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Open, Randomised, Controlled Study to Evaluate a Dietary Supplement with *Pelargonium sidoides* Extract, Honey, Propolis, and Zinc in Children with Acute Tonsillopharyngitis

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Abstract: Background/Objectives: Acute tonsillopharyngitis (ATR), typically caused by viruses, is a common reason for consultation with pediatricians. A dietary supplement with *Pelargonium sidoides* extract, propolis, honey, and zinc has been proposed as an effective adjuvant for the treatment of respiratory tract infections. This study aimed to verify, in a setting of standard clinical practice, the efficacy of this dietary supplement plus standard of care (SoC) versus SoC alone in a pediatric population affected by ATR. **Methods**: This open randomized study (registered on ClinicalTrials.gov: NCT 04899401) involved three Romanian sites specialized in pediatric care. The primary endpoints were changes in Tonsillitis Severity Score and the number of treatment failures (evaluating the use of ibuprofen or high-dose paracetamol as rescue medicine). One hundred and thirty children, divided into two groups, were enrolled and treated for six days. **Results**: The results showed an overall better performance in terms of efficacy of dietary supplement + SoC, compared to SoC alone, with lower total Tonsillitis Severity Score ratings and lower sub-scores (swallowing on day 4 and throat pain and erythema on day 6). No adverse events were reported. Investigators found compliance to be optimal. **Conclusions**: The administration of the dietary supplement + SoC in pediatric patients with ATR was found to be safe and superior to the administration of SoC alone in terms of efficacy. The results confirmed the tested dietary supplement as an optimal effective adjuvant in the treatment of respiratory tract infections and useful in the daily clinical practice of the paediatricians.

Keywords: dietary supplements; tonsillitis; *Pelargonium sidoides*; honey; propolis; zinc

1. Introduction

Acute tonsillo-and rhinopharyngitis (ATR) are acute infections of the pharynx, palatine tonsils, or both. They mainly affect children and adolescents [**1**] and are one of the most common reasons for consulting a family physician. Diagnosis is generally made clinically, with symptoms including sore throat, dysphagia, cervical lymphadenopathy, and fever. In a few cases, the diagnosis should be supplemented by culture or rapid antigen tests. According to common opinion, the vast majority of ATR cases are caused by viral infections, including adenoviruses, Epstein Barr virus, human boca virus, influenza, para-influenza, rhinoviruses, and enteroviruses including Coxsackie viruses [**2**].

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Bacterial etiology, therefore, represents a much smaller percentage, estimated by various authors to account for no more than 30% of all cases of ATR [**2**]. Obviously, only ATR of bacterial origin is an indication for antibiotic therapy. As indicated by the guidelines [2–4], antibiotics should not be used in the pediatric population for apparent viral respiratory illness (sinusitis, pharyngitis, bronchitis). Although the overall antibiotic prescription rates for children have fallen in many countries, this unnecessary administration remains high. The risk of individual antibiotic resistance, potential adverse events (AEs), and an increase in social care costs represent a real and actual danger. In Italy, the number of children treated with antibiotics at home is four times that in the United Kingdom (52% vs. 14%) [**5**]. Common cold, non-specific upper respiratory infection (URI), acute cough, and acute bronchitis are conditions in which antibiotic therapy is not indicated for the predominant viral etiologies [**6**]. Acute bronchitis is diagnosed during more than two million pediatric office visits annually, and doctors prescribe antibiotics for approximately 70% of the cases. As observed [**6**] using the diagnostic clinical criteria for acute otitis media (AOM), sinusitis, and pharyngitis, clinicians should be able to evaluate the etiology correctly and prescribe the appropriate treatment. Only in a few cases is antitussive medication (e.g., dextromethorphan) suggested [**7**], although it must be remembered that it is not recommended in children under 6 years of age. Consequently, research is focused on new products that can be used in daily clinical practice by pediatricians to treat respiratory diseases of viral etiology, avoiding the inappropriate use of antibiotic therapy. The scientific literature currently available shows that the extract of *Pelargonium sidoides* may be effective in the treatment of disorders affecting the respiratory tract [**8,9**]. In particular, its appropriateness and its effectiveness has been demonstrated in the treatment of pediatric populations, both in acute non-streptococcal tonsillopharyngitis compared to placebo [**10**], and in acute bronchitis outside the strict indication for antibiotics [**9**]. Additionally, other substances, including honey, propolis, and zinc, are currently used as dietary supplements (DSs) in clinical practice for pulmonary diseases and are supported by a substantial body of literature [11–13]. In particular, a Cochrane meta-analysis on honey published in 2018 included six clinical trials involving 900 children and demonstrated that honey alleviated cough symptoms compared to no treatment or diphenhydramine but failed to show improvement over dextromethorphan [**11**]. The anti-inflammatory effect of honey has been correlated with the typical reduction in free radicals produced at the site of inflammation. Honey also showed improved immune system and also promoted human peripheral blood B, neutrophils and T lymphocytes [**14,15**]. A systematic review showed that propolis is a safe DS with a positive effect on glutathione peroxidase, glutathione, and total antioxidant capacity levels and can be used as an adjuvant therapy in diseases where oxidative stress is a key factor in the etiology [**16**]. Studies have demonstrated a significant improvement (i.e., reduction in the number of days necessary for remission of symptoms) with the administration of propolis in patients with uncomplicated bacterial and viral upper respiratory tract infections (URTI). [**17**]. Recently, a combination of propolis and N-acetylcysteine was used in randomized double-blind trials in patients with chronic obstructive pulmonary disease (COPD) [**18**]. These studies demonstrated an improvement in symptoms and quality of life and a reduction in the incidence of COPD exacerbations in treated patients. During the recent pandemic, propolis has also been tested in randomized placebo/standard-care controlled trials [**19**]. This dietary supplementation showed limited, but statistically significant, beneficial effects in improving the signs and symptoms of COVID-19 (dry cough, shortness of breath, sore throat, and chest pain). A comprehensive systematic review [**20**] evaluated data from 8.526 treated patients and demonstrated the beneficial effects of zinc supplementation in reducing the duration of the common cold. In the same review, the majority of the 15 studies (out of 34) focused on prevention failed to demonstrate a positive effect of zinc on the prevention of colds, and an increase in non-serious adverse events (SAE) was noted in the patients who received the supplement. The positive results in this important review of zinc supplementation and the fact that it was often used in conjunction with other treatments underscore that zinc can be considered a useful therapeutic tool in clinical practice and that physicians can safely administer it in conjunction with various therapeutic products.

