



R Wave Amplitude in V1 and its Association with Mortality in Cardiac Resynchronization Therapy

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ABSTRACT

Objective: In addition to QRS duration, a new marker is needed for selection of patients who will respond favorably to cardiac resynchronization therapy (CRT) in patients with non-left bundle branch block (LBBB) morphology. This study aimed to analyze the predictive ability of R-wave amplitude in V1 and compare its efficacy to previously suggested criteria in patients with non-LBBB morphology.

Material and Methods: We retrospectively included 433-patients with heart failure (HF) diagnosis, QRS \geq 120 ms, NYHA II-IV, LVEF $<$ 35% and previous CRT implantation. Patients were divided into three-groups as patients with LBBB (Group-I), patients with right-bundle branch block (Group-II) and patients with nonspecific-intraventricular conduction delay (Group-III).

Results: The R-wave amplitude in V1, presence of R $>$ S in V1 and RV1S1 were higher in Group-I than in the other two-groups (p $<$ 0.05, for all). R-wave amplitude in V1, presence of R $>$ S in V1, RV1S1, R $<$ S in D1-aVL, QS in V5-V6-D1 were lower in patients with mortality (p $<$ 0.05, for all). Only R-wave amplitude in V1 was found to be independently associated with mortality in logistic regression analysis (p $<$ 0.001, OR= 0.575). Every 1-mV decrease in R-wave amplitude in V1 was associated with 42.5% increase in the risk of mortality. The cut-off value of R-wave amplitude in V1 obtained by ROC curve analysis was 2.5 mV for prediction of mortality (sensitivity= 81.5%, specificity= 81.8%).

Conclusion: R-wave amplitude in V1 is negatively and independently associated with mortality. Strong predictive ability of the R-wave amplitude in V1 gives the operator the chance to intraoperatively improve prognosis by orienting the implantation process according to the biggest possible R-wave in coronary sinus (CS) branches.

Keywords: Cardiac resynchronization therapy, R wave amplitude in V1, right bundle branch block

ÖZ

V1'deki R Dalga Amplitüdü ve Kardiyak Resenkronizasyon Tedavisinde Mortalite ile İlişkisi

Giriş: LBBB morfolojisine sahip olmayan hastalarda kardiyak resenkronizasyon tedavisi (KRT)'ye olumlu yanıt verecek hastaların seçimi için QRS süresine ek olarak yeni bir belirteç gereklidir. V1'de R-dalga amplitüdünün prognostik değerini değerlendirmeyi ve bunun LBBB morfolojisine sahip olmayan hastalardaki etkinliğini daha önce önerilen kriterlerle karşılaştırmayı amaçladık.

Gereç ve Yöntemler: Bu retrospektif çalışmaya kalp yetmezliği (KY) tanısı olup, QRS \geq 120 ms, NYHA II-IV, LVEF $<$ %35 ve KRT implantasyonu yapılmış olan 433 hastayı dahil ettik. Hastalar LBBB olan hastalar (Grup-I), RBBB olan hastalar (Grup-II) ve non-spesifik intraventriküler ileti gecikmesi olan hastalar (Grup-III) olmak üzere üç gruba ayrıldı.

Bulgular: V1 derivasyonunda R dalga amplitüdü ve R $>$ S varlığı ile RV1S1 varlığının Grup-I hastalarda diğer iki gruba göre yüksek olduğu belirlendi (p $<$ 0.01 her biri için). Mortalitesi olan hastalarda V1'de R dalga amplitüdü ve R $>$ S varlığı, RV1S1 varlığı, D1-aVL'de R $<$ S varlığı ile V5-V6-D1'de QS varlığının daha az olduğu belirlendi (p $<$ 0.05 her biri için). Lojistik regresyon analizinde sadece V1'deki R dalga amplitüdünün mortalite ile bağımsız olarak ilişkili olduğu bulundu (p $<$ 0.001, OR= 0.575). V1'de R dalgası amplitüdündeki her 1 mV azalma, mortalite riskinde % 42.5 artışla ilişkilendirildi. ROC analizinde, V1'deki R dalga amplitüd sınırı değeri 2.5 mm olarak alındığında %81.5 duyarlılık ve %81.8 özgüllükle mortaliteyi öngördüğü saptandı.

