Hypotensive and Renal Effects of Diazoxide, a Sodium-Retaining Benzothiadiazine Compound

By C. Bartorelli, M.D., N. Gargano, M.D., G. Leonetti, M.D., and A. Zanchetti, M.D.

The well-known hypotensive effect of several saluretic compounds of the benzothiadiazine series has commonly been ascribed to their saluretic properties, hypotension resulting either from hypovolemia consequent to extracellular sodium loss or from various changes in body water and electrolyte balance. Hypotension has recently been raised by the report that 3-methyl-7-chloro-1,2,4-benzothiadiazine-1,1-dioxide (SRG 95213, diazoxide), a compound differing from the known benzothiadiazine drugs by the elimination of the sulfamyl group from the benzenoid moiety, has no natriuretic or diuretic activity, although retaining definite hypotensive properties. The following is a report of our clinical experience with this drug during the last months: particular attention has been paid not only to the blood pressure effects of diazoxide, but also to its action on kidney function and on water and electrolyte balance.

Materials and Methods

The drug has been tested either by single or by repeated administration in 20 patients with primary arterial hypertension of mild to moderate degree. Several subjects had some secondary vascular, myocardial, or kidney involvement, but none had history or signs of congestive heart failure. All subjects except three were hospitalized.

Arterial blood pressure was measured with the auscultatory method each morning (or each second morning in the three outpatients) by the same observer, with the patient in the lying and then in the standing position. Sodium and potassium were measured in blood and urine by flame photometry; urine chloride was determined by the method of Volhard and Harvey, as modified by Peters and van Slyke; urine bicarbonates were measured in a manometric van Slyke apparatus, and titratable acidity was measured according to Henderson and Palmer. Plasma volume was determined in the fasting subject by the Evans-blue dye method, specimens being drawn at 15 minutes for determining dye dilution. Body weight was measured daily with a precision scale. Glomerular filtration rate and renal plasma flow were computed by measuring inulin and paraaminohippurate clearances. Statistical analysis was carried out according to Snedecor.

Results

Effects of Diazoxide on Arterial Pressure

Single Dose Administration

Diazoxide was administered to eight hypertensive patients in single oral doses of 100 to 300 mg. Control blood pressure was taken in the lying position after the patient had become accustomed to the experimental conditions, and when at least four consecutive readings had given concordant results. In all subjects but one, arterial pressure started to decrease after 30 minutes and reached the lowest level within 60 to 90 minutes after oral administration, the maximal effect being maintained for at least 4 hours. The decrease in mean arterial pressure ranged from 10 to 55 mm. Hg (fig. 5). It should be noted that the only patient who did not show a hypotensive response was the only one receiving a 100-mg. dose of the drug.

Prolonged Administration

Prolonged administration of diazoxide was tested in 17 hypertensive subjects, 14 of them being hospitalized and three outpatients. The experimental period was delayed for 5 to 7 days after admission until the blood pressure remained at the same level for 3 consecutive days. During the experimental period, which ranged from 20 to 102 days in the different patients, placebo and diazoxide were administered according to a crossed design, and in a few subjects hydrochlorothiazide, alone or in association with diazoxide, was also tested.
Diazoxide was administered by the oral route in single daily doses of 200 to 400 mg, for periods extending up to 49 days.

For the whole period of treatment with diazoxide there was a decrease of recumbent systolic blood pressure averaging 32, of recumbent diastolic pressure averaging 17, of standing systolic pressure averaging 27, and of standing diastolic pressure averaging 15 mm. Hg. An analysis of variance with multiple classification of average pressure values during prolonged placebo or diazoxide administration (table 1) shows that both systolic and diastolic blood pressures were consistently and durably decreased by the drug both in the lying and the standing positions ($p < .01$). A further analysis of variance indicates that in our patients the drug was equally effective either in the lying or in the standing position (table 2).

All patients tested responded favorably, at least to a 400-mg. dose of diazoxide, except one subject. It should be noted, however, that only cases of mild to moderate severity were studied. Blood pressure decrease was always rapidly achieved upon taking the drug, and promptly disappeared when diazoxide was substituted by a placebo (fig. 1). No instance of tolerance to the drug was noticed, at least for the relatively short periods of our observations (up to 49 days); however, phases of transient escape from blood pressure control, lasting 1 or 2 days, occurred not infrequently during prolonged treatment. Although a statistical analysis was not attempted, due to the limited number of our observations, in the six patients tested, the addition of hydrochlorothiazide (25 to 50 mg. daily) to diazoxide had hypotensive effects larger than those induced by either drug alone (fig. 2).

