ColdZyme® Mouth Spray reduces duration of upper respiratory tract infection symptoms in endurance athletes under free living conditions.

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GD conceived and designed the research. GD, EP, AWJ, GMS, ARJ, HR, and KD conducted the research. GD wrote the manuscript and all authors read, edited and/or approved the final manuscript**.**

Acknowledgements and Declaration of Interests

This study was funded by Enzymatica AB (Sweden), manufacturer of ColdZyme® Mouth Spray. GD has also provided expert opinion to Enzymatica AB (Sweden).

Abstract

Upper respiratory tract infection (URTI) can compromise athlete preparation and performance, so countermeasures are desirable. The aim of this study was to assess the effects of ColdZyme® Mouth Spray (ColdZyme) on self-reported upper respiratory tract infection in competitive endurance athletes under free-living conditions.

One hundred and twenty-three endurance-trained, competitive athletes (recruited across 4 sites in England, UK) were randomised to control (no treatment, n = 61) or ColdZyme (n = 62) for a 3-month study period (between December 2017 – February 2018; or December 2018 – April 2019). They recorded daily training and illness symptoms (Jackson common cold questionnaire) during the study period.

A total of 130 illness episodes were reported during the study with no difference in incidence between groups (episodes per person: 1.1 ± 0.9 Control, 1.0 ± 0.8 ColdZyme, P = 0.290). Episode duration was significantly shorter in ColdZyme compared to Control: Control 10.4 ± 8.5 days vs ColdZyme 7.7 ± 4.0 days, P = 0.016). Further analysis to compare episodes with poor vs good compliance with ColdZyme instructions for use (IFU) within the ColdZyme group showed a further reduction in duration of URTI when compliance was good (9.3 ± 4.5 days in ColdZyme poor IFU compliance vs 6.9 ± 3.5 days in ColdZyme good IFU compliance, P = 0.040).

ColdZyme may be an effective countermeasure to reduce URTI duration, which was significantly lower (by 26-34%) in the ColdZyme treatment group (with no influence on incidence). This may have implications for athlete performance.

Key words

Common Cold, Illness, Training, Exercise, Immunology, Countermeasure

Abbreviations

ANOVA	analysis of variance
IFU	instructions for use
OTC	over-the-counter
PCR	polymerase chain reaction
sRPE	session rating of perceived exertion
URS	upper respiratory symptoms
URT	upper respiratory tract
URTI	upper respiratory tract infection

Introduction

The incidence of upper respiratory illnesses is higher than normal in some groups of athletes, and such infections can compromise training and/or competition performance.[1] Endurance athletes are often associated with a higher than normal incidence of infections, especially of the upper respiratory tract (URTI).[2,3] This is typically related to a high training load and/or heavy competition schedule. More recent debates have questioned whether athletes do experience a higher incidence, compared to the general adult population (i.e. one to three individual episodes of upper respiratory tract infection per year[4,5]). However, it is clear that reporting of upper respiratory symptoms (URS) in athletes cluster around periods of intensive training and/or competition.[6,7] Experiencing URTI or illness symptoms can result in a loss of training days and a performance decrement ([6,8,9]) so strategies to reduce the risk of contracting these illnesses may be of direct benefit to athletes. This may also limit the risks of spreading infection to others (i.e. teammates). The possible links between URTI incidence and athletic performance is highlighted by research showing World Championship and Olympic medal winning athletes reported fewer URS than less successful athletes.[6,10,11] This is likely related, at least in part, to the ability (and resource) to successfully implement strategies that reduce URTI risk.[12,13]

Strategies to minimise the risk of contracting a URTI and/or reduce time taken to clear an infection have focussed on avoidance of exposure and minimising the controllable risk factors that are associated with lowered immune defence (e.g. intensified training, life stressors), but these may be difficult to avoid for many athletes.[6,14] Other strategies have focussed on nutritional interventions purported to reduce the immune perturbations caused by strenuous exercise and training. Unfortunately, many such strategies have limited success.[11–15] An alternative strategy that has received little attention in athletic populations, is the use of products that may inhibit viral infectivity (for example, via limiting viral entry or replication/propagation after initial exposure). Most URTIs are caused by viral infection, with over 200 known viruses, the most common being rhinoviruses, coronaviruses, influenza viruses, adenoviruses, parainfluenza viruses, respiratory syncytial viruses and enteroviruses.[16] Infection is initially established in the mucosa of the nasopharynx before spreading anteriorly, through the nasal region ([17]), with local symptoms typically beginning in the throat before nasal congestion, rhinorrhoea, sneezing and cough tend to develop.[18]