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The above-mentioned considerations have prompted Pediatrica Srl (Livorno, Italy) to develop the dietary supplement PediaFlù® (DSPP), an oral solution of *Pelargonium sidoides* extract (Pelagon P 70™) specifically for the pediatric age, as an adjuvant for the well-being of the respiratory tract, honey, a formulation of propolis (PropolNext® Plus), and zinc. The DSPP product is currently available in Italy and other European countries as an adjuvant for seasonal diseases. In the present clinical trial, we tested the product DSPP to confirm its effectiveness, which has been evidenced in the daily clinical practice of thousands of pediatricians in this country.

Therefore, the aim of this study was to explore whether in pediatric patients affected by ATR, a 6-day administration of the tested DSPP along with standard of care (SoC) could improve ATR symptomatology in comparison with patients receiving SoC alone. We have recently performed a preliminary and partial evaluation of the study results focused on the honey component by analyzing a limited subset of the parameters monitored during the study [**21**]. In contrast, the present article offers a comprehensive overview of the study outcomes and results with the aim of demonstrating the potential contribution of this trial to the enhancement of DSPP safety and performance. In addition, it provides pediatricians with the opportunity to autonomously assess whether the administration of DSPP in conjunction with SoC yields clinical benefits in children with ATR.

2. Materials and Methods

2.1. Study Design

The objective of this multicenter trial was to evaluate the efficacy as curative treatment and safety of a commercially available DS containing *Pelargonium sidoides* extract, honey, propolis and zinc administered with SoC versus SoC alone to improve the symptom severity of ATR in a population of pediatric patients. This study used a randomized, controlled, parallel-group superiority design. We followed the methods described by Fabio Cardinale et al., [**22**] available at https://www.protocols.io/view/a-randomized-open-controlled-study-to-evaluate-the-cmtqu6mw.html. The primary objectives of the study were to evaluate the efficacy through changes in the Tonsillitis Severity Score (TSS), the assessment of rescue medicine administration considered a treatment failure, and safety through the evaluation of AE and SAE incidence during the study. The secondary objectives included overall symptom improvement, overall safety during the study, and compliance.

The three participating sites were Romanian private clinics specializing in pediatric care located in Timisoara region (#01 Cabinet Medical Medicina de Familie Dr Morariu Bordea, #02 Cabinet Medical Dr Herteg Dorina, and #03 Cabinet Medical Dr Cristian-Radu Matei). The trial was approved by the local Independent Ethics Committees on April 27, 2021 (site #01 and #02) and April 23, 2021 (site #03). The three Romanian sites were coordinated by the Department of Clinical Trials of the University of Medicine and Pharmacy Victor Babes in Timisoara, Romania, which is an academic research organization. The Sponsor charged an independent Contract Research Organization (Opera CRO, Timisoara, Romania) to perform the sites monitoring, data management, and statistical analysis. On the other hand, Sponsor designated Prof. Fabio Cardinale (University of Bari, Italy) as the scientific coordinator of the study. In this role, he assured the application of ethics requirements and the finalization of the protocol and shared the scientific information of the tested product to the Investigators. The Sponsor limited its activity to send to the sites the quantity of DSPP to perform the trial and to offer a partial grant.

The trial was conducted in accordance with the Declaration of Helsinki and complied with the International Conference on Harmonization, Good Clinical Practice, and Italian and Romanian regulatory requirements. Written informed consent was obtained from all participants before inclusion in the study, and no financial incentives were offered to any of them. This trial was registered in the clinicaltrials.gov database (NCT04899401) and the protocol was deposited in protocols.io [**22**]. A detailed description of the study protocol has recently been published [**23**] and its graphical abstract is available here as Supplementary Material.

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2.2. Participants

The patients were male and female children aged 3-10 years old, suffering from ATR with a TSS score ≥ 8 for no more than 48 hours, showing negative results on rapid tests for Group A beta-hemolytic *Streptococcus* (GABHS) and Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) infection, and negative nasal and/or pharyngeal exudate culture. Exclusion criteria included evidence of lacunar or follicular angina, increased hemorrhagic diathesis or chronic diseases, more than two episodes of tonsillitis within the previous 12 months, or close contact with SARS-CoV-2 infected individuals within ten days from symptoms onset. Patients with known or suspected hypersensitivity to trial products, those treated with antibiotics within four months prior to trial inclusion, presenting a mandatory indication for therapy with antibiotics, or currently in treatment with agents potentially influencing the trial outcomes or having interactions with the trial products were also excluded. Participation in another clinical trial within the previous three months was also an exclusion criterion.

Prior to the start of the study and in the presence of parents, the Investigators trained the children to respond correctly to the tests administered, particularly the Patient Global Assessment of Efficacy (PGAE). In addition, parents were asked to complete a temperature log and report in the patient diary the amount of rescue medication used during the study, were instructed to return the unused medication at the final visit, and to call the investigator in case of AE. The enrollment period started on June 3, 2021, and ended on August 6, 2021. The study was concluded on August 12th 2021 (last patient last visit).

