsible the observed effects, and how they are related to biochemical and/or histological alterations.

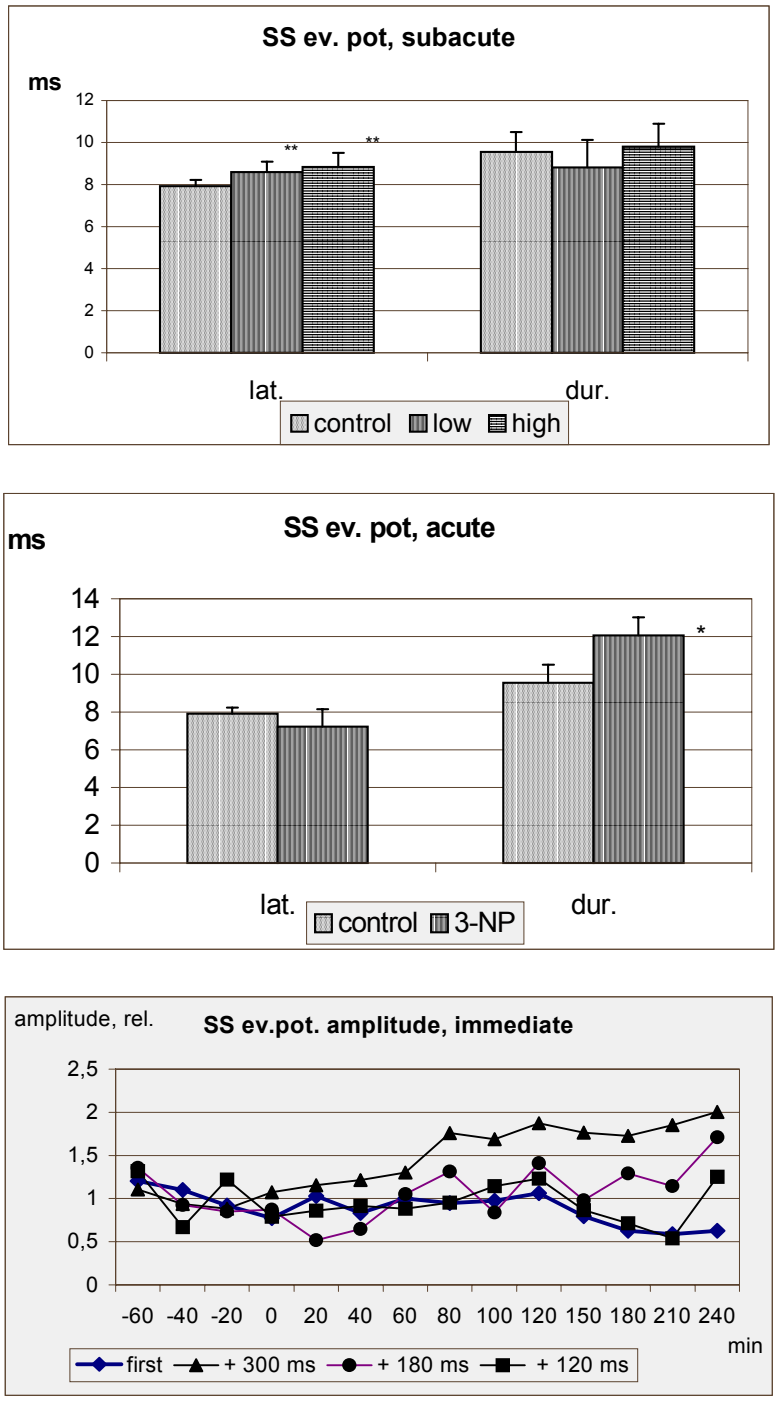
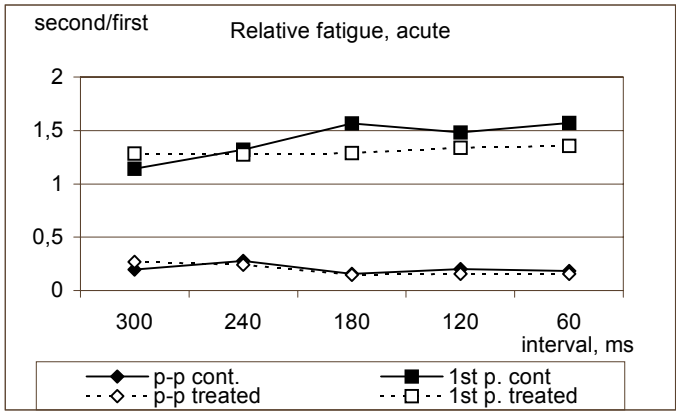
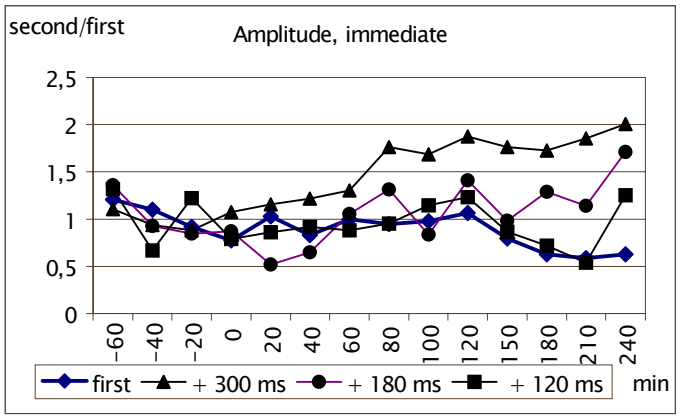
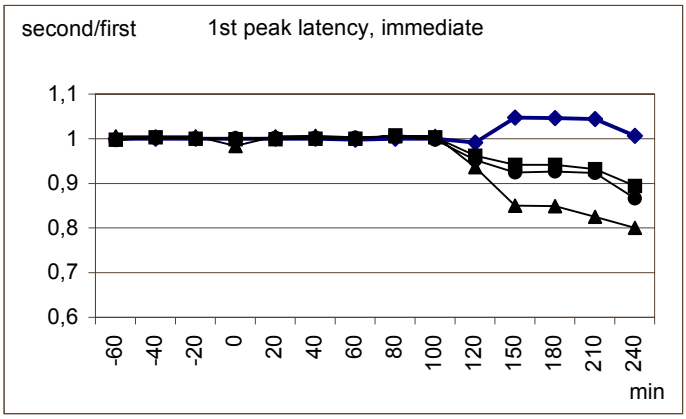


***Fig. 2**.*
Alteration of the somatosensory evoked potential in subacute and acute treatment (top, latency and duration) and in immediate effect (bottom, amplitude)







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NEUROTOXICITY OF LEAD AND MERCURY IN ACUTE EXPOSURE

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Abstract:

The use of heavy metals can lead to considerable emission and, hence, harmful levels at workplaces - affecting employees - and in the general environment - resulting in airborne, foodborne etc. population exposure. Lead and mercury represent an environmental health hazard including nervous system damages. Beyond human studies on exposed individuals, animal experimentation is required in order to elucidate toxic mechanisms and develop biomarkers for early detection of the adverse effects.

The aim of this study was to see the short-term effect of inorganic lead and mercury on the activity of the somatosensory system of rats. Weak electric shocks to the whiskers served as stimuli, and the evoked nervous activity was recorded from the cortical and subcortical focus of the brain. From the cortex, spontaneous activity was also recorded. Both lead and mercury caused alterations in the recorded stimulus-evoked and spontaneous bioelectric activity. These alterations probably reflect a specific action of the heavy metals on the nervous system so they have a potential use in human health protection.

Keywords:

lead, mercury, environment, neurotoxicity, rat

1. INTRODUCTION

A number of heavy metals are known to affect the activity of the nervous system of animals and humans, as indicated by the multitude of neurological signs following occupational exposure, or by the effects of airborne, foodborne etc. exposure of the population.

Lead has been used in large amounts in metal and inorganic forms (in batteries, piping, paints, solders etc), and tetraethyl lead was, and in a number of counties still is, used as a petrol additive. Lead in any form is accumulated in the central nervous system, first of all in the cortex and hippocampus [15]. Pb^{2+} interferes with Ca-dependent regulation of protein kinase C, calmodulin, ATPases, etc. due to the competition of with Ca^{2+} ions [3,27]. Partly due to the interference with Ca^{2+}, lead also affects several transmitter systems. GABA uptake was decreased and dopamine uptake increased in synaptosomes from lead-treated rat brains [16]. Alterations in the dopaminergic, cholinergic and glutamatergic control of behavior were observed in lead-treated animals [10]. In humans, alterations of various forms of central and peripheral evoked activity, like sensory evoked potentials and nerve conduction

velocity, were described in lead-exposed individuals [2,19]. In our earlier studies, similar changes were found in rats after up to 12 weeks oral exposure by Pb^{2+} [22].

Mercury is another heavy metals known to be harmful for the nervous system. In occupational exposure to inorganic mercury, alterations of the spontaneous [25] and stimulus-evoked [19] cortical electrical activity have been reported. Mercury in animal experiments affected a number of ion channels in the peripheral and central nervous system [29]. Hg^{2+} also interfered with calcium homeostasis, by disturbing Ca uptake to the endoplasmic reticulum [13]. In rats treated with ionic mercury, higher than normal levels of the transmitters noradrenaline [14] as well as dopamine and serotonin [18] were seen. In vitro ligand binding of rat cortical muscarinic receptors also was negatively affected by Hg^{2+} [8]. In earlier studies of our group on mercury effects on the cortical activity, rats receiving subchronic $HgCl_2$ treatment showed alterations in the spontaneous [11] and stimulus evoked [28] cortical activity.

The aim of the present study was to see the short-term effect of inorganic lead and mercury on the activity of the somatosensory system of rats.

2. METHODS

Adult male Wistar rats of ca. 350 g b.w. were used in the experiments. After urethane anaesthesia (1000 mg/kg b.w., ip.) the animals' head was fixed in a stereotaxic frame and the left hemisphere was exposed. Wounds were sprayed with 10 % lidocaine and the exposed cortex was covered with warm paraffin oil. Recording of spontaneous activity (electrocorticogram, ECoG) and evoked potentials (EPs) commenced after an hour of recovery. Somatosensory stimulation was done by a pair of needles delivering weak electric shocks to the whiskery part of the skin (rectangular pulses: 3-4 V, 0.2 msec). EPs were recorded from the primary somatosensory cortical focus (ball-tipped silver electrode) and from the thalamic relay nucleus VPM (steel needle electrode placed to stereotaxic coordinates [24]. The pattern of recording consisted of a five minutes ECoG taken from the cortical sufrace, then EPs by applying one train of 20 stimuli to the whiskery skin. This pattern was repeated every 20 minutes.

Recording and evaluation of the electrical activity was done by a PC and the NEUROSYS software (Experimetria, UK). After 5 control records, mercury ($HgCl_2$, 7 mg/kg) or lead ($Pb(CH_3COO)_2$, 1000 mg/kg) was administered via a peritoneal cannula and the recording was continued for further ca. 2 hours. After averaging the individual EPs, peak-to-peak amplitude and latency of the main peaks (from the stimulus artefact) was measured. From the ECoGs, band activity (standard, delta to gamma [17] was automatically determined and the so-called ECoG index calculated (relation of the low and high frequencies in the recorded ECoG; delta+theta/beta1+beta2).

3. RESULTS

The effect of the heavy metals mostly started immediately after administration and developed in the next 2 hours.

On the spontaneous activity (ECoG index), a trend to decreased activity was seen (Fig. 1). The effect was significant when Hg, but not when Pb, was administered.

The amplitude of the evoked potentials increased. This effect was also stronger with Hg than with Pb, and was generally more pronounced on the cortical

projection area than at the thalamic relay site (Fig. 2). Both metals increased the latency of the EPs (Fig. 3), the effect being significant both at the thalamic and at the cortical recording site.

In case of Hg administration, there was a significant correlation between the alteration of the spontaneous cortical activity (ECoG index) and the evoked potential amplitude, as shown by the correlation diagram (Fig. 4, left). In case of Pb, the correlation was poor (Fig. 4, right).

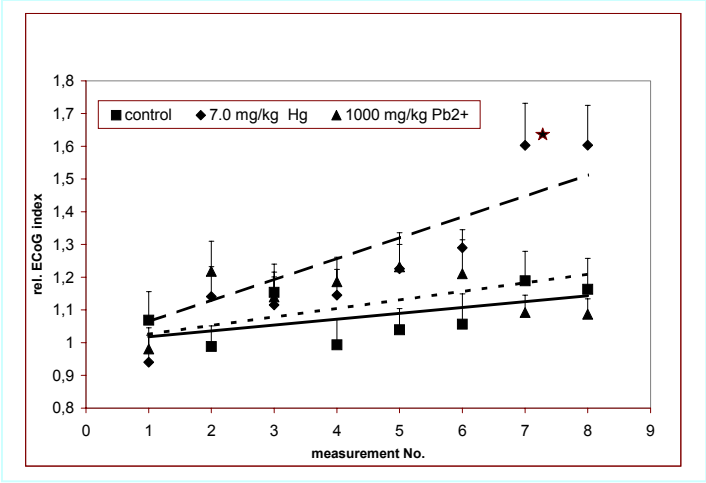

Fig. 1. *Effect of Hg and Pb on the spontaneous cortical activity of rats (mean+SD, n=8).*
Abscissa: measurements (metal given just before measurement 1).
Ordinate: relative change of the ECoG index (treated/averaged control).
*Linear trend lines fitted by EXCEL. *: p<0.05*

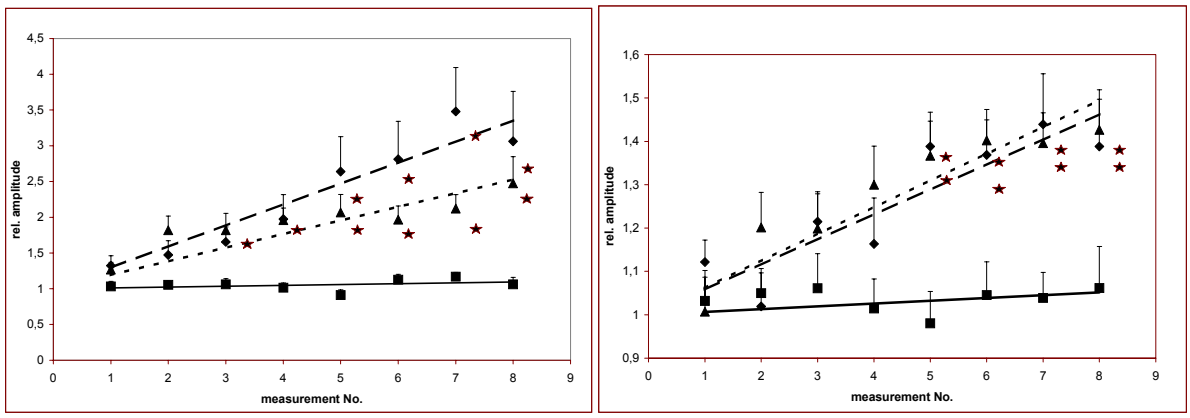

Fig. 2. *Increase of the evoked potential amplitude on Hg and Pb administration in the cortical focus (left) and the thalamic relay site (right). Displayed as in Fig. 1.*

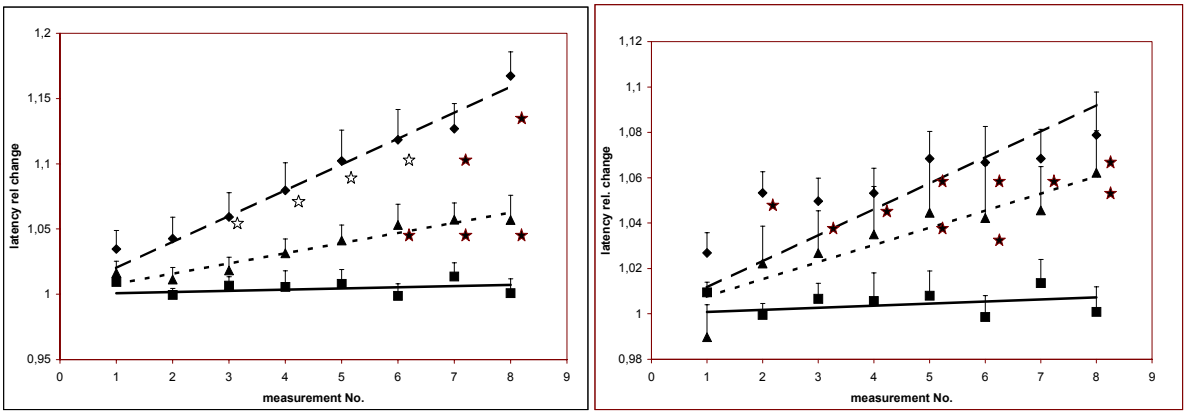

Fig. 3. *Increase of the evoked potential latency (1st peak) on Hg and Pb administration in the cortical focus (left) and the thalamic relay site (right). Displayed as in Fig. 1.*

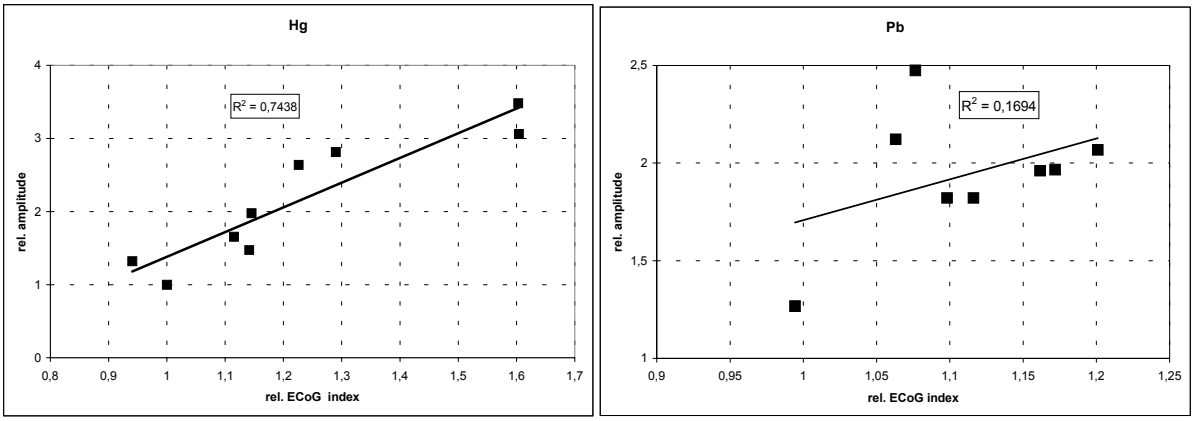

Fig. 4*. Diagrams of the correlation between the changes of the spontaneous (abscissa) and the evoked (ordinate) cortical activity in case of Hg (left) and Pb (right). trend lines fitted and correlation coefficients calculated by EXCEL.*

4. DISCUSSION

Following exposure to inorganic mercury compounds, accumulation of mercury within the CNS has been demonstrated [1,21], in spite of its tendency to bond to plasma proteins. Pb^{2+}, although forms low solubility salts with physiological anions like Cl^-, is readily absorbed by various routes of exposure [31]. Entering the blood stream, Pb^{2+} passes the blood-brain barrier above a concentration threshold [4].

The alterations of the cortical evoked potentials found in our work (increased latency and amplitude) were similar to those seen in chloralkali workers [9] which has several possible explanations. The excitatory thalamocortical input is glutamatergic, and Hg^{2+} inhibits the glial uptake of Glu [7]. Lead, too, interferes with the spontaneous and stimulus-evoked release of Glu and GABA [5,6,30]. In case of Hg^{2+}, an effect on the ascending cholinergic activation [20] is also likely. Hg^{2+} inhibits choline acetyltransferase [12] and decreases the binding of ACh on the muscarinic receptors [26], resulting in less activation. Inorganic lead could possibly interfere with the cholinergic activation of the cortex by increasing the spontaneous and decreasing the

stimulus-evoked synaptic release of ACh [30]. In our results, however, the correlation between spontaneous and evoked cortical activity was firm for Hg^{2+} but poor for Pb^{2+}.

These alterations probably reflect a specific action of the heavy metals on the nervous system so they have a potential use in human health protection.

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TEENAGERS` EATING BEHAVIOUR - A COMPARATIVE STUDY BETWEEN TWO SCHOOLS IN ARAD AND TURNU SEVERIN, ROMANIA

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Abstract:

Teenage is a period of intense physical and psychical development of a person. The aim of this study was to establish the measure in which eating behaviour presents differences between two schools in Arad and Turnu Severin, in Romania. The study was realized on two samples formed of 50 students, each of them, of the Germany Secondary School in Arad and of the Economic Secondary School in Turnu Severin. We applied the questionnaire method. In Arad, the teenagers` diet consisted of milk and milk products, hamburger and sandwich. The same students didn't consume fruits, green salad, fried potatoes, cooked legumes, and fruit juice. In Turnu Severin, the teenagers` diet consisted of fried potatoes, milk and milk products, biscuits, pasta and fruit juice. More than half of the students didn't consume sandwich or hamburger, cooked vegetables. There is a difference between two schools in different towns: Arad and Turnu Severin.

Keywords: eating behaviour, teenagers, secondary school

1. INTRODUCTION

In different regions of Romania differences between eating habits exist. This can act on children growth in the teenage period especially, when happens a growth spurt occurs. Nutrition can influence teenagers' growth. [4] Teenage is the children` period of development between 12 and 19 years and it can be divided into two stages: puberty (12-15 years) and post puberty (16-19 years). In the period 16-19 years the energetic needs are high. In the post puberty stage the boys need 3100 calories daily and the girls 2300 calories daily.[6] Of the caloric values of the ration, at this age proteins represent 13-14%, fats – 28-30% and carbohydrates 56 - 60%. [1-3] These nutritive factors and others are

taken from foods, which are divided into 16 groups. The people of these areas include by these student groups frequently consume: milk and milk products, meat and meat products, cereals and pasta, vegetables, fruits and juices, sugar products.[5]

2. PURPOSE

The aim of this study was to establish if the eating behaviour, as a risk factor, presents differences between the two schools in Turnu Severin and Arad, which are situated in two different geographical areas, in Romania.

3. MATERIAL AND METHOD

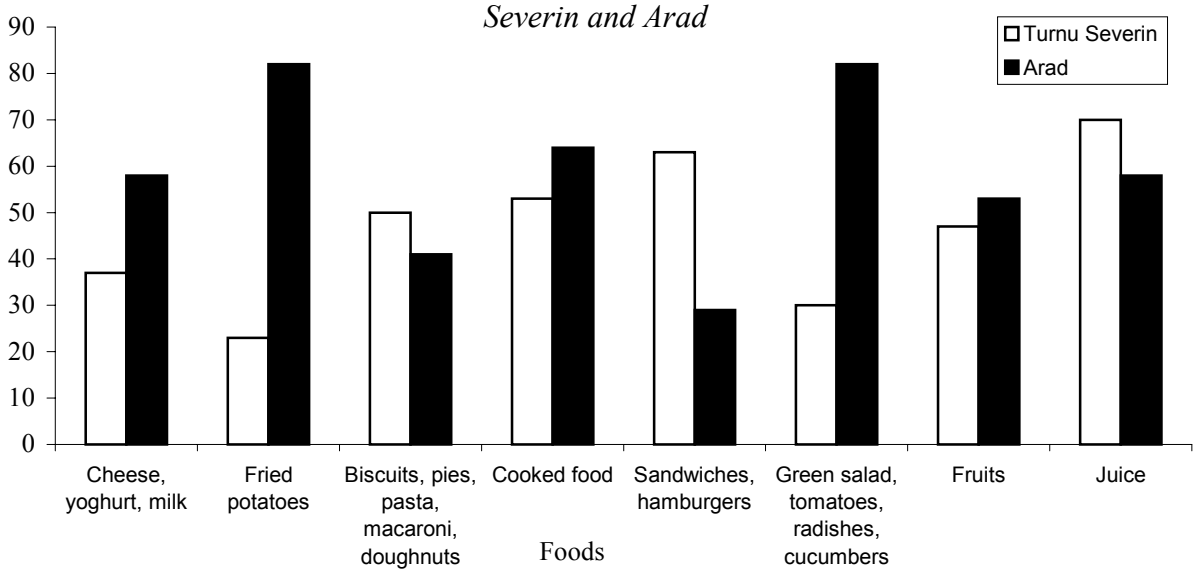
The study was realised with one questionnaire with items for nutrition, applied in two homogenous samples formed of 50 students, each of them, of the German Secondary School, Arad and the Economic Secondary School, Turnu Severin. We analysed and interpreted the obtained results.

4. OBTAINED RESULTS

Students of the Economic Secundary School of Drobeta Turnu-Severin don't consume juices (70% students), sandwiches, hamburgers (63%), cooked food (53%), biscuits, pies, pasta, macaroni, doughnuts (50%).