Sonuç: V1'deki R dalga amplitüdü negatif yönde ve bağımsız olarak mortalite ile ilişkilidir. V1'deki R dalga amplitüdünün güçlü prediktif yeteneği; implantasyon esnasında operatörü CS dallarındaki mümkün olan en büyük R-dalgasına göre yönlendirerek prognozu intraoperatif olarak yükseltme şansı verir.

Anahtar Kelimeler: Kardiyak resenkronizasyon tedavisi, V1 R dalga amplitüdü, sağ dal bloğu

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INTRODUCTION

The primary goals of cardiac resynchronization therapy (CRT) such as symptomatic relief, reduction in mortality and morbidity cannot be achievable in every patient. QRS morphology and duration are basically the most important parameters that determine patients who will benefit from CRT (1). As a matter of fact, according to the latest published data and guidelines, CRT is not recommended in patients with QRS duration < 130 ms (1-4). Several studies have shown that patients with left bundle branch block (LBBB) morphology are more likely to respond favorably to CRT, whereas there is less certainty about patients with non-LBBB morphology (1,2,5,6).

Electrocardiography (ECG) is a useful tool for evaluating biventricular stimulation. In ECG, QRS morphology in some leads can serve as a guide for the presence and location of LV, RV capture. Some ECG criteria such as $R > S$ in V1 (7-9), $R < S$ in D1 and aVL (10), presence of RV1S1 ($R > S$ in V1 + $R < S$ in D1) (7), QS in V5-V6-D1-aVL (11,12) and shortening of QRS duration (13) have been previously reported to be indicative of favorable response to CRT and effective LV stimulation in patients with baseline LBBB morphology.

There is not enough information about the effectiveness of CRT treatment in patients with non-LBBB morphology. According to the limited number of studies conducted, CRT is known to be less effective in patients without LBBB (14-20). However, according to the latest guidelines, CRT is recommended in patients with QRS duration ≥ 150 ms in patients with non-LBBB (1,14). Therefore, in addition to QRS duration, a new marker is needed for selection of patients who will respond favorably to CRT in patients with baseline non-LBBB morphology. To the best of our knowledge, this is the first study to investigate ECG changes and their clinical implications after CRT implantation in patients with baseline non-LBBB morphology (right bundle branch block [RBBB] and nonspecific intraventricular conduction delay [NICD]).

In our study, it was aimed to analyze the predictive ability of R wave amplitude in V1 and compare its effectiveness to previously suggested criteria in patients with baseline non-LBBB morphology.

MATERIALS and METHODS

Study Protocol and Study Population

A total of 498 patients with HF diagnosis, QRS ≥ 120 ms, NYHA II-IV, LVEF < 35% and previous CRT implantation were the initial candidates for the present study. Patients in the pediatric age group (<18), patients without appropriate ECG for evaluation, patients with known dysfunction in leads,

electrolyte abnormality, pulmonary embolism, thyroid dysfunction, active malignancy, severe kidney and liver dysfunction and patients with previous AV node ablation for AF rate control were excluded. Of the 498 patients, 65 were excluded from the final analysis because of having one of the exclusion criteria. Therefore, we retrospectively included 433 patients with HF diagnosis, QRS ≥ 120 ms, NYHA II-IV, LVEF < 35% and previous CRT implantation (256 males, 177 females, mean age 64.1 ± 7.57 years). Patients were divided into 3 groups as patients with LBBB (Group-I), patients with RBBB (Group-II) and patients with NICD (Group-III). The NICD was defined as 'wide QRS without left or right bundle block' (21). The Local Ethics Committee approved the study protocol (Adana City Hospital Local Ethics Committee, 28.08.2019/534), and each participant provided written informed consent.

Detailed medical history and a complete physical examination, the baseline characteristics of patients including age, sex, hypertension, diabetes, hyperlipidemia, smoking status, family history of cardiac disease and medications were obtained from medical records. ECG, telecardiography, complete blood count, fasting blood glucose, uric acid, high sensitive C reactive protein, high sensitive cardiac troponin I, NT-proBNP, serum electrolytes, serum lipids, renal and liver function tests were obtained from medical records. All patients were followed up 370 to 940 (mean 670 ± 242) days for cardiac death after hospital discharge.

