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ORIGINAL RESEARCH

In vitro antimicrobial activity of an oral spray combining 0.15% benzydamine hydrochloride and 0.5% cetylpyridinium chloride

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Abstract

Background: Reducing the microbial load in the upper respiratory tract can reduce the risk of transmission and spread of respiratory tract infections.

Methods: The in vitro antimicrobial activity of a new oral spray combining 0.15% benzydamine hydrochloride and 0.5% cetylpyridinium chloride (Tantum Verde DUO®, Angelini Pharma S.p.A., spray duo) was investigated.

Results: Spray duo showed bacterial, yeasticidal and virucidal activity against *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Escherichia coli*, *Enterococcus hirae* (1 minute contact), *Haemophilus influenzae*, *Moraxella catarrhalis*, *Streptococcus pyogenes* (30 second contact), *Candida albicans* (5 minute contact), modified vaccinia Ankara (3 minute contact), influenza A virus subtype H1N1, herpes simplex virus 1, and SARS-CoV-2 (1 minute contact).

Conclusions: Spray duo showed antimicrobial activity under in vitro conditions. Further investigations are warranted to evaluate the antimicrobial activity in clinical practice.

Keywords: antimicrobial activity, bacteria, benzydamine hydrochloride, cetylpyridinium chloride, enveloped virus, fungi, oral spray formulation.

Citation

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Introduction

Respiratory tract infections (RTIs) are amongst the three most common human diseases, characterized by typical symptoms such as rhinitis, cough, fever and sore throat.[1] Viruses represent the prevalent cause of RTIs arising in the oral cavity and throat area,[2] whereas bacterial RTIs are less common and often develop after having a viral infection.[3]

It has been estimated that about 200 different viruses can infect the human airways, including influenza viruses, coronaviruses, rhinoviruses and adenoviruses.[4] In particular, human coronaviruses are considered responsible for 10–15% of all upper RTIs.[5] Human pathogenic viruses are mainly represented by enveloped viruses with a lipid bilayer, sharing the ability to infect their target cells by inducing the fusion of the viral envelope with the cell membrane.[6] Human pathogenic viruses replicate in the nasopharynx and cause mild, self-limited upper RTIs; however, lower RTIs and pneumonia have been described, especially after the emergence of more virulent coronaviruses such as SARS-CoV-1 in 2002 and Middle East respiratory syndrome coronavirus in 2012.[5,7] As of 2020, SARS-CoV-2 has been added to the list of human coronaviruses with potentially severe consequences since COVID-19 caused a global pandemic with over 6–7 million deaths globally.[8]

The principal transmission route of respiratory microbes is the secretion of droplets during exhalation; indirect contact with contaminated surfaces has also been shown to spread infection.[9] Therefore, reducing the microbial load at the site of infection, such as the upper respiratory tract, can be considered a relevant approach

to lowering the risk of transmission via both routes, simultaneously reducing symptoms and the potential spread of infection to the lower respiratory tract.

Benzydamine hydrochloride (BNZ) and cetylpyridinium chloride (CPC) are well known drugs, widely distributed worldwide and with extensive clinical and safety data available.[10,11] The proven anti-inflammatory effect of BNZ acts by inhibiting the production and activity of mediators involved in the inflammatory process and by stabilizing the biological membranes of platelets and other pro-inflammatory cells.[12,13] BNZ also exhibits antimicrobial and antifungal activity in vitro.[14,15] CPC is a quaternary ammonium compound with properties and uses typical of cationic surfactants. It exhibits bactericidal activity against many Gram-positive bacteria and, in higher concentrations, some Gram-negative bacteria.[11] CPC also has variable antifungal activity and is effective against some viruses.[16–18] Due to its surfactant properties, it has prolonged activity in the oral cavity as it binds to glycoproteins covering the teeth and oral mucosa.[3]

The primary objective of the present study was to evaluate the in vitro antimicrobial activity of a new oral spray formulation combining 0.15% BNZ and 0.5% CPC (Tantum Verde DUO®, Angelini Pharma S.p.A., hereinafter the spray duo). In addition to bacteria and *Candida albicans*, which are included in the minimum requirements for evaluating the antimicrobial activity of antiseptic products, modified vaccinia Ankara (MVA), influenza A virus subtype H1N1, and herpes simplex virus 1 (HSV1) were also tested to assess the antiviral properties of the formulation. Moreover, the spray duo was tested against SARS-CoV-2.

