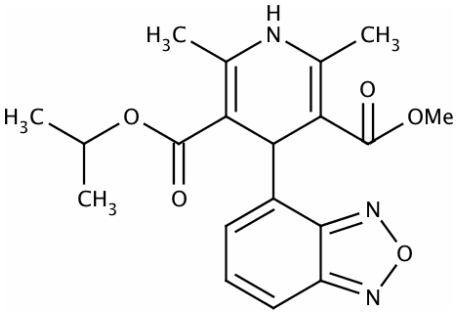
CINAPS COMPOUND DOSSIER

Isradipine



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NATIONAL INSTITUTE OF NEUROLOGICAL DISORDERS AND STROKE

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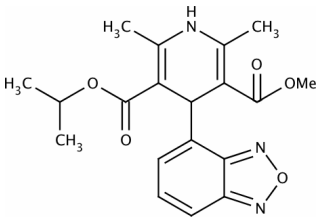
I. Compound Information

Common name: Isradipine

Structure:



Pubchem ID: 3784 **Mol. Formula:** $C_{19}H_{21}N_3O_5$ **FW:** 371.39

CASRN: 75695-93-1 **Polar surface area:** 103.55 **logP:** 2.07

IUPAC name: 3-Methyl 5-propan-2-yl 4-(2,1,3-benzoxadiazol-4-yl)-2,6-dimethyl-1,4-dihydropyridine-3,5-dicarboxylate

Other names: PN 200-110; DynaCirc

Drug class: Antihypertensive; Calcium Channel Blocker; Vasodilator

Medicinal chemistry development potential: High

II. Rationale

IIa. Scientific Rationale / Mechanism

When properly controlled, calcium fluxes across the plasma membrane and between intracellular compartments play critical roles in fundamental functions of neurons, including the regulation of neurite outgrowth and synaptogenesis,[1-2], synaptic transmission and plasticity, [3] and cell survival (i.e., calcium-mediated gene expression is the principle link between extracellular cues and enduring changes in cell function[4]). During aging, and in neurodegenerative disorders, cellular calcium-regulating systems are compromised resulting in synaptic dysfunction, impaired plasticity, neuronal degeneration, and death. Alterations of calcium-regulating proteins in the plasma membrane (ligand- and voltage-gated calcium channels, ion-motive ATPases, and glucose and glutamate transporters), endoplasmic reticulum (presenilin-1, Herp, and ryanodine and inositol triphosphate receptors), and mitochondria (electron transport chain proteins, Bcl-2 family members, and uncoupling proteins) are implicated in age-related neuronal dysfunction and disease.[5] The resulting oxidative stress, perturbed energy metabolism and aggregation of disease-related proteins (amyloid β-peptide, α-synuclein, huntingtin, etc.) further compromise cellular homeostasis. Ultimately, this renders neurons vulnerable to calcium-mediated apoptosis via induction/activation of pro-apoptotic proteins such as Bax, Par-4, and p53 leading to mitochondrial membrane permeability changes, release of cytochrome c and caspase activation.[6-8] It is important to note that the adverse effects of "normal" aging on neuronal calcium regulation are subject to modification by genetic (mutations in presenilins, α-synuclein, huntingtin, or Cu/Zn-superoxide dismutase; apolipoprotein E isotype, etc.) and environmental (dietary energy intake, exercise, exposure to toxins, etc.) factors that may cause or affect the risk of neurodegenerative disease.[5]

Although there are signs of distributed neuropathology, as judged by Lewy body (LB) formation, the motor symptoms of Parkinson 's disease, including bradykinesia, rigidity and resting tremor, are clearly linked with the degeneration and death of dopamine neurons in the substantia nigra pars compacta (SNc).[9-10] Unlike most neurons in the brain, these nigral dopaminergic neurons are autonomously active, with action potentials at 2–4 Hz in the absence of synaptic input. More importantly, these neurons are further differentiated from other neurons based upon their reliance upon L-type calcium channels to govern pacemaking, whereas most neurons use channels that allow sodium ions across the membrane to govern their activity. Further differentiating the L-type channels underlying the pacemaking activity in dopaminergic fibers in the SNc, these neuronal channels have a pore-forming Cav1.3 subunit, which constitute less than one quarter of the L-type channels in the brain. As a consequence, these neurons have a low affinity for dihydropyridines, and open at more hyperpolarized membrane potentials than do L-type calcium channels with a pore-forming Cav1.2 subunit, the more predominant neuronal type

