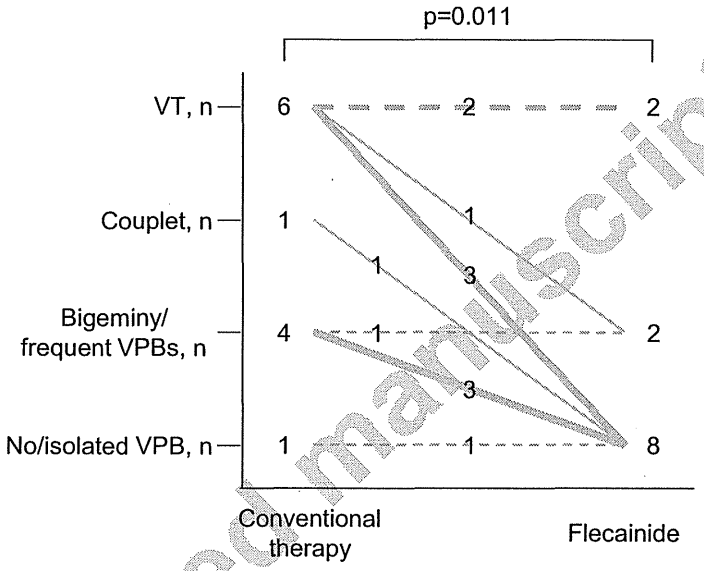

Figure. Effects of flecainide on exercise-induced ventricular arrhythmias. Ventricular arrhythmias during exercise testing were compared between conventional therapy and flecainide in genotype-negative patients. Green line indicates suppression of ventricular arrhythmias by flecainide; blue line, no change. VT denotes ventricular tachycardia; VPB, ventricular premature beat.

Heart Vessels
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CASE REPORT

Improvement in non-tachycardia-induced cardiac failure after radiofrequency catheter ablation in a child with a right-sided accessory pathway

Hideo Fukunaga · Katsumi Akimoto · Takeshi Furukawa · Ken Takahashi · Masahiko Kishiro · Toshiaki Shimizu · Hiroshi Kamiyama · Naokata Sumitomo

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Abstract A 6-year-old boy was referred for an evaluation of intolerance to physical activity at his elementary school. The patient had no episodes of palpitations. He was diagnosed as Wolff-Parkinson-White syndrome with a right-sided accessory pathway (AP) and dilated cardiomyopathy (DCM). Ventricular dyskinesis was detected mostly in the ventricular septum. Because the asynchronous septal motion caused by pre-excitation through a right-sided AP might deteriorate his cardiac function, he underwent an AP ablation, after which the asynchronous ventricular wall motion disappeared and the wall thickness improved. We suggest that an AP ablation may be the treatment of first priority in patients who have DCM-like dyskinesis even without sustained tachyarrhythmias.

Keywords Child · Wolff-Parkinson-White syndrome · Dilated cardiomyopathy · Accessory pathway · Dyskinesis

Introduction

Atrioventricular re-entrant tachycardia (AVRT) using an accessory pathway (AP) is the most common supraventricular tachycardia (SVT) in children. However, in patients with a manifest AP (Wolff-Parkinson-White (WPW) syndrome), the ventricles are electrically and mechanically pre-excited through an AP, which directly connects the atria and ventricles. This may cause eccentric ventricular activation via the AP and normal conduction system, resulting in an asynchronous ventricular depolarization, and cardiac dysfunction in some patients. The cardiac function in patients with a manifest AP markedly depends on the degree and site of the pre-excitation [1–3]. Recently, some patients, either children or adults, with overt ventricular pre-excitation have been reported to develop left ventricular (LV) dysfunction and dilated cardiomyopathy (DCM) [4–6]. In this report, we present the case of a male patient with cardiac failure caused by a right-sided AP with no documented episodes of SVT, who completely recovered from his cardiac failure after radiofrequency catheter ablation (RFCA).

Case report

A 6-year-old boy was referred for an evaluation of intolerance to physical activity at his elementary school. He had had no prior episodes of palpitations, presyncope, or syncope. He had no family history of sudden cardiac death, arrhythmias, or cardiomyopathy. His electrocardiogram revealed a right-sided AP (Fig. 1a). Although he had no SVT or heart failure symptoms, we performed a careful follow-up. The left ventricular ejection fraction (LVEF) on echocardiography was within the normal range during the first medical examination, but gradually deteriorated over 5 years. Mild mitral valve regurgitation, thinning of the basal segments of the interventricular septum, a ventricular aneurysm with dyskinetic motion during systole, a reduced LVEF of 45 % with Simpson rule, and left ventricular dilation (Fig. 2a) were detected by an echocardiogram

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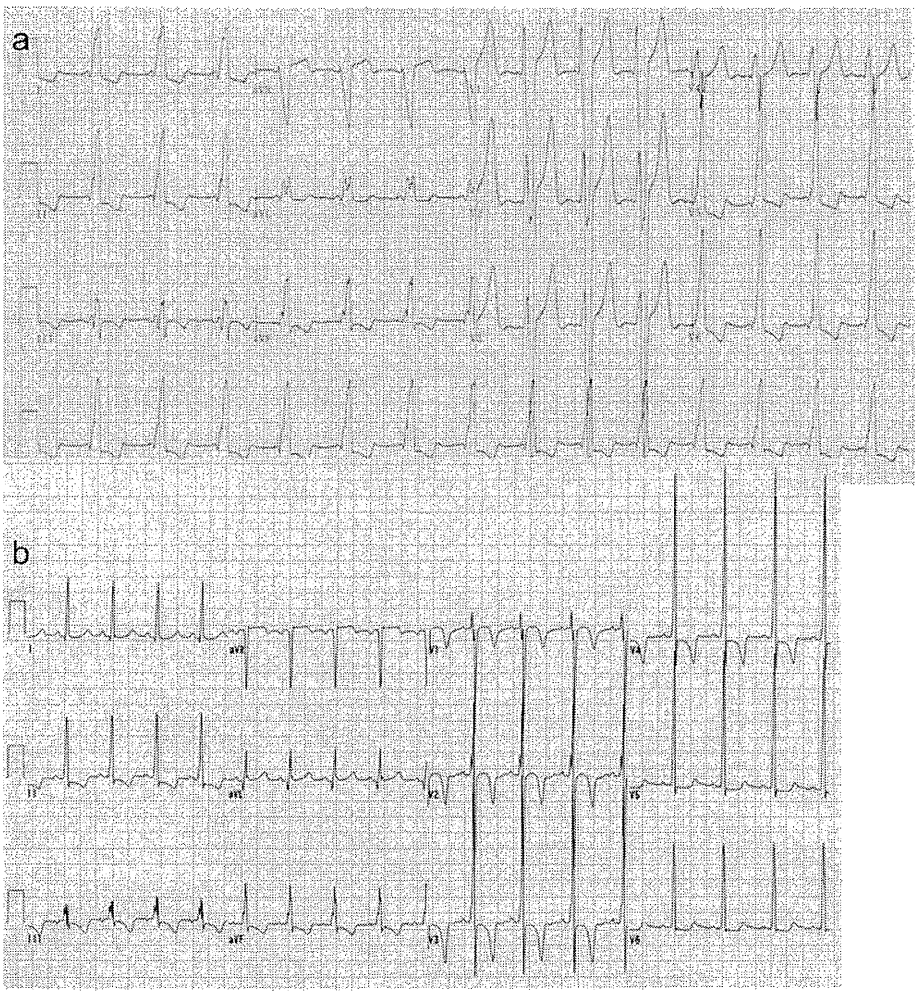

Fig. 1 Electrocardiograms before and after ablation of the accessory pathway.
a Electrocardiogram before the ablation. From the vector of the delta wave in this electrocardiogram, a right anterior accessory pathway was suggested. **b** Electrocardiogram after the ablation Although the T wave was negative in leads II, III, aVF, and V1–V6, and mild ST-segment depression was noted in II, III, aVF, V5, and V6, the delta wave disappeared after ablating the accessory pathway

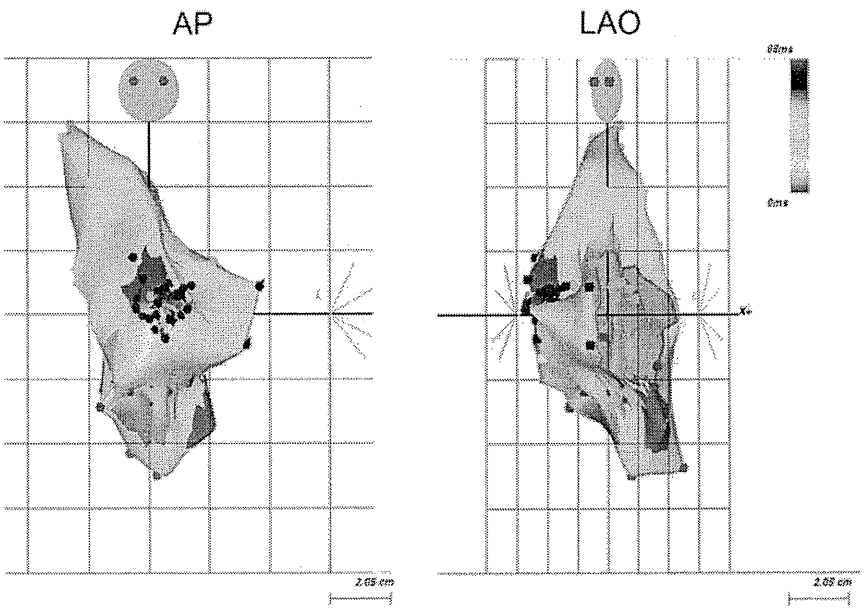

Fig. 2 Ventricular activation map before the radiofrequency catheter ablation (RFCA). Before the RFCA, the earliest ventricular activation was observed in the right lateral wall. The sites of the RFCA are represented by the *red points*. *AP* anterior posterior view, *LAO* left anterior oblique view

(Vivid 7 ultrasound system; GE Medical Systems, Milwaukee, WI, USA; or Acuson Sequoia 512 ultrasound system; Siemens Medical Solutions, Mountain View, CA, USA). There were no abnormal findings for the blood cell count and biochemical data, and the brain natriuretic peptide (BNP) level (9 pg/ml) was also within normal limits. Consequently he was diagnosed as having DCM. The dyskinesis was mostly detected in the ventricular septum. There were no abnormal findings in the coronary arteries by selective angiography; however, the LVEF was revealed to have decreased to 0.50 by left ventricular angiography. In addition, 201-thallium myocardial scintigraphy revealed no abnormal findings to suggest ischemic cardiomyopathy. Magnetic resonance imaging also revealed thinning of the interventricular septum, and the average EF decreased to 0.45. Based on these results, we considered that the left ventricular dyssynchrony was caused by pre-excitation due to the right-sided AP that may consequently have resulted in DCM-like dysfunction.

