SENSITIVITY AND RESISTANCE OF ANAEROBIC MICROORGANISMS TO ANTIBIOTICS  
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SENSITIVITY AND RESISTANCE OF ANAEROBIC MICRO-ORGANISMS TO ANTIBIOTICS

K. A. Sarkisov and F. I. Fagan

PROTECTIVE INOCULATION OF SWINE AGAINST HOG CHOLERA, Erysipelas AND LEPTOSPIROSIS

A. I. Ulendeyev and A. P. Stepochkin

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1. To establish the sensitivity of anaerobic microorganisms, foreign and local researchers used a series of mixtures in liquid medium, and the disc method on hard agar. Most researchers show that the bacteriolytic dose of antibiotics to anaerobic microorganism increases greatly the bacteriostatic concentration. Many antibiotics are universal and deaden, in small concentrations the growth of the majority of the stimulants of anaerobic infections.

2. Microscopic tests of smears prepared from lungs of deceased pigs (mainly from minute bronchial contents) painted with Romanov stains, continuously showed microorganisms typical to leptospirosis. Besides erysipelas and leptospriosis, paratyphoid was found primarily in new born pigs.
<table>
<thead>
<tr>
<th>KEY WORDS</th>
<th>LINK A</th>
<th>LINK B</th>
<th>LINK C</th>
</tr>
</thead>
<tbody>
<tr>
<td>inoculation</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>leptospirosis</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>microorganisms</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>erysipelas</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>paratyphoid</td>
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</table>
SENSITIVITY AND RESISTANCE OF ANAEROBIC MICROORGANISMS TO ANTIBIOTICS

Antibiotics are used against diseases involving anaerobic microorganisms. However, published literature has shown that antibiotic resistance in bodies of anaerobic microorganisms are markedly lower than in aerobic. This has been established after considerable work with anaerobic microorganisms linked with their breathing capacities. They develop under low acid-establishing potentials.

To establish the sensitivity of anaerobic microorganisms, foreign and local researchers used a series of mixtures in liquid medium, and the disc method on hard agar.

Authors maintain different opinions on the effects of the same antibiotics on aerobic microorganisms. A. K. Ageyev (1951) informed about the action of penicillin on Clostridium perfringens and Clostridium oedematos; others (M. R. Nechaevskaya 1956, N. G. Chernikova, 1950, N. F. Kalinichenko, 1957) did not mention such actions on the above microorganisms.

N. F. Kalinichenko (1957) Bittner and co-authors (1961) showed that combinations of antibiotics have stronger synergetic action. Iveland and co-authors (1955) noted that, while using two antibiotics, the weaker of the two did not increase nor did it decrease the activity of the stronger antibiotic. M. R. Nechaevskaya (1956) in her "in vivo" experiments established that for the treatment of experimental diseases in animals, an increased dosage of some antibiotics are required because the albumens of the organism lower the activity of these medications.

However, N. F. Kalinichenko (1957) and others, feel that the addition of normal human and horse serum, as well as of whole blood, does not have any effect on the bacteriostatic and bacteriocytic titer of penicillin.
The majority of the researchers based on their tests show that the bacteriocytic dose of antibiotics to anaerobic microorganisms increases greatly the bacteriostatic concentration. Many antibiotics are universal and deaden, in small concentrations, the growth of the majority of the stimulants of anaerobic infections.

