The influence of the low-intensity electromagnetic radiation of mm-range on the ulcerogenic damages, induced by chronic hypokinetic stress

M Yu Ravaeva1*, E N Chuyan1 and S Ye Chornobay1

1 V.I. Vernadsky Crimean Federal University, Simferopol, the Republic of Crimea, the Russian Federation

*ravaevam@yandex.ru

Abstract. The antiulcerogenic effectiveness of the low-intensity electromagnetic radiation of the millimetric range was studied in the model of chronic hypokinetic stress. It was found out that chronic hypokinetic stress causes the development of the ulcerogenesis, the intensity of which depends on the duration of the stress factor, which for animals is being in hypokinesia. Along with this, the maximum ulcerogenic damages are registered after 10 days of the mobility restriction; this is evident by the increase of the affected animals’ number, and by the growth of Pauls’ index by 586 % (p≤0.05) in comparison with the first day of the mobility restriction. The application of the low-intensity electromagnetic radiation of the millimetric (mm) range has the gastroprotective effect in the conditions of hypokinesia. The effect depends on the length of the impact, the maximum ulceroprotective effect was observed after the ten-fold mm-impact: the number of the affected animals decreases by 50 %, Pauls’ index decreases by 70 % (p≤0.05) in relation to the same in the group of animals, having undergone the hypokinesia, and the index of antiulcerogenic activity of mm-impact was 3.3, (>2), this indicates the occurrence of the antiulcerogenic activity of the mm-radiation.

1. Introduction
Stress ulcers are the gastritis or gastropathy, caused by stress; it occurs when the gastric and sometimes esophageal or duodenal barrier of mucous membrane is broken owing to acute stressor damage or severe disease [1, 2]. The death rate of the patients without surgery is almost 100% [3, 4]. In relation to this the therapy and prophylaxis of the stress ulcer is the center of the discussion for many national and international societies of the intensive therapy. However, till nowdays the recommendations concerning the indications, the choice of the medicine, the therapy length, and prophylaxis peculiarities are not clear [5]. Many researchers are inclined to take “aggressive prophylactic measures” for the corresponding population of patients with the risk of stress ulceration development; this indicates the importance of prophylaxis, which is the cornerstone in the treatment for gastropathy, caused by stress [6, 7].

In our previous work [8] we researched the antiulcerogenic effectiveness of the electromagnetic radiation of extremely high frequency (EMR EHF) or the millimetric (mm) range during the preventive and combined with the acute stress action. It was shown that mm-impact restricts the development of all the ulcerous destructions of the stomach mucous membrane and limits the stress-reaction development in the test of the forced swimming. These findings lead us to the further research
of antilulcerogenic effectiveness of mm-impact during the development of chronic hypokinetic stress or hypokinesia (HK), which is the objective of the given research.

2. Materials and methods

2.1. Animals
All the researches on the animals were carried out according to the principles set out in the Directive 2010/63/EU of the European Parliament and of the EU Council of 22.09.2010 on the protection of animals used for scientific purposes.

2.2. Design of research
The experiment was performed on 120 sexually mature male laboratory Wistar rats, weighing 200-220 gr, which were kept under the vivarium conditions with the natural 12-hour day-and-night cycle at the temperature of 18–22°C with free access to water and full-fledged granulated food (the State Standard 33215-2014).

For the experiment the animals of the same age were selected; they were characterized by an average motor activity and low emotionality in the “open field” test, and they made up the majority of the population. Such a selection allowed forming homogeneous groups of animals with the same type peculiarities and which react identically to the action of many factors.

After the preliminary selection, the animals were divided into 4 groups of 30 individuals in each:

- the animals of Group 1 were the biological control and were in the ordinary vivarium conditions – K;
- the animals of Group 2 were under constant chronic hypokinetic stress – HK;
- the animals of Group 3 were affected by the isolated mm-impact – MM;
- the animals of Group 4 were under the combined action of the chronic hypokinetic stress (1-10 days of the experiment) and MM-impact (1-10 days of the experiment) – HK+MM.

The chronic stress was modelled by the restriction of the mobility (hypokinesia, HK), which was achieved by putting the rats into special cartridges made from perspex (140 × 60 × 60 mm for each rat), where they were during 10 days for 20 hours. During the remaining 4 hours the experimental researches, the mm-impact (for the animals of Group 4), feeding and grooming were done. It is known that the restriction of the rats’ mobility in the cages-cartridges causes the stress reaction, the intensity of which depends on the degree of HK “rigidity”. The obtained experimental model allowed creating the same degree of the “rigidity” of HK for all the animals; this is the necessary requirement for obtaining the comparable results.