Echocardiographic assessment was made by using a 2.5-3.5 MHz transducer (Philips HD11 ultrasound system, Bothell, USA) with parasternal long and short axis, apical two and four chamber views. Baseline transthoracic echocardiography measurements were performed before CRT implantation. LV end-diastolic dimension (onset of the Q wave of the electrocardiogram) and LV end-systolic dimensions were measured from M-mode recordings. LV end-systolic volume, LV end-diastolic volume, and ejection fraction (LVEF) were assessed using Simpson's equation using the apical 4-chamber view.

12-lead Electrocardiography

The 12-lead ECG, which was recorded prior to discharge after CRT implantation, was obtained from medical records and used for evaluation. MAC 2000 ECG Machine (GE Medical, Milwaukee, WI, USA) was used for all ECG procedures. ECG recordings of all patients were performed at 25 mm/sec speed and 10 mm/mV amplitude. In addition to R wave amplitude in V1, several ECG criteria from previous studies such as i) $R > S$ in V1, ii) $R < S$ in D1, iii) presence of RV1S1, iv) $R < S$ in aVL, v) QS in V5, vi) QS in V6, vii) QS in D1, viii) QS in aVL and ix) QRS duration were also evaluated in terms of

CRT response and prognosis (Figure 1-4). All ECG parameters were evaluated by at least two experienced electrophysiology specialists.

Statistical Analysis

Statistical analyses were conducted using SPSS, version 22.0, (SPSS Inc. Chicago, Illinois). Data were expressed

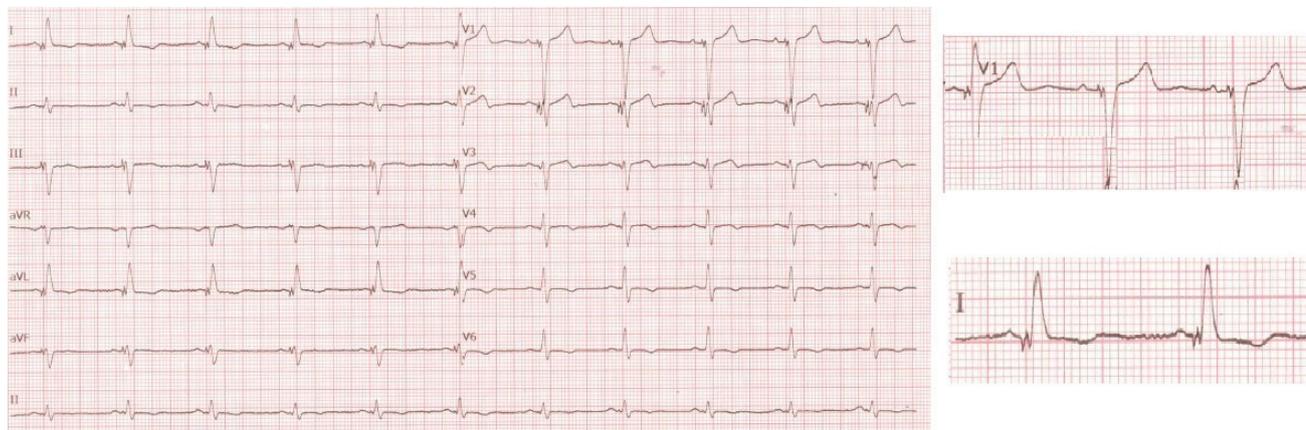


Figure 1. Ineffective biventricular pacing findings after CRT implantation in a patient with HFrEF and NICD. QRS duration 128 msn, r wave amplitude 1 mm in lead V1, so there is no prominent R wave ($R > S$) in V1 lead. Also, no obvious S wave ($S > R$) is seen in DI lead.



Figure 2. Ineffective biventricular pacing findings after CRT implantation in a patient with HFrEF and RBBB. QRS duration 132 msn, r wave amplitude 2.5 mm in lead V1, so there is no prominent R wave ($R > S$) in V1 lead. Also, no obvious S wave ($S > R$) is seen in DI lead.

