Клінічна педіатрія / Clinical Pediatrics

UDK 616.98-053.2-08-039.57 *DOI: 10.22141/2224-0551.12.2.2017.99767*

O.M. Mukvich, T.M. Kaminskaya
State Institution "Institute of Pediatrics, Obstetrics and Gynecology of NAMS of Ukraine", Kyiv, Ukraine

Preventive and therapeutic effectiveness of bioflavonoids in children with recurrent respiratory infections

For cite: Zdorov'ye Rebenka. 2017;12:124-9. doi: 10.22141/2224-0551.12.2.2017.99767

Introduction

Health status analysis of the child population of Ukraine proves the current tendency towards a constant increasing proportion of school-aged children with recurrent and long-term acute respiratory infections (ARI) [6, 7]. Children suffering from common conditions of the upper respiratory tract – acute recurrent respiratory infections (ARRI) – pose a special problem for paediatricians and general practitioners. They account for the majority of visits to paediatricians, disruptions in school attendance, and hospitalizations; they are administered a wide range of drugs, but the lack of efficient and timely preventive measures in this category of children results in chronic conditions in 70.0% of them, among which the leading place is occupied by the otolaryngologic and bronchopulmonary diseases [13]. This group of children is very heterogeneous. It is necessary to distinguish those patients who have episodes of repeated viral infections, and children with chronic diseases of the ENT organs and respiratory tract, developmental anomalies of the respiratory tract, congenital or acquired immune dysfunctions, etc.

There is a fact to consider that the definition of "recurrent respiratory tract infections" (RRTI) today remains quite controversial, arbitrary and is too general in nature, with a lack of explicit medical consensus. For the RRTI diagnosis in children, we adhered to the recommendations of the Italian Society of Pediatric Allergy and Immunology with the detection of one of the following criteria [13]:

– ≥6 respiratory infections per year;

– ≥1 respiratory infection per month with the upper respiratory tract involvement from September to April;

– ≥3 respiratory infections per year with the lower respiratory tract involvement.

Data of currently conducted studies show that though the immune system of children with ARRI does not have rough primary and acquired defects, it is characterised by extreme intensity of the immune response system, intercellular cooperation violation and insufficiency of reserve capacity, which are the consequence of a long-term and massive antigenic exposure on the child's organism [9-11].

Our studies suggest that any serious abnormalities in the health status, immune or non-immune pathologies were not revealed in the majority of children with ARRI, which allowed to diagnose the physiological status of their respiratory system. This means that in certain cases these changes can be considered as an adaptive reaction of the child organism's response to the excessive load of infectious antigens, unfavourable environmental factors and socio-environmental challenges. A partial decrease in certain immunity indices, which was revealed in some children with ARRI, was temporary and de facto reflected an increased tendency to response against infectious agent, which can confirm its secondary, post-infectious nature. Nonspecific changes in immunity indices are a consequence of reinvasions of infectious agents, rather than favourable factor that leads to their development. According to our data, 83.3% of children with ARRI had latent and recurrent intracellular infections (herpes virus type 1 and type 2, Epstein-Barr virus, cytomegalovirus, chlamydia and mycoplasma), which significantly burdened the

inflammatory process and reduced its treatment efficacy [6].

An issue of ARRI prevention and treatment during the formation period of organised school groups, which are rightfully classified as high-risk groups, is particularly topical, as far as they are notable for the cross infection due to their active transference and characterised by an increased incidence, which, in turn, may be an indirect proof of immune changes in this adaptation period [7]. Flu vaccination is an efficient preventive measure; however, due to the serologic plasticity of the pathogen, the continuous monitoring and development of new vaccine strains circulating in the human population in each specific epidemic season are required.

In recent years, the properties of Imunoflazid® syrup have investigated in experimental and clinical studies, the active ingredients of which are the bioflavonoids (BF) of such herbs as bushgrass (Calamagrostis epigeios L.) and tufted hair grass (Deschampsia caespitosa L.) – the stable molecular complexes of aglycone flavonoid compounds, consisting of free aglycones (tricine, apigenin, luteolin, quercetin, rhamnazin), O-glycosides and C-glycosides [1, 2]. The major characteristic of these bioflavonoids is the direct antiviral action due to their biological properties; in addition, they function in the human body as antioxidants [1, 2, 8].