Methods

Tests were run according to the EP 5.1.11. 'Determination of bactericidal, fungicidal, or yeasticidal activity of antiseptic medicinal products' (EU Pharma 07/2017:50111; EN 13727 bactericidal and EN 13624 yeasticidal guidelines[19–21]) or according to the EN 14476:2013+A2:2019 standard (Chemical disinfectants and antiseptics – Quantitative suspension test for the evaluation of virucidal activity in the medical area – Test method and requirements (Phase 2/Step 1)[22]) (see Table 1 for details).

Determination of bactericidal and yeasticidal activities of the spray duo

The bactericidal and yeasticidal activities were determined by adding the study product (0.15% BNZ and 0.5% CPC; batch number 00368IP02) to a defined number (1–5 × 10^8 colony-forming unit (CFU)/mL) of bacteria, namely *Staphylococcus aureus* (ATCC 6538), *Enterococcus hirae* (ATCC 10541), *Pseudomonas aeruginosa* (ATCC 15442), *Escherichia coli* K12 (DSM 11250), *Haemophilus influenzae* (ATCC 49766), *Moraxella catarrhalis* (ATCC 8176), and *Streptococcus pyogenes* (ATCC 12344). For yeast, the study product was added to 1–5 × 10^7 CFU/mL of *C. albicans* (ATCC 10231).

Current EP 5.1.11 EU Pharma 07/2017:50111

The study product was tested as is (undiluted; 80% in-test). Bovine serum albumin (BSA) solution with a final concentration of 0.03% was used as the interfering substance.

One millilitre of each microorganism suspension was mixed with an equivalent volume of interfering substance in sterile tubes for 2 minutes. Subsequently, 8 mL of disinfectant was added to the tubes without mixing and the tubes were kept at 33 ± 1°C for the chosen contact times (1 or 5 minutes). At the end of contact time, an aliquot was taken, and the antimicrobial/yeasticidal activity was immediately stopped by a validated membrane filtration process; 1 mL of the mixture was transferred to a new tube containing 8 mL of neutralizer, and 1 mL of sterile distilled water. The tubes were mixed by vortex for 10 seconds. After the neutralization process, the living microorganisms were enumerated using the pour plate technique. Inoculated petri dishes were incubated at 37°C for bacteria and 30°C for fungi for 48 hours. All studies were performed in triplicate.

Finally, the selected experimental conditions and the membrane filtration method were verified.

EN norm 13727 of disinfection bactericidal tests

The study product was tested based on EN 13727[20] as is (undiluted; 80% in-test). The inocula were prepared with the tripartite soil load as the interfering substance, which consists of BSA solution, mucin bovine submaxillary gland type I-S, and yeast extract according to the Organization for Economic Co-operation and Development guidance document on Quantitative Methods for Evaluating the Activity of Microbicides Used on Hard Non-Porous Surfaces.[23] The reaction mixtures were held with the study product for contact times of 30 seconds, 1 minute, 3 minutes, and 5 minutes at 20°C. Following contact times, a 1 mL aliquot of the reaction mixture was drawn up and neutralized in 1 mL of ice-cold neutralizer, which was further quenched 300-fold with ice-cold DM within 30 minutes. The tubes were mixed by vortex for 10 seconds. After the neutralization process, the living microorganisms were counted using the spread plate technique. Inoculated petri dishes were incubated at 37°C for bacteria. Finally, the selected experimental conditions and the membrane filtration method were verified.

Reduction in viability

The reduction in the viability (R) of the microorganism was calculated by applying the following formula: $R = \log N_0 - \log Na$, where N_0 is the number of CFU/mL of the test suspension/10, and Na is the number of CFU of the suspension of microorganisms that survive the assay. For cases where the Na value was 'not countable', R values were expressed as <5-log for bactericidal activity and <4-$\log_{10}$ for yeasticidal activity. The study product was considered to have bactericidal activity if the number of CFU was reduced by at least 5-$\log_{10}$ within 5 minutes when the test microorganisms were bacteria. The study product was considered to have yeasticidal activity if the number of CFU was reduced by at least 4-$\log_{10}$ within 5 minutes at the test temperature of 33°C with a 0.03% final concentration of BSA solution when the test microorganism was *C. albicans*. Experimental condition control must be >0.5 × Nv/10 (Nv: number of CFU/mL in the validation suspension), and membrane filtration method control must be >0.5 × Nv/10/10 for a valid test.

