

Anti-Ischemic Effects of an 80-mg Tablet of Isosorbide Dinitrate in Sustained-Release Form Before and After 2 Weeks Treatment with 80 mg Once Daily or Twice Daily

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Summary: The purpose of this study was to investigate whether the anti-ischemic effects of a single 80-mg tablet of isosorbide dinitrate in sustained-release form are attenuated after 2 weeks of twice-daily administration. In order to follow a double-blind protocol with respect to the 12-h interval, we also evaluated, in randomized order, another group of patients receiving the tablets once daily. Parameters for assessment of the anti-ischemic effects were changes in exercise-induced ST-segment depression and the simultaneously recorded left ventricular ejection fraction, as determined by radionuclide ventriculography. The ST-segment depression was measured completely blind; the left ventricular ejection fraction was calculated automatically with a clinically validated software. In the group that received the tablets once daily ($n = 10$) none of the patients showed any signs of attenuation of the beneficial anti-ischemic effects. However, seven of the 12 patients who received the tablets twice daily demonstrated significant attenuation. Thus, based on analysis of the presently available studies, in order to guarantee maintenance of the anti-ischemic benefits, a once-daily high dose of ISDN in sustained-release form (80 mg or even higher) is recommended.

Zusammenfassung: Ziel der vorliegenden Untersuchung war es, festzustellen, ob die antiischämische Wirkung einer 80-mg-Einzeltablette Isosorbiddinitrat (ISDN)-retard nach zweiwöchiger Einnahme in 12stündigen Abständen noch voll erhalten ist. Um ein – im Hinblick auf das 12stündige Einnahmeintervall – doppelblindes Protokoll durchzuführen, untersuchten wir zusätzlich randomisiert eine zweite Patientengruppe, die während zweier Wochen morgens eine 80-mg-Tablette ISDN-retard und abends eine äußerlich identische Placebotablette erhielt. Als Prüfparameter der antiischämischen Wirkung dienten die Änderungen der belastungsinduzierten ST-Streckensenkung und die der gleichzeitig mittels Radionuklidventriculographie bestimmten linksventrikulären Auswurffraktion. Die Auswertung der ST-Streckensenkung erfolgte blind, die linksventrikuläre Auswurffraktion wurde automatisch anhand eines klinisch erprobten Computerprogramms berechnet. Bei allen Patienten, die das ISDN-retard 1× täglich erhielten ($n = 10$), war nach chronischer Gabe die antiischämische Wirkung unverändert erhalten.

Dagegen zeigten 7 der 12 Patienten, die das ISDN-retard in 12stündigen Intervallen einnahmen, eine deutliche Wirkungsabschwächung bzw. einen Wirkungsverlust. Somit scheint zur Aufrechterhaltung der vollen antiischämischen Wirkung von ISDN in retardierter Form die 1× tägliche Gabe einer relativ hohen Einzeldosis (80 mg oder sogar höher) empfehlenswert.

Key words: tolerance development, coronary artery disease

The discussion about the development of tolerance to the anti-ischemic effects of nitrates becomes increasingly controversial, as recent placebo-controlled clinical studies in patients with coronary artery disease have demonstrated a complete loss of the initial positive effects on exercise-induced ST-segment depression after 8 weeks of treatment with 3×20 mg, 3×40 mg, and 3×60 mg/day isosorbide dinitrate (ISDN) in sustained-release form (4), as well as a remarkable temporal reduction of the initial benefits on development of angina during exercise after chronic administration of 4×15 mg, 4×30 mg, 4×60 mg, and 4×120 mg/day ISDN (40). Another clinical study with isosorbide-5-mononitrate (5-ISMN), the main metabolite of ISDN, reported the failure to diminish the exercise-induced increase of pulmonary artery pressure, combined with a reduced exercise duration after 4 weeks of treatment with 3×50 mg/day (19). These studies have been confirmed by other authors who had earlier been doubtful about the antianginal benefits of chronic therapy with ISDN, administered three or four times daily (2, 24). A recently published animal study investigated the dependence of tolerance development on the intervals between administration of 5-ISMN, and found no change in response after chronic treatment with 2×20 mg/day administered every 12 h – a relatively high

dosage for dogs (37). Since tolerance studies applying relatively high single doses at 12-h intervals had not yet been performed with human subjects, we investigated the anti-ischemic effects of a single 80-mg tablet of ISDN administered twice daily in sustained-release form after 2 weeks of treatment. In order to follow a double-blind protocol with respect to the 12-h intervals, we also studied another group of patients, who received the 80-mg tablet only once daily.

