ISRCTN13055033 https://doi.org/10.1186/ISRCTN13055033

Assessing the Impact on dental biofilm of Heyndrickxia coagulans administered through sugar-free chewing gum

Submission date 03/11/2025	**Recruitment status** No longer recruiting	[_] Prospectively registered [_] Protocol
Registration date 04/12/2025	**Overall study status** Completed	[_] Statistical analysis plan [_] Results
Last Edited 05/11/2025	**Condition category** Oral Health	[_] Individual participant data [X] Record updated in last year

Plain English summary of protocol

Background and study aims
This study is looking at whether a probiotic called Heyndrickxia coagulans (H. coagulans), delivered through sugar-free chewing gum, can settle in dental plaque and possibly improve oral health. Researchers want to understand how this probiotic affects the mix of bacteria in dental plaque.

Who can participate?
Healthy adults aged 18 to 64 years can take part, as long as they have at least 24 natural teeth (excluding wisdom teeth), good gum and plaque scores, and a normal saliva flow. People with certain health conditions, pregnant or breastfeeding women, smokers, those with braces, or anyone allergic to ingredients in the gum cannot take part.

What does the study involve?
Participants will be randomly assigned to one of two groups: one will chew gum containing the probiotic, and the other will chew gum without it. Everyone will receive a toothbrush and fluoride toothpaste and be asked to follow specific oral hygiene instructions. They'll chew the gum five times a day for four weeks, after a two-week preparation period. There will also be a one-week follow-up phase. Participants will keep a diary of their gum use and bring back empty packs to help researchers track compliance. Dental plaque samples will be collected and analysed to see how the bacteria change over time.

What are the possible benefits and risks of participating?
The study may help improve understanding of how probiotics affect oral health, which could benefit future dental care. Risks are minimal but may include mild reactions to ingredients in the gum. Anyone who experiences side effects or needs to take antibiotics or other restricted products during the study will be withdrawn.

Where is the study run from?
University of Milan, Italy.

When is the study starting and how long is it expected to run for?
January 2024 to April 2025

Who is funding the study?
Investigator initiated and funded

Who is the main contact?
Dr Silvia Cirio, silvia.cirio@unimi.it

Contact information

Type(s)
Public, Scientific, Principal investigator

Contact name
Dr Silvia Cirio

ORCID ID
https://orcid.org/0000-0002-3531-2595

Contact details
Via Beldiletto, 1
Milan
Italy
20121
+39 3478436387
silvia.cirio@unimi.it

Additional identifiers

Clinical Trials Information System (CTIS)
Nil known

ClinicalTrials.gov (NCT)
Nil known

Protocol serial number
Nil known

Study information

Scientific Title
Assessing the Impact on dental biofilm of Heyndrickxia coagulans administered through sugar-free chewing gum: a double-blind randomized controlled trial

Study objectives
The present randomized controlled trial aims to evaluate the ability of the probiotic H. coagulans, administered through sugar-free chewing gum, to modified dental biofilm

Ethics approval required
Ethics approval required

Ethics approval(s)
approved 13/02/2024, Ethics Committee of the University of Milan (Via Festa del Perdono, 7, Milan, 20121, Italy; +39 2 5032 5032; comitato.etico@unimi.it), ref: 24/24

Study design
Double blind interventional randomized controlled trial

Primary study design
Interventional

Study type(s)
Other

Health condition(s) or problem(s) studied
Change in dental plaque microflora after administration of probiotics in healthy adult patients

Interventions
Participants will be randomized into two groups using a computer-generated randomization system. Both participants and investigators will be blinded to group allocation. All study participants will give their written consent to participate.

The study will last a total of seven weeks. All enrolled subjects will receive instructions for at-home oral hygiene, along with a manual toothbrush and a fluoride toothpaste (1450 ppm F) to be used throughout the study period. Participants will be instructed not to use any mouthwash or other oral hygiene products aside from those provided during the experimental phase. Additionally, subjects will be asked to refrain from using antibacterial or antibiotic medications (either topical or systemic), from taking probiotics, and from consuming chewing gum or other products containing xylitol. Should the use of any of these products become necessary, participants will be required to notify the investigators and will subsequently be excluded from the study sample.