The patient underwent an electrophysiological study (EPS) when he was 11 years old after obtaining informed consent, using an electroanatomical mapping system (CARTO XP; Biosense Webster, Diamond Bar, CA, USA). No tachycardia could be induced by programmed atrial or ventricular pacing. Retrograde atrial conduction was confirmed via the atrioventricular node by the earliest atrial activation being noted in the His bundle recording area, and prolongation of the ventricular-atrial conduction

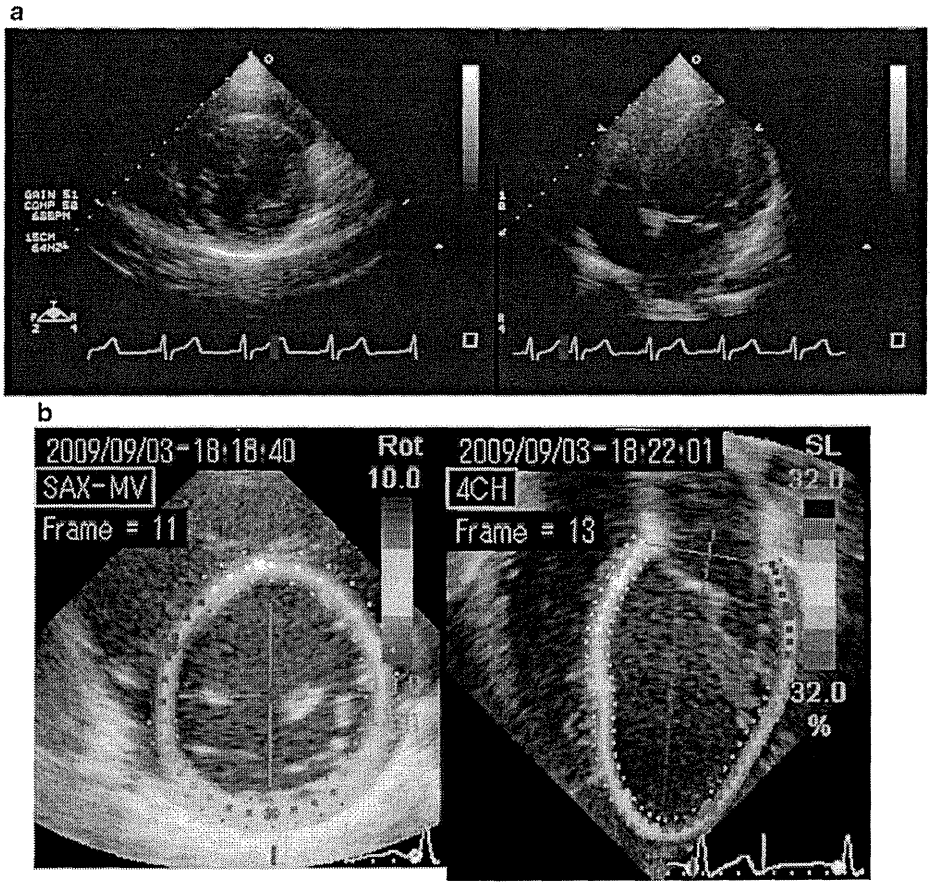

Fig. 3 Echocardiograms before and after ablation of the accessory pathway. **a** Echocardiogram before the ablation: LVDd = 61.8 mm, LVDs = 46.7 mm, IVSd = 7.0 mm, IVSs = 8.2 mm, LVPWd = 8.2 mm, LVPWs = 9.9 mm, LVEF = 0.45, LVFS = 0.24. The echocardiogram shows mitral valve regurgitation, thinning of the basal segments of the interventricular septum, a ventricular aneurysm with dyskinetic motion in systole, dilation of the left ventricular cavity, and an LVEF that decreased to 0.45. **b** Echocardiogram after the ablation: LVDd = 51.2 mm, LVDs = 31.3 mm, IVSd = 11.4 mm, IVSs = 14.2 mm, LVPWd = 10.4 mm, LVPWs = 20.9 mm, LVEF = 0.69, LVFS = 0.39. In the echocardiogram 6 months after the RFCA, the left ventricular wall motion markedly improved and the LVEF was 0.69. The thinning of the basal segments of the interventricular septum and ventricular aneurysm with dyskinetic motion also disappeared. *LVDd* left ventricular end-diastolic dimension, *LVDs* left ventricular end-systolic dimension, *IVSd* intraventricular end-diastolic septal thickness, *IVSs* intraventricular end-systolic septal thickness, *LVPWd* left ventricular end-diastolic posterior wall dimension, *LVPWs* left ventricular end-systolic posterior wall dimension, *LVEF* left ventricular ejection fraction, *LVFS* left ventricular fractional shortening

interval by incremental ventricular pacing and the use of 10 mg adenosine triphosphate. Consequently, we decided to map the earliest ventricular activation site during sinus rhythm. The earliest ventricular activation was observed in the right lateral wall, and we successfully ablated the AP (Fig. 1b). We recreated the activation map after the ablation. No premature excitation according to the site of the AP was found, and the atrioventricular node was the only conduction route involved in the atrioventricular conduction (Fig. 2).

During the follow-up echocardiography 6 months after the RFCA, it was observed that the LVEF was 0.69 using the Simpson method, the thinning of the basal segments of the interventricular septum returned to within normal limits, and the ventricular aneurysm with dyskinetic motion disappeared (Fig. 3b). For further evaluation, we analyzed the left ventricular wall motion using an Acuson Sequoia 512 ultrasound system with Velocity Vector Imaging analysis software (TomTec imaging systems, Unterschleissheim, Germany) before the RFCA, and a Vivid 7

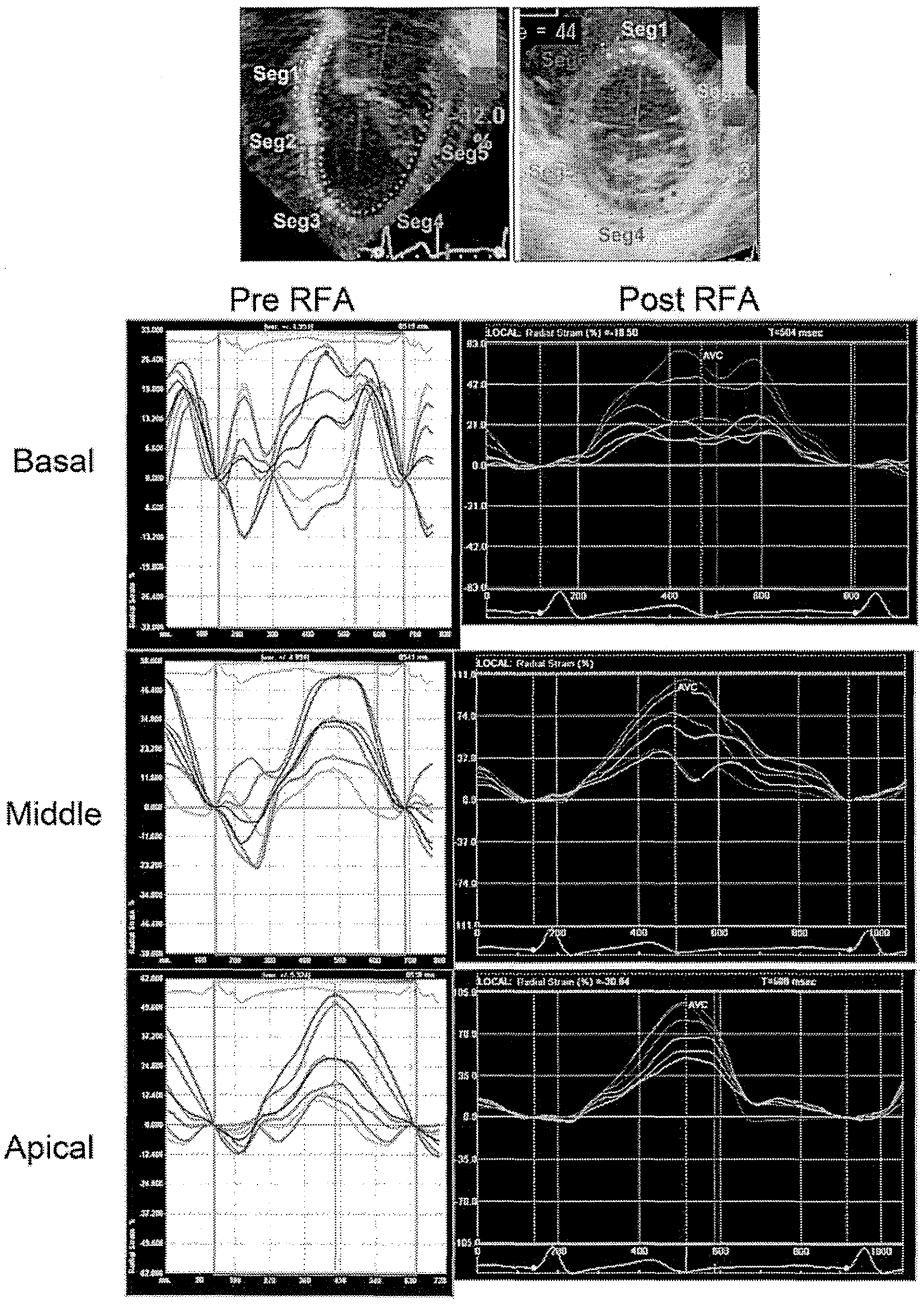

Fig. 4 Localized wall motion analyzed with speckle-tracking imaging. Analyzed by the radial strain, the time delay on the basal septal wall improved from 232 to 90 ms, and the Δ time delay was 142 ms. The Δ time delay of the midseptal and apical-septal radial strain became 31 and 47 ms, respectively

ultrasound system with EchoPAC analysis software (GE Medical Systems) after the RFCA. Intraventricular dyssynchrony was quantified as the time delay between the peak septal systolic motion and left posterior wall systolic motion (septal-to-posterior wall motion delay). The zero reference point was adjusted to the onset of the electrical systole (delta wave or QRS complex). Analyzed by the radial strain, the time delay at the basal septal wall improved from 232 to 90 ms, and the Δ time delay was 142 ms. The Δ time delay during the midseptal and apical-septal radial strain shortened by 31 and 47 ms, respectively (Fig. 4). Furthermore, the thinning of the interventricular septum and the mitral valve regurgitation had disappeared. The ventricular aneurysm with dyskinetic motion decreased, resulting in the recovery of the LVEF to 69 %.

Discussion

Paradoxical movement of the interventricular septum due to premature contractions of the right ventricle through the AP is occasionally observed by echocardiography in patients with WPW syndrome. The first report of cardiac resynchronization using RFCA of a right-sided AP in a child with palpitation attacks was reported by Shan et al. [4]. This patient was diagnosed with DCM and a severely reduced cardiac function. A possible pathophysiological linkage between APs and ventricular dysfunction has been proposed in patients with WPW syndrome, without any evidence of tachyarrhythmias [4, 6–9]. There have been several reports concerning the efficacy of RFCA in patients with preexcitation-induced cardiomyopathy or dyskinesis [4, 6, 7, 10, 11]. In these reports, 26 patients had a decreased ejection fraction, and all the locations of the APs in those patients who had a decreased ejection fraction existed in the right septal portion. In the present report, the location of the AP was on the right anterolateral portion, and to the best of our knowledge this is a rare report of a follow-up study by echo for this location of an AP. In previous reports, the heart function evaluated by tissue Doppler imaging showed an improvement in the ventricular dyssynchrony after RFCA. In this report, we confirmed the improvement of the regional movement after RFCA by speckle-tracking echocardiography. In most patients with WPW syndrome, the asynchronous ventricular motion would not cause any symptoms because the global cardiac function usually remains normal [12]. We speculated that an early septal activation may induce paradoxical or hypokinetic interventricular septal motion even if the AP is located in the right lateral portion. In a recent report, left ventricular dyssynchrony was also caused by a left-sided AP [13]. Right ventricular pacing has already been reported to be one of the risk factors for heart failure [14]. Although the electromechanical asynchrony is greater in septal APs, the location, which more likely participates in the dyssynchrony of the left ventricle and impairment of the cardiac function, is still controversial [15].

In conclusion, sudden death in asymptomatic WPW syndrome is rare; therefore, generally the indication of RFCA for APs has mostly been for patients with episodes of AVRT. However, RFCA should be the first priority for the treatment of patients who have DCM-like dyskinesis even without AVRT.

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Conflict of interest None declared.