THE SENSITIVITY OF STIMULANTS OFANAEROBIC INJECTIONS TO CERTAIN ANTIBIOTICS

<table>
<thead>
<tr>
<th>Type of Microbe</th>
<th>Penicillin</th>
<th>Streptomycin</th>
<th>Cytomycin</th>
<th>Chlorotetra mycin</th>
<th>Erythromycin</th>
<th>Authors, year</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cl.chauvoei</td>
<td>0.011-0.03</td>
<td>-</td>
<td>-</td>
<td>0.1-0.5</td>
<td>0.011-0.08</td>
<td>Trishkina,E.T.1961</td>
</tr>
<tr>
<td>(0.05)</td>
<td></td>
<td>2.5</td>
<td></td>
<td>0.05</td>
<td></td>
<td>Nilikantan 1965</td>
</tr>
<tr>
<td>Cl.septicum</td>
<td>0.6</td>
<td>-</td>
<td>-</td>
<td>0.4-0.8</td>
<td>0.3-2.5</td>
<td>Kagan, F.I. 1955</td>
</tr>
<tr>
<td>(0.02-0.07)</td>
<td>-</td>
<td>-</td>
<td>0.2-0.5</td>
<td>-</td>
<td></td>
<td>Safarov, U.B. 1961</td>
</tr>
<tr>
<td>(0.15)</td>
<td>250</td>
<td>-</td>
<td>-</td>
<td>0.05</td>
<td></td>
<td>Trishkina,E.T.1961</td>
</tr>
<tr>
<td>Cl.histolyticum</td>
<td>0.6</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>0.1</td>
<td>Nilikantan 1965</td>
</tr>
<tr>
<td>(0.025)</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td></td>
<td>Trishkina,E.T.1961</td>
</tr>
<tr>
<td>Cl.botulinum</td>
<td>1.25</td>
<td>-</td>
<td>10</td>
<td>-</td>
<td>1.25</td>
<td>Kieser 1952</td>
</tr>
<tr>
<td>(0.2 ed/ml)</td>
<td>-</td>
<td>5 ed/ml</td>
<td>5 ed/ml</td>
<td>5</td>
<td></td>
<td>Kieser 1952</td>
</tr>
<tr>
<td>Cl.perfringens</td>
<td>0.02</td>
<td>-</td>
<td>2.5</td>
<td>-</td>
<td>0.02</td>
<td>Kagan, F.I. 1955</td>
</tr>
<tr>
<td>(0.07-0.5)</td>
<td>-</td>
<td>1:32 000</td>
<td>1.64 000</td>
<td>0.02</td>
<td></td>
<td>Kagan, F.I. 1955</td>
</tr>
<tr>
<td>(0.3)</td>
<td>250</td>
<td>-</td>
<td>-</td>
<td>0.02</td>
<td></td>
<td>Larina, I.A.1958</td>
</tr>
<tr>
<td>(0.02)</td>
<td>-</td>
<td>2.5</td>
<td>0.16</td>
<td>-</td>
<td></td>
<td>Trishkina,E.T.1961</td>
</tr>
<tr>
<td>Cl.tetani</td>
<td>0.6</td>
<td>-</td>
<td>-</td>
<td>0.08</td>
<td>0.01</td>
<td>Ahmed 1956</td>
</tr>
<tr>
<td>(0.1 ed/ml)</td>
<td>10-50</td>
<td>-</td>
<td>-</td>
<td>0.16</td>
<td>0.16</td>
<td>Shabell 1959</td>
</tr>
<tr>
<td>Bacterium</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>1.64 000</td>
<td></td>
<td>Kagan, F.I. 1955</td>
</tr>
<tr>
<td>necrophorum</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>0.03-0.02</td>
<td></td>
<td>Volkova, A.A. 1965</td>
</tr>
</tbody>
</table>
The following, also, belong to the above mentioned group: Erythromycin (Grundi 1955, E. T. Trishkina 1961) the Tetracycl-
*antibiotic family and especially Chlorotetracycline (Kieser and coauthors 1952); Iveland 1955; F. I. Kagan 1955; Thomas 1962; Nilakantar: 1965; Chloramphenicol (Maiselle 1954; Bittner 1962 and others) although some of the researchers discount the high bacteriological activity of the last antibiotic mentioned (Kieser, Iveland).

Anaerobic microorganisms, however, are immune to the following antibiotics: Streptomycin (Maiselle 1954; Shaibell 1959; A. M. Pasechnik 1958; Thomas 1962) Sanasin (N. F. Kalinichenko 1957) polym (Santo 1962) Kanamycin, Novomycin and Nemoicin (Thomas 1962). Through manure tests, some authors discovered resistance of some microbe bodies to these antibiotics.

I. A. Larina (1958) through her experiments proved that Grisemin even in large doses generally does not have bacteriocytic effects on anaerobic microorganisms.

According to data from C. G. Barskaya and Robert (1955) freshly developed anaerobic bodies are more susceptible to antibiotics than museum cultures. According to Steli (1947), Katishu (1953), N. F. Kalinichenko (1955 and M. R. Khechayevskaya (1956), toxins of anaerobic microorganisms are secure against antibiotic actions.