2.3. Treatment and Allocation

Patients who met the eligibility criteria were enrolled and randomized into one of two groups in a 1:1 ratio: one group received DSPP in addition to SoC, whereas the other group received SoC alone. Randomization was conducted centrally by the CRO managing the study using a permuted block of random sizes. The package was GNU Library General Public License, v 2.1 from R statistical software v 3.5. Patients were allocated a unique 5-digit patient randomization number using an online web service (https://edc.operacro.com:4xx), which was available 24 h a day in a 1:1 group assignment ratio. Personnel involved in the allocation process were not involved in the evaluation of subjects in the study. It should be noted that no one (patient or parent) was blinded to treatment allocation. The tested DSPP was orally administered 5 ml × 3 times per day for six days for children under six years, and 10 ml × 3 times per day for six days for children over six years old. The DSPP contains Pelagon P-70™ (equal to 133.3 mg of *Pelargonium sidoides* extract per 100 ml), PropolNext® Plus (equal to 7.7 mg of propolis extract per 100 ml), zinc (13.3 mg per 100 ml), honey (5.5 g per 100 ml).

SoC treatment included the following products:

- Benzydamine hydrochloride (throat spray 0.15%): children over six and below twelve: 4 sprays 2-6 times a day; children (under six years): 1 spray per 4kg of body weight, a maximum of 4 sprays at once, 2-6 times a day according to the leaflet. Each spray was equals 0.17 ml of solution.
- Paracetamol (120 mg/5 ml) per os, in case of >38,5°C, 10 mg/kg/dose, per need every 6-8 hours. Maximum dosage under a 30 mg/kg dose
- Nasal decongestion through hydration with fluids, aspiration of secretions, use of saline solution for nasal irrigation, nasal sprays with seawater, and nasal spray with an active compound (this last one was only a special indication of the medical doctor).

Coumarin-based products and antibiotics were not permitted during the trial. The administration of paracetamol (>30 mg/kg) or ibuprofen (100 mg/5 ml) during the study period was considered rescue medicine. There were no restrictions on treatments previously taken by the subjects for medical conditions unrelated to this trial protocol.

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Four visits were scheduled for the study. At each visit, eligibility and physical examinations were carried out, including disease assessment and TSS score evaluation, as well as concomitant medications and AEs and SAEs assessment and recording. At the screening visit (visit 1; day -2 to day -1), GABHS and SAR-COV-2 were detected, medical history and demographics were recorded, and eligible patients (parents or legal guardians) were asked to sign informed consent. At the baseline visit (visit 2, day 0), patient characteristics were recorded, and the study products and patient diaries were supplied. At the intermediate visit (visit 3, day 4 from recruitment), diary verification was performed. Final evaluations, symptoms, and compliance evaluations were performed at the end of the study visit (visit 4; day 6 from recruitment). The study products and their diaries were also collected.

2.4. Primary and Secondary Outcomes

The primary efficacy outcomes were the change in TSS [**10**] and the number of treatment failures, comparing the use of rescue medicines.

TSS is composed of five sub-scores reflecting the nature of acute non-GABHS tonsillopharyngitis as a virus-induced inflammatory infection of the upper airways, namely difficulty in swallowing, sore throat, increased salivation, pharyngeal erythema, and fever. At each visit, the Investigator scored each symptom (difficulty in swallowing, sore throat, increased salivation, pharyngeal erythema) using a 4-point scale (3= severe, 2= moderate, 1= mild, 0= not present). Finally, a fever score was added, comprising the categories < 37.5°C = 0; 37.5°C to < 38.5°C = 1; 38.5°C to < 39.5°C = 2; and ≥ 39.5°C = 3, thus creating a possible TSS range between 0 to 15. The results were compared in terms of absolute change in score from baseline to final visit, between groups, and intragroup. TSS was assessed at each visit, and the total and sub-scores were evaluated.

The use of rescue medicines (paracetamol >30 mg/kg or ibuprofen 100 mg / 5 ml) by a patient was considered treatment failure.

The primary safety outcome was the incidence of AE/SAE during the trial.

The following secondary outcomes were recorded:

Investigator Global Assessment of Efficacy (IGAE): Investigators performed a global assessment of treatment efficacy using a 4-point scale (1 = excellent, 2 = good, 3 = fair, 4 = poor), with a comparison between groups at the end of the trial.

PGAE: Patients performed a global assessment of treatment efficacy using a 5-point scale (1 = very satisfied, 2 = satisfied, 3 = adequate, 4 = unsatisfied, 5 = very unsatisfied), with a comparison between groups at the end of the trial.

Investigator Global Assessment of Safety (IGAS): Investigators performed a global assessment of the safety of treatments using a 4-point scale (1 = very good safety, 2 = good safety, 3 = moderate safety, and 4 = poor safety), with a comparison between groups at the end of the trial.

Compliance with the administered treatments: comparison between groups at the end of the trial.

2.5. Sample Size Determination and Statistical Methods

The sample size was calculated based on the primary outcome TSS and based on the results of a similar investigation [**10**]. The minimal clinical difference between the tested group DSPP in addition to SoC and the control group SoC alone, after six days of treatment was considered to be 2 points decrease in mean TSS. Therefore, based on the sample size formula for the comparison of two means (2-sample) at a significance level of 5%, a power of 80%, and a minimally clinically important difference of 2 ±3.85 points, 120 patients were required to be enrolled in this study. To obtain 120 evaluable patients, approximately 150 patients should be screened (including potential screening failure and estimated dropout patients).

All statistical analyses were performed using a specific statistical software (version 4.1.0; R Foundation for Statistical Computing). The final analysis was completed after all patients had

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concluded the trial, all queries were resolved, and the database was locked. The overall Type I error rate was maintained at 5%. All tests were two sided. Data from unscheduled visits were excluded from analysis. Statistical analyses were conducted on all patients who had successfully completed the trial without a protocol deviation, which is regarded as impacting the assessment of the key variables (as per the trial protocol). The quality and completeness of the collected data were preliminarily evaluated and compared using data analysis. If a patient had missing information for one or more variables, the missing data were not replaced even after the resolution of the queries. If a patient violated the inclusion/exclusion criteria, their data were excluded from analysis. Quantitative variables (i.e., demographic information) were described using mean and standard deviation (SD) if they were normally distributed. Non-normally distributed variables were described using median and interquartile ranges. The Student's t-test and Mann-Whitney U test were used to perform a comparative analysis, depending on the distribution of these variables. Factorial variance analysis was also employed to evaluate any interactions between quantitative variables and linear progression models to relate possible confounding bias to independent variables. Categorical variables were described using frequencies and percentages, and a comparative analysis was performed using the chi-square test.