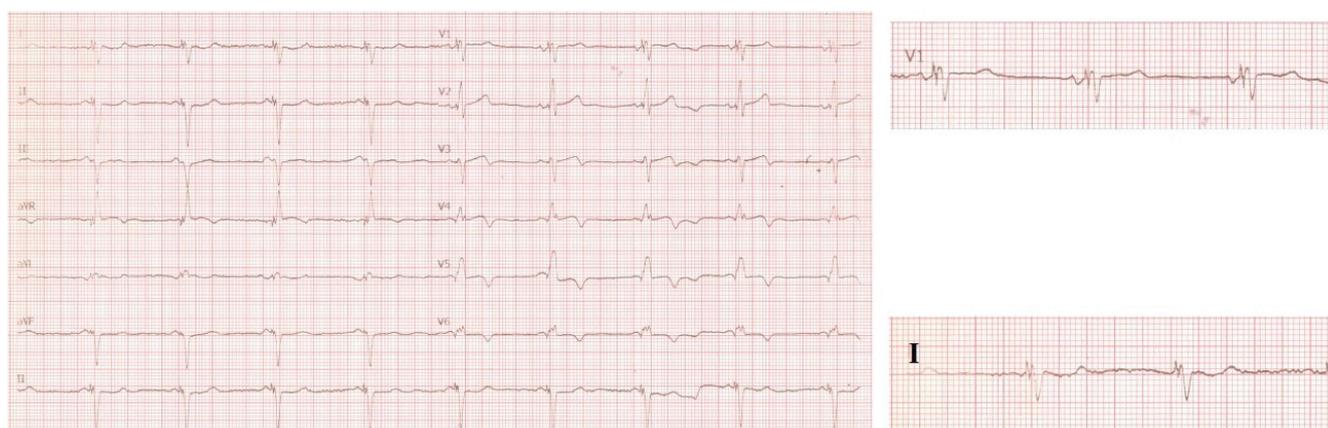


Figure 3. Relatively effective biventricular pace findings after CRT implantation in a patient with HFrEF and LBBB. QRS duration 118 msn, r wave amplitude 1.5 mm in lead V1, so there is no prominent R wave ($R > S$) in V1 lead. However, there is a prominent S wave ($S > R$) in the DI lead.

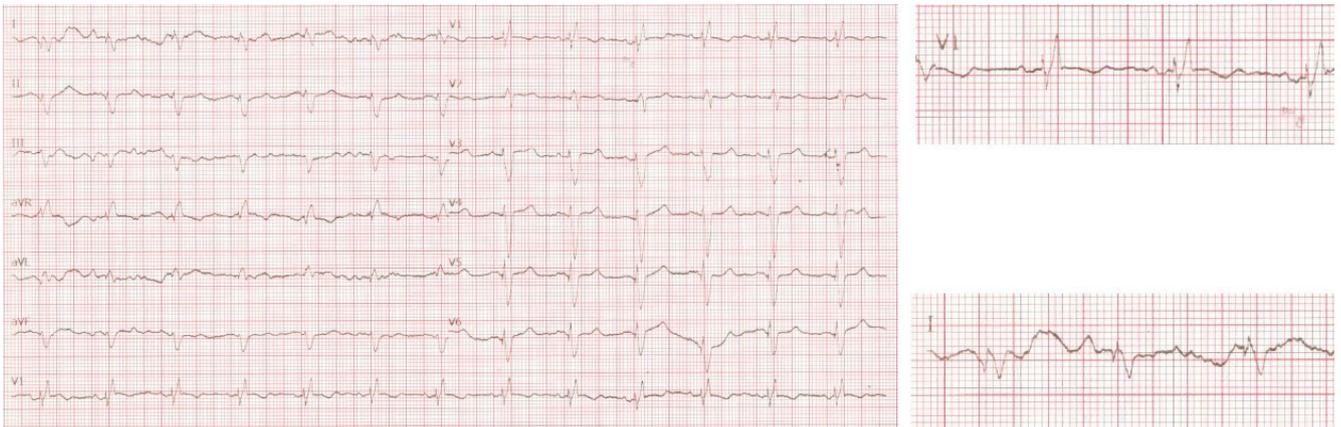


Figure 4. Effective biventricular pace findings after CRT implantation in a patient with HFrEF and RBBB. QRS duration 124 msn, R wave amplitude is 5 mm therefore there is a prominent R wave ($R > S$) in V1 lead and there is a prominent S wave ($S > R$) in DI lead.

