Heart Failure

Growth Hormone Prolongs Survival in Experimental Postinfarction Heart Failure

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OBJECTIVES	We evaluated the effects of growth hormone (GH) on survival in experimental heart failure (HF).
BACKGROUND	Growth hormone has been beneficial in various models of experimental HF. Whether GH also affects HF progression and survival is not known.
METHODS	A total of 119 rats with moderate myocardial infarction were randomized to receive either GH (3.5 mg/kg every other day) or placebo for 28 days. Treatment was initiated one month after coronary ligation; the follow-up lasted 13 months. In the surviving animals, Doppler echocardiography and closed-chest Millar left ventricular (LV) catheterization were performed. Apoptosis, collagen volume fraction, and capillary density in the LV zone remote from infarction were measured. The early effects of GH on apoptosis were also assessed in a subgroup of eight infarcted rats, treated as specified earlier and euthanized at one month.
RESULTS	Survival rate was 68% in GH-treated rats and 48% in the placebo group (p = 0.0377). Growth hormone had no effect on myocardial architecture, systolic function, and sarcoplasmatic reticulum calcium ATPase-2 messenger ribonucleic acid. Growth hormone improved LV relaxation; this was associated with a 50% reduction in collagen volume fraction and a 27% increase in capillary density. Growth hormone reduced the apoptotic index by 50% at one month and by 33% at 13 months.
CONCLUSIONS	Growth hormone prolonged survival of rats with postinfarction HF. This effect was associated with marked attenuation of cardiomyocyte apoptosis and pathologic interstitial remodeling in the surviving myocardium and enhanced LV relaxation. (J Am Coll Cardiol 2003;41:2154–63) © 2003 by the American College of Cardiology Foundation

Growth hormone (GH) has gained growing attention in the past decade as an important regulator of myocardial structure and function (1). Because of its growth-promoting, vasodilating, and positive inotropic properties, GH has been recently proposed as an adjunctive therapy in chronic heart failure (CHF) (2). Indeed, a number of animal studies have consistently documented the efficacy of GH and its mediator, insulin-like growth factor-I (IGF-I), in attenuating left ventricular (LV) remodeling and improving myocardial energetics and function in experimental myocardial infarction (MI) (3–5). For reasons not completely understood, clinical studies of GH administration to patients with CHF have not provided unequivocal results (2,6–9). However, the issue of whether GH is capable of affecting heart failure progression and survival is unsettled.

The present study was designed to evaluate the impact of four-week GH administration on LV remodeling and mortality in a rat model of post-MI heart failure (HF). Because cardiomyocyte loss has been hypothesized to be an important mechanism of myocardial remodeling, and IGF-I has previously shown to be endowed with powerful antiapo-ptotic properties (10–12), we also evaluated the effects of GH treatment on myocardial apoptosis.

METHODS

All methods described conformed to the "Position of the American Heart Association on Research Animal Use," and the protocol was approved by the Animal Care Committee of the University Federico II of Naples. Myocardial infarction was induced according to previously described methods (3) in male Sprague-Dawley rats (Charles River Italy, Calco, Lodi, Italy), weighing 150 to 200 g, and anesthetized with pentobarbital (60 mg/kg intraperitoneal) and orally intubated. Perioperative mortality rate was approximately 35%. One month after coronary ligation, a total of 119 surviving rats were randomized to receive either GH (58 rats), 3.5 mg/kg/every other day for 4 weeks, or placebo (61 rats). The survival study was designed to last 13 months. During the treatment period, the cages were inspected daily for dead animals. A postmortem examination was performed, and the lungs were inspected for gross signs of consolidation. The lungs and heart were placed in formalin for subsequent necropsy studies. An additional group of eight infarcted rats was used to assess the early effects of GH on cardiomyocyte apoptosis. These animals were treated with GH (n = 4) or placebo (n = 4) for four weeks, as described earlier, and were euthanized one month after start of treatment. This was done because apoptotic death is known to vanish with time after MI (12), and its occurrence might have

Abbreviations and Acronyms	
CHF	= chronic heart failure
CVF	= collagen volume fraction
DNA	= deoxyribonucleic acid
GH	= growth hormone
HF	= heart failure
IGF-I	= insulin-like growth factor I
LV	= left ventricular
MI	= myocardial infarction
RNA	= ribonucleic acid
SERCA-2	= sarcoplasmatic reticulum calcium ATPase-2
TUNEL	= terminal UTP nick end labeling

been too small to detect potential intergroup differences at 13 months.

Echocardiographic and hemodynamic studies. Transthoracic echocardiograms were performed in all surviving animals, according to previously described methods (3), with an electronic system equipped with a 7 or 12 MHz probe (Agilent Technologies, Palo Alto, California). All measurements, performed with an off-line analysis system by one observer who was blind to prior results, were based on the average of three to six consecutive cardiac cycles.

Within 6 h from the final echocardiogram, rats underwent closed-chest LV catheterization using a calibrated 2 F micromanometer-tipped catheter (SPR-407, Millar Inst. Inc., Houston, Texas), as previously described (3). The time-constant of LV pressure decay (tau) was calculated by the variable asymptote method.

Morphometric histology and immunohistochemistry. Twelve hearts from each group were rapidly excised, gross examined, immersion fixed in formalin 4%, and embedded in paraffin (3,13). In each animal the following parameters were assessed from transverse sections (6 μm thick) taken at similar level, by two observers blind to the treatment groups: 1) percentage of infarcted areas, Mallory's trichrome, picrosirius red (3,13,14); 2) cardiomyocyte diameter, hematoxylin, and eosin (50 fields per animal) (3); 3) collagen volume fraction (CVF) in myocardial areas remote from infarction, picrosirius red and collagen I and III immunostaining (Monosan, Uden, The Netherlands), 40× magnification, 50 fields per animal; CVF was expressed as the mean percentage of connective tissue areas divided by total tissue area in the same field (14); 4) capillary density taken as the number of capillaries per mm^2 (staining GSLI) (4); and 5) perivascular collagen. Collagen I and III isoforms were also detected by SDS-PAGE electrophoresis-Western blot according to standard methods (15). Slides were observed with a Nikon Microphot FXA light microscope equipped with a polarized set and analysed with Zeiss KS300 software.