3. Results

3.1. Patient Disposition and Characteristics

Of the 135 patients screened, 130 were considered eligible for the trial completion. Patients were randomly assigned to either the DSPP + SoC group (n = 66) or SoC alone group (n = 64). All 130 enrolled subjects completed the trial and were included in the Intention-To-Treat (ITT) and safety populations. One-hundred and twenty-nine patients were included in the Per Protocol (PP) population: one patient belonging to the DSPP + SoC group dropped out.

The reporting of this study conforms to the CONSORT Trials checklist: https://www.equator-network.org/reporting-guidelines/consort/. Patient disposition is graphically displayed in Figure 1.

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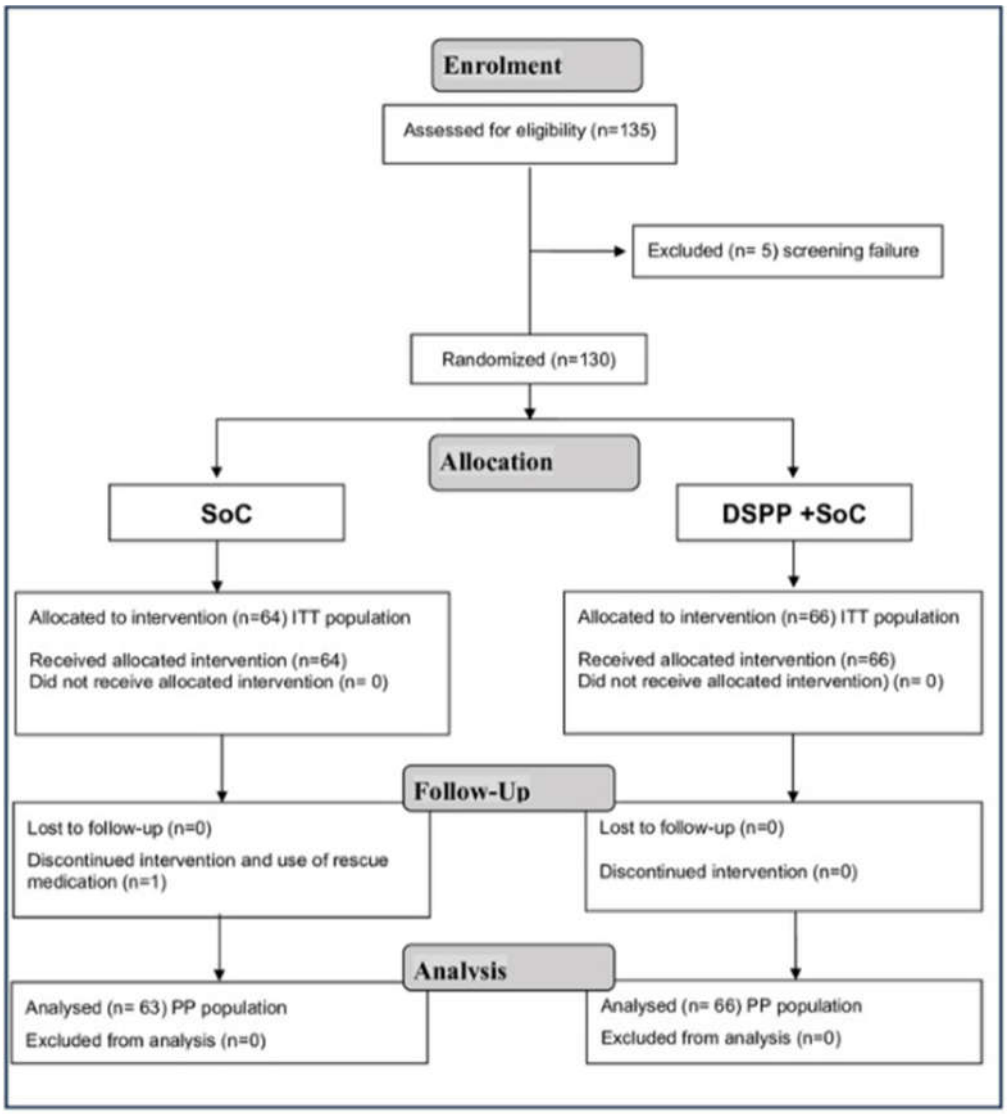

Figure 1. Consort 2010 Flow Diagram. Standard of Care (SOC); Dietary Supplement Pediaflù® (DSPP).

The patient demographics presented in Table 1 revealed no significant differences ($p > 0.05$) in baseline demographic and anthropometric measures.

Table 1. Demographics characteristics of the Per Protocol (PP) population by assignment group at the baseline visit.

		All	DSPP + SoC	SoC	p
Age (years)	N	129	66	63	ns
	Mean (SD)	5.52 (2.07)	5.8 (2.01)	5.22 (2.11)	
	Median	5	5.5	5	
	Range	3 - 10	3 - 10	3 - 10	
BMI (kg/m^2)	N	129	66	63	ns
	Mean (SD)	16.41 (3.06)	16.71 (3.0)	16.1 (3.11)	
	Median	15.54	15.97	15.42	
	Range	10.42 - 30.26	10.42 - 25.69	11.76 - 30.26	
Height (cm)	N	129	66	63	ns
	Mean (SD)	114.51	116.56 (15.36)	112.37 (14.31)	

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		(14.95)			
	Median	114	116	108	
	Range	89 - 160	89 - 160	90 - 148	
	N	129	66	63	
Weight	Mean (SD)	22.1 (8.29)	23.32 (8.74)	20.83 (7.66)	ns
(kg)	Median	20	22	19	
	Range	11 - 60	12 - 60	11 - 54	

ns: non-significant ($p > 0.05$) Mann-Whitney U tests.