It is possible that inhibiting viral propagation in this area during the incubation period, may prevent or shorten the duration of viral URT infection. ColdZyme® Mouth Spray (ColdZyme) consists of a hyperosmotic glycerol solution containing cold-adapted trypsin from the Atlantic cod (*Gadus*

morhua) and has been shown to reduce URTI duration in a number of studies in healthy and clinical (i.e. non-athletic) populations.[19,20] It is suggested that orally spraying of the solution forms a temporary barrier on the pharynx that prevents viral binding and entry. The idea that local effects in this part of the URT can successfully reduce URTI duration is also supported by other studies showing effective protection against the common cold with substances administered orally (e.g. zinc lozenges [21]). ColdZyme spray solution has demonstrated broad antiviral activity *in vitro*, deactivating 64-100% of virus activity for common URTI-causing pathogens (influenza virus, rhinovirus, adenovirus and coronavirus).[22] Clarsund et al. ([19]) found that ColdZyme treatment was effective against the common cold in healthy adults inoculated with rhinovirus-16: most notably the duration of illness was reduced by 54% in those who were infected. Also, Clarsund et al. ([20]) reported a case study of a 12-year old boy with common variable immunodeficiency, and found a reduction in reported common cold infection and a 3-fold decrease is missed school days when using ColdZyme. However, no randomised controlled trials have examined whether such products can reduce URTI/URS incidence or duration in athletic populations. One recent study ([23]) did examine ColdZyme in athletes, but it lacked a control group and made comparisons with retrospective historical data from athlete's own training diaries, which has obvious limitations for establishing efficacy. The aims of this study were, therefore, to assess the efficacy of ColdZyme on URTI incidence, symptom ratings, and missed (or reduced) training in competitive endurance athletes under free-living conditions, in a prospective randomised controlled trial.

Methods

Type of Study:

Prospective, open label, parallel groups, randomised controlled trial.

Participants:

Endurance-trained; competitive athletes (e.g. long-distance runners; triathletes; cyclists) were recruited. Participants were excluded if on long term medication; currently smoking; allergic to any of the ingredients in ColdZyme; had any other current medical conditions that may be aggravated by use of the product; were currently using any medication (except for contraceptives), or food supplements; were currently using any other relevant products or supplements (nutritional or otherwise) that may influence the common cold; were currently taking part in another study that may compromise results of this study; or were pregnant, breast-feeding or planning to become pregnant during the study.

Ethical approval:

The study was conducted in accordance with the Declaration of Helsinki and approved by the ethics committees of the authors' Universities. All participants were informed, both verbally and in writing, of the nature and risks of the study before giving their written consent to take part.

Design:

Athletes were monitored over a 3-month period of using ColdZyme product (or control). During this period they completed self-report training logs, and the Jackson common cold questionnaire ([24]). The ColdZyme group were also required to keep a personal record of product usage. The study period was in UK winter months. Tranche 1 took place between December 2017 – March 2018 and Tranche 2 took place between December 2018 and April 2019. Tranche 1 took place in Southeast England (East Kent and Medway areas). Tranche 2 took place in Southeast England, as in Tranche 1, plus at 3 additional sites: Tonbridge, UK; Merseyside, UK, and Lincolnshire, UK. Participants were stratified by sport type, sex, age, and usual training volume and randomised to either control or ColdZyme group by random number generation (www.randomization.com). Randomisation was also applied at each site independently so that groups were matched within each location. The allocation schedule was concealed from investigators involved in recruiting the participants, hence group allocations were only provided one at a time.

Treatment:

Participants in the treatment (ColdZyme) group were asked to use the ColdZyme product in accordance with manufacturer instructions for treatment of suspected common cold/URTI (at first self-perceived signs of URTI). Briefly, this included instruction to spray 2 times (1 dose) every second hour up to 6 times daily.

Participants in the control group were asked to continue with their normal training, to log all activities and URTI (Jackson questionnaire) but were not provided with ColdZyme. Participants were not restricted from using over-the-counter (OTC) medication if they felt it necessary, but were required to record any usage in their illness log.

Training monitoring:

Participants were required to log s every exercise training session. They were required to provide details on exercise type, duration and a single overall rating of their perceived exertion for the session (using the session rating of perceived exertion method, sRPE [25]).

Monitoring of upper respiratory illness:

Participants were required to complete the Jackson questionnaire daily ([24]). Completion of this prospective questionnaire first requires participants to indicate if they believe they are suffering from a common cold/URTI. If they answer yes to the initial question, they must then rate which of the 8 Jackson score symptoms they are suffering (headache, chilliness, sneezing, sore throat, malaise, cough, nasal discharge, nasal obstruction) and give a rating of the severity of each symptom experienced (0, none; 1, mild; 2, moderate; 3, severe). An episode of URTI was defined using the Jackson criteria (as applied by Martineau et al.[26]): scores for each of 8 symptoms were summed for each day to generate a total Jackson score, and an episode was defined as those lasting ≥ 3 days and with either i) a total Jackson symptom score of ≥14 + subjective impression of having a cold (question 1), or ii) a total Jackson symptom score of ≥14 + nasal discharge for at least 3 days, or iii) a total Jackson symptom score <14 + subjective impression of having a cold + nasal discharge for at least 3 days.