MM-impact was done by the apparatus of EHF-therapy “EHF-ND”. The technological characteristics of the generator are the following: the working length of the wave is 7.1 mm, the density of the radiation power flow is 4-12 mVt/cm² (Production: LLS “Scientific commercial firm RESLA”, Russia, Declaration of conformity № ROSS Ru. ME67.D00227; the registered ID № FSR 2007/00763 from 18.09.2007). The impact was done during 30 minutes on the cervical-neck area daily in the morning during 10 sessions.

After ten-day’s HK and ten-fold mm-impact the animals were killed. The slaughter of the animals was done under the ether narcosis by the method of cranio-cervical dislocation. After that the animals’ stomachs were retrieved and opened by the lesser curvature and washed thoroughly with the cold physiological solution. Then, macroscopically with the help of the magnifying glass under the bright lighting, the following signs of the damages were indicated:

- the general area of the pointed ulcerous damages, mm square (the area of each is taken being equal to 1 mm²);
- the general area of erosive damages, mm square (in the case of linear erosions they were measured by the length (in mm), and the width was taken being equal to 1 mm).

During the research results’ processing the following aspects were taken into consideration: an average number of ulcerations per one animal in the group, the percentage of the animals with ulcers...
and erosions, as well as Pauls’ index and antiulcerogenic activity of the experimental factors were calculated. Pauls’ index (PI) was defined as the integral index of the number of destructions according to the formula:

$$PI = \frac{\text{general amount of ulcers} \times \text{the amount of animals with ulcers}}{100}$$

(1)

The antiulcerogenic activity (AA) of the experimental impact was determined as the ratio of Pauls’ index in the control group to Pauls’ index in the experimental group. Along with this, if AA≥2, it indicated the presence of the acting factor’s antiulcerogenic effect.

2.3. Statistical methods and analysis

The statistic processing of the results was done by using the program package StatSoft \ STATISTICA 8. The credibility of the statistical differences between the experimental groups was determined by the Whitney-Mann criterion, the data are presented as an average ± a standard mistake.

3. Results

The carried out experiments showed that the animals from the control group did not have the ulcerogenic damages. At the same time the animals from Group HK underwent the following changes: after 24 hours of producing the stressor factor and, correspondingly, after the first day of animals being in hypokinesia, 50 % of animals demonstrated the ulcerogenic damages of the stomach mucous membrane. The average area of damage was 3.2±1.15 mm², Pauls’ index was equal to 1.6±0.58 (Table 1). On the 5th day of HK the ulcerogenesis was registered in 100% of animals, the average number of ulcers increased by 168 % (p≤0.05), and PI increased by 437 % (p≤0.05) in comparison with the same of the animals after the 1st day of HK. After 10 days of HK the average number of ulcers increased by 243 % (p≤0.05), and PI increased by 587 % (p≤0.05) in comparison with the same of the animals after the 1st day of HK.

Thus, the chronic hypokinetic stress leads to the damage of the animals’ stomach mucous membrane, the intensity of which depends on the length of the stressor factor’s presence – animals’ being in the conditions of the mobility restriction. The maximum ulcerogenic damages are registered after 10 days of the mobility restriction.

The animals of Group 3, which were affected by ten-fold mm-impact during all the research period, did not expose ulcerations of the stomach mucous membrane.

In the animals of Group 4 a successive mm-impact with HK-stress decreased the number of the animals with ulcerogenesis: in the group HK+MM it was 30 % (under HK-stress this index was 50 %), and by the 10th day it increased up to 50 %, this is twice less than in the HK group of animals (see Table 1, Figure 1).

Besides, MM-impact restricted the development of all types’ ulcerous destructions already after the first session; the antiulcerogenic effectiveness depended on the repetition factor of the impact (see Table 1). So, the antiulcerogenic effect was observed after single mm-impact, this was evident by the 40 % decrease of the general number of stomach mucous membrane (SMM) damages due to the decrease, first, of the area of pointed ulcerous damages (by 59 %) in comparison with the indices of the animals in HK group. Pauls’ index after single mm-impact decreased for sure and was 35.6 % (p≤0.05) in comparison with the same in HK group. The index of antiulcerogenic activity of mm-impact was 2.8 (>2), which indicates the presence of antiulcerogenic activity of mm-radiation.
Table 1. The indices’ dynamics of ulcerous destructions under the experimental chronic stress and its combination with mm-impact (x±Sx).