3.2. *Administered Treatments*

All patients were deemed to comply with the prescribed regimens for DSPP, low-dose paracetamol, and benzydamine hydrochloride. No significant intergroup differences were observed with regard to the administration of benzydamine hydrochloride, as evidenced by the comparable numbers of doses and quantities. In Table 2, a summary of low-dose paracetamol use as part of SoC during the 6-day study period is shown. A statistically significant difference in the mean number of doses administered was observed between the two groups ($p < 0.01$).

Table 2. Number of doses of paracetamol used during the study.

	DSPP + SoC	**SoC**	**p**
N	66	63	
Mean (SD)	13.67 (4.34)	15.92 (3.02)	< 0.01
Median	14	18	
Range	5 - 18	6 - 18	

3.3. *Efficacy Analysis*

3.3.1. TSS

The absolute change in the TSS score from baseline to the final visit indicated that patients treated with DSPP + SoC showed lower TSS ratings than patients treated with SoC alone (Figure 2). The difference between groups was statistically significant at days 4 ($p = 0.034$) and 6 ($p = 0.002$), as determined by the Mann-Whitney U test for independent samples. The evaluation of TSS subscores revealed a statistically significant difference between the DSPP + SoC and SoC alone groups for sore throat ($p < 0.001$) and pharyngeal erythema ($p < 0.05$).

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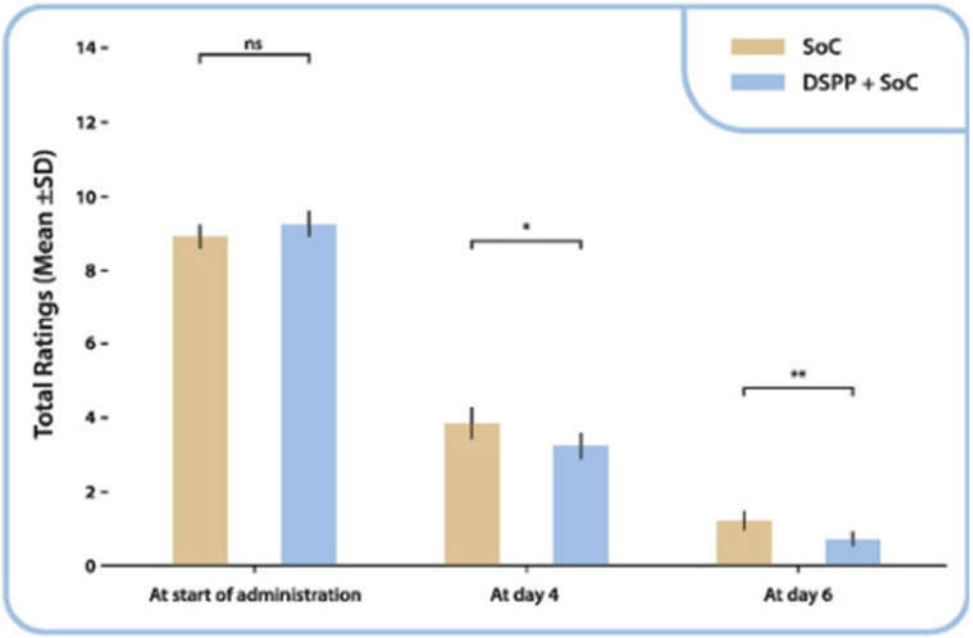

Figure 2. Tonsillitis Severity Score (TSS) total mean±SD ratings between administration groups, at day 0, day 4 and day 6. *** $p < 0.001$; ** $p < 0.01$; * $p < 0.05$; ns non-significant ($p > 0.05$).

3.3.2. Use of Rescue Medicine

Ibuprofen was administered as rescue medicine to only one patient in the SoC alone group (1.6% of patients in the SoC alone group and 0.8% of total patients), who withdrew prematurely due to persistent symptoms and increased fever. No patients were administered a dose of paracetamol above 30 mg/kg/dose, which was considered in the protocol as the threshold for rescue medicine.

3.3.3. PGAE

The evaluation carried out using the PGAE on day 6 showed that the efficacy of the treatment was evaluated as very good by 80.3% (n = 53) of the patients in the DSPP + SoC group and 55.6% (n = 35) of the patients in the SoC alone group (Figure 3).

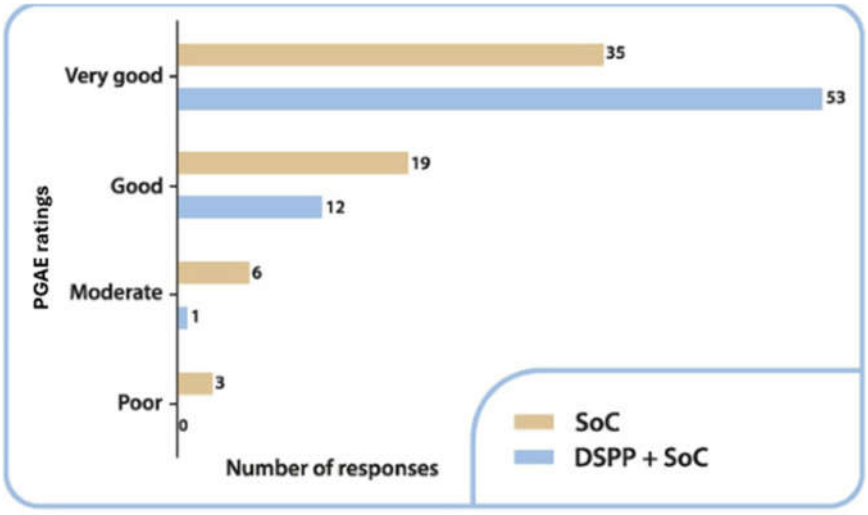

Figure 3. PGAE ratings in the two groups at final visit.

The difference was considered statistically significant ($p = 0.013$, as determined using the chi^2 test of independence). On day 6, none of the patients in the DSPP + SoC group rated the treatment as poor, whereas 4.8% of the patients (n = 3) in the SoC alone group rated the treatment as poor.