Patients and Methods

Patients

In this study patients were included only if they fulfilled all of the following criteria: 75 % or more stenosis of at least one of the major coronary arteries, documented by selective coronary angiography; left ventricular ejection fraction ≥ 55 % without signs of prior myocardial infarction in the biplane contrast ventriculogram; chronic, stable, exercise-inducible angina; reproducible ST-segment depression ≥ 1.5 mV (measured .08 s after the J-point in one of the precordial leads), associated with the development of angina. The ST-segment depression had to have been shown to respond to nitrates. Only patients were included with sinus rhythm, and without bundle branch block, preexcitation syndromes, ST-T depression at rest, and complex arrhythmias, documented by 24-h Holter monitoring.

Further prerequisites were a history of no or minimal nitrate headache and a high likelihood of good cooperation. Thus, between October 1982 and May 1983, after informed consent was obtained, 22 patients were studied. Nine had one-vessel, seven had two-vessel, and six had three-vessel disease. Ten patients received the 80-mg ISDN tablet once daily and 12 twice daily.

Study Design

Each patient first had a single-blind, inpatient placebo period of 4 days, ingesting tablets visually identical with the 80-mg ISDN tablets twice daily. During this period none of the patients complained of resting angina. On the morning (usually at about 7:00 a.m.) of the 5th day, hereafter called the 1st day of the study, exercise electrocardiography and radionuclide ventriculography were performed simultaneously to determine the placebo ST-segment depression and the placebo exercise left ventricular ejection fraction (Fig. 1).

After the end of exercise patients received their first tablet of 80 mg ISDN in sustained-release form (Isoket Retard 80, Pharma Schwarz, Monheim, FRG). Simultaneous recordings of exercise ECG and exercise radionuclide ventriculography were repeated 2 h, 6 h, and 12 h after ingestion. After the 12-h exercise patients were randomized to two groups receiving 80-mg tablets of ISDN either once or twice daily as outpatients for a period of 14 days. As the patients of

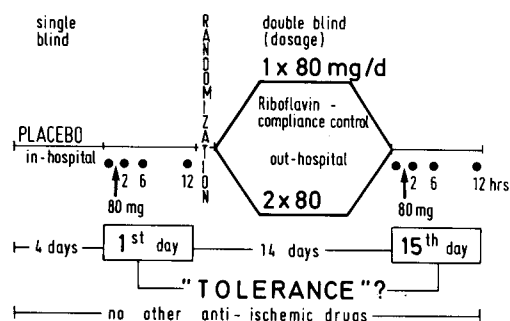


Fig. 1. Graphic representation of study design. Dots refer to simultaneously recorded exercise ECG and radionuclide ventriculography.

the once-daily group received placebo tablets every evening, this phase was performed in a double-blind manner with respect to the dosage. To enhance compliance each patient received the tablets in a package individually inscribed with the date, day, and time for each single tablet to be ingested. In addition, a diary for documentation of the ingestion of each single tablet was given to each patient. Compliance was further enhanced during this period by telephone calls with the patients as well as with their physicians. For measurement of compliance from the 2nd to the 14th day (the tablets of the first and fifteenth day were ingested in the presence of the investigators), the riboflavin-fluorescence method was used: Each 80-mg ISDN tablet (and each placebo tablet) was put into a capsule to which 10 mg riboflavin was added. The patients were asked to avoid all food with relatively high riboflavin content and to fill all of the provided vials with urine about 2–4 h after each capsule ingestion. At the end of the 2 weeks, exercise ECG and exercise radionuclide ventriculography were performed (15th day) in exactly the same way as described above for the 1st day: After the first exercise recordings, representing the values for 12 h after ingestion of the last 80-mg tablet in the twice-daily group and the values for 24 h after the last 80-mg administration in the once-daily group, an 80-mg tablet of ISDN was given again to evaluate the anti-ischemic response after chronic treatment. The results of the 15th day were compared with those of the 1st day to assess the possible development of tolerance. With the exception of sublingually administered nitroglycerin or ISDN at the onset or just before expected angina, no other anti-ischemic drugs were administered during the study, in particular no beta-blockers and no calcium antagonists.