**Effects of Diazoxide on Water and Electrolyte Excretion, Plasma Volume, and Body Weight**

**Single Dose Administration**

This study was carried out in six hypertensive patients maintained in a metabolic ward at a constant diet containing 140 mEq. of sodium, 45 mEq. of potassium, 135 mEq. of chloride, and 1,500 ml. of water. Sodium, potassium, chloride, bicarbonate, hydrogen ion, and water excretion was measured in three 2-hour samples, taken at 11 a.m., 1 p.m., and 3 p.m. and in three 6-hour samples, taken at 9 p.m., 12 p.m., and 6 a.m. Both placebo and drug were given in a single dose at 9 a.m.

<table>
<thead>
<tr>
<th>Source of variation</th>
<th>Degrees of freedom</th>
<th>Syst. BP</th>
<th>Diast. BP</th>
<th>Syst. BP</th>
<th>Diast. BP</th>
</tr>
</thead>
<tbody>
<tr>
<td>Patients</td>
<td>16</td>
<td>18,082**</td>
<td>3,223*</td>
<td>18,007**</td>
<td>2,429</td>
</tr>
<tr>
<td>Diazoxide vs. placebo</td>
<td>1</td>
<td>12,364**</td>
<td>3,486**</td>
<td>9,194**</td>
<td>2,339**</td>
</tr>
<tr>
<td>Interaction</td>
<td>16</td>
<td>4,574</td>
<td>421</td>
<td>3,984</td>
<td>442</td>
</tr>
<tr>
<td>Error</td>
<td>16</td>
<td>3,556</td>
<td>1,158</td>
<td>4,919</td>
<td>1,408</td>
</tr>
</tbody>
</table>

$^*P < 0.05; \ ^{**}P < 0.01$

<table>
<thead>
<tr>
<th>Source of variation</th>
<th>Degrees of freedom</th>
<th>Syst. BP placebo</th>
<th>Syst. BP diazoxide</th>
<th>Diast. BP placebo</th>
<th>Diast. BP diazoxide</th>
</tr>
</thead>
<tbody>
<tr>
<td>Patients</td>
<td>16</td>
<td>51,168**</td>
<td>20,471</td>
<td>3,763**</td>
<td>2,586</td>
</tr>
<tr>
<td>Lying vs. standing</td>
<td>1</td>
<td>100</td>
<td>0</td>
<td>63</td>
<td></td>
</tr>
<tr>
<td>Interaction</td>
<td>16</td>
<td>1,316</td>
<td>483</td>
<td>361</td>
<td>149</td>
</tr>
<tr>
<td>Error</td>
<td>16</td>
<td>936</td>
<td>7,539</td>
<td>540</td>
<td>2,026</td>
</tr>
</tbody>
</table>

$^*P < 0.05; \ ^{**}P < 0.01$

### Table 1

**Analysis of Variance of Systolic and Diastolic Blood Pressure: Diazoxide vs. Placebo Treatment**

### Table 2

**Analysis of Variance of Systolic and Diastolic Blood Pressure: Lying vs. Standing Values**
EFFECTS OF DIAZOXIDE

The excretion values for at least 4 days after initiation of the diet were discarded, and only those of the 2 days immediately preceding diazoxide administration were used as controls. In each patient, 200 and 400 mg. of diazoxide were successively tested, a week interval being allowed between the two trials (table 3). Excretion of water, as well as of sodium, chloride, and bicarbonate ions was decreased by administration of diazoxide, a fact confirmed by analyses of variance with multiple classification (tables 4 and 5). There was no clear change in potassium and hydrogen ion excretion, but the limited size of our sample does not allow any definite conclusion on this point.

The effects on the water and electrolyte balance of diazoxide and hydrochlorothiazide are compared in figure 3, which refers to one of the patients of the study. While hydrochlorothiazide is a powerful natriuretic, as well as chloruretic agent, diazoxide is a sodium and chloride-retainig drug (chloride is not shown in figure 3, but runs parallel to sodium); while hydrochlorothiazide induces a slight, though significant, alkalinization of the urine associated with an increase in bicarbonate excretion, diazoxide administration results in urine acidification and in decrease of bicarbonate output. Thus, at least for single doses, the patterns of urinary excretion induced by the two types of benzothiadiazine compounds appear altogether opposite.