Data analysis:

All data analysis was conducted using IBM SPSS statistics version 25 (IBM, Armonk, NY). Data were checked for normal distribution prior to analysis. Data that did not have a normal distribution (sRPE-based training load) were normalised via log transformation prior to analysis. Data that could not be normalised by log or square root transformation (missed and reduced training) were analysed with non-parametric tests. Group comparisons were made using independent samples t-test (or for missed and reduce training data, Mann-Whitney U test; and for OTC medication use, chi-squared analysis). Training load variables were compared between groups, and across repeated time-points (weekly over study period, and weeks before, during and after URTI episodes) with 2-way mixed ANOVA (between factor: group, and within group/repeated factor: time point). Post hoc paired t-tests were used, where necessary, to compare within/repeated time-points following significant main effects in this factor.

Results

A total of 130 subjects were enrolled, but 7 were lost to follow-up (n = 3 [2 Control, 1 ColdZyme] due to injury meaning no regular training during study period; n = 2 [1 Control, 1 ColdZyme] voluntarily withdrew before completion; n = 2 [1 Control, 1 ColdZyme] did not return logs and could no longer be contacted/did not reply to communications). Analysis was completed on n = 123 (n = 61, age 39.3 ± 11.5 years, control and n = 62, age 39.5 ± 12.1 years, ColdZyme; male n = 60, female n = 63). Athletes were all competitive endurance athletes in current training, ranging from

club level athletes to age-group international level, with comparable average training load in each group (see below). The majority of athletes were runners (n = 38 control, n = 44 ColdZyme), then triathletes (n = 13 control, n = 13 ColdZyme) and cyclists (n = 8 control, n = 4 ColdZyme) with a small number from other sports (swimming n = 1 control, n = 1 ColdZyme; and rowing n = 1 ColdZyme).

At least one URTI episode was recorded during the study period in 76.4% of all participants (77.0% control, 75.8% ColdZyme). In total 130 episodes were recorded by all participants over the study period with no difference between groups in the incidence rate (mean incidence rate per person over study period was: 1.1 ± 0.9 Control, 1.0 ± 0.8 ColdZyme, P = 0.290). Symptom duration and severity ratings were also lower in the ColdZyme group (see Table 1).

Please insert Table 1 near here

Symptoms duration

Overall Control vs ColdZyme mean episode duration was 10.4 ± 8.5 days in Control and 7.7 ± 4.0 days in ColdZyme (P = 0.016, see Table 1). On further examination of usage records and diaries from Tranche 1 (December 2017-February 2018) it became apparent that, despite being instructed to follow the manufacturer's IFU (i.e. to use 1 dose every second hour up to 6 times daily), not all of the participants in the ColdZyme group followed the recommendations for use (~37% of recorded episodes were not treated according to the ColdZyme IFU). For Tranche 2 (December 2018-March 2019) we provided additional information and regular reminders to participants to overcome this. These procedures appear to have been effective as poor compliance with IFU was only evident in 14% of reported episodes. Nevertheless, these subjects/episodes (with poor IFU compliance) from both Tranches do provide a useful comparator group for some statistical comparisons, which may overcome some of the limitations that arise in open label trials (i.e. potential placebo effects in treatment group). Poor compliance with IFU was considered as not using the ColdZyme product in accordance with guidelines (i.e. less than 4 doses per day). When those randomised to ColdZyme but with poor compliance (Poor IFU comp) were separated from those with good compliance (Good IFU comp) the observed effect between Control and ColdZyme groups becomes even more evident (episode duration 10.4 ± 8.5 days in Control vs 6.9 ± 3.5 days in ColdZyme Good IFU comp, P = 0.004). Direct comparison between compliance groups also shows a significantly shorter episode duration with good compliance (episode duration 9.3 ± 4.5 days in ColdZyme poor IFU comp vs 6.9 ± 3.5 days in ColdZyme Good IFU comp, P = 0.040).

Jackson Symptom Score

***Total symptom score during episode*:** This parameter shows the overall symptom impact (product of number of Jackson symptoms and symptom severity ratings, and accounting for episode duration). Overall Control vs ColdZyme mean Jackson symptom score was 74.9 ± 72.0 in Control and 43.6 ± 30.1 in ColdZyme (P = 0.003, see Table 1). When comparing episodes with good and poor IFU compliance, the ColdZyme Good IFU comp group had a significantly lower symptom score (40.0 ± 26.5) than the Control group (P = 0.002), whereas the Poor IFU comp group (52.6 ± 35.7) were not significantly different from control (P = 0.100). Direct comparison between the episodes with bad and good IFU compliance did not show any statistically significant difference however (P = 0.152).

***Average symptom score per day during episode*:** This parameter is directly related to the severity rating and number of symptoms experienced on average (per day) in each episode. Overall Control vs ColdZyme mean Jackson symptom score was 6.9 ± 2.8 in Control and 5.5 ± 2.4 in ColdZyme (P = 0.006, see Table 1). When comparing episodes with good and poor IFU compliance, the ColdZyme Good IFU comp group had a significantly lower symptom score (5.6 ± 2.7) than the Control group (P = 0.014), and so did the ColdZyme Poor IFU comp group (5.3 ± 1.7) (P = 0.033). Direct comparison between the episodes with bad and good IFU compliance did not show any statistically significant difference (P = 0.481).