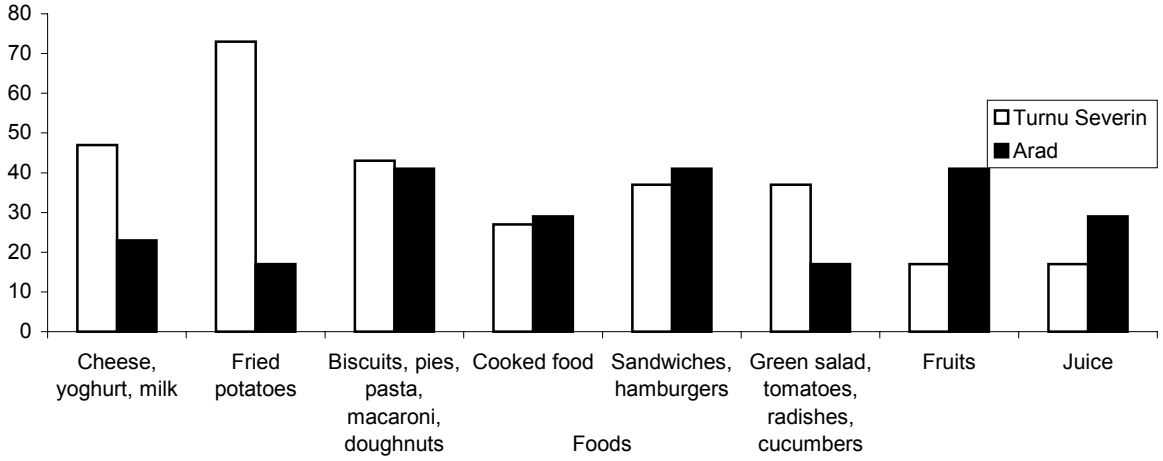
Students of the German Secondary School do not consume fried potatoes, green salad, tomatoes, radishes, cucumbers (82%), cooked food (64%), cheese, yoghurt, milk (58%) and juice (58%) (figure 1).

Figure 1. *The frequency (%) of the students, who do not consume these types of food and food products, in both schools studied in Turnu Severin and Arad*



Students of Turnu Severin consume once/day fried potato (73%), cheese, yoghurt (47%) and biscuits, pies, pasta, macaroni, doughnut (43%). The students in Arad consume once/day: sandwiches, hamburgers, pies, pasta, macaroni, and doughnuts (41%) (figure 2).

Figure 2. *The frequency (%) of the students, who consume once/day these types of food and food products, in both schools studied in Severin and Arad*



The students in Turnu Severin consume more times/day fruits (36%), green salad, tomatoes, radishes, cucumbers (33%) in comparison with the students in Arad, who consume especially sandwiches and hamburgers (29%) (figure 3).

Figura 3. *The frequency (%) of the students, who consume more times/day these types of food and food products, in both schools studied in Severin and Arad*

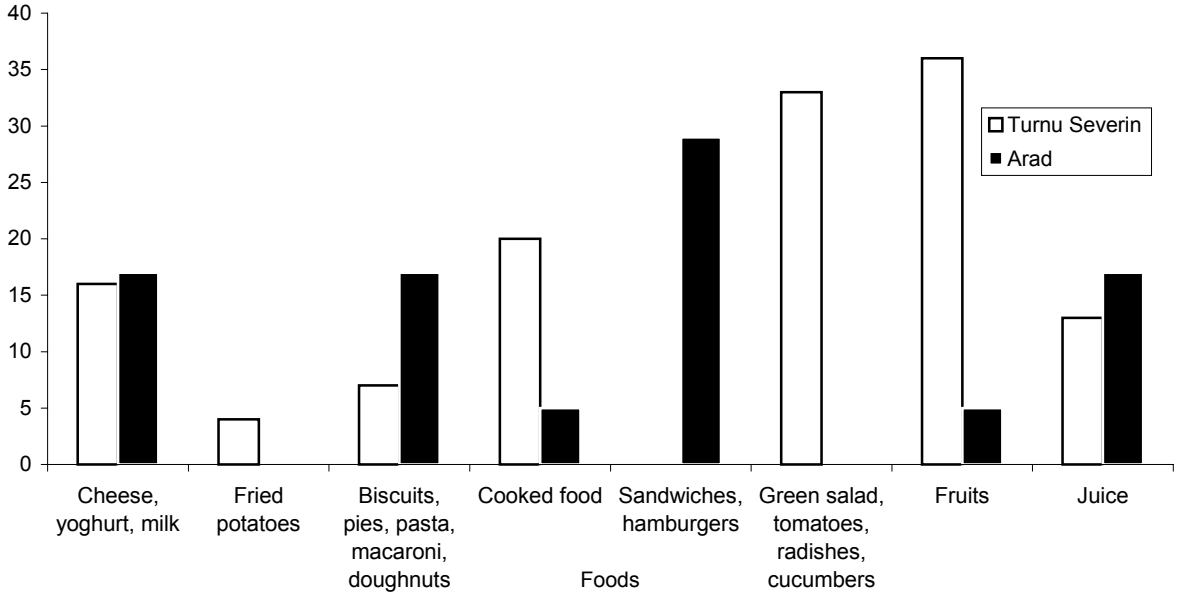
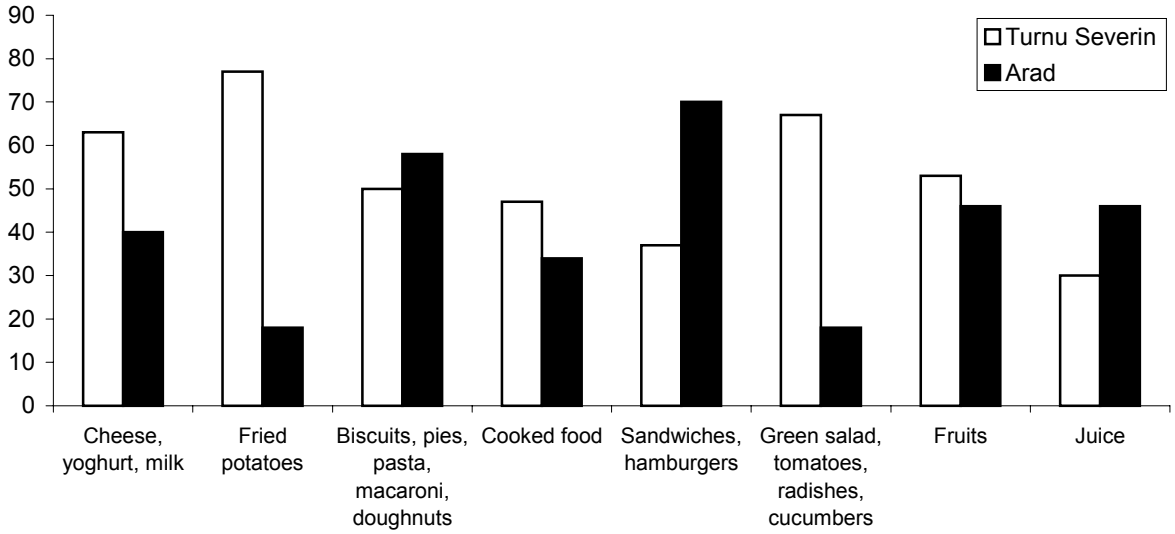


Figure 4. The frequency of the students (%), who consume once or more times/day these types of food and food products, in both schools studied in Severin and Arad



There are differences in the menu diet between the two schools in Turnu Severin and Arad. The first foods that are consumed by the students in Turnu Severin are fried potatoes (77%), vegetables (67%) and milk products (63%), while in Arad the students consume sandwiches, hamburgers (70%) especially, biscuits, pies, pasta (58%) and juice (46%) (figure 4).

It is very interesting to observe that two kinds of eating habits in two schools from different geographical areas are completely different and almost opposite.

In conclusion, in both situations the diet are unbalanced: in Turnu Severin the students receive small quantities of superior quality proteins, which have as sources meat and meat product, and in Arad they receive small quantities of vitamins, minerals and indigestible carbohydrates obtained from vegetables.

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HOUSEHOLD CADMIUM POISSONING

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***Abstract**:*

In household lethal intoxication with Cadmium is rarely met. In medico-legal practice we found 2 cases of lethal cadmium poisoning. In first case we found out that the members of the family consumed grape juice that was kept in a metal recipient.

In second case a man presented a light alteration of the general state. He was brought to the hospital where the death was established.
Conclusions – In this two cases of unexpected death the source of cadmium poisoning was unexpected in household.

***Keywords:** cadmium poisoning, accidental death, household*

The lethal intoxication with Cadmium is rarely met in legal medicine practice. We present here a case of a child and an adult poisoning that showed different major microscopic manifestations.

Case 1: The four-year-old girl, PF, is hospitalised for throw-ups, 38.7^{0}C temperature, and the refusal of food and sleepiness. From the anamnesis we found out that the members of the family consumed grape juice that was kept in a metal recipient and then sieved with a galvanised (with cadmium) sieve. It appears that the girl consumed most of the juice, a twin brother and her father consumed a smaller quantity of the juice. At hospitalisation the girl presented profound altered general state, coughs, bronhic hipersecretion, hepatomegaly, coma of I and II degrees, absence of pupilar reflexes, highlighted ROT, tonico-clonic convulsions.

An acute intoxication atropine type was suspected with an unknown substance (toxicologically unconfirmed). Autopticly we observed a hepatomegaly 25/15/21 cm, sharp anterior edge, light brown parenchyma, on which there were round clear areas yellow orange, with a

diameter between 1.5 and 2 cm, friable zones in comparison with the rest of the parenchyma, in a section, the area was micronodular, and the lobular drawing is highlighted. Minimum renal modifications took place, pale cortical with a wipe cortico-medular limit.

The toxicological investigation infirm the atropine intoxication, but highlights the toxic concentrations of cadmium: 185μg/100 gr. in the liver, and 615μg/100 gr. in the renal tissue. The determinations were made using spectrofotometry of atomic absorption.

The tests for other toxic substances were negative pesticides, drugs that have a SNC and neurovegetative affect, alkaloids, anestesics, anorganic, volatile, metallic (Pb, As, Hg, Zn) toxins were absent.

The histopthological test established the presence of massive hepatic modification with panlobular vacuolar dystrophy, infiltration with lymphocytes and plasmocits on unequal intensity, relatively well determined in some areas.

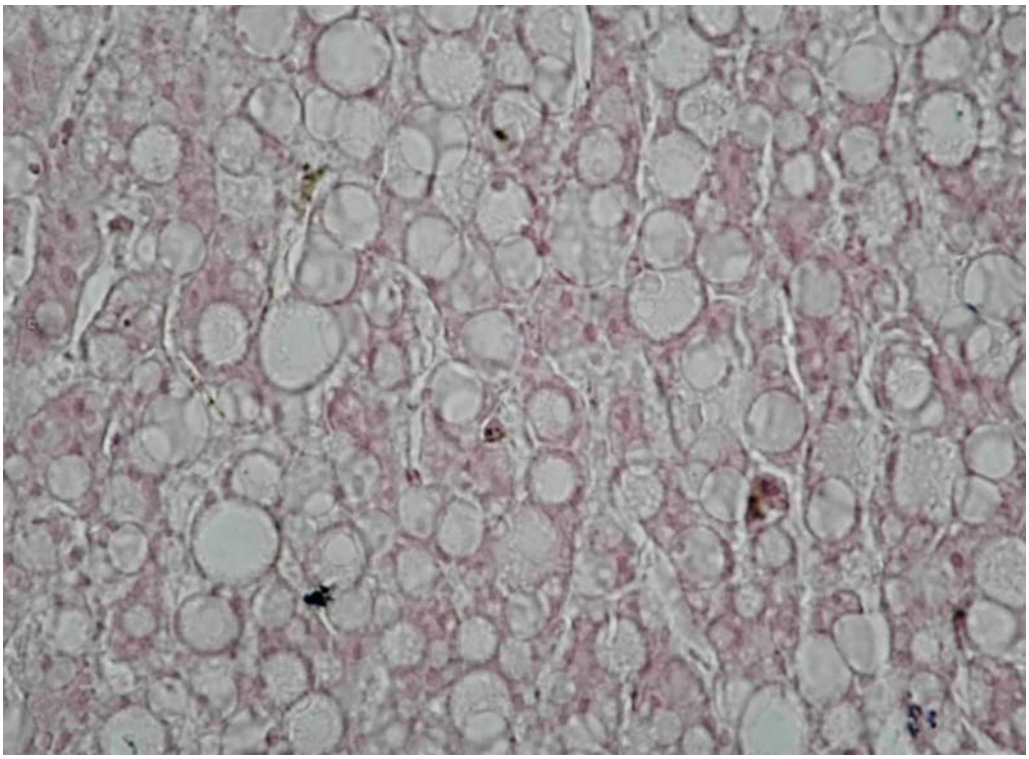

Fig. 1. Panvacuolar hepatic dystrophy col. HE x200

The renal affectation was stasis on medullar and at cortico-medularae junction, haematic thrombi, reduce perivascular haemorrhage, tubular necrosis and necrobiosis. In glomerular capillary we found unequal stasis, hipercelullarity, narrower of glomerular filtration space.

Case II: The 47 year old man presented a light alteration of the general state, for which he was recommended aspirin and paracetamol a day before his death. He presented macroscopically haematuria, for which he was recommended to be hospitalised for the next day. At his home, he

felt sick and threw-up. He was brought to the hospital where the death was established.

Autopticly at the deck, there was a small haemorrhage of 0.2-0.3 cm, and at the right cerebellum lob in the white substance, an area with a low consistence, reddish substance was found. The cerebral ventricles contained a reddish liquid.

At visceral pleura some little haematic petechiae and moderate acute pulmonary oedema (fig. 2).

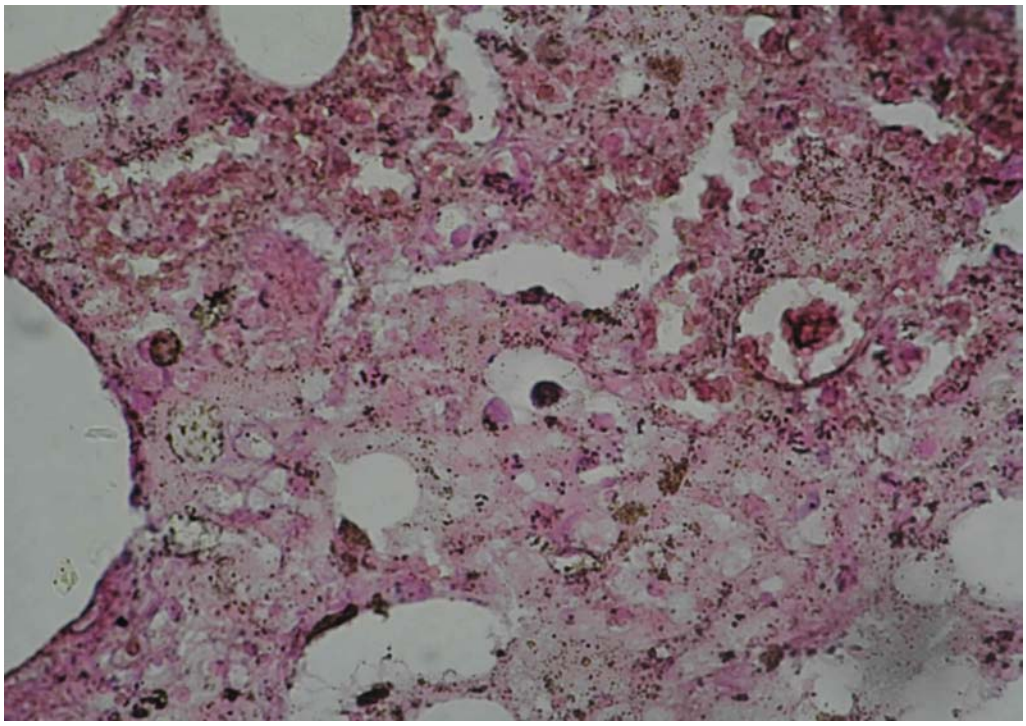

Fig. 2. Acute pulmonary oedema col. HE x200

At the hepatic level, the consistence was a little smaller, with chronic hepatitis modifications. The kidneys presented, at the cortical area, round haemoragic zones with a diameter between 1.5-2 cm, these zones continued at the cortical level affecting partially the medullar. At the renal pelvis on the left we observed a black reddish clot.

The histologic exam showed a cerebellum menigeal haemorrhage and recent cerebellum haemoragic areas, chronic emphysema and moderate interstitial oedema. Chronic persistent hepatitis with dystrophy. The most particular modifications are the renal modifications - necroses and proximal tubular pluricelular necrobioses, associated with proliferative glomerulo-nephrite. Isolated cortical hematic suffusions and more frequent at the renal papillae are also observed.

The toxicological exam was negative for alcohol, drugs that affect SNC, organofungicides, volatile toxic, aromatic hydrocarbons, halogenated organic solvents, metals (copper, Zn, Pb, As, Hg). The amoniphenasone and paracetamol in gastric tissue were highlighted. Through

spectrofotometry of atomic absorption we identified, 240 μg/100 gr. in the hepatic tissue and 1290 μg/100 gr. in the renal tissue.

Conclusions - The cases we presented were the only ones met in the Institute of Forensic Medicine from Timisoara in a 14-year period.

The macroscopically and microscopically findings were different: hepatic necrosis (at the girl) and modifications of the vascular permeability at the adult, which both determined multiple haemorrhages. The microscopically modifications proved the affection of the three target organs lung, liver and kidney.

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Study of synthesis and characterisation possibilities of β-2 ethylhexyl (EH) polyethylenoxy ($\bar{n}$ = 0-20) propionic acid amides

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Abstract:

This paper aims at analysing the opportunity for expanding synthesis intermediary range for ***fat substitutes*** (F.S.) as potential precursors of foodstuff additive products with controlled functionality. The researches are part of research project of the Department for foodstuff technologies and of experimental work as doctoral studies of the authors and they aim at synthesising β-2-ethylhexyl (EH) polyethylenoxy ($\bar{n}$ = 0 – 20) propionic acid amides

$$[i\text{-}C_8H_{17}\text{-}O(CH_2CH_2O)n\text{-}CH_2CH_2CONH_2\ (I)]$$

by nucleophilous basic catalysis addition of 2-ethyl-hexyl (EH) polyetoxylate alcohols ($\bar{n}$ = 0 – 20) alcohol industrially to monomer acrylamide followed by purification and physico-chemical and chemical characterisation of derivation products.

Keywords:

β-2 ethylhexyl (EH) polyethylenoxy ($\bar{n}$ = 0 – 20) propionic acid amides, fat substitutes, foodstuff additive products

1. Presentation of the matter

The increasing interest in the diversifying of food additive range, in general, in ecological lipid and technologically performing substitutes in food processing asked for the reassessment of new bases of raw materials and chemical reaction schemes. This paper, part of a doctoral thesis in progress, allows such a practical approach of the matter.

2. Experimental approach

In a reaction vase, endowed with mechanical stirring, thermometer, dripping funnel, refrigerant, inert atmosphere at a proper temperature (30-35°C for the EH alcohol), we introduce 0.1 mols of superior polyethoxylate, purified alcohol, 0.0025 mols of basic catalyst ($CH_3O^-Na^+$), we suspend 0.0025 mols (0.38 g) of anhydrous, finely divided $FeSO_4$ and we cautiously add and stir for 30-45 min (0.11 mols) of monomer acryl-amide, so that mixture temperature does not go above the prescribed value (if necessary, the reaction vase is cooled in a water-ice bath). We leave in the same conditions for about 2 h to perfection, we neutralise the catalyst with about 4.8 cm^3 of CH_3COOH 5%, we filter in warm conditions the precipitated salts; the fluid residue of a light yellow is purified if necessary from oligomer by elution on silica gel column. Yields as compared to superior polyethoxylated alcohol are practically quantitative.

3. Results, discussions

Primarily, we worked without using an oligomerisation inhibitor of the acrylic monomer, in order to follow the evolution of the carbamoilation and oligomerisation yields. Subsequently, under optimal parameters, we introduced $FeSO_4$ as oligomerisation inhibitor.

In equimolar conditions AN/EH-O-H, the raise of temperature in the interval 25-45° C causes the increase of yields in nucleophilous addition, after which in the domain 40-60 °C yields decrease. In monomer excess, yield evolution follow the same steps. In these conditions in the interval 25-40 °C oligomer amount formed is below 1%, no matter de monomer excess used, and in the interval 40-60 °C it increases significantly. For i-$C_8H_{17}OH$(EH), in equimolar conditions and monomer excess AN/EH-O-H, the increase of temperature between 45-55 °C (below this interval yields are very low) favour nucleophilous addition yields the greater monomer excess. In the interval 55-70 °C, yields decrease in the same conditions, and oligomer content is higher. In carbamoilation polyethoxylated EH-O-$(EO)_nH$ (n = 3), in equimolar conditions or in monomer excess, addition yields increase in the interval 25-35 C°, after which in the interval 45-60 °C they decrease considerably. Oligomer amount formed follows approximately the same evolution as in the case of EH-OH for the whole temperature range. For polyethoxylated alcohol (n = 3) in equimolar or monomer conditions addition yields increase in the interval 30-40 °C after which they decrease between 45-65 °C. The content in oligomers increases with temperature and monomer excess. These behaviours suggest that carbamoilation reaction also has a reversible character, polyethoxylated chain favours the addition, and increasing hydro-carbonated chain decreases yields.

Gas chromatographic analysis (though diminished volatility for n ≥ 5 limits the method), relative distribution of polyoxylated chain homologues distribution for a series of polyethoxylated alcohols probes (Hüls) (n = 3) purified only by polyethyleneglycols proved that the share of superior free alcohols vary between 15-40% for the same polyetheric chain.

The difference between hydrocarbonated chain homologues distribution values determined by acid splitting (HI) and the evolution of assessed gas

chromatographically distribution is apparent because chromatographic value mean is identical with analytical assessment. These conclusions are of interest for supplementary enlightening of the polyoxyethylation mechanism but in the context of this paper it suggests a greater reactivity of the chain with an increase of polyetheric chain. Gas chromatographic follow-up of the relative evolution of polyethoxylated (n = 3) alcohol addition process yields allowed us to see that modifying polyetheric chain in isothermal conditions (30 or 40°) and the same molecular ratio of the reactants decreases the ratio of hydrocarbonated chain homologues.

In the process of carbamoilation reaction duration favours the formation of β-EH-oxypropionamides up to 180 minutes, and monomer oligomerisation for the whole duration of the process. After this interval, yields decrease, suggesting the reversible character in the presence of a prolonged contact between reactants.
In the series of polyethoxylated (n = 3) alcohols EH maximum value of cyanoethylation yield is reached for a shorter duration of reaction that can suggest the intervention of polyoxyethylene chain in the process. On the whole, increasing the duration of reaction up to about 180 minutes favours all the steps of the reaction, including AN oligomer formation (***Table 1***).

"***Table 1.*** *Dependence of carbamoilation yield of polyethoxylated alcohols EH (%)/ oligomers (%), on duration, solvent, temperature 30°, molar ratio AN/EH-$(EO)_n$-H 1,1/1, catalyst concentration [$CH_3O^-Na^+$] 5 · 10^{-3} mol/l, without an oligomerisation inhibitor*"

Nr.	Symbol	Hydroxyl substratum	Duration of the reaction (minutes)							
			30	60	90	150	180	210	240	270
1	EH-$(EO)_n$-H	n = 0	53.537	66.500	70.003	80.608	89.443	86.256	76.084	69.175
			–	–	0.118	0.338	0.456	0.518	0.738	0.886
2		n = 3	55.331	75.420	83.110	92.181	95.938	96.547	94.718	90.175
			–	–	0.103	0.251	0.288	0.381	0.665	0.728
3		n = 9	58.437	85.528	94.523	100.00	100.00	100.00	100.00	100.00
			–	–	0.096	0.175	0.211	0.296	0.334	0.508
4		n = 12	69.387	91.368	98.033	100.00	100.00	100.00	100.00	100.00
			–	–	0.084	0.098	0.179	0.213	0.299	0.308
5		n = 20	78.263	96.297	100.00	100.00	100.00	100.00	100.00	100.00
			–	–	0.031	0.121	0.147	0.131	0.243	0.281

After about 180 minutes, the content in β-EH-polyethyleneoxy-propionitrile decreases because of the prolonged contact with the basic environment.

Increasing catalyst amount above the optimal value (4-5 · 10^{-3} mol/l) increases the alkalinity of the reaction environment and favours oligomerisation reactions of the acrylic monomer. In the field of concentrations 15-50 · 10^{-3} mol/l, the content in β-EH-polyethyleneoxy (n = 3) propionamides decreases, together with the outstanding increase of the content in oligomers. In the series of polyoxyethylene chain homologues maximum yield is reached at greater values of catalyst concentration. For the same catalyst concentration the increase of polyetheric chain causes not only a sensible increase of the nucleophilous addition yields but also a diminution of oligomer amount, maybe due to the solvatation of acrylic monomer in polyetheric chain (***Table 2***).

