logic halitosis, and pathologic halitosis is subclassified as an oral or nonoral pathologic halitosis. The treatment of physiologic halitosis primarily involves dental and oral care, oral hygiene instruction, and counseling. Oral pathologic halitosis is caused largely by periodontal disease, and its treatment requires periodontal treatment in addition to the measures used to treat physiologic halitosis. The goal of this treatment regime is to acquire healthy oral condition, including a normal microflora.

The aim of this open-label pilot study was to evaluate whether the oral administration of probiotic *L. salivarius* WB21 could reduce oral malodor in patients with actual malodor complaining of halitosis and, subsequently, to improve the oral conditions associated with oral malodor.

MATERIALS AND METHODS

Probiotic product

The MINNA NO ZENDAMAKIN WB21 TABLET (Wakamoto Pharmaceutical, Tokyo, Japan) contains 6.7×10^8 colony-forming units (CFU) of *L. salivarius* WB21 and 280 mg of xylitol per tablet. Strain WB21 is an acid-tolerant lactobacillus that was isolated from *L. salivarius* WB1004.[19] The dosage throughout the test period was maintained at 3 tablets per day, taken orally after eating.

Subjects and study design

The study population consisted of 20 genuine halitosis subjects (6 males, 14 females; mean age 50.9 ± 12.1 years, range 30-66 years) who complained of halitosis and presented to the Oral Malodor Clinic of Fukuoka Dental College Medical and Dental Hospital, Japan, between July 2008 and August 2009. None of these patients had received antibiotics within 3 months before participating in the study. All of the subjects who participated understood the nature of the research project and provided informed consent. Permission for this study was obtained from the Ethics Committee for Clinical Research of Fukuoka Dental College and Fukuoka College of Health Sciences (approval number 125).

The subjects took 3 tablets of MINNA NO ZENDAMAKIN WB21 TABLET every day, containing a total of 2.01×10^9 CFU *L. salivarius* WB21 and 840 mg xylitol. They were directed to place a tablet in the mouth for a few minutes and allow it dissolve. They were also instructed not to change their oral hygiene regimens and not to take other probiotic products throughout the study period. Neither professional prophylaxis nor tooth-brushing instruction was performed during or before the experimental period. Maintenance of this regime was confirmed at days 15 and 29 of the study. Malodor was assessed, clinical parameters recorded, and saliva samples obtained from all subjects on days 1, 15, and 29 from 10 subjects with oral pathologic halitosis.

Malodor assessment

For each patient, malodor was assessed and a clinical examination performed at the same time of day at least 5 hours after eating, drinking, chewing, smoking, and brushing or rinsing the mouth. The severity of oral malodor in each individual was determined using an organoleptic test (OLT) and gas chromatography (model GC14B; Shimadzu Works, Kyoto, Japan). For the OLT, each patient was instructed to exhale through the mouth with moderate force into a Teflon sampling bag (GL Science, Tokyo, Japan) for 2 to 3 seconds to prevent the dilution of mouth odor with lung and room air. This procedure was repeated until approximately 1 L of breath sample was obtained. Two of the 3 evaluators (with training and experience in calibration tests) then estimated the odor at a distance of 10 cm from the sampling bag. The OLT scores were estimated on a scale of 0 to 5 (0, no odor; 1, questionable odor; 2, slight malodor; 3, moderate malodor; 4, strong malodor; 5, severe malodor),[18] and mean scores given by the different judges were used. The percentage agreement in the OLT scores among the 3 evaluators always exceeded 75.0% ($\kappa = 0.50$). The threshold level for genuine halitosis was defined as an OLT score of 2 or higher according to the experimental criteria.[18] For the gas chromatographic measurements, the subjects were asked to remain quiet with a closed mouth for 30 seconds, after which mouth air (10 mL) was aspirated using a gas-tight syringe. These samples were injected onto a gas chromatograph column at 70°C. A glass column was packed with 25% β, β 9-oxydipropionitrile on a 60-

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