References

1. Klein GJ, Yee R, Sharma AD (1989) Longitudinal electrophysiologic assessment of asymptomatic patients with the Wolff-Parkinson-White electrocardiographic pattern. N Engl J Med 320:1229–1233
2. Hishida H, Sotobata I, Koike Y, Okumura M, Mizuno Y (1976) Echocardiographic patterns of ventricular contraction in the Wolff-Parkinson-White syndrome. Circulation 54:567–570
3. DeMaria AN, Vera Z, Neumann A, Mason DT (1976) Alterations in ventricular contraction pattern in the Wolff-Parkinson-White syndrome. Detection by echocardiography. Circulation 53:249–257
4. Shan Q, Jin Y, Cao K (2007) Reversible left ventricular dyssynchrony and dysfunction resulting from right ventricular preexcitation. Europace 9:697–701
5. Nelson GS, Curry CW, Wyman BT, Kramer A, Declerck J, Talbot M, Douglas MR, Berger RD, McVeigh ER, Kass DA (2000) Predictors of systolic augmentation from left ventricular preexcitation in patients with dilated cardiomyopathy and intraventricular conduction delay. Circulation 101:2703–2709
6. Udink ten Cate FE, Kruessell MA, Wagner K, Trieschmann U, Emmel M, Brockmeier K, Sreeram N (2010) Dilated cardiomyopathy in children with ventricular preexcitation: the location of the accessory pathway is predictive of this association. J Electrocardiol 43:146–154
7. Tomaske M, Janousek J, Razek V, Gebauer RA, Tomek V, Hindricks G, Knirsch W, Bauersfeld U (2008) Adverse effects of Wolff-Parkinson-White syndrome with right septal or posteroseptal accessory pathways on cardiac function. Europace 10:181–189
8. Cadrin-Tourigny J, Fournier A, Andelfinger G, Khairy P (2008) Severe left ventricular dysfunction in infants with ventricular preexcitation. Heart Rhythm 5:1320–1322
9. Cai Q, Shuraih M, Nagueh SF (2012) The use of echocardiography in Wolff-Parkinson-White syndrome. Int J Cardiovasc Imaging 28:725–734
10. Iwasaku T, Hirooka K, Taniguchi T, Hamano G, Utsunomiya Y, Nakagawa A, Koide M, Ishizu T, Yamato M, Sasaki N, Yamamoto H, Kawaguchi Y, Mizuno H, Koretsune Y, Kusuoka H, Yasumura Y (2009) Successful catheter ablation to accessory atrioventricular pathway as cardiac resynchronization therapy in a patient with dilated cardiomyopathy. Europace 11:121–123

11. Kwon BS, Bae EJ, Kim GB, Noh CI, Choi JY, Yun YS (2009) Septal dyskinesia and global left ventricular dysfunction in pediatric Wolff-Parkinson-White syndrome with septal accessory pathway. J Cardiovasc Electrophysiol 21:290–295
12. Yamanaka S, Shirayama T, Inoue K, Kawata K, Yagi T, Azuma A, Inoue D, Nakagawa M (1998) Improved cardiac function after catheter ablation in a patient with type B Wolff-Parkinson-White syndrome with an old myocardial infarction. Jpn Circ J 62:860–862
13. Park HE, Chang SA, Kim JH, Oh IY, Choi EK, Oh S (2012) Left ventricular dyssynchrony in pre-excitation syndrome: effect of accessory pathway location and reversibility after ablation therapy. Heart Vessels. doi:10.1007/s00380-012-0233-x
14. Freudenberger RS, Wilson AC, Lawrence-Nelson J, Hare JM, Kostis JB (2005) Permanent pacing is a risk factor for the development of heart failure. Am J Cardiol 95:671–674
15. De Boeck BW, Teske AJ, Leenders GE, Mohamed Hoesein FA, Loh P, van Driel VJ, Doevendans PA, Prinzen FW, Cramer MJ (2010) Detection and quantification by deformation imaging of the functional impact of septal compared to free wall preexcitation in the Wolff-Parkinson-White syndrome. Am J Cardiol 106:539–546 e2

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CLINICAL RESEARCH

Public access defibrillation improved the outcome after out-of-hospital cardiac arrest in school-age children: a nationwide, population-based, Utstein registry study in Japan

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Aims The purpose of this study was to determine whether implementation of public access defibrillation (PAD) improves the outcome after out-of-hospital cardiac arrest (OHCA) in school-age children at national level.

Methods and results We conducted a prospective, nationwide, population-based Japanese Utstein registry study of consecutive OHCA cases in elementary and middle school children (7–15 years of age) who had a bystander-witnessed arrest of presumed cardiac origin during 2005–09 and received pre-hospital resuscitation by emergency responders. The primary endpoint was a favourable neurological outcome 1 month after an arrest. Among 230 eligible patients enrolled, 128 had ventricular fibrillation (VF) as an initial rhythm. Among these 128 patients, 29 (23%) children received a first shock by a bystander. Among these 29 patients, the proportion of the favourable neurological outcome after OHCA was 55%. During the study period, the proportion of patients initially shocked by a bystander among eligible patients increased from 2 to 21% ($P = 0.002$ for trend). The proportion of patients with a favourable neurological outcome after OHCA increased from 12 to 36% overall ($P = 0.006$). The collapse to defibrillation time was shorter in bystander-initiated defibrillation when compared with defibrillation by emergency responders (3.3 ± 3.7 vs. 12.9 ± 5.8 min, $P < 0.001$), and was independently associated with a favourable neurological outcome after OHCA [$P = 0.03$, odds ratio (OR) per 1 min increase, 0.90 (95% confidence interval 0.82–0.99)]. A non-family member's witness was independently associated with VF as the initial rhythm [$P < 0.001$, OR 4.03 (2.08–7.80)].

Conclusion Implementation of PAD improved the outcome after OHCA in school-age children at national level in Japan.

Keywords Cardiopulmonary resuscitation • Sudden unexplained death • School health • Public access defibrillation • School-age children

Introduction

Sudden cardiac death in elementary and middle school children is a rare but tragic event, which has tremendous impact on the family, school, communities, and health-care providers, and which may be relevant to cardiopulmonary resuscitation (CPR)/automated external defibrillator (AED) programmes in the public environment surrounding these children.[1,2] Recently, implementation of public

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What's new?

- This is the first population-based study, which specifically addressed the impact of public access defibrillation on the outcome of out-of-hospital cardiac arrests (OHCAs) in elementary and middle school children.
- Among 230 eligible patients, 128 (56%) had ventricular fibrillation (VF) as an initial rhythm. Among these 128 patients, 29 (23%) children received a first shock by a bystander. Among these 29 patients, the proportion of the favourable neurological outcome after OHCA was 55%.
- During the study period 2005–09, the proportion of patients initially shocked by a bystander among eligible patients increased from 2 to 21%. The proportion of patients with a favourable neurological outcome after OHCA increased from 12 to 36% overall.
- A non-family member's witness was independently associated with VF as the initial rhythm. The collapse to defibrillation time was independently associated with a favourable neurological outcome, the survival at 1 month, and the prehospital return of spontaneous circulation after OHCA.

access defibrillation (PAD) improved outcomes among adults after out-of-hospital cardiac arrest (OHCA) in public locations, by reducing the time interval from the patient's collapse to defibrillation.[3–6] However, the impact of PAD on the outcome after OHCA in such school-age children was unclear. This question is challenging two-fold. First, paediatric patients of different ages have diverse aetiologies of OHCA; relatively poor survival has been reported in this heterogeneous group of patients.[7,8] The reported incidence of ventricular fibrillation (VF) as an initial rhythm in paediatric OHCA is lower than that reported in adults, and the effectiveness of early defibrillation programmes even for paediatric patients in VF arrest has been questioned.[7,8] Secondly, although school-age children are reported to spend a large part of their active daytime in public locations,[9] it is uncertain whether PAD programme, if any, would be effective for ordinary children in the children's public environment, including schools.[2,9–11] Recently, VF was found to be present in a higher percentage of high school-age athletes with sudden arrest, and recent small series have noted improved survival when early defibrillation with CPR was provided for such patients in high schools.[11–13] However, the limited deployment of AED devices in elementary and middle schools, and other public locations, a small sample size of OHCA in this age population in local studies, and the lack of an appropriate reporting system of OHCA, may have hampered any investigations involving the epidemiological basis of the benefit of PAD for OHCA in such school-age children.[11,12,14,15]

In Japan in July 2004, the Ministry of Health, Labour and Welfare approved AED use by citizens. By 2009, the number of AED devices in public places increased to 203 924 (106.6/100 000 population).[16,17] Of note, up to 28.9% of public access AED devices in Japan were placed in schools; by 2009, AEDs were placed in 72% of elementary schools and 89.8% of middle schools.[18,19] In January 2005, the Fire and Disaster Management Agency of Japan launched a prospective, nationwide, population-based, Utstein-style registry involving consecutive OHCA victims in all the age groups.[20] A recent study, using the Utstein registry database, demonstrated that there was a temporal increase in public access AED application and improved outcomes after OHCA in adults at the national level.[20] However, the impact of the national PAD programme on outcomes of OHCA in elementary and middle school children has not been reported. We therefore investigated whether PAD may have an impact on the outcome after OHCA in such school-age children at the national level, by using the Japanese Utstein registry database.[20,21]

Methods

Study design

The All-Japan registry of the Fire and Disaster Management Agency of Japan is a prospective, nationwide, population-based registry of OHCA, which is based on the standardized Utstein style, as reported in detail previously.[20,21] Briefly, this cohort enrolled all consecutive patients who suffered OHCA all over Japan, and were treated by emergency medical service (EMS) personnel and transported to hospitals. Specific enrolment process was described in Supplementary material online, Supplementary methods.[20,21] Among these patients, who had OHCA during January 2005–December 2009, we identified eligible patients who were 7–15 years of age, because we would include school-age students in compulsory education, which corresponds to the elementary and middle schools in Japan: high school students were thereby excluded. We identified those school-age victims with bystander-witnessed OHCA of presumed cardiac origin occurring during the entire day. Cardiac arrest was defined as the cessation of cardiac mechanical activity as confirmed by the absence of signs of circulation.[22–24] The arrest was presumed to be of cardiac origin unless it was caused by non-cardiac (respiratory disease, malignant tumours, and central nervous system disorders), external (trauma, hanging, drowning, drug overdose, and asphyxia) , or any other non-cardiac factors.[22–24] The data form was filled out by the EMS personnel in cooperation with the physicians in charge of the patients, and the data were integrated into the registry system on the database server. The working group for All-Japan Utstein registry designed the study protocol; collected and managed the data; and the authors analysed the data and wrote the manuscript. The protocol for analyses was approved by the Ethics Committee of Mie University Graduate School of Medicine.

Study setting

Emergency medical service and training system in Japan was previously reported in detail.[20,21] Briefly, Japan has an area of ~378 000 km^2, and its population was 127 million, including 3 666 839 male and 3 496 405 female 7–12-year-old children (elementary school students), and 1 871 134 male and 1 780 230 female 13–15-year-old children (middle school students) in 2005.[25] Placement of AEDs in public locations was driven by either public or private initiatives.[17] The cumulative number of public access AEDs, excluding those in medical facilities and EMS institutions, as estimated from sales of AEDs, increased from 9906 to 203 924 during the 5-year study period (see Supplementary material online, *Table*).[16] A total of 96.5% of public access AEDs are located in public locations (28.9% in schools, 20.6% in workplaces, 8.8% in nursing homes, 5.7% in sports facilities, 4.8% in cultural facilities, 2.6% in public transportation facilities, and 25.1% in other public locations), 1.4% in residential areas, and 2.1% in others.[18] From 2007 to 2009, the

percentage of elementary or middle schools equipped with at least one AED device increased from 18.1 to 72.9% in elementary schools and from 38.3 to 89.8% in middle schools (see Supplementary material online, *Table*).[19] School teachers and other staff were trained in CPR programmes by EMS providers or other instructors, under the guidance of local school boards, in which paediatric and adult PADs were generally recommended for children at 7 years of age and older, respectively, in accordance with the Japanese CPR guidelines.[26] In Japan, ~1.4–1.5 million citizens per year participated in the CPR/AED training programmes, generally provided by local fire departments.[27]

Data collection

Procedure of data collection was described previously.[20,21] Briefly, registry data were prospectively collected in accordance with the Utstein-style reporting guidelines for OHCA, which is a standardized form (uniform definitions, terminology, and recommended data sets) for clinical investigators to report human resuscitation studies.[20,21,23,24] Specific data sets and data collecting process were described in Supplementary material online, Supplementary methods.[20,21,23,24] In the present Japanese Utstein reporting system, a patient initially shocked by a bystander was defined as one in which a public access AED was used and the shock was delivered; if the public access AED was applied but the shock was not delivered, the patient was not included in this category.[20,21] In this analysis, an OHCA witnessed by non-family members was presumed to be an event in a public location, because of a lack of data with respect to specific locations in the registry; when a bystander delivered shocks with an AED, the initial rhythm of the patient was regarded as VF, including pulseless ventricular tachycardia.

Endpoints

The primary endpoint was survival at 1 month with minimal neurological impairment, which was defined as a Glasgow–Pittsburg cerebral performance category of 1 (good performance) or 2 (moderate disability).[23,24] Secondary endpoints were survival at 1 month and return of spontaneous circulation (ROSC) before arrival at the hospital.