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3.3.4. IGAE

The IGAE among the groups are summarized in Figure 4. On Day 6, treatment was evaluated as very good in 83.3% of patients in the DSPP + SoC group and 58.7% of patients in the SoC alone group. The difference between the groups was statistically significant (p = 0.04).

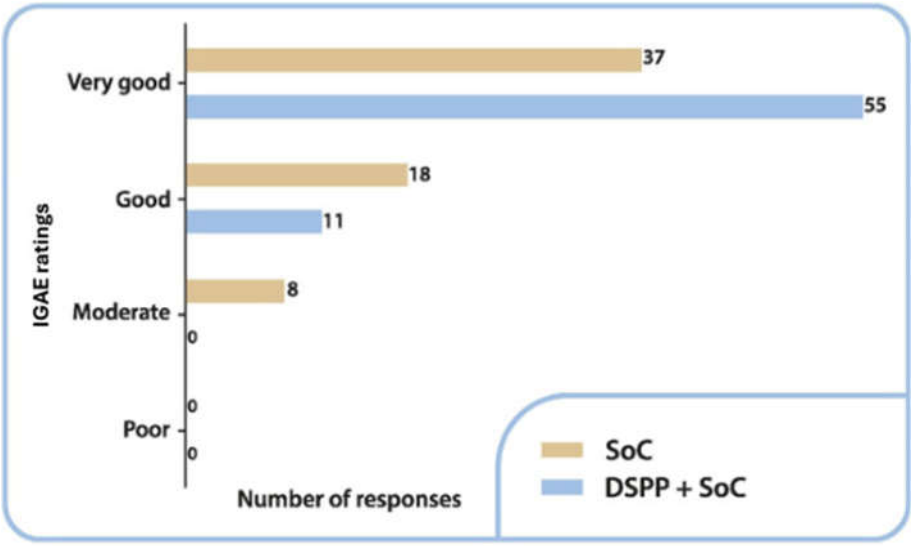

Figure 4. IGAE ratings in the two groups at final visit.

3.4. Safety Analysis

No AE or SAE were reported during the clinical trial. The investigators did not record any concomitant medications. Based on results from IGAS performed at day 6, the safety of treatment was evaluated as "very good" or "good" by 93.9% (n= 62) and 6.1% (n = 4) of investigators in the DSPP + SoC group, and 77.8% (n= 49) and 22.2% (n= 14), of investigators in SoC alone group (Figure 5). The difference between the DSPP + SoC and SoC alone groups was statistically significant (p = 0.008).

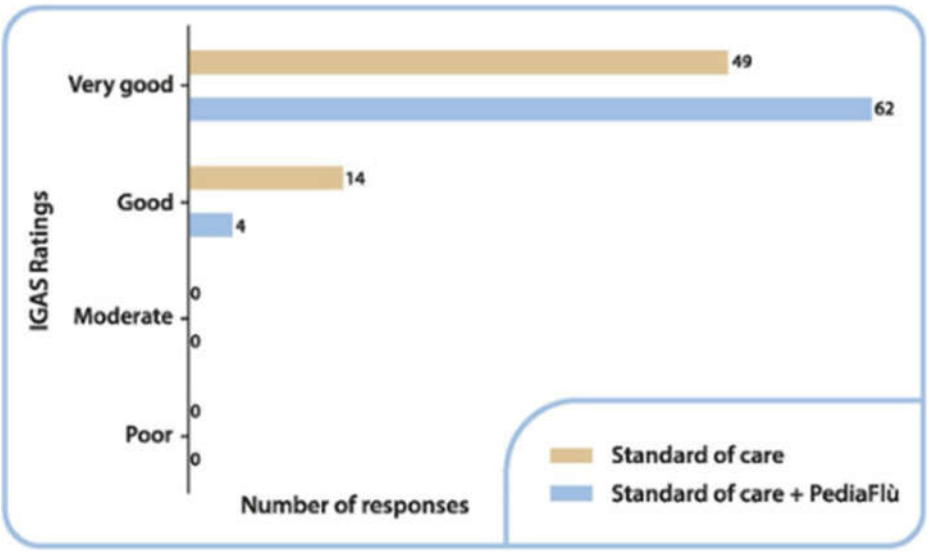

Figure 5. IGAS ratings by number of responses at final visit.

4. Discussion

A key question in children affected by tonsillitis-pharyngitis is differentiating between GABHS infections such as *Streptococcus pyogenes* and viral tonsillitis [**6**]. In fact, clinical symptom scoring

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systems (Modified Centor or McIsaac Scores) can help in identifying the disease, but none of the available scoring systems are sufficiently accurate to identify GABHS pharyngitis with reasonable certainty. Therefore, the diagnosis of GABHS infection should be confirmed by laboratory testing (either a rapid antigen detection test or culture). It is therefore extremely important that pediatricians can easily and quickly identify the infant population in which it is appropriate to perform the rapid test. The presence of more than two criteria in the Centor score (absence of cough, fever above 38°, enlarged lymph nodes, presence of tonsillar exudate, and age under 15 years) suggest the performance of the rapid test. In contrast, if the McIsaac score is greater than two (presence of rhinitis, cough, rhinorrhea, diarrhea, stomatitis, oral ulcers), it is more likely to be a viral infection [**24**]. In addition, the rapid test is not recommended in children less than three years old, because the risk of acute articular rheumatism should be very low at this age. In our trial, only children suffering from ATR for no more than 48 h and with a negative rapid test for GABHS and SARS-CoV-2 infection were included. In addition, to avoid the inclusion of any subjects affected by bacterial diseases, a negative culture of the nasal/pharyngeal exudate was required.

The main limitation of the study (due to economic issues) is the open-controlled design instead of a double-blind design.

In our opinion, the positive results in terms of reduction of TSS total rating, significant improvement of symptoms (sore throat and pharyngeal erythema), and reduction of used doses of paracetamol clearly demonstrate that the administration of DSPP along with SoC produces a clinical benefit in children affected by viral acute tonsillopharyngitis. In addition, the absolute safety of DSPP (no AEs during the entire duration of the trial) and its optimal compliance represent the fundamental requirements for a product to be administered in such young children.