Daily training logs

Participants typically trained between 4 and 10 h per week. Participants were asked to record every training session (duration and sRPE) in their training logs. Training 'load' was quantified as the product of sRPE and duration for each session (and summed each week). Sufficient detail to allow full analysis of training data was provided by 93 participants (whereas 19 participants [n = 9 Control, n = 10 ColdZyme] failed to record sRPE but recorded details on training type and duration, and 11 participants [n = 9 Control, n = 10 ColdZyme] failed to record either sRPE or duration).

For the 93 subjects with complete training logs, there were no significant between-group differences in training load or the profile of training load across the study period (2-way mixed ANOVA: group P = 0.925; time P = 0.055; group × time P = 0.626). For the n = 19 who did record duration only there was no difference in average weekly training time (Control 6.0 ± 1.5 h; ColdZyme 5.9 ± 1.8 h, P = 0.851).

For analysis of training load data (see Figure 1) each subject's average healthy (i.e. when not experiencing or recovering from a URTI episode or injury) value was calculated and training load profile across the 12-week period expressed as a percentage of this. There was a trend for training

load to increase across the study period although this did not reach statistical significance, but importantly this pattern did not differ between groups. There was a significant reduction in training load during the weeks in which URTI episodes were experienced ($P < 0.01$ compared to other, none URTI weeks, see Figure 2), however, this did not differ between groups (2-way mixed ANOVA: group $P = 0.424$; time $P < 0.001$; group × time $P = 0.269$). There was also no difference in the rate of return to normal training load, following reported URTIs, between groups.

Please insert Figure 1 near here

Please insert Figure 2 near here

Missed and reduced training days

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Missed training

The number of missed training days (caused by URTI episode/symptoms etc) was significantly lower in the ColdZyme compared to Control group ($P = 0.013$, see Table 2). When considering IFU compliance, there were significant differences between Control and ColdZyme Good IFU comp ($P = 0.021$) and ColdZyme Poor IFU comp ($P = 0.045$). There was no significant difference between ColdZyme Good and Poor IFU comp however ($P = 0.406$).

Reduced training

The number of days on which training was reduced, as a consequence of an episode, was not significantly different between the ColdZyme and Control groups ($P = 0.475$ see Table 2). When considering IFU compliance, there were no significant differences between Control and ColdZyme Good IFU comp ($P = 0.288$) or ColdZyme Poor IFU comp ($P = 0.336$). There was also no significant difference between ColdZyme Good and Poor IFU comp ($P = 0.269$).

Use of OTC medication

Participants felt the need to use OTC medication for 48% of cases (32/67 episodes) in Control and 38% of cases (24/63 episodes) in ColdZyme, with no difference between groups (χ^2 $P = 0.266$). The OTC medications used were most commonly cold and flu formulations (e.g. Lemsip; Control = 14, ColdZyme = 11), followed by analgesics (e.g. paracetamol (acetaminophen); Control = 12,

ColdZyme = 12), with others used rarely (throat lozenges or cough mixture, Control = 5, ColdZyme = 0; and decongestants, Control = 1, ColdZyme = 1).

Adverse events reporting

One participant reported a potential adverse event (unsettled stomach) during the study, in the ColdZyme (treatment) group. It was not serious and no special treatment was necessary. No other adverse events were reported by any participant.

Discussion

This is the first randomised controlled trial to examine the efficacy of ColdZyme mouth spray on URTI outcomes in athletes (and the first to study any intervention of a product purported to act via local mechanisms of inhibiting viral infectivity and propagation in the URT). The main finding from this study is that ColdZyme did not alter the chances of contracting a URTI (no effect on URTI incidence) but it was able to reduce the duration for which URTI symptoms persisted, and reduce mean daily 'severity' ratings, in competitive endurance athletes. This benefit was evident in the ColdZyme group overall, with further analysis showing that the benefit was significantly more apparent when compliance was good (i.e. ~26% vs ~34% shorter episode duration) but was lost for duration if compliance with the IFU was poor. The ColdZyme group also reported fewer missed training days as a consequence of URTI episodes. The average episode duration was ~10.4 days in the control group, 7 days of which (~66%) resulted in compromised training (~3.4 d reduced training and 3.5 d missed training per episode). In the ColdZyme group episodes of URTI had less of an effect on compromising training (4.6 of 7.7 d [~60%]), with the difference resulting from significantly less missed training days (Control 3.5 of 10.4 d [~34%] missed training days per episode, and ColdZyme 1.6 of 7.7 d [~21%] missed training days per episode). These results suggest that ColdZyme mouth spray can reduce symptom duration and severity ratings (during a self-reported URTI) in endurance athletes, consequently reducing the number of missed training days. This may help to reduce the negative impact of an illness episode on athlete performance, since a loss of training days is associated with performance decrement in athletes ([6,8,9]). ColdZyme is a potential countermeasure to reduce the negative impact caused by such illness.