Statistical analysis

The age-stratified annual incidence of OHCA was calculated with the use of 2005 census data.[25] Continuous variables between two groups were assessed by the unpaired *t*-test. Trends in categorical and continuous variables were analysed with the use of univariate regression models and linear tests, respectively, in overall and subgroups of eligible patients, determined by the relation of bystanders to the victims (family or non-family member). The planned subgroup analysis was intended to determine the impact of PAD on trends in outcome parameters of arrest in presumed public locations (non-family member-witnessed arrests) in comparison with the non-public location arrest (family member-witnessed arrests). Univariate and multivariable logistic regression analyses were performed to assess the factors associated with VF as the initial rhythm, and outcome parameters. Adjusted and unadjusted odds ratios with their 95% confidence intervals and *P* values were reported. Potential confounding factors adjusted for VF as the initial rhythm included the calendar year, the age, gender, the relation of the bystander to the patient (family or non-family member), the type of CPR initiated by a bystander (compression-only or conventional CPR), and the time from the witnessed collapse to the EMS arrival, in accordance with previous reports.[20,21,28] Potential confounding factors for outcome parameters included VF as an initial rhythm, bystander's AED use at the first shock, and the time from the witnessed collapse to the first shock, in addition to the potential confounders for VF, in accordance with previous reports.[20,21,28] All statistical analyses were performed with the use of the SPSS statistical package, version 16.0J (SPSS). Data were reported as mean ± standard deviation. All tests were two-tailed, and *P* values of <0.05 were considered to indicate statistical significance.

Results

Among 2072 OHCA children, 522 were of presumed cardiac origin; 230 of 522 arrests were witnessed by bystanders (*Figure 1*). Among a total of 230 eligible patients, 128 (56%) children had VF as the initial rhythm. Among these 128 patients, 29 (23%) children received a first shock by bystanders using a public access AED before the arrival of EMS personnel and 96 (75%) children received a first shock by EMS personnel (32 with a monophasic and 64 with a biphasic defibrillator). In addition, among 102 patients without VF as the initial rhythm, none received bystander's defibrillation, but 13 (13%) received a shock by EMS personnel following CPR. Among 128 children with VF as the initial rhythm, 53 (41%) survived with a favourable neurological outcome, 67 (52%) survived 1 month after the arrest, and 55 (43%) had pre-hospital ROSC. Among the subset of 29 school-age children with OHCA who received initial AED shock by bystanders, 16 (55%) survived with favourable neurologic outcome, 19 (66%) survived 1 month after the arrest and 19 (66%) had prehospital ROSC. Among 102 patients without VF as the initial rhythm, 8 (8%) survived with favourable neurological outcome, 15 (15%) survived 1 month after the arrest and 11 (11%) had pre-hospital ROSC. The time interval from collapse to the initiation of CPR was shorter in bystander-initiated CPR than EMS-initiated CPR (3.2 ± 4.9 vs. 8.9 ± 6.8 min, $P < 0.001$). The interval from collapse to the initiation of AED use was shorter in bystander-initiated AED use than EMS-initiated one (3.3 ± 3.7 vs. 12.9 ± 5.8 min, $P < 0.001$). Clinical and outcome parameters in the overall, family and non-family member witnessed arrests were reported in *Table 1*. The population-based age-stratified incidence of bystander-witnessed OHCA of presumed cardiac origin in children was constant during the study period (see Supplementary material online, *Table*).

Trends in clinical and outcome parameters

During the study period (*Table 2*), the proportion of patients initially shocked by a bystander's AED among total patients increased from 2% in 2005 to 21% in 2009 ($P = 0.002$). Such a temporal increase was observed in non-family member-witnessed arrests, from 4% in 2005 to 37% in 2009 ($P = 0.001$), but not in family member-witnessed arrests. The collapse to AED time tended to become shorter only in non-family member-witnessed arrests, from 11.1 min in 2005 to 8.3 min in 2009 ($P = 0.07$). The proportion of any other categorical and continuous variables investigated in either subgroup of patients did not change significantly (see Supplementary material online, *Appendix 1*). As the outcome parameters (*Figure 2*), the proportion of patients with a favourable neurological outcome among total patients increased from 12% in 2005 to 36% in 2009 ($P = 0.006$). Such a temporal improvement

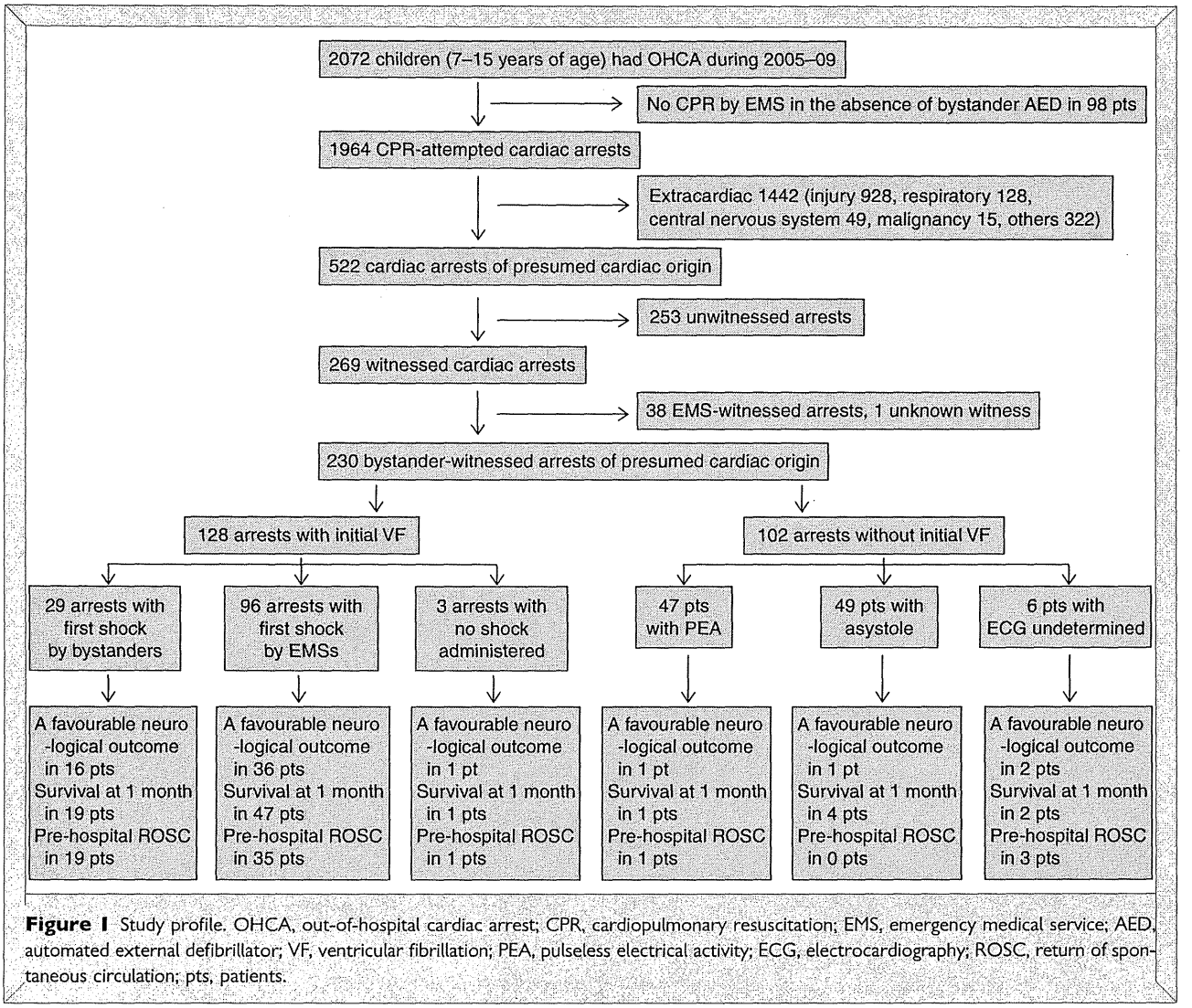

Figure 1 Study profile. OHCA, out-of-hospital cardiac arrest; CPR, cardiopulmonary resuscitation; EMS, emergency medical service; AED, automated external defibrillator; VF, ventricular fibrillation; PEA, pulseless electrical activity; ECG, electrocardiography; ROSC, return of spontaneous circulation; pts, patients.

was observed only in non-family member-witnessed arrests, from 9% in 2005 to 53% in 2009 ($P = 0.001$). The proportion of survival at 1 month after OHCA ($P = 0.008$) and ROSC before arrival at the hospital ($P = 0.046$) increased only in non-family member-witnessed arrests, from 17 and 17% in 2005 to 53 and 42% in 2009, respectively. Trends in specific values in all the clinical and outcome parameters investigated in overall and subgroups of patients were reported (see Supplementary material online, *Appendix 1*).

Multivariable analysis

In multivariable analysis (*Table 3*), a non-family member's witness [$P < 0.001$, adjusted odds ratio (OR) 4.03 (2.08–7.80)] was independently associated with the presence of VF as the initial rhythm. The collapse to AED time, either by a bystander or an emergency responder [$P = 0.03$, OR per 1 min increase, 0.90 (0.82–0.99)], and female gender [$P = 0.008$, 3.20 (1.35–7.56)] were independently associated with a favourable neurological outcome. The collapse to AED time was the only variable independently associated with the survival at 1 month [$P = 0.045$, 0.92 (0.85–0.99)] and pre-hospital ROSC [$P = 0.001$, 0.82 (0.73–0.92)]. Results of univariate analysis were reported in the see Supplementary material online, *Appendix 2*.

Discussion

Although the epidemiological data related to the impact of disseminating PAD programmes on OHCA in elementary and middle school children were limited,[7,8] the present Utstein registry study would supply evidence supporting that implementation of PAD programmes increases the likelihood of early defibrillation by bystanders, and improves the outcome after OHCA in such school-age children. These findings may underscore the benefit of PAD in the prevention of sudden cardiac death in school-age children.

Table 1 Clinical and outcome parameters

Parameters	Total (n = 230)	Family witnessed (101)	Non-family witnessed (129)
Age, years of age	12.2 ± 2.5	11.3 ± 2.7	12.8 ± 2.1
Male gender, *n* (%)	145 (63)	63 (62)	82 (64)
Ventricular fibrillation, *n* (%)	128 (56)	37 (37)	91 (71)
CPR initiated by bystanders, *n* (%)	161 (70)	55 (55)	106 (82)
Conventional CPR, *n* (%)	102 (64)	30 (55)	72 (69)
Collapse to CPR time (min)	4.9 ± 6.1	5.3 ± 6.3	4.6 ± 6.0
Shock initiated, *n* (%)			
by bystanders	29 (13)	2 (2)	27 (21)
by EMS	109 (47)	38 (38)	71 (55)
Collapse to AED time (min)	10.9 ± 6.7	13.1 ± 7.0	10.0 ± 6.4
Collapse to EMS arrival (min)	34.6 ± 17.2	35.0 ± 16.5	34.3 ± 17.7
Favourable neurological outcome, *n* (%)	63 (27)	15 (15)	48 (37)
Survival at 1month, *n* (%)	84 (37)	22 (22)	62 (48)
Prehospital ROSC, *n* (%)	66 (29)	20 (20)	46 (36)

Favourable neurological outcome denotes cerebral performance category 1 or 2 at 1 month.
Conventional CPR indicated chest compression with rescue breathing, as a type of bystander-initiated CPR. Percentages were calculated on the basis of the available data in overall or each subgroup of arrests (family or nonfamily witnessed). Plus–minus values are means ± SD.
CPR, cardiopulmonary resuscitation; EMS, emergency medical service; AED, automated external defibrillator; ROSC, return of spontaneous circulation.

