Introduction and objectives. In patients with systolic dysfunction, different ventricular filling patterns are associated with different prognoses. The load changes resulting from nitroprusside infusion or long-term therapy for heart failure induce alterations in filling pattern that have been shown to serve as outcome markers. Our aim was to investigate the prognostic value of the Doppler-detected change in pseudonormal or restrictive left ventricular filling pattern induced by the Valsalva maneuver in patients with systolic dysfunction.

Material and method. The study included 36 patients in sinus rhythm with a depressed ejection fraction and an E/A ratio greater than 1. Filling velocities were recorded before and after 3 Valsalva maneuvers.

Results. The E/A ratio remained greater than 1 in 12 patients (group A); in 24 patients (group B), it fell below 1, indicating an abnormal relaxation pattern (i.e., a reversible pattern). During a mean follow-up period of 18 months, 8 patients died: 6 in group A (50%) and 2 in group B (8.3%; \( P = .005 \)). Moreover, 12 patients either died or suffered severe heart failure: 8 in group A (67%) and 4 in group B (17%; \( P = .003 \)). A reversible filling pattern was associated with lower risks of death (hazard ratio [HR]=0.06; 95% confidence interval [CI], 0.01-0.48) and of hospitalization or death (HR=0.11; 95% CI, 0.03-0.43).

Conclusions. Change of a pseudonormal or restrictive left ventricular filling pattern into an abnormal relaxation pattern after the Valsalva maneuver in patients with systolic dysfunction predicts a lower risk of death or severe heart failure.

Key words: Heart failure. Echocardiography. Prognosis.

Valor pronóstico de los cambios inducidos por la maniobra de Valsalva en el llenado ventricular registrado con Doppler en pacientes con disfunción sistólica

Introducción y objetivos. Los patrones de llenado ventricular registrados con Doppler tienen valor pronóstico en la disfunción sistólica y también los cambios que sufren al modificar la carga mediante tratamiento crónico o nitroprusiato. Nuestro propósito fue investigar el valor pronóstico de los cambios inducidos por la maniobra de Valsalva sobre los patrones restrictivo y seudonormal del llenado ventricular, registrados con Doppler en pacientes con disfunción sistólica.

Material y método. Se estudió a 36 pacientes en ritmo sinusal, disfunción sistólica y cociente entre las ondas E y A > 1, en situación clínicamente estable. Las velocidades de llenado se registraron antes y después de tres maniobras de Valsalva parcialmente estandarizadas.

Resultados. El cociente E/A se mantuvo mayor de 1 en 12 pacientes (grupo irreversible) y se transformó en uno de relajación anormal (E/A menor de 1) en 24 (grupo reversible). Durante un seguimiento medio de 18 meses fallecieron 8 pacientes, 6 en grupo irreversible (50%) y 2 en grupo reversible (8,3%; \( P = 0,005 \)). Doce murieron o presentaron insuficiencia cardíaca severa, 8 en el grupo irreversible (67%) y 4 en el grupo reversible (17%; \( P = 0,003 \)). La reversibilidad se asoció a un menor riesgo tanto de defunción como de defunción y hospitalización por insuficiencia cardíaca (hazard ratio [HR] = 0,11; intervalo de confianza [IC] del 95%, 0,03-0,43, y HR = 0,06; IC del 95%, 0,01-0,48, respectivamente).

Conclusiones. La transformación de un patrón de llenado seudonormal o restrictivo en otro de relajación anormal tras la maniobra de Valsalva predice un mejor pronóstico en la disfunción sistólica.

Palabras clave: Insuficiencia cardíaca. Ecocardiografía. Pronóstico.
INTRODUCTION

Analysis of left ventricular diastolic function has conclusively proven to be useful as a tool to separate patients with heart failure due to systolic dysfunction into different prognostic groups. Among the various diastolic parameters used for this purpose, analysis of the E wave to A wave (E/A) ratio and transmitral valve filling by Doppler study offers highly relevant information. As compared to patients with an abnormal relaxation pattern, patients with restrictive or pseudo-normal filling patterns show poorer evolution. These two patterns are related with clinical events in patients with heart failure, and with inadequate remodeling and worse outcome in patients who have undergone coronary revascularization surgery.

