INTRODUCTION

The prevalence of dysphonia is approximately 6% among children aged eight years (1). Dysphonia develops mainly in children aged 7-12 years, predominantly boys, and vocal nodules are the most common cause of dysphonia in childhood (2). Vocal abuse and incorrect use of the voice are the main predisposing factors for vocal nodules (3). Vocal nodules are diagnosed using video-stroboscopic examinations in adult patients. However, pediatric patients hardly accept the examination because of their lack of collaboration and patience (4), which prevents regular follow-up of patients' laryngeal findings. For the assessment of voice disorders in children, multi-dimensional assessments, including perceptual and objective voice evaluations, have been recommended by the European Laryngological Society (5). In addition, a minimum protocol for the pediatric voice clinic recommended measuring acoustic parameters of voice in pediatric patients with dysphonia (4). Recently, the objective and quantitative acoustic analysis of voice, such as pitch period perturbation quotient (PPQ), amplitude perturbation quotient (APQ), and noise-to-harmonic ratio (NHR), were measured. Results : In phonation at a loudness of over 80 dBA, the PPQ, APQ, and NHR values of the voice significantly increased in children with vocal nodules than in the control children without dysphonia. The sensitivities and specificities of PPQ, APQ, and NHR for prediction of vocal nodules in children were 62.86% and 84.38%, 74.29% and 75.00%, and 31.43% and 93.75%, respectively. Discussion : The present findings suggest that vocal nodules affect vocal fold vibration, resulting in impaired control of pitch and loudness leading to increased noise components. NHR could be used to evaluate the efficacy of treatment, such as voice rehabilitation, in pediatric patients with vocal nodules because of its high specificity for prediction of vocal nodules in children.
a loudness of over 80 dBA, confirmed using a sound-level meter (Audio Analyzer PAA3, Phonic, Japan) and at a comfortable loudness with an instruction of conversational pitch and loudness. The sound pressure level of the recorded voice was calibrated by recording of a pure tone of 62 dBA at 1 kHz generated by a sound calibrator (NC-74, Rion, Tokyo, Japan). Brockmann-Bauser, et al. reported the voice SPL in phonation at a comfortable loudness ranged from 62.7 to 93.8 dBA in children (10).

Experiment 2
Participants
In Experiment 2, 35 pediatric patients with vocal nodules (31 boys and 4 girls, mean age: 7.46 ± 1.46 years) and 32 control children with no vocal symptoms or history of phoniatric disorders (24 boys and 8 girls, mean age: 8.53 ± 1.61 years) were enrolled. Vocal nodules in these pediatric patients were diagnosed by otolaryngologists using video-laryngoscopic examination. Stroboscopic illumination was not applied because all patients could not sustain the vowels in a relaxed manner during the examination. The voice quality of the control children was evaluated as euphonic by otolaryngologists in auditory-perceptual evaluation. There was no sex ratio difference between pediatric patients with vocal nodules and control children (p = 0.15). This study was approved by the Committee for Medical Ethics of Tokushima University Hospital. Written informed consent was obtained from a parent of each pediatric patient with vocal nodules and control child prior to the experiment.

Recordings
The parameters of the voice recordings were similar to those in Experiment 1. Three samples of sustained vowel /e/ recordings for 3 seconds were obtained for all the participants under the instruction to phonate at a loudness of over 80 dBA.

Acoustic analysis
All recordings were analyzed using the Multi-Dimensional Voice Program (MDVP, Kay Pentax, USA) (11) in both experiments. A second mid-vowel portion of each sample without the onset and offset of phonation was prepared for analysis. The acoustic parameters of PPQ, APQ, and NHR were measured in three samples from each child, and the average value was used for statistical analyses.

Statistical analysis
The F test and paired t-test were used in Experiment 1, and Chi-square for independence test, Student’s t-test and ROC analysis were used in Experiment 2 for statistical analysis using Statcel version 4 (OMS Publishing Inc., Saitama, Japan) and statistical software EZR (12). p < 0.05 was considered statistically significant. The optimum cut-off values were defined using the points closest to (0, 1) on the ROC curve.