as mean \pm SD for continuous variables and percentage for categorical variables. The Shapiro-Wilk test was used to test normality and a $p > 0.05$ was defined as normally distributed data. Continuous variables that showed normal distribution were compared using the Student's t test and ANOVA, whereas the Mann-Whitney U test and Kruskal-Wallis test were used for nonnormally distributed samples. Categorical variables and frequencies were compared by means of the chi-square test. Statistical significance was defined as a $p < 0.05$ for all comparisons. Pearson's and Spearman's correlations were used to examine the relationship between continuous variables. To decrease the possibility of type I error, Bonferroni correction was used after tests with multiple comparisons, such as one-way ANOVA.

The factors associated with cardiovascular mortality were tested by univariate and multivariate analyses. Variables with a $p < 0.05$ in the univariate and bivariate analysis were tested in the multivariate model. Results were expressed as the p value and odds ratio (OR) in CI of 95%. ROC analysis was made to determine the cut-off value of R wave amplitude in V1 to predict cardiovascular mortality. Statistical significance p value was defined as < 0.05 for all comparisons.

RESULTS

Previously suggested ECG parameters to predict CRT response were successfully measured in all patients included in the study. Cohen kappa values that evaluate interobserver and intra-observer variability were over 90% for all ECG criteria.

All demographic, clinical and laboratory data were similar between the groups. Although cardiovascular mortality was higher in Group-II and Group-III patients, this difference was not statistically significant (Table 1).

The presence of $R > S$ in V1 and RV1S1 were found to be less frequent in Group-II and Group-III patients than in Group-I patients (Table 2). When ECG parameters of patients with and without mortality are compared; R wave amplitude in V1, the presence of $R > S$ in V1, $R < S$ in D1, RV1S1, $R < S$ in aVL, QS in V5, QS in V6 and QS in D1 were found to be lower in patients with mortality (Table 3). Logistic regression analysis was done to determine independent parameters to predict mortality. As a result of this analysis, only R wave amplitude in V1 was found to be independently associated with mortality ($p < 0.001$, OR= 0.575 and 95% CI= 0.478-0.693). Every 1 mV decrease in R wave amplitude in V1 was found to be associated with 42.5% increase in the risk of mortality.

The cut-off value of R wave amplitude in V1 obtained by ROC curve analysis was 2.5 mV for prediction of mortality (sensitivity: 81.5%, specificity: 81.8%). The area under the curve (AUC) was 0.754 (95% CI= 0.685 - 0.799) ($p < 0.001$) (Figure 5).

DISCUSSION

To the best of our knowledge, this is the first study to investigate ECG changes and their clinical implications after CRT implantation in patients with baseline non-LBBB morphology. Our study made several significant contributions to the literature in terms of CRT treatment to patients with non-LBBB baseline ECG morphology. First, R wave amplitude in V1 and the presence of $R > S$ in V1, which are the most important ECG parameters for CRT response, were found to be lower in patients with non LBBB baseline morphology. Another important finding is that only R wave amplitude in V1 was found to predict mortality. Every 1 mV decrease in R wave amplitude in V1 was found to be associated with 42.5% increase in the risk of mortality. When the cut-off value was taken as 2.5 mV, R wave amplitude in V1 was found to pre-

Table 1. Clinical, demographic, laboratory and medical treatment findings according to study groups

