Comparative experimental evaluation of the bactericidal effect of an antiseptic „ANK-neutral”

1Terekhov G., 2Chukhraev N., 3Zukow W., 1Savitska I., 2Shcherbakov S., 2Sytnik V.

1National Institute of Surgery and Transplantation after A.A. Shalimov NAMS (National Academy of Medical Sciences) of Ukraine, Kyiv, Ukraine
2ANK Technologies LLC, Kyiv, Ukraine
3Nicolaus Copernicus University in Torun, Torun, Poland

Annotation.

The article presents the results of an experimental study of the bactericidal action of the disinfectant "ANK-neutral" on bacterial cultures: ESCHERICHIACOLI, KLEBSIELLA PNEUMONIAE, PSEUDOMONAS AERUGINOSA, STAPHYLOCOCCUS AUREUS, CANDIDA ALBICANS. Options for the use of "ANK-neutral" for preventive and therapeutic purposes are proposed.

Key words: infectious diseases, disinfectants, ANK-neutral.
According to the WHO (World health organization) experts, infectious diseases continue to be a serious health problem and the fight against them, in many countries, absorbs most of the budget, allocated to health care. Acute diarrheal diseases are responsible for more than 30% of deaths among children under 5 years of age, at least 4 million people die from such diseases each year. Acute respiratory infections (primary pneumonia) are another very important cause of death. Every year, more than 2.5 million people die prematurely from them in the world.

No less dangerous is the infection of wounds, which leads to significant lethal losses in the conditions of combat operations.

The danger of epidemic or even pandemic spread of infectious diseases caused by viral or bacterial agents may be due to both reasons - inadequate epidemiological surveillance and the lack of adequate measures for the prevention and treatment of such diseases.

The aim of the work is an experimental study of the bactericidal effect of the disinfectant "ANK-neutral" on bacterial cultures: ESCHERICHIA COLI, KLEBSIELLA PNEUMONIAE, PSEUDOMONAS AERUGINOSA, STAPHYLOCOCCUS AUREUS, CANDIDA AlbICANS and the development of methods for its use for disinfection.

For carrying out the research, the disinfectant "ANK neutral" (LLC "ANK technologies", Kyiv, Ukraine), the disinfectant "Terralin® protect" (“Schülke and Mayr” GmbH, Germany) and the disinfectant "Clean-stream" were used as the base (LLC “Clean Stream”, Ukraine).
The choice of disinfectants was carried out on the basis of studying the information about the most effective bactericidal agents currently used in medical institutions in Ukraine.

Based on the analysis of the disinfectants, the following were selected:
- "Terralin® protect" ("Schülke and Mayr GmbH", Germany);
- "Clean-stream" (LLC "Clean Stream", Ukraine);
- "ANK-neutral" (LLC "ANK technology", Ukraine).

According to professor Leonov B.I. co-author. (1991-2019) ANK, unlike 0.5 - 5.0% hypochlorite solutions, which have only a disinfecting effect, is also a sterilizing solution with an oxidant concentration in the range of 0.005 - 0.05%. Even after relaxation time, ANK anolyte is a much stronger antimicrobial agent than sodium hypochlorite [1].

The sum of active oxygen and chlorine compounds in ANK (total content of oxidants) ranges from 100 to 500 mg/l, which is tens times less than in most working solutions of modern disinfectants. ANK does not cause coagulation of the protein that protects microorganisms and, due to its loose structure, easily penetrates into the microchannels of living and inanimate matter. ANK is non-toxic, due to the low concentration of active substances, therefore, it does not require removal from the treated surfaces after the end of the treatment.

An important conclusion follows from this: it is possible to exclude the development of resistance of microorganisms to a liquid antimicrobial agent only by using solutions with metastable active substances, the spontaneous decomposition of which during exposure ensures the multiplicity and unpredictability (for microorganisms) of the development pathways of reactions that disrupt their vital processes.

RESEARCH METHOD.

The assessment of the bacteriological effect of the studied antiseptic agents was carried out using the “cups” method [2-7].