Cardiomyocyte apoptosis. Apoptosis was assessed both in the rats euthanized at one month and in the rats surviving at 13 months. Four independent methods were employed to assess the prevalence of cardiomyocyte apoptosis: 1) the terminal deoxynucleotidyltransferase-mediated dUTP nick

end labeling (TUNEL, in situ Cell Death Kit, Roche Diagnostic, Basel, Switzerland); 2) hairpin probe with single-base 3′ overhangs (in situ oligo ligation technique, Apoptag ISOL kit, Intergen Co., Serological, Norcross, Georgia, plus streptavidin-fluorescin, Pierce Laboratories, Rockford, Illinois) (11); 3) caspase-3 activity detection by immunochemistry (M30, Roche Diagnostic); and 4) deoxyribonucleic acid (DNA) laddering by agarose gel electrophoresis, to confirm that immunohistochemical detection of DNA fragments reflected internucleosomal DNA cleavage (12).

Cardiomyocytes were identified by incubation with anti-alpha-actin monoclonal antibody (1:20, Sigma, Saint Louis, Missouri) and subsequently with anti-mouse IgG TRITC (tetramethyl rhodamine isothiocyanate) labeled. All the nuclei were also stained with DAPI (11,12). Positive and negative controls were performed as previously described (12). Myocyte apoptotic nuclei were identified and counted as previously described (10–12). One hundred high-power fields (120×) were evaluated in each section for a total of 1,500 fields per sample. The number of myocyte nuclei labeled by terminal deoxynucleotidyl transferase (TdT) or hairpin probe divided by the numerical density of myocyte nuclei represents the apoptotic index.

SERCA-2 measures. RNAse protection assay (RPA II kit Ambion, Austin, Texas) and the solution hybridization RNAse protection assay were performed according to the manufacturer's instructions with 20 μg of total ribonucleic acid (RNA), prepared as previously described (16).

Statistical analysis. All results, except survival, are given as mean ± SEM. Statistical analysis was performed using the SPSS statistical package (SPSS, version 11.0, Chicago, Illinois). After testing for normal distribution, comparisons between the two study groups were performed with the unpaired two-tailed Student *t* test. The survival curve for each treatment was determined using the Kaplan-Meier method. Comparison of the survival distribution between GH- and placebo-treated rats was performed by a log-rank test.

In planning the study, sample size calculations were performed with the intent to detect a 20% difference (such as 60% vs. 80% for ≈1-year survival) between treatments with a two-sided significance level of 0.05 and 80% power. A value of p < 0.05 was statistically considered significant.

RESULTS

There were no significant differences in body weights between the two study groups at baseline, at end of GH therapy, and at 13 months. Body weight increments at one month and at 13 months were 18% and 50% in the placebo group and 24% and 48% in the GH group, respectively (Table 1). Infarct sizes of rats that died spontaneously were slightly higher than those of rats euthanized at 13 months: 30 ± 3% versus 28 ± 2% (dead during the trial) and 27 ± 2% versus 25 ± 3% (euthanized at 13 months) in GH and placebo groups, respectively (Table 1). Thus, randomization resulted in a balanced distribution of infarct sizes in both groups. No rat died of lung infection.

Table 1. Animal Characteristics, Morphometric Histology, and SERCA-2 Myocardial Levels

	MI-Placebo	MI-GH
Body weight$_{BASELINE}$, g	311 ± 12 (n = 61)	309 ± 15 (n = 58)
Body weight$_{4\ weeks}$, g	367 ± 13 (n = 61)	384 ± 15 (n = 58)
Body weight$_{13\ months}$, g	467 ± 19 (n = 29)	460 ± 24 (n = 39)
LV wet weight, g	1.32 ± 0.09 (n = 29)	1.23 ± 0.11 (n = 39)
LV wet/body weight, mg/g	2.79 ± 0.20 (n = 29)	2.80 ± 0.32 (n = 39)
Infarct size, %	27 ± 3 (n = 61)	28 ± 3 (n = 58)
Collagen volume fraction, %	4.42 ± 1.9 (n = 12)	2.22 ± 0.47* (n = 12)
Capillary density, n/mm^2	983 ± 160 (n = 12)	1247 ± 183* (n = 12)
Oxygen diffusion distance, μm	16.0 ± 1 (n = 12)	14.1 ± 1.1 (n = 12)
Perivascular collagen ratio	1.5 ± 0.2 (n = 12)	1.4 ± 0.2 (n = 12)
SERCA-2, pg/μg RNA	1.004 ± 0.05 (n = 7)	1.075 ± 0.02 (n = 7)

Values are mean ± SE. *$p < 0.05$ vs. MI-placebo group.

n = number of animals studied; GH = growth hormone; LV = left ventricular; MI = myocardial infarction; RNA = ribonucleic acid; SERCA = sarcoplasmic reticulum calcium ATPase.