RESULTS
Experiment 1
The variances of PPQ and NHR, but not APQ, in phonation at a loudness of over 80 dBA were significantly lower than those in phonation at a comfortable loudness in children without dysphonia (p < 0.01) (Table 1). The mean values of PPQ, APQ, and NHR in phonation at a loudness of over 80 dBA were significantly lower than those in phonation at a comfortable loudness in children without dysphonia (p < 0.01) (Table 1).
suggest that vocal nodules on bilateral vocal folds affect the control of pitch and intensity of the voice. It was reported that the value of PPQ of the voice was affected by the width of the vocal nodules in pediatric patients (15) and that vocal polyp, a unilateral vocal mass lesion, had greater effect on vocal vibration to induce higher elevations of jitter and shimmer (similar to PPQ and APQ, respectively) than bilateral vocal nodules (16). Taken together with the present findings, it is suggested that vocal nodules, as a mass lesion on bilateral vocal folds, affect the vibration of vocal folds, resulting in impaired control of pitch and loudness of the voice. Gramuglia et al. also reported that children with vocal nodules showed elevations in PPQ and APQ of voice (17).

The elevation in the value of NHR of the voice in children with vocal nodules in the present study is consistent with previous reports (17, 18). Because NHR is an index of the noise component, the findings suggest that vocal nodules on bilateral vocal folds produce a noise component, an essential element of dysphonia. It has been reported that the value of HNR (a semantic inverse of NHR) of voice is affected by the mediolateral width of the vocal nodules in children (15). Therefore, the increased NHR in the present study suggests that glottal insufficiency due to protruded vocal nodules exacerbates noise components in voice signals.

Although the video-stroboscopic examination is considered the best modality for diagnosing and assessing vocal nodules, it is difficult for pediatric patients to cooperate with an uncomfortable examination (4). Therefore, there is an essential need for quick, non-invasive, and well-tolerated methods, such as computer-assisted voice analysis, for pediatric patients. In general, the clinical evaluation of voice disorders consists of diagnosis and monitoring steps. It would be ideal for both steps to be examined using a non-invasive modality. For this purpose, the degree of sensitivity of the examination is crucial for the diagnosis. In the present study, sensitivities and specificities of the three acoustic parameters were evaluated to predict vocal nodules in children. Among the three vocal parameters, sensitivity was highest in APQ. However, its value of 74.28% is too low for screening and diagnosing the presence of vocal nodules in children. Therefore, acoustic analysis seemed not to be suitable for the diagnosis of pediatric voice disorders. That is, an invasive method of video-stroboscopy/laryngoscopy is still necessary for diagnosis. Because the highest specificity of 93.75% in NHR demonstrates its ability to designate an individual who does not have the disease, it is suggested that NHR can monitor the efficacy of treatment, such as voice rehabilitation, in pediatric patients with dysphonia. Moreover, it was reported that jitter (similar to PPQ), shimmer (similar to APQ), and NHR improved after voice therapy in children with vocal nodules (19, 20). Therefore, regular follow-up using NHR combined with PPQ and APQ could be acceptable for monitoring pediatric patients with vocal fold nodules if an occasional video-stroboscopy/laryngoscopy is performed.

**CONCLUSION**

Because the variances of PPQ and NHR, but not APQ in phonation at a loudness of over 80 dBA were smaller than those in phonation at a comfortable loudness in children, we used phonation at a predefined loudness of over 80 dBA in acoustic analysis of voice using MDVP in pediatric patients with vocal nodules. Of the three acoustic parameters, NHR could be used to evaluate the efficacy of voice rehabilitation in pediatric patients with vocal nodules because of its high specificity for prediction of vocal nodules in children.
CONFLICT OF INTEREST

The authors declare no conflicts of interest in this study.