Subjects in the intervention arm will be administered a chewing gum containing 5×10^8 CFU of Heyndrickxia coagulans SNZ1969®; subjects in the control arm will receive a placebo chewing gum, identical in appearance and composition to that of the intervention group but without the probiotic.

The experimental period will be structured as follows: an initial two-week washout phase, followed by a four-week Intervention phase, and concluding with a one-week post-Intervention phase. During the four-week Intervention, participants will be instructed to consume the assigned chewing gum five times per day (after breakfast, mid-morning, after lunch, mid-afternoon, and in the evening after dinner), at least 30 minutes after brushing their teeth.

Follow-up assessments will be conducted at the following time points: after the initial two-week washout period (T0), after two weeks of chewing gum use (T1), after four weeks of chewing gum use (T2), and at the end of the post-Intervention period (T3).

Intervention Type
Supplement

Primary outcome(s)
The composition of the dental plaque microbial ecosystem is measured using metataxonomic analysis of 16S rRNA gene sequencing at T0, T2, and T3

Key secondary outcome(s))
1. Presence and quantity of *H. coagulans* SNZ1969® in dental plaque is measured using strain-specific qPCR at T0, T1, T2, and T3 in the intervention arm, and at T0 only in the control arm
2. Volatile sulphur compounds (VSCs) in exhaled breath are measured using a halimeter (OralChroma™) at T0, T1, T2, and T3
3. Halitosis status is measured using threshold values of $H_2S \geq 112$ ppb and $CH_3SH \geq 26$ ppb from halimeter readings at T0, T1, T2, and T3
4. Alpha diversity of the dental plaque microbiota is measured using observed features, Faith's phylogenetic diversity, Pielou's evenness, and Shannon entropy from QIIME 2™ analysis at T0, T2, and T3
5. Beta diversity of the dental plaque microbiota is measured using weighted UniFrac, unweighted UniFrac, Jaccard, and Bray–Curtis dissimilarity metrics from QIIME 2™ analysis at T0, T2, and T3
6. Differential abundance of bacterial taxa in dental plaque is measured using DESeq2 analysis of metataxonomic data at T0, T2, and T3

Completion date
10/04/2025

Eligibility

Key inclusion criteria
1. Adult subjects aged 18 to 64 years
2. At least 24 natural teeth (excluding third molars)
3. Gingival index and plaque index scores ≤ 2
4. Stimulated salivary flow rate between 1.5 and 2.0 mL/min.

Participant type(s)
Healthy volunteer

Healthy volunteers allowed
No

Age group
Adult

Lower age limit
18 years

Upper age limit
64 years

Sex
All

Total final enrolment

Key exclusion criteria
1. Presence of systemic diseases
2. Pregnancy or lactation
3. History of drug abuse
4. Smoking habits
5. Use of fixed orthodontic appliances
6. Allergies to any ingredients in the chewing gums used

Date of first enrolment
01/09/2024

Date of final enrolment
30/09/2024

Locations

Countries of recruitment
Italy

Study participating centre
Department of Biomedical, Surgical and Dental Sciences, University of Milan
Via Beldiletto, 1
Milan
Italy
20121

Study participating centre
Perfetti Van Melle spa
Via Angelo Clerici 30
Lainate (MI)
Italy
20045

Sponsor information

Organisation
University of Milan

ROR
https://ror.org/00wjc7c48

Funder(s)

Funder type
Other

Funder Name
Investigator initiated and funded

Results and Publications

Individual participant data (IPD) sharing plan
The datasets generated during and/or analysed during the current study will be stored in a publicly available repository
https://doi.org/10.13130/RD_UNIMI/VATL7W (link to be activated once data is added)

IPD sharing plan summary
Stored in publicly available repository

Study outputs

Output type	Details	Date created	Date added	Peer reviewed?	Patient-facing?
Participant information sheet	First participant info sheet. In Italian		05/11/2025	No	Yes
Participant information sheet	Second participant info sheet. In Italian		05/11/2025	No	Yes
Participant information sheet	Participant information sheet	11/11/2025	11/11/2025	No	Yes