The survival curves are depicted in Figure 1. Of the 119 rats entered into the study, 68 (39 GH-treated and 29 in the placebo arm) survived at 13 months. Thus, the 13-month survival rate was 48% in the placebo group and 68% in GH-treated rats ($p = 0.0377$ with log-rank test). The beneficial effect of GH treatment on survival was also supported by the increase in the mean survival time from 277 days in the placebo group to 323 days in the GH group.
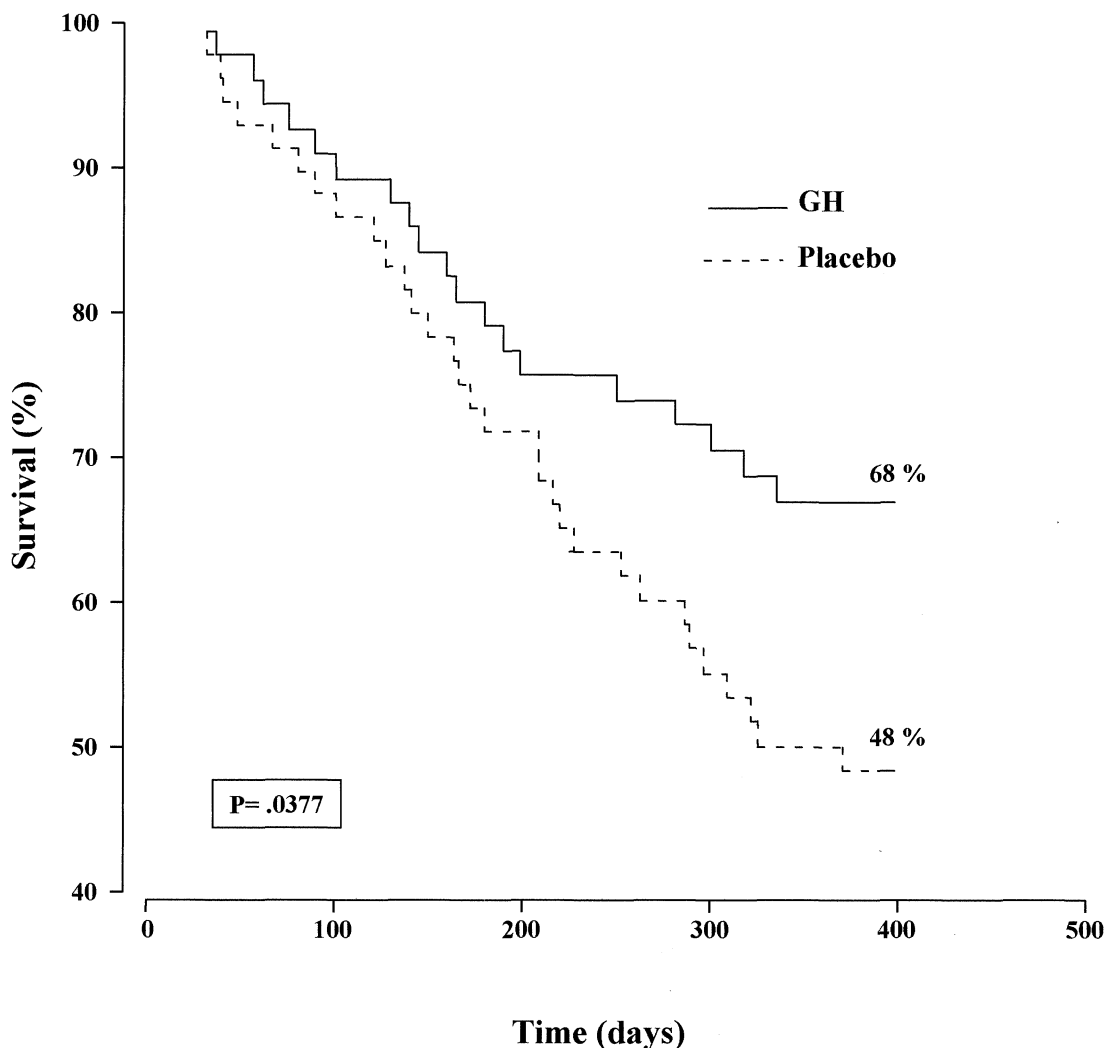
Figure 1. Kaplan-Meier curves in rats with myocardial infarction treated with placebo or growth hormone (GH). Thirteen-month mean survival time was 267 and 323 days in placebo and GH-treated rats, respectively.

Table 2. Echocardiographic Data at 13 Months

	MI-Placebo n = 24	MI-GH n = 34
Anterior wall diastole, mm	1.20 ± 0.06	1.18 ± 0.07
Anterior wall thickening, %	42 ± 6	36 ± 7
Posterior wall diastole, mm	1.31 ± 0.07	1.42 ± 0.08
Posterior wall thickening, %	67 ± 8	58 ± 10
LV diastolic diameter, mm	8.03 ± 0.32	8.53 ± 0.29
LV systolic diameter, mm	5.3 ± 0.3	5.5 ± 0.28
LV diastolic/body weight, cm/kg	1.72 ± 0.1	1.85 ± 0.14
LV systolic/body weight, cm/kg	1.13 ± 0.09	1.19 ± 0.1
Relative wall thickness	0.33 ± 0.02	0.33 ± 0.02
LV fractional shortening, %	34 ± 4	35 ± 3
Heart rate, beats/min	239 ± 22	249 ± 10
Stroke volume, ml	0.40 ± 0.05	0.45 ± 0.04
Stroke volume/body weight, ml/kg	0.86 ± 0.1	0.99 ± 0.1
Cardiac output, ml/min	97 ± 13	113 ± 19
Cardiac output/body weight, ml/min/kg	204 ± 35	246 ± 28

All data are mean ± SE.
   Abbreviations as in Table 1.

Assessment of LV function in vivo. Echocardiography was performed in all surviving rats. However, the data of only 58 animals were included in the analysis (24 placebo-treated and 34 GH-treated rats). In the remaining 10 rats, the recording was not of optimal quality. Similarly, the LV hemodynamic study was performed in 13 placebo-treated and 15 GH-treated rats. No significant differences were found as to LV architecture and systolic function between the two study groups. There was a tendency toward a reduction of end-diastolic pressure and posterior wall diastolic load index in the active treatment group compared with placebo. The time constant of LV relaxation was shortened by 20% in the GH-treated rats compared with placebo ($p < 0.05$), indicating enhanced LV relaxation (Tables 2 and 3).

SERCA-2 myocardial expression. The two infarcted groups displayed a significant downregulation of myocardial SERCA-2 mRNA expression (−26%) when compared with an age-matched control group (1.359 ± 0.03 pg/μg RNA), measured by a solution hybridization assay. However, no significant differences in SERCA-2 transcript levels were found between the two study groups (Table 1).