Virucidal activity

The study product was evaluated against MVA (Centre of Veterinary Public Health of the University Leipzig; ATCC VR-1518), influenza virus A/Michigan/45/2015 (H1N1; ATCC VR-1469), HSV1 strain MacIntyre (ATCC VR-539) and SARS-CoV-2 strain USAWA1/2020 (BEI Resources NR-52281).

Each virus was propagated in a specific cell line that supports its optimal replication. In detail, the stock viruses were prepared by infecting MDCK (H1N1), BHK-21 (MVA) or embryonic lung fibroblasts (HSV1) cell lines. The stock SARS-CoV-2 was prepared by infecting Vero E6 cells. The selection of these cell lines aligns with standard virological methods to ensure reliable viral propagation and assessment of antiviral activity. The cultures were frozen at −60°C to −90°C several days after infection. After freezing and thawing, cell-free stocks were prepared by centrifugation. The stock virus was then aliquoted and stored at −60°C or below until used in testing.

EP 5.1.11 EU Pharma 07/2017:50111

A concentration of the study product was tested: neat (80% in-test). BSA solution with a final concentration of 0.03% was used as the interfering substance.

First, 1 mL of the suspension for each microorganism was mixed with an equivalent volume of interfering substance in sterile tubes for 2 minutes. Subsequently, 8 mL of disinfectant was added to the tubes without mixing, and the tubes were kept at 33 ± 1°C for the chosen contact times (30 seconds and 3 minutes). At the end of the contact time, an aliquot was taken, and the virucidal activity was immediately stopped by filtration with Sephadex LH20 columns and dilution in an iced culture medium; 1 mL of the mixture was transferred to a new tube containing 8 mL of neutralizer and 1 mL of sterile distilled water. The tubes were mixed by vortex for 10 seconds. After the neutralization process, the living microorganisms were enumerated using the endpoint titration method. In order to evaluate the cytotoxic effect of the test item on BHK-21 (C-13) ATCC CCL-10, a preliminary cytotoxicity test was performed. Despite using MicroSpin™ S400 HR and Sephadex LH-20 columns, residual cytotoxicity was observed on the BHK-21 (C-13) ATCC CCL-10 cell line at the test item concentration. Therefore, the large-volume plating (LVP) method was used to determine the residual virus at the test concentration of 80%. Using the LVP method, the lowest apparently non-cytotoxic dilution (1:100,000 in this case) was added to a defined number of wells containing the host cell line. This method improved the detection of residual virus by plating a large sample volume. In parallel, the test procedure was conducted following the standard Spearman–Kärber method.

EN 14476 norm of disinfection virucidal tests

A concentration of the study product was tested as is (80% in-test; 90% in-test for SARS-CoV-2). The inoculates were prepared with the tripartite soil load (interfering substance), which consists of BSA solution, mucin bovine submaxillary gland type I-S, and yeast extract according to the Organization for Economic Co-operation and Development guidelines.[23]

Tests were conducted once at 20 ± 1°C according to EN 14476 and repeated at 33°C following EU Pharmacopoeia standards for antiseptics, simulating oral mucosa conditions. The test assay comprised 100 µL of virus suspension, 100 µL of interfering substance and 800 µL of the test product (undiluted). A virus control mixture was also evaluated using double-distilled water instead of the test product. After the specified contact time (30, 60 or 180 seconds), virucidal activity was immediately suppressed by dilution with nine volumes of ice-cold medium (minimal essential medium +2.0% fetal calf serum) and serially diluted 10-fold. Infectivity was determined by end-point dilution titration in microtiter plates. Aliquots of 100 µL from each dilution were added to six 200 µL samples of cells. Cultures were examined microscopically for cytopathic effects after the respective incubation time of the cell line used. The LVP assay, according to EN 14476:2019-10,[22] is sometimes necessary to detoxify the test mixtures to achieve at least a 4-$\log_{10}$ reduction in the virus titre. Using the LVP method, the lowest apparently non-cytotoxic dilution of the test mixture was added to a large volume of ice-cold medium after the specified contact time and then added to 96-well plates with 90% confluent cells.