| Group       | The number of rats with ulcers, % | The area of ulcers, mm² | The area of erosions, mm² | The area of damages, mm² | Pauls’ index | Antiulcerogenic activity |
|-------------|----------------------------------|-------------------------|--------------------------|-------------------------|---------------|--------------------------|
|             | 1st day                          |                         |                          |                         |               |                          |
| Control     | 0                                | 0                       | 0                        | 0                       | 0             | -                        |
| MM          | 0                                | 0                       | 0                        | 0                       | 0             | -                        |
| HK          | 50                               | 1.2±0.63                | 2.0±0.94                 | 3.2±1.15                | 1.6±0.58      | -                        |
| HK+MM       | 30                               | 0.50±0.34               | 1.4±0.67                 | 1.9±0.85                | 0.57±0.25     | 2.81                     |
|             | 5th day                          |                         |                          |                         |               |                          |
| Control     | 0                                | 0                       | 0                        | 0                       | 0             | -                        |
| MM          | 0                                | 0                       | 0                        | 0                       | 0             | -                        |
| HK          | 100                              | 3.7±0.83                | 4.9±0.46                 | 8.6±0.93                | 8.6±0.93      | -                        |
| HK+MM       | 50                               | 1.3±0.56                | 2.9±0.97                 | 4.2±1.11                | 2.1±0.55      | 4.09                     |
|             | 10th day                         |                         |                          |                         |               |                          |
| Control     | 0                                | 0                       | 0                        | 0                       | 0             | -                        |
| MM          | 0                                | 0                       | 0                        | 0                       | 0             | -                        |
| HK          | 100                              | 3.0±0.3                 | 8.06±0.67                | 11.0±0.8                | 11.0±0.8      | -                        |
| HK+MM       | 60                               | 2.0±0.56                | 4.7±1.30                 | 6.7±1.53                | 4.02±0.92     | 3.28                     |

Note: \( p_{HK} \) is the level of credibility of animals’ differences indices in the animals of HK group on the corresponding day of the experiment in comparison with the same on the 1st day of HK; \( p_{HK/MM} \) is the level of credibility of indices’ differences according to Whitney-Mann criterion between the animals of groups HK and HK+MM on the corresponding day of the experiment. HK is the chronic hypokinetic stress, MM is mm-impact.

On the 5th day of the combined action of hypokinesia and mm-impact the credible decrease of all the indices of stomach mucous membrane ulcerous destructions was observed. Besides, there was the most significant decrease in the number of pointed ulcers (by 65 %, \( p \leq 0.05 \)), PI was 24.4 % (\( p \leq 0.05 \)) in comparison with the same in HK group of animals (see Figure 1).
Figure 1. The dynamics of the ulcerous destructions indices in the animals of HK+MM group under the experimental chronic hypokinetic stress and its combination with mm-impact in comparison with the same, taken as 100 %, in HK group.

Note: * - credibility of the indices’ differences in HK+MM group in comparison with the same in HK group.

By the 10th day of HK and, correspondingly, after ten-fold mm-impact there was a credible decrease in the number of linear erosions (by 41 %, p≤0.05), and in the average number of SMM damages (by 39.1 %, p≤0.05), however, the number of pointed ulcers did not change for sure. PI was 30.4 % (p≤0.05) in relation to the same in HK group (see Figure 1).

Thus, the impact of EMR mm-range has a gastroprotective effect, the intensity of which increases with the growth of mm-impact sessions’ repetition factor. The index of antiulcerogenic activity is in favor of this; after single mm-impact this index was 2.8, after the 5th session of mm-impact it was 4.1, and after the 10th session it was 3.3.

4. Discussion
The results of the given research indicate that mm-impact has an evident gastroprotective effect during the chronic stress. Ten-fold mm-impact lowers the amount and the character of ulcer-erosive damages and prevents the development of the dystrophic changes in the mucous membrane.

Thus, the mm-impact can be referred to the groups of measures, increasing the regenerative activity of the stomach mucous membrane and its resistance to the aggressive factors.

Our previous researches show that EMR of mm-range has an evinced antistressor effect, the basis of which is the decrease of hyperactivity of stress-implementing and activation of stress-limiting systems [9]. The results of the given research confirm and complement the data on antistressor action of EMR mm-range, proving its antiulcerogenic activity. The obtained data prove the reasonability of applying mm-impact as stress- and gastroprotective medicine in the ulcer therapy; the ulcer is the most wide-spread disease of the digestive system, it first damages the people, who are of the active and able-bodied age, and quite often it leads to the disability.

5. Conclusion
1. Chronic hypokinetic stress leads to the development of ulcerogenesis, the intensity of which depends on the length of the stressor factor. The maximum ulcerogenic damages are registered after 10
days of mobility restriction. This is indicated by the increase of the affected animals (100 %), Pauls’ index growth by 586 % (p≤0.05) in comparison with the day’s hypokinesia.

2. Low-intensity mm-radiation has a gastroprotective effect, depending on the repetition factor; the maximum effect was observed after ten-fold mm-impact: Pauls’ index decreased by 70 % (p≤0.05) in relation to the same in HK group; and the index of antiulcerogenic activity of mm-impact was 3.3, (>2), which indicated the presence of mm-radiation antiulcerogenic activity.

6. References
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