Variable	Group I n= 328	Group II n= 52	Group III n= 53	p
Age (year)	64.6 ± 8.3	62.1 ± 9.1	63.2 ± 8.4	0.165
Gender (female)	132	20	25	0.553
Hypertension, n (%)	182 (56%)	27 (52%)	34 (64%)	0.524
Diabetes mellitus, n (%)	128 (39%)	26 (50%)	25 (47%)	0.132
Current smoker, n (%)	39 (12%)	7 (14%)	10 (19%)	0.096
Coronary artery disease, n (%)	205 (63%)	29 (56%)	30 (57%)	0.875
Systolic blood pressure (mmHg)	122 ± 17	121 ± 15	125 ± 16	0.382
Diastolic blood pressure (mmHg)	78 ± 11	75 ± 9.1	78 ± 8.8	0.158
Basal heart rate (pulse/minute)	78 ± 12	80 ± 11	79 ± 10	0.142
Body mass index (kg/m ²)	26.3 ± 3.8	25.6 ± 3.8	26.3 ± 3.4	0.517
White blood cell (µl)	8.43 ± 2.28	8.76 ± 2.73	8.23 ± 2.68	0.512
Hemoglobin (g/dl)	12.7 ± 1.7	12.5 ± 1.6	13.0 ± 1.8	0.400
Blood urea nitrogen (mg/dl)	54.5 ± 32	57 ± 30	50 ± 21	0.258
Creatinine (mg/dl)	1.29 ± 1.19	1.24 ± 1.47	1.21 ± 1.10	0.346
hs-CRP (mg/L)	2.24 ± 2.89	2.38 ± 2.77	2.41 ± 2.22	0.656
Total cholesterol (mg/dl)	171 ± 45	168 ± 59	174 ± 35	0.844
LDL cholesterol (mg/dl)	109 ± 38	105 ± 42	113 ± 34	0.546
HDL cholesterol (mg/dl)	39.5 ± 14	40.1 ± 13	38.0 ± 12	0.709
Triglycerides (mg/dl)	153 ± 84	163 ± 90	167 ± 99	0.461
Uric aside (mg/dl)	6.88 ± 2.15	6.99 ± 2.41	7.23 ± 2.23	0.539
NT-proBNP (pg/ml)	1059 ± 533	999 ± 404	964 ± 355	0.341
hs-cTnI (ng/L)	0.103 ± 0.335	0.089 ± 0.237	0.082 ± 0.168	0.881
LV end-diastolic dimension (mm)	59.5 ± 9.9	61.6 ± 8.8	60.1 ± 9.2	0.370
LV end-systolic dimension (mm)	51.8 ± 10	53.5 ± 9.4	52.1 ± 9.8	0.606
LV ejection fraction (%)	25.1 ± 5.9	25.5 ± 6.2	25.9 ± 6.7	0.641
Cardiovascular mortality, n (%)	44 (13%)	12 (23%)	10 (19%)	0.077

HDL: High-density lipoprotein, hs-CRP: High sensitive C reactive protein, hs-cTnI: High sensitive cardiac troponin I, LDL: Low-density lipoprotein, LV: Left ventricle, NT-proBNP: N-terminal pro-brain natriuretic peptide. Group I: Left bundle branch block group, Group II: Right bundle branch block group, Group III: Nonspecific intraventricular conduction delay group.

dict mortality with a sensitivity of 81.5% and a specificity of 81.8%.

In the evaluation of biventricular stimulation, QRS morphology in V1 is the basic assessment. In patients with baseline LBBB, the presence of R > S in V1 is observed in 78-93% of the patients after CRT implantation (7-9). In addition, the R/S ratio ≥ 1 in V1 indicates that there is LV or biventricular stimulation (10). In our study, similar to previous studies, 81% of the patients with LBBB had R > S in V1 after CRT implantation. R/S ratio in D1 is another criterion to be evaluated. R/S ratio < 1 in D1 supports biventricular stimulation (10). In a study conducted by Coverstone E. et al. (7), the RV1S1 (R > S in V1

lead + R < S in D1 lead) has been reported to be 60.7% in patients with LBBB. In our study, the frequency of RV1S1 in patients with LBBB was 64%, consistent with the previous study. Similarly, in other studies, QS wave in D1 has been reported to be a strong finding for biventricular stimulation (11,12). In previous studies, the incidence of QS wave in D1 and aVL in patients with biventricular stimulation has been reported to be 71-85%, respectively (10-12). In our study, it was shown that the frequency of R/S ratio of < 1 in D1 was 72% in accordance with previous literature. Similarly, the incidence of QS wave in D1 and aVL was 74%. Although some studies have shown a decrease in the mean QRS duration by 20-40 milli-