For SARS-CoV-2, tests were conducted according to the European Standard EN 14476:2013+A2:2019 guidelines.[22] The reaction mixture (1 mL) was added to 9 mL of the

Table 1. Bactericidal, yeasticidal and virucidal tests.

Norm	Microorganisms	Results
According to current EP 5.1.11 EU Pharma 07/2017:50111*		
Current EP 5.1.11 EN 13727 bactericidal	*Staphylococcus aureus* *Enterococcus hirae* *Pseudomonas aeruginosa* *Escherichia coli*	Study product active at 1 minute
Current EP 5.1.11 EN 13624 yeasticidal	*Candida albicans*	Study product active at 5 minutes
Current EP 5.1.11 EN 14476 virucidal	MVA H1N1	Study product active at 3 minutes Study product active at 30 seconds
According to the EN disinfectant norm**		
EN 13727 bactericidal	*S. aureus* *Haemophilus influenzae,* *Moraxella catarrhalis* *Streptococcus pyogenes*	Study product active at 3 minutes Study product active at 30 seconds Study product active at 30 seconds Study product active at 30 seconds
EN 14476 virucidal	MVA H1N1 HSV1 SARS-CoV-2	Study product active at 3 minutes Study product active at 30 seconds Study product active at 30 seconds Study product active at 30 seconds

*Tested at 33°C with 0.3 g/L bovine serum albumin solution as interfering condition.
**Tested at 20°C with Organization for Economic Co-operation and Development tripeptide solution as the interfering condition.[23]
H1N1, influenza A virus subtype; HSV1, herpes simplex virus 1; MVA, modified vaccinia Ankara.

study product and mixed. The reaction mixture was then held for contact times of 1, 3 and 5 minutes at 22°C. Following contact times, a 1 mL aliquot of the reaction mixture was drawn up and neutralized in 1 mL of ice-cold neutralizer, which was further quenched 300-fold with ice-cold dilution medium within 30 minutes. This post-neutralized sample was considered undiluted.

The 50% Tissue Culture Infectious Dose per mL value ($TCID_{50}$/mL) was calculated by the Spearman-Kärber method and converted to $\log_{10}$ $TCID_{50}$ viral load. The viral titre of each sample was reported as a ± 95% confidence interval. According to the guidelines, the study product was considered to possess virucidal activity if there was at least a ≥4-$\log_{10}$ reduction in titre above the cytotoxicity level. Controls included the neutralization control, the cytotoxicity control determining whether the study product was cytotoxic to the culture cells, a viral recovery control quantifying viable virus after simulating the assay process, and a cell viability control verifying whether cells were viable for the duration of the incubation.

Results

Details and main results for each test are summarized in Table 1.

Bacteria and yeast

Under the tested conditions, the study product could be considered with bactericidal activity at the contact time of at least 1 minute and with both bactericidal and yeasticidal activities at the contact time of at least 5 minutes (Table 2). All controls met the valid test criteria according to the European norm of disinfectants and antiseptics (EN 14885:2022 Chemical disinfectants and antiseptics – Application of European Standards for chemical disinfectants and antiseptics).[24]

Viruses

MVA as a surrogate virus for enveloped viruses

Under the tested conditions, the study product was active against MVA after at least 3 minutes of contact, resulting in >4.3-$\log_{10}$ reductions in viral recovery (Table 3). All controls met the valid test criteria according to the European norm of disinfectants and antiseptics.

Influenza A virus subtype H1N1

Under the tested conditions, the study product was active against seasonal influenza virus A starting from 30 second contact, resulting in >4-$\log_{10}$ reductions in viral recovery (Table 3). All controls fulfilled the required test criteria in accordance with the European standards for disinfectants and antiseptics.

Table 2. Spray duo bactericidal and yeasticidal activities.