Supplementary Materials: The following supporting information can be downloaded at the website of this paper posted on Preprints.org., Figure S1: Graphical Abstract. Figure S2: Synopsis Pitch Deck.

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Institutional Review Board Statement: The study was conducted in accordance with the Declaration of Helsinki and approved by the Ethics Committees of Cabinet Medical Medicina de Familie Dr Morariu Bordea (code #043 on April 27, 2021), by the Ethics Committees of Cabinet Medical Dr Herteg Dorina (code #044 on April 27, 2021), and by the Ethics Committee of Cabinet Medical Dr Cristian Radu Matei (code #046 on April 23, 2021).

Informed Consent Statement: Informed consent was obtained from all subjects involved in the study.

Data Availability Statement:. The study methods [22] (p. 4), has been deposited at protocols.io (source at: https://www.protocols.io/view/a-randomized-open-controlled-study-to-evaluate-the-cmtqu6mw.html), a platform for developing and sharing reproducible protocols. The description of the study protocol was recently published [23]. In addition, the full study protocol (final version 2.0, dated March 3, 2021) and the data sets analysed in the study are available from the corresponding author on reasonable request.

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Conflicts of Interest: F.C., M.M.B., D.H., and C.R.M. certify that there is no conflict of interest with any financial organization regarding the material discussed in the manuscript. D.F.B. is employed at Opera CRO, the Contract Research Organization that performed monitoring of the study. L.B. and M.M. are former employed at TIGERMED Italy, the company that performed the data management of the study. A.C. and G.G. are employed at Pediatrica Srl. The funder Pediatrica Srl had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript; or in the decision to publish the results.

Abbreviations

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ATR	Acute Tonsillopharyngitis/Rhinopharyngitis
SoC	Standard of Care
AOM	Acute Otitis Media
DS	Dietary Supplement
URTI	Upper Respiratory Tract Infection
AE/SAE	Adverse Event/Serious Adverse Event
DSPP	Dietary Supplement Pediaflù®
TSS	Tonsillitis Severity Score
CRO	Contract Research Organization
GABHS	Group A beta-hemolytic Streptococcus
SARS-CoV-2	Severe Acute Respiratory Syndrome Coronavirus 2
PGAE	Patient Global Assessment of Efficacy
IGAE	Investigator Global Assessment of Efficacy
IGAS	Investigator Global Assessment of Safety
SD	Standard Deviation
ITT	Intention-To-Treat
PP	Per Protocol

References

1. Shaikh, N.; Leonard, E.; Martin, J.M. Prevalence of streptococcal pharyngitis and streptococcal carriage in children: a meta-analysis. *Pediatrics* **2010**, 126, e557-564. doi: 10.1542/peds.2009-2648. Epub 2010 Aug 9. PMID: 20696723.
2. Windfuhr, J.P.; Toepfner, N.; Steffen, G.; Waldfahrer, F.; Berner, R. Clinical practice guideline: tonsillitis I. Diagnostics and nonsurgical management. *Eur Arch Otorhinolaryngol* **2016**, 273, 973–987. doi: 10.1007/s00405-015-3872-6. Epub 2016 Jan 11. PMID: 26755048; PMCID: PMC7087627
3. ESCMID Sore Throat Guideline Group; Pelucchi, C.; Grigoryan, L.; Galeone, C.; Esposito, S.; Huovinen, P.; Little, P.; Verheij, T. Guideline for the management of acute sore throat. *Clin Microbiol Infect* **2012**, 18, Suppl 1:1–28. doi: 10.1111/j.1469-0691.2012.03766.x. PMID: 22432746.
4. Vicentini, C.; Vola, L.; Previti, C.; Brescia, V.; Dal Mas, F.; Zotti, C.M.; Bert, F. Antimicrobial Stewardship Strategies Including Point-of-Care Testing (POCT) for Pediatric Patients with Upper-Respiratory-Tract Infections in Primary Care: A Systematic Review of Economic Evaluations. *Antibiotics (Basel)* **2022**, 22, 1139. doi: 10.3390/antibiotics11081139. PMID: 36010008; PMCID: PMC9404955.
5. Clavenna, A.; Bonati, M. Differences in antibiotic prescribing in paediatric outpatients. *Arch Dis Child* **2011**, 96, 590-5. doi: 10.1136/adc.2010.183541. Epub 2011 Jan 12. PMID: 21233078.
6. Hersh, A.L.; Jackson, M.A.; Hicks, L.A. American Academy of Pediatrics Committee on Infectious Diseases. Principles of judicious antibiotic prescribing for upper respiratory tract infections in pediatrics. *Pediatrics* **2013**, 132, 1146-54. doi: 10.1542/peds.2013-3260. Epub 2013 Nov 18. PMID: 24249823.
7. American Academy of Pediatrics. Committee on Drugs. Use of codeine- and dextromethorphan-containing cough remedies in children. *Pediatrics* **1997**, 99, 918–920. doi: 10.1542/peds.99.6.918. PMID: 9190557.
8. Careddu, D.; Pettenazzo, A. *Pelargonium sidoides* extract EPs 7630: a review of its clinical efficacy and safety for treating acute respiratory tract infections in children. *Int J Gen Med* **2018**, 11, 91-98. doi: 10.2147/IJGM.S154198. PMID: 29563828; PMCID: PMC5849386.
9. EMA/HMPC/765656/2022 Committee on Herbal Medicinal Products (HMPC) Assessment report on *Pelargonium sidoides DC. Pelargonium reniforme Curt., radix.* Final - Revision 2 29 - May 2024 Available online: https://www.ema.europa.eu/en/documents/herbal-report/final-assessment-report-pelargonium-sidoides-dc-pelargonium-reniforme-curt-radix-revision-2_en.pdf (accessed on 24 December 2024).
10. Bereznoy, V.V.; Riley, D.S.; Wassmer, G.; Heger, M. Efficacy of extract of *Pelargonium sidoides* in children with acute non-group A beta-hemolytic streptococcus tonsillopharyngitis: a randomized, double-blind, placebo-controlled trial. *Altern Ther Health Med* **2003**, 9, 68–79. PMID:14526713.
11. Oduwole, O.; Udoh, E.E.; Oyo-Ita, A.; Meremikwu, M.M. Honey for acute cough in children. *Cochrane Database Syst Rev* **2018**, 4, CD007094. PMID: 29633783. PMCID: PMC6513626. doi:10.1002/14651858.CD007094.pub5.

