



**Conference** Paper

# Study of Antibiotic Resistance of the Oropharyngeal Hemolytic Microflora in Preschool Children

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#### Abstract

It is impossible to imagine modern medical practice without antibiotic therapy. However, the rapid development of the pharmaceutical industry expands free access of the population to antibacterial drugs. At the same time, the illiteracy of people with respect to the principles of rational antibiotic therapy also increases. The problem of microbial resistance to antibacterial drugs remains relevant to this day. Special attention should be paid to rational antibiotic therapy applied to children.

The purpose of this work was to study the resistance of hemolytic microorganisms, which are often the cause of upper respiratory infection in preschool children, to the main antibacterial drugs used in pediatric practice.

The results of this scientific research can be advisory and useful to pediatricians and other specialists whose professional activities are related to children's health.

**Keywords:** hemolytic active microorganisms, bacterial carriage, antibiotic resistance, children's health

#### 1. Introduction

Many parents consider antibiotics as a panacea. However, few of them are aware of how important it is to correctly select a drug depending on the pathogen type, drug dosage and treatment duration. In addition, in July 2014, I.A. Dronov, N.A. Geppe, A.B. Malakhov and others, conducted an interregional study among pediatricians and revealed their insufficient proficiency in antibiotic therapy [1]. Pediatricians tend to consider treatment of respiratory diseases as a routine task, for it is the most common pathology in children [2]. As a consequence, it is not infrequently in clinical practice that the nature of infectious agents is not established and, while acute respiratory viral infections (ARVI) are very common, ineffective antibiotics may be prescribed. Pediatricians prescribe antibacterial drugs in more than 80% of respiratory diseases while they

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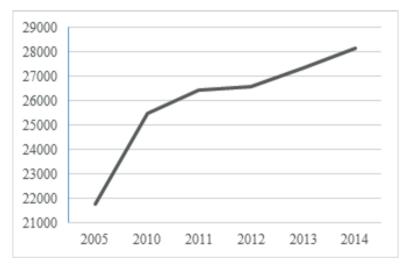
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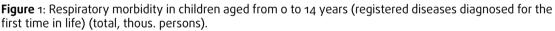




are really necessary only in 6–8% of cases [3, 4]. Together, these circumstances lead to a global problem, i.e., the development of microbial antibiotic resistance in children from an early age.

According to the Russian Federal State Statistics Service for 2015, the number of children aged from 0 to 14 years with newly registered respiratory diseases is continually increasing (Fig. 1) [5]. Respiratory infections are uppermost in the childhood morbidity patterns. According to the WHO for 2016, the second cause of infant mortality up to five years after the premature birth is pneumonia of bacterial etiology [6]. No treatment regimen for this disease is complete without antibacterial drugs.





The urgency of the problem of microbial drug resistance remains indisputable up to date. Against this background, the purpose of this scientific research was to study the resistance of hemolytic microorganisms, which are often the cause of upper respiratory infection in preschool children, to the main antibacterial drugs used in pediatric practice.

### 2. Materials and methods

The object of the study were hemolytic forms of microorganisms isolated from 51 children attending preschool educational institutions.

The isolated hemolytic strains of microorganisms were tested for their susceptibility to the following antibacterial chemotherapeutic agents: amoksiklav (group of inhibitorprotected penicillins); ceftazidime (group of third-generation cephalosporins); azithromycin (group of macrolides); clindamycin (group of lincosamides); co-



trimoxazole (a group of sulfanilamides). The drugs were selected based on the survey results and taking into account the clinical experience of pediatricians. At present, the following groups of drugs are primarily used for the treatment of upper respiratory tract infections in children:  $\beta$ -lactam antibiotics (inhibitor-protected penicillins, second-and third-generation cephalosporins) and macrolides. Alternatively, lincosamides can be used [2].

Sampling was carried out using the pharyngeal swab method. The sampling material was inoculated on the blood agar medium. The medium was prepared in advance according to the prescriptions from Order No. 535 [7]. The samples were incubated at 37 °C for 24 hours in a TSVL-160 thermostat (Kasimov).

After incubation of the cultures, the cultural properties of the grown colonies possessing hemolytic activity were taken into account, and the morphological properties of these microorganisms were examined.

In order to accumulate a pure culture of microorganisms, the examined colonies were reinoculated on the slant blood agar. The cultures were incubated at 37 °C for 24 hours in a thermostat.

The pure culture of isolated hemolytic microorganisms was identified according to the following biochemical properties: the ability to break down sugars, the production of cytochrome oxidase enzymes and catalase.