Microorganisms	$LogN_0^*$ (Log N/10)	Contact times	$LogN_a^*$	Reduction in viability (R)
Bactericidal and yeasticidal activities of the study product according to current EP 5.1.11 EU Pharma 07/2017:50111				
Staphylococcus aureus	7.64	1 minute 5 minutes	<2.00	>5.64
Pseudomonas aeruginosa	7.66	1 minute 5 minutes	<2.00	>5.66
Escherichia coli	7.69	1 minute 5 minutes	<2.00	>5.69
Enterococcus hirae	7.54	1 minute 5 minutes	<2.00	>5.54
Candida albicans	6.61	1 minute 5 minutes	N.A. 2.52	<4.00 4.09
Bactericidal tests according to EN norm of disinfectants and antiseptics				
Staphylococcus aureus	8.57	30 seconds 1 minute 3 minutes 5 minutes	>4.22 >4.22 2.54 <2.15	>3.35 >3.35 5.02 >5.42
Haemophilus influenza	8.60	30 seconds	<2.15	>5.42
Moraxella catarrhalis	8.69	30 seconds	<2.15	>5.42
Streptococcus pyogenes	8.37	30 seconds	<2.15	>5.42

*Reported log_{10} values are the mean of the three replicates.

HSV1

Under the tested conditions, the study product was active against HSV1 starting from a 30 second contact, resulting in >4-log_{10} reductions in viral recovery (Table 3). All controls complied with the validity criteria established by the European standards for disinfectants and antiseptics.

SARS-CoV-2

Under the tested conditions, the study product was an effective virucidal agent against SARS-CoV-2 starting from 1 minute contact, resulting in >4.15-log_{10} reductions in viral recovery (Table 3). All controls met the acceptance criteria defined by the European standards for disinfectants and antiseptics.

Discussion

The transmission route of RTIs is currently considered to be via respiratory droplets, and microbial particles can be viable in aerosols for up to 3 hours.[25,26] Assuming that the throat is the main site of microbial replication in the early stages of infection, the use of topical agents that can damage or destroy microbes has the potential to reduce the viral load in the oropharynx. Consistent with this hypothesis, several clinical cases have confirmed the efficacy of mouth rinses in reducing the SARS-CoV-2 load in saliva.[27–30]

In this in vitro study, the antimicrobial effect of an oral spray formulation combining 0.15% BNZ and 0.5% CPC (spray duo) was tested against bacteria, yeast and enveloped viruses. To the best of our knowledge, this is the first in vitro study to examine the virucidal effect of a spray formulation containing both BNZ and CPC on SARS-CoV-2, influenza A virus H1N1 and HSV1.

Our findings support the bacterial and yeasticidal activity of the spray duo against *S. aureus*, *P. aeruginosa*, *E. coli*, *E. hirae* after 1 minute contact, against *H. influenzae*, *M. catarrhalis and S. pyogenes* after 30 second contact and against *C. albicans* after 5 minute contact.

The study product also showed virucidal activity, reporting a ≥4-log_{10} reduction in titre above the cytotoxicity level against MVA (3 minute contact), H1N1 (30 second contact), HSV1 (30 second contact) and SARS-CoV-2 (1 minute contact).

Previous studies have shown that the concentration of SARS-CoV-2 in saliva samples of infected individuals is on the order of 4–6-log_{10} genome copies per millilitre,[25,31]

Table 3. Spray duo virucidal activity.

Virus	Log $TCID_{50}$ titre of the virus control (log_{10} $TCID_{50}$/mL)	Contact times	Titre of the 'residual virus' inactivation (log_{10} $TCID_{50}$/mL)	Log reduction*
Current EP 5.1.11 EU Pharma 07/2017:50111				
MVA (LVP method)	7.67 ± 0.346	3 minutes 5 minutes	≤3.33 ≤3.33	≥5.00 ± 0.346
Influenza virus A subtype (H1N1; LVP method)	7.67 ± 0.346	30 seconds 3 minutes	≤3.26 ≤3.26	≥5.24 ± 0.000
According to EN 14476:2019				
Virus	**Titre of the virus control (log_{10} $TCID_{50}$/mL) with 95% CI**	**Contact times**	**Titre of the 'residual virus' inactivation (log_{10} $TCID_{50}$/mL) with 95% CI**	**Reduction factor with 95% CI***
MVA	7.83 ± 0.42	30 seconds 1 minute 3 minutes	5.45 4.62 3.48	2.38 ± 0.42 3.21 ± 0.42 4.35 ± 0.42
Influenza virus A subtype (H1N1)	6.67 ± 0.33	30 seconds 1 minute 3 minutes	2.64 2.63 2.63	4.03 ± 0.33 4.04 ± 0.33 4.04 ± 0.33
HSV1 (LVP method)	6.67 ± 0.33	30 seconds 1 minute 3 minutes	2.58 2.58 2.58	4.09 ± 0.33 4.09 ± 0.33 4.09 ± 0.33
SARS-CoV-2	7.57 ± 0.18	1 minute 3 minutes 5 minutes	≤3.35 ≤3.35 ≤3.35	≥4.15 ≥4.15 ≥4.15