Transmitral filling velocities are determined by several dynamic phenomena that lead to the variability of this parameter. Preload and afterload changes affect the filling pattern in different ways in the various patient groups. This fact, which is often considered a limitation for applying the technique, has high clinical value. Induction of certain changes in the filling pattern by means of controlled manipulation of ventricular loading provides added value to the Doppler prognostic study of heart failure. Pozzoli et al demonstrated that patients with systolic dysfunction and a restrictive filling pattern had a better prognosis when the filling pattern changed to abnormal relaxation after modifying loading with the use of nitroprusside infusion. These authors’ results indicate that there is a progressive sequence of prognostic deterioration starting with the abnormal relaxation pattern and progressing with the pseudonormal and reversible restrictive patterns.

The Valsalva maneuver is another means for modifying ventricular loading that has been proposed for the analysis of ventricular filling pressures. It can be easily performed as part of the echocardiography laboratory routine to reduce the afterload, and can be used to assess the stability of the filling patterns in the same way as nitroprusside infusion. Nevertheless, to our knowledge, there are no published data on the prognostic value of this method. The aim of this study was to analyze the prognostic value of the filling pattern changes induced by the Valsalva maneuver in patients with left ventricular systolic dysfunction.

MATERIAL AND METHODS

Study Group

The study was conducted in a second-level hospital with no cardiac transplantation program. Among 177 consecutive patients evaluated in our echocardiography laboratory and diagnosed with left ventricular systolic dysfunction between June 2001 and May 2002, 37 patients were selected for participation in the study. All participants fulfilled the following conditions: a) sinus rhythm; b) ejection fraction less than 50%; c) end-diastolic diameter greater than 3.5 cm/m²; d) visible E and A waves, with the E/A ratio greater than 1; and e) E wave propagation velocity in the left ventricle less than 50 cm/s, as measured by color M-mode Doppler. This condition was required to confirm the existence of abnormal ventricular relaxation and to be able to distinguish between normal and pseudonormal filling patterns.

Ventricular dysfunction was due to an ischemic etiology in the majority of cases (26), 6 patients had dilated cardiomyopathy and 5 had other conditions (alcoholic or hypertensive heart disease). Thirty patients were men and the mean age was 65 years (range, 35-83); 18 were recruited during hospitalization and all patients had presented clinical signs and symptoms of heart failure at some time. The study was undertaken with the patients in stable clinical status under the best treatment possible; the hospitalized heart failure patients were included in the study only after they had completely stabilized. Informed consent for participation was obtained in all cases.

Echocardiographic Study

The echo-Doppler study was performed with a Vingmed CFM 800 system, with the patients in the left lateral decubitus position. The images were recorded on videotape for later analysis. Left ventricular and left atrial measurements were made from the parasternal long axis view using two-dimensional guided M-mode imaging. The ejection fraction was estimated from the apical view using the modified Simpson’s biplane method. Filling velocities were recorded from the apical four-chamber view with pulsed-Doppler. A single 5-mm sample volume was placed between the tips of the mitral leaflets, oriented parallel to the left ventricular filling wave recorded with color Doppler. From the same view, the E wave propagation velocity in the left ventricle was measured by color M-mode according to the method proposed by García et al. The E wave deceleration time was calculated from the peak E wave velocity to the point where the deceleration slope intercepted the Doppler baseline. A restrictive pattern was established when the E/A ratio was greater than 1 and the E wave deceleration time was less than or equal to 130 ms. A pseudonor-
nal pattern was defined as one in which the time was greater than this fixed limit. Both cases had to meet the condition of presenting an E wave propagation velocity less than 50 cm/s.

The baseline filling measurements were performed after the patient had rested for at least 10 min in a decubitus position while breathing normally. The patient was then asked to perform a Valsalva maneuver as deeply as possible for at least 6 s. The maneuver was repeated at least 3 times and was considered adequate when there was a reduction of at least 20% in the E or A wave velocities, or both. The position of the sample volume between the mitral leaflet tips was monitored during the maneuver, with readjustments performed when necessary. One investigator, unaware of the patients' clinical status, performed the measurements a posteriori, after meticulously calibrating the tracings. The maneuver resulting in the greatest filling pattern change with respect to baseline was used for the analysis. For the remaining parameters the mean of at least three measurements was used.