To determine the saccharolytic properties, the cultures were inoculated on Hiss media with 10% horse serum containing glucose, sucrose, fructose, maltose and lactose, followed by incubation in a thermostat at 37 °C.

Testing for cytochrome oxidase was carried out with the help of paper indicator systems; catalase was tested by a standard method using 3% hydrogen peroxide [7].

The sensitivity of the isolated hemolytic forms to the selected antibacterial agents was determined by diffusing to agar using standard paper discs [8]: amoksiklav (amoxicillin, 20  $\mu$ g + clavulanate, 10  $\mu$ g); ceftazidime (30  $\mu$ g); azithromycin (15  $\mu$ g); clindamycin (2  $\mu$ g); co-trimoxazole (trimethoprim, 1.25  $\mu$ g + sulfamethoxazole, 23.75  $\mu$ g).

### 3. Results and discussion

It has been established that about half of the children (49%) considered healthy at the time of visiting the kindergarten are carriers of hemolytic microorganisms, among which are both opportunistic and pathogenic species, mainly represented by cocci forms. Morphological, cultural and biochemical features of the isolated hemolytic active forms (Tab. 1) show that they belong to the species: *Staphylococcus aureus* 



(52%), Streptococcus pyogenes (12%), Streptococcus pneumoniae (8%), Neisseria perflava (12%), Aerococcus viridans (8%), Gemella haemolysans (8%).

TABLE 1: Morphological, cultural and biochemical features of experimentally isolated strains of hemolytic active microorganisms.

Feature	S.aureus	S.pyogenes	S.pneumoniae	N.perflava	A.viridans	G.haemolysans
Cell shape	cocci	cocci	cocci	cocci	cocci	cocci
Mutual arrangement	as a bunch of grapes	a chain	a couple	a couple	a couple or a tetrad	a couple or a tetrad
Tinctoriality	Gr+	Gr+	Gr+	Gr-	Gr+	Gr+
Hemolysis	β	β	α	β	α	α
Glucose	+	+	+	+	+	+
Sucrose	+	+	+	+	+	+
Maltose	+	+	+	+	+	+
Fructose	+	+	+	+	+	+
Lactose	+	+	+	-	+	+
Catalase	+	-	-	+	-	-
Cytochrome oxidase	-	-	-	+	-	-

As a rule, it is these representatives of microflora that cause inflammatory diseases of the upper respiratory tract in children when their immunity is weakened. In addition, it should be noted that almost all these representatives can cause infectious-allergic bronchial asthma in children with the impaired immune status. Unfortunately, the true infectious nature of catarrhal diseases and bronchial asthma is most often not investigated.

Based on the research findings, the most effective antibacterial drugs for opportunistic and pathogenic pharyngeal microbiota possessing hemolytic activity in children of preschool age are clindamycin, amoksiklav and azithromycin. In addition, the expressed resistance of several hemolytic forms of pharyngeal microorganisms is established in preschool children to modern antibiotics used in pediatrics, i.e.: *Staphylococcus aureus* is resistant to amoksiklav and ceftazidime (Fig. 2); *Streptococcus pyogenes* and *Gemella haemolysans* are resistant to ceftazidime and co-trimoxazole (Fig. 3, 4), *Streptococcus pneumoniae* and *Aerococcus viridans* are resistant to ceftazidime (Fig. 5, 6). *N.perflava* showed susceptibility to all the antibacterial drugs under study in different degrees of manifestation (Figure 7).





Figure 2: S.aureus hemolytic strain antibiotic susceptibility.



Figure 3: S.pyogenes antibiotic susceptibility.



Figure 4: G.haemolysans antibiotic susceptibility.



Figure 5: S.pneumoniae antibiotic susceptibility.

Symbols: 1. amoksiklav; 2. azithromycin; 3. ceftazidime; 4. co-trimoxazole; 5. clindamycin.





Figure 6: A.viridans antibiotic susceptibility.



Figure 7: N.perflava antibiotic susceptibility.

# 4. Conclusions

The confirmed resistance of opportunistic and pathogenic microorganisms to the antibiotics most often prescribed by pediatricians further exacerbates the problem with the incidence of upper respiratory tract infections and their treatment. The administration of a "useless" antibiotic, which is in fact a poison in a small dose, undermines the unformed immunity of a child and can have serious consequences. Thus, the problem of uncontrolled intake of antibiotics and non-compliance with the principles of rational antibiotic therapy still requires special attention.

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