Prolonged Administration

Although urinary electrolyte excretion was measured only in acute experiments to avoid the uncertainties of long-lasting balance studies, there is evidence that the sodium-retaining effect of diazoxide is not short-lived. Indeed, it is apparent from the examples of figures 1 and 2 that prolonged administration of either 200 or 400 mg. of diazoxide daily resulted in an important augmentation of plasma volume and body weight. While mean increase in plasma volume has been 488 ml. in several instances, especially when 400-mg. doses were used, plasma-volume expansions exceeding 1 liter were observed. Maximal increase in body weight was 3.7 Kg., and the mean change 1.2 Kg. One patient had a decrease in plasma...
Table 3
Effects of 200 and 400 mg of Diazoxide on Water and Electrolyte Excretion during 12 or 24 Hours Following Oral Administration of the Drug

<table>
<thead>
<tr>
<th>Treatment</th>
<th>H(_2)O (ml.)</th>
<th>Na(^+) (mEq.)</th>
<th>K(^+) (mEq.)</th>
<th>Cl(^-) (mEq.)</th>
<th>HCO(_3^-) (mEq.)</th>
<th>H(^+) (mEq.)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Placebo, 200 mg.</td>
<td>802 ± 239</td>
<td>1469 ± 226</td>
<td>9.2 ± 10.9</td>
<td>68.2 ± 16.8</td>
<td>3.09 ± 2.71</td>
<td>5.91 ± 3.63</td>
</tr>
<tr>
<td>Diazoxide, 200 mg.</td>
<td>692 ± 120</td>
<td>1209 ± 383</td>
<td>19.7 ± 5.5</td>
<td>4.19 ± 3.73</td>
<td>3.78 ± 1.95</td>
<td>7.04 ± 2.55</td>
</tr>
<tr>
<td>Placebo, 400 mg.</td>
<td>769 ± 273</td>
<td>1409 ± 305</td>
<td>18.3 ± 3.4</td>
<td>2.89 ± 3.68</td>
<td>1.90 ± 0.64</td>
<td>4.92 ± 2.27</td>
</tr>
<tr>
<td>Diazoxide, 400 mg.</td>
<td>589 ± 253</td>
<td>931 ± 229</td>
<td>17.0 ± 3.5</td>
<td>84.3 ± 28.2</td>
<td>17.0 ± 3.5</td>
<td>5.76 ± 2.92</td>
</tr>
</tbody>
</table>

Means (±s) and standard deviations (s) of six hypertensive patients. Values indicated under “placebo” are means of values measured during the 2 days preceding administration of diazoxide.

Table 4
Analysis of Variance of Water and Electrolyte Excretion: Diazoxide, 200 mg., Treatment (24 Hours Excretion)

<table>
<thead>
<tr>
<th>Source of variation</th>
<th>Degrees of freedom</th>
<th>H(_2)O</th>
<th>Na(^+)</th>
<th>K(^+)</th>
<th>Cl(^-)</th>
<th>HCO(_3^-)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Patients</td>
<td>5</td>
<td>882,215</td>
<td>3,400</td>
<td>935</td>
<td>1,717</td>
<td>37</td>
</tr>
<tr>
<td>Diazoxide 200 mg. vs. Placebo</td>
<td>1</td>
<td>272,798</td>
<td>3,502**</td>
<td>69</td>
<td>2,242**</td>
<td>8</td>
</tr>
<tr>
<td>Interaction</td>
<td>5</td>
<td>366,474</td>
<td>2,925</td>
<td>49</td>
<td>2,927</td>
<td>8</td>
</tr>
<tr>
<td>Error</td>
<td>6</td>
<td>767,884</td>
<td>1,136</td>
<td>291</td>
<td>1,169</td>
<td>10</td>
</tr>
</tbody>
</table>

*P <0.05; **P <0.01

Table 5
Analysis of Variance of Water and Electrolyte Excretion: Diazoxide, 400 mg., Treatment (24 Hours Excretion)