A reproducibility study was performed by 2 observers blindly assessing the first ten studies. After the first observer had finished, the patient rested for at least one-half hour and then the protocol was repeated with the second observer. The results for the mean changes were compared to analyze between-observer variability of the maneuver.

Follow-up

Patient follow-up was done by two cardiologists who were unaware of the changes induced by the Valsalva maneuver. The patients were contacted at least once every 4 months to record data on their functional class, the events that had occurred, and the treatment followed. The hospital records, primary care center data, and the death certificate in 1 case were also examined. Events were defined as sudden death or death due to a cardiac cause, and hospital admissions for heart failure. The data compiled on death or death due to a cardiac cause, and hospital admissions for heart failure were used for the analysis. For the remaining parameters the mean of at least three measurements was used.

A stratified survival analysis (time to first event) was done with the Kaplan-Meier method. For the multivariate analysis, the Cox regression method was used. For each of the 2 variables (death and events) a maximum model of 5 independent echocardiographic variables was established (peak E velocity, peak A velocity, ejection fraction, E wave deceleration time and filling reversibility following the Valsalva maneuver). Variables with a statistical significance of less than .05 were selected for each model using a backward approach. All the studies were two-tailed when possible, and required a significance level of .05.

RESULTS

The mean follow-up time was 18 months (range, 1-27). Eight patients died during follow-up, and 12 died or were hospitalized for severe heart failure. Six patients showed a restrictive filling pattern and the remaining patterns were considered pseudonormal. Three patients died in the restrictive group and 5 in the pseudonormal group (50% vs 17%; P=.073). The difference was smaller for the comparison of events between the groups (50% vs 30%; P=.34).

There were no significant differences between the E and A wave variations in the first 10 cases, as assessed by two independent observers. According to the first observer, the mean E wave reduction was 65%±18% and the mean A wave change was 104%±30%. The changes recorded by the second observer were 61%±17% and 108%±29%, respectively. Moreover, classification of all the cases was identical by both operators into reversible pattern (when there was a change to a normal relaxation pattern) or irreversible pattern (when the E/A value persisted at more than 1).

Overall, the E wave decreased by 0.33±0.16 m/s and the A-wave increased by 0.07±0.16 m/s.

The filling pattern was reversible following the Valsalva maneuver in 24 patients, whereas the E/A ratio remained higher than 1 in 12. Six patients died in the group with an irreversible restrictive or pseudonormal filling pattern (50%) and 2 in the reversible group (8.3%); the difference was significant (P=.005). An even greater difference was found when the total of

Statistical Analysis

All statistical analyses were done with SPSS®, version 8.0. The follow-up endpoints were considered to be death and the combined variable death and severe heart failure with hospital admission, which are referred to hereinafter as death and events, respectively.

Continuous variables were compared between the various groups using the Mann-Whitney U test and analysis of variance. Nonparametric continuous variables were analyzed with the Wilcoxon test or Kruskal-Wallis test, where appropriate. Differences among categorical variables were studied with the χ² test. The stratified survival analysis (time to first event) was done with the Kaplan-Meier method. For the multivariate analysis, the Cox regression method was used. For each of the 2 variables (death and events) a maximum model of 5 independent echocardiographic variables was established (peak E velocity, peak A velocity, ejection fraction, E wave deceleration time and filling reversibility following the Valsalva maneuver). Variables with a statistical significance of less than .05 were selected for each model using a backward approach. All the studies were two-tailed when possible, and required a significance level of .05.
events was considered, with 8 events in the irreversible group and 4 events in the reversible pattern group (67% vs 17%; \( P = .003 \)). The distribution of deaths and events according to the type of pattern and response to the Valsalva maneuver is shown in Figure 1. Death occurred in all patients with an irreversible restrictive pattern, none of those with a reversible restrictive pattern, 3% of those in the irreversible pseudonormal group and 9.5% in the reversible pseudonormal group. The differences in the distribution of these events among the four groups was not significant for deaths alone, but was significant for the total of events (\( P=.01 \)).