ColdZyme has been shown to have broad-range antiviral activity against common URTI-causing viruses *in vitro*, deactivating 64-100% of virus activity for influenza virus, rhinovirus, adenovirus, and coronavirus ([22]). A possible mechanism for the present results, therefore, is that the oral application via spraying forms a temporary barrier on the oropharynx that prevents viral binding and entry, and that the regular reapplication (i.e. up to 6 times per day, in line with manufacturer IFU) can inhibit viral propagation to a sufficient extent so as to reduce viral load and allow a more

rapid clearance of infection in endurance athletes. The fact that the spray is applied to the throat suggests that the oropharynx is an important area for propagation and target for treatment. This is further supported, for example, by the reduction in common cold duration that has been observed with the use of zinc gluconate lozenges ([21]). Indeed, this is in line with previous research on ColdZyme using the quarantine human viral challenge model with rhinovirus-16 ([19]). The present study, however, is the first to examine ColdZyme in free living/real-world conditions in a randomised controlled trial with athletes.

It is possible that some findings of this study may be influenced by the open label nature of the trial. For the primary outcomes (URTI reporting) previous research ([27]) has shown that the magnitude of the placebo effect has a small influence on URTI reporting (see further discussion in limitations below). For the other (secondary) parameters, such as missed or reduced training, we are not aware of any research on how the placebo effect may influence these. Ultimately, the decision to deviate from planned training is a choice made by the athlete so it is possible that the placebo effect influences this to a greater extent. However, a possible explanation for our findings for missed training days could also be that both poor IFU compliance and good IFU compliance groups reported significantly lower average daily severity ratings than the control group. Indeed, it would seem logical that symptom severity would be the most important factor influencing athletes' decisions about whether to train as normal, reduce training or miss training altogether. We also cannot exclude the possibility that those in the treatment group felt more willing to continue with training as they knew they were receiving an intervention that may help.

Limitations

The main limitation is the open label nature of this study, which presents the possibility of a placebo effect in the treatment group and/or a nocebo effect in the control group. At the time of commencing the study, the manufacturer was not able to provide an appropriate placebo. Future research would be enhanced with a full placebo so that a fully placebo-controlled, double-blind design could be implemented. Although previous research has shown that knowledge of the treatment does not influence objective biomarkers of immune function in response to exercise (e.g. immune cell functions [29, 30]), self-report results for URTI parameters are likely more prone to influence. Research on the placebo effect has shown only modest placebo effects for common cold symptoms, with an episode duration comparable to the present study ([27]). In this study they observed an effect size below the minimal threshold for a small effect (for common cold duration [27]), whereby the placebo effect influenced average reported duration by less than 0.7 days (i.e. <10%) and severity ratings by 8-17%. However, the effect was larger for individuals who had a higher belief in the possible beneficial effects (i.e. expectation) at the point of enrolment. It is

possible therefore that some individuals in the ColdZyme group had positive expectations, which could have influenced their URTI scores. Unfortunately we did not capture data on expectations in the treatment group, and this would be beneficial to include in future studies (although an appropriate placebo is preferable).

On examination of usage records and diaries from Tranche 1 (December 2017-February 2018) it became apparent that, despite being instructed to follow the manufacturer IFU (i.e. 1 dose every second hour up to 6 times daily), not all of the participants in the ColdZyme group followed the recommendations for use (~37% of recorded episodes were not treated according to the ColdZyme IFU). For Tranche 2 (December 2018-April 2019) we provided additional information and regular reminders to participants to overcome this. These procedures appear to have been effective as poor compliance with IFU was only evident in 14% of reported episodes, but compliance was still not entirely optimal (a common problem with free-living human trials). On the one hand, this is a limitation - possibly reducing the magnitude of effect in the ColdZyme group. However, on the other hand, separate analysis of the good and bad compliance episodes does provide a useful comparator condition which may overcome some of the limitations that arise in open label trials (i.e. potential placebo effects in treatment group), as can be seen in the ColdZyme Good IFU comp vs ColdZyme Poor IFU comp and ColdZyme Good IFU comp vs Control comparisons.

The use of self-report methods and illness questionnaires has the limitation of being subjective and presents the possibility of athletes reporting symptoms/self-reporting illness in the absence of a true infection. Some studies have reported an increased occurrence of allergy-type symptoms that are often mistaken by athletes as URTI, although this tends to be more common in spring (Northern hemisphere), when responses to environmental allergens such as pollen are more common ([28]). The current study was conducted in the winter months when URTI-incidence is known to be at a peak. In addition, the validated Jackson questionnaire and scoring criteria were used, which help to protect against false positive episode counts (although these can never be completely prevented). It was not feasible in the present study to confirm infection via laboratory-based diagnostic methods (e.g. polymerase chain reaction (PCR) detection of URTI-causing pathogens from throat and nasal swabs and/or bioligical fluids). It would be beneficial for future studies to include these measurements to 1) confirm the presence of URTI during self-reported episodes, and 2) monitor viral load during the course of an episode to provide insight into the mechanisms of action (for example, this would allow greater insight into the pattern of propagation, spread and infection/virus clearance rate at different sites within the URT).