Morphometric histology and immunohistochemistry. Morphometric histology showed no differences in myocyte diameter between the two groups (22 ± 3 in placebo-treated vs. 25 ± 5 μm in GH-treated animals, p = NS). Sirius red staining showed a 50% reduction of CVF in GH-treated versus untreated rats. Perivascular collagen ratio did not differ significantly between the two groups. Capillary density increased by 27% in rats receiving GH. Accordingly, oxygen diffusion distance decreased significantly when compared with controls (14.1 ± 0.4 vs. 16.1 ± 0.5 μm in GH and control group, respectively; $p < 0.01$). Collagen phenotype differed between the infarcted animals: the collagen I/III ratio was approximately 30% lower in the GH-treated animals than in the placebo group (5.4 ± 0.9 vs. 7.7 ± 1.2; $p < 0.01$). Also, the histopathologic appearance of collagen fibers differed in the two study groups. Specifically, in both the infarcted and noninfarcted areas, the GH-treated group displayed better preservation of the collagen framework, with more uniform deposition of collagen I fibers and a reduction of the typical disarray exhibited by the placebo infarcted group. Figures 2 and 3 depict representative photographs from the two study groups.

Apoptosis. The extent of apoptosis did not differ when measured by TdT or hairpin probe (Figs. 4 to 6), as previously reported (11). At 13 months, the apoptotic index was reduced by 33% in the GH-treated rats (20 and 22 cardiomyocyte nuclei/10^6) compared with the placebo group

Table 3. Hemodynamic Data at 13 Months

|	MI-Placebo n = 13	MI-GH n = 15
LV systolic pressure, mm Hg	104 ± 4	110 ± 7
Mean aortic blood pressure, mm Hg	86 ± 3	92 ± 5
LV end-diastolic pressure, mm Hg	11 ± 2	8 ± 2
Peak positive dP/dt, mm Hg/s	3884 ± 444	4255 ± 945
Peak negative dP/dt, mm Hg/s	2317 ± 252	2640 ± 390
Systemic vascular resistance, mm Hg/ml/min/kg	0.42 ± 0.02	0.37 ± 0.03
Posterior wall systolic load index, kdynes/cm^2	118 ± 10	126 ± 13
Posterior wall diastolic load index, kdynes/cm^2	26 ± 4	19 ± 4
tau, ms	30 ± 2	24 ± 2*

All data are mean ± SE; tau = time constant of LV pressure decay; *$p < 0.05$ vs. MI-placebo group.
   Abbreviations as in Figure 1.
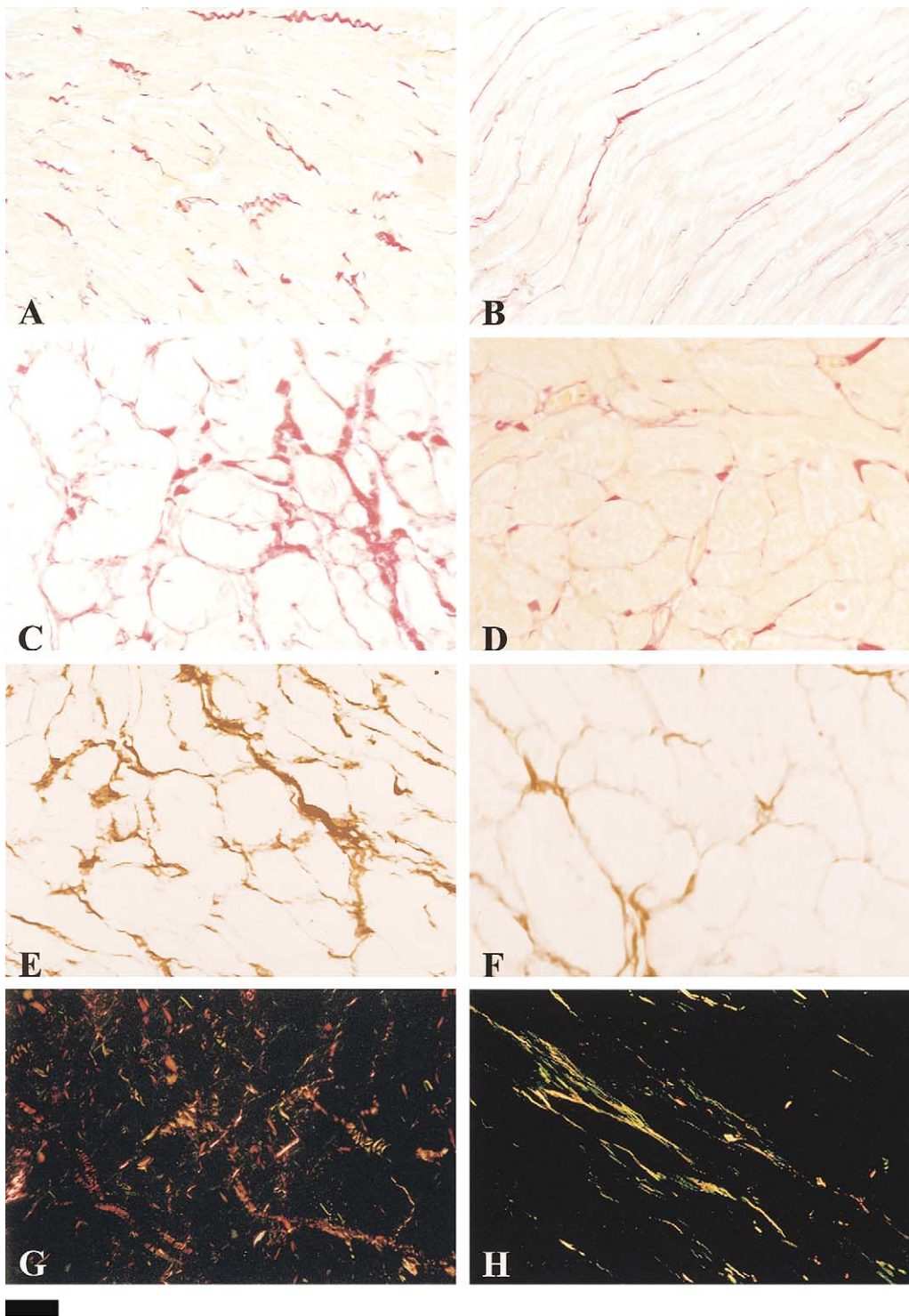
Figure 2. Collagen content and morphology in remote zones from infarction in placebo **(left column)** and growth hormone (GH)-treated rats **(right column)**, picrosirius red staining. In the placebo group **(A)**, collagen fibers **(in red)** appear shorter and thicker than in the GH group **(B)**. Compared with **panel D**, **panel C** exhibits a marked deposition of collagen fibers that encapsulate cardiomyocytes and fill the interstitial gaps. **Panels E and F** show collagen I antibody stain in the placebo and GH groups, respectively. In **panels G and H** collagen I appears **red-yellowish**, whereas collagen III appears **green** under polarized light. Collagen III is substantially reduced in **panel G** (placebo) compared with **panel H** (GH). Collagen I is the major component of thick and short collagen fibers in **panel G** and of long and thin fibers in **panel H**, where also a weak collagen III network can be appreciated. **Bar** = 20 μm.