of calcium channel. Indeed, the calcium channels underlying nigral dopaminergic pacemaker activity are more closely related to the L-type channels found in cardiac and smooth muscle. This particular composition of calcium channel and their use in dopaminergic neurons in the SNc (where the calcium channels are open much of the time because they are responsible for pacemaking) causes the magnitude of the intracellular calcium flux to be much larger than in other neuronal populations. As a result, the calcium flux entering these Cav1.3 containing neurons presents a greater burden on these cell, in that the calcium must be rapidly sequestered either in organelles lying below the plasma membrane or through ionic interactions with mobile buffering proteins before being escorted out of the cell. Because of the larger flux of calcium and the slow kinetics of the plasma membrane ion transporters and their restriction to the cellular surface, mitochondria and the endoplasmic reticulum are the principal organelles involved in sequestering the intracellular calcium, and these stores come at a considerable energetic cost to the cell as the endoplasmic reticulum utilizes ATP-dependent transporters and the mitochondrial calcium storage dissipate the electrochemical gradient created by respiratory metabolism along the electron transport chain. Thus, it appears that while in most neurons calcium flux and energy demands are relatively infrequent and episodic, the dopaminergic neurons of the SNc experience greater and more prolonged calcium flux, and the resulting cellular energy demands and oxidative stress are much more sustained. Ultimately, the calcium overload can act as a trigger for apoptosis, resulting in the death of these neuronal fibers critical for normal basal ganglia function and control of movement. Based on the critical role of calcium in apoptosis, any intervention that inhibits the influx of calcium into cells might prove to be protective for apoptosis-related human disorders.[10-12]

If Parkinson's disease is a long-term consequence of the accelerated oxidative stress or apoptosis in neurons that rely on calcium channels for pacemaking activity, than reducing this dependence should delay its onset and slow its progression. Evidence to support this hypothesis can be found in laboratory animals and in man. For example, in mice, the reliance of nigral dopaminergic neurons on calcium channels to drive autonomous activity is developmentally regulated. Specifically, young animals have neurons that are pacemakers, but generate this activity with channels that allow sodium ions to cross the membrane, not calcium. This juvenile mechanism appears to be retained in adult neurons, but becomes latent, since long term blockade of the L-type Cav1.3 calcium channels that underlie normal adult pacemaking reveals the juvenile mechanism, which still allows normal functioning. Most importantly, although the effect on mitochondrial oxidative stress can only be inferred at this time, switching the pacemaking mechanism of SNc dopaminergic neurons confers resistance to the mitochondrial toxins used to study Parkinson's disease. For example, pretreatment of brain slices for 2 hours with isradipine at a concentration shown to rejuvenate SNc DA neurons dramatically attenuated the rotenone-induced dendritic loss and fragmentation.[13] In agreement with the hypothesis that

toxicity was dependent upon pacemaking-related calcium loading, rotenone-induced dendritic fragmentation was not significantly attenuated by tetrodotoxin but was dramatically diminished in juvenile and Cav1.3-/- SNc DA neurons. Additional evidence is afforded by *in vivo* studies in laboratory rodents, where mice were systemically administered isradipine with slow release, subcutaneous pellets, mimicking oral administration in humans with controlled release format tablets. SNc DA neurons in brain slices obtained from mice treated for 7 days with slow-release iradipine were virtually insensitive to isradipine, in sharp contrast to age-matched, naive neurons, demonstrating that the switch in pacemaking could be accomplished *in vivo*. However, slow-release isradipine treatment failed to protect SNc DA terminals and cell bodies against large intrastriatal injections of the toxin 6-hydroxydopamine (6-OHDA), although it did provide a robust protection against a more modest intrastriatal 6-OHDA challenge.[11, 13] Isradipine-treated adult mice were also refractory to repeated injections of the toxin MPTP over the course of 5 weeks, where stereological analysis of TH-immunoreactive and Nissl-stained sections revealed that isradipine treatment reduced the MPTP-induced loss of SNc dopaminergic neurons by nearly half and prevented the development of motor deficits.[13] In man, evidence to support a calcium -mediated onset and progression of Parkinson's is seen in clinical studies with calcium channel modulators, like the dihydropyridines, which are commonly used in the treatment of hypertension. Specifically, treatment of hypertension with dihydropyridines was found to significantly reduce the observed rate of developing Parkinson's disease in a recent study in which the duration of treatment was factored into the analysis.[14] Another retrospective study of Parkinson's patients and calcium channel antagonists suggests that their use is less common among diseased patients than in the general population, again raising the possibility that these agents may exert a neuroprotective effect on Parkinson's disease onset and progression.[15]