*>4 denotes a complete inactivation of the challenged virus.
HSV1, Herpes simplex virus 1; LVP, large-volume plating; MVA, modified vaccinia Ankara.

with the highest concentration occurring 5–6 days after symptom onset. In our in vitro study, the virus concentration was comparable, ranging from 6.6-log_{10} to 7.8-log_{10} $TCID_{50}$/mL, suggesting the potential in vivo activity of the study product against saliva viral load as well.

Our findings align with previous literature evidence showing the in vitro virucidal effect of the fixed combination of BNZ and CPC as a throat lozenge on SARS-CoV-2.[3] However, further clinical studies are required to strengthen the indication for the use of the study product as the currently available evidence on the two components refers mainly to in vitro studies[3,32] whilst in vivo evidence is contrasting.[30,33]

Results obtained with the fixed combination of BNZ and CPC align with, or even exceed, the effectiveness reported for other CPC-based formulations. For instance, Peiter et al. showed that lozenges containing 1.4 mg CPC and 10 mg benzocaine exhibited bactericidal activity within 5 minutes against various pharyngitis-associated microorganisms as well as virucidal activity against influenza A virus and bovine coronavirus.[34] However, in that formulation, CPC is present at a lower concentration (approximately 0.07%), and benzocaine acts purely as a local anaesthetic, without anti-inflammatory properties.

Similarly, Donath et al. evaluated lozenges combining lidocaine with 2 mg CPC (Mebucaine CL) and found modest symptomatic relief but no significant improvement in virucidal efficacy compared to higher-CPC formulations.[35]

Furthermore, Meister et al. systematically assessed several antiseptic agents in vitro and confirmed that CPC at concentrations between 0.05% and 0.1% significantly reduced SARS-CoV-2 infectivity, likely via disruption of the viral lipid envelope.[36] This supports the observed efficacy of the spray duo, in which CPC is present at an optimal concentration for broad-spectrum antimicrobial activity within clinically relevant contact times. In contrast, lozenges containing amyl-

metacresol and 2,4-dichlorobenzyl alcohol exhibited only limited virucidal activity against respiratory viruses such as influenza A and human coronavirus OC43, with notable variability depending on the specific formulation and excipients.[37] Only selected amylmetacresol/2,4-dichlorobenzyl alcohol lozenges demonstrated moderate antiviral effects in vitro, and their activity was significantly lower – up to 1368-fold – compared to carrageenan-containing formulations.[37]

Overall, the spray duo appears advantageous, coupling the anti-inflammatory and analgesic properties of benzydamine with the antiseptic efficacy of CPC. At the same time, some limitations can be reported. Indeed, this study was conducted exclusively under in vitro conditions, which do not fully replicate the complex environment of the oral cavity and the dynamics of viral or bacterial colonization in vivo. Furthermore, the antimicrobial activity was assessed at predefined contact times that may not directly reflect the exposure conditions during real-life use of the spray.

Conclusion

In conclusion, the new oral spray formulation combining 0.15% BNZ and 0.5% CPC showed bactericidal, yeasticidal and virucidal activity under in vitro conditions. Further investigations are warranted to evaluate the study product's antimicrobial activity in clinical practice and its clinical effects. If further clinical data confirm the properties of the new oral spray formulation, the investigated product will certainly be a useful therapeutic tool for the treatment of upper RTIs.

Contributions: Study design: ACdJ and ME; data collection and interpretation: all; manuscript writing: ACdJ; manuscript editing: all. All named authors meet the International Committee of Medical Journal Editors (ICMJE) criteria for authorship for this article, take responsibility for the integrity of the work as a whole, and have given their approval for this version to be published.

Data availability: The datasets generated during and/or analysed during the current study are available from the corresponding author upon reasonable request.

Disclosure and potential conflicts of interest: ACdJ and LB are employees at Angelini Pharma S.p.A. The International Committee of Medical Journal Editors (ICMJE) Potential Conflicts of Interests form for the authors is available for download at: https://www.drugsincontext.com/wp-content/uploads/2025/07/dic.2024-9-3-COI.pdf

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