<table>
<thead>
<tr>
<th>Source of variation</th>
<th>Degrees of freedom</th>
<th>H(_2)O</th>
<th>Na(^+)</th>
<th>K(^+)</th>
<th>Cl(^-)</th>
<th>HCO(_3^-)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Patients</td>
<td>5</td>
<td>1,180,134</td>
<td>6,879</td>
<td>220</td>
<td>4,815</td>
<td>16</td>
</tr>
<tr>
<td>Diazoxide, 400 mg. vs. Placebo</td>
<td>1</td>
<td>933,297*</td>
<td>18,881*</td>
<td>19</td>
<td>5,274*</td>
<td>105*</td>
</tr>
<tr>
<td>Interaction</td>
<td>5</td>
<td>245,785</td>
<td>3,098</td>
<td>74</td>
<td>741</td>
<td>9</td>
</tr>
<tr>
<td>Error</td>
<td>6</td>
<td>600,385</td>
<td>9,734</td>
<td>115</td>
<td>3,675</td>
<td>51</td>
</tr>
</tbody>
</table>

*P <0.05
volume and body weight, suggesting a natriuretic effect.

It should be pointed out that no constant correlation was found between changes in plasma volume and body weight in the patients receiving diazoxide (fig. 4). The correlation coefficient is only $r = 0.363$, and its significance is 0.101. The two regression coefficients are $b_{2,1} = 1.29$ and $b_{1,2} = 0.101$. The lack of relationship between the two sets of data is also indicated by partitioning the sum of squares of one variate (body weight changes), which demonstrates that the variation cannot effectively be reduced by the regression (table 6). As plasma volume is known to be a fixed portion of the extracellular fluid compartment, the changes in body weight induced by diazoxide do not appear only to reflect variations in extracellular fluid volume, but are likely to result from additional disturbances in water balance.

We are aware that the interpretation of daily changes in body weight as variations in body water is not without uncertainty, but in the absence of more direct measurements body weight changes smaller than those expected from the variations in plasma volume might tentatively suggest that during treatment with diazoxide water retention is associated to fluid transfer from the intracellular to the extracellular compartment.

Finally, it can be remarked that, in spite of these readjustments in water balance, significant changes in the plasma concentrations of sodium, potassium, and chloride have never been observed.

**Effects of Diazoxide on Renal Function**

**Single Dose Administration**

In eight hypertensive patients, glomerular filtration rate and renal plasma flow have been measured by clearance technics immediately preceding and following oral administration of 100 to 300 mg. of diazoxide. Simultaneously,
Likewise, nitrogen excretion was definitely increased; no decrease; also pressure was reduced; GFR increased after administration of the drug, and filtration rate was unchanged in one, and slightly decreased in four. Prolonged administration of the drug, generally increased arterial blood pressure and urinary sodium excretion were also measured (fig. 5). Glomerular filtration rate increased, although moderately, in four subjects, was practically unchanged in three, and was slightly decreased in one. Renal plasma flow greatly increased in three subjects, had a slighter increment in one, and did not show any consistent change in the remaining four. Arterial pressure and urinary sodium excretion definitely decreased in all subjects, except in the one patient who received 100 mg. of the drug. The various changes occurred at rather different times in the different patients, but they generally became evident within 1 hour from the administration of the drug, persisting 2 or more additional hours.

**Prolonged Administration**

In six patients glomerular filtration rate and renal plasma flow were measured before and after several days of diazoxide administration (table 7). Although mean arterial pressure was markedly decreased in all cases, in no instance was glomerular filtration rate also decreased; to the contrary, it was definitely increased in at least four patients. Likewise, renal plasma flow increased strikingly in two subjects and slightly in two, and was decreased, although moderately, in the remaining two.

### Table 6

<table>
<thead>
<tr>
<th>Source of variation</th>
<th>Degrees of freedom</th>
<th>Sum of squares</th>
<th>Variance</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean</td>
<td>1</td>
<td>36,966,400</td>
<td></td>
</tr>
<tr>
<td>Deviations due to regression</td>
<td>1</td>
<td>4,494,489</td>
<td></td>
</tr>
<tr>
<td>Random deviations from regression</td>
<td>23</td>
<td>29,940,444</td>
<td>1,301,758</td>
</tr>
<tr>
<td>Total</td>
<td>25</td>
<td>71,460,000</td>
<td></td>
</tr>
</tbody>
</table>