REFERENCES

1. Carding PN, Roulstone S, Northstone K, ALSPAC Study Team: The prevalence of childhood dysphonia: a cross-sectional study. J Voice 20: 623-630, 2006
2. Tavares EL, Bruscolotto A, Santana MF, Padovan CA, Martins RH: Epidemiological study of dysphonia in 4-12 year-old children. Braz J Otorhinolaryngol 77: 736-446, 2011
3. Martins RH, Hidalgo Ribeiro CB, Fernandes de Mello BM, Branco A, Tavares EL: Dysphonia in children. J Voice 26: 674.e17-20, 2012
4. Cohen W, Wynne DM, Kubba H, McCartney E: Development of a minimum protocol for assessment in the paediatric voice clinic. Part 1: Evaluating vocal function. Logoped Phoniatr Vocol 37: 33-38, 2012
5. Dejonckere PH, Bradley P, Clemente P, Cornut G, Crevier-Buchman L, Friedrich G, Van De Heyning P, Remacle M, Woisard V, Committee on Phoniatrics of the European Laryngological Society: A basic protocol for functional assessment of voice pathology, especially for investigating the efficacy of (phonosurgical) treatments and evaluating new assessment techniques. Guideline elaborated by the Committee on Phoniatrics of the European Laryngological society (ELS). Eur Arch Otorhinolaryngol 258: 77-82, 2001
6. Bhuta T, Patrick L, Garnett JD: Perceptual evaluation of voice quality and its correlation with acoustic measurements. J Voice 18: 299-304, 2004
7. Glaze LE, Bless DM, Susser RD: Acoustic analysis of vowel and loudness differences in children’s voice. J Voice 4: 37-44, 1990
8. Kakei Y, Takahashi H, Calcatera TC: Acoustic measures for detecting laryngeal pathology. Acta Otolaryngol 84: 105-117, 1977
9. Yamoto E, Gould WJ, Baer T: Harmonics-to-noise ratio as an index of the degree of hoarseness. J Acoust Soc Am 71: 1544-1549, 1982
10. Brockmann-Bauser M, Beyer D, Bohlender JE: Clinical relevance of speaking voice intensity effects on acoustic jitter and shimmer in children between 5;0 and 9;11 years. Int J Pediatr Otorhinolaryngol 78: 2121-2126, 2014
11. Smits I, Ceuppens P, De Bodt MS: A comparative study of acoustic voice measurements by means of Dr. Speech and Computerized Speech Lab. J Voice 19: 187-196, 2005
12. Kanda Y: Investigation of the freely available easy-to-use software ‘EZR’ for medical statistics. Bone Marrow Transplantation 48: 452-458, 2013
13. Brockmann M, Drinnan MJ, Storck C, Carding PN: Reliable jitter and shimmer measurements in voice clinics: the relevance of vowel, gender, vocal intensity, and fundamental frequency effects in a typical clinical task. J Voice 25: 44-53, 2011
14. Nardone HC, Recko T, Huang J, Roger C, Nuss RC: A retrospective review of the progression of pediatric vocal fold nodules. JAMA Otolaryngol Head Neck Surg 140: 233-236, 2014
15. Bilal N, Selcuk T, Sarica S, Alkan A, Orhan I, Doganer A, Sagiroglu S, Kilic MA: Voice Acoustic Analysis of Pediatric Vocal Nodule Patients Using Ratios Calculated With Biomedical Image Segmentation. J Voice 33: 195-203, 2019
16. Jiang JJ, Zhang Y, MacCallum J, Sprecher A, Zhou L: Objective acoustic analysis of pathological voices from patients with vocal nodules and polyps. Folia Phoniatr Logop 61: 342-349, 2009
17. Gramuglia AC, Tavares EL, Rodrigues SA, Martins RH: Perceptual and acoustic parameters of vocal nodules in children. Int J Pediatr Otorhinolaryngol 78: 312-316, 2014
18. Niedzielska G, Glijer E, Niedzielski A: Acoustic analysis of voice in children with noduli vocales. Int J Pediatr Otorhinolaryngol 60: 119-122, 2001
19. Tezcaner CZ, Ozturksoy SK, Sati I, Dursun G: Changes after voice therapy in objective and subjective voice measurements of pediatric patients with vocal nodules. Eur Arch Otorhinolaryngol 266: 1923-1927, 2009
20. Valadez V, Ysunza A, Ocharan-Hernandez E, Garrido-Bustamante N, Sanchez-Valerio A, Pamplona MC: Voice parameters and videoendolaryngoscopy in children with vocal nodules: a longitudinal study, before and after voice therapy. Int J Pediatr Otorhinolaryngol 76: 1361-1365, 2012