Exercise

The patients exercised on a bicycle ergometer in semisupine position (30° inclination), with legs below heart level. Based on previous exercise tests, the first workload was 50 or 80 watts for 3 min. If there was no or only minimal angina, workloads were increased every 3 min by steps of 30 watts (Bosch apparatus, ELP 500). Thus, the end point of exercise during placebo was always angina combined with a ST-seg-

ment depression $\geq .15$ mV. The end point of exercise after ingestion of the 80-mg ISDN tablet was the termination of the maximal placebo workload. The heart rate at each workload was determined from the ECG recordings, the blood pressure was measured using the standard cuff sphygmomanometry, always on the right arm and always by the same person.

Electrocardiography

After removing patients' chest hair at the sites of V_1 - V_6 , special silver electrodes were used to reduce radiation attenuation, which could influence the results of radionuclide ventriculography. The electrodes fixed in the morning remained attached throughout the day, thus eliminating variations due to electrode placing within the same day. Electrocardiograms were derived from the standard 12-lead system with a three-channel recorder (Schiller, Switzerland) at a paper speed of 50 mm/s at 1-min intervals. ST-segment depression was determined .08 ms after the J-point, only in the precordial lead, which showed the maximal ST-segment depression during the placebo exercise test. Measurements of ST-segment depression were performed completely blind, i.e., without knowledge of the patient's name, hour of registration, or whether the ECG was being recorded on the 1st or the 15th day of the study.

Radionuclide Ventriculography

Radionuclide ventriculography was performed with a 10 inch-crystal mobile gamma camera (Picker, DynaMo) and an on-line computer system (Medtronic-MDS/A²) according to the standard equilibrium multiple-gated technique, after labeling patients' red blood cells with about 25 mCi (ca. 9×10^8 Bq) 99m-Technetium. Images were obtained at 21 frames per cardiac cycle in a 64×64 modified byte-mode matrix (for further details see (35)). During exercise, registration started 1 min after the beginning of each workload; thus, acquisition time per workload was 2 min, while a relatively steady state was maintained with respect to the heart rate. The data were stored on 80-Mbyte discs and evaluated with commercially available software (MDS). Briefly, the left ventricular border was automatically determined by the computer within a rectangle placed by the operator arbitrarily around the left ventricle. The algorithm used identifies the left ventricular border on a frame-by-frame basis, using constant thresholds and second derivative criteria. Each frame was reviewed by the operator. Background was selected by the computer 5 pixels away from the inferolateral segment of the left ventricle. Ejection fraction was calculated from the background-corrected counts of the end-diastolic frame, determined from the peak of the left ventricular time-activity curve, and from the counts at the end-systolic frame, determined at the nadir of the curve. Ejection fractions calculated with this computer program correlate well with those determined from biplane con-

trast ventriculography, and are highly reproducible (28, 29).

Data Analysis

All comparisons of 1st-day values with those of the 15th day were made for identical levels of exercise, i.e., the maximal possible workload for each patient in the placebo exercise test. The data are expressed as mean \pm one standard deviation. Comparisons of the mean values for the 1st and 15th days as well as for different times of registration were made using the Student's t-test for paired data. The criterion of significance was $P < .05$. To answer the question of tolerance development by intraindividual analysis it is necessary to set a cutoff for definition of a "diminished effect". For reasons of accuracy and reproducibility of the parameters used in this study, we made the cutoff-threshold for ST-segment depression at a difference of .1 mV between the 1st and the 15th day and at left ventricular ejection fraction during exercise of 10 % ("ejection fraction units"). Thus, individual tolerance was reached when the differences of the ST-segment depression and/or the left ventricular ejection fraction during exercise between the 1st and the 15th day were above the corresponding thresholds 2 h and/or 6 h after drug administration.

Results

Compliance

Of 572 urine specimens (22 patients \times 13 days \times 2 vials/day), 543 revealed unequivocal positive fluorescence, 16 were equivocal, and two were unequivocally negative; 11 vials were not turned in. Assuming even the worst case, i.e., all but the unequivocally positive were due to noningestion of the tablets, the resulting compliance for the interval from the 2nd to the 14th day was 95 %.