IIb. Consistency

n/a

III. Efficacy (Animal Models of Parkinson's Disease)

IIIa. Animal Models: Rodent

The chronic administration of MPTP along with the adjuvant probenecid (MPTP/p), that blocks the clearance of the toxin and its metabolites, rapidly and persistently depletes ATP and increases reactive oxygen and nitrogen species molecules that induce cell death pathways. Over time this oxidative stress appears to select against adult dopamine neurons in the substantia nigra because their basal activity is driven by the energetically demanding, relatively rare, voltage-dependent, L-type calcium channel, thereby producing many of the pathological hallmarks and motor deficits of Parkinson's disease. It is thus an excellent choice for studies of pathogenesis, for testing neuroprotective therapies and developing biomarkers to detect the disease presymptomatically.[16]

in vivo experiments using adult mice implanted with extended release pellets with biodegradable-carrier bound isradipine (60 days, 3µg/g/day) or inert placebo pellets 1 week before MPTP/p toxin or vehicle treatment revealed that isradipine treatment improved motor performance compared to mice implanted with placebo. Additionally, although MPTP/p-treated mice with isradipine lost significantly more SNpc neurons than those treated with vehicle, isradipine attenuated the loss compared to MPTP/p-treated, placebo-implanted mice. Protection was not due to isradipine affecting MPTP metabolism, because brain 1-methyl-4-phenylpyridinium ion (MPP^+) levels did not differ between toxin-treated groups.[13, 16]

In similar studies in rats using the neuronal toxin 6-hydroxy-dopamine, isradipine was subcutaneously delivered to rats at the doses of 0.05, 0.1, or 0.2 mg/kg/day, for 4 weeks, starting the day after a unilateral nigrostriatal 6-OHDA lesion. Fourteen days later, L-DOPA treatment was initiated. In these studies, isradipine-treated animals displayed a dose-dependent reduction in L-DOPA-induced rotational behavior and abnormal involuntary movements. Dendritic spine counting at electron microscopy level showed that isradipine (0.2 mg/kg/day) prevented the 6-OHDA–induced spine loss and normalized preproenkephalin-A messenger RNA expression. However, involuntary movement induced by L-DOPA was not reduced by concomitant isradipine treatment.[17]

IIIb. Animal Models: Non-human Primates

n/a

IV. Efficacy (Clinical and Epidemiological Evidence)

IVa. Clinical Studies

There is one ongoing Phase II clinical trial of isradipine in Parkinson's disease entitled "Safety, Tolerability and Efficacy Assessment of Dynacirc CR in Parkinson Disease (STEADY-PD)" (ClinicalTrials.gov Identifier NCT00909545). This study is currently recruiting participants and is a research study of a controlled release formulation of isradipine (Dynacirc CR) to find out if it can be used safely, is tolerated by patients with early stage Parkinson Disease (PD) and if it slows the progress of the disease. This is a randomized, double blind, placebo controlled study lasting approximately 54 weeks. Each study participant is randomly assigned to one of the 4 doses being studied: placebo (inactive medication) or isradipine CR 5, 10 or 20 mg administered p.o. daily for 12 months.