"***Table 2.*** *Dependence of carbamoilation yield on polyethoxylated EH alcohols (n = 3-20) (amine / oligomer AN %), on catalyst excess [$CH_3O^-Na^+$] toluene solvent, duration 60 minutes, temperature 30°, molar ratio AN/EH-$(EO)_n$-H 1,1/l*"

Nr.	Hydroxyl substratum	Concentration of basic catalyst (mol/l) · 10^{-3}							
		2	5	9	15	25	30	40	50
1	n=0	64.111	66.501	68.821	73.474	70.531	62.098	56.621	57.411
		–	–	0.773	1.530	8.543	13.182	15.324	18.460
2	n=3	74.980	75.420	69.320	76.453	72.332	67.567	62.631	49.684
		–	–	0.534	1.063	7.148	12.834	13.733	16.920
3	n=9	85.171	88.639	90.285	82.446	73.437	72.142	65.733	50.818
		–	–	0.263	0.631	5.314	11.968	12.420	14.789
4	n=12	87.186	90.333	92.263	93.521	78.534	75.565	69.432	58.326
		–	–	0.095	0.287	4.868	11.524	12.081	13.920

"***Table 3.*** *Dependence of carbamoilation yield of EH polyethoxylated alcohols (n = 3-20) on the content in superior alcohol, molar ratio AN/EH-$(EO)_n$-H 1,1/l, duration 30 minutes, solvent toluene, temperature 30°, catalyst [$CH_3O^-Na^+$] 5 · 10^{-3} mol/l*"

Nr.	Superior alcohol added (%)	Number of ethylene oxide units (n)	Yields of amides (%)
1	5	3	85.08
2	10	3	80.18
3	20	3	78.32
4	30	3	75.38
5	5	20	98.44
6	10	20	95.62
7	20	20	92.32
8	30	20	88.81

The presence of superior alcohols in variable amounts (5-40%) in polyethoxylated superior alcohols negatively influences yields through monomer equivalent use and diminished speed (***Table 3***). This phenomenon can be explained on one side through steric effects of hydrocarbonated chain and through lower nucleophilous activity of alkaline superior alcoxides as compared to polyethoxylated alcohols.

The presence of polyethyleneglycols in variable amounts (1-10%) in superior polyethoxylated alcohols influences yields in two directions:

- ✓ high consumption of monomer (1-2 mols/l mol PEG) with the formation of polyethyleneglycol (n = 3-30) dipropionamide;
- ✓ participation of carbamoilated polyethyleneglycols in the process as oligoglymes mono- or di-protected CTF (activation of superior alkaline alcoxides through the coordination of superior cation). The favourable contribution on yields depends on the average degree pf polyethoxylation of the superior alcohol (n) as polyoxyethylene chain homologue distribution is the wider the superior alcohol is

more polyethoxylated and the distribution of PEG homologues (n = 3-30) is wider than that of $EH(EO)_n$-H (n = 3-20) homologies for the same average content in ethylene oxide.

In this paper we can see:

- ✓ the increase of content in oligomers due to the fact that alkaline polyethyleneglycols (n = 3-30) as bidentate nucleophilous agents have a higher basicity than alkaline superior alcoxides;
- ✓ the increase if content in β-EH-polyethyleneoxypropionamides, proportionally with the amount of polyethyleneglycols ($\bar{n}$ = 3-30) on the ground of interphase transfer catalysis (***Table 4***).

"***Table 4.*** *Dependence of carbamoilation yield of 2-ethyl-hexilic alcohol on polyethyleneglycol amount PEG ($\bar{n}$ = 9), molar ratio AN/EH-$(EO)_n$ 1,1/l, duration 60 minutes, toluene solvent, catalyst concentration $[CH_3O^-Na^+]$ 5 · 10^{-3} mol/l, temperature 25°, without an oligmerisation inhibitor*"

Nr.	Content in polyethyleneglycol in the superior polyethoxylated alcohol (%)	Yield ratio amide / oligomer (%)
1	0	67.48/ -
2	3	75.09/0.198
3	5	76.38/0.226
4	8	80.03/0.318
5	10	85.62/0.376
6	12	88.71/0.434
7	15	92.25/0.618

In this paper we have avoided the formation of vinyl oligomers through the introduction of anhydrous ferrous sulphate ($FeSO_4$) as polymerisation inhibitor of the acrylic monomer. For 1% polymerisation inhibitor cyanoethylation yield of the alcohol increases to over 10% without oligomer formation. Similar results can also be obtained in the series of superior polyethoxylated alcohols ($\bar{n}$ = 3-20).

4. Conclusions

Researches carried out and results obtained allow us to be confident in pursuing our studies on the valorising of 2-ethyl-hexilic alcohols (EH) as such and polyethoxylated ($\bar{n}$ = 0-20) with a view to extinguishing food additives range through guided building of new fat suppliers.

5. References

C. Jianu, unpublished data (doctoral thesis)

THE DYNAMICS OF THE VETERINARIAN PERSONNEL IN ROMANIA, HUNGARY, SERBIA AND MONTENEGRO IN 1997-2001 PERIODS

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Abstract:

In the paper is presented the evolution of the veterinarians and technical personnel from veterinary field from Romania, Hungary and Serbia and Montenegro in five years.

On the whole, in the period 1997-2001, in these studied countries, the number of the veterinarians increased in Romania and Hungary, but this number in Serbia and Montenegro decreased. The technical personnel remain constant in Hungary and Serbia and Montenegro, but this number in Romania decreased.

***Keywords*:**

veterinarians, technical personnel

From adhere Romania to E.U., the number of the veterinarians and technical personnel has a different evolution, compare with other countries, these differences can be observed in the tables and graphics bellow.

MATERIALS AND METHODS

For realization this paper, the data were taking over from Internet by the site of OIE [1], the web address: *http://www.oie.int/hs2/report.asp*

RESULTS AND DISCUSSION

After the processing of the information were obtained results concerning the evolution of the veterinarians and technical personnel in these countries.

In Romania, between 1997-2001 is observed high oscillation of the number of veterinarians and technical personnel, both, but, at the end of this period the number of the veterinarians was close from the values from the beginning of the studied period (8.400), contrary, the technical personnel was drastically diminishable (5.272 vs. 8.100).

In the table 1 is present the evolution of the veterinarians and technical personnel in Romania between 1997-2001.

Table 1. *The number of the veterinarians and technical personnel in Romania*

Year	Number of	
	veterinarians	**technical personnel**
1997	8260	8100
1998	8320	7280
1999	8015	1825
2000	7614	1348
2001	8403	5272

In Hungary, the number of the veterinarians increases from 2920 in 1997 to 4088 in 2001, while the increase of the technical personnel was insignificant in this period.

In the table 2 is present the evolution of the veterinarians and technical personnel in Hungary between 1997-2001.

Table 2. *The number of the veterinarians and technical personnel in Hungary*

Year	Number of	
	veterinarians	**technical personnel**
1997	2920	490
1998	3250	530
1999	3772	537
2000	3939	542
2001	4088	548

In Serbia and Montenegro, the number of the veterinarians in the first three years of the studied period was round about 2.700, has an important decrease in 2000 and 2001 till 1400, while the technical personnel was constantly over 2.000.

In the table 3 is present the evolution of the veterinarians and technical personnel in Serbia and Montenegro between 1997-2001.

Table 3. *The number of the veterinarians and technical personnel in Serbia and Montenegro*

Year	Number of	
	veterinarians	technical personnel
1997	2616	2016
1998	2687	2000
1999	2730	2090
2000	1483	2008
2001	1442	2049

On the whole, in the countries studies in 1997-2001 periods, the number of the veterinarians increases in Romania and Hungary and decrease in Serbia and Montenegro (table 4 and figure 1). The veterinary technical personnel remain constant in Hungary and Serbia and Montenegro and decrease in Romania (table 5 and figure 2).

Table 4. *The evolution of veterinarians in Romania, Hungary and Serbia and Montenegro*

Country / Territory	1997	1998	1999	2000	2001
Hungary	2920	3250	3772	3939	4088
Romania	8260	8320	8015	7614	8403
Serbia and Montenegro	2616	2687	2730	1483	1442

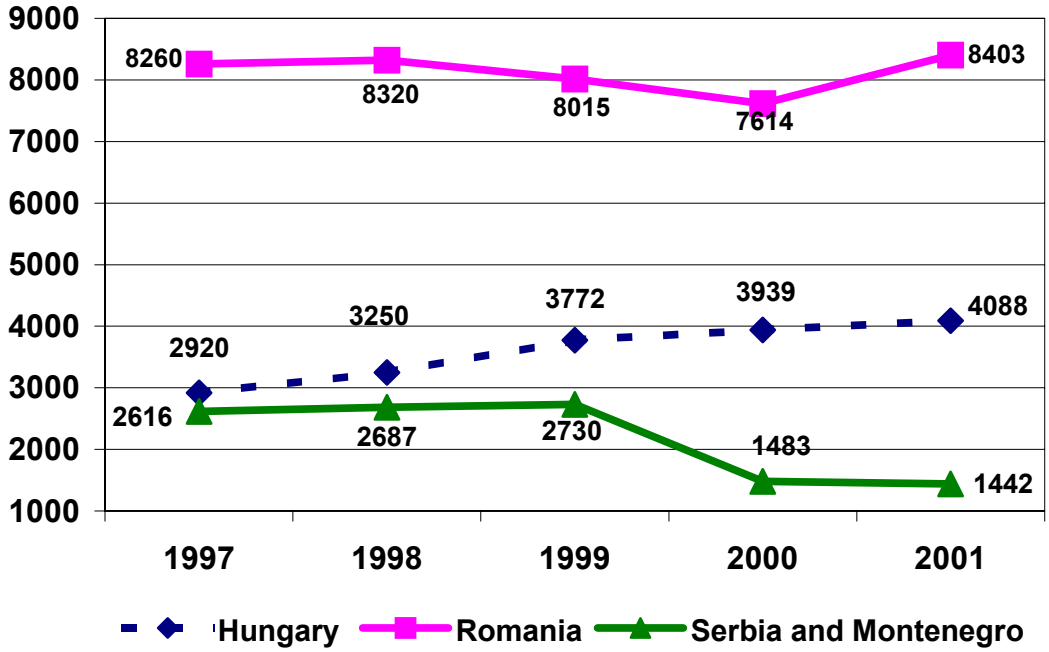

Fig. 1. *The dynamics of the veterinarians' number from Romania, Hungary and Serbia and Montenegro*

Concerning the total number of veterinarians the decrease order is: Romania, Hungary and Serbia and Montenegro.

Table 5. *The evolution of veterinary technical personnel in Romania, Hungary and Serbia and Montenegro*

Country / Territory	1997	1998	1999	2000	2001
Hungary	490	530	537	542	548
Romania	8100	7280	1825	1348	5272
Serbia and Montenegro	2016	2000	2090	2008	2049

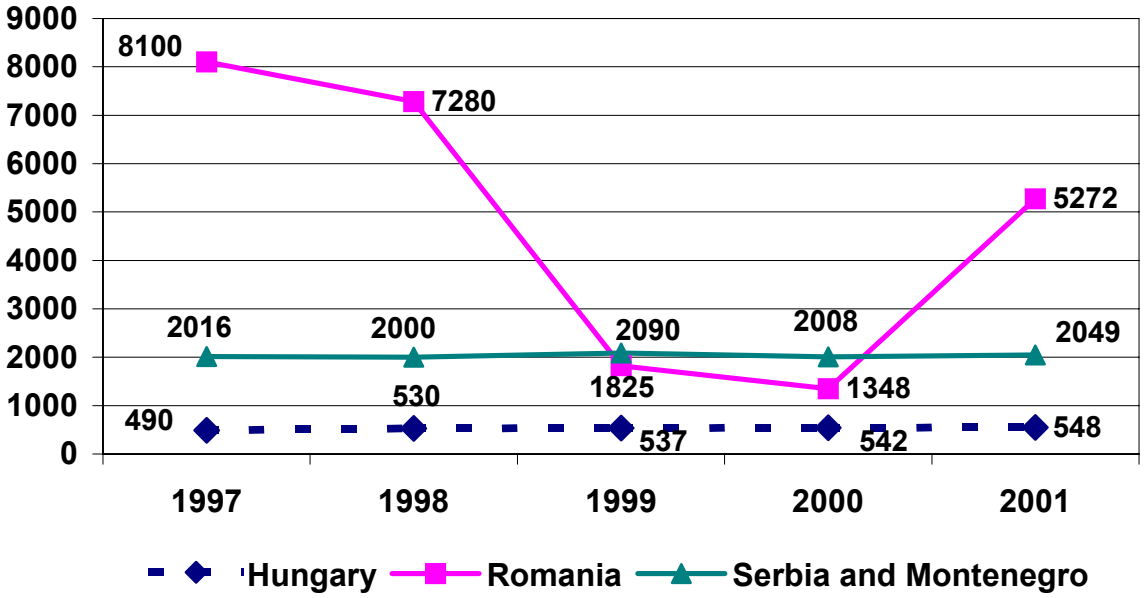

Fig. 2. *The dynamics of veterinary technical personnel number from Romania, Hungary and Serbia and Montenegro*

Concerning the total number of veterinary technical personnel the decrease order is: Romania, Serbia and Montenegro and Hungary.

CONCLUSIONS

In the end of this study can be observe difference between the evolution of the veterinarians and technical veterinarians from these three countries, even they are in the same part of the Eastern Europe.

In the countries studies in 1997-2001 periods, the number of the veterinarians increases in Romania and Hungary and decrease in Serbia and Montenegro. The technical personnel remain constant in Hungary and Serbia and Montenegro and decrease in Romania.

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DIRECT PHOTOLYSIS OF 2-AMINO-5-CHLOROPYRIDINE

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Abstract:

Kinetics of direct photolysis of a pyridine pesticide analogue, 2-amino-5-chloropyridine, have been investigated at different initial concentrations of the substrate. Kinetics were studied by monitoring the reaction of chloride generation by direct potentiometry. Upon comparing the reaction of direct photolysis with the reaction in the presence of TiO_2 it was found that the reaction of chloride elimination, i.e. degradation of the initial substrate takes place at a greater rate in case of direct photolysis than it does in the presence of TiO_2.

Keywords:

direct photolysis, TiO_2, 2-amino-5-chloropyridine, water remediation

1. INTRODUCTION

Contamination of waterstreams by different organic pollutants is becoming an increasing environmental problem. Because water supplies are limited in the world, there is a need for development of new methods for water remediation. These methods should be efficient and cost-effective. There have been attempts to apply the direct photolysis process to these purposes [2,3,5,7-12]. The direct photolytic process is based on the use of UV-light for degradation of organic compounds. In many cases, direct photolysis is not an efficient process regarding complete mineralization, but causes degradation of organic compounds to some extent. Heterogeneous photocatalysis, on the other hand, has proven to be an efficient method to completely mineralize organic compounds [1,4,6,10,12,14,15]. For this reason, the aim of this work was to investigate the kinetics of direct photolysis of a pyridine pesticide analogue [13], 2-amino-5-chloropyridine, at different initial concentrations of the substrate and compare them to the kinetics of photocatalytic degradation. Potentiometric monitoring of chloride generated during the process was used for these purposes.

2. EXPERIMENTAL

Reagents and solutions

All chemicals used were of reagent grade. 2-amino-5-chloropyridine, as well as KNO_3, were purchased from Merck. NaCl was purchased from Zorka (Šabac, Serbia and Montenegro). Titanium dioxide Degussa P25 (specific surface area 50 ± 15 m^2/g, nonporous) was used as photocatalyst. In all experiments doubly-distilled water was used.

For the investigation of the effect of initial substrate concentration, a stock solution of 2-amino-5-chloropyridine ($c \sim 2.5$ mmol/dm^3) was prepared. This solution was appropriately diluted to obtain solutions of lower concentrations (0.5 – 2.0 mmol/dm^3).

For calibration of the chloride electrode, a stock solution of NaCl (100 mmol/dm^3) was prepared. By multiple dilution of this solution, a series of standards was prepared (0.01 – 3 mmol/dm^3). Since titanium dioxide contains chlorides, by manufacturers declaration, it was added to the standard solutions in the same amount (2 mg/cm^3) as for the photodegradation procedure.

Photodegradation procedure

For the experiments 20.0 cm^3 of the investigated compound solution were measured into a double-walled photochemical cell made of Pyrex glass, equipped with a magnetic stirring bar. KNO_3 (100 mmol/dm^3) was then added to keep the ionic strength constant during irradiation. For the photocatalytic degradation experiments, 40 mg of titanium dioxide were added and the solution was sonified to make the particles uniform. Thus obtained solution was then thermostated at 40 ± 0.5 °C in a stream of oxygen. A 125 W Philips HPL-N mercury lamp, with the highest emitted intensity in the UV region at 365 nm, was used as the irradiation source.

Analytical procedure

Concentration changes of chloride generated during the degradation were monitored by a chloride ion selective electrode (Mettler Toledo Me-51340400) coupled to a saturated calomel electrode (Iskra K401) via a potassium nitrate electrolytic bridge and connected to a pH-meter (Radiometer PHM62).

3. RESULTS AND DISCUSSION

Since photocatalytic degradation is in most cases a more efficient method for degradation of organic compounds than direct photolysis [1,3,8,12], the efficiency of photocatalytic degradation of 2-amino-5-chloropyridine was primarily investigated. Kinetics of the degradation were monitored by determination of chloride generated in the reaction, since these two processes take place simultaneously.

Upon investigation of the degradation of the substrate in presence of titanium dioxide (Fig. 1), it was found that the reaction is of the first-order, in the entire investigated concentration range. However, the values for the reaction rate constants (Table 1) were significantly higher than those found in literature [1,4,6,8,15]. According to the mechanism which involves the reaction with hydroxyl radicals formed by excitation of TiO_2, these values should be independent from the compound type [16]. This discrepancy indicates that the reaction of chloride elimination takes

place by another mechanism – one not involving the hydroxyl radicals formed on the surface of titanium dioxide. It can also be concluded from Table 1 that the decrease in the reaction rate constant is more pronounced at lower concentrations.

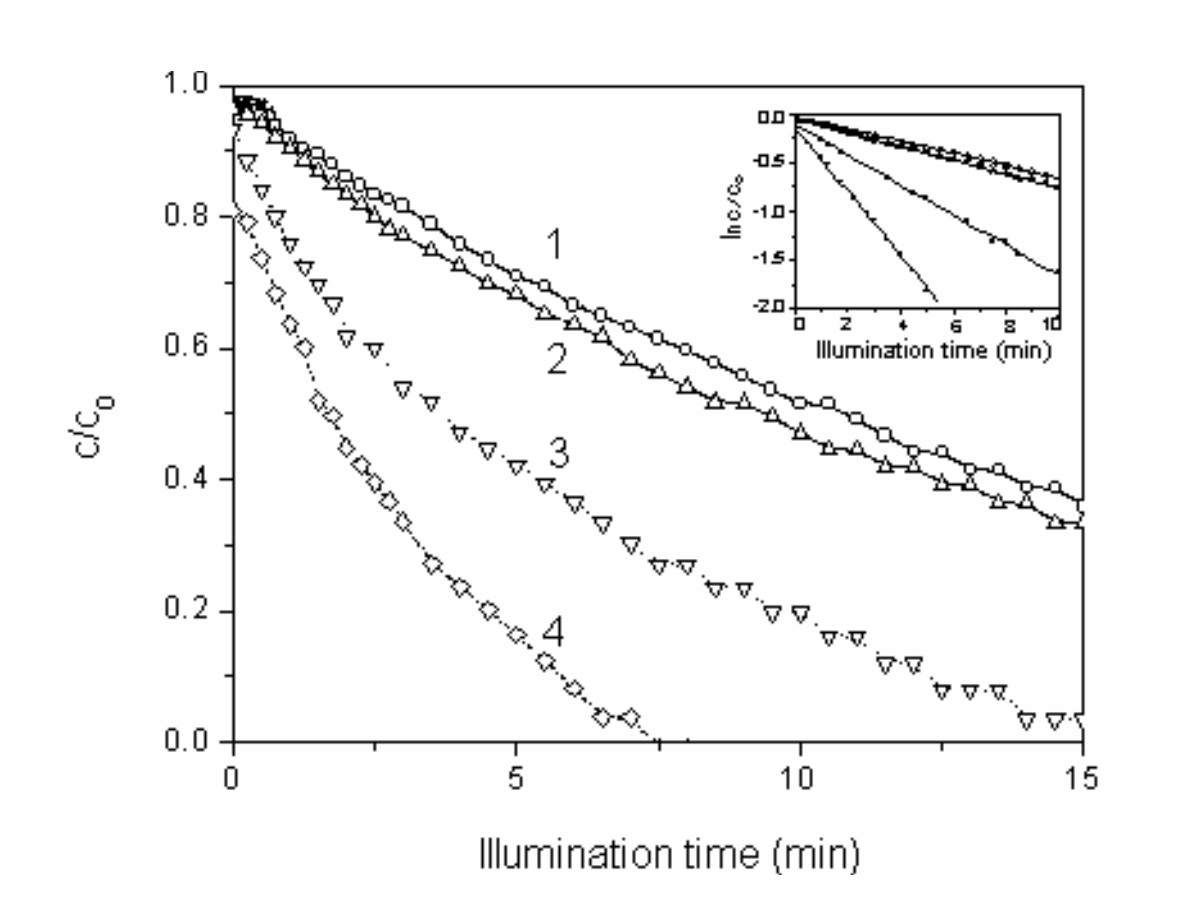

Fig. 1. *The effect of the initial concentration of 2-amino-5-chloropyridine (mmol/dm^3) on the rate of photodegradation* in the presence of TiO_2 (2 mg/cm^3)*: (1) 2.5; (2) 2.0; (3) 1.0; (4) 0.5. The insert represents the linear transform ln(c/c$_0$) = f(illumination time).*

Table 1. *Effect of initial concentration (c$_0$) of 2-amino-5-chloropyridine on the rate of photodegradation in the presence of TiO_2 (2 mg/cm^3)*

c_0 (mmol/dm^3)	$10^2\,k$ (min^{-1})§	$r^{\dagger}$	$t_{1/2}$ (min)‡
0.5	34	0.998	2.0
1.0	15	0.999	3.8
2.0	7.1	0.999	9.3
2.5	6.3	0.999	10.7

§ first-order rate constant; † linear regression coefficient; ‡ half-life.

For this reason kinetics of photodegradation were studied in the absence of titanium dioxide, i.e. under the conditions of direct photolysis. The effect of the initial substrate concentration is presented in Fig. 2. It was found that not only does the reaction take place in the absence of titanium dioxide, but it is somewhat faster and

of the zero-order (Table 2), which further supports the fact that the reaction doesn't take place via hydroxyl radical mechanism generated from TiO_2, but by another mechanism. Namely, although the half-lives of the reactions are similar, the time necessary for chloride elimination is mainly lower. Lower reaction rate of chloride elimination in the presence of titanium dioxide is probably a consequence of the opacity of the suspension and light scattering by titanium dioxide particles.

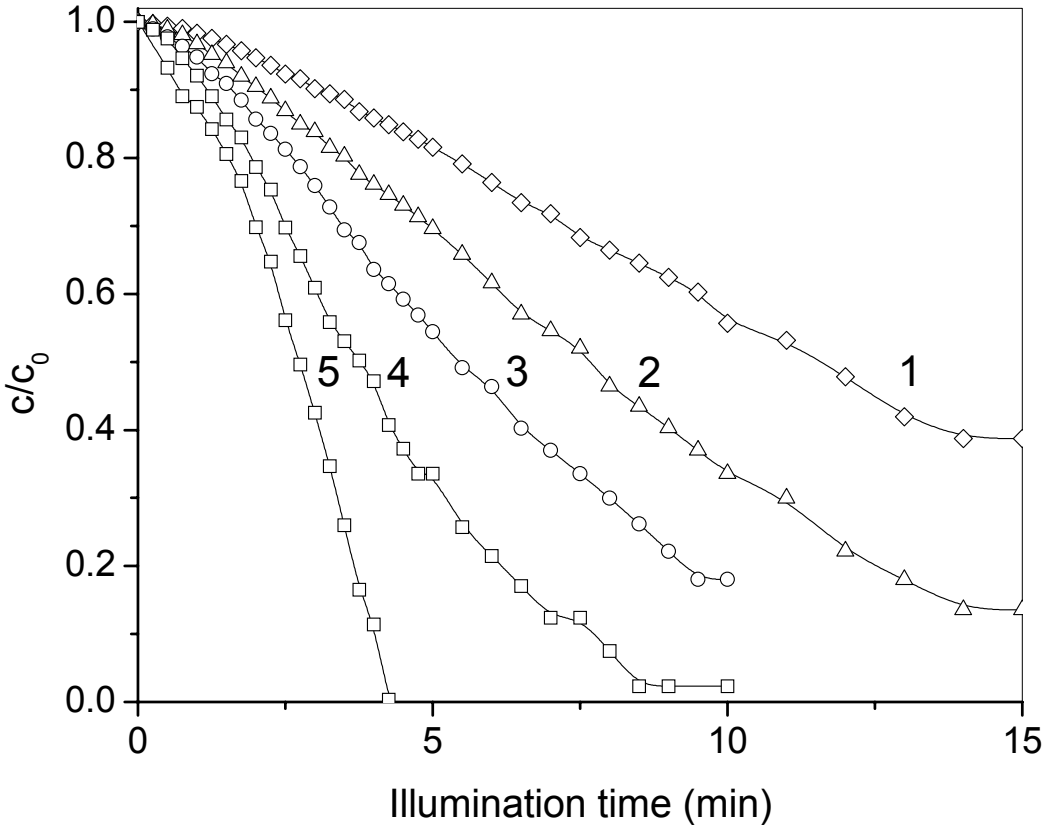

Fig. 2. *Effect of initial concentrations of 2-amino-5-chloropyridine (mmol/dm^3) on the rate of direct photolysis: (1) 2.5 ; (2) 2.0; (3) 1.5; (4) 1.0; (5) 0.5.*

Table 2. *Effect of initial concentration (c_0) of 2-amino-5-chloropyridine on the rate of direct photolysis*

c_0 (mmol/dm^3)	10^4 k (mol dm^{-3} min^{-1})[§]	r[†]	$t_{1/2}$ (min)[‡]
0.5	1.0	0.991	2.6
1.0	1.3	0.998	3.8
1.5	1.3	0.999	5.8
2.0	1.3	0.999	7.8
2.5	1.1	0.999	11.7

[§] zero-order rate constant; [†] linear regression coefficient; [‡] half-life.