Anti-ischemic Effects

Once-daily Group

In this group of ten patients the placebo values (1st day) and the values for 24 h after ingestion of the last 80-mg ISDN tablet (15th day) were identical for both ST-segment depression ($.21 \pm .06$ mV versus $.20 \pm .07$ mV) and exercise ejection fraction ($52 \% \pm 8 \%$ versus $54 \% \pm 8 \%$). There was a remarkably identical reduction in ST-segment depression 2 h after administration of an 80-mg ISDN tablet on the 1st and the 15th day ($.03 \pm .05$ mV versus $.03 \pm .06$ mV), combined with a highly significant unattenuated increase in exercise ejection fraction to $67 \% \pm 9 \%$ on the 1st and to $65 \% \pm 11 \%$ on the 15th day. The effects on ST-segment depression and exercise ejection fraction were nearly the same 6 h after ingestion as 2 h after: $.03 \pm .04$ mV versus $.03 \pm .05$ mV and $70 \% \pm 11 \%$ versus $70 \% \pm 9 \%$. After 12 h a comparison of the values of the 1st and the 15th days also showed no differences in ST-segment depression ($.13 \pm .09$ mV versus $.12 \pm .09$

mV) and ejection fraction ($68\% \pm 6\%$ versus $64\% \pm 6\%$). Compared with the values at 2 h and 6 h after drug administration, the anti-ischemic effects were diminished 12 h after ingestion, but still significantly present on both the 1st and the 15th day. Analysis of individual data revealed the unattenuated anti-ischemic effects of a single 80-mg ISDN tablet in each patient with respect to both parameters.

Twice-Daily Group

In this group of 12 patients the placebo values (1st day) and the values 12 h after ingestion of the last 80-mg ISDN tablet on the 15th day showed no significant differences for ST-segment depression ($.24 \pm .07$ versus $.22 \pm .08$ mV) or for ejection fraction during exercise ($52\% \pm 16\%$ versus $54\% \pm 11\%$). However, 2 h and 6 h after drug administration there was a significant attenuation of the positive effects on ST-segment depression between the 1st and the 15th day, with $.07 \pm .07$ versus $.13 \pm .08$ mV and $.09 \pm .06$ versus $.15 \pm .08$ mV respectively. In addition, the ejection fraction during exercise was significantly lower on the 15th day compared with that of the 1st day, 2 h after ingestion ($62\% \pm 13\%$ versus $56\% \pm 12\%$). After 6 h the ejection fraction of the 15th day showed a tendency towards decreasing effects compared with that of the 1st day ($62\% \pm 12\%$ versus $58\% \pm 12\%$, n.s.). After 12 h the ejection fraction during exercise still revealed an attenuation ($61\% \pm 12\%$ versus $56\% \pm 14\%$). The ST-segment depression, which only reached the level of significance 12 h after administration on the 1st day ($.20 \pm .06$ mV) was not different after the 2 weeks of treatment, either in the morning of the 15th day (12 h after ingestion the night before: $.22 \pm .08$ mV) or in the evening of the 15th day (12 h after following ingestion: $.22 \pm .08$ mV). Analysis of the individual changes in ST-segment depression between the 1st and 15th day revealed a diminished anti-ischemic effect in seven of the 12 patients, ranging between 33% and 88% (mean: 58%). Three of these seven patients demonstrated an attenuation 2 h and 6 h after drug administration, three patients only 2 h after and one patient only 6 h after. Five of these seven patients also demonstrated a partial loss – most of them even a total loss – of the effects on ejection fraction during exercise: an attenuation of 82% was present in one patient 2 h after the administration. The remaining four patients had a complete loss of the effects on ejection fraction during exercise either 2 h (two patients) or 6 h (two patients) after administration.

Discussion

This study shows that the anti-ischemic effects of a single 80-mg tablet of isosorbide dinitrate (ISDN) in sustained-release form were considerably attenuated after 2 weeks of twice-daily administration at 12-h intervals, whereas the anti-ischemic effects were com-

pletely maintained if a once-daily regimen was followed (Fig. 1 + 2).

We did not assess the anti-ischemic effects according to such parameters as the rate of anginal attacks and nitrate consumption, since these parameters are highly subjective and depend additionally on changes in the environment. The parameter of "exercise tolerance" (i.e., the time between the beginning of a stress test and the point at which angina develops or becomes of "moderate severity") is also influenced by subjective information and the determination may be biased unless a true double-blind protocol is followed, which is nearly impossible in tolerance studies, since the investigators are usually always aware of the acute or chronic moment of the exercise test. We therefore assessed only the more objective parameters at identical levels of exercise. In addition to the generally accepted and easy-to-measure ST-segment depression we determined the left ventricular ejection fraction, as it gives direct information on pump function during exercise and seems – according to several working groups – to be more sensitive than ST-segment depression in the evaluation of myocardial ischemia. Based on intraindividual analysis, the maximal attenuation of ST-segment depression after administration of ISDN was 88%, with a mean of about 60%, whereas the maximal attenuation of left ventricular ejection fraction during exercise was 100%, with a mean of 96%. The smaller number of patients showing attenuation of ejection fraction as compared with the number showing attenuation of ST-segment depression is explained by the choice of the cutoff threshold. Nevertheless, this does not influence the maximal values of attenuation (i.e., only partial loss of the effects on ST-segment depression but complete loss of the effects on ejection fraction) present in four of five patients showing attenuation.