Also flavonoids have anti-inflammatory, antibiotic-like potential: acting as antimicrobial agents that inhibit the pathogenic microorganism functioning [12, 15].

It is proved that the antioxidative activity of BF of Imunoflazid® syrup is achieved by inhibition of the activated oxygen metabolites (AOM) generation by cells of the macroorganism. The generation rate of superoxide anion radical after the 2-hour cell incubation with BF decreased by 20-30%, after the 4-hour incubation – by more than 50% reduction observed, and after the 24-hour incubation, the generation of AOM by the macroorganism's cells was almost completely inhibited [8]. Flavonoids affect the antioxidant system: at the stage of free radical and peroxide formation, they act as traps for free radicals, suppress a number of enzymes, provoke an oxidative process and protect antioxidant enzymes from damage [16].

BFs potentiate the apoptosis inducers effect, activate initiation caspase-9 by intracellular mechanisms and contribute to the elimination of affected cells, thereby an apoptosis-modulating effect achieved, which is important for preventing the development of chronic processes associated with the latent viral infection persistence [8].

In recent years, the effect of BF on the cell receptors, including receptors stimulating detoxification processes, has been determined [17]. Such properties are of paramount importance for children with ARRI, since persistent inflammatory processes, accompanied by increased proinflammatory cytokine production and the immune balance violation, lead to the chronic intoxication and a decrease in the adaptive properties of the organism [9].

For preventing polypragmasy in this category of children, efficient use of combined drugs that, except for specific antiviral action, should have a wide range of immunomodulating and general tonic properties, which results in search for new means of prevention and treatment of respiratory infections and determines the relevance of the study.

Objective. To improve treatment and prevention efficacy of acute recurrent respiratory infections by using Imunoflazid ® syrup in the primary school-aged children.

Materials and methods

To perform the assigned tasks 50 children aged 7-10 years with recurrent respiratoty infections were examined, who suffered from ARIs during the academic year more than six times and had 35-40 days of absenteeism on average due to the disease during the academic year. The comprehensive examination and treatment in this group of children was agreed upon with their parents.

The followed-up children had no clinically significant organic respiratory and otolaryngologic diseases, congenital and acquired immune deficiencies. All children were followed-up for 12 months; they were administered prophylactic (anti-relapsing) treatment courses, which included Imunoflazid® syrup at the age-related doses for three weeks twice per year (September–October and February–March). Additionally, in case of clinical symptoms of respiratory infection, this group of children was administered a 5-day treatment course with the indicated above drug (no more than twice per year).

The efficacy of Imunoflazid® syrup was determined by comparing the dynamics of clinical symptoms during the year, the features of saliva micro-crystallization (SMC), the bacterial composition of the pharyngeal exudate at the baseline and a year after the treatment and prevention courses.

The study of the crystallization function of mixed saliva was carried out according to P.A. Leus [4]. Dried saliva drops were examined microscopically. Depending on the arrangement of crystals, three types of saliva microcrystallization were distinguished:

– Type I of SMC is represented by a large centrally located arboraceous or crystal-like pictures ("fern"); the percentage of oral cavity and tonsils diseases in children with this type is no more than 12-15%;

– Type II of SMC – occasional crystal-like conglomerates or needle-like crystals across the visual field of microscope; the percentage of oral cavity and tonsils diseases in children with this

type does not exceed 25-30%;

– Type III of SMC – fragmented occasional fine crystals without a specific orientation (sometimes in the form of drops); this type is accompanied by multiple noncompensated inflammatory processes of the oropharynx.

The results of a comparative evaluation of the microcrystallization indices of three biological fluids (serum, oral fluid, urine) of experimental animals and humans prove that microcrystallization of saliva is a general index of the organism's homeostasis, which can be used to evaluate the efficacy of preventive, curative and rehabilitative measures, and predicting the disease course as well [3]. A number of studies [5, 14] deals with the investigation of the saliva micro-crystallization in various somatic pathologies.

Statistical processing of the study outcomes was carried out using Excel. The main characteristics are given as quantitative observations (*n*), arithmetic mean (*M*), standard error of mean (*m*), absolute and relative values(abs.,%), and statistical significance (*p*). Comparison of statistical characteristics in different groups and during follow-up care was carried out using parametric and non-parametric tests. When determining the statistical significance of differences between independent groups of study by comparison the mean values of the studied indices, unpaired t-test was used.