To conduct research, microbes were cultivated according to the method of R. Koch using the nutrient medium "AGAR-AGAR". The use of agar gels in
microbiology is unique because of their great resistance to enzymatic degradation, not characteristic of other gel formers. The ability to form gels in the absence of cations allow the use of agars to create a nutrient medium with controlled osmotic pressure. Due to the special properties of physical gels derived from agar-agar, the gelation and melting temperature point, as well as their huge hysteresis and reversibility, their use allows to get the most reliable results.

For experimental studies, 5 sets of 5 Petri dished with prepared nutrient medium based on "AGAR-AGAR" were used. Bacterial cultures: ESCHERICHIA COLI, KLEBSIELLA PNEUMONIAE, PSEUDOMONAS AERUGINOSA, STAPHYLOCOCCUS AUREUS, CANDIDA ALBICANS were added dropwise into the standard nutrient medium "AGAR-AGAR" in the amount of 0.5 (ICF - International Classification of Functioning).

After inoculation, a "cup" made of a biologically inert material - stainless steel with a diameter of 5 mm and a height of 15 mm was placed in the center of the Petri dish.

0.5 ml of the appropriate disinfectant was added into the “cups”. For the treatment of bacteria of the 1st sample (ANK-1), an ANK-neutral solution prepared during the day was used as a disinfectant. For the treatment of bacteria of the 2nd sample (ANK-2), an ANK-neutral solution 8 days after its production was used as a disinfectant. For the treatment of bacteria of the 3rd sample (ANK-3), an ANK-neutral solution 15 days after its production was used as a disinfectant. For the treatment of bacteria of the 4th sample - "Terralin® protect" was used. For the treatment of bacteria of the 5th sample, the “Clean Stream” solution was used as a disinfectant.

After applying the bacterial cultures and filling the “cups” containing solutions for disinfection, the Petri dishes were placed into a thermostat for a 24 hours at a temperature of 37°C. A day later, the result was evaluated by measuring the diameter of the absence of growth of the bacterial agent around the "cup".

The work was performed on the basis of the bacteriological research department of the National Institute of Surgery and Transplantation named after
A.A. Shalimov of the National Academy of Medical Sciences of Ukraine by the staff of the NIST after A.A. Shalimov NAMS of Ukraine and “ANK-Technologies” LLC. In the process of conducting an experimental study, a comparative assessment of the effectiveness of using the ANK-neutral disinfectant (“ANK Technologies” LLC, Kyiv, Ukraine) depending on the time of its production, the “Terralin® protect” disinfectant (“Schülke and Mayr” GmbH, Germany) and the “Clean-stream disinfectant” (LLC “Clean Stream”, Ukraine) was carried out.

**Results and their discussion.**

The results of the conducted studies are presented in table 1.

<table>
<thead>
<tr>
<th>Solutions for disinfection</th>
<th>ANK-1</th>
<th>ANK-2</th>
<th>ANK-3</th>
<th>Terralin</th>
<th>Clean-stream</th>
</tr>
</thead>
<tbody>
<tr>
<td>Escherichia coli</td>
<td>46</td>
<td>60</td>
<td>36</td>
<td>22</td>
<td>31</td>
</tr>
<tr>
<td>Klebsiella pneumoniae</td>
<td>54</td>
<td>60</td>
<td>44</td>
<td>30</td>
<td>20</td>
</tr>
<tr>
<td>Pseudomonas aeruginosa</td>
<td>35</td>
<td>60</td>
<td>44</td>
<td>40</td>
<td>45</td>
</tr>
<tr>
<td>Staphylococcus aureus</td>
<td>40</td>
<td>48</td>
<td>42</td>
<td>35</td>
<td>25</td>
</tr>
<tr>
<td>Candida albicans</td>
<td>56</td>
<td>70</td>
<td>58</td>
<td>36</td>
<td>44</td>
</tr>
</tbody>
</table>

**Conclusions and practical recommendations:**

1. Conducted bacteriological studies have proven the advantages of "ANK-neutral" in relation to the generally accepted means of disinfection in medical institutions of Ukraine.

2. Completed clinical studies have proven the possibility and high efficiency of using "ANK-neutral" in aerosol treatment of wounds and/or wound surfaces.

3. According to the results of experimental and clinical studies on laboratory animals, "ANK-neutral" can be recommended for use in surgical and outpatient practice.
Literature.