(30 and 33 cardiomyocyte nuclei/10^6), as measured by TUNEL and hairpin assay, respectively, p = 0.069. In the subgroup of rats euthanized at one month (n = 8), there was a 50% reduction of the apoptotic index in the GH compared with the placebo group. The TUNEL apoptotic index was 40 and 45 cardiomyocyte nuclei/10^6 versus 80 and 82 cardiomyocyte nuclei/10^6, as measured by TUNEL and hairpin assay, respectively (p < 0.001). Caspase activity was
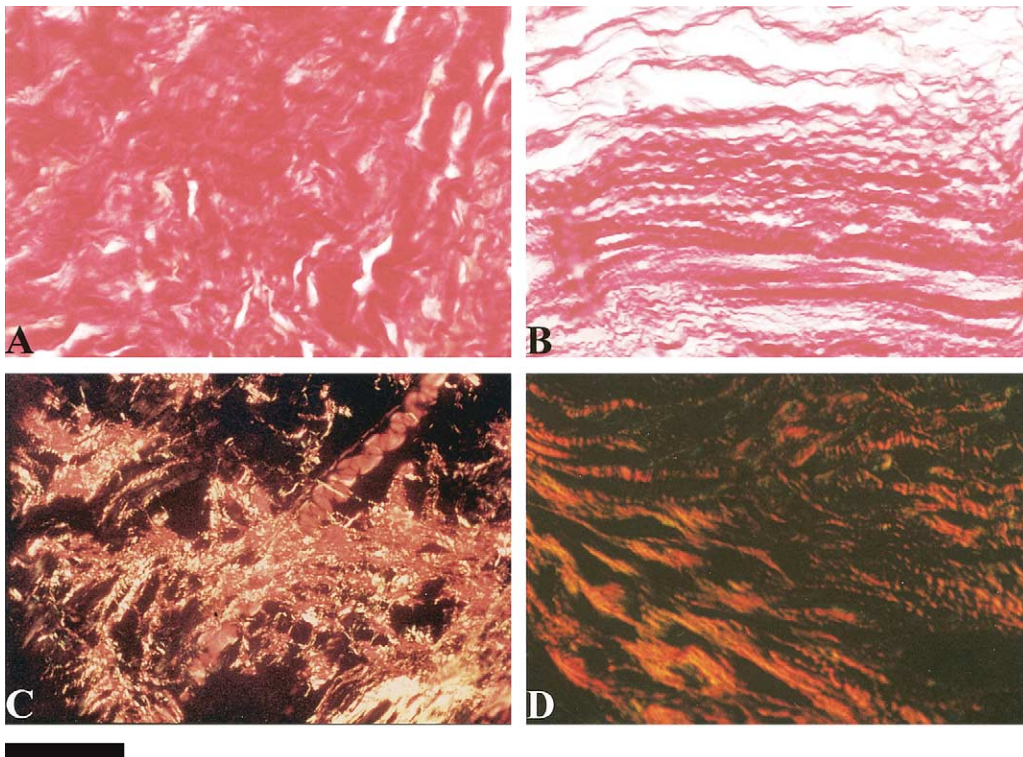
Figure 3. Collagen morphology in infarcted areas from placebo **(left)** and growth hormone (GH)-treated rats **(right)**. **Panels A and B** depict picrosirius **red** staining observed under white light. Note that the placebo group **(A and C)** exhibits disarrayed collagen deposition, whereas in GH group **(B and D)** the collagen network appears better preserved, with collagen scar fibers organized in parallel bundles. In both groups, scars are mainly made of collagen I fibers, as shown under polarized light microphotographs. **Bar** = 20 μm.

found to co-localize with TUNEL-positive cardiomyocytes, with 95% of TUNEL-positive nuclei that were also anti-cleaved caspase-3 positive (Fig. 5). The typical DNA laddering was apparent in all study groups, confirming the presence of mono-oligonucleosomes cleavage of genomic DNA in the observed TUNEL/hairpin-labeled cardiomyocytes (not shown).

DISCUSSION

The present study demonstrates that GH prolongs survival in a rat model of post-MI HF, and that this effect is associated with marked attenuation of interstitial fibrosis and cardiomyocyte apoptosis.

Current study. The most striking difference between the two study groups was the attenuation of interstitial remodeling and cardiomyocyte apoptosis in the non infarcted myocardium of GH-treated rats. Cardiac fibrosis is regarded as one of the major biological determinants of poor prognosis in HF, insofar as it is strongly associated with severe arrhythmias, diastolic dysfunction and progression of chronic heart failure (CHF), and sudden death.

In the current study, a broad spectrum of structural abnormalities of the extracellular matrix was improved by GH treatment. Specifically, there was a 50% decrease in CVF, whereas capillary density was significantly increased. Growth hormone lowered the collagen I/III ratio, which is usually increased in animal models of CHF, contributing to the elevation of operating chamber stiffness (15). The

changes induced by GH in the nonmyocyte compartment were associated with enhanced LV relaxation. It is a well-established concept that relaxation abnormalities occur in ischemic heart disease because of multiple defects, including inhibition of Ca^{2+} uptake by the SR, impaired detachment of force-generating sites between actin and myosin, nonuniformity, and interstitial fibrosis (17). In the current study, the enhanced relaxation found in the GH-treated group appears to depend on the reduced interstitial fibrosis rather than improvement of SERCA-2, which was previously reported in post-MI HF following short-term GH administration (4).