Kaplan-Meier analysis showed a significant reduction in survival in those patients in whom the Valsalva maneuver did not cause a change from a restrictive or pseudonormal pattern to another type of abnormal relaxation pattern, as is seen in Figure 2. The patients’ survival results according to their response to the Valsalva maneuver are shown in Table 1. Patients with a reversible pattern presented a reduction in the risk of cardiac death (HR=0.06, 95% confidence interval [CI], 0.01-0.48; \( P=.0003 \)). In addition, the risk of the total of events was lower in this group (HR=0.11; 95% CI, 0.03-0.43; \( P=.001 \)).

The survival analysis demonstrated that patients with a pseudonormal pattern had a lower risk of death than those with a restrictive pattern (HR=0.21; 95% CI, 0.08-0.55; \( P=.0003 \)).

### Table 1. Mean Survival and Hazard Ratios for Cardiac Deaths and Total Events According to the Response to the Valsalva Maneuver: Reversible or Irreversible

<table>
<thead>
<tr>
<th></th>
<th>Mean Survival (95% CI), Months</th>
<th>Hazard Ratio (95% CI)</th>
<th>( P )</th>
</tr>
</thead>
<tbody>
<tr>
<td>Events</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Reversible</td>
<td>33 (29-37)</td>
<td>0.11 (0.03-0.43)</td>
<td>.010</td>
</tr>
<tr>
<td>Irreversible</td>
<td>14 (9-20)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Deaths</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Reversible</td>
<td>35 (33-38)</td>
<td>0.06 (0.01-0.48)</td>
<td>.0003</td>
</tr>
<tr>
<td>Irreversible</td>
<td>16 (10-22)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

CI indicates confidence interval

---


Figure 1. Classification of patients according to a restrictive or pseudonormal filling pattern and response to the Valsalva maneuver. The clinical evolution is shown for each group.

Figure 2. Survival curves for patients with a reversible or irreversible filling pattern following the Valsalva maneuver, according to the variable death. CI indicates confidence interval.
univariate analysis for the E wave velocity, E/A wave included on the basis of their statistical value in the application of the Valsalva maneuver). The first 3 were E wave deceleration time, and filling reversibility after treated (peak E velocity, peak A velocity, ejection fraction, and filling reversibility after application of the Valsalva maneuver). The first 3 were included on the basis of their statistical value in the univariate analysis for the E wave velocity, E/A wave percent changes, E/A ratio, and heart rate before and during the Valsalva maneuver were also analyzed, but no significant relationship was found with the endpoints of the analysis. For each of the two variables (death and events) a model with a maximum of 5 independent echocardiographic variables was contemplated (peak E velocity, peak A velocity, ejection fraction, E wave deceleration time, and filling reversibility after application of the Valsalva maneuver). The first 3 were included on the basis of their statistical value in the univariate analysis for the E wave velocity, E/A wave

Results are presented in Table 3. The E and A wave

left atrial velocity; DD, left ventricular end-systolic diameter; E, peak E wave velocity; EF, ejection fraction; MR, grade of mitral regurgitation; NS, non-significant; EDT, E wave deceleration time; EVp, E wave propagation velocity; A%, A wave percent change following the Valsalva maneuver.

**TABLE 3. Results of Cox Univariate Analysis for the Echocardiographic Measures Performed as Independent Variables, With the Dependent Variables Events and Deaths as Endpoints**