Table 2 Trends in clinical parameters

Variables	2005	2006	2007	2008	2009	*P* value for trend
Type of bystanders, (*n*)						
Total	41	46	51	48	44	
Family member	18	18	21	19	25	0.27
Non-family member	23	28	30	29	19	
CPR initiated by bystanders, *n* (%)						
Total	26 (63)	33 (72)	36 (72)	37 (77)	29 (66)	0.65
Family member	8 (44)	9 (50)	12 (60)	12 (63)	14 (56)	0.35
Non-family member	18 (78)	24 (86)	24 (80)	25 (86)	15 (79)	0.90
Shock initiated by bystanders, *n* (%)						
Total	1 (2)	1 (2)	10 (20)	8 (17)	9 (21)	0.002
Family member	0 (0)	0 (0)	0 (0)	0 (0)	2 (8)	0.99
Non-family member	1 (4)	1 (4)	10 (33)	8 (28)	7 (37)	0.001
Shock initiated by EMS, *n* (%)						
Total	18 (44)	24 (52)	26 (51)	19 (40)	22 (50)	0.94
Family member	6 (33)	3 (17)	10 (48)	7 (37)	12 (48)	0.14
Non-family member	12 (52)	21 (75)	16 (53)	12 (41)	10 (53)	0.22
Collapse to AED time (min)						
Total	11.8 ± 4.8	12.5 ± 5.4	10.4 ± 7.2	10.2 ± 6.2	10.3 ± 8.4	0.22
Family member	13.5 ± 7.2	14.3 ± 4.6	13.3 ± 4.4	12.7 ± 3.0	12.6 ± 10.3	0.71
Non-family member	11.1 ± 3.2	12.2 ± 5.5	9.2 ± 7.8	9.4 ± 6.8	8.3 ± 5.9	0.07

Percentages were calculated on the basis of the available data in overall or each subgroup of arrests (family or non-family witnessed) in the respective year. Plus-minus values are means ± SD.
CPR, cardiopulmonary resuscitation; EMS, emergency medical service; AED, automated external defibrillator.

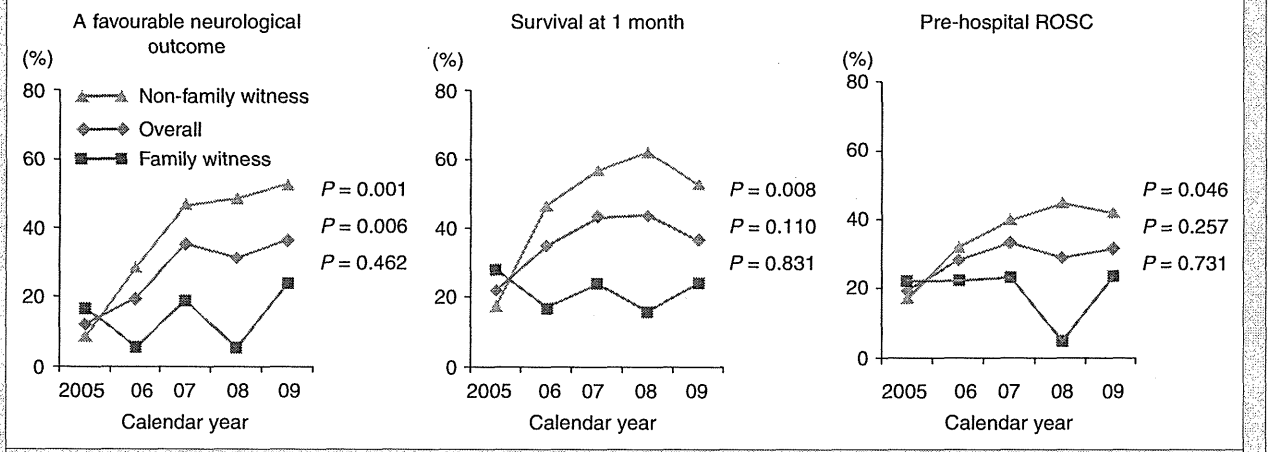

Figure 2 Trends in outcome parameters in arrests, by the relationship of a bystander to the victim. P values are for trend; ROSC, return of spontaneous circulation.

Table 3 Multivariable analyses of factors associated with ventricular fibrillation as the initial rhythm and outcome parameters

Variable	Ventricular fibrillation	Favourable neurological outcome	Survival at 1 month	Pre-hospital ROSC
	Adjusted OR (95% CI)	Adjusted OR (95% CI)	Adjusted OR (95% CI)	Adjusted OR (95% CI)
Year (per 1-year increase)	1.09 (0.87–1.37)	1.19 (0.87–1.63)	0.97 (0.73–1.30)	0.80 (0.58–1.10)
P value	0.46	0.29	0.86	0.17
Age≥13 years	1.83 (0.96–3.48)	1.79 (0.76–4.20)	1.60 (0.72–3.54)	1.60 (0.67–3.80)
P value	0.07	0.18	0.25	0.29
Female gender	0.76 (0.39–1.47)	3.20 (1.35–7.56)	1.80 (0.79–4.10)	2.17 (0.91–5.19)
P value	0.41	0.008	0.16	0.08
Non-family witnessed	4.03 (2.08–7.80)	1.53 (0.58–4.03)	1.68 (0.70–4.02)	0.86 (0.32–2.29)
P value	<0.001	0.39	0.27	0.76
CPR				
Not bystander-initiated	reference	reference	reference	reference
Bystander-initiated				
Conventional	0.76 (0.35–1.65)	1.01 (0.34–3.00)	1.24 (0.45–3.43)	0.90 (0.29–2.78)
P value	0.49	0.98	0.68	0.86
Compression only	0.79 (0.34–1.83)	1.55 (0.51–4.71)	1.08 (0.38–3.06)	1.77 (0.58–5.46)
P value	0.58	0.44	0.88	0.32
Collapse–EMS time (per 1 min increase)	0.99 (0.97–1.37)	1.00 (0.98–1.03)	1.00 (0.98–1.03)	1.01 (0.99–1.04)
P value	0.30	0.74	0.89	0.33
Ventricular fibrillation as the initial rhythm		2.03 (0.43–9.46)	1.30 (0.34–4.91)	0.76 (0.18–3.20)
P value		0.37	0.70	0.71
Bystander's AED		0.49 (0.12–2.02)	0.59 (0.15–2.23)	0.61 (0.14–2.76)
P value		0.32	0.43	0.53
Collapse–AED time (per 1-min increase)		0.90 (0.82–0.99)	0.92 (0.85–0.99)	0.82 (0.73–0.92)
P value		0.03	0.045	0.001

Favourable neurological outcome denotes cerebral performance category 1 or 2 at 1 month.
OR, odds ratio; ROSC, return of spontaneous circulation; EMS, emergency medical service; CPR, cardiopulmonary resuscitation; AED, automated external defibrillator.

Impact of public access defibrillation on out-of-hospital cardiac arrest in school-age children

Between 2005 and 2009 in Japan, there was a remarkable increase in the availability of AED in public spaces surrounding school children, including schools.[16–19] During this period, there was an increase in the proportion of OHCA in which the victim was initially shocked by a bystander, and this was temporally associated with an improvement in the neurological outcome in children with OHCA. In subgroup analyses, (i) temporal trends in these parameters were evident in non-family member-witnessed arrests, but not in family member-witnessed arrests, (ii) similar trends in secondary outcome parameters were observed in non-family member-witnessed arrests, and (iii) trends in other clinical parameters were not affected in either subgroup of patients during the same period. Therefore, trends in relevant variables, together with multivariable analysis data, consistently support that introduction of PAD programmes would increase the likelihood of early defibrillation by bystanders, and improve the outcomes of school-age children after public location arrest. Such an impact of PAD on OHCA in school-age children is consistent with that reported in adults.[20] In an adult study (≥18 years of age) by using the same Japanese Utstein registry data during 2005–07, 32% of patients with bystander-witnessed OHCA of presumed cardiac origin with initial rhythm of VF who received bystander AED shock delivery had a favourable neurological outcome.[20] In the present study during the corresponding years 2005–07 (data not shown), 58% (7/12) of children who received bystander-initiated shock had a favourable neurological outcome. In other adult studies, the survival rate of OHCA patients initially shocked by a bystander was ~60%.[3,4,6,13] Thus, the survival to 1 month with good neurological outcome of school-age children who experience witnessed OHCA with bystander CPR and AED shock delivery appears to equal or surpass that reported in adults. The more favourable outcome in this paediatric population may result from the higher rate of bystander CPR, and the shorter collapse to CPR and collapse to AED shock delivery intervals than those observed in adults with OHCA during the same period in Japan. This may be explained in part by factors in the school environment, such as constant visual observation of the children and focused training of teachers and staff.

Frequency of ventricular fibrillation as the initial rhythm in out-of-hospital cardiac arrest in school-age children

The frequency of VF in OHCA in children has been debated for a decade, and has been negatively influenced by the young age (<1 year of age), and traumatic and respiratory aetiologies.[7,8,14,15] In the present study, as high as 56% of bystander-witnessed arrests of presumed cardiac origin in school-age children were associated with VF. This is consistent with the results in local studies (in King county of USA, and in a province of the Netherlands), in which the frequency of VF has been positively associated with the advanced age (≥8 years of age), witnessed arrest, and cardiac aetiology, and a half of arrest patients had an initial rhythm of VF among adolescents aged 13–18 years with witnessed arrest.[11,29] In our study, we could further demonstrate that the non-family member-witnessed arrest was independently associated with VF as the initial rhythm, which is consistent with the results in an adult study.[28] The relatively low proportion of initial VF in adolescent OHCA in ROC study may be related to the difference of witness status, aetiology, and the reporting system.[14] The present study suggests that the relatively high proportion of initial VF in bystander-witnessed OHCAs of presumed cardiac origin in public locations in school-age children may confer an epidemiological basis for early defibrillation in this age population.

Limitations

Several limitations could be acknowledged in this study, in addition to those, as described previously.[20,21] First, the proportion of OHCA patients in schools among total eligible samples is unknown, because of the lack of data with respect to school as a specific location in the registry. Secondly, there might be unmeasured confounding factors (i.e. quality of bystander's CPR) that might influence the association between bystander's defibrillation and outcomes. Thirdly, information on in-hospital treatment (ie, hypothermia) is unavailable, which might affect survival after OHCA. Fourthly, it is unknown whether the present information can be generalized to other communities with different emergency response programmes at schools and other public locations surrounding children,[18,19] or different EMS systems.[20] Fifthly, the present investigation is not a cost-effectiveness analysis, although a previous study of cardiac arrests in high schools indicated that PAD may be cost-effective in schools.[12] Sixthly, specific data on the scope of the budgetary barriers and logistic issues (i.e. the locations of AED placement, training schedule for teachers) in implementing and refining AED/CPR programmes at the national level in Japan is unavailable.[17]

Conclusions

Although the impact of PAD has been largely elusive in overall children of different ages after etiologically diverse OHCAs in their public environmnent,[7,8] the present study would supply evidence which could dissect an epidemiological basis of the benefit of PAD in school-age children after bystander-witnessed OHCA of presumed cardiac origin. We believe that these findings are relevant to medical emergency response and CPR/AED programmes in the public environment surrounding school-age children.[1,2]

Supplementary material

Supplementary material is available at *Europace* online.

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Conflict of interest: none declared.