### Table 7

**Effects of Prolonged Administration of Diazoxide on Renal Functions**

<table>
<thead>
<tr>
<th>Patient</th>
<th>Before treatment</th>
<th>After treatment</th>
<th>Days</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>MBP mm. Hg</td>
<td>GFR ml./min.</td>
<td>RPF ml./min.</td>
</tr>
<tr>
<td>M.V.</td>
<td>128</td>
<td>65</td>
<td>307</td>
</tr>
<tr>
<td>B.I.</td>
<td>140</td>
<td>66</td>
<td>255</td>
</tr>
<tr>
<td>B.A.</td>
<td>163</td>
<td>42</td>
<td>184</td>
</tr>
<tr>
<td>F.G.</td>
<td>130</td>
<td>80</td>
<td>396</td>
</tr>
<tr>
<td>M.C.</td>
<td>150</td>
<td>78</td>
<td>273</td>
</tr>
<tr>
<td>F.N.</td>
<td>127</td>
<td>78</td>
<td>282</td>
</tr>
</tbody>
</table>

MBP, mean arterial pressure; GFR, glomerular filtration rate; RPF, renal plasma flow.

Side Effects of Prolonged Treatment with Diazoxide

In spite of the conspicuous increase of plasma volume, no patient developed pitting edema during treatment with diazoxide. This may have resulted from our careful selection of subjects with no sign of congestive heart failure. However, a patient with ischemic heart disease had exacerbation of cardiac pain and worsening of the electrocardiographic signs of altered myocardial repolarization during diazoxide treatment with important expansion of plasma volume. Only one patient had some gastrointestinal troubles; no subject complained of weakness, tachycardia, or other disturbances. One subject who had received diazoxide (200 to 400 mg. daily) for 20 days in the hospital and had subsequently been discharged on a mixed treatment with diazoxide (400 mg.) and hydrochlorothiazide (50 mg.), returned to our hospital 3 days later with symptoms of acute pancreatic necrosis. Because of his death, plasma diastase activity was estimated in seven hypertensive patients.
EFFECTS OF DIAZOXIDE

receiving diazoxide. Diastase activity was found to increase above the upper normal level (160 Somogyi units) in three subjects, but some increase, though within the normal range, occurred in two other patients. No consistent change in fasting blood glucose concentration was observed. Increased diastase activity above 160 units occurred also in seven hypertensive patients treated with hydrochlorothiazide for various time, thus confirming a previous study on the subject.12

Discussion

While we do not know of any extensive report on the clinical action of diazoxide, our observations seem well consistent with a few reports that have recently appeared in abstract form.13, 14 These observations on the hemodynamic and renal effects of the drug are worthy of comment. In mild and moderate hypertensive patients diazoxide appears to be an effective antihypertensive agent, affecting diastolic as well as systolic blood pressure with no appreciable difference either in the lying or the standing position. This finding suggests that hypotension is mainly obtained through arteriolar vasodilatation rather than through reduction of cardiac output. Expansion of plasma volume hints that cardiac output may even be increased during diazoxide treatment.

The sodium-retaining action of diazoxide is of some interest. It is quite marked and associated with water retention, and tends to have cumulative effects during prolonged treatment. While other antihypertensive drugs, for example guanethidine, are known to retain sodium and water,15 the action of diazoxide is peculiar: indeed, the sodium-retaining effect of guanethidine results from a simultaneous decrease in glomerular filtration rate, but sodium retention is induced by diazoxide in spite of an increase, or at least no change, in glomerular filtration. This suggests that diazoxide affects tubular handling of electrolytes, at the same site, though in the opposite way as the classical benzothiadiazine drugs with saluretic activity. Furthermore, our observation that during prolonged treatment with diazoxide the increase in plasma, and presumably in extracellular fluid volume, does not parallel the increase in body weight, and presumably in total body water, suggests that diazoxide administration may induce a shift in water from the intracellular to the extracellular compartment. Should this hypothesis be confirmed by more direct measurements of intracellular and extracellular fluid volumes, diazoxide might be postulated to act more broadly on the ion exchange at the cell membrane rather than strictly on renal tubular mechanisms only.