<table>
<thead>
<tr>
<th>E/A</th>
<th>Hazard Ratio (95% CI)</th>
<th>P</th>
<th>E/A</th>
<th>Hazard Ratio (95% CI)</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>A wave</td>
<td>1.03 (1.01-1.05)</td>
<td>.023</td>
<td>A wave</td>
<td>1.02 (0.99-1.05)</td>
<td>.012</td>
</tr>
<tr>
<td>E</td>
<td>1.06 (1.01-1.12)</td>
<td>.003</td>
<td>E</td>
<td>1.05 (1.01-1.09)</td>
<td>.023</td>
</tr>
<tr>
<td>EDT</td>
<td>1.02 (1.01-1.10)</td>
<td>.045</td>
<td>EDT</td>
<td>1.01 (0.99-1.10)</td>
<td>.116</td>
</tr>
<tr>
<td>EVp</td>
<td>1.9 (9.4-2.3)</td>
<td>.036</td>
<td>EVp</td>
<td>1.6 (9.2-8.1)</td>
<td>.012</td>
</tr>
<tr>
<td>MR</td>
<td>2.5 (0.94-5.90)</td>
<td>NS</td>
<td>MR</td>
<td>2.4 (1.03-5.57)</td>
<td>.040</td>
</tr>
<tr>
<td>A%</td>
<td>0.95 (0.89-1.02)</td>
<td>NS</td>
<td>A%</td>
<td>1.07 (0.96-1.19)</td>
<td>NS</td>
</tr>
<tr>
<td>EDT</td>
<td>1.02 (1.01-1.10)</td>
<td>.045</td>
<td>EDT</td>
<td>1.02 (0.99-1.10)</td>
<td>.116</td>
</tr>
<tr>
<td>EVp</td>
<td>1.9 (9.4-2.3)</td>
<td>.036</td>
<td>EVp</td>
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<td>.012</td>
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<tr>
<td>MR</td>
<td>2.5 (0.94-5.90)</td>
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<td>MR</td>
<td>2.4 (1.03-5.57)</td>
<td>.040</td>
</tr>
<tr>
<td>A%</td>
<td>0.95 (0.89-1.02)</td>
<td>NS</td>
<td>A%</td>
<td>1.07 (0.96-1.19)</td>
<td>NS</td>
</tr>
</tbody>
</table>

*MR indicates grade of mitral regurgitation; NS, non-significant; NYHA, New York Heart Association; EDT, E wave deceleration time; EVp, E wave propagation velocity.

**TABLE 4. Cox Multivariate Analysis Results. Among the Values Included in the Model (Peak E Wave Velocity, Peak A Wave Velocity, Ejection Fraction, E Wave Deceleration Time and Filling Reversibility Following the Valsalva Maneuver), Those With an Independent Predictive Value Are Shown**

<table>
<thead>
<tr>
<th>Dependent Variables</th>
<th>Independent Variables</th>
<th>Hazard Ratio</th>
<th>95% Confidence Interval</th>
</tr>
</thead>
<tbody>
<tr>
<td>Death</td>
<td>A wave</td>
<td>1.04</td>
<td>1.01-1.07</td>
</tr>
<tr>
<td></td>
<td>Reversible with</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Valsalva</td>
<td>0.06</td>
<td>0.01-0.48</td>
</tr>
<tr>
<td>Events</td>
<td>A wave</td>
<td>1.03</td>
<td>1.00-1.06</td>
</tr>
<tr>
<td></td>
<td>Reversible with</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Valsalva</td>
<td>0.11</td>
<td>0.03-0.43</td>
</tr>
</tbody>
</table>

ratio and ejection fraction, whereas E wave deceleration time was included because of its proven prognostic value. Other possible variables, such as clinical or demographic factors, were not included because of the limited size of the sample. The analysis showed that A wave velocity and the results of the Valsalva maneuver were the only independent predictors of death due to a cardiac cause and of total events, as is shown in Table 4.
DISCUSSION

This study shows that the type of response produced with the use of the Valsalva maneuver in patients with a pseudonormal or restrictive filling pattern can serve to separate patients with systolic dysfunction into different prognostic groups. This finding, which, to our knowledge, is demonstrated for the first time, supports the use of this method echocardiographic assessment of heart failure.

The prognostic value of similar changes in response to various manipulations to modify loading, such as long-term treatment for heart failure and nitroprusside infusion, have been reported previously. The results obtained with the 3 methods (Valsalva maneuver, chronic heart failure treatment, and nitroprusside treatment) confirm the hypothesis of Pozzoli et al., suggesting that the type of transmitral filling pattern response provides an estimation of cardiovascular reserve in patients with elevated left ventricular stiffness.