Conclusions

It is clear that reporting of upper respiratory illness symptoms in athletes cluster around periods of intensive training and/or competition ([6,7]). This can have a direct and significant impact on athletes' performance in competition, preparation and training and general wellbeing ([6,8,9]). In this study we provide the first evidence from a randomised trial in athletes under free-living conditions on the effects of a proposed non-nutritional countermeasure (ColdZyme Mouth Spray) to self-reported URTI in athletes, which may have implications for athletes' absence from training and athlete performance. We show that ColdZyme mouth spray used in accordance with manufacturer instructions can reduce self-reported URTI episode duration (by 3.5 days: ~34% reduction in episode duration). This was also associated with a reduction in lost training days (~54% reduction, from 3.5 days lost per episode in Control to 1.6 days lost per episode in ColdZyme). This may provide an effective strategy to reduce the impact of upper respiratory illness on training and competition.

References

1. Pyne DB, Hopkins WG, Batterham AM, Gleeson M, Fricker PA. Characterising the individual performance responses to mild illness in international swimmers. *Br J Sports Med*. 2005;39(10):752-756. doi:10.1136/bjsm.2004.017475
2. Nieman DC. Exercise, infection, and immunity. *Int J Sports Med*. 1994;15 Suppl 3:S131-41. doi:10.1055/s-2007-1021128
3. Walsh NP, Gleeson M, Shephard RJ, et al. Position statement. Part one: Immune function and exercise. *Exerc Immunol Rev*. 2011;17:6-63. http://www.ncbi.nlm.nih.gov/pubmed/21446352.
4. Bayer C, Remschmidt C, an der Heiden M, et al. Internet-based syndromic monitoring of acute respiratory illness in the general population of Germany, weeks 35/2011 to 34/2012. *Euro Surveill*. 2014;19(4). doi:10.2807/1560-7917.es2014.19.4.20684
5. Fendrick AM, Monto AS, Nightengale B, Sarnes M. The economic burden of non-influenza-related viral respiratory tract infection in the United States. *Arch Intern Med*. 2003;163(4):487-494. doi:10.1001/archinte.163.4.487
6. Svendsen IS, Taylor IM, Tønnessen E, Bahr R, Gleeson M. Training-related and competition-related risk factors for respiratory tract and gastrointestinal infections in elite cross-country skiers. *Br J Sports Med*. 2016;50(13):809-815. doi:10.1136/bjsports-2015-095398
7. Hellard P, Avalos M, Guimaraes F, Toussaint J-F, Pyne DB. Training-related risk of common illnesses in elite swimmers over a 4-yr period. *Med Sci Sports Exerc*. 2015;47(4):698-707. doi:10.1249/MSS.0000000000000461
8. Cunniffe B, Griffiths H, Proctor W, Davies B, Baker JS, Jones KP. Mucosal immunity and illness incidence in elite rugby union players across a season. *Med Sci Sports Exerc*. 2011;43(3):388-397. doi:10.1249/MSS.0b013e3181ef9d6b
9. Reid VL, Gleeson M, Williams N, Clancy RL. Clinical investigation of athletes with persistent fatigue and/or recurrent infections. *Br J Sports Med*. 2004;38(1):42-45. doi:10.1136/bjsm.2002.002634
10. Raysmith BP, Drew MK. Performance success or failure is influenced by weeks lost to injury and illness in elite Australian track and field athletes: A 5-year prospective study. *J Sci Med Sport*. 2016;19(10):778-783. doi:10.1016/j.jsams.2015.12.515
11. Keaney LC, Kilding AE, Merien F, Dulson DK. Keeping Athletes Healthy at the 2020 Tokyo Summer Games: Considerations and Illness Prevention Strategies. *Front Physiol*. 2019;10. doi:10.3389/fphys.2019.00426
12. Walsh NP. Recommendations to maintain immune health in athletes. *Eur J Sport Sci*. 2018;18(6):820-831. doi:10.1080/17461391.2018.1449895
13. Williams NC, Killer SC, Svendsen IS, Jones AW. Immune nutrition and exercise: Narrative review and practical recommendations. *Eur J Sport Sci*. 2019;19(1):49-61. doi:10.1080/17461391.2018.1490458
14. Walsh NP, Gleeson M, Pyne DB, et al. Position statement. Part two: Maintaining immune health. *Exerc Immunol Rev*. 2011;17:64-103.
15. Davison G, Kehaya C, Jones AW. Nutritional and Physical Activity Interventions to Improve Immunity. *Am J Lifestyle Med*. 2016;10(3):152-169. doi:10.1177/1559827614557773
16. Heikkinen T, Järvinen A. The common cold. *Lancet (London, England)*. 2003;361(9351):51-59. doi:10.1016/S0140-6736(03)12162-9
17. Winther B, Gwaltney JM, Mygind N, Turner RB, Hendley JO. Sites of rhinovirus recovery after point inoculation of the upper airway. *JAMA*. 1986;256(13):1763-1767. http://www.ncbi.nlm.nih.gov/pubmed/3018306.
18. Witek TJ, Ramsey DL, Carr AN, Riker DK. The natural history of community-acquired common colds symptoms assessed over 4-years. *Rhinology*. 2015;53(1):81-88. doi:10.4193/Rhin14.149
19. Clarsund M, Fornbacke M, Uller L, Johnston SL, Emanuelsson CA. A A Randomized,