Results

The study outcomes showed that there was a decrease in signs of prolonged asthenic and intoxication syndromes, manifested in increased physical activity and performance efficiency in 25 (50.0%) children, fatigue reduction in 33 (66.0%) patients, increased sleep duration in 16 (32.0%) cases, improved appetite in 26 (52.0%) children ($p \leq 0.05$) after 12 months of the follow-up care.

Reduced number of ARVI episodes during the follow-up period compared to the previous year was recorded in 16 (32.0%) children, which is illustrative of a high efficiency of the preventive courses (Table 1). One or two episodes of respiratory disease were observed in 6 (12.0%) children. The follow-up care of these children showed that they had uncomplicated course of ARI, reduced duration and severity of the key symptoms that did not require any additional drug administration or inpatient treatment. In 42.0% of children, normalization of haemoglobin and WBC were observed, and the rheological parameters of blood improved.

In 13 (26.0%) children, the clinical efficiency of preventive therapy was found to be low. While analyzing the past medical history of this group of children, there were no specific features of the respiratory infection course concerning its severity, complications or

Table

Table 1. Dynamic pattern of certain clinical and laboratory parameters in children with ARRI (n=50)

Clinical symptom	Abs.	%
Increase physical activity and performance efficiency	25	50.0
Fatigue reduction	33	66.0
Sleep duration increase	16	32.0
Improved appetite	26	52.0
Haemoglobin level normalization	21	42.0
Reduced number of ARVI episodes	16	32.0
Reduced ARI severity and main symptoms duration	22	44.0

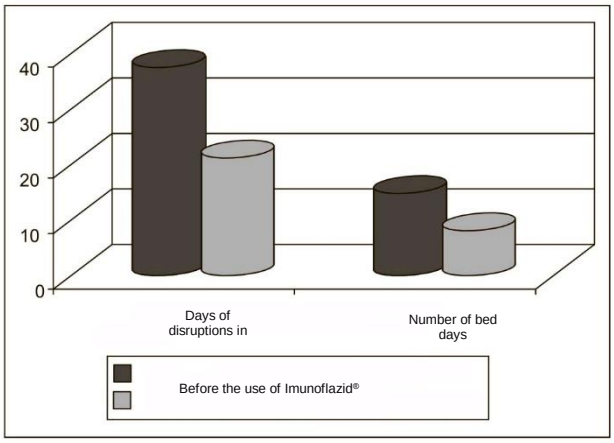

Figure 1. Number of days of the disruptions in school attendance and bed-days during the follow up care.

severe concomitant conditions, but these were children from multi-member families with low socioeconomic status.

Children who were administered Imunoflazid® syrup with prophylactic and treatment purpose throughout the year had a 3-fold decrease in the incidence of the herpes virus infection reactivation, a decrease in the hospitalization rate by 19.0% and its duration – by 27.0%.

When estimating the number of days of the disruptions in school attendance, it was established that in the previous year this number was 37.5±3.7 days per year, and at the end of the follow-up period – 21.2±1.5 days ($p \leq 0.05$). In addition to the above, the number of bed-days in case of hospitalization for the complicated ARI decreased from 14.8±1.3 to 8.1±1.9 days; $p \leq 0.05$ (Fig. 1).

A microbiological study of the oropharynx in 92.0% of children during the period with no clinical

manifistations of the disease, the following pathogenic microorganisms and commensals were cultured: *St. aureus* – 68.0 %, *Str. haemolyticus* – 48.0 %, *Str. pyogenes* – 52.0 %, or associations of coccal flora or their combination with *E. coli* – 18.0 %, *C. albicans* – 34.0 % and *P. aeruginosa* – 10.0 %. The persistence of potentially pathogenic bacteria in the oropharynx should be considered as an evidence of the local defence mechanisms exhaustion, which determines the risk of chronic processes formation associated with increased viral load.

During the follow-up control, the oropharynx microbiocenosis indices showed a great improvement after such preventive and treatment courses: the number of children with bacterial overgrowth and moderate growth of commensals and microbial associations was significantly reduced (the moderate and poor growth prevailed (grade III and II)), which was due to the immunological processes normalization associated with the antigenic viral load reduction; $p<0.01$ (table. 2).