As can be seen from Table 2, in the case of the reaction of direct photolysis, the reaction rate constants have similar values as expected, which as was discussed, is not the case with photodegradation in the presence of TiO_2.

4. CONCLUSION

During the investigation of direct photolysis of 2-amino-5-chloropyridine it was found that its dechlorination is a fast process, in the entire investigated concentration range, as well as that it follows zero-order kinetics. Kinetics of chloride evolution for direct photolysis and photodegradation in the presence of TiO_2 were compared. It was found that the reaction of chloride evolution is retarded by TiO_2 presence due to the opacity of the suspension and light scattering by its particles. However, high rate of the initial compound degradation doesn't necessarily indicate its complete mineralization. Namely, formation of much more stable, and what's probably worse more toxic intermediates is very common.

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TRILATERAL COOPERATION IN THE FIELDS OF UNIVERSITY ACTIVITIES

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Abstract: *The Faculties of Pharmacy in Szeged and in Targu-Mures take part in the ac-tive trilateral cooperation in the graduate and postgraduate education of students and pharmacists, and in the scientific cooperation and medical treatment of the population in this region. The staff and students of the two Universities have excellent connections in practically all possible fields of cooperation.*

Keywords: *cooperation, education, research, medical treatment, management*.

1. INTRODUCTION

The important Hungarian town of Szeged is located on the banks of the River Tisza, very close to the borders with Romania and Yugoslavia, in an excellent position for international cooperation. One of the universities that participates in active cooper-ation with the University of Szeged is the University of Medicine and Pharmacy in the central Romanian town of Targu-Mures. This inter-university collaboration includes the graduate and the postgraduate education, joint research, the joint publication of scien-tific results, medical treatment of the population and management. The present article relates to the cooperation as concerns the field of pharmacy.

2. EDUCATION OF STUDENTS

The education of students at the Faculty of Pharmacy in Szeged started in 1921. During the last 80 years, more than 5000 pharmacists have completed their university pharmacy studies. Since 1964, students from abroad may also take part in the educa-tion, and since 1986, they may study pharmacy in English language courses, too. So far, 370 foreign pharmacists from 37 countries have received their pharmacy diploma in Szeged. Most of those taking part in the Hungarian course were citizens of Yugoslavia or Romania: 82 and 27 persons, respectively, while 55 and 45 students on the English course were from Greece or Jordan, respectively.

More than 50% of the pharmaceutical students of Targu-Mures can speak Hungarian. They may take part in partial education at the universities in Budapest

and Szeged. For them, it is important to carry out laboratory experiments and to prepare highlevel diploma work.

Students from Targu-Mures may spend their summer practice at the Departments of the Faculty of Pharmacy in Szeged. They generally perform scientific investi-gations and make acquaintance with the cultural and pharmaceutical background in Szeged and the county of Csongrád. They already number nearly 100.

For the best students, pharmaceutical factories such as Chinoin and G. Richter in Budapest provide possibilities for industrial practice during 4 weeks.

The Scientific Student Association organizes large-scale, nationwide and now in-ternational meetings each year in Targu-Mures, where pharmaceutical and medical stu-dents from Budapest and Szeged may also present their results. The best Hungarian students and young teachers from the Faculty in Targu-Mures may also make presenta-tions at the Rozsnyay scientific meetings in Hungary.

As concerns the postgraduate education of pharmacists in Szeged, during the past 80 years 494 pharmacists have received their Dr.Pharmacy, and 40 their PhD in the past decade. Of the foreign students, 12 have received their Dr.Pharmacy and 10 their PhD (earlier CSc); one of them has also received his DSc and 2 have become professors. Examples of the young PhD candidates who prepared excellent theses in Szeged are Ildikó Fejér (Timisoara) and Ljiljana Tasic (Beograd).

3. SCIENTIFIC COOPERATION and SCHOLARSHIPS

Cooperation between scientific researchers is also very important. Since 1990, Prof. Gyéresi in Targu-Mures has carried out joint research work on the stability and ap-plication of 1,4-dihydropyridine dye derivatives and their inclusion complexes with Judith Hohmann and Gábor Nagy at the Department of Pharmacognosy (TLC, HPLC and UV spectrometry), with György Dombi at the Department of Pharmaceutical Chemistry (nu-clear magnetic resonance) and with Géza Regdon sen. and M. Kata at the Department of Pharmaceutical Technology in Szeged.

Prof. Gyéresi also conducts joint HPLC experiments on the separation of stereo-isomers of β-receptor blockers with Ferenc Fülöp and Mária Péter at the Department of Pharmaceutical Chemistry in Szeged and with Miklós Józan, György Szász and Béla Noszál at the Department of Pharmaceutical Chemistry of Semmelweis University in Budapest. Prof. Gyéresi was recently awarded a Domus Hungarica Fellowship for 3 months.

The scholarship systems in Hungary, such as the ″Domus Hungarica Artium et Scientiarium″ of the Hungarian Academy of Sciences, ensure good possibilities for ex-perimental and bibliographical studies during periods of 1-3 months and they give help for participation in different conferences. These possibilities sometimes prove very im-portant in the preparation of the doctoral dissertations of young colleagues, as was the case for Emese Sípos, Hajnal Kelemen and Erzsébet Varga, who worked with Géza Regdon jr. and Erzsébet Csányi at the Department of Pharmaceutical Technology in Szeged.

Prof. Imre Máthé and Prof. Károly Csedő, heads of the Departments of Pharma-cognosy in Szeged and Targu-Mures, collaborate actively on topics of pharmaceutical vegetables. They work jointly in the framework of agreements between the Hungarian
and Romanian Academies of Sciences. In recent years, many young Hungarian researchers have taken part in botanical expeditions in Transylvania in Romania.

4. PARTICIPATION IN SCIENTIFIC PROGRAMMES

In the past 14 years, many experts from Targu-Mures have taken part in profes-sional programmes in Hungary, very often as invited lecturers at congresses or on visits to the University of Szeged. The most important such events included:

- The participation of Transylvanian pharmacists in the Symposium of Clinical Pharmacy in Szeged in 1994.
- The meeting of pharmacists and veterinarians in Szeged in 1996, when Prof. Gyéresi made a professional presentation.
- The annual meetings of the Society of Phytotherapy, with participation by Prof. Csedő and H. Mária Péter.
- Prof. Gyéresi lectured on the ″Separation technology of isomers of chiral drug compounds″ at the Department of Pharmaceutical Chemistry in Szeged.
- The Congressus Pharmaceuticus Hungaricus in 1993, 1999 and 2003.
- The Rozsnyay Memorial Competitions. Since 1995, lecturers have been invited from Transylvania, such as Prof. Gyéresi, Prof. Csedő, H. Mária Péter and two young participants, as guests of the Hungarian Pharmaceutical Society and the Hunga rian Chamber of Pharmacists. In 2003, we had a lecturer from Bratislava, too.
- The Ottó Clauder Memorial Competitions. Since 1996, Prof. Gyéresi has been among the invited lecturers from Transylvania.
- Participation in the symposium ″Medicines at the turn of the millenium″ in Sopron by Emese Sípos and Hajnal Kelemen.
- Participation in the History of Pharmacy meetings in Kőszeg by Prof. Gyéresi, H. Mária Péter and Hajnal Kelemen.

Many Hungarian lecturers have also taken part in the work of the meetings in Transylvania or in Targu-Mures:

- The presentation by Prof. Kata at the Faculty of Pharmacy in Targu-Mures in 1994.
- The annual meetings of the Medical and Pharmaceutical Sections of the Museum So- ciety of Transylvania since 1994, the invited Hungarian guest lecturers including Prof. Kata, Géza Regdon sen. and jr., Lajos Simon and László Tóth, all from Sze- ged, and Prof. Noszál, Zoltán Hankó and Imre Klebovich from Budapest.
- The presentation by Sándor Szabó, President of the Hungarian Chamber of Pharma- cists, at the Faculty of Pharmacy in Targu-Mures.
- Prof. Nyiredy and Klára Mikita took part as invited guests at the Romanian National Conference of Pharmacists in Targu-Mures in 1996.
- The presentation by Prof. István Erős to pharmacists at the Museum Society of Tran- sylvania in 1998.
- The presentation by Prof. Zoltán Vincze (Budapest) at the meeting of the Scientific Association of Hungarian Students in Targu-Mures in 1999.
- The presentation by Prof. György Falkay, Dean of the Faculty of Pharmacy in Szeged, in Targu-Mures in 2000.
- The participation of guest lecturers at the annual meetings of the Scientific Association of Hungarian Students at Targu-Mures: Prof. Erős and Prof. Falkay from Szeged, and Prof. Vincze from Budapest.
- Miklós Gyarmathy, an expert from the Béres Pharmaceutical Works, has held a number of very interesting scientific lectures in Transylvania.

• The Society of Hospital Pharmacists organized a 2-day meeting in Szeged and in Subotica in 1991.

Within the framework of the Hungarian Pharmaceutical Days, the Hungarian Chamber of Pharmacists received professors and students from Targu-Mures as guests in the towns of Gyula and Balatonfüred in 2001 and 2002.

5. JOINT PUBLICATIONS

As a result of joint scientific cooperation, many manuscripts have been published in excellent professional periodicals, such as Acta Pharmaceutica Hungarica, Farmacia, Gyógyszerészet (Pharmacy), Orvosi és Gyógyszerészi Szemle (Medical and Pharma-ceutical Review), Orvostudományi Értesítő (Bulletin of Medical Sciences), Revista de Medicina si Farmacie, etc. The joint publications and abstracts number more than 20. The number of joint lecture presentations is about 50.

Thin-layer chromatographic (TLC) methods are frequently used, important ana-lytical methods for the rapid control of basic drug materials. On the basis of seven sig-nificant Pharmacopoeias (the American, British, European, Germain, Hungarian, Italian and Romanian Pharmacopoeias), the authors earlier compiled a book describing the TLC analyses of 840 basic drug materials. The title: ″Thin-layer chromatographic tech-niques for quality control of pharmaceutics″, published by Prof. Gyéresi and Prof. Kata in 2000 (pp. 545).

6. REGULAR CONNECTIONS

In 1990, János Szilárd, Rector, and Emil Minker, Vice-Rector of Albert Szent-Györgyi Medical University in Szeged, paid a visit to Targu-Mures to initiate cooperation between the two universities.

In 1994, a collaboration contract was signed between the two Faculties and some Departments, too.

In 2001, the management of the Hungarian Chamber of Pharmacists visited the Faculty of Pharmacy in Targu-Mures.

In the autumn of 2001, the Dean and management of the Faculty of Pharmacy at the University of Szeged entertained a delegation from the Faculty of Pharmacy at Targu-Mures to improve and enhance the connections between the Faculties.

7. MISCELLANEOUS

Hungarian pharmaceutical authors regularly publish their manuscripts in the ac-credited journal Orvostudományi Értesítő (Bulletin of Medical Sciences), a perodical of the Museum Society of Transylvania.

To improve the education of the students in Targu-Mures, assistance amounting to one million HUFs has been provided by the Hungarian Chamber of Pharmacists.

Since 1997, one of the professors in Szeged each year offers a financial prize for the best young lecturers at the meetings of the Museum Society of Transylvania.

The Faculty of Szeged has provided the Faculty in Targu-Mures with university hand-outs, laboratory instruments, apparatus and personal computers. New hand-outs too will be handed over.

Prof. Kálmán Szendrei from Szeged, a well-known WHO expert on narcotic drugs, earlier delivered professional presentations in Transylvania. In 2001, he was

awarded an honorary doctorate by the University of Medicine and Pharmacy in Targu-Mures.

Professors from the Universities in Targu-Mures, in Novi Sad and in Szeged of-ten meet and cooperate in the framework of the World Association of Hungarian Profes-sors.

The clinical departments at the Medical Faculty of the University of Szeged also play a very important role in the medical treatment of the population, providing health care service in this geographical area. Visiting professors from Targu-Mures, including Á. Gyéresi, physicians and pharmacists often take part in the postgraduate education at the departments and clinics in Szeged.

Closing opinion: this trilateral cooperation is really international and successful, as far as possible.

EFFECT OF EXTREMELY LOW FREQUENCY ELECTROMAGNETIC FIELD ON BLOOD VESSELS OF ADENOHIPOHYSIS IN RAT

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Abstract

Potential health risk of extremely low frequency electromagnetic field [ELF-EMF] exposure are of considerable public interest. In the present study we investigated the effect of three months exposure to ELF-EMF of frequency [50 Hz] and intensity [50 μT to 500 μT] to which humans can be exposed in their home and work environments on the blood vessels morphology and number in adenohypophysis of male Wistar rats. Results of histological and stereological analysis of five micrometer thick adenohypophysal slides showed increased number and volume of blood capillaries in adenohypophysis compared to corresponding controls what can indicate influence of these fields on microcirculation

Keywords:

adenohypophysis, blood vessels, extremely low frequency electromagnetic field [ELF-EMF]

1. INTRODUCTION

Extremely low frequency [<300 Hz] electromagnetic fields [ELF-EMFs] and its biological effects and their consequences on human health are receiving increasing scientific interest and have become the subject of great public debate. The controversy has been stimulated by some epidemiological studies that have reported a relation between magnetic field and human disease [17,4,12,16]. However, so far no accepted, biologically plausible mechanism has been advanced to explain how fields interact with biological systems to yield observed in vitro responses, much less disease in an organism. Considering ELF EMFs interactions from the purely point of view several mechanisms have been proposed to account for the initial interactions with cells [8,9,1,5], but these models have been limited by their inability to account for the wide range of experimental observations.

One of the robust biological effects of ELF-EMFs is its influence on angiogenesis. From that reason this study was designed to look for possible effects of long-term exposed to ELF-EMFs at the levels found in residential, occupational and general community on rat adenohypophysal blood capillary network.

2. MATERIAL AND METHODES

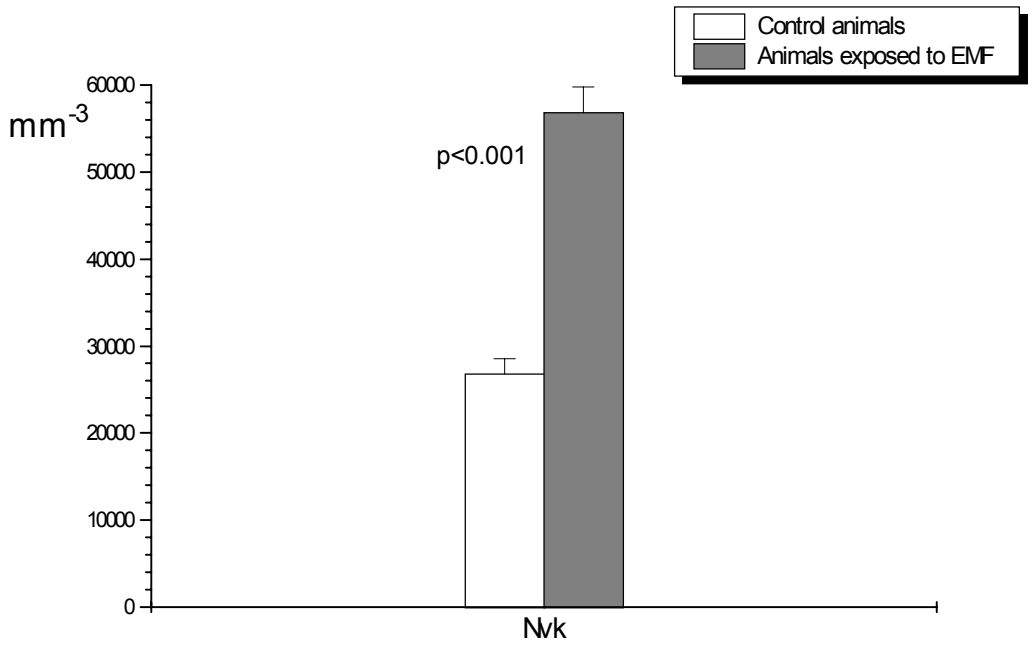
A total of 20 male Mill Hill rats were used in these experiments. All animals were maintained under controlled laboratory conditions. Ten animals were exposed to the influence of ELF-EMFs [50 Hz] for 7 hours a day, 5 days a week, beginning from 24 h after birth until the end of third month of postnatal life. Control animals were housed under identical conditions except for the ELF-EMFs.

ELF-EMF-inducing system consisted of a single coil of 2.5 mm thick wire winded in 1320 turns on wooden frame. The coil was energized from standard 220 V, 50 Hz, 16 A outlets via an autotransformer, which provided 60 V output and was used in order to reduce the electric field, measured to be less then 10 V/m anywhere in the room. The cages with animals were placed symmetrically on both sides of the coil. Along the cages, the coil produced a magnetic field of decaying intensity from 500 μT to 50 μT.

After sacrificing, the hypophyses, were removed and fixed in Romeis's solution. Paraffin-embedded glands were cut serially and stained after the method of Hurduc, and El-Etrery Tüshaus. Three adenohypophysal gland sections per animal were subjected to histological and stereological analysis. The volume density [Vvk] and numerical density of adenohypophysal blood capillaries with grid M42 were determined. The results were statistically analyzed by Student's *t*-test.

3. RESULTS

The mast prominent morphological characteristics of the adenohypophysal capillary network in rats sacrifices after 3 months of exposure to ELF-EMFs compared to the controls were increase of number of adenohypophysal blood capillary and appearance of its dilatation, especially in the center of gland. These histological findings were substantiated with the results of stereological analysis.
In animals exposed to ELF-EMFs influence numerical density of adenohypophysal capillary network was significantly [$p<0.001$] increased [Fig. 1]. At same time the volume density of adenohypophysal capillary network was, also, significantly [$p<0.01$] increased compared to the non exposed animals [Fig. 2].



"Fig. 1. Numerical density of adenohypophysal blood capillary network. Mean±SE".

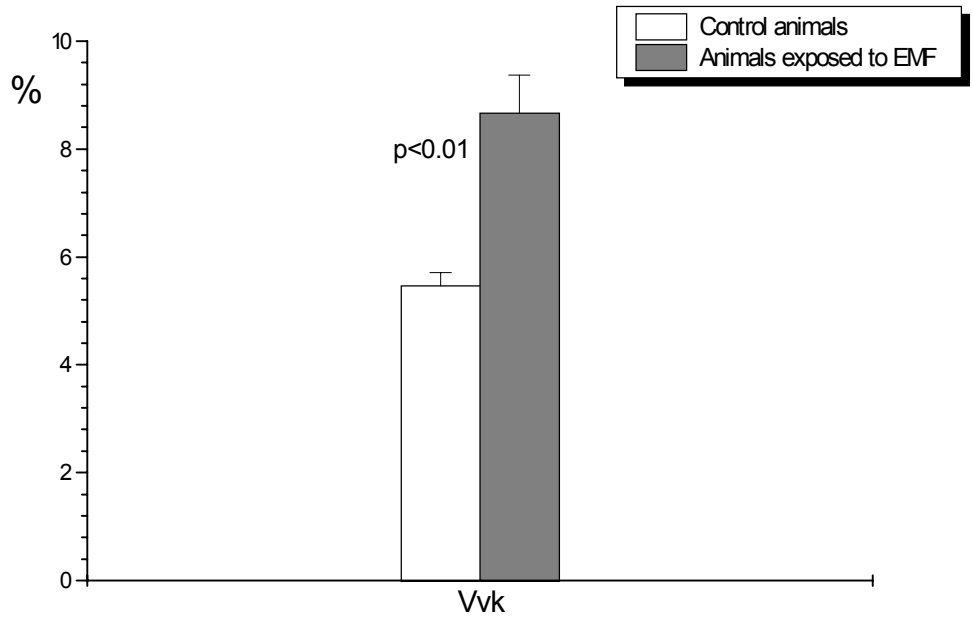


Fig. 2. Volume density [Vvk] of adenohypophysal blood capillary network. Mean±SE"

4. DISCUSSION

As the present results show, long term exposition to ELF-EMFs can change the morphology of adenohypophysal capilary network.

The available data imply that the response of blood vessels to ELF-EMFs is extremely heterogeneous with regard to the strength and frequency of the field. Beyond this divergences of the frequency scale, the use of pulsed or modulated fields raise additional problems. The contradictoriness of the results may also be due to differences in experimental design and procedures.

A mathematical model for changes in several typical blood vessels [aorta, artery, arteriole, capillary, venule, vein] under action of an alternating magnetic field has been developed [15]. It has been demonstrated that the blood serum ions interacting with this field have the largest impact in large vessels; changes in capillary parameters are small and can be caused mostly by neuronal factors and redistribution of the blood from arteries and arterioles; the venous vessels are more susceptible the influence by the EMFs as compared to the arterial ones.

Investigations of he effects of three waveforms of pulsed electromagnetic fields on blood vessel growth in the rabbit ear chamber showed that first, a pulse burst waveform, produced a significant increase, but second and third, two different single pulse waveforms, had, in contrast, no significant effect on rate of vascular growth [5].

Also effect of global system for mobile communication radiofrequency fields [RFs] on vascular permeability in the brain was studied using a purpose-designed exposure system. Shimacher et al. [2000] were show that EMF of 1.8 GHz increase permeability to sucrose of the blood-brain barrier in vitro, while 30 min of exposure to RF [900 MHz] significantly increase blood pressure [2]. Opposite of this, transcranial pulsed magnetic stimulation on blood-brain barrier no effect on its permeability [13].

Our results indicated that ELF-EMFs can increased both numerical and volume density of adenohypophysal blood capillary network. Influence of ELF-EMFs on blood vessels in some other endocrine gland we already described and discussed earlier [11]. According some other authors EMF has influence on angiogenesis and they concluded that some of observed effects of EMF on tissue healing may be mediated trough a primary effect on vascular growth [6]. Once can postulated that EMFs influence angogenesis through cell cycle of endothelial cells. It is well known that growth of cells can be controlled by the interaction of growth factors with their receptors on the plasma membrane. Such interaction may, for example, cause quiescent, somatic cells to leave G_0, traverse G1, and enter S phase, whereupon they are normally committed to at list one round of the cell cycle. Activated growth factor generate one or more primary signals that promote a sequence of metabolic events. It is possible that EMFs may stimulated this cycle on the some step of endothelial cell cycle.

From reason that, still remains difficult to correlate exposure to ELF-EMFs with health risk, once has supposed that factors other than electromagnetic waves may be involved [10]. According some authors harmful effects, if any, should therefore be induced by an indirect effect with these fields acting as a promoting, or co promoting agent rather than as an initiator. This could for example be by favoring the amount and longevity of free radicals [3] or by enhancing the effect of a chemical or physical mutagen [7]. It is, therefore, very often concluded that this quite certainly points to an absence of any major ELF-EMFs-releated health hazards, but as almost everybody is exposed to different types of ELF-EMFs uncertainties still justify further investigations.

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CHANGES IN THE D-AMINO ACID CONTENT OF SHEEP MILK RELATED TECHNOLOGIES

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Abstract

We have studied the free D-aspartic acid and free D-glutamic acid content of ewe's milk, heat-treated sheep milk at various temperatures and various products of ewe's milk. Raw ewe's milk didn't contain free D-aspartic and D-glutamic acid in remarkable amount. Whereas, all of the investigated products contained high level of investigated free D-amino acids. The free D-aspartic acid content of the products was 16,9-39,5%, while the free D-glutamic acid content was 13,3-27% in the percent of total free amino acids. The D-amino acid content of fermented milk products was higher than in the case of different cheeses.

Key words*: free D-aspartic acid, free D-glutamic acid, ewe's milk, dairy products*

INTRODUCTION

Milk and dairy products provide very good examples of the occurrence of D-amino acids in the processing of raw foods. Although a consensus has not been reached on D-amino acids, at present their negative consequences outnumber their positive effects. The presence of D-amino acids in protein reduces digestibility and affects that of other amino acids. Research to date has indicated that it is rather racemisation that is affected, first and foremost, by the pH of the substance, by heat treatment and by alkalisation time as well as by the structure of certain amino acids. The D-amino acid content of the food, we eat is determined by the original D-amino acid content of the raw material, by production methods and by microbiological processes

The D-amino acid content of the food we eat is determined by the original D-amino acid content of the raw material, by production methods and by microbiological processes. Several D-amino acid isomers may have a toxic effect; some may change the biological effect of lisinoalanine as well. On the other hand, certain D-amino acids may be useful (e.g. in pain relief), and proteins containing D-amino acids with reduced digestibility may be used, e.g. in dieting (7.)