IVb. Epidemiological Evidence

Evidence that calcium channel modulation holds therapeutic promise in humans is seen in clinical studies with calcium channel blockers, like the dihydropyridines, which are commonly used in the treatment of hypertension. Specifically, treatment of hypertension with dihydropyridines was found to significantly reduce the observed rate of developing Parkinson's disease in a recent study in which the duration of treatment was factored into the analysis.[14] Another retrospective study of Parkinson's patients and calcium channel antagonists suggests that their use is less common among diseased patients than in the general population, again raising the possibility that these agents may exert a neuroprotective effect on Parkinson's disease onset and progression.[15]

V. Relevance to Other Neurodegenerative Diseases

Cellular mechanisms for maintaining the homeostasis of cytosolic calcium concentration play an important role in aging, and it has been hypothesized that sustained changes in calcium homeostasis represent a common pathway for several neuropathological conditions.[18-19] For example, chronic elevation of calcium influx has been implicated in age-associated neuronal loss in the hippocampus and the learning and memory deficits observed in Alzheimer's disease. Electrophysiological studies of cultured hippocampal neurons have noted an increase in voltage-gated calcium current with age [20], as well as a significant positive temporal correlation between calcium current density and neuronal death.[21] Consistent with this relationship, reducing prolonged calcium-activated currents through chronic exposure to the L-type calcium channel antagonist nimodipine blocks a significant amount of hippocampal cell loss in culture. Moreover, age-related learning impairments in rabbits are similarly reversed by the specific L-type calcium channel blocker nimodipine.[22] Interestingly, the molecular basis for the increase in L-type channel activity during aging is not fully defined. Studies have demonstrated a ≈20% increase in expression of the Cav1.3 protein subunit α_{1D} in area CA1 of the hippocampus of aged rats; that these age-related increases in α_{1D} expression in area CA1 correlate with working memory impairment; and that chronic nimodipine treatment ameliorates the age-related working memory deficits and reduces expression of α_{1D} protein in the hippocampus.[23] In addition to these observations, a two-fold increase in cAMP-dependent protein kinase (PKA)-mediated phosphorylation of the L-type channel Cav1.2 has also been observed over time in laboratory animals, which may also contribute to the age-related increase in calcium influx.[24]

VI. Pharmacokinetics

VIa. General ADME

The pharmacokinetics of isradipine have been reported following oral administration of immediate-release or sustained-release formulations in healthy volunteers [25-29], and hypertensive patients.[30] At doses between 2.5 and 10 mg, isradipine in oral solution formulation was rapidly absorbed with the mean peak concentration occurring within 0.40 hours for all doses. A biphasic pattern of elimination was exhibited. No significant dose effect occurred with respect to tmax, elimination half-life, or apparent oral clearance, and mean elimination half-lives and mean residence times were similar across doses.[26] Similarly, a study in 18 normal male volunteers who received orally a single 5-mg dose, a single 20-mg dose, or repeated administration of 5 mg every 8 h for 13 doses of [^{14}C]isradipine demonstrated that the drug was rapidly and almost completely (90–95%) absorbed from the gastrointestinal tract, although the estimated bioavailability was only 17% due to extensive first-pass metabolism.[27] As before, the pharmacokinetics of isradipine appeared to be linear in the 5 to 20-mg dose range, as indicated by the dose-proportional blood levels of total radioactivity as well as the parent drug. Repeated administration of [^{14}C]isradipine showed no change in pharmacokinetic characteristics. During the 5 mg thrice daily regimen, steady-state blood levels of parent drug were reached in 2 days, while those of total radioactivity were reached in approximately 4 days, indicating effective half-lives of 8.8 h and 16 h. Once absorbed, isradipine appears to be completely metabolized prior to excretion. The recovery of radioactivity after both the 5 and the 20-mg dose was virtually complete within the experimental period, with a renal:fecal excretion ratio of ca. 70:30.[27]