The type definition of the saliva microcrystallization, presented in Table 3, indicates that the type III of SMC was detected in 35.4% of children with ARRI, which indirectly evidenced a decrease in the mineralizing function, saliva pH, violation of its antioxidative properties and determined the decreased protective functions of the respiratory mucous membranes secondary to increased proliferative activity of potentially pathogenic flora. Moreover, the type I of SMC corresponded to the standard data, at the baseline of the observational study was found only in 6.3% of this cohort of children, and at the end of the year – in 39.6% of patients; $p<0.01$.

Table 2. Dynamics of the oropharyngeal microflora growth rate in the examined children (n=50)

Growth rate of microflora	Before the use of Imunoflazid® syrup, n (%)	After usage of Imunoflazid® syrup, n (%)	Parameter
I	-	-	-
II	4 (8.0)	20 (40.0)	+32
III	10 (20.0)	26 (52.0)	+32
IV	36 (72.0)	4 (8.0)	-64
Comparison during the follow-up care	$\chi^2 = 43.378$; $p < 0.01$		

Note: p<0.05 – a statistically significant improvement of the parameter for the 5% level of significance value by the signed-rank test.

Table 3. Types of saliva microcrystallization in children with recurrent respiratory infections (n=48)

Type of saliva microcrystallization	Before the use of Imunoflazid® syrup, n (%)	After usage of Imunoflazid® syrup, n (%)	Dynamics of parameter, abs. (%)
I type	3 (6.3)	19 (39.6)	+16 (33.3)
II type	28 (58.3)	17 (35.4)	-11 (22.9)
III type	17 (35.4)	12 (25.0)	-5 (10.4)
Comparison during the follow-up care	$\chi^2 = 15.9$; $p < 0.01$		

Note: p<0.05 – a statistically significant improvement of the parameter for the 5% level of significance value by the signed-rank test.

Discussion

When analyzing the research data, we can conclude that the proposed method of prevention using Imunoflazid® syrup can be successfully used in children with acute recurrent respiratory infections, accompanied by a violation of antioxidative balance. This management of patients with ARRI allows reducing the disease incidence, its severity and duration, which helps to improve the quality of life of this cohort of patients. The method is easy to use, has no adverse effects, along with a high compliance; it can be used for comprehensive treatment of ARVIs in the acute period to reduce recovery time and in remission to prevent and control over the disease course and improve rehabilitation.

The data obtained make it possible to positively evaluate the organization of preventive courses during the academic period, which will allow conducting efficient prevention and treatment of respiratory diseases, reducing the severity of acute diseases, the incidence of complications and hospitalization, drug administration and preventing the chronic condition formation in this group of children.

Conclusions

1. It has been established that administration of Imunoflazid® syrup in cycles throughout the year with a treatment and prevention purpose results in the children's overall health improvement, a decrease in the intoxicating and asthenic syndromes manifestations, the ARI incidence and severity, the number of days of absenteeism at school, the number of hospitalizations and hospital length of stay ($p< 0.01$).

2. In 35.4% of children with ARRI, the type III of saliva microcrystallization was determined, which is indicative of a decreased mineralizing function, acidity (pH) of saliva, its antioxidative properties violation and determines the decreased protective functions of the mucous membranes of the respiratory tract associated with the increased proliferative activity of the potentially pathogenic flora (St. aureus, St. haemolyticus, St. pyogenes, E. coli, C. albicans, P. aeruginosa). Imunoflazid® syrup in this group of children contributes to an increase in the mineralizing potential of saliva (in 39.6% of children with the type I of SMC) associated with a decrease in the microbial load of the oral cavity ($p<0.01$).

3. The antiviral and antioxidative effects, along with interferonogenic properties, make it feasible of Imunoflazid® syrup administration, both during the treatment of ARRI in children, as well as preventive courses. In prophylactic and therapeutic use, Imunoflazid® is well tolerated by children, does not cause complications, allergic reactions development or exacerbation that determines its high compliance.

Conflict of interest statement

The authors have no conflicting interests to report.

Reference

1. Abaturov AYe, Vysochina IL. Realization of antiviral and antioxidant effects of bioflavonoids in treatment of acute respiratory viral infections. Zdorov'ye Rebenka. 2016;5(73):42-8. (in Russian).

2. Beketova GV, Khaitovich NV, Grinevich AI. Immunoflazidum in pediatrics: system analysis of effectiveness and safety. Pediatrija. Vostochnaja Evropa. 2014;2(07):141-52. (in Russian).