Double-Blind, Placebo-Controlled Pilot Clinical Study on ColdZyme® Mouth Spray against Rhinovirus-Induced Common Cold. *Open J Respir Dis*. 2017;07(04):125-135. doi:10.4236/ojrd.2017.74013

20. Clarsund M, Blom U, Gardulf A. Evaluation of ColdZyme® Mouth Spray on prevention of upper respiratory tract infections in a boy with primary immunodeficiency: a case report. *J Med Case Rep*. 2016;10(1):302. doi:10.1186/s13256-016-1085-2
21. Eby GA, Davis DR, Halcomb WW. Reduction in duration of common colds by zinc gluconate lozenges in a double-blind study. *Antimicrob Agents Chemother*. 1984;25(1):20-24. doi:10.1128/aac.25.1.20
22. Stefansson B, Gudmundsdottir Á, Clarsund M. A medical device forming a protective barrier that deactivates four major common cold viruses. *Virol Res Rev*. 2017;1(5). doi:10.15761/VRR.1000130
23. Clarsund M. Evaluation of ColdZyme Mouth Spray for the Protection against Common Cold in Elite Athletes to Reduce Unwanted Absence from Training and Competition. *Open J Respir Dis*. 2017;07(03):103-109. doi:10.4236/ojrd.2017.73010
24. Jackson GG, Dowling HF, Spiesman IG, Boand A V. Transmission of the common cold to volunteers under controlled conditions. I. The common cold as a clinical entity. *AMA Arch Intern Med*. 1958;101(2):267-278. doi:10.1001/archinte.1958.00260140099015
25. Foster C, Florhaug JA, Franklin J, et al. A new approach to monitoring exercise training. *J strength Cond Res*. 2001;15(1):109-115. http://www.ncbi.nlm.nih.gov/pubmed/11708692.
26. Martineau AR, Hanifa Y, Witt KD, et al. Double-blind randomised controlled trial of vitamin D3 supplementation for the prevention of acute respiratory infection in older adults and their carers (ViDiFlu). *Thorax*. 2015;70(10):953-960. doi:10.1136/thoraxjnl-2015-206996
27. Barrett B, Brown R, Rakel D, et al. Placebo Effects and the Common Cold: A Randomized Controlled Trial. *Ann Fam Med*. 2011;9(4):312-322. doi:10.1370/afm.1250
28. Robson-Ansley P, Howatson G, Tallent J, et al. Prevalence of allergy and upper respiratory tract symptoms in runners of the London marathon. *Med Sci Sports Exerc*. 2012;44(6):999-1004. doi:10.1249/MSS.0b013e318243253d
29. McFarlin BK, Hutchison AT, Kueht ML. Knowledge of carbohydrate consumption does not alter natural killer cell activity following an acute bout of high-intensity aerobic exercise. *Appl Physiol Nutr Metab*. 2008;33(5):1007-12. doi: 10.1139/H08-076.
30. Navalta JW, McFarlin BK, Lyons S, Arnett SW, Schafer MA. Cognitive awareness of carbohydrate intake does not alter exercise-induced lymphocyte apoptosis. *Clinics (Sao Paulo)*. 2011;66(2):197-202.

Table headings

Table 1: Overview of self-report URTI data

Table 2: Days that training was affected by URTI episodes/symptoms

Figure titles and captions

Figure 1. Pattern of training load change across study period (% of healthy average).

Figure 2. Pattern of training load in 2 weeks before URTI onset, during URTI and 3 weeks after URTI subsided.

*Significantly different to 'healthy' weeks (** $P < 0.01$)*

Table 1: Overview of self-report URTI data

	Control	ColdZyme (overall)	ColdZyme (Poor IFU comp)	ColdZyme (Good IFU comp)
URTI episodes (per person)	1.1 ± 0.9	1.0 ± 0.8	*#n/a*	*#n/a*
URTI episode duration (d)	10.4 ± 8.5	*7.7 ± 4.0	9.3 ± 4.5	**†6.9 ± 3.5
Jackson Symptom Score (episode total)	74.9 ± 72.0	**43.6 ± 30.1	52.6 ± 35.7	**40.0 ± 26.5
Jackson Symptom Score (daily episode average)	6.9 ± 2.8	**5.5 ± 2.4	*5.3 ± 1.7	*5.6 ± 2.7

NOTE: #n/a = Not relevant or calculated since compliance can only be analysed when an episode exists
ColdZyme Poor IFU comp = Poor compliance with ColdZyme IFU (e.g. less than 4 doses per day);
ColdZyme Good IFU comp = Good compliance with ColdZyme IFU.
*Significantly different to Control: * P < 0.05; ** P < 0.01*
Significant difference between poor and good IFU compliance groups † P < 0.05; ‡ P < 0.01