The anti-ischemic effects of the nitrates are caused essentially by an improvement in the loading condi-

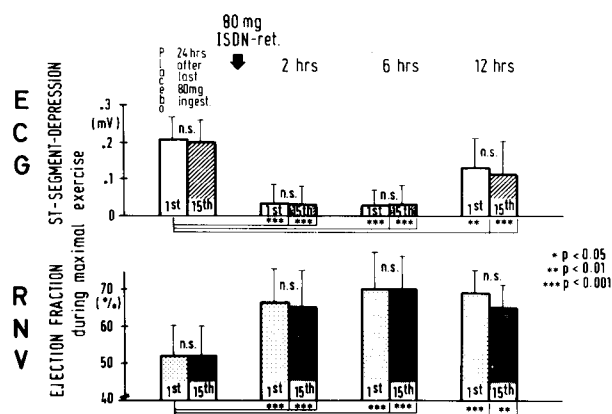


Fig 2. Anti-ischemic effects of an 80-mg tablet of sustained release ISDN before (1st day) and after (15th day) two weeks of treatment with these tablets once daily. The upper panel represents the ST-segment depression (ECG), the lower panel the left ventricular ejection fraction (RV). The bars show the mean values \pm one standard deviation of the initial values as well as 2, 6, and 12 h after ingestion of the 80-mg tablet ISDN on the 1st and 15th day.

Nevertheless, the existence and importance of nitrate tolerance development with respect to anti-ischemic effects is a problem of everyday practice, and a matter of highly controversial discussion: while only a few studies (4, 19, 40, this study) have demonstrated a diminution or absence of the anti-ischemic effects after chronic treatment with orally administered nitrates, the majority of published papers report sustained antianginal action (10, 22, 42), unattenuated improvement in "exercise tolerance" (9, 22, 42), persistent reduction in exercise-induced ST-segment depression (3, 22, 27, 33, Schneider et al., this volume) and unchanged beneficial effects on pulmonary artery pressure and left ventricular end-diastolic pressure (27). After cutaneous application as well there were no signs of diminished anti-ischemic effects (8, 11, 16, 30). The initial study by Blasini et al. (4), which demonstrated a complete loss of the effects on ST-segment depression after 8 weeks of treatment with 3×20 , 3×40 , and 3×60 mg ISDN, was recently criticized by Abrams (1), because "apparently not each individual subject was tested acutely with ISDN". A consecutive study, however, in which every patient was tested acutely (Rudolph et al., this volume) revealed the same result after 2 weeks of treatment with 4×40 mg ISDN/day, even in non-sustained-release form. The study of Thadani et al. (40), which demonstrated diminished nitrate effects on the duration of the improvement of "exercise tolerance" after sustained treatment with 4×15 , 4×30 , 4×60 , and 4×120 mg ISDN/day may also have some limitations because of the restrictions concerning the parameter of "exercise tolerance" mentioned above. The same objection, however, must then also

What, then, are the possible reasons for the contradictory findings? The contradictory results are explained neither by the different use of sustained- and non-sustained-release forms of nitrates, nor by the differing intervals between administration and (first) measurements of 30 min (19), 1 h (33; Rudolph et al., Schneider et al., this volume), 2 h (22, this study), 3 h (4), or 4 h (42). Apart from one study in which the nitrate-free interval of 24 h before retesting could have restored vascular responsiveness (3), there have been different protocols of exercise with constant or graduated workloads in combination with walking on a treadmill (22, 40, 42), arm-assisted step test (33; Schneider et al., this volume), the use of bicycle ergometers in sitting position (9), semi-supine position with the legs below heart level (this study), or supine position with elevated legs (4). For elevation of the legs above heart level a significant attenuation of the nitrate-induced venodilatation has been demonstrated (43). The controversial issues may also be due to different distributions in the populations investi-