12. Braakhuis, A. Evidence on the Health Benefits of Supplemental Propolis. *Nutrients* **2019**, 8, 2705. doi: 10.3390/nu11112705. PMID: 31717277; PMCID: PMC6893770.
13. Singh, M.; Das, R.R. Zinc for the common cold. *Cochrane Database Syst Rev* **2013**, 18, CD001364. doi: 10.1002/14651858.CD001364.pub4. Update in: Cochrane Database Syst Rev. 2015;(4):CD001364. PMID: 23775705.
14. Abuharfeil, N.; Al-Oran, R.; Abo-Shehada, M. The effect of bee honey on the proliferative activity of human B- and T lymphocytes and the activity of phagocytes. *Food Agri Immunol* **1999**, 11, 169–177. doi.org/10.1080/09540109999843.
15. Khan, R.U.; Naz, S.; Abudabos, A.M. Towards a better understanding of the therapeutic applications and corresponding mechanisms of action of honey. *Environ Sci Pollut Res Int* **2017**, 24, 27755-27766. doi:10.1007/s11356-017-0567-0.
16. Nazari-Bonab, H.; Jamilian, P.; Radkhah, N.; Zarezadeh, M.; Ebrahimi-Mameghani, M. The effect of propolis supplementation in improving antioxidant status: A systematic review and meta-analysis of controlled clinical trials. *Phytother Res* **2023**, 37, 3712-3723. doi:10.1002/ptr.7899.
17. Esposito, C.; Garzarella, E-U.; Bocchino, B.; D'Avino, M.; Caruso, G.; Buonomo, A.R.; Sacchi, R.; Galeotti, F.; Tenore, G.C.; Zaccaria, V.; et al. A standardized polyphenol mixture extracted from poplar-type propolis for remission of symptoms of uncomplicated upper respiratory tract infection (URTI): A monocentric, randomized, double-blind, placebo-controlled clinical trial. *Phytomedicine* **2021**, 80, 153368. doi:10.1016/j.phymed.2020.153368.
18. Buha, I.; Mirić, M.; Agić, A.; Simić, M.; Stjepanović, M.; Milenković, B.; Nagorni-Obradović, L.; Škodrić-Trifunović, V.; Ilić, B.; Popević, S.; et al. A. A randomized, double-blind, placebo-controlled study evaluating the efficacy of propolis and N-acetylcysteine in exacerbations of chronic obstructive pulmonary disease. *Eur Rev Med Pharmacol Sci* **2022**, 26, 4809-4815. doi:10.26355/eurrev_202207_29206.
19. Silveira, M.A.D.; De Jong, D.; Berretta, A.A.; Galvão, E.B.D.S.; Ribeiro, J.C.; Cerqueira-Silva T, Amorim TC, Conceição LFMRD, Gomes MMD, Teixeira MB; et al. BeeCovid Team. Efficacy of Brazilian green propolis (EPP-AF®) as an adjunct treatment for hospitalized COVID-19 patients: A randomized, controlled clinical trial. *Biomed Pharmacother* **2021**, 138, 111526. doi:10.1016/j.biopha.2021.111526.
20. Nault, D.; Machingo, T.A.; Shipper, A.G.; Antiporta, D.A.; Hamel, C.; Nourouzpour, S.; Konstantinidis, M.; Phillips, E.; Lipski, E.A.; Wieland, L.S. Zinc for prevention and treatment of the common cold. *Cochrane Database Syst Rev* **2024**, 5, CD014914. doi:10.1002/14651858.CD014914.pub2.
21. Cardinale, F.; Barattini, D.F.; Martinucci, V.; Bordea, M.M.; Barattini, L.; Rosu, S. The Effectiveness of a Dietary Supplement with Honey, Propolis, *Pelargonium sidoides* Extract, and Zinc in Children Affected by Acute Tonsillopharyngitis: An Open, Randomized, and Controlled Trial. *Pharmaceuticals (Basel)* **2024**, 17, 804. doi:10.3390/ph17060804.
22. Cardinale, F.; Barattini, D.F.; Bordea, M.M.; Herteg, D.; Matei, C.R. A randomized, open, controlled study to evaluate the efficacy and safety of Pediaflù® (dietary supplement) along with standard of care in children with acute tonsillopharyngitis / rhinopharyngitis versus standard of care only. Protocols.io. Available online: https://dx.doi.org/10.17504/protocols.io.kqdg3988eg25/v2. (accessed on 24 December 2024)
23. Cardinale, F.; Barattini, D.F.; Sbrocca, F.; Centi, A.; Giuntini, G.; Morariu Bordea, M.; Herteg, D.; Rosu, S.; Matei, C.R. The Effects of a Dietary Supplement (PediaFlù®) Plus Standard of Care in Children With Acute Tonsillopharyngitis/Rhinopharyngitis: Protocol for a Randomized Controlled Trial. *JMIR Res Protoc* **2024**, 13:e53703. doi: 10.2196/53703.
24. Chiappini, E.; Principi, N.; Mansi, N.; Serra, A.; De Masi, S.; Camaioni, A.; Esposito, S.; Felisati, G.; Galli, L.; Landi, M. et al. Italian Panel on the Management of Pharyngitis in Children. Management of acute pharyngitis in children: summary of the Italian National Institute of Health guidelines. *Clin Ther* **2012**, 34, 1442-1458.e2. doi: 10.1016/j.clinthera.2012.04.028. PMID: 22691611.