Table 2: Days that training was affected by URTI episodes/symptoms

	Control	ColdZyme (overall)	ColdZyme (poor IFU comp)	ColdZyme (good IFU comp)
Days missed	3.5 ± 5.0	*1.6 ± 2.5	*1.6 ± 2.9	*1.6 ± 2.3
Days reduced training	3.4 ± 5.1	3.0 ± 3.4	3.1 ± 3.1	2.9 ± 3.7

Average days missed/reduced per URTI episode
ColdZyme Poor IFU comp = Poor compliance with ColdZyme IFU (e.g. less than 4 doses per day);
ColdZyme Good IFU comp = Good compliance with ColdZyme IFU.
*Significantly different to Control: * P < 0.05*

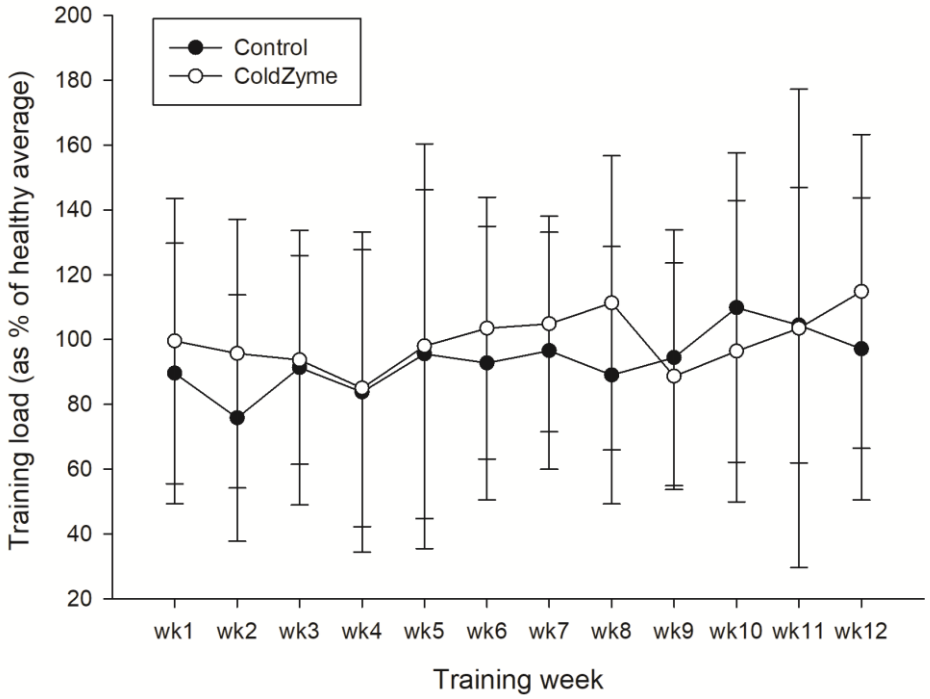

Figure 1

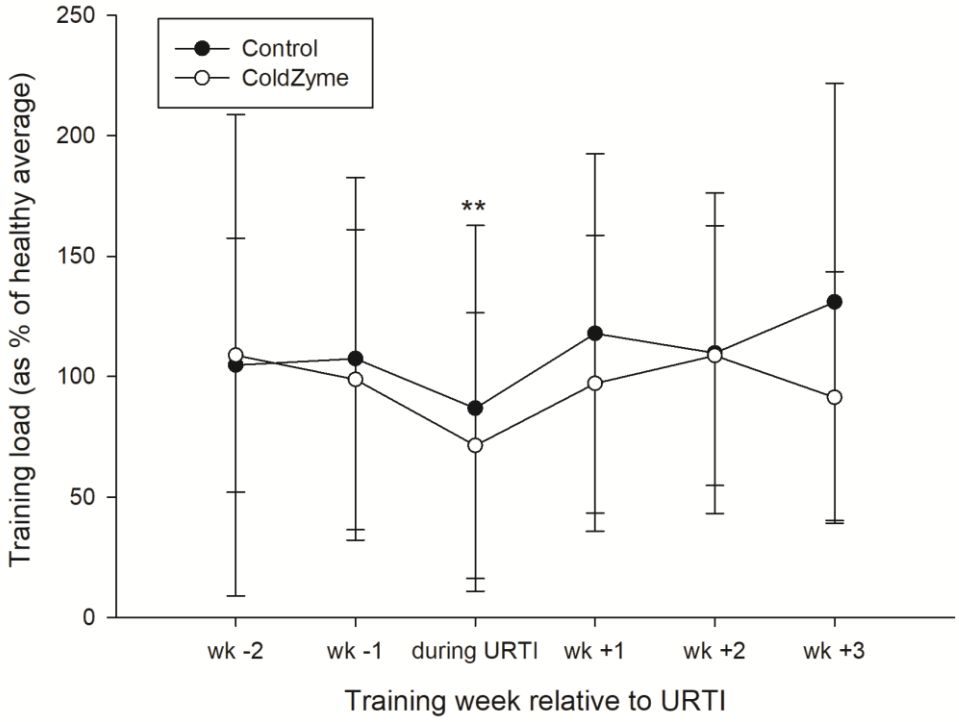

Figure 2