A number of researchers have analysed the D-amino acid content of milk and various dairy products and concluded that D-amino acid content increases significantly during processing. In their study of the racemisation of free amino acids, Bada (1.) determined that at 100° C with pH between 7 and 8 the half time of

racemisation (the period during which the D/L ratio reaches 0.33) for serine is 3 days; they also found that this figure is 30 days for aspartic acid, 120 days for alanine and 300 days for isoleucine.

Payan (12.) studied the changes brought about during milk treatment by measuring the concentration of D-aspartic acid. Raw milk contained the smallest amount of D-aspartic acid (1.48%). However, this amount increased in direct proportion to the number of treatments (acidophilus milk: 2.05%; low fat milk powder: 2.15%; kefir: 2.44%; evaporated milk: 2.49%; yoghurt: 3.12%; milk-based baby formula: 4.95%).

Gandolfi (9.) analysed the effects of heat treatment and bacteria on the content of free D-amino acid in milk and D-amino acid bonded in protein. They determined that the free D-amino acid content did not grow in raw milk under the effects of pasteurisation, ultra-high pasteurisation or sterilization. In contrast, they discovered that the free D-amino acid content of the raw milk samples grew significantly when stored at 4° C and thus recommended that the figure for D-alanine content should be used in checking potential bacterial contamination in milk.

Palla (11.) found the free D-aspartic acid content of milk powder to be between 4-5% and that of D-alanine to be between 8-12%. They measured the D-alanine content of yoghurt at 64-68%, D-aspartic acid at 20-32% and free D-glutamic acid at 53-56%. These values in aged cheese were between 20-45%, 8-35% and 5-22%, respectively. They measured the free D-phenylalanine content of aged cheese as being between 2-13% and even managed to demonstrate the presence of a minimal amount of D-leucine in the aged cheese. Based on their figures, they point out that it is not those foods that are subjected to long periods of heat treatment which contain large amounts of D-amino acids but rather those that undergo microbiological fermentation.

In their study of the free D-amino acids in milk, fermented milk, lactic cheese and quarg, they (2.) determined that a significant amount of D-amino acid occurs both in raw milk and in fermented dairy products.

Csapó et al. (4., 5., 6.) studied cow's milk from healthy and mastitic udders. They determined that during milking both samples from the initial streams of milk and those from the diseased udders contained large amounts of D-Asp, D-Glu, D-Ala and D-Ile. The amount and proportion of D-amino acids in the milk from the diseased udders grew in line with the mastitest degrees. These studies prove that the first streams during milking and the milk of cows suffering from subclinical mastitis play a significant role in the amount of D-amino acid in various types of market milk produced from cow's milk .

Csapó (7.) investigated the free D-amino acid content of cheeses made using various processes, it was determined that the following free D-amino acids occurred in the following concentrations on average in the various cheeses: D-Asp at 58 □mol/100g (30.3%), D-Glu at 117 □mol/100g (15.8%) and D-Ala at 276 □mol/100g (37.2%). A larger D-amino acid content was measured in Cheddar cheese samples, which were made using species of Lactobacillus as well.

MATERIAL AND METHODS

For our experiments the ewe's milk was heat-treated and the fermented product (yoghurt) was made at the experimental dairy of the College Faculty of Food Engineering at the University of Szeged. We heat-treated the raw ewe's milk at 60,

70, 80, 90 and 120° C. The yoghurt was pasteurised at 75° C and homogenised; it was produced using a Lactobacillus bulgaricus-Streptococcus thermophilus culture.

The D-amino acid content of the freeze-dried samples was determined at the Institute of Chemistry of the Faculty of Veterinary Science at the University of Kaposvár by high performance liquid chromatography using fluorenyl-ethyl-chloroformate (3.) and by precolumn derivation using chiral reagents o-phthalaldehide/tetra-O-2,3,4,6-tetra-O-acetyl-thio-β-D-glucopyranose (8.).

RESULTS AND DISCUSSION

The ratio of D-aspartic acid and D-glutamic acid was higher in free amino acids, therefore we report only our results related free D-amino acids. The change in D-amino acid content resulting from heat treatment is illustrated in figure 1. We determined that the amount of D-aspartic acid and D-glutamic acid increases in ewe's milk as a result of heat treatment.

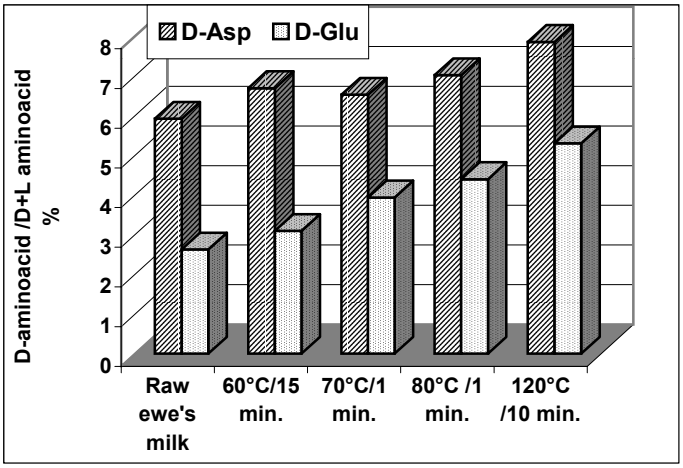

Fig. 1. *The free D-aspartic acid and D-glutamic acid content of raw ewe's milk and ewe's milk heat-treated at various temperatures (all data for total D-Asp and D-Glu in %)*

However, the heat sensitivities of the two amino acids appear to diverge. Free aspartic acid shows nearly the same D-amino acid content growth at 60 and 70° C whereas the D-amino acid content clearly grew as of 80° C. The free D-glutamic acid content growth, however, was unambiguous and continuous at each successive temperature. In the case of both amino acids, the highest free D-amino acid content resulted from the highest temperature. The divergent heat sensitivities are supported, however, by the fact that in raw milk the existing 3.3% difference in favour of D-aspartic acid decreased to 2.5% after heat treatment at 120° C, 10 min.

Based on the data, we can state that heat treatment alone does not bring about a major change in the D-aspartic acid and D-glutamic acid content of ewe's milk compared to the total given amino acid content (max.: 7.8% D-aspartic acid; 5.3% D-glutamic acid). The effect of investigated heat treatments compared to raw milk is demonstrated in Table 1.

Table 1. *The growth rate (%) of free D-Aspartic acid and Glutamic acid content resulting from various heat treatments (Value of raw ewe's milk=100%)*

	Heat treatment			
Amino acid	**60° C 15 min.**	**70° C 1 min.**	**80° C 1 min.**	**120° C 10 min.**
D-aspartic acid	113.0 %	110.2 %	119.0 %	132.6 %
D-glutamic acid	117.8 %	149.9 %	167.5 %	201.9 %

Heat treatment at 60° C for 15 minutes brought about roughly the same change for the two amino acids, but glutamic acid growth was comparatively greater at 70° C. Heat treatment at 120° C (sterilization) effected a 32% increase in D-aspartic acid content while D-glutamic acid content grew by almost 102% (roughly double). Based on the findings of heat treatments at 70 and 80° C, we can state that a temperature increase of 1° C results in an appr. 0.9% increase in D-aspartic acid content and an appr. 1.7% increase in D-glutamic acid content. For glutamic acid, the speed with which D-enantiomers occurred caused by the same degree of temperature increase was double that of aspartic acid.

The data in the table (1.), therefore, prove that when subjected to heat treatment under the same conditions glutamic acid exhibits a greater tendency for racemisation. In this case the D-enantiomer occurs more rapidly and in larger amounts than in the case of aspartic acid. The higher D-aspartic acid content of raw milk suggests, however, that the microflora prevailing in the udder and/or grown in the milk during cold storage have a greater effect on aspartic acid.

Figure 2 demonstrates the findings of our analyses of D-amino acid content in traditionally aged ewe cheese, fused cheese (Kashkaval), cream white cheese made by ultrafiltration and the fermented product (yoghurt).

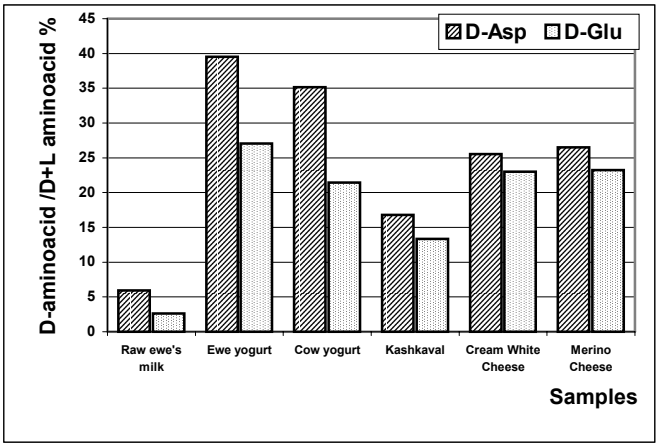

Figure 2. *The free D-aspartic acid and D-glutamic acid content of raw ewe's milk and certain dairy products (all data for total free D-Asp and D-Glu in %)*

We can state that all these products contain a significantly higher proportion of D-enantiomers than raw ewe's milk. Our findings therefore reinforce conclusions in the literature according to which fermentation with cultures greatly increases the D-amino acid content in dairy products. Values and ratio of D-aspartic acid and D-glutamic acid is represented in Table 2.

***Table 2**. The ratio of free D-aspartic acid and free D-glutamic acid in raw ewe's milk and certain products of ewe's milk*

Samples	D-Asp/D-Glu
Raw sheep milk	2.26
Yoghurt from sheep milk	1.46
Yoghurt (from cow's milk)	1.64
Kashkaval	1.26
Cream white cheese	1.11
Merino cheese	1.14

Of the two amino acids, a higher D-aspartic acid content and a lower D-glutamic acid content was found in all the dairy products. D-amino acid content was roughly the same for traditionally aged cheese (merino) and acid rennet cream white cheese. The lower values for Kashkaval cheese may have resulted from the heat effect of soaking in warm brine as well as the lower water activity of the cheese.

The yoghurts contained significantly more D-amino acid than the cheeses. This may be a result of the higher CFU value and the more intensive bacterial activity. Interestingly, the yoghurts representing a pH value of appr. 4.4 exhibited a significantly higher D-Asp/D-Glu ratio than the cheeses (at 1.11-1.26). This ratio is 1.46 for ewe yoghurt and 1.64 for yoghurt made from cow's milk. Also of interest is the fact that the D-Asp/D-Glu ratio is greater for yoghurt made from cow's milk than for ewe yoghurt. However, as the two products were not made under identical conditions we can offer no explanation for this at present.

At the same time, ewe yoghurt has a significantly higher D-amino acid content, which can be explained in part by the fact that the total number of microbes is a great deal higher in ewe's bulk milk than in cow's milk. It may also be concluded that the highest D-Asp/D-Glu ratio is brought about by the natural, or common, micro flora in raw ewe's milk, whereas this value for one of the products made from ewe's milk does not even approach 1.5. In other words, the few cultures commonly used to make dairy products from ewe's milk bring about a D-Asp/D-Glu ratio of between 1.1 and 1.5.

CONCLUSION

Many have studied the presence of D-amino acids in cow's milk and the products of cow's milk. However, we have found no research concerning ewe's milk. We have therefore studied the D-amino acid content of ewe's milk, ewe's milk heat-treated at various temperatures and various products of ewe's milk. According to our findings, raw ewe's milk does not have a high D-aspartic acid (5.92%) and D-glutamic acid (2.62%) content.

Heat treatment brings about no meaningful change in the D-amino acid content of ewe's milk. In the case of the strongest heat treatment D-aspartic acid content increased to 7,85 % and the D—glutamic acid content increased to 5,30 %. In the case of the common pasteurisation of milk we can state that a temperature increase of 1° C results in an appr. 0.9% increase in D-aspartic acid content and an appr. 1.7% increase in D-glutamic acid content.

However, a significant change was detected in the D-amino acid content of every product investigated. The products contained 16.8-39.5% D-aspartic acid and 13.3-27.0% D-glutamic acid. We measured the highest D-amino acid content in those products were made by lactic acid fermentation (yoghurts).

These findings and those of the analyses of the various heat treatments do not enable us to make any generalisations at this point. They call for further study, in particular on temperature and holding time as well as to gain a better understanding of the precise effects of certain cultures and even individual species of bacteria in order to be able to maintain the D-amino acid content at an acceptably low level.

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THE FREQUENCY OF VARICOUSE DISEASE IN SPECIALIZED AMBULATORY UNIT AND FAMILY DOCTORS PRACTICE

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ABSTRACT:

In the last years the ambulatory surgery has developed in Timisoara. The ambulatory practice shows us a higher frequency of varicouse disease, so between 30 and 40% of patients treated ambulatory suffered of varicouse disease. In these cases we have performed ambulatory flebectomy Muller, thrombectomy and sklerotherapy.

The postoperative evolution was very good and encourages us to develop this surgical technique. The phlebologic patients represent an important part of activity for the family doctors. Even with the Romanian costs in medicine, the costs of venous disease are important. In conclusion we must prevent the varicoase disease by all means because the treatment is expensive.

KEYWORDS

varicouse disease, flebectomy

1. INTRODUCTION

The changes brought by the last years of practice in specialized units led to a modern approach to the ambulatory surgical treatment of patients whit venous problems.

In this paper we share our experience in the field of varicous disease ambulatory surgery, presents aspects regarding the surgical treatment of varicose veins in specialized ambulatory units, by performing Muller phlebectomy and/or sclerotherapy.

In our town there are 11 private Clinics and 4 Policlinics in witch the surgeons of Timisoara can performed ambulatory techniques of phlebology and proctology surgery.

2. METODS

Over the last 5 years we have treated a number of patients with a varicous disease. We have performed Muller minimal phlebotomy (in 16 of the cases) or phlebectomy of the thrombosed varicosis (in 21 of the cases), according to the clinical findings, and the evolutive stage of the disease.

In addition to the flebectomy, in 22 cases we also performed catgut endovenous inclusion, according to the Brinzeu procedure.

Local anesthesia has been used in all the interventions.

We have performed sclerotherapy for reticular varices and for the recidive of the varices after saphenectomy in 50 cases.

3. RESULTS

Few post-surgical complications appeared, e.g. small haematoms developed in 5 of the cases, but they didn't require surgical reintervention, as we solved the problem by medical care.

The prognosis after five years is good, with no clinical symtomatology and minimal scars.

In all cases we also took in consideration the patient’s desire for an aesthetic outcome.

4. CONCLUSIONS

The ambulatory venous surgery is well accepted in all cases, being less expensive that hospital surgery.

Surgical ethics imposed that we take in consideration, first of all, the functional result, both in establishing the surgical technique and the treatment protocol.

The aesthetic results were very good in all cases of varicous disease.

In selected cases is a very good method of therapy, the best results being obtained using associated methods of varicous therapy.

This approach is a new alternative for varicous disease surgery and hemorroidal disease surgery in our country. The good results obtained encourage us to develop the ambulatory surgery.

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ANTIBIOTICS AS AN ECOLOGICAL FACTOR

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ABSTRACT

Uncontrolled using of antibiotic in one geographic area for therapy, prophylaxis, animals' growth promotion and food preservation, have unforeseeable consequences. Antibiotics act as an ecological factor; eliminate susceptible and favor resistant species and strains from the microecologica ecosystems of human and animal organisms, which form commensals and opportunistic pathogens. This selective pressure has consequences such as complication of therapy after infection by resistant strains, disturbing balance of microbial communities and spreading the resistant bacteria and their genes worldwide.

The aim of our study was to test antibiotic susceptibility of nonpathogenic and pathogenic bacterial strains from genus Enterobacteriaceae (E.coli and Salmonella sp.), isolated from different specimens. The results show very high percent of resistant strains to the most commonly used antibiotics (penicillin, ampicillin, tetracycline, erythromycin, etc.).

We also discovered that antibiotics have mutagenic effect to bacteria, because strains treated with low concentration of amoxicillin and penicillin in laboratory, show significant level of increasing the resistance to other antibiotics. Our results show that antibiotics are strong ecological factor that has influence on animals and people by changing characteristic of microbial communities and whole life of ecosystems.

KEYWORDS: *resistance, antibiotics, enterobacteria*

1. INTRODUCTION

The great number of studies that engage in problem of antibiotic resistance of human pathogenic bacteria has opened many questions about usage and role of antibiotic in agriculture and medicine. Main reason for phenomenon of resistance is inadequate use of antibiotics. Scientist generally agree with opinion that bacterial resistance is not only result of antibiotics usage in therapeutic purposes, but also in other fields, especially in agriculture.

Usage of antibiotics as an additive in feed, for prophylaxis and growth promotion, has had impact on spreading of resistance. There are a lot of evidences that spreading of antibiotics resistance is connected with commensal bacteria, especially with commensals of gastrointestinal tract (7). Antibiotics act as an ecological factor; eliminate susceptible and favour resistant species and strains from

the micro ecological ecosystems of human and animal organisms, which form commensals (non-pathogenic bacteria) and opportunistic pathogens. So, intestinal micro flora is carrier of resistance genes to many antibiotics and these bacteria reach into milk, meat, water and other mediums by fecal contamination. According to this, intestinal bacteria are one of the reservoirs of resistance genes and they are able to transmit these genes to sensitive pathogenic bacteria.

Quantity of antibiotics that are used in the world decrease every day, and one half is produced for human usage.. Bacteria from Enterobacteriaceae in animal gastrointestinal tract are especially exposed to antibiotics, because this therapeutics are being added into water and food. Also, these bacteria, when expire by feces or urine in environment, could be exposed to antibiotics taht are excreted in their active form from body and could cumulate in environment (2, 7). For example, during fish feeding has been estimated that 70-80% of antibiotics used for therapy of fish, could be detected in the water sediment. About $25x10^6$ kilograms of antibiotics are used every year for prevention infectious diseases and promotion of animal growth. In European Union, 3000 tones per year are used by veterinarian for therapy. In the US this quantity is about 8.500-11.200 tones (3). One part of these antibiotics is degraded in intestinal tract, but other is excreted in environment.

Because of this disturbing situation we examined resistance of strains of *E. coli* and *Salmonella typhimurium and S. enteritidis* isolated from different speciment. Also, we examined changes in sensitivity of some strains when these are exposed to low doses of antibiotics.

2. MATERIALS AND METHODS

Strains of *S. enteritidis*, *S. typhimurium* and *E. coli* were isolated from different materials (feed, animal feces, pigs, poultry and human infants) during routine bacteriological control. For isolation and subcultivation of test strains we used Endo agar, brilliant green agar, MacConkey agar, and nutrient broth. Identification of strains was performed by standard bacteriological methods.

Test strains of *S. enteritidis* and *E. coli* have been isolated from feed and feces. Strains were inoculated in nutrient broth and incubated at 37^0C 24 hours. After this period all strains were tested by Kirby Beyer method. One milliliter of culture suspensions were spread on Muller Hinton plate. Susceptibility of strains was tested to penicillin, streptomycin, ampicillin, neomycin, erythromycin, tetracycline, cephalexin, chloramphenicole, gentamicin, and linkomycin.

Susceptibility of two test strains of *E. coli* and *S. enteritidis* was examined in vitro before and after incubation with low doses of antibiotics. Selected strains of *S. enteritidis* and *E. coli* were suspended in 1% peptone water and physiological saline, with dense of 8 MC. In these suspensions was added low concentration of penicillin or amoxicillin, they were stored at 37^0C 48 h and than tasted to antibiotic susceptibility. The experiment was repeated four times. Susceptibility of strains was detected by Kirby Beyer method.

3. RESULTS AND DISCUSSION

Results of susceptibility of *E. coli, S. entritidis* and *S. tiphimurium* are showed in table 1 and 2.

Table 1. *Resistance (%) of S.enteritidis and S.typhmurium isolated from different animal speciments*

	S.enteritidis			*S.typhmurium*		
Terapeutics	Poultry	Feed	Animals' feces	Poultry	Pigs	Feed
penicilin	60,15	82,14	60,12	69,14	98,18	60,12
streptomicin	50,6	49,9	52,11	49,2	45,8	50,00
ampicillin	75,16	75,15	88,18	75,16	78,17	75,00
neomicin	82,17	74,14	89,18	72,14	65,13	76,16
erythromycin	100	90,19	86,18	82,17	98,19	88,17
tetracycline	100	78,17	76,16	100,00	98,19	75,00
cephalexin	62,14	64,13	50,00	51,11	50,00	62,12
chloramphenicol	90,19	88,18	99,19	60,12	75,00	74,14
gentamicin	43,7	39,6	25,5	49,95	49,99	39,70
linkomicin	100,0	99,19	100,00	88,18	78,16	99,19

Table 2. *Resistance (%) of E. coli isolated from diifferent speciments*

Terapeutics	Poultry	Pigs	Feed	Animals' feces
penicilin	69,14	89,18	50,00	79,80
streptomicin	60,12	44,90	71,14	59,11
ampicillin	98,18	77,72	65,13	77,17
neomicin	67,13	68,17	50,00	62,12
erythromycin	98,19	99,19	91,18	98,19
tetracycline	98,19	75,00	74,14	81,17
cephalexin	26,00	49,90	45,80	25,00
chloramphenicol	88,18	98,29	76,00	99,00
gentamicin	49,20	47,35	49,00	38,00
linkomicin	78,17	88,25	76,16	77,17

Our investigation showed high percent of resistant strains from animal, food and feces and can be explained by prolonged usage of these antibiotics on animal farms. This opinion is supported by studies of geneticists that showed that the same resistance genes can be found in the bacteria of animals and humans (12). Sunde et al. (10) was examined about 1200 strains of *E. coli* from health and ill pigs and found that the least resistance for an antibiotic was 100%, and that the most resistance was to streptomycin, sulfonamide and tetracycline. Similar results of *E. coli, Haemophylus influenzae* and *S.aureus* resistance was obtained from volunteers (4). Our experiment showed that 44,90 - 71,4% of tested isolates was resistant to streptomycin. Our results are in correlation whit these findings. Resistance of *S. tiphimirium*, *S. enteritidis* and *E. coli* isolated from human infants is shown in table 3.

Table 3. *Resistance of S.enteritidis, S. typhimurium i E.coli isolated from human infants*

antibiotics	***S.enteritidis***	***S.typhimurium***	***E.coli***
ampicillin	3,2	75	16,7
cephalexin	15,9	32,9	33,3
cefotaxime	0	0	16,7
gentamicin	0	0,9	0,9
trimetoprim	1,8	1,8	2
norfloxacin	0	0	0
ciprofloxacin	0	0	0

The low percent of resistant strains from human infants is probably resulting from infection with strains that are highly sensitive and that could not infect adults that use some of these antibiotics time to time. Some of these strains are commensal and opportunistic pathogens, especially to infants, and because there is no time to develop resistance they are still sensitive. Our opinion is that during the life, some of these bacteria, persisting as commensal, have a great chance to develop resistance or to gain resistance genes from other bacteria that will reach to gastrointestinal tract. Also, antibiotics used in this test are less common used.

Results of susceptibility of S. enteritidis and *E. coli* strains before and after exposure to low doses of antibiotics are showed in table 4 and 5.

Table 4. *Susceptibility of S. enteritidis stored in 1% peptone water and physiological saline with antibiotics*

Terapeutics	**1% peptone water**			**Physiological saline**		
Antibiotics	**control**	**penicilin**	**amoxicillin**	**control**	**penicillin**	**amoxicillin**
Penicillin	I	R	R	I	R	R
Streptomycin	S	R	I	S	I	R
Ampicillin	I	I	I	I	I	I
Neomycin	S	R	R	S	R	R
Erythromycin	I	I	I	I	R	R
tetracyckine	I	R	R	R	R	R
Cefalexin	I	I	I	I	I	I
Chloramphenicol	S	I	S	S	I	S
Gentamicin	I	I	I	I	I	I
Linkomycin	R	R	R	R	R	R

R- *resistant*, I- *intermediary susceptible* S- *susceptible*

Tested strains showed different susceptibility to used therapeutics. Resistance to linkomycin was detected before and after exposure to low doses of antibiotics. Supplementation of low doses of penicillin did not have any effect to change resistance of tested strain to penicillin, cephalexin, erythromycin and tetracycline. These results can be explained with fact that these four antibiotics are wide used in veterinarian medicine. Amoxicillin, added in physiological saline had effect on change of resistance to erythromycin and tetracycline. Resistance to erythromycin and tetracycline can be explained with their frequent usage to prevention and therapy of animals' infectious diseases from our area. Susceptibility of strains to chloramphenicol decreased after supplementation of penicillin. Susceptibility of strains stored in 1% peptone water was considerably greater to streptomycin and gentamicin than susceptibility of strains stored in the physiological saline.