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References

1. Hazinski MF, Markenson D, Neish S, Gerardi M, Hootman J, Nichol G *et al.* Response to cardiac arrest and selected life-threatening medical emergencies: the medical emergency response plan for schools: a statement for healthcare providers, policymakers, school administrators, and community leaders. *Circulation* 2004;**109**:278–91.
2. Cave DM, Aufderheide TP, Beeson J, Ellison A, Gregory A, Hazinski MF *et al.* Importance and implementation of training in cardiopulmonary resuscitation and automated external defibrillation in schools: a science advisory from the American Heart Association. *Circulation* 2011;**123**:691–706.
3. Valenzuela TD, Roe DJ, Nichol G, Clark LL, Spaite DW, Hardman RG. Outcomes of rapid defibrillation by security officers after cardiac arrest in casinos. *N Engl J Med* 2000;**343**:1206–9.
4. Page RL, Joglar JA, Kowal RC, Zagrodzky JD, Nelson LL, Ramaswamy K *et al.* Use of automated external defibrillators by a U.S. Airline. *N Engl J Med* 2000;**343**:1210–6.
5. Hallstrom AP, Ornato JP, Weisfeldt M, Travers A, Christenson J, McBurnie MA *et al.* Public-access defibrillation and survival after out-of-hospital cardiac arrest. *N Engl J Med* 2004;**351**:637–46.
6. Caffrey SL, Willoughby PJ, Pepe PE, Becker LB. Public use of automated external defibrillators. *N Engl J Med* 2002;**347**:1242–7.
7. Donoghue AJ, Nadkarni V, Berg RA, Osmond MH, Wells G, Nesbitt L *et al.* Out-of-hospital pediatric cardiac arrest: an epidemiologic review and assessment of current knowledge. *Ann Emerg Med* 2005;**46**:512–22.
8. Young KD, Gausche-Hill M, McClung CD, Lewis RJ. A prospective, population-based study of the epidemiology and outcome of out-of-hospital pediatric cardiopulmonary arrest. *Pediatrics* 2004;**114**:157–64.
9. Nitta M, Iwami T, Kitamura T, Nadkarni VM, Berg RA, Shimizu N *et al.* Age-specific differences in outcomes after out-of-hospital cardiac arrests. *Pediatrics* 2011;**128**:e812–20.
10. Lotfi K, White L, Rea T, Cobb L, Copass M, Yin L *et al.* Cardiac arrest in schools. *Circulation* 2007;**116**:1374–9.
11. Bardai A, Berdowski J, van der Werf C, Blom MT, Ceelen M, van Langen IM *et al.* Incidence, causes, and outcomes of out-of-hospital cardiac arrest in children. A comprehensive, prospective, population-based study in the Netherlands. *J Am Coll Cardiol* 2011;**57**:1822–8.
12. Berger S, Whitstone BN, Frisbee SJ, Miner JT, Dhala A, Pirrallo RG *et al.* Cost-effectiveness of project ADAM: a project to prevent sudden cardiac death in high school students. *Pediatr Cardiol* 2004;**25**:660–7.
13. Drezner JA, Rao AL, Heistand J, Bloomingdale MK, Harmon KG. Effectiveness of emergency response planning for sudden cardiac arrest in United States high schools with automated external defibrillators. *Circulation* 2009;**120**:518–25.
14. Atkins DL, Everson-Stewart S, Sears GK, Daya M, Osmond MH, Warden CR *et al.* Epidemiology and outcomes from out-of-hospital cardiac arrest in children: the Resuscitation Outcomes Consortium Epistry-Cardiac Arrest. *Circulation* 2009; **119**:1484–91.
15. Zideman DA, Hazinski MF. Background and epidemiology of pediatric cardiac arrest. *Pediatr Clin North Am* 2008;**55**:847–859, ix.
16. Report on a study on social system development to improve survival from emergency cardiovascular disease using automated external defibrillator (Marukawa's report)(in Japanese)(23 June 2011). http://kouroukaken-kyukyusosei.Info/wpm/archivepdf/21/2_11a.Pdf
17. Mitamura H. Public access defibrillation: Advances from Japan. *Nat Clin Pract Cardiovasc Med* 2008;**5**:690–2.
18. Japanese Foundation for Eemergency Medicine: A search for information on placement of AEDS (in Japanese) (23 June 2011). http://www.Qqzaidan.Jp/aed/aed.Htm
19. *National Survey on AED Placement at Schools* (in Japanese) (23 July 2011). http://www.Mext.Go.Jp/a_menu/gakkouanzen/syousai/__icsfiles/afieldfile/2009/06/17/1267499_2.Pdf
20. Kitamura T, Iwami T, Kawamura T, Nagao K, Tanaka H, Hiraide A. Nationwide public-access defibrillation in Japan. *N Engl J Med* 2010;**362**:994–1004.
21. Kitamura T, Iwami T, Kawamura T, Nagao K, Tanaka H, Nadkarni VM *et al.* Conventional and chest-compression-only cardiopulmonary resuscitation by bystanders for children who have out-of-hospital cardiac arrests: a prospective, nationwide, population-based cohort study. *Lancet* 2010;**375**: 1347–54.
22. Zaritsky A, Nadkarni V, Hazinski MF, Foltin G, Quan L, Wright J *et al.* Recommended guidelines for uniform reporting of pediatric advanced life support: the pediatric Utstein style. A statement for healthcare professionals from a task force of the American Academy of Pediatrics, the American Heart Association, and the European Resuscitation Council Writing Group. *Circulation* 1995;**92**: 2006–20.
23. Cummins RO, Chamberlain DA, Abramson NS, Allen M, Baskett PJ, Becker L *et al.* Recommended guidelines for uniform reporting of data from out-of-hospital cardiac arrest: the Utstein style. A statement for health professionals from a task force of the American Heart Association, the European Resuscitation Council, the Heart and Stroke Foundation of Canada, and the Australian Resuscitation Council. *Circulation* 1991;**84**:960–75.
24. Jacobs I, Nadkarni V, Bahr J, Berg RA, Billi JE, Bossaert L *et al.* Cardiac arrest and cardiopulmonary resuscitation outcome reports: update and simplification of the Utstein templates for resuscitation registries: a statement for healthcare professionals from a task force of the International Liaison Committee on Resuscitation (American Heart Association, European Resuscitation Council, Australian Resuscitation Council, New Zealand Resuscitation Council, Heart and Stroke Foundation of Canada, Inter-American Heart Foundation, Resuscitation Councils of Southern Africa). *Circulation* 2004;**110**:3385–97.
25. *Vital Statistics of Japan 2005.* Tokyo: Health and welfare statistics association, 2011 (in Japanese) (23 June 2011). http://www.Stat.Go.Jp/data/kokusei/2005/index.Htm.
26. *Japanese Guidelines for Emergency Care and Cardiopulmonary Resuscitation.* Tokyo: Health Shuppansha; 2007.
27. Ministry of International Affairs and Communications. *Ambulance Service and Emergency Responses in 2010.* (in Japanese) (23 June 2011). http://www.Fdma.Go.Jp/neuter/topics/houdou/2212/221203_1houdou/01_houdoushiryou.Pdf).
28. Weisfeldt ML, Everson-Stewart S, Sitlani C, Rea T, Aufderheide TP, Atkins DL *et al.* Ventricular tachyarrhythmias after cardiac arrest in public versus at home. *N Engl J Med* 2011;**364**:313–21.
29. Moler FW, Donaldson AE, Meert K, Brilli RJ, Nadkarni V, Shaffner DH *et al.* Multicenter cohort study of out-of-hospital pediatric cardiac arrest. *Crit Care Med* 2011; **39**:141–9.

nature genetics

Common variants at *CDKAL1* and *KLF9* are associated with body mass index in east Asian populations

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Obesity is a disorder with a complex genetic etiology, and its epidemic is a worldwide problem. Although multiple genetic loci associated with body mass index, the most common measure of obesity, have been identified in European populations, few studies have focused on Asian populations. Here we report a genome-wide association study and replication studies with 62,245 east Asian subjects, which identified two new body mass index–associated loci in the *CDKAL1* locus at 6p22 (rs2206734, $P = 1.4 \times 10^{-11}$) and the *KLF9* locus at 9q21 (rs11142387, $P = 1.3 \times 10^{-9}$), as well as several previously reported loci (the *SEC16B, BDNF, FTO, MC4R* and *GIPR* loci, $P < 5.0 \times 10^{-8}$). We subsequently performed gene-gene interaction analyses and identified an interaction ($P = 2.0 \times 10^{-8}$) between a SNP in the *KLF9* locus (rs11142387) and one in the *MSTN* (also known as *GDF8*) locus at 2q32 (rs13034723). These findings should provide useful insights into the etiology of obesity.

Obesity is a major risk factor for a number of chronic diseases, and its recent rise in prevalence worldwide has imposed serious medical and economic burdens[1]. It is well known that obesity is a highly heritable trait, and around 40–70% of the inter-individual variation in obesity is attributable to genetic factors[2]. Recently, genome-wide association studies (GWAS) identified dozens of genetic loci associated with body mass index (BMI), the most common measure of obesity[3–12]. However, most of these studies were conducted in European populations, and few studies have assessed Asian populations[5,11], which account for two-thirds of the world's population. The degree of adiposity and the risks of diseases exacerbated by obesity are greater in Asians than in Europeans when evaluated with the same BMI[13]. Thus, the study of Asian populations might lead to the identification of new obesity-associated loci and provide insight into the genetic architecture of obesity. We report here a large-scale GWAS and replication studies of BMI examining a total of 62,245 subjects from east Asian populations.

In the GWAS for BMI, we enrolled 26,620 Japanese subjects under the support of the BioBank Japan Project[14] (**Supplementary Table 1** and **Supplementary Fig. 1**). We applied stringent quality control criteria, including principal component analysis (PCA) for evaluating potential population stratifications, as previously described[15]. To extend the coverage to the genomic region, we performed whole-genome imputation for SNPs, and we obtained the genotype data for 2,178,018 autosomal SNPs with minor allele frequency (MAF) ≥ 0.01. We evaluated each SNP for association with BMI using a linear regression model assuming additive effects of the allele dosages using the rank-based inverse-normal–transformed values of BMI. Although no significant population stratification was suggested in our study population (**Supplementary Fig. 2**) or in our previous studies of Japanese subjects[15], for robustness, we applied genomic-control corrections to the results of the GWAS using a genomic control inflation factor (λ_{GC}) of 1.123 (referenced $\lambda_{GC\_1,000} = 1.005$)[16]. The quantile-quantile plot of the resulting *P* values indicated a remarkable discrepancy in its tail from the null hypothesis (**Supplementary Fig. 3**), which suggested the presence of statistically significant associations in this GWAS. We identified significant associations in three chromosomal loci (the *KLF9* locus at 9q21, the *BDNF* locus at 11p14 and the *GIPR* locus at 19q13) that satisfied the genome-wide significance threshold of $P < 5.0 \times 10^{-8}$ (**Table 1** and **Fig. 1a**).

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Table 1 Associations of the GWAS and the replication studies for BMI

rs ID[a]	Chr.	Position[b]	Cytoband	Nearest gene	Class	A1/A2[c]	East Asian populations							European populations		
								GWAS		Replication study[f]		Combined			GIANT consortium[h]	
							Frequency[d]	β (SE)[e]	*P*	β (SE)[e]	*P*	β (SE)[e]	*P*	Explained variance[g]	β (SE)[e]	*P*
Significantly associated SNPs ($P < 5.0 \times 10^{-8}$)																
rs12149832	16	52,400,409	16q12	*FTO*	Intron	A/G	0.20	0.056 (0.011)	3.2×10^{-7}	0.090 (0.011)	5.1×10^{-17}	0.073 (0.008)	4.8×10^{-22}	0.20	0.077 (0.005)	5.6×10^{-58}
rs2030323	11	27,685,115	11p14	*BDNF*	Intron	C/A	0.60	0.054 (0.008)	1.3×10^{-9}	0.040 (0.008)	1.8×10^{-7}	0.046 (0.006)	3.8×10^{-16}	0.08	0.042 (0.006)	5.7×10^{-13}
rs11671664	19	50,864,118	19q13	*GIPR*	Intron	G/A	0.45	0.051 (0.008)	7.4×10^{-9}	0.041 (0.009)	5.6×10^{-6}	0.046 (0.006)	6.8×10^{-14}	0.08	0.029 (0.009)	0.0012
rs2206734	6	20,802,863	6p22	*CDKAL1*	Intron	C/T	0.59	0.043 (0.008)	8.3×10^{-7}	0.035 (0.008)	6.2×10^{-6}	0.039 (0.006)	1.4×10^{-11}	0.06	0.017 (0.006)	0.0049
rs2331841	18	55,979,617	18q21	*MC4R*	Intergenic	A/G	0.25	0.045 (0.011)	1.2×10^{-5}	0.047 (0.009)	1.9×10^{-7}	0.046 (0.007)	1.8×10^{-11}	0.08	0.035 (0.005)	1.2×10^{-13}
rs11142387	9	72,188,152	9q21	*KLF9*	Intergenic	C/A	0.46	0.048 (0.008)	4.6×10^{-8}	0.028 (0.011)	0.0084	0.040 (0.007)	1.3×10^{-9}	0.04	0.003 (0.005)	0.50
rs516636	1	176,122,140	1q25	*SEC16B*	Intergenic	A/C	0.22	0.053 (0.011)	4.2×10^{-7}	0.044 (0.014)	0.0014	0.050 (0.008)	3.4×10^{-9}	0.07	0.023 (0.027)	0.40
SNPs with suggestive associations ($5.0 \times 10^{-8} \leq P < 5.0 \times 10^{-5}$)																
rs4377469	3	42,278,078	3p22	*CCK*	Intron	T/G	0.69	0.050 (0.010)	6.4×10^{-7}	0.022 (0.012)	0.058	0.039 (0.007)	1.6×10^{-7}	–	0.018 (0.033)	0.58
rs10993160	9	96,108,747	9q22	*ZNF169*	Intergenic	A/G	0.83	0.061 (0.014)	7.9×10^{-6}	0.041 (0.016)	0.012	0.053 (0.011)	5.5×10^{-7}	–	0.025 (0.012)	0.035

Chr., chromosome.