Table 2. Electrocardiographic findings according to study groups

Variable	Group I n= 328	Group II n= 52	Group III n= 53	p
R > S in V1, n (%)	267 (81%) ^α	29 (56%) ^β	31 (59%) ^β	<0.001
R < S in D1, n (%)	235 (72%)	43 (83%)	40 (76%)	0.173
R > S in V1 + R < S in D1, n (%)	211 (64%) ^α	27 (52%) ^β	23 (43%) ^β	0.002
R wave in V1 (mm)	3.78 ± 1.63 ^α	3.60 ± 1.66	3.06 ± 1.47	0.010
R < S in aVL, n (%)	242 (74%)	42 (81%)	38 (72%)	0.734
QS wave in V5, n (%)	203 (62%)	37 (71%)	31 (59%)	0.735
QS wave in V6, n (%)	231 (70%)	41 (79%)	35 (66%)	0.857
QS wave in D1, n (%)	242 (74%)	39 (75%)	39 (74%)	0.937
QS wave in aVL, n (%)	241 (74%)	44 (85%)	34 (64%)	0.869
QRS time (msn)	140 ± 8.8	141 ± 9.1	142 ± 9.3	0.884

Group I: Left bundle branch block group, Group II: Right bundle branch block group, Group III: Nonspecific intraventricular conduction delay group.

α: The significant association between the Group I and Group III (p < 0.05).

β: The significant association between the Group I and Group II (p < 0.05).

γ: The significant association between the Group II and Group III (p < 0.05).

Table 3. Electrocardiographic findings of patients with and without cardiovascular mortality

Variable	Mortality (+) n= 66	Mortality (-) n= 367	p
R > S in V1, n (%)	33 (50%)	294 (80%)	<0.001
R < S in D1, n (%)	37 (56%)	281 (77%)	0.001
R > S in V1 + R < S in D1, n (%)	22 (33%)	239 (65%)	<0.001
R wave in V1 (mm)	2.53 ± 1.25	3.88 ± 1.60	<0.001
R < S in aVL, n (%)	39 (59%)	283 (77%)	0.002
QS wave in V5, n (%)	27 (41%)	244 (67%)	<0.001
QS wave in V6, n (%)	37 (56%)	270 (74%)	0.004
QS wave in D1, n (%)	37 (56%)	283 (77%)	<0.001
QS wave in aVL, n (%)	43 (65%)	276 (75%)	0.062
QRS time (msn)	147 ± 7.4	146 ± 12	0.630

The values were shown as mean ± standard deviation or n (%).

seconds with biventricular stimulation, this effect cannot be observed in every patient (13). In addition, although some studies have shown a relationship between the degree of QRS shortening and clinical response, this relationship has not been established in some other studies (13). There is also information in the opposite direction that the post-implantation QRS duration does not have prognostic significance (22). In our study, it was shown that QRS durations after CRT implantation were similar in patients with and without LBBB. In addition, QRS durations after implantation were similar in patients with and without mortality.

Previous CRT studies have mostly included patients with LBBB. In our study, we evaluated the patients with RBBB and non-specific IVCD which are the most common forms non-

LBBB morphologies in clinical practice. Data on ECG findings of biventricular stimulation in patients with RBBB are quite limited. In our study, ECG changes previously evaluated in patients with LBBB were tested in patients with non-LBBB. We found that the presence of R < S in leads D1-aVL, the presence of QS wave in the leads V5-V6-D1-aVL, and QRS duration were similar between the LBBB and non-LBBB groups. R wave amplitude in V1, the presence of R > S in V1 and RV1S1 were significantly lower in patients with non-LBBB baseline morphology. In a study with 213 patients, similar to our study, percentage of the patients with RV1S1 sign has been found to be lower in patients with RBBB baseline morphology (7). It has been suggested that less frequent RV1S1 sign in patients with RBBB may be associated with poor CRT response and prognosis (7).