There are substantial differences between the populations included in our study and those of Pozzoli et al. and Temporelli et al. Whereas these authors studied only patients with restrictive patterns, our patients had both restrictive and pseudonormal patterns. Patients with systolic dysfunction show a spectrum of filling patterns ranging from abnormal relaxation to restrictive, corresponding to increasing ventricular stiffness and left atrial pressure. Along this continuum, the cut-off between pseudonormal and restrictive is somewhat arbitrary. In our series the filling pattern changes provided a more precise reflection of the patients' evolution than the mere differentiation between pseudonormal and restrictive. In addition, we considered the analyses of evolution in the group with a pseudonormal pattern to be interesting since little attention has been paid to this population. This group is at a lower risk than those with restrictive filling, but at a higher risk than patients with abnormal relaxation. The effect of loading modifications has not been studied in these patients; hence, once again, the present study is the first to assess this factor, to the best of our knowledge. The fact that a load reduction in this group also resulted in filling changes with prognostic value is in keeping with the classification proposed by Xie and Smith. These authors suggested that cases with restrictive or pseudonormal patterns found to be reversible can be considered a group with an “intermediate prognosis” between the abnormal relaxation and irreversible restrictive pattern groups.

The various methods used to modify loading show considerable differences. Whereas the Valsalva maneuver mainly reduces preload, nitroprusside reduces both preload and afterload. We did not compare these methods in the present study and cannot judge whether they are equivalent.

The subgroup with an irreversible pattern included patients with many other factors associated with a poor prognosis, such as greater ventricular dilation, more elevated E/A ratios, and particularly, a poorer functional class. The findings are expected and we do not believe they detract from the results. The Valsalva maneuver would maintain its value both for predicting the response to treatments that reduce the load as well as to confirm or substitute the prognosis provided by other data with recognized prognostic value, such as functional class.

The E wave propagation velocity was used to confirm the existence of abnormal ventricular relaxation and verify as pseudonormal the cases with this diagnosis, as was done by Moller et al. in a previous study. The relaxation disturbance should appear in all the patients, since all have systolic dysfunction, and this was confirmed by the detection of a depressed E wave propagation velocity in all the patients. Nevertheless, the E wave propagation velocity did not demonstrate prognostic value. This was not surprising since the diastolic function parameters most closely related to the prognosis are those reflecting deterioration of ventricular stiffness and increased end-diastolic pressure, and E wave velocity propagation is relatively independent of these factors. It was more surprising to find that the ratio between E wave velocity and E wave propagation velocity as measured by color M-mode Doppler showed no relationship with survival. This ratio is known to correlate with pulmonary capillary pressure and is associated with the prognosis of patients experiencing a first infarction. Patients in the study cited were included in an unstable phase during the first 24 hours after infarction and therefore, were a different population than those in our series, analyzed after they had been treated in the best way possible and in a stable condition. Nonetheless, we cannot rule out that the absence of a relationship was due to the small size of our sample.

Other studies on the prognosis of patients with systolic dysfunction have reported other echocardiographic predictors of events, such as E wave deceleration time and atrial size, which did not appear as independent factors in our study. These differences can be explained by several facts. First, only patients with an E/A greater than 1 were selected for participation in our study. We excluded patients with an abnormal relaxation filling pattern, which are the majority and have a better prognosis; hence the sample is different than the population with systolic dysfunction. In addition, this strict selection of the sample reduced the number of candidates for inclusion, such that there were relatively few events and the relationship with the variables studied might be less consistent. The size of the sample is one of the main limitations of the study, and made it impossible to analyze the clinical

and demographic variables together with the echocardiographic variables.

The Valsalva maneuver could have been standardized by recording the pressure exerted. This was not done because our objective was to assess the maneuver as it is usually performed. To reduce the variability of the response, we believe that a minimum apnea time should be required, the maneuver should be repeated until the patient has learned to do it properly and there should be evident changes in the filling waves. The efficacy of these measures is reflected in the prognostic value of the results obtained and the fact that the changes observed were similar to those described for patients with elevated filling pressures.

In conclusion, the results of this study suggest that persistence of a restrictive or pseudonormal pattern after the Valsalva maneuver performed with minimal conditions of standardization indicates a poorer prognosis than reversibility of the pattern. These findings confirm the value of this fast, simple tool to assess prognosis in patients with systolic dysfunction.

In conclusion, the results of this study suggest that persistence of a restrictive or pseudonormal pattern after the Valsalva maneuver performed with minimal conditions of standardization indicates a poorer prognosis than reversibility of the pattern. These findings confirm the value of this fast, simple tool to assess prognosis in patients with systolic dysfunction during the standard echocardiographic examination.

REFERENCES