[a]SNPs that satisfied $P < 5.0 \times 10^{-5}$ in the combined study are indicated. [b]Position in bp. [c]The allele that increased BMI is denoted as allele 1 (A1) and is indicated based on forward strand and NCBI Build 36. [d]Frequency of allele 1. [e]Effect size of allele 1 on the normalized BMI (mean, 0; s.d., 1). [f]Combined results of three independent replication sets (**Supplementary Fig. 1**). [g]Estimated variance (%) based on the effect sizes in the replication studies and the allele frequencies in HapMap east Asian populations. [h]Referenced using the results of the genome-wide meta-analysis for BMI in European populations[12].

To further validate the associations identified in the GWAS, we performed replication studies on three independent sets (**Supplementary Table 1** and **Supplementary Fig. 1**). The first and second sets consisted of 3,763 and 4,147 Japanese subjects from the BioBank Japan Project[14], respectively. The third set consisted of 27,715 subjects enrolled in the concurrently conducted meta-analysis of GWAS for BMI with cohorts of east Asians[17]. First, we evaluated the 36 SNPs most significantly associated in each of the loci having $P < 5.0 \times 10^{-5}$ in the GWAS in the first replication set and then further evaluated 11 SNPs with $P < 5.0 \times 10^{-5}$ in the combined study of the GWAS and replication set 1 in replication sets 2 and 3. Using the combined results of the GWAS and the replication studies, we identified a total of seven loci that satisfied the genome-wide significance threshold ($P < 5.0 \times 10^{-8}$; **Table 1**), which included the three loci that originally satisfied this threshold in the GWAS. Among the seven identified loci, five were previously identified to be associated with BMI in Europeans (the *SEC16B*, *BDNF*, *FTO*, *MC4R* and *GIPR* loci at 1q25, 11p14, 16q12, 18p21 and 19q13, respectively)[3–9,11,12]. The associations at the remaining two loci (6p22 and 9q21) had not been previously reported, including in the large-scale study of Europeans[12], and were new findings, to our knowledge. The landmark associated SNP at 6p22 (rs2206734, $P = 1.4 \times 10^{-11}$; **Fig. 1b**) is located in the coding region of *CDKAL1*, the gene encoding cyclin-dependent kinase 5 (CDK5) regulatory subunit associated protein 1-like 1. The other associated SNP at 9q21 (rs11142387, $P = 1.3 \times 10^{-9}$; **Fig. 1c**) is located in the promoter region of *KLF9*, the gene encoding Krüppel-like factor 9 (also known as basic transcription element-binding protein 1, or BTEB1). The linkage disequilibrium block that includes rs11142387 also covers several genes, including *MAMDC2*, *SMC5* and *TRPM3* (**Fig. 1c**). Although we examined tag copy number variants and performed an expression analysis of rs11142387 in the locus using a publicly available database, no significant findings were observed (**Supplementary Fig. 4**). Our study showed suggestive associations ($5.0 \times 10^{-8} \leq P < 5.0 \times 10^{-5}$) in the *CCK* (encoding cholecystokinin) locus at 3p22 (rs4377469, $P = 1.6 \times 10^{-7}$) and in the *ZNF169* (encoding zinc finger protein 169) locus at 9q22 (rs10993160, $P = 5.5 \times 10^{-7}$). The combination of these seven identified loci ($P < 5.0 \times 10^{-8}$) explained 0.72% of the inter-individual variance in BMI; the *FTO* locus explained the largest proportion of the inter-individual variance in BMI (0.20%; **Table 1**).

To evaluate ethnic differences in the genetics of obesity, we further evaluated the associations of the loci with confirmed or suggestive associations ($P < 5.0 \times 10^{-5}$) in the east Asian studies in Europeans using the results of a meta-analysis for 123,865 subjects performed by the GIANT consortium (**Table 1**)[12]. We found the same directional effects of the alleles in this European population as we found in the east Asian population at all of the nine evaluated loci. We observed significant associations in five loci ($P < 0.028$, false discovery rate (FDR) < 0.05), including the *CDKAL1* locus ($P = 0.0049$), but we saw no association in the *KLF9* locus ($P = 0.50$) in the European population.

Using our data, we then evaluated the associations of the previously reported BMI-associated loci, most of which had been identified in Europeans (**Supplementary Table 2**)[3–12,18]. Our study replicated the associations with the same directional effects of the alleles as the previous studies at ten loci, including the *TMEM18*, *DNAJC27* (also known as *RBJ*)-*ADCY3*-*POMC*, *GNPDA2*, *POC5* (also known as *FLJ35779*)-*HMGCR*, *TFAP2B*, *TRHR*, *MTCH2*, *MAP2K5*-*SKOR1* (also known as *LBXCOR1*), *SH2B1*-*ATP2A1* and *BMP2* loci, in addition to the five loci that were already replicated in our GWAS ($P < 0.02$, FDR < 0.05). In regards to loci previously reported in Koreans[11], we replicated the

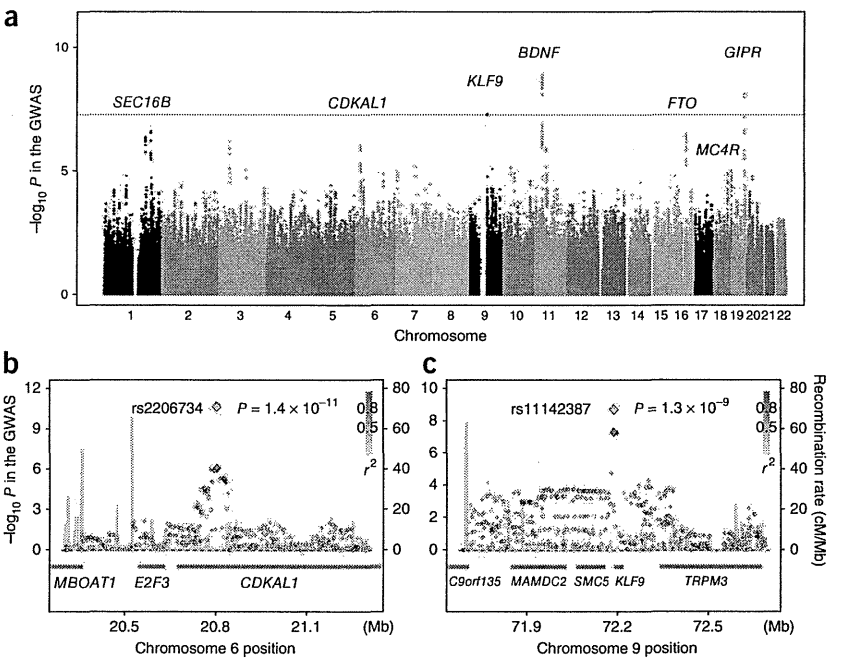

Figure 1 Results of the GWAS for BMI. (**a**) Manhattan plot showing $-\log_{10} P$ of the SNPs in the GWAS for BMI in 26,620 Japanese subjects. The genetic loci that satisfied the genome-wide significance threshold of $P < 5.0 \times 10^{-8}$ in the combined study of the GWAS and the replication studies are labeled. The gray horizontal line represents the significance threshold. (**b**,**c**) Regional plots of the SNPs in the *CDKAL1* locus (**b**) and in the *KLF9* locus (**c**). The red diamond-shaped dots represent the $-\log_{10} P$ values for the SNPs in the GWAS, and the green dots represent the *P* value for the most significantly associated SNP in each of the loci in the combined study. The intensity of the red color in the small-sized dots represents the r^2 value with the most significantly associated SNP, which is shown as a large-sized red dot. The blue lines show the recombination rates given in the HapMap Phase II east Asian populations (release 22). The genes listed below the plots indicate the RefSeq genes in the loci.

reported association in *FTO* but not *LOC729076* at 6q24 (**Supplementary Table 2**).

Because obesity is a polygenic trait and epistasis may help dissect its genetic background[19], we performed a gene-gene interaction analysis of BMI. We evaluated gene-gene interactions assuming additive × additive effects of SNPs[20] between each of the seven SNPs confirmed to be associated with BMI and all of the 2,178,018 genome-wide SNPs (**Supplementary Fig. 5**) using the subjects enrolled in the GWAS, and we then conducted a replication study. We found an interaction that satisfied the significance threshold ($P < 5.0 \times 10^{-8}$) between rs11142387 at *KLF9* and a SNP located in the promoter region of *MSTN* (myostatin, also known as growth and differentiation factor-8 or *GDF8*) at 2q32 (rs13034723, $P = 2.0 \times 10^{-8}$; **Table 2** and **Fig. 2a**). Notably, the association of rs13034723 at the *MSTN* locus with BMI was not significant ($P = 0.56$; **Supplementary Table 3**). When we stratified the association of rs11142387 by the genotypes of rs13034723, we observed a significant association of rs11142387 with BMI in the subjects with the AA genotype at rs13034723 ($P = 8.7 \times 10^{-15}$; **Supplementary Tables 4** and **5** and **Fig. 2b**), although we observed no significant association at this SNP for the subjects with the AG or GG genotype ($P = 0.0029$ and $P = 0.30$, respectively).

Through the GWAS and the replication studies, we identified two new loci, in *CDKAL1* and *KLF9*, associated with BMI in east Asians. These two loci also had significant associations with the risk of obesity (BMI ≥ 27.5; $P = 1.1 \times 10^{-5}$ for *CDKAL1* and $P = 4.2 \times 10^{-4}$ for *KLF9*; **Supplementary Table 6**). The association in the *CDKAL1* locus, but not the *KLF9* locus, was shared between east Asians and Europeans[12]. Because the study in Europeans[12] had enough power to detect the *KLF9* locus (>99%), under the assumption that this locus has the same effect size in Europeans as east Asians and given the allele frequency in Europeans (0.51), these results suggest that there exists ethnic heterogeneity in the effect of the *KLF9* locus for BMI. We also performed a gene-gene interaction analysis and showed an interaction between the *KLF9* and *MSTN* loci. Although the substantial role of epistasis in polygenic traits has been recognized, the approach to elucidate actual epistatic associations has been challenging[20]. Our findings are one of the first pieces of evidence for the presence of epistatic associations.

Recent studies reported associations of the *CDKAL1* locus with BMI at an age of 8 years (rs4712526)[21] and with birthweight (rs7756992)[22], and these two SNPs also had significant associations in our GWAS ($P < 5.0 \times 10^{-5}$). To our knowledge, our study is the first to report on the association with adult BMI. The *CDKAL1* locus has been reported to be a risk locus for type 2 diabetes (T2D)[23,24]. We found that the T allele of rs2206734, which was associated with a lower BMI, significantly increased T2D risk in our study subjects ($P < 1.4 \times 10^{-18}$; **Supplementary Table 6**). *CDKAL1* risk variants for T2D were previously associated with decreased insulin secretion[23]; therefore, the observed effects of the *CDKAL1* risk variant on lower BMI might be mediated by decreased insulin secretion. Notably, a recent study identified similar patterns of associations in the *GIPR* locus[25], and we observed that the A allele of rs11671664 at *GIPR*, which is also associated with lower BMI, increased T2D risk ($P < 1.5 \times 10^{-5}$; **Supplementary Table 6**). These findings suggest that further studies

Table 2 Gene-gene interaction for BMI between the *KLF9* and *MSTN* loci

Independent variables in the regression model				Associations with BMI in the regression model					
				GWAS		Replication study[c]		Combined	
rs ID	Cytoband	Gene	A1/A2[a]	β (SE)[b]	*P*	β (SE)[b]	*P*	β (SE)[b]	*P*
rs11142387[d]	9q21	*KLF9*	C/A	0.088 (0.012)	8.3×10^{-14}	0.117 (0.027)	1.7×10^{-5}	0.093 (0.011)	1.1×10^{-17}
rs13034723[d]	2q32	*MSTN*	A/G	−0.065 (0.015)	2.8×10^{-5}	−0.068 (0.029)	0.018	−0.065 (0.014)	1.5×10^{-6}
rs11142387 × rs13034723[e]	–	–	–	0.064 (0.013)	1.7×10^{-6}	0.073 (0.025)	0.0031	0.066 (0.012)	2.0×10^{-8}

[a]Based on the forward strand and NCBI Build 36. [b]Effect size of allele 1 (A1) on the normalized BMI (mean, 0; s.d., 1). [c]Consisted of replication sets 1 and 2 (**Supplementary Fig. 1**). [d]The allele dosage of allele 1 was used as an independent variable. [e]The product of the allele dosages of allele 1 of rs11142387 and allele 1 of rs13034723 was used as the independent variable.