3. Gonchar FL, Pokhodenko-Chudakova IO. Microcrystalliza-tion of oral fluid as a general homeostasis parameter. In: Proceeding of Republic Scientific Research Conference: Innovative approaches to practical decisions of topic problems in maxillofacial surgery and dentistry. Minsk; 2010. p. 70-2. (in Russian).

4. Leus PA. Diagnosticheskoe znachenie gomeostaza sljuny v klinike terapevticheskoj stomatologii: uchebno-metodicheskoe posobie [Diagnostic value of saliva homeostasis in clinical picture of therapeutic dentistry: textbook]. Minsk: Belorussian State Medical University; 2011. 67p. (in Russian).

5. Kobiyasova IV, Savushkina NA. Features of mineralizing saliva function in adolescents of pubertal age and methods for its correction. Sovremennajastomatologija. 2006;2:64-7. (in Russian).

6. Mukvich OM, Kaminska TM. Organization of preventive-rehabilitation measures at school. Sovremennaya pediatriya. 2016;3(75):15-18. doi: 10.15574/SP.2016.75.15. (in Ukrainian).

7. Nedilko VP, Kaminska TM, et al. Ways for improvement of health in schoolchildren. Sovremennaya pediatriya. 2010;3(31):81-4. (in Ukrainian).

8. Rybalko SL. Otchet "Izuchenie mehanizmov dejstvija bio-logicheski aktivnih veshhestv lechebnojsubstancii Proteflazid"[Report "Study of mechanisms of biologically active substances of remedial Proteflazidum]; 2010. 84p. (in Russian).

9. Yulish YeI, Ivanova LA, Chernyshova OE. Immune impairments in sickly children and chronic children. Pediatrija, akusherstvo ta ginekologija. 2003;4:109. (in Russian).

10. Hosakote YM, Liu T, Castro SM, et al. Respiratory syncytial virus induces oxidative stress by modulating antioxidant enzymes. Am J Respir Cell Mol Biol. 2009 Sep;41(3):348-57. doi: 10.1165/ rcmb.2008-0330OC.

11. Hosakote YM, Jantzi PD, Esham DL, et al. Viral-mediated inhibition of antioxidant enzymes contributes to the pathogenesis of severe respiratory syncytial virus bronchiolitis. Am J Respir Crit Care Med. 2011 Jun 1;183(11):1550-60. PMID: 21471094. PMCID: PMC3137144. doi: 10.1164/rccm.201010-1755OC.

12. Lyu SY, Rhim JY, Park WB. Antiherpetic activities of fla-vonoids against herpes simplex virus type 1 (HSV-1) and type 2 (HSV-2) in vitro. Arch. Pharm Res. 2005 Nov;28(11):1293-301. PMID:16350858.

13. de Martino M, Ballotti S. The child with recurrent respiratory infections: normal or not? Pediatr Allergy Immunol. 2007;18 Suppl 18:13-8. doi: 10.1111/j.1399-3038.2007.00625.x. PMID: 17767600.

14. Stensson M, Wendt LK, Koh G, et al. Oral health in pre-school children with asthma — followed from 3 to 6 years. Int J Paediatr Dent. 2010;20(3):165-72. PMID: 20409196. doi: 10.1111/j.1365-263X.2010.01037.x.

15. Ozcelik B, Kartal M, Orhan I. Cytotoxicity, antiviral and antimicrobial activities of alkaloids, flavonoids, andphenolic-acids. Pharm Biol. 2011;49(4):396-402. PMID: 21391841. doi: 10.3109/13880209.2010.519390.

16. Gescher K, Kühn J, Lorentzen E, et al. Proanthocyanidin-enriched extract from Myrothamnus flabellifolia Welw. exerts antiviral activity against herpes simplex virus type 1 by inhibition of viral adsorption and penetration. Journal of Ethnopharmacology. 2011 Mar 24;134(2):468-74. PMID:21211557. doi: 10.1016/j.jep.2010.12.038.

17. Odenthal J, van Heumen BW, Roelofs HM, et al. The influence of curcumin, quercetin, and eicosapentaenoic acid on the expression of phase II detoxification enzymes in the intestinal cell lines HT-29, Caco-2, HuTu 80, andLT97. Nutr. Cancer. 2012;64(6):856-63. PMID: 22830632. doi: 10.1080/01635581.2012.700994.

Received 24.03.2017