Table 5. *Susceptibility of E. Coli strains stored in 1% peptone water and and physiological saline with antibiotics*

Terapeutics	**1% peptone water**			**Physiological saline**		
Antibiotics	**control**	**penicilin**	**amoxicillin**	**control**	**penicillin**	**amoxicillin**
Penicillin	R	R	I	R	R	R
Streptomycin	S	R	I	S	I	S
Ampicillin	I	I	I	I	R	R
Neomycin	I	I	I	I	R	R
Erythromycin	I	R	R	R	R	R
tetracyckine	I	R	R	R	R	R
Cefalexin	R	R	I	I	R	I
Chloramphenicol	S	S	S	S	I	S
Gentamicin	I	R	I	S	I	I
Linkomycin	R	R	R	R	R	R

R- *resistant*, I- *intermediary susceptible* S- *susceptible*

All tested *E. coli* strains were resistant to linkomycine. Strains from both medium have showed lower susceptibility to amoxicillin, erythromycin, caephalexin, and gentamicin. This susceptibility was not changed after exposure of test strains to low doses of antibiotics, except when they were treated with erythromycin. Streptomycin and neomycin showed the most similar effects to change resistance pattern of tested strains. These results can be consequence of their common origin (they are product of Streptomycetes). According to Egorov (5), effect of neomycine to many bacteria is greater. So, these results confirm similarity of this therapeutics.

Resistance, impact of antimicrobial additives, as well as monitoring of change of susceptible bacteria to chemotherapeutics have been interesting to scientist for a long period (9,12). Other studies also support our results and statements. Kelley et al. (6) tested resistance of enterobacteria and *P. aeruginosa* isolated from cover for chicken treated with 12 different antibiotics. Results of this experiment showed that strains were multi resistant to them. Bailey et al.(1)discovered that different antimicrobial additives in different combination had impact on decreasing of number of *Salmonella sp.* in chicken caecum. Tassios at al. (11) monitored decreasing in number of infections caused by *S. enteritidis* during seven years in Greek. They found that strains resistant to ampicillin and doxycyclin showed cross resistance whit therapeutics from other classes, especially sulphonamide and streptomycin. Examination of other bacteria also showed that low doses of antibiotics have impact on resistance occurrence. For example, after usage of subinihibitory doses of linkocyn for treatment diseases caused by *Staphilococcus aureus* in cow, isolates from milk show resistance and change in morphology (8).

4. CONCLUSION

According to our results, it is obvious that resistance problem is ecological. In the competition between resistant and sensitive bacteria, antibiotics act as an ecological factor that encouraged growth of resistant strains. Also, antibiotics have a mutagenic effect to bacteria, because strains treated with low concentration of amoxicillin and penicillin in laboratory, show significant level of increasing the resistance to other antibiotics.

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THERAPEUTICALLY COMMUNITY. FROM HISTORY TO THE THEORETICALLY MODEL
COMUNITATEA TERAPEUTICA. DE LA ISTORIE LA MODELUL TEORETIC

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Comunitatea terapeutica este o modalitate de tratament pentru abuzul de droguri ce cunoaste o larga acceptare in momentul de fata. Aparuta in urma cu mai mult de 40 de ani dintr-o arie neprofesionista doar recent a castigat un spatiu in lumea academica si in cea a cercetarii. Valoarea sa sociala este contundenta si indiscutabila iar asta se face evident observand marea oferta de programe terapeutice pentru abuzul de droguri care adera la principiile C.T.

Acest articol are doua parti: in prima vom trece in revista istoria C.T. iar in a doua vom expune o definire a modelului C.T.

De unde apar CT?

Intr-un studiu 1977, dr. Frederick Glaser a mers pe urmele C.T. pana spre originile sale cele mai indepartate. In acest studiu este citat Phito Judaeus care in anul 25 i.e.n. scrie despre activitatile unei comunitati care locuia in Egipt, aproape de Alexandria. De asemenea, povesteste ca, intr-un Rollo din Marea Moarta, numit Regula Comunitatii sau Manualul de disciplina se descriu problemele pentru care oamenii intrau in respectivele Comunitati si care, spre surprinderea noastra, aproape se suprapun pe ceea ce vedem astazi ca mod de viata, cultura si suferinta a sutelor de tineri dependenti ce sunt asistati in C.T. Inclusiv codurile (regulile) de convietuire ne aduc aminte de cele ce sunt valabile astazi in noile C.T.

In acest fel dr. Glaser concluzioneaza ca, daca dam credit istoriei, putem deduce ca CT au doua milenii vechime si puternice radacini religioase.

Daca facem un drum retrospectiv si plecam de la C.T., firul conductor ne va face sa trecem mai intai prin Alcoolicii Anonimi (AA), apoi prin grupul Oxford si prin Asociatia Crestina a Tinerilor, ne vom interna apoi in Reforma Protestanta si in anumite grupuri primitive crestine pentru a ajunge la Esenios, despre care se vorbeste in Rollo din Marea Moarta descris mai sus.

Pentru a gasi C.T. specifica dependentilor de droguri trebuie, fara indoiala, sa ajungem la jumatatea secolului XX. Aceasta pentru ca, desi este documentat faptul ca omul dintodeauna a consumat substante psihoactive (SPA) ce i-au favorizat modificari in starea sa de constiinta, folosirea lor era limitata la anumite contexte, in principal anumite ritualuri religioase, festivitati, privilegii legate de caste, pregatiri pentru razboaie etc, si nu exista dovezi ale unui consum masiv care sa depaseasca aceste limite si care sa preocupe autoritatile sau care sa pretinda vreun tip de interventie din partea societatii.

Localizandu-ne in lumea occidentala, etapa in care aceasta iesea din lunga noapte a Evului Mediu, cand se facea simtita greutatea Inchizitiei, se stie ca consumul nereglementat de SPA, cel care nu era pentru motive medicale, era intotdeauna cel putin condamnat, iar consumatorii erau considerati posedati, vinovati de ceva, cu un singur destin final posibil, inchisoarea, ospiciul sau rugul.

Incepand secolul al XIX si pana in 1950 a avut loc un proces gradual pe parcursul caruia consumatorii de SPA nu au mai fost considerati pacatosi sau pleava societatii ci au fost inclusi printre cetatenii respectuosi cararoa societatea trebuia sa le ofere un raspuns terapeutic. Aceasta evolutie a conceptului de consumator de SPA a avut loc datorita noului loc pe care l-a dobandit religia dar si progreselor stiintei.

In acelasi timp, in aceasta perioada 1800-1950, si in campul Sanatatii Mintale, s-a renuntat la considerarea bolnavului psihic ca persoana care sufera de fenomene supranaturale si care trebuie inchis in azile si inchisori (Pinel a scos lanturile „nebunilor” in 1793 in Paris) si a fost considerat persoana bolnava caruia societatea trebuie sa-i ofere un raspuns terapeutic.

Influenta Psihanalizei in aceasta schimbare a viziunii bolnavului psihic a fost determinanta.

Aceasta coincidenta temporala a viziunii fata de consumatorul de droguri si fata de bolnavul psihic, conduce in mod natural la ideea ca ambele vor evolutiona in mod concomitent si isi vor suma aporturile si vor da raspunsri comune: dar aceasta nu s-a intamplat, iar tratamentul dependentilor nu a beneficiat de noile abordari si instrumente pe care campul Sanatatii Mintale le – a adus tratamentului nevrozelor sau psihozelor.

Ne spune Nyswander in 1956 aratandu-si descurajarea lucrand cu dependenti de opiacee in Centrul Lexington pentru Tratamentul Narcoticilor: „intr-o cautare temeinica a bibliografiei nu s-a putut gasi o singura informatie in care psihoterapia a permis unui numar semnificativ de pacienti dependenti sa revina in comunitate traind o viata ca un idivid normal”.

Explicatia este destul de simpla: dependentii au demonstrat a fi ireductibili si nu au raspuns tratamentelor existente in acel moment. Si chiar daca nu a existat o declaratie formala, faptic dependentii au fost considerati „intratabili”, cel putin cu instrumentele existente in acel moment.

In anii 40-50 a avut loc o interesanta evolutie: AA, ce erau o miscare de autoajutor, o retea de sprijin informala, au primit recunoasterea ca programe de sprijin in tratamentul alcoolicilor; a fost o schimbare de calificare interesanta si importanta in acelasi timp ce a determinat ceea ce a urmat.

In anii 50 se intra din plin in istoria moderna a CT, prin mainile initiatorului lor **Charles E. Dederich**, ce s-a nascut in 1914 in Ohio, SUA. Acesta era un director de succes. In anii 20 fusese un bautor excesiv de alcool si spre sfarsitul acestei perioade devenise in mod clar un alcoolic. Cum se intampla cu majoritatea alcoolicilor, curand si-a pierdut slujba si a ajuns sa-si petreaca timpul inchis in casa sa din Los Angeles, intr-un mod simplu, band. Catre 1957, situatia devenisa atat de extrema ca sotia sa l-a dat afara din casa. Incepand cu acea zi , Dederich a contactat AA si a reusit sa-si recastige sobrietatea. A inceput sa asiste la grupuri, participa la diverse grupuri in aceeasi zi. Incetul cu incetul a inceput sa vorbeasca in grupuri si atat a vorbit incat au ajuns sa-i ceara sa taca din gura, ajungandu-se chiar la situatia de a fi eliminat din grup, lucru pe care el nu putea sa il accepte.

Aceasta caracteristica a sa – necesitatea de a vorbi, de a fi protagonistul – a fost decisiva pebtru ceea ce a urmat: a creat un gruop de AA in casa sa din los Angekles. Dupa cum povesteste Naza Abiter: CD participa la AA dar isi dorea o abordare mai provocatoare si mai interactivapentru obtinerea abstinentei. A inceput sa gazduiasca in casa sa grupuri de AA apoi de heroinomani si observa ca acestia stimulau discutiile. Imediat – din motive economice – heroinomanii in recuperare au inceput sa traiasca impreuna: astfel s-a creat prima CT. Dederich a observat ca impotriva tutror prezicerilor, heroinomaniise puteau mentin sobrii pe perioade ce erau din ce in ce mai lungi. Din amvon, ii stimula sa-si prelungeasca perioadele de abstinenta si ii pedepsea cand recadeau. Astfel, in acesta maniera aproape intamplatoare, Dederich a demonstrat ca prin acest tip diferit de organizare sociala ce incepuse sa se autodenumeasca Synanon, se putea obtine abstinenta la heroina fara interventii medicale, raspuns pe care societate din exterior nu il avea si mai mult il cauta si il aprecia.

Senatorul Thomas J. Dodd in declaratia sa in fata senatului SUA din septembrie 1962 spunea: ingredientul principal al Synanon este tesatura inchisa a comunitatii sau clima sociala, tip familial, unde drogodependentii se ajuta unul pe altul pentru a face fata vietii intr-o alta maniera. In Synanon ei gasesc o familie, un grup uman, o societate unde fiecare individ poate trai ca un membru al comunitatii in loc de a trai ca un pacient sau un prizonier. Este acest mediu protejat, acest tip de atmosfera familiala, ceea ce face ca, cu fiecare zi, sa fie recunoscut ca necesar pentru stabilitatea emotionala a fiintei umane.

Anii 60 au marcat inceputul unui lung drum de cautare, elaborare, construire a C.T.; aceste prime grupuri (dintre care am remarca Daytop) s-au transformat in piatra de temelie a C.T. ca metoda de tratament. Faptul ca pentru prima data s-a avut succes in recuperarea dependentilor de droguri, le-au situat intr-o situatie paradigmatica. Adevarul este ca de

la 2-4% din reusitele de pana atunci s-a ajuns la 40-60 % in C.T. Sa amintim ca in acel moment, multi dintre rezidentii din Synanon si bineinteles cei care le conduceau - care erau rezidenti recuperati – trecusera prin lungi tratamente de psihoterapie frustrante, fare nici un rezultat. Aceasta a condus la neintroducerea psihologiei in miscarile de autoajutor; si mai mult, in unele cazuri sa ajunga sa fie contraindicata si in opozitie cu activitatile de recuperare, si toate acestea datorita experientei anterioare prin care au trecut unii dintre ei. Bresa dintre Sanatatea Mintala si tratamentele pentru consumatorii de droguri a continuat sa se adanceasca in anii 70. Recent, la jumatatea anilor 80, recunoscand rezultatele obtinute in tratamentul dependentilor de droguri, s-au initiat primele miscari de apropiere dinspre partea Sanatatii Mintale catre cel al Autoajutorului in general, cautand - probabil - sa extinda catre alte patologii tehnicile sale si strategiile gestaltice, cognitive, conductiste, de confruntare, sistemice etc.

Aceasta miscare s-a consolidat in anii 90 iar profesionistii au facut diverse stagii (mai mult sau mai putin prelungite) in C.T., s-au integrat in echipele lor, au invatat din autoajutor si au reusit sa adapteze tehnicile lor; din partea C.T., operatorii lor, la randul lor, au valorificat input-ul ce le venea din partea profesionistilor. Insa integrarea reala intre ambele campuri s-a realizat odata cu 'epidemia' abuzului de cocaina, deoarece puternica sa componenta psi si regimul ambulator i-a obligat sa lucreze impreuna, daca se doreau reusite terapeutice; in asa fel incat putem asigura ca astazi bresa intre cele doua practici este de domeniul istoriei si ca nu exista C.T. care sa nu aiba in componenta sa operatori, voluntari si profesionisti din diverse ramuri academice.

Dupa aceasta scurta si fugara trecere prin istoria C.T., vom prezenta in continuare unele aspecte teoretice despre modelul C.T., subliniind ca fiecare va continua sa faca interpretarea sa particulara.

Aspecte teoretice ale Modelulului C.T.

Dupa cum afirma **George de Leon** in diferite lucrari ale sale, elementul distinctiv al C.T., ceea ce il diferentiaza de alte modalitati de tratament, este folosirea deliberata si intentionata a comunitatii ca metoda primara pentru facilitarea schimbarii psihologice si sociologice in indivizi.

In C.T., terapeutul, metoda, maestrul, este comunitatea.

Folosirea comunitatii ca metoda implica ca un individ sa vrea sa se schimbe si sa invete mai mult prin intermediul apropierii sale cu altii ce se afla implicati in aceeasi lupta: din aceasta perspectiva perceptia de comunitate este fundamentala pentru participanti, staff, prefesionisti, voluntari.

Astfel, comunitatea nu este un loc unde se gasesc dependentii paziti si unde merg prefesionistii pentru a face terapie; comunitatea nu este un loc unde dependentii isi petrec ziua departe de tentatiile strazii, asteptand grupul urmator. Comunitatea reprezinta un mediu social compus din operatori, profesionisti, directori care, fiind un model de schimbare

personala de succes sau simple modele de viata, servesc de ghid in procesul de recuperare. Toate activitatile din C.T. sunt elaborate in scopul producerii schimbari terapeutice si educationale la participanti si toti participantii sunt mediatori ai acestor schimbari terapeutice si educationale.

Din perspectiva teoretica C.T. contine un concept original si propriu despre:

1. Abuzul de droguri
2. Rezident
3. Viata sanatoasa
4. Recuperare

1. Despre abuzul de droguri

C.T. intelege abuzul de droguri ca o problema a oricarei persoane, unde adictia este o componenta, nu esenta.

Cu privire la efecte, pentru C.T., odata instalat abuzul, acesta va avea impact asupra uneia sau tuturor ariilor de functionare astfel ca putem gasi distorsiuni in plan afectiv, cognitiv, comportamental, in caracter, probleme medicale si psihologice; valorile sunt confuze, nu exista sau sunt antisociale. In mod frecvent exista deficiente in abilitatile necesare pentru a functiona la un loc de munca.

Cu privire la cauze, abuzul de substante este vazut ca un comportament cu multiple cauze determinante, unde dependenta psihologica este secundara unei game largi de circumstante care influenteaza si care preiau controlul asupra comportamentelor subiectului si care fac ca, in mod invariabil, problemele si situatiile ce sunt asociate cu un anumit disconfort sa se transforme in semnale pentru a se recurge la droguri. De aceea se considera abuzul de droguri ca o problema a oricarei persoane.

2. Despre rezident

Exista doua mari grupuri de persoane care cer ajutor intr-o C.T. Primul este format din persoane care nu au avut niciodata un stil de viata satisfacator, conventional; valorile generale ale culturii lipsesc sau nu sunt luate in considerare; provin dintr-un mediu unde abuzul de droguri este mai mult un raspuns social decat o problema personala.

Pentru acest grup experienta dintr-o C.T. poate fi numita 'abilitare' adica posibilitatea de a apartine unei societati organizate si de a dezvolta un mod de viata productiv si conventional, pentru prima data in viata lor.

Pentru al doilea grup abuzul de droguri este o problema psihopatologica, de personalitate sau de instabilitate emotionala. Pentru ei termenul este 'reabilitare', pentru ca este vorba de reintoarcerea la un stil de viata precedent, cunoscut, dar lasat deoparte temporar.

Fara indoiala, cu toate diferentele de provenienta, odata instalat consumul si dependenta, cele doua grupuri prezinta similitudini: au probleme de socializare, in atitudinile lor cognitive si emotionale, si probleme psihologice ce pun in evidenta imaturitate, autostima scazuta, tulburari de caracter si de comportament si caracteristici antisociale.

In C.T. nu se dezbate daca toate acestea sunt cauza sau consecinta consumului: pentru C.T. trebuie doar schimbate pentru a obtine o recuperare stabila.

De aceea, rezidentii vor urma in mod fundamental acelasi regim general: diferentele individuale sunt recunoscute in planuri specifice ale tratamentului care modifica anumite etape, dar nu cursul general al experientei in C.T.

3. Despre ce este un stil de viata sanatos

Obiectivul general al C.T. esta ca rezidentii sai sa ajunga sa duca un stil de viata sanatos. Pentru a-l obtine, rezidentul este orientat in mod specific prin intermediul autoajutorului / ajutorului mutual (self help) sa-si asume responsabilitati personale.

Se pune emfaza pe prezent (aici si acum) ca opozant al trecutului; comportamentele trecute sunt explorate doar pentru a ilustra pattern-uri de comportament disfunctionale, atitudini negative si aspecte care trebuie schimbate acum, in prezent.

Cumpatarea, sobrietatea este o cerinta pentru a duce o viata sanatoasa, dar o viata sanatoasa este necesara pentru a mentine sobrietatea.

Conceptul de viata sanatoasa pune accent pe valori explicite, acestea folosesc indivizilor in relatiile intrapersonale si interpersonale, cu partenerii, cu persoanles iubite, cu intreaga societate. Aceste valori includ: adevarul, onestitatea, solidaritatea, etica in munca, a invata sa inveti, independenta economica, preocupare pentru ceilalti, responsabilitate, responsabilitate familiala, compromisul cu comunitatea si chiar a fi un bun cetatean.

4. Despre recuperare

Obiectivele recuperarii in C.T. sunt globale, iar dezintoxicarea, intreruperea consumului sunt mai degraba o pre-conditie decat un obiectiv. Obiectivul primar individual este schimbarea pattern-urilor de comportament negativ, cognitive si a sentimentelor care predispun la consumul de droguri; obiectivul primar social este dezvoltarea unui stil de viata sanatos, fara folosirea drogurilor. Recuperarea stabila este integrarea cu succes a acestor doua obiective

COMENTARII

Incheiem acest articol cu sublinierea faptului ca, la fel ca orice organism viu, C.T. s-a nascut din predecesori ce i-au lasat anumite mostenirii, ca a fost tanara si ca a crescut iar din acel moment este un proces, o dezvoltare, este schimbare, este viitor deschis.

In actualitate fundamentele sale teoretice si actiunile concrete pe care le intreprinde au multe puncte comune cu diversele abordari profesionale actuale si din aceasta pozitie devine foarte valoroasa pentru ca profesionistii si terapeutii sa se incorporeze si sa se poata imbogati reciproc.

THE DIAGNOSIS, RISK FACTORS AND TREATMENT OF THE EXTENDED THROMBOSIS OF THE TRUNK AND CROSSA OF THE GREAT SAPHENOUS VEIN

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ABSTRACT:

In our experience in First Clinic of Surgery, County Hospital Timişoara, we followed 36 cases with great saphenous vein thrombosis, aged 28-79 years, period 1.01.2001-15.04.2003.

The investigations: continuous Doppler, echo Doppler and phlebography.

We applied initially non-steroid anti-inflammatory and anticoagulant treatment with heparin and LMWH. In the cases with extended venous thrombosis we continued with surgical treatment as a delayed urgency and we performed saphenectomy by stripping and phlebectomies, crossectomy with femoral thrombectomy, femoral-iliac thrombectomy. Postoperative we continued the anticoagulant treatment for 3 months with oral anticoagulant drugs at the patients with deep venous thrombosis respectively aspirin 100 mg/day, anti-inflammatory and phlebotonic treatment in cases with superficial thrombophlebitis.

Keywords: *thrombosis of the saphena.*

1. INTRODUCTION

The varicose disease is an evolving vascular pathology, frequent and affecting an important percent of population. The frequency is 1 of 2 women respectively 1 of 4 men.

The varicose disease untreated in time evolves continuously and slowly in many years. Suddenly a lot of acute complications may appear: chronic venous insufficiency after 15-20 years of evolution; varicose thrombophlebitis; spontaneous or posttraumatic injury of the varicose veins; thrombosis of the great saphenous vein and its crossa with or without extension at the deep venous system.

The thrombosis of the great saphenous vein and its crossa is a complication of the varicose disease with an increasing frequency in the last period – approximately 1 of 8 cases with hydrostatic varices.

The risk factors of the thrombosis of saphenous vein are:

1. Exogenous: mechanical factors (traumas), long immobilization in bed or due to a immobilization for a fracture; smoking; medical factors (contraceptives, vitamin K, diuretics);

2. Endogenous: age 60-70 years old, primary varices, hyper coagulation, pregnancy, sex (females), heredity.

The determinant factors known as Wirchow triad are:

- The venous parietal lesion done by external traumas, catheterizations and compressions;
- The venous stasis in the dilated varicose packets - it creates conditions for thrombosis by hypoxic lesions at the venous endothelium modifying the vascular electro negativity and determining plackets adherence (due to long immobilization in bed);
- The hyper coagulation of the blood in the varicose packets determines the extension of the thrombosis to the trunk and crossa of the great saphenous vein (deficit of S-protein, C-protein).

All these factors together determine the appearance of localized thrombosis (the white thrombus made of placket aggregates and fibrin fibres) followed by the extension of the thrombosis to the first collateral vein (mixed thrombus made of erythrocytes caught in a fibrin and leucocytes network). The evolution of the thrombosis, spontaneous or under treatment consists of the retraction of the clot and the reorganizing process with recanalization or extension of the thrombus in the deep venous system. It determines deep venous thrombosis at the calf through the perforant veins in the femoral vein, through the great saphenous crossa and then in the common iliac vein and inferior cava vein. In this case the edema from the calf extends to the unilateral thigh and then at the opposite inferior limb.

The clinic diagnosis: localized pains at the level of varicose packets; hard, thrombosed venous cordon with eritematous and edematous skin; the edema of the calf and thigh due to the extended thrombosis in the deep venous system (through the perforans veins and the saphenous crossa).

Investigations: continuous Doppler ultrasonography, Echo Doppler and Duplex Doppler used as noninvasive investigations. In selected cases phlebography allows to visualize the deep venous system.

Clinic and anatomic forms:

- Thrombosis localized at varicose packets;
- Great saphenous vein thrombosis;
- Great saphenous vein and crossa thrombosis;
- Trunk and crossa of the great saphenous vein thrombosis + extension at femoral vein.

The treatment differs with the extension of the thrombosis:

- Superficial confined venous thrombosis: anti-inflammatory drugs, with hialuronidase, heparin with low molecular weight (LMWH), surgical treatment;
- Extensive venous thrombosis: “delayed” urgency surgery – saphenectomy, phlebectomies, crossectomy, thrombectomy followed by anticoagulation 3 month.

2. MATERIAL AND METHOD

Prospective study – 01.01.2001- 30.06.2003: 516 patients with hydrostatic varices and with ages between 19-75 years old have entered the hospital in this period in the First Clinic of Surgery University of Medicine and Pharmacy Timisoara. 361 (61.25%) were females and 155 (38,75%) were males. Unilateral varices were observed in 336 cases (65,11%) and bilateral in 181 cases (34,89%).

Etiology: primary varicose disease – 423 cases (81,97%); secondary varices due to postthrombotic syndrome – 39 cases (7,55%); congenital varices – 2 cases (0,4%); relapsed varices after saphenectomy – 52 cases (10,07%).

Using CEAP classification the patients are grouped in the following classes: C4 – 348 cases (67,45%); C5 – 128 cases (24,8%); C6 – 40 cases (7,75%).

The treatment consisted in:

- saphenectomies – 124 cases (24,03%),
- saphenectomies and phlebectomies, ligatures of perforating veins, sclerotherapy with catgut – 231 cases (44,76%),
- phlebectomies, ligations of perforating veins, sclerotherapy with catgut – 138 cases (26,74%),
- crossectomy and thrombectomy – 12 cases (2,32%),
- Chiva operation – 6 cases (1,16%),
- SEPS – 4 cases (0,77%)
- an operation for decreasing the diameter of the great saphenous vein near its crossa – 1 case (0,19%).