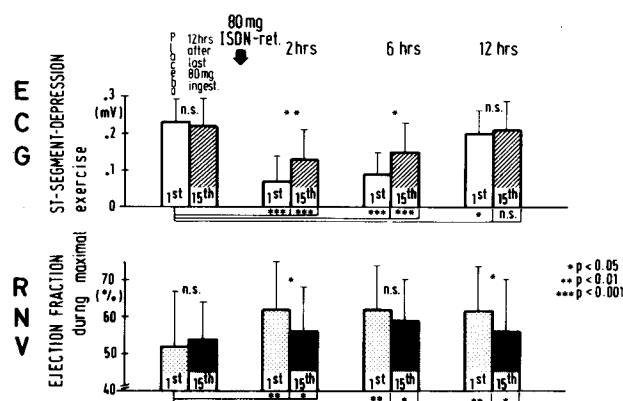


Fig. 3. Anti-ischemic effects before (1st day) and after (15th day) 2 weeks of treatment with 80-mg tablets of ISDN twice daily at 12-h intervals. See Fig. 2 for explanation.

gated with respect to the development of different degrees of attenuation, thus influencing the mean values and standard deviations. Unfortunately, there is no other study in which an intra-individual analysis based on cutoff thresholds was made to clarify this point. In the population of this study, the anti-ischemic effects of the 80-mg tablet after 2 weeks of twice-daily ingestion were significantly attenuated, but the mean values still showed a significant difference compared with placebo (Fig. 3). It is also conceivable that another group observed a tendency to attenuation after chronic treatment with ISDN, which, however, did not reach the level of significance (33).

One of the most encumbering points in interpreting presently available studies is the absence of documented patients' compliance (2, 3, 8, 9, 10, 11, 16, 19, 24, 27, 40). Assuming a rapid (i.e., within 36 h (5), or even about 15 h (Rudolph et al., this volume) restoration of vascular responsiveness, it is easy to imagine why irregular ingestion, which is very likely with patients receiving more than two tablets each day (41), blurs the problem of anti-ischemic tolerance. Simple methods for documentation of patients' compliance, such as tablet counting, or a diary for daily recording of ingestion, are not very reliable and can therefore only be of supportive value. Since determination of plasma-concentrations cannot be made on a day-by-day basis, we decided to use the riboflavin-fluorescence method, which permits the estimation of patients' compliance on a tablet-by-tablet basis, if they are administered at 12-h intervals. Thus, in our study the compliance was documented by the doctor and not by the patient. The limitations of the riboflavin-fluorescence method are that the patients must avoid food with high riboflavin content and that only a qualitative assessment can be made of the urine specimens. Nevertheless, the result of this study itself proves the reality of the calculated compliance of 95 %.

It is not clear why five of the 12 patients receiving the 80-mg tablets twice daily did not develop tolerance to the anti-ischemic effects. This may be explained only partially by the choice of the cutoff threshold. It is suspected that the plasma concentrations of the 5-ISMN metabolite dropped below a "critical level" in only some patients 12 h after ingestion of the 80-mg ISDN tablet – this level may be individually different – with subsequent restoration of vascular responsiveness. Since the metabolism of ISDN in the liver does not seem to depend on food intake and cigarette smoking (Fung and Parker, this volume) additional interindividual differences in the time needed for passage of the ISDN tablet through the small gut may be taken into account.

The problem of tolerance development might not be so essential if increasing dosages could circumvent the attenuated vascular response. However, animal studies did not reveal a mere parallel shift, but rather a diminished steepness of the dose-response curve (37).

Clinical data consistently indicate that the anti-ischemic effects after chronic treatment cannot be increased, even after eightfold higher dosages are administered (40).

Undoubtedly, long-term treatment with nitrates is desirable, because anginal pain probably represents only the tip of the iceberg compared with the number of silent episodes of transient myocardial ischemia (12, 34, 42). But since obviously not all patients can be tested for individual development of attenuation, a once-daily high dosage of ISDN in sustained-release form is recommended, based on analysis of the presently available studies, in order to guarantee complete maintenance of the anti-ischemic benefits. The optimal time of day for administration of this dose (80 mg or even higher) can be determined from the patient's history, possibly with the assistance of ST-segment Holter monitoring.

Acknowledgement. The authors wish to express their appreciation to Margot Vogel for her expert and skillful technical assistance.

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