In spite of the variety of techniques used by foreign and Soviet researchers in determining the sensitivity of anaerobic microorganisms to antibiotics, it can be noted that stimulants of malignant inflammation (Cl. septicum, Cl. perfringens, Cl. oedematis, Cl. Histolyticum and others) are more sensitive to antibiotics (C. G. Barskaya, 1948); A. A. Agayev, 1956 and others) than stimulants of microbacilli (Y. R. Kovalenko, 1955; F. I. Kagan 1955; R. A. Ahmedjanov, 1956; A. A. Volkov, 1955) Tetanus (Ito 1951: Maiselle 1954; Ahmed 1956; G. I. Kremnev 1965) and Botulism (Kieser 1952; Anderson 1953).

Growing spores are more sensitive to Penicillin than spores in a quiet stage (Lund 1953). These tests have been confirmed by tests made by Maiselle (1954), Shaibell (1959) and others.

Foreign and Soviet scientific literature have published a series of works on the obtaining of resistance to antibiotics of bodies of anaerobic microorganisms under special circumstances through consecutive strains.

N. F. Kalinichenko (1955) in strains (up to 70 times) with penicillin, Sanacyn and Sintomycin, obtained highly resistant bodies of stimulants of gaseous gangrene; the resistance of which was 30 times higher than those of the original.
L. B. Pasachnik and A. P. Lisogor (1958) by means of lengthy strains (up to 70 times) discovered that Cl. perfringens types A and B bodies were 10 to 20 times more resistant to Streptomycin.

Bittner, after 36 passages of bodies of VP K Cl. perfringens type A in the Pope medium, with growing concentrations of penicillin, found the stabila variant of the body which during the following 55 passages through the same medium maintained a property characteristic of the L-forms and was biochemically, morphologically and virulently different from the parent body.

According to V. V. Kusmin (1962), G. V. Smith was able to isolate from feces of pigs that had received Terramycin, 58% microbe bodies, Cl. perfringens that were highly resistant to this antibiotic.

However, N. T. Kalinichenko advised (1957) that during isolation from dead laboratory animals of stimulants of anaerobic infections which were subjected to the effects of antibiotics, he did not get a single resistant body.

In the literature there is only a small number of published works on the discovery of experimental resistance of anaerobics and the research was done "in vitro".

That is why for the purpose of long range prognosis on the origin of anaerobic resistance in the organism of animals, a much more detailed research work is needed to be done through a more unified method.
PROTECTIVE INOCULATION OF SWINE AGAINST HOG CHOLERA, ERISIPELAS AND LEPTOSPIROSIS

The effectiveness of protective inoculations against infectious diseases depends not only on the bio-preparations, but, also, on how they are used. We arrived at this conclusion during an experiment in immunization against cholera, erisipelas and leptospirosis in hogs at the training farm of our Institute.

The hog research study farm was continuously unsuccessful with regards to the diseases of paratyphoid, leptospirosis and hog cholera. Although the plan of protective inoculations against these diseases was carried out annually, diseases among the hogs did not cease.

Grown pigs were inoculated against erisipelas with deposited vaccine twice a year. Little pigs were given inoculations 14 days after birth as shown in our table below. However, sickness was noticed to be quite frequent and only stopped after we started to immunize the animals in accordance with the scheme we worked out.

Leptospirosis in farm hogs was first established in 1959. Planned inoculations against this disease were started in 1960. The mother pigs and the breeder hogs were inoculated twice and the offspring after reaching one month were given vaccine once a year. The inoculations helped to liquidate abortions due to leptospirosis, but did little to help in the preservation of the little pigs. In 1960, deaths from pneumonia and gastro-intestinal diseases were 3.6% of the birth figures; in 1961 4.4%, in 1962 4.3% and in 1963 the death rate reached 11.7%.

Microscopic tests of smears prepared from lungs of deceased pigs (mainly from minute bronchial contents) painted with Romanowsky stains, continuously showed micro-organisms typical to leptospirosis. Besides erisipelas and leptospirosis, paratyphoid was found primarily in new born pigs.
In April of 1964, cholera threatened hog farming and all the animals were immunized against the disease. The animals were first inoculated with glycerined crystal-violet vaccine and after one month were given dry avirulent virus vaccine (ASV). From then on the animals were regularly given vaccine against cholera, erisipelas and leptospirosis (see table). When the pigs are 44 days old, the males are castrated and at 65 days of age they are taken away from the mothers.