We studied prospectively from all these cases an homogenous group of 174 patients, with ages between 19-69 years, 124 females (71,26%) and 50 males (28,74%), with unilateral disease – 115 cases (66,09%) and bilateral – 59 cases (33,91%).

The etiology of their varicose disease was: primary varices – 137 cases (78,73%); secondary varices due to postthrombotic syndrome – 17 cases (9,77%); congenital varices – 2 cases (1,14%); relapsed varices after saphenectomy in antecedents – 18 cases (10,34%).

After CEAP classification, we grouped them in: C4 class – 102 cases (58,62%); C5 class – 37 cases (21,26%); C6 class – 35 cases (20,11%).

The surgical treatment consisted in:

- Intern saphenectomy – 46 cases (26,43%);
- Intern saphenectomy + phlebectomies, ligations of perforating veins, sclerotherapy with catgut – 68 cases (39,08%);
- Phlebectomies, ligations of perforating veins, sclerotherapy with catgut – 45 cases (25,86%);
- Crossectomy + thrombectomy – 10 cases (5,74%);
- SEPS – 4 cases (2,29%);
- Operation for decreasing the diameter of the great saphenous vein near its crossa – 1 case (0,57%).

3. RESULTS AND DISCUSSIONS

We observed 39 cases (24,41%) with hydrostatic varices complicated with extended thrombosis at the great saphenous vein, patients selected from this group of 174 cases. There were 15 males (38,47%) and 24 females (61,53%) with ages between 19-75 years old. They entered the hospital for: pain at the level of varicose packets, inflammation in the presence of a hard superficial venous cord, important eritema and edema of the calf and/or thigh, recent pulmonary embolias (2 cases).

The extension of the thrombosis was varied: 14 cases (35,89%) – confined at calf; 8 cases (20,51%) – extended at thigh; 12 cases (30,76%) – thrombosis of the crossa, 3 of them with floating thrombus in the femoral vein; 5 cases (12,82%) – thrombosis extended in the femoral and iliac vein.

Anticoagulant treatment has been started before a surgeon consulted them. Surgical treatment was performed at all these 39 cases and it consisted of: saphenectomy and crossectomy – 22 cases (56,41%); crossectomy and femoral thrombectomy – 12 cases (30,76%); crossectomy and femoral-iliac thrombectomy – 5 cases (12,82%).

The surgical treatment is preceded by heparin administration 5000UI, i.v. at 6 hours stopping it 6 hours before the surgery, at cases with femoral and femoral-iliac extended thrombosis. After we extracted the thrombus and after the vein has become permeable, we administrated intraoperatively 5000UI, i.v. heparin and we continued this treatment 7 days after surgery followed by oral anticoagulants (with 5 days of association of these 2 types of anticoagulants drugs). The treatment with oral anticoagulant drugs must continue minimum 2 years with a periodic evaluation of INR.

The incidents and accidents in surgery were minor.

The postoperative evolution was favorable with remission of symptoms at 5-7 days postoperatory; the period of admission in hospital was about 10-15 days and the time of returning to normal life and work was 1-2 months.

The postoperatory complications were minor and local and we observed them in 4 of our cases. A major complication – pulmonary embolia was occurred in 1 case (it was treated with unfractionated heparin preoperatory then crossectomy, thrombectomy, saphenectomy and LMWH postoperatory).

4. CONCLUSIONS:

- Prophylaxis – surgery in time for varicose disease;
- The thrombosis of the saphenous vein – significant frequency;
- Diagnosis – it is sufficient the Echo Doppler exam;
- Treatment - surgical: delayed urgency
- Medical: anti-inflammatory, anticoagulant and phlebotonic drugs.

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INTRAPERITONEAL BILIARY CALCULI – EXPERIMENTAL AND CLINICAL STUDY

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ABSTRACT:

„Lost" intraperitoneal biliary stones are described as laparoscopic cholecistectomy complication. The result of this complication and the requirement of putting them out by exploratory laparatomy are in discussion.

The experimental study was carried out on 40 white rats, Wistar race. We carried out a median mini-laparatomy in general anesthesia and we introduced biliary stones or fragments of them.

Postoperative evolution was good and we re-operated them after 6 weeks respectively 3 month. When we looked in the peritoneal cavity we observed the blocking of the biliary stones by the omentum and their omento-parietal adherence.

The histopathologic exam showed: lax conjunctive tissue, which presents an important histiomacrophagic infiltration with foaming cells of xantomatous type (which have incorporated apparent lipidic substance).

In our recent experience we had 2 cases with intraperitoneal biliary stones.

We consider, on the basis of our experimental and clinical study that the intraperitoneal "loosing" of the little biliary stones is an "incident" and it does not require exploratory laparatomy for putting them out.

KEYWORDS: *biliary intraperitoneal stones, laparoscopic cholecistectomy.*

1. INTRODUCTION

In Romania we cure biliary lithiasis almost always by surgical means- so laparoscopic cholecistectomy is the preferred technique used (preferred by patients and also by the physician because this method became an alternative and very popular method). One of the intraoperatory incidents is the loose of the biliary stones in the peritoneal cavity.

The evolution and the intraoperatory attitude for this incident are discussed and the necessity for the finding of the "lost" intraperitoneal stones and their extraction is in the debate.

The methods for extraction are: direct with the tourniquet, using the “basket” procedure, by laparotomy.

In the medical literature some complications in these cases were cited like:

-granuloma
-peritoneal and retroperitoneal abscess
-extern biliary fistula
-late intestinal occlusion
-cholelithoptisis

2. MATERIALAND METHOD

We performed an experimental study at the Laboratory of Experimental Surgery and Morphopathology University of Medicine and Pharmacy Timisoara. We used white mice – Wistar race.

We performed median minilaparotomy under intraperitoneal general anesthesia for implantation of biliary stones, clips and suture threads. We performed another laparatomy after 3-6 months and we observed the local structural changes.

We used 4 groups of mice, 20 mice for each group:

- 1-st group: test group;
- 2-nd group: with intraperitoneal biliary stones;
- 3-rd group: with titan clips;
- 4-th group: with suture threads (10.0).

The results of the study were:

- 0 mortality;
- 0 complications;
- a good tolerance of the organism for the biliary calculi;
- the macroscopic anatomo-pathological results: the blocking of the calculi by the epiploon and by the adherence between the epiploon and parietal peritoneum;
- microscopic findings: important histiomacrophages infiltrate with foamy xanthomatous cells, granulomatous reaction with multinucleate giant cells and discreet cells infiltrate.

We observed recently 2 cases with intraperitoneal biliary “lost” stones in our clinical experience:

1. The patient K.S. (F.O.35838/04.10.2002), 52 years old was operated by laparoscopic method (colecistectomy) 2.5 years ago. The patient was diagnosed with uterine fibroma and we performed total hysterectomy with bilateral anexectomy. When we opened the peritoneal cavity we observed multiple calculi of small sizes (2-5 mm in diameter) adhered at the epiploon, the surface of the both ovaries, salpinxes and at the pelvic parietal peritoneum. They were tolerated well and there was no macroscopic inflammatory reaction. The hystopatological exam of the ovary revealed hyaline transformation and calcification regions with blackberry like aspect without inflammatory infiltrate.
2. The patient S.M. (F.O. 23581/14.05.2001), 40 years old, operated laparoscopic for biliary stones (cholecistectomy), 3 years ago, were hospitalized for policystic left ovary. We performed left ovarectomy. When we explored the peritoneal cavity we observed 4 solid bodies

with biliary stones aspect (with 2-5 mm diameter) on the left ovary, 5 calculi in Douglas, without local inflammatory reaction.

Concerning the "lost" intraperitoneal biliary stones after laparoscopic surgery, we observed that abdominal symptoms determined by the adherence of the calculi at different viscera didn't appear. We discovered them when we performed laparatomy for another pathologies. We state that the intraperitoneal "loosing" of some little calculi after laparoscopic cholecistectomy is an " incident" and it doesn't require exploratory laparatomy for their extraction. Also the inflammatory reaction, which is minimal and rare, is not a reason for the conversion of the surgery to a classical mean.

3. CONCLUSIONS

In the First Clinic of Surgery we observed, studying a group of over 1500 patients who underwent laparoscopic cholecistectomy, the advantages of this procedure comparing with the classic approaches: minor and rare intraoperatory complications, short period of hospitalization (3-5 days), the decreased cost for the postoperatory medical assistance and the quick cure and returning to normal life. Another advantage is the decreased operatory trauma comparing with the classic approach. The patients even in the presence of another associated pathology more easily tolerate the method.

The factors, which may influence the rate of postoperatory complications, are the advanced age, the anatomo-pathologic form of cholecistitis and another associated diseases.

The laparoscopic cholecistectomy was for a long time contraindicated at cases with acute cholecistitis because of the difficulties of identifying the anatomic elements and also because of the increased risk of iatrogenous lesions.

Now this method is used also for these cases due to the experience obtained in time by the surgeons.

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THE NATURAL HISTORY AND THE EVOLUTION OF THE TREATED AND NOT TREATED VARICOSE DISEASE

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ABSTRACT:

The varicose disease represents a very frequent cause of morbidity: 50% of population.

The main etiological factors are: prolonged orthostatic position, genetic factors, pregnancy, raised intraabdominal pressure.

The treatment is initially conservative: postural drainage, elastic stockings, drugs treatment. This treatment is not curative; the varicose disease is progressive so subsequently we perform surgical operations as saphenectomy by stripping, phlebectomies, venous catgut inclusions and sclerotherapy.

We studied a group of 72 patients with relapsed varicose disease in different stages of evolution. We performed the stripping of the great saphenous vein associated with phlebectomies, the cuffing of the great saphenous crossa and phlebectomies centered on the varicose packages. We used this method of cuffing the crossa because the varicose disease was in an early stage and the patients were diagnosed with chronic arteritis obliterans at the controlateral limb for preserving the great saphenous vein for an eventual arterial by-pass.

KEYWORDS: *varicose disease, relapses of varices.*

1. INTRODUCTION

The real relapses of the varicose disease after a previous surgery for this pathology consists of the varices appearance in the reflux regions, which were interrupted at the primary operation.

The development of the varicose veins in another region closer or farther from the previously operated areas, corresponds to the chronic evolution of the varicous disease. We have to take into account 3 elements when we observed varices at a previously operated patient for the varicose disease: real relapsed varices, residual varices and varices due to the evolution of the disease. In the surgery of the varicose disease there are a lot of relapses (15-30% of cases) if we follow the

patients for a long time. This disease is a chronic evolving pathology determined by a defect in the venous walls in many cases, the veins having a predisposition for dilatation.

Clinic arguments for relapsed varices: free interval of time after surgery; postoperatory scars; disseminated varicose veins with fragile and tortuous veins.

When we know the physiopathological aspects of the surgical treatment we can easily recognize the causes of the relapses. The persistence of some reflux points will determine the relapse because the functional objective of the varices treatment was not solved (the suppression of the pathologic reflux which determines the increased orthostatic venous pressure). The reflux may persist if the sclerotherapy is used exclusively for the treatment of all types of varices. If some varicose packets and their collaterals are not excised there will exist an important source of relapses.

The relapses may also appear, when a correct surgical treatment is applied, because of the evolving character of the disease due to the genetic factors (family history of varicose disease). These patients must be controlled regularly and carefully and when the first varicose veins appear we must start a therapeutic procedure (mainly sclerotherapy and phlebectomies).

In the period 2001-2002 at The 1st Clinic of Surgery Timisoara, there were 72 cases with relapsed varicose disease, 17 men and 55 women.

The location of the relapses were: calf: 27 cases (37.5%); thigh: 3 cases (4.1%); calf and thigh: 42 cases (58.3%).

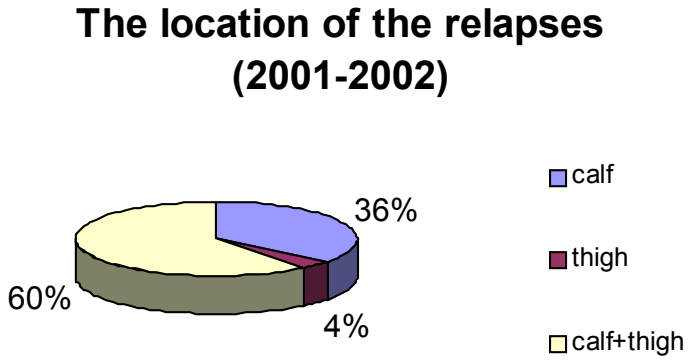

***Fig.1**. The location of the relapses*

The relapses appear most frequent in association at calf and thigh.

Unilateral varices were observed in 41 cases (56.9%) and bilateral – in 31 cases (43.1%) so the unilateral disease was predominant.

The relapses had the following locations (as regions of superficial venous system): in the intern saphenous vein territory: 8 cases (11.1%); in the extern saphenous vein territory: 2 cases (2.7%); in both these 2 territories: 2 cases (2.7%); unsystemathized varices: 60 cases (83.3%).

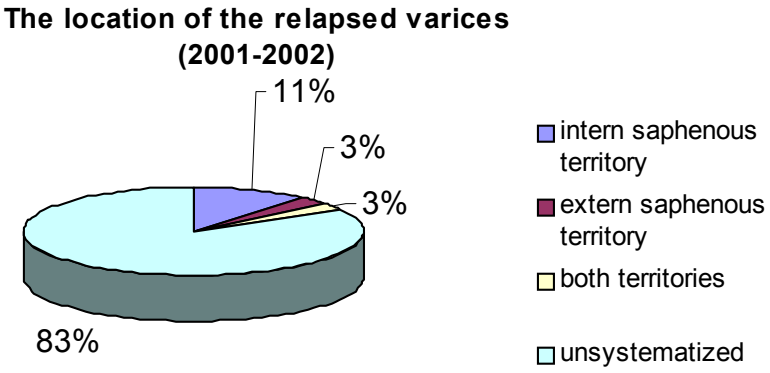

Fig. 2. *The affected venous territories*

The majority of the relapses were unsystemathized.

In some cases, the relapses were associated with signs and symptoms of chronic venous insufficiency as calf ulcer. So we observed at 2 cases (2.7%) trophic changes of the skin (pigmentations, atrophies of the) and active calf ulcers in 16 cases (22.2%).

Another associations were with acute superficial thrombophlebitis in 8 cases (11.1%), acute deep thrombophlebitis in 3 cases (4.1%) and with postthrombotic syndrome in 6 cases (8.3%).

The previous surgeries, carried on before the appearance of the relapses, were: crossectomy of the intern saphenous vein associated with phlebectomies: 2 cases (2.7%); intern saphenectomy: 42 cases (58.3%); extern saphenectomy: 1 case (1.3%); intern and extern saphenectomy: 3 cases (4.1%); phlebectomies: 13 cases (18%); sclerotherapy: 8 cases (11.1%); intern saphenectomy associated with sclerotherapy: 1 case (1.3%); intern saphenectomy associated with phlebectomies: 2 cases (2.7%).

The types of therapeutic methods applied for the relapses were: sclerotherapy: 5 cases (6.9%); intern saphenectomy associated with phlebectomies: 5 cases (6.9%); intern saphenectomy associated with perforating veins ligations: 1 case (1.3%); phlebectomies: 9 cases (12.5%); phlebectomies associated with intraoperatory sclerotherapy with catgut: 21 cases (29.1%); plastic reconstructive surgery for the calf ulcer: 1 case (1.3%); medical treatment (anti-inflammatory and anticoagulant drugs): 7 cases (9.7%); perforating veins ligations associated with sclerotherapy with catgut: 2 cases (2.7%); intern saphenectomy associated with phlebectomies and sclerotherapy with catgut: 3 cases (4.1%); without treatment (patients that entered the hospital for another surgical pathology): 3 cases (4.1%); perforating veins ligations associated with valvuloplasty of the popliteal vein: 1 case (1.3%); subfascial endoscopic perforating veins ligations at calf: 5 cases (6.9%); perforating veins ligations associated with phlebectomies and sclerotherapy: 2 cases (2.7%); perforating veins ligations (calves and thighs): 2 cases (2.7%); perforating veins ligations associated with phlebectomies: 2 cases (2.7%); perforating veins ligations associated with intern saphenectomy and phlebectomies: 1 case (1.3%); CHIVA operation associated with phlebectomies and sclerotherapy: 1 case (1.3%); crossectomy of the intern saphenous vein associated with phlebectomies: 1 case (1.3%).

So in these cases with relapsed varicose disease phlebectomies associated with intraoperatory sclerotherapy with catgut and only phlebectomies were performed in the majority of cases.

2. CONCLUSIONS

In the period 2001 – 2002 we had 72 patients with relapsed varicose disease. This pathology was observed predominantly at women, the venous dilations were located especially at calf and thigh, unilateral and unsystematized. In 22.2% of cases there was an association with active calf ulcer and in 8.3% of cases with postthrombotic syndrome. The previous surgeries were predominantly intern saphenectomies (58.3%) and phlebectomies (18%). Phlebectomies associated with intraoperatory sclerotherapy with catgut and only phlebectomies were the surgical procedures preferred for these cases.

Recent studies observed an increased incidence of relapses (15-30%) for patients operated in antecedents for varicose disease. In fact we must make the difference between the real relapsed varices, the residual unexcized varices and the varices due to the natural evolution of the disease. The relapsed varices can be determined by surgical causes but there is also a complex phenomenon of angiogenesis of the varicose vessels. We observed relapsed varices caused by technique deficiencies but also by neoangiogenesis, studying these cases from the 1st Clinic of Surgery. The varicose veins caused by neoangiogenesis are sinuous, fragile and form a real venous sponge, which is excised with difficulty.

The prophylaxis of the relapses in the varicose disease is made by selecting the cases for surgery as well as by choosing the appropriate type of operation taking into account the clinic, laboratory and imagistic exams. A new method for neoangiogenesis prevention is the invagination of the saphenous stump to avoid the contact of the venous endothelium with the surrounding tissues and so the regeneration of the veins. Postoperatory, the complex treatment for relapsed varicose disease includes periodic controls, sclerotherapy and elastic stockings. The results of this surgery are better if the patients are cooperative and if they come regularly at the controls and periodical treatments, also if they respect the appropriate regime of life.

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PESTICIDES AND HEALTH

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ABSTRACT

Pesticides became a very important means of the chemisation, inevitable in developed and developing regions in order to obtain the necessary amount of agricultural products for the continuously increasing population. They are also crucial in the prevention some communicable diseases transmitted by vectors (tick-borne encephalitis, malaria, yellow fever, etc.).

At the same time, their harmful effects on humans and the environment cannot be neglected. Some of them are highly or moderately toxic, and cause acute or chronic intoxication in their users during dilution, spraying without protective garments etc.

Certain pesticides (e.g. organochlorines) accumulate in different organs of the exposed workers, and, due to their residues in food, also in the organism of the population. Some of them can alter the hormonal status of the exposed persons affecting the spermiogenesis, and several are embriotoxic or cause different alterations of the newborns.

In spite of the mentioned and other hazards, pesticides will be used in the future. That is why it is absolutely necessary to work out and apply the preventive measures. In case of efficient prevention, the majority of harmful effects of pesticides on human health can be avoided.

PATHOPHYSIOLOGY OF ALCOHOLISM

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In the past 25 years substantial progress has been made in elucidation of effect of ethanol in the brain. These fundamental discoveries include a proper understanding of transmission of chemical messages in the brain by neurotransmitters, cell receptors to which these neurotransmitters bind.

Ethanol effects on neurotransmitters are described on emphasis on ion channels, neurotransmitter transporters and enzymes that affect these activities. Exposure of the brain to alcohol initiates an adaptation, which is reflected in development of tolerance, dependence and alcohol withdrawal syndrome. Alcohol profoundly alters the communication within and between neurons, the ultimate result being changes in brain activity and behaviour.

The present summary focuses on the new developments in neurotransmitters (NMDA, glycin and GABA), inhibitory ligand-gated and voltage-gated ion channels, and protein phosphorylation, localization and gene expression during acute and chronic effect of alcohol on the brain. Reward and reinforcing effects will be discussed in the context of craving, dependence and relapse and the role extended amygdale as a possible neural structure for reward.

Genetic studies from knockout and transgenic mice gather data on possible candidate genes involved in alcoholism.

ILL EFFECTS OF INORGANIC METAL POLLUTANTS

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ABSTRACT:

The presence of heavy metals in the ecosystem presents a major problem in environmental and occupational medicine. Repeated exposure of humans by Hg and Mn compounds and the resulting pathological changes have been described.

In the present experiments, male Wistar rats were treated for 10 weeks by gavage with low-doses $HgCl_2$ (0.5 and 2.0 mg/kg) and $MnCl_2 \cdot 4H_2O$ (14.8 and 59.4 mg/kg). It was tested how these doses of the two metals affected various elements of spatial learning and short- and long-term memory, spontaneous exploratory locomotion, and sensorimotor performance with psychomotor gating. Metal-specific functional neurotoxic effects in the CNS in general and in centers with special role in learning (hippocampus) were also looked for.

Both metals caused a dose-dependent significant decrease in the memory performance and in the local locomotor activity. In the sensorimotor (startle) reaction, the number of responses dose-dependently and significantly (high dose vs. control) decreased. In the Hg^{2+}-treated animals, spontaneous cortical activity was shifted to higher frequencies. The effect on the evoked cortical activity was below significance.

The Hg and Mn doses applied altered in the higher nervous functions of the treated adult rats. In cases of human exposure, similar effects can be expected.

KEYWORDS:

mercury, manganese, neurotoxicity, 8-arm radial maze; spontaneous locomotor activity; acoustic startle response

1. INTRODUCTION

Manganese is toxic in high but an essential micronutrient in low dose. Occupational Mn exposure comes from ore and metal dusts [11] while Mn burden of the population originates from the environment (due to Mn-containing waste, methylcyclopentadienyl manganese tricarbonyl used as anti-knock petrol additive, and organo-Mn agricultural fungicides). The brain is among the primary target organs in chronic Mn exposure [20]. A "Parkinson-like syndrome" develops with functional [22] and structural [26] damages of the dopaminergic systems. The release of other transmitters (Glu and GABA) is also reduced by Mn^{2+} in moderate doses [24]. In Mn-exposed humans, impairment of several memory and motor function parameters was observed [14]. In rats, increase or decrease of the spontaneous motor activity was seen depending on the dose and duration of Mn treatment [2].

The important forms of mercury from neurotoxicological viewpoint are the metallic form, and the divalent inorganic and organic forms. Humans are exposed to mercury by the industry, from dental fillings, by the use of mercury-based fungicides and bactericides, and via food. Inorganic mercury is known to diminish mental performance in humans (especially children) and in young experimental animals by inducing deficits in coordination, emotionality and other behavioural features, and causing neurological disorders [17]. Mercury induces pathomorphological changes, affecting the higher order functions of the central nervous system. By stimulating serotonin receptors, Hg causes increased motivation, aggressiveness and impulsive behaviour [5], and by stimulating striatal dopamine release, decreases the intraneuronal dopamine degradation [7]. Several authors described the decrease of spontaneous locomotion [10,21] and startle response [5]. In animal experiments, Hg^{2+} was found to inhibit presynaptic Ach release and postsynaptic muscarinic receptor activation [4], and to damage motor axons [18]. The irreversible inhibition of transmitter release is supposed to depend on generation of disulfide bridges [13]. Effects of Hg^{2+} on GABA receptors [15] have likewise been described. Hg^{2+} and Mn^{2+} block Ca^{2+} channels [3], and Hg^{2+} also affects Na^{+}/K^{+} ATP-ase [23,25]. In occupational exposure to inorganic mercury, alterations of the spontaneous cortical activity [19] and delayed waves in the brainstem auditory evoked potential [6] were found.

The aim of the present work was to investigate the effects of subacute (10 weeks) oral exposure of rats to inorganic Mn^{2+} and Hg^{2+} with behavioral and electrophysiological methods.

2. METHODS

Male Wistar rats (220-250 g body weight at start) were used in both treatments. The animals were treated with 14.8 and 59.4 mg/kg b.w. Mn^{2+}, or 0.5 and 2.0 mg/kg b.w. $HgCl_2$ (low and high dose, respectively) per os by gavage for 10 weeks (5 days a week). Control animals received distilled water. The animals were housed under controlled conditions of temperature (22 to 24°C) and photoperiod (12-hour light/dark cycle with light starting at 06:00), with free access to drinking water. The memory test used required that during the 10 weeks of treatment the animals had a restricted access to food (1 hour/day) resulting in a mild (ca. 20-25%) body weight loss [1]. Body weight was measured and the animals' general state was observed every day. All behavioral tests were performed, in a room different from that used for keeping and treating the animals, between 08:00 and 14:00.