As to the mechanisms by which GH attenuated fibrosis in our model, one possibility is that GH acts directly by reducing collagen synthesis or by increasing its breakdown. This interpretation is unlikely because GH is a powerful wound-healing stimulator and activates fibroblasts in vitro (1,18). Another possibility is that GH, by improving hemodynamics and attenuating LV wall stress, reduces the accumulation of extracellular matrix proteins in the interstitial space. This interpretation is equally unlikely, given the similarity of the hemodynamic profile exhibited by the two study groups. A third possibility is that the well-known antiapoptotic properties of the GH/IGF-I system (10–12) may have limited the ongoing cell loss that occurs during pathologic remodeling. Although apoptosis per se does not induce fibrosis, it leaves myocardial defects that may become
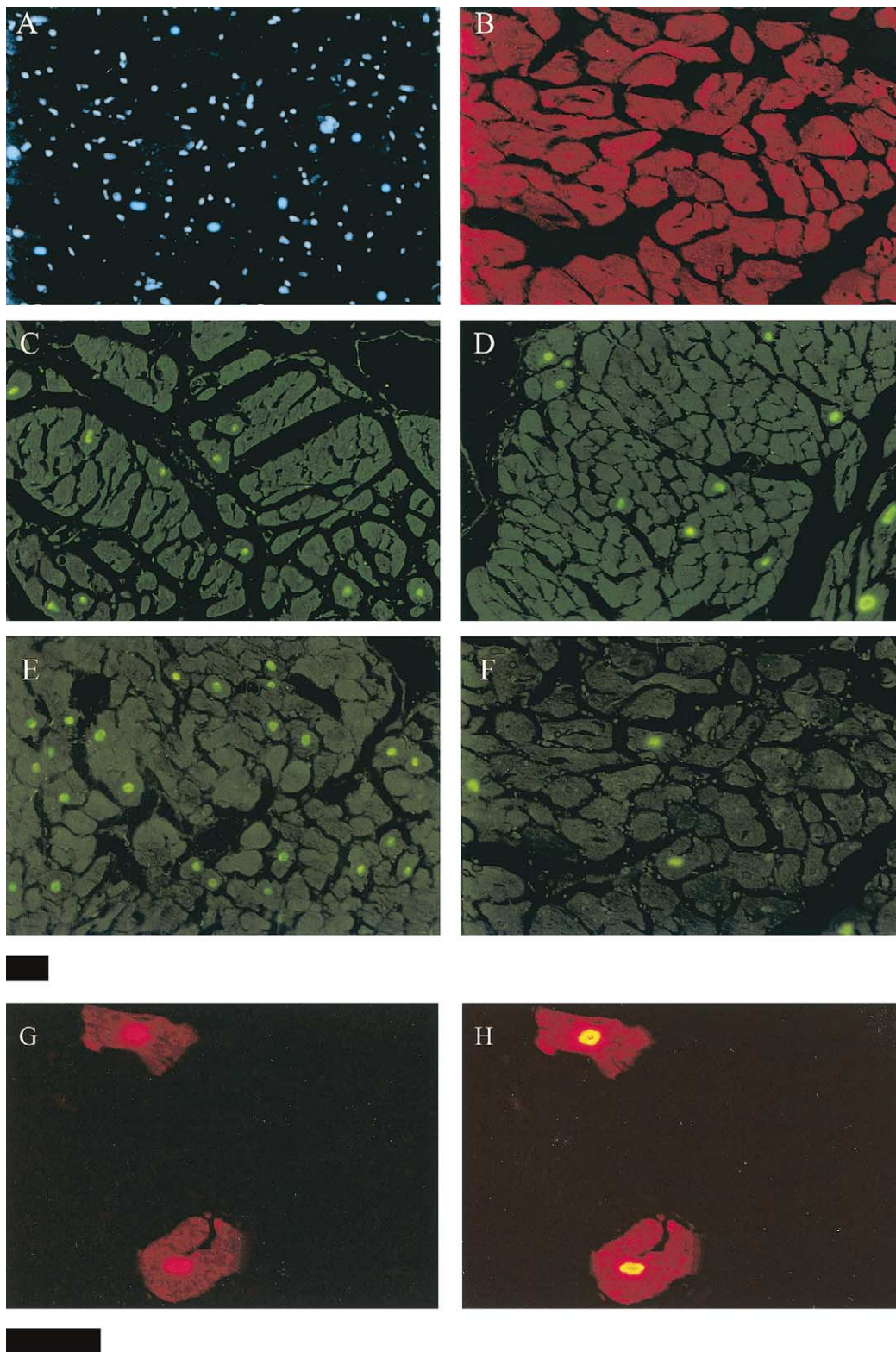
Apoptotic index at 13 months
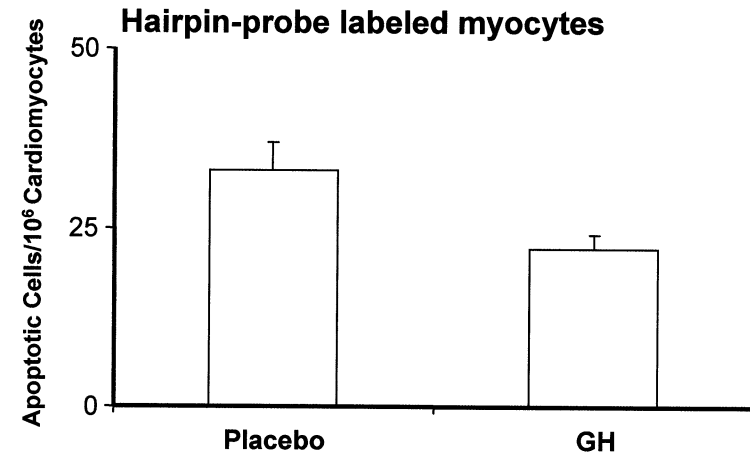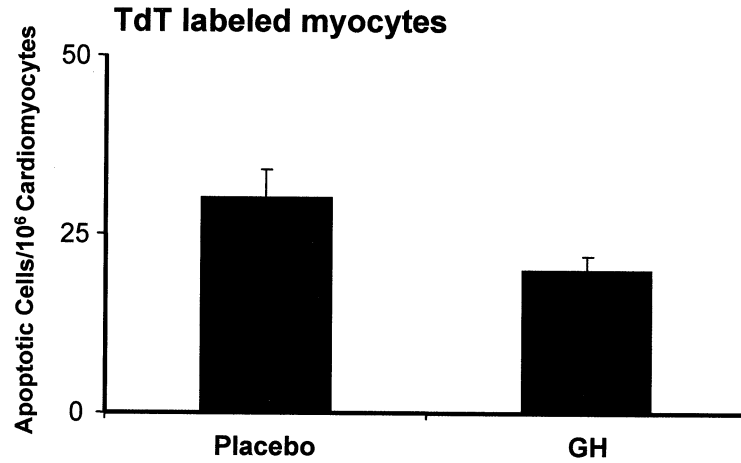