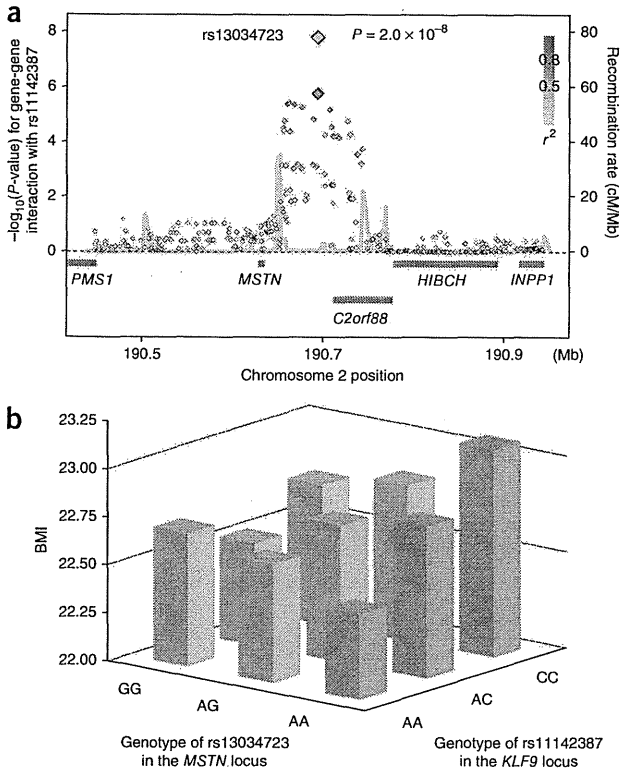

Figure 2 Gene-gene interactions between the *KLF9* and *MSTN* loci. (**a**) Regional plots of the SNPs. Diamond-shaped dots represent the $-\log_{10}$ *P* values for the SNPs for the gene-gene interaction with the landmark SNP in the *KLF9* locus (rs11142387). The green dot indicates the *P* value of the most significantly associated SNP in the combined study, and the red dot indicates the *P* value of this same SNP in the genome-wide gene-gene interaction analysis. The intensity of the red color in the small-sized dots represents the r^2 value with the most significantly associated SNP, which is shown as a large-sized red dot. The blue lines show the recombination rates given in HapMap. The genes listed below the plot indicate the RefSeq genes in the locus. (**b**) Mean BMI values of the subjects stratified by the genotypes of rs13034723 in the *MSTN* locus and rs11142387 in the *KLF9* locus.

that comprehensively assess genetic associations with T2D risk, BMI and insulin secretion should be performed. When we excluded the subjects affected with T2D (n = 12,234), the association of rs2206734 with BMI was not obvious (effect size = 0.031, $P = 1.4 \times 10^{-6}$). We evaluated the association of the *CDKAL1* and *GIPR* loci with other related traits, including systolic and diastolic blood pressure and serum lipid levels (total cholesterol, high-density–lipoprotein cholesterol, low-density–lipoprotein cholesterol and triglycerides), however, we observed no significant associations with any of these traits ($\alpha = 0.05$; **Supplementary Table 6**).

KLF9 is a member of zinc-finger transcription factors involved in various physiological processes. A recent study indicated *KLF9* as a pro-adipogenic transcription factor acting through the transactivation of PPARγ[26], a key component of adipocyte differentiation that has been implicated in obesity[27]. Researchers from a previous study[18] reported the association of the *KLF7* locus with obesity in the Danish population, although we could not test the relevance of this locus here because the SNP reported in that study, rs7568369, is not polymorphic (**Supplementary Table 2**). It is known that *KLF5*, a gene belonging to the *KLF* family and also known as *BTEB2*, regulates adipocyte differentiation[28]. Considering these observations, the association of the *KLF9* locus with BMI would be plausible from a biological aspect. Contrary to the results from the *CDKAL1* locus, we observed no significant associations of *KLF9* with T2D risk or with other related traits ($\alpha = 0.05$; **Supplementary Table 6**).

MSTN encodes a member of the transforming growth factor-β (TGF-β) superfamily that regulates mesenchymal stem cell proliferation[29]. A loss-of-function mutation in *MSTN* causes muscle hypertrophy and decreased body fat[29,30]. In our study, the SNP in the *MSTN* locus was not associated with BMI, but its genotypes clearly stratified the association in the *KLF9* locus. This would imply a regulatory role of *MSTN* on the effect of *KLF9* on BMI, and further studies evaluating functional epistasis should be performed. Notably, researchers from a previous study[31] identified similar conserved promoter and enhancer architecture in *KLF9* and *MSTN* through a search of evolutionary conserved regions, which suggests that these two genes may form a synexpression group[31]. Other genes located near *MSTN*, such as *C2orf88*, could also be candidates, and the relatively small sample size used in the replication studies provided limited evidence of this.

Researchers at concurrent study[17] have reported a genome-wide meta-analysis for BMI using data of eight cohorts of east Asians. The subjects enrolled in these two studies overlapped because of their reciprocal replication approaches, and newly identified loci were shared at *CDKAL1*, whereas some loci were specifically identified in each study, such as *KLF9*. These differences could be attributed to differences in study designs, effects of different compositions of the populations in the discovery phases and the probability of study-specific bias induced by the winner's curse effect.

In summary, our study identified new associations of the *CDKAL1* and *KLF9* loci with BMI in east Asians. We also found a gene-gene interaction between the *KLF9* and *MSTN* loci. Our study should contribute to understanding of the genetic architecture of obesity.

METHODS

Methods and any associated references are available in the online version of the paper at http://www.nature.com/naturegenetics/.

Note: Supplementary information is available on the Nature Genetics website.

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AUTHOR CONTRIBUTIONS

Y.O. and T. Tanaka designed the study and drafted the manuscript. N.H. and M.K. performed the genotyping. Y.O., H.O., A.T., N. Kumasaka and T. Tsunoda performed the statistical analyses. Y.O. and M.K. managed the clinical information. W.W., R.D., M.J.G., W.Z., N. Kato, J.-Y.W. and Q.L. managed replication study set 3. The GIANT consortium managed the association study in Europeans. S.M., K.Y., Y.N., N. Kamatani and T. Tanaka supervised the study.

COMPETING FINANCIAL INTERESTS

The authors declare no competing financial interests.

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1. Kopelman, P.G. Obesity as a medical problem. *Nature* **404**, 635–643 (2000).
2. Maes, H.H., Neale, M.C. & Eaves, L.J. Genetic and environmental factors in relative body weight and human adiposity. *Behav. Genet.* **27**, 325–351 (1997).

3. Frayling, T.M. *et al.* A common variant in the *FTO* gene is associated with body mass index and predisposes to childhood and adult obesity. *Science* **316**, 889–894 (2007).
4. Liu, Y.J. *et al.* Genome-wide association scans identified *CTNNBL1* as a novel gene for obesity. *Hum. Mol. Genet.* **17**, 1803–1813 (2008).
5. Chambers, J.C. *et al.* Common genetic variation near *MC4R* is associated with waist circumference and insulin resistance. *Nat. Genet.* **40**, 716–718 (2008).
6. Loos, R.J. *et al.* Common variants near *MC4R* are associated with fat mass, weight and risk of obesity. *Nat. Genet.* **40**, 768–775 (2008).
7. Thorleifsson, G. *et al.* Genome-wide association yields new sequence variants at seven loci that associate with measures of obesity. *Nat. Genet.* **41**, 18–24 (2009).
8. Willer, C.J. *et al.* Six new loci associated with body mass index highlight a neuronal influence on body weight regulation. *Nat. Genet.* **41**, 25–34 (2009).
9. Meyre, D. *et al.* Genome-wide association study for early-onset and morbid adult obesity identifies three new risk loci in European populations. *Nat. Genet.* **41**, 157–159 (2009).
10. Liu, X.G. *et al.* Genome-wide association and replication studies identified *TRHR* as an important gene for lean body mass. *Am. J. Hum. Genet.* **84**, 418–423 (2009).
11. Cho, Y.S. *et al.* A large-scale genome-wide association study of Asian populations uncovers genetic factors influencing eight quantitative traits. *Nat. Genet.* **41**, 527–534 (2009).
12. Speliotes, E.K. *et al.* Association analyses of 249,796 individuals reveal 18 new loci associated with body mass index. *Nat. Genet.* **42**, 937–948 (2010).
13. Deurenberg, P., Deurenberg-Yap, M. & Guricci, S. Asians are different from Caucasians and from each other in their body mass index/body fat per cent relationship. *Obes. Rev.* **3**, 141–146 (2002).
14. Nakamura, Y. The BioBank Japan Project. *Clin. Adv. Hematol. Oncol.* **5**, 696–697 (2007).
15. Okada, Y. *et al.* Genome-wide association study for C-reactive protein levels identified pleiotropic associations in the *IL6* locus. *Hum. Mol. Genet.* **20**, 1224–1231 (2011).
16. Freedman, M.L. *et al.* Assessing the impact of population stratification on genetic association studies. *Nat. Genet.* **36**, 388–393 (2004).
17. Wen, W. *et al.* Meta-analysis identifies common variants associated with body mass index in east Asians. *Nat. Genet.* Advance online publication (12 February 2012), doi:10.1038/ng.1086.
18. Zobel, D.P. *et al.* Variation in the gene encoding Kruppel-like factor 7 influences body fat: studies of 14,818 Danes. *Eur. J. Endocrinol.* **160**, 603–609 (2009).
19. Hinney, A., Vogel, C.I. & Hebebrand, J. From monogenic to polygenic obesity: recent advances. *Eur. Child Adolesc. Psychiatry* **19**, 297–310 (2010).
20. Cordell, H.J. Detecting gene-gene interactions that underlie human diseases. *Nat. Rev. Genet.* **10**, 392–404 (2009).
21. Winkler, C. *et al.* BMI at age 8 years is influenced by the type 2 diabetes susceptibility genes *HHEX-IDE* and *CDKAL1*. *Diabetes* **59**, 2063–2067 (2010).
22. Andersson, E.A. *et al.* Type 2 diabetes risk alleles near *ADCY5*, *CDKAL1* and *HHEX-IDE* are associated with reduced birthweight. *Diabetologia* **53**, 1908–1916 (2010).
23. Steinthorsdottir, V. *et al.* A variant in *CDKAL1* influences insulin response and risk of type 2 diabetes. *Nat. Genet.* **39**, 770–775 (2007).
24. Yamauchi, T. *et al.* A genome-wide association study in the Japanese population identifies susceptibility loci for type 2 diabetes at *UBE2E2* and *C2CD4A-C2CD4B*. *Nat. Genet.* **42**, 864–868 (2010).
25. Saxena, R. *et al.* Genetic variation in *GIPR* influences the glucose and insulin responses to an oral glucose challenge. *Nat. Genet.* **42**, 142–148 (2010).
26. Pei, H., Yao, Y., Yang, Y., Liao, K. & Wu, J.R. Kruppel-like factor KLF9 regulates PPARγ transactivation at the middle stage of adipogenesis. *Cell Death Differ.* **18**, 315–327 (2011).
27. Kadowaki, T. & Yamauchi, T. Adiponectin and adiponectin receptors. *Endocr. Rev.* **26**, 439–451 (2005).
28. Oishi, Y. *et al.* Kruppel-like transcription factor KLF5 is a key regulator of adipocyte differentiation. *Cell Metab.* **1**, 27–39 (2005).
29. Elkasrawy, M.N. & Hamrick, M.W. Myostatin (GDF-8) as a key factor linking muscle mass and bone structure. *J. Musculoskelet. Neuronal Interact.* **10**, 56–63 (2010).
30. Schuelke, M. *et al.* Myostatin mutation associated with gross muscle hypertrophy in a child. *N. Engl. J. Med.* **350**, 2682–2688 (2004).
31. Grade, C.V., Salerno, M.S., Schubert, F.R., Dietrich, S. & Alvares, L.E. An evolutionarily conserved Myostatin proximal promoter/enhancer confers basal levels of transcription and spatial specificity *in vivo*. *Dev. Genes Evol.* **219**, 497–508 (2009).