The marked hypotensive effect of a benzothiadiazine compound deprived of saluretic action might appear to weaken considerably

![Figure 5](http://circ.ahajournals.org/content/XXVII/5/901/F5.large.jpg)

Changes in arterial pressure and renal function produced by a single oral dose of diazoxide. The drug was given at 0 minute. The numbers on the ordinates are the differences between the control values and those read at the time indicated on the abscissa.
the current hypothesis that hypotension induced by chlorothiazide and its analogues is due to sodium and water loss.\textsuperscript{1, 2} Although the latter hypothesis has already been shown to hold only during early treatment,\textsuperscript{2-4, 6} the late hypotensive effects of the saluretic benzothiadiazines have been postulated to result from the continued loss of potassium from the intracellular environment and redistribution of ions across the cell membrane.\textsuperscript{6} While a direct vasodilator effect of diazoxide cannot be ruled out in the absence of crucial evidence against it, it does not seem without significance that this drug is also endowed with a primary action on electrolyte and water exchange at the renal tubular, and probably at the cell membrane, level. The hypothesis that both natriuretic and sodium-retaining benzothiadiazine compounds affect blood pressure through some ion disturbance seems worthy of further investigation.

Finally, the clinical usefulness of diazoxide as an antihypertensive agent should be discussed. On the one hand, its definite effectiveness, at least in mild and moderate cases, on both systolic and diastolic pressures with no difference either in the lying or the standing position, the frequently observed increase in glomerular filtration rate and in renal plasma flow suggest it as a very convenient drug in the treatment of moderate hypertension. On the other hand, the consequences of sodium retention may cause some concern. Although we have not observed any case of edema, it is conceivable that edema and congestive heart failure might well develop in hypertensive patients with impaired heart function. Prolonged administration of diazoxide for several months should help in clarifying whether sodium retention and increase in plasma volume are continuing during progressive treatment or are gradually compensated in late periods.

The problem of pancreatic toxicity should be carefully investigated, though it seems to be common to treatment with all benzothiadiazine compounds.\textsuperscript{12, 16, 17} If an aggravation of this potential danger is discounted, the association of sodium-retaining and saluretic benzothiadiazine drugs might be of some advantage in permitting a good hypotensive response without gross fluid retention or electrolyte loss.

Summary

Diazoxide, a new compound of the benzothiadiazine group, has been shown to decrease both systolic and diastolic arterial pressure in mild or moderate hypertensive patients, even though it lacks the saluretic properties characteristic of the other known compounds of the group.

Instead of promoting sodium excretion, diazoxide appears to be a powerful sodium- and chloride-retaining agent, its prolonged use leading to important increases in plasma volume and body weight, and probably to some redistribution between extracellular and intracellular fluid. Glomerular filtration rate and renal plasma flow are often increased during treatment with the drug.

Although diazoxide was in several respects a useful hypotensive agent, at least in hypertensive patients without heart failure, it may share with other benzothiadiazine compounds some pancreatic toxicity, a side effect that should be carefully considered during benzothiadiazine treatment.

Acknowledgment

We are grateful to Schering Corporation, Bloomfield, New Jersey and to Schering Co., Milan, Italy, for kindly providing us with diazoxide.

References


Circulation, Volume XXVII, May 1963
EFFECTS OF DIAZOXIDE


Claude Bernard and Medical Science

Not until after the turn of the century did the movement which Claude Bernard had foreseen make itself felt. To-day it is well established and should be generally recognized. The result has already been a remarkable increase of experimental investigation and of rational theorizing in the clinic. For the first time mathematics, physics, chemistry and physical chemistry, as aids to physiology, have passed into the hospitals. I believe that, for the reasons which Claude Bernard has explained, this will long remain the way of medical progress and that we have now definitely entered upon the epoch of experimental medicine.—L. J. Henderson. Introduction. Claude Bernard, M.D. An Introduction to the Study of Experimental Medicine. New York, The Macmillan Company, 1927, p. xi.
Hypotensive and Renal Effects of Diazoxide, a Sodium-Retaining Benzothiadiazine Compound

C. BARTORELLI, N. GARGANO, G. LEONETTI and A. ZANCHETTI

Circulation. 1963;27:895-903
doi: 10.1161/01.CIR.27.5.895

Circulation is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 1963 American Heart Association, Inc. All rights reserved.
Print ISSN: 0009-7322. Online ISSN: 1524-4539

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://circ.ahajournals.org/content/27/5/895

Permissions: Requests for permissions to reproduce figures, tables, or portions of articles originally published in Circulation can be obtained via RightsLink, a service of the Copyright Clearance Center, not the Editorial Office. Once the online version of the published article for which permission is being requested is located, click Request Permissions in the middle column of the Web page under Services. Further information about this process is available in the Permissions and Rights Question and Answer document.

Reprints: Information about reprints can be found online at:
http://www.lww.com/reprints

Subscriptions: Information about subscribing to Circulation is online at:
http://circ.ahajournals.org//subscriptions/