Apoptotic index at 1 month
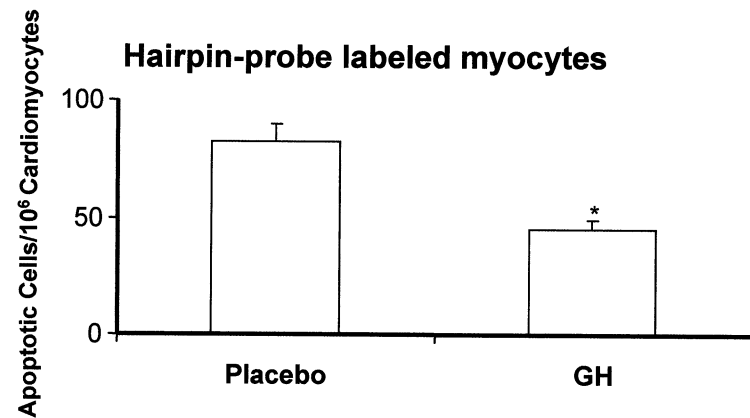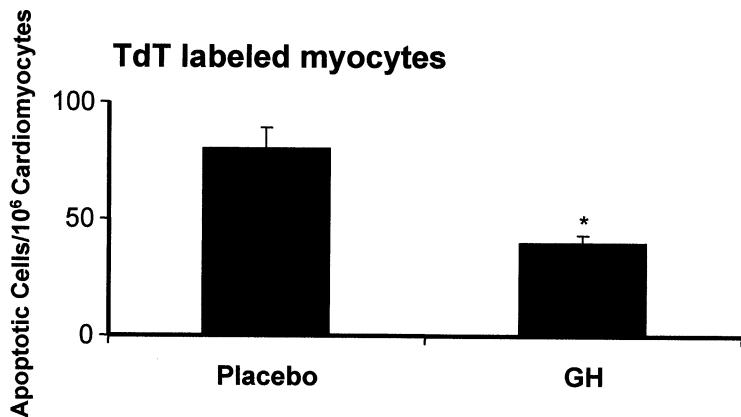
Figure 4. Prevalence of cardiomyocyte apoptosis in the two study groups as measured by terminal deoxynucleotidyl transferase (TdT) and hairpin assays. The apoptotic index was assessed as described in Methods and yielded similar results with the two techniques. *p < 0.05 vs. placebo group. GH = growth hormone.

Figure 5. Cardiomyocyte apoptosis in remote areas from infarction observed under fluorescent light. **Panels A, B, and F** depict representative triple staining obtained in the same microscopic fields from a growth hormone (GH)-treated rat euthanized at one month. **Panel A** shows cardiomyocyte and non-cardiomyocyte nuclei stained with DAPI. **Panel B** depicts cardiomyocyte immunostained with anti-sarcomeric actin. **Panels C to F** depict detection of terminal UTP nick end labeling (TUNEL)-positive cardiomyocyte nuclei. **Panels C and D** are representative images from placebo and GH group, respectively, after the 13-month observation period. **Panels E and F** are representative images from placebo and GH rats, respectively, euthanized at one month. Note the higher percentage of positive nuclei in the placebo-treated animals. **Panel G**: Immunostaining for activated caspase-3 antibody. **Panel H**: Double-exposure photomicrograph of dual labeled cardiomyocytes for activated caspase-3 (red signal) and TUNEL. Note the co-localization of caspase activity and TUNEL-positivity in the same cardiomyocyte. Overlapped signals result in **yellow** fluorescence. **Bars** = 20 μm.
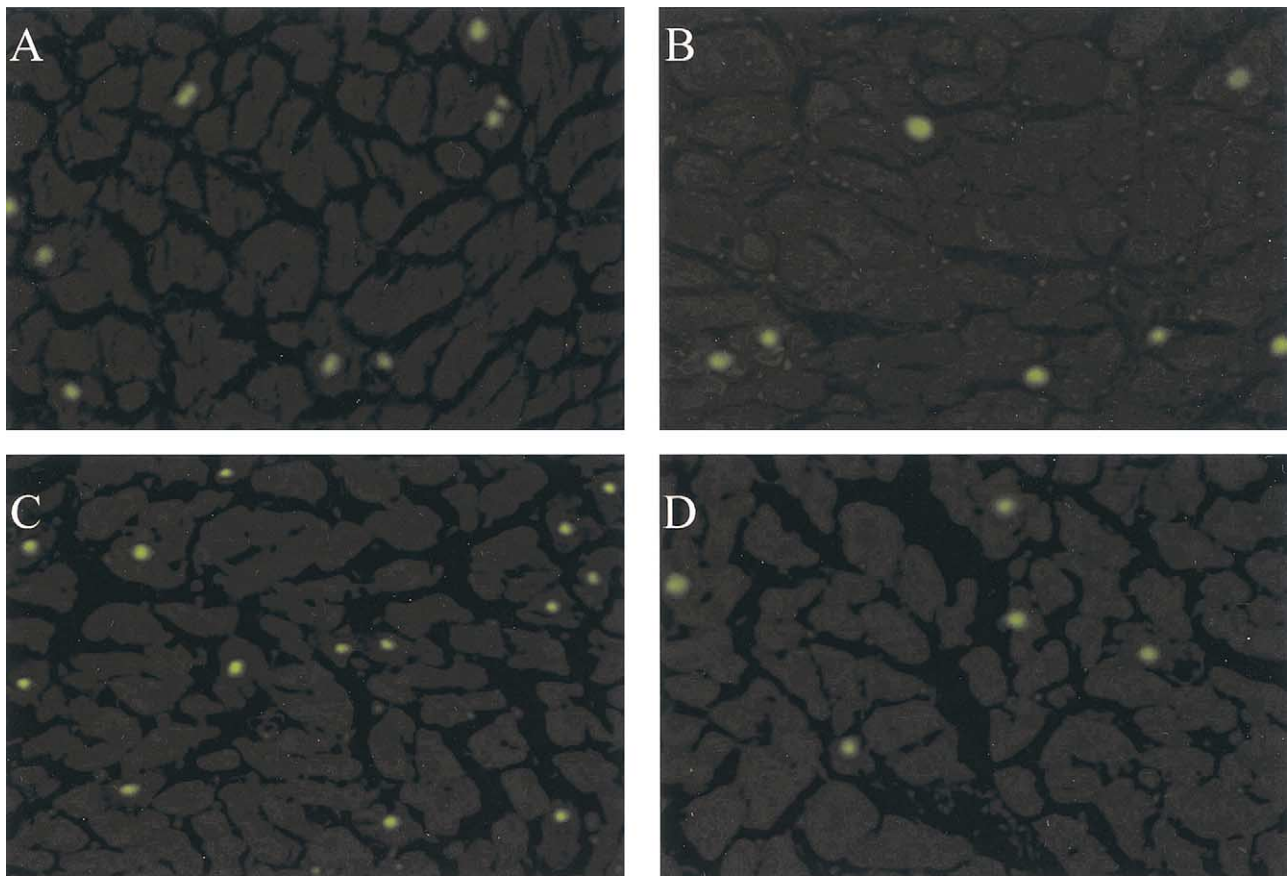
Figure 6. Detection of apoptosis in cardiomyocyte nuclei of left ventricular area remote from infarction by hairpin probe. The oligo was recognized by a streptavidin fluorescin conjugate. **Panels A and B** are representative images from the placebo and the growth hormone (GH) group, respectively, after the 13-month observation period. **Panels C and D** are representative images from placebo and GH rats euthanized soon after the active treatment period (1 month). **Bar** = 20 μm.