The animals' spatial learning ability was tested in an 8-arm radial maze. In the first week of treatments, the rats were adapted to find food pellets in the maze arm ends. During acquisition (2^{nd} week), the rats learned to visit the farthest points of each arm. All animals had a run performance of over 85%. In the spatial short-term working memory test (3^{rd} and 5^{th} week) the rats were allowed in the first run to enter four of the arms, and their task in a second run was to enter only arms not entered 2 or 4 hours ago (the “event-to-be-remembered”). Reference memory was tested on the 4^{th} week; here food reward was put only in the 4 arms preferred by the individual rats. Long-term retention memory test: following 2 weeks of rest, memory return was observed on the 8^{th} week. Then, the 2- and 4-hour spatial working memory was tested again (9^{th} and 10^{th} week). In all tests with the 8-arm maze, run performance was calculated from the proportion of errors (entering a false arm) to all responses (entering any arm).

Locomotor activity was tested on the 5th and 10th weeks of treatments. Spontaneous horizontal, vertical and local exploratory activity was scored automatically by means of a PC during a 10-minute session in a dimly lit open field box (40x40x40 cm) equipped with two arrays (3 and 15 cm above floor level) of infrared movement detectors with 1.1 cm distance between the beams.

Acoustic startle response (ASR) and prepulse inhibition (PPI) of the rats was measured on the 5th and 10th weeks, after the open field sessions, using a commercially available acoustic reflex monitor. The animals were one by one put in the test box. After a 10-min accommodation, a series of 10 consecutive tones (5 kHz, 110 dB, 200 ms, 15 s interval) as test stimuli were applied. In another series following 15 min rest, the test stimuli were by 200 ms preceded by inhibiting prepulses (1 kHz, 73 dB, 500 ms). A whole body twitch resulting in more than 50 g force to the cage floor was accepted as positive response.

Electrophysiological investigation. After finishing all behavioural tests (i.e. after 5 and 10 weeks of Hg- and Mn-administration), the animals were anesthetized with 1000 mg/kg urethane ip. and the left hemisphere was exposed by removing the bony skull. Following a recovery of 30 minutes minimum, surface electrodes were placed on the primary somatosensory, visual and auditory cortical focus and a steel needle electrode was inserted into the hippocampal CA1 region. Spontaneous electrical activity (electrocorticogram, ECoG) was recorded for 5 min, and subsequently analysed for the relative power distribution among the standard frequency bands (delta to gamma). Cortical evoked potentials were recorded subsequently via the same electrodes. (Somatosensory stimulation - square electrical pulses {1 Hz, 3-4 V, 0.2 msec} to the whiskers, visual stimulation - flashes {1 Hz, 60 lux} to the contralateral eye, acoustic stimulation - clicks {1 Hz, 40 dB}to the contralateral ear.) Fifty stimuli per modality per rat were applied. After averaging, latency and duration of the main waves was measured manually. All recording of spontaneous and evoked activity and off-line analysis was performed by a PC using the NEUROSYS 1.11 software (Experimetria Ltd., U.K.).

All data were analyzed by ANOVA or Kruskal-Wallis- and Mann-Whitney U-test following a Kolmogorov-Smirnov normality analysis.

3. RESULTS

The manganese and mercury doses applied in the present investigation had no general toxic effect.

During all phases of the maze learning test, both $MnCl_2$ treated groups showed, compared to control animals, a decrease in the average memory performance (*Fig.1*). Acquisition (7th-12th days of the treatment) was dose-dependently impaired in $MnCl_2$ treated rats (high dose vs. control: $0.001<p<0.01$; low: $0.01<p<0.05$). The reference memory of the animals' spatial learning (4th week) showed in both treated groups a significant (high dose vs. control: $p<0.001$; low dose vs. control: $0.001<p<0.01$; and high dose vs. low dose: $0.01<p<0.05$) performance deficit. During the short-term (4 hours) working memory test (5th week), the error frequency of the treated rats was significantly and dose-dependently higher that in the controls (high dose vs. control: $p<0.001$; low dose vs. control: $0.01<p<0.05$; high vs. low dose: $0.01<p<0.05$). After 2 weeks rest period, the control and low dose group both showed a memory return to the level on the 2nd week of treatment but the level reached by the high dose animals was about 20 % below that. In the long-term retention test (from the 43rd day on) both $MnCl_2$ treated groups showed a further

significant memory deficit vs. control ($p<0.001$) and the high vs. low difference was also significant ($0.001<p<0.001$). Comparison of the long-term (9^{th} and 10^{th} week) and short-term (3^{rd} and 5^{th} week) memory retention showed that the error level of both treated groups remained nearly unchanged but the difference vs. control group increased in both long-term memory tests.

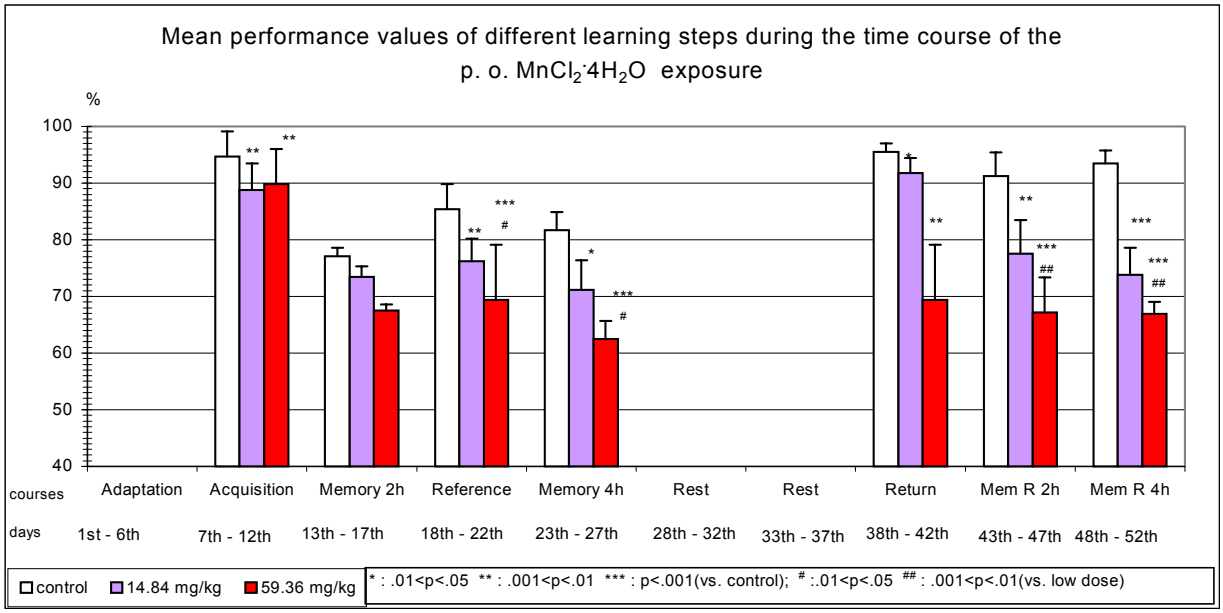

Fig.1. Memory alteration of male Wistar rats treated with $MnCl_2$ p.o. by gavage.

During acquisition and short-term maze learning (2^{nd}- 5^{th} week of treatment), the $HgCl_2$ treated and control animals showed dissimilar trends in the averaged memory performance (*Fig.2*). On the 13^{th} to 17^{th} and 23^{rd} to 27^{th} day of behavioural investigation (short-term retention tests), a significant (high dose vs. control: $p<0.001$, low dose vs. control: $0.01<p<0.05$) memory deficit developed in the groups treated with $HgCl_2$. In the long term retention test (43^{rd} to 57^{th} day of treatment) the 2 and 4 hours memory performance of the treated groups decreased by further ca. 10 %. The reference memory of the animals (4^{th} week of treatment) showed also a significant dose-dependent alteration (high dose vs. control: $p<0.01$, low dose vs. control: $p<0.001$).

Open field tests revealed a decreased locomotor activity in the treated animals on the 5^{th} and 10^{th} weeks of $MnCl_2$ administration (*Fig.3*). The diminished spontaneous exploratory activity of the animals was mainly due to decreased vertical and horizontal activity, and was significant (both doses vs. control: $p<0.01$). On the 10^{th} week, local motor activity was significantly reduced in both treated groups ($0.001<p<0.01$ vs. control).

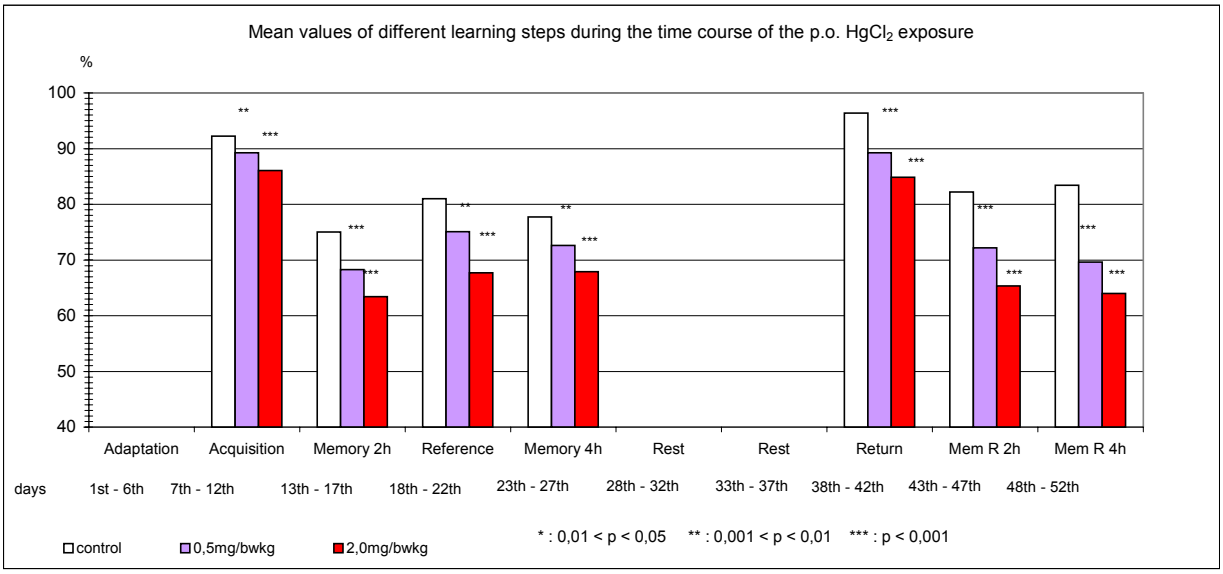

Fig.2. Memory alteration of male Wistar rats treated with $HgCl_2$ p.o. by gavage.

Habituation in the exploratory activity (over the 10 min session) was increased in both treated groups vs. control in the 5th and 10th weeks. Fig. 3 shows the three different elements of locomotor activity - motility, rearing and grooming - on the 10th week. Habituation in the exploratory activity (over the 10 min. session) was increased in both treated groups vs. control.

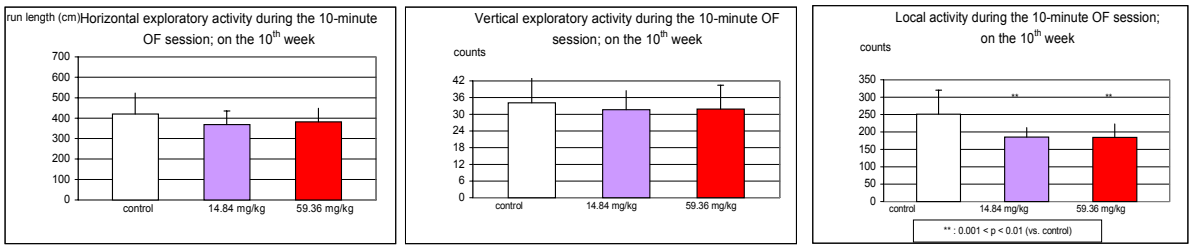

Fig.3. Effects of $MnCl_2$ on horizontalm exploratory activity, rearing activity and local activity of rats over the 10 min open field session in the 10th week of treatment.

Decreased locomotor activity was seen on the 5th and 10th week of hg^{2+} treatment, too. The diminished spontaneous locomotor activity was mainly due to decreased vertical and horizontal activity. *Fig. 4* shows the 1st, 5th and 10th min locomotor activity on the 5th week. Grooming activity/local exploration was reduced by 25 % in the low and 38 % in the high dose group in the 5th week. Continued treatment, however, did not increase this effect. The habituation in the exploratory activity (over the 10 min. session) was decreased in both treated groups vs. control.

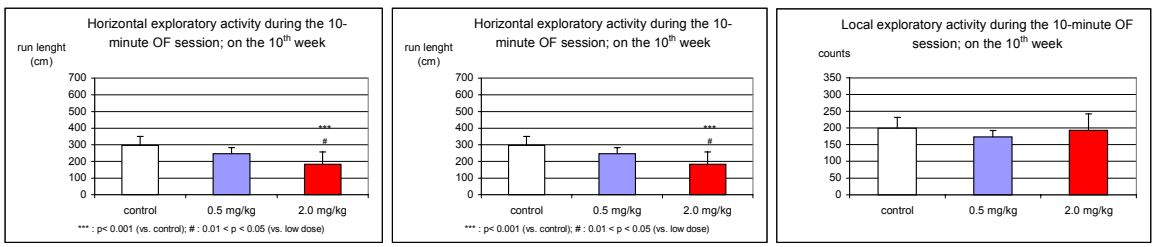

Fig.4. Effects of $HgCl_2$ on spontaneous motility, rearing activity, and grooming activity of rats in the 10th week of treatment.

The number of positive ASR responses decreased significantly ($p<0.01$) in the $MnCl_2$ treated groups by the 10^{th} week (in he 5^{th} week, the difference was not significant). With prepulse (PPI), the number of responses of the treated rats increased, while in the controls, it decreased (significant difference, $p<0.01$ vs. control for both).

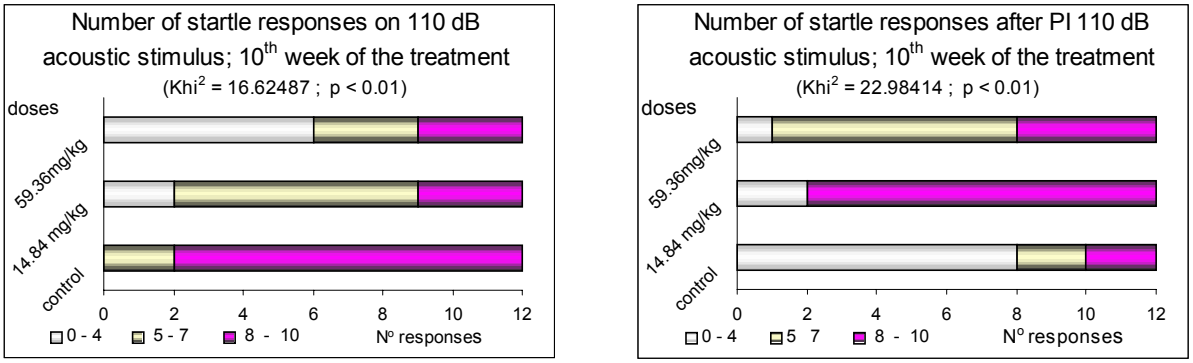

Fig.5. Psychomotor (ASR) and sensorimotor gating (PPI) performance on the 10^{th} week of the $MnCl_2$-treatment

In the high dose $HgCl_2$ group, the number of positive responses was significantly decreased both in the 5^{th} (high dose vs. control $p<0.01$ - not shown) and 10^{th} week ($p<0.05$; *Fig.6*). In the low dose group, the number of startle responses was significantly less than in the control in the 5^{th}, but not in the 10^{th}, week. The number of responses with PPI was not different in the treated and control groups in the 5^{th} week. In the 10^{th} week, however, the low dose rats gave significantly ($p<0.05$) more responses.

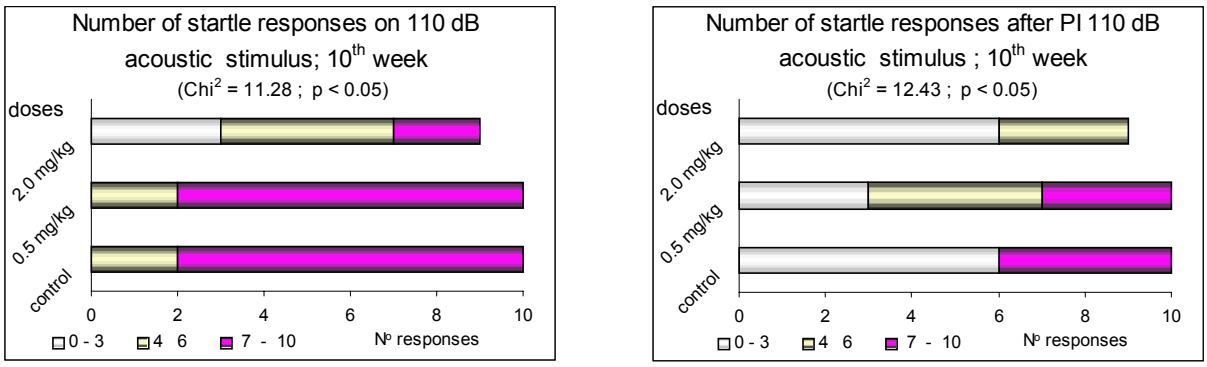

Fig.6. Psychomotor (ASR) and sensorimotor gating (PPI) performance on the 10^{th} week of the $HgCl_2$-treatment

Spontaneous cortical and hippocampal activity was shifted to higher frequencies in the Hg^{2+} treated animals. In the $MnCl_2$ treated ones, the spontaneous activity in the delta and gamma bands decreased, and in the theta and beta1 bands, increased.

4. CONCLUSION

Recall of acquired memory contents was more affected by the high than by the low dose Mn. In the long-term retention, however, the low dose group showed a more severe impairment. Manganese is known to affect several transmitter systems, including those involved in memory functions [12,22]. The effect of Mn on the

cholinergic system [16] can be reflected in altered spatial learning [8]. Spontaneous motor activity in the rat involves both the mesolimbic and nigrostrial dopaminergic system [9], Mn-dependent dysfunction of which is also known.

Hg affects transmitter systems involved in memory functions. The changes of vertical and horizontal motor activity may arise from alterations in the serotoninergic and dopaminergic transmission, respectively [5,7]. Hippocampal muscarinic receptors are extremely sensitive to Hg [4] which explains its strong memory effect and supports our finding that hippocampal spontaneous electrical activity was more affected than cortical activity. The effect of Hg on the cholinergic system may explain the diminished reactions of the treated animals in the ASR test and memory processes [8]. The reduction of prepulse inhibition, as it was found in our experiments, can be due to the effect of Hg on GABAergic synapses [15].

The heavy metal doses applied caused alterations in the higher nervous functions of the treated adult rats. In case of human exposure, similar defects can be expected.

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THE STIMULATION OF IMMUNE REACTIVITY IN POULTRY AS CONSEQUENCE TO THE ADMINISTRATION OF PROBIOTICS

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ABSTRACT

The researches served as purpose the establishment of the immunostimulator effect of probiotics in poultry vaccinated with an immunogen against the New Castle disease. The experiments were performed on 60 broilers Ross hibrids, grouped in four experimental batches that have had the benefit of the same conditions of alimentation and maintenance.

The immunomodulating effect of the probiotic has been appreciated on the basis of the modification of lyzozyme concentration and serum properdin concentration, as well as the antibody titre. The obtained results have shown an intensification in the synthesis of immune effectors, specific and unspecific, in all batches to which the immunomodulating substances were given. Significant results were registered in the case of antibody synthesis and from the unspecific factors it could be said that the probiotic influences mostly the serum lyzozyme production.

KEY WORDS: *probiotic, poultry, antibody, lysozyme, properdin*

1. INTRODUCTION

The studies that promote the modern micro-technologies to obtain probiotics are extremly preoccupied to reestablish the natural balance between the probiotic microbiocenosis and antibiotics microbicenosis, starting from the level of ecosystems, as elements of the external environment or of the inner environment, characteristic for the animal and human organisms, disturbed by the use, and mostly by the irrational abuse of antibiotics(1).

The results obtained in zootechnics with probiotics are obvious, and the experiments performed revealed a positive influence on the immune system in animals to which probiotics were administered(2, 3, 4).

2. MATERIALS AND METHODS

Researches were performed on 60 chicks, grouped in four experimental batches, as following:

- the witness batch (M) - unvaccinated and fed with mixed standard fodder;
- the experimental batch 1 (E1) - unvaccinated and fed with standard mixed fodder in association with the immunomodulating product;
- the experimental batch 2 (E2) - vaccinated and fed with mixed standard fodder;
- the experimental batch 3 (E3) - vaccinated and fed with mixed standard fodder in association with the immunomodulating product.

The immunomodulating product was administered in quantity of 250 ppm.

The immunomodulating effect of the probiotic was appreciated after the determination of the concentration of the unspecific immune effectors (lysozyme and serum properdine) and specific (the antibody titre). In order to carry out the serological testing, blood samples were gathered from 10 chicks from each batch, as following:

R1 - the day of the inoculation
R2 - seven days after inoculation
R3 - fourteen days after inoculation
R4 - twenty-one days after inoculation

The samples gathered were tested in the local laboratory of Immunology and Immunopathology of FMV Timisoara.

The serum lyzozyme was determined by using the simple radial spreading test, in agar gel 2% in which was planted a culture of Micrococcus lysodeicticus. The diameter of the area of lysis of the germs included in the environment is directly proportional to the concentration of the serum lysozyme.

The quantification of the antibodies against the virus of New Castle disease has been performed through the reaction of inhibition of hemaglutinins (IHA).

The chicks, from the four batches have been checked daily, checking on general status and mortality.

3. RESULTS AND DISCUSSIONS

The immune reaction of the chicken is conditioned by the morphological and functional integrity of the immune system, in which the bursae of Fabricius plays an essential part.

The three parameters monitored indicated relevant values, the results obtained being centered and systematized in tables and graphics.

Analyzing the post inoculation reaction evolution, regarding the unspecific immune parameters, for the batches of poultry taken into study, were noticed significant differences from one batch to another and from an immune parameter to another.

In what regards the serum lysozyme, the higher values were registered in the vaccinated batches E2 and E3, with the mention that the maximal value 54,65 µg/cm3 was registered in batch E3, three weeks after vaccination (table 1). High values were noticed also in batch E1, unvaccinated but which received in its food probiotics, which proves the immunomodulating part of the probiotics.

Table 1.

The average values of the serum lysozyme

Parameter	Batch	Experimental period			
		Initially (R_1)	After 7 days (R_2)	After 14 days (R_3)	After 21 days (R_4)
Lysozyme (µg/cm$^{3)}$	M	10,65	12,30	13,55	14,95
	E_1	12,45	19,60	25,30	25,20
	E_2	14,05	39,70	31,90	31,40
	E_3	22,50	41,80	47,40	54,65

The properdine concentration proved a resembling dynamics, with the mentioning that the registered values for the vaccinated batches were quite similar between them but significantly greater than in batch E1, unvaccinated batch who received in food the immunomodulating substances.

Thus, the properdine concentration increases from 18,30 mg/100 ml serum to 29,98 mg/100 ml serum, after 21 days from vaccination in the case of batch E1, while the maximal value is registered in batch E2, after 21 days from vaccination (35,93 mg/100 ml serum).

To underline that the maximal value was registered in batch E2 not in batch E3, vaccinated batch to which was administered also the immunomodulating substances which proves that the probiotic influences on a smaller scale the synthesis of properdine as compared to the lysozyme (table 2).

Table 2.

Average values of serum properdine

Parameter	Batch	Experimental period			
		Initially (R_1)	After 7 days (R_2)	After 14 days (R_3)	After 21 days (R_4)
Properdin (mg/100 ml ser)	M	17,60	17,93	16,58	19,84
	E_1	18,30	20,67	27,65	29,98
	E_2	22,10	23,33	32,26	35,93
	E_3	22,05	24,61	31,37	35,03

The presence of specific antibodies, shown through the reaction of inhibition of hemaglutination is presented in table 3. The antibody concentration, expressed in hemaglutionation inhibition units, increased progressively in all chicks from the vaccinated batches, but the higher values were registered after 14 days from vaccination. The highest average value (144,0) was registered in batch E3 at 21 days after vaccination, while in batch E2, vaccinated batch, which did not receive probiotic, the titre was 137,6 (UIHA).

Considering the superior results obtained in batches to which probiotic was administered, regarding both the unspecific immune effectors and specific immune effectors, we consider that the probiotic has a benefic effect on the immune reactivity in poultry.

Table 3.
The effect of the probiotic on the antibody synthesis

The collecting	Concentration of antibodies (D.O.)			
	Witness batch	Batch E_1	Batch E_2	Batch E_3
R_1	167,9	211,8	198,7	242,5
R_2	174,6	219,3	216,3	278,2
R_3	235,3	279,9	334,1	472,1
R_4	258,5	332,4	355,5	401,3

4. CONCLUSIONS

The laboratory tests performed have proved a progressive increasing of all the determined immune effectors, the maximal values being registered in poultry from batch E3. Puoltryzyme TM 250 stimulates especially the production of lysozyme and on a smaller scale the production of serum properdin.

The antibody titre presents a pronounced individual variability and maintains at significant values for over 21 days from vaccination.

The results obtained prove the immunomodulating effect of probiotic Poulrtyzyme TM 250 both on the specific immune system and unspecific one.

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