During the period immediately after birth, as a preventive measure against coli-intestinal diseases and paratyphoid, the pigs are given Furasolidon 0.15 - 0.2 gm. to each pig three times a day for a period of three - four days. Little pigs are not vaccinated against paratyphoid.

The mother pigs and breeders are immunized against cholera once every ten months with ASV virus. The vaccination is given intramuscularly in a dose of 2 mil. milliliters in a solution of 1:100.

Mother pigs in their first pregnancy and breeders are given polyvalent vaccine against leptospirosis each time for one to one and a half months away from giving birth. The vaccine is given subcutaneously in two doses seven days apart: the first 3 mil. and the second 5 mil.

Against erisipelas the pigs are immunized after the age of one year, twice yearly during Spring and Fall.

Besides the prophylactic inoculations, the farms are subjected to veterinary-sanitation precautions (daily cleaning of the area, disinfection of sties at least once a month). The disinfection solution is made from a hot solution of 2% Soda into which 3% creoline has been added.

As a preventive measure against skin diseases, the breeding areas are washed twice in summer, with a warm solution of 2% creoline and 0.5% chloride.

The precautionary inoculation for animals shown in the tables has been used on the farm for three years. During this period the pigs have not once been sick with cholera; and sickness from erisipelas, leptospirosis and paratyphoid has dropped. Loss of pigs each year is diminishing and birth gains are seen every year.

During the past three years deaths as compared to previous years have been reduced 2.5 times and births have shown considerable increase.
<table>
<thead>
<tr>
<th>Age (days)</th>
<th>Innoculation</th>
<th>Biopreparation</th>
<th>Method Applied</th>
<th>Dose</th>
</tr>
</thead>
<tbody>
<tr>
<td>10</td>
<td>Against cholera</td>
<td>Serum, virus-vaccine</td>
<td>Subcutaneously</td>
<td>10-15 ml in culture of 1:50</td>
</tr>
<tr>
<td></td>
<td>(simultaneously)</td>
<td>ASV</td>
<td>Intramuscularly</td>
<td></td>
</tr>
<tr>
<td>30</td>
<td>Ditto, revaccination</td>
<td>Virus vaccine ASV</td>
<td>&quot;</td>
<td>2 ml. in culture of 1:100</td>
</tr>
<tr>
<td>40</td>
<td>Against leptospirosis</td>
<td>Polyvalent vaccine</td>
<td>Subcutaneously</td>
<td>1 ml.</td>
</tr>
<tr>
<td>47</td>
<td>&quot;</td>
<td>&quot;</td>
<td>&quot;</td>
<td>2 ml.</td>
</tr>
<tr>
<td>60</td>
<td>Against Erisipelas</td>
<td>Deposited vaccine</td>
<td>&quot;</td>
<td>0.3 ml.</td>
</tr>
<tr>
<td>72</td>
<td>&quot;</td>
<td>&quot;</td>
<td>&quot;</td>
<td>0.5 ml.</td>
</tr>
<tr>
<td>105</td>
<td>Against cholera (revaccination)</td>
<td>Virus ASV</td>
<td>Intramuscularly</td>
<td>2 ml. in culture of 1:100</td>
</tr>
<tr>
<td>120</td>
<td>Against Erisipelas (revaccination)</td>
<td>Deposited vaccine</td>
<td>Subcutaneously</td>
<td>0.3 ml.</td>
</tr>
<tr>
<td>132</td>
<td>&quot;</td>
<td>&quot;</td>
<td>&quot;</td>
<td>0.5 ml.</td>
</tr>
<tr>
<td>280</td>
<td>&quot;</td>
<td>&quot;</td>
<td>&quot;</td>
<td>0.3 ml.</td>
</tr>
<tr>
<td>292</td>
<td>&quot;</td>
<td>&quot;</td>
<td>&quot;</td>
<td>0.5 ml.</td>
</tr>
</tbody>
</table>