filled with interstitial fluid arising from myocardial edema, which subsequently leads to accumulation of fibrous tissue. In other words, GH might have the potential to prevent rather than reduce fibrosis (18). The marked reduction of the apoptotic index exhibited by GH-treated animals strongly supports this hypothesis. Our findings of decreased apoptotic death following GH/IGF-I activation are congruent with recent data obtained both in animals treated with exogenous GH or IGF-I and in transgenic mice overexpressing IGF-I (10–12).

Comparison with previous work. The magnitude of survival benefit in the current study (20%) was lower than that obtained with high doses of angiotensin-converting enzyme inhibitors, which ranged from 25% to 40% (14,19). Noteworthy, in our study the two survival curves began to clearly diverge around day 200 and continued to do so thereafter, showing the largest separation during the last weeks of observation. This implies that GH in some way altered the underlying mechanisms of the disease progression. In contrast, the survival curves, observed when angiotensin-converting enzyme inhibitors or beta-blocking agents were used in animal or human CHF (20), start to diverge at an

early stage, but then become parallel. This suggests attenuation or loss of effectiveness of neurohormonal antagonism as heart failure progresses.

Previous studies addressed the effects of GH on the nonmyocyte compartment (21,22). High-dose GH administration to normal rats for 80 days slightly decreased CVF, without modifying the collagen I/III ratio (22), in line with earlier observations in normal and failing rats (3,4). In the aggregate, the data available suggest that only long-term GH excess, such as that occurring in acromegaly, induces fibrosis with histopathologic signs of inflammation of the myocardial interstitium (1), whereas in the short term GH does not stimulate myocardial fibrosis, as shown by animal and human studies (23).

A very recent study examined the long-term effects of GH in a rat model of acute MI (24). At variance with the present study, GH treatment was initiated very early (on the day of MI). Growth hormone reduced the infarct size and increased the relative survival rate by 36% at 52 weeks. The study did not look at the effects of GH on myocardial histology and LV function. However, the interesting observation was made that GH downregulated the expression of

fetal genes regarded as markers of CHF progression, such as the atrial natriuretic factor, alpha-smooth muscle actin, and beta-myosin heavy chain. In agreement with our data, SERCA-2 was unaffected by GH. Particularly interesting was the observation that GH prevented the overexpression of extracellular matrix genes, such as collagen I, collagen III, and fibronectin. The data are in close agreement with the current study and complement our observations based on direct examination of myocardial tissue structure.

Clinical implications. Whereas GH and IGF-I have almost invariably been beneficial in experimental heart failure, previous studies in CHF have provided inconsistent results (2,6–9). The most likely explanation for this discrepancy resides in the fact that patients with CHF present with a wide spectrum of GH/IGF-I conditions, encompassing GH deficiency and severe GH resistance (25). In the former, GH administration only acts as a replacement therapy, whereas in the latter GH is unable to cause sufficient generation of its effector IGF-I. Indeed, when the authors of the largest clinical study of GH in CHF (6) re-analyzed their data by discriminating those patients who did not respond to GH administration with substantial IGF-I production, they could demonstrate significant effects of GH on LV function (9).

As discussed in a recent review (26), the perfect surrogate end point in CHF does not exist yet, and mortality is still regarded as the standard for assessing the efficacy of a new therapeutic strategy. In the present study, GH did not improve LV systolic function despite its effect to prolong survival. This finding bears several implications. First, it supports the stance that questions the validity of hemodynamic changes as surrogate end points of mortality. Second, the data point to the importance of structural changes in the myocardium and apoptosis, events that are not usually addressed in clinical studies. Ultimately, the effects of GH and IGF-I on the interstitium and apoptosis here reported are among those most consistently found in previous studies, and this encourages testing of the hypothesis that GH may affect the progression of clinical CHF.

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