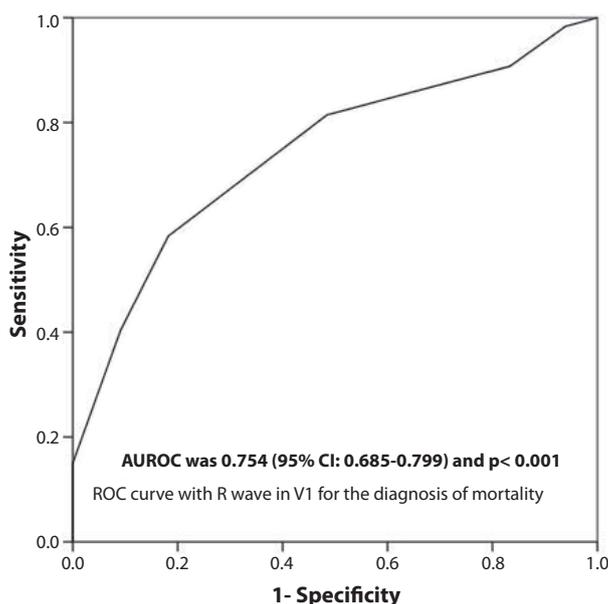


Figure 5. ROC analysis of R wave amplitude in V1 in predicting the presence of mortality after the CRT implantation for patients with heart failure with reduced ejection fraction.

Previous studies have demonstrated mortality and morbidity benefit in patients who underwent CRT treatment with appropriate indication. In our study, long term mortality was 15.1%. Patients with non-LBBB baseline morphology was included in a small number of CRT studies, which investigated the prognostic effects of CRT rather than ECG findings of biventricular stimulation. In these studies, it was reported that CRT treatment had a positive effect on prognosis in patients with non-LBBB morphology, although not as much as the patients with LBBB morphology. Therefore, although patients with non-LBBB do not benefit from CRT as much as patients with LBBB, it is recommended to perform CRT according to certain clinical, electrocardiographic and imaging criteria, especially in selected patients with QRS duration \geq 150 ms (1,2,14-20).

In our study, the effects of ECG changes of CRT treatment on mortality were also evaluated, and it was shown that only low R wave amplitude in V1 was independent predictor of mortality. Every 1 mV decrease in R wave amplitude in V1 was found to be associated with 42.5% increase in the risk of mortality. In the study of Coverstone E. et al (7), it has been reported that the absence of RV1S1 is associated with unpredictable hospitalization. In the same study, however, this parameter has not been found to be related with mortality (7). In our study, the presence of RV1S1 was higher in patients with mortality, but it was not an independent predictor. In

our study, we found a cut-off value of > 2.5 mV for R wave amplitude in V1 to predict good response to CRT treatment and post-implantation prognosis. It was concluded that there is need for larger trials including more patients with non-LBBB morphology to determine the prognostic effects of $R > S$ in V1, R wave amplitude in V1 and RV1S1 after CRT implantation.

There were some limitations in our study. As a single-center study, our patient cohort might be different from that in other centers. Another important limitation was the assessment of the ECG before discharge. There may be a change in ECG morphology in patients with CRT over time. Since the main purpose in our study was to detect ECG changes after CRT implantation, we only evaluated the relationship between these changes and mortality. We did not evaluate other clinical, laboratory and echocardiographic parameters in terms of predicting mortality. There was no a standard lead type, similar localization of lead and similar technique.

CONCLUSION

After CRT implantation, R wave amplitude in V1 is negatively and independently associated with mortality. Strong predictive ability of the R wave amplitude in V1 gives the operator the chance to intraoperatively improve CRT response and prognosis by orienting the implantation process according to the best possible R wave in CS branches.

Ethics Committee Approval: The study was approved from adana City Hospital Local Ethics Committee (Date: 28.08.2019, Decision No: 19/534).

Author Contributions: Concept/Design: HK; Analysis/Interpretation: HK; Data Acquisition: HK; Writting: HK; Critical Revision: HK; Final Approval: HK.

Conflict of Interest: There is no conflict of interest.

Financial Disclosure: The authors declared that this study has received no financial support.

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