Alternative formulations of isradipine have also been studied. For example, pharmacokinetic studies were carried out in 48 healthy male volunteers to determine the pharmacokinetic properties of a new modified-release formulation of isradipine (SRO) and to compare these with the pharmacokinetic properties of the original oral formulation of isradipine (NR). Studies were carried out in the fasting state and also following a light meal. Following the administration of an equivalent dose, the time required to reach maximum plasma concentration was extended from approximately 1.5 h with NR to 5 to 7 h with SRO, and the maximum plasma concentration was reduced by around 50%. The bioavailability of SRO was increased by less than 20% compared with the same daily dose given as NR, indicating that the two forms are bioequivalent and that no adjustment in the total daily dose of isradipine is necessary when switching from one form to the other. Administration with a light meal altered the pharmacokinetic parameters by less than 20% compared with administration without food.[28] The pharmacokinetic variables and antihypertensive effect of the calcium antagonist isradipine were similarly investigated in 30 hypertensive patients. isradipine was given orally in parallel group design in plain and slow release formulations in doses of 2.5 mg twice daily and 5.0 mg once daily, respectively. isradipine concentration in serum

was measured by a sensitive RIA method after the first dose and after 6 weeks of treatment. The pharmacokinetics and concentration/effect relationship after the first dose and after 6 weeks of treatment were compared. No differences in pharmacokinetics were observed between single and multiple dosing. Data were in accordance with results from studies in healthy volunteers. Rate, but not extent of bioavailability differs between the two isradipine formulations. Antihypertensive efficacy of the two formulations was similar (16/11 and 19/15 mmHg), and a significant time dependent increase in Emax from 10/3 mmHg to 23/14 mmHg after 6 weeks of treatment was observed.[30]

From these studies and others it has been shown that isradipine is 90%-95% absorbed and is subject to extensive first-pass metabolism, resulting in a bioavailability of about 15%-24%. isradipine is detectable in plasma within 20 minutes after administration of single oral doses of 2.5-20 mg, and peak concentrations of approximately 1 ng/mL/mg dosed occur about 1.5 hours after drug administration. Administration of isradipine with food significantly increases the time to peak by about an hour, but has no effect on the total bioavailability (area under the curve) of the drug. Both peak plasma concentration and AUC exhibit a linear relationship to dose over the 0-20 mg dose range. The elimination of isradipine is biphasic with an early half-life of 1½-2 hours, and a terminal half-life of about 8 hours. The total body clearance of isradipine is 1.4 L/min and the apparent volume of distribution is 3 L/kg.

Isradipine is 95% bound to plasma proteins such as human serum albumin and α1-acid glycoprotein.[31] It is completely metabolized prior to excretion, and no unchanged drug is detected in the urine. Six metabolites have been characterized in blood and urine, with the mono acids of the pyridine derivative and a cyclic lactone product accounting for > 75% of the material identified.[32-34]

VIb. CNS Penetration

Isradipine and other, structurally similar dihydropyridines are relatively lipophilic substances, highly plasma protein bound, and rapidly and efficiently cleared by the liver. However, these compounds cross the blood brain barrier.[35-38] Although it is often assumed that a highly plasma protein-bound drug is not available for transport through the blood brain barrier, studies have shown that many plasma protein-bound drugs are available for transport through the brain capillary endothelial wall. Since plasma proteins, per se, undergo negligible transcapillary transport in brain, transport through the blood brain barrier from the plasma protein bound pool represents a mechanism of enhanced dissociation of the drug from the plasma protein binding site within the brain microcirculation *in vivo*.[39] Indeed, while the brain extraction of isradipine in the presence of either albumin or α1-acid glycoprotein is inversely related to the protein concentrations in the presence of either albumin or α1-acid glycoprotein, it is higher than expected from the *in vivo* measurement of the dialyzable fraction.[40]

VIc. Calculated log([brain]/[blood])

–1.08 (Clark Model[41])

VII. Safety, Tolerability, and Drug Interaction Potential

VIIa. Safety and Tolerability

Headache, dizziness, tachycardia, nausea and tinnitus have been reported after isradipine administration.[26] Other side-effects and warnings are summarized in the *Dynacirc* (the GlaxoSmithKline isradipine formulation) drug package insert:

Blood Pressure:

While symptoms like syncope and severe dizziness have rarely been reported in hypertensive patients administered isradipine at the recommended dosage, because isradipine decreases peripheral resistance it may produce symptomatic hypotension.

Use in Patients with Congestive Heart Failure:

Acute hemodynamic studies in patients with congestive heart failure have shown that isradipine reduced afterload without impairing myocardial contractility. Nevertheless, it has a negative inotropic effect at high doses *in vivo*, and possibly in some patients. Therefore, caution should be exercised when using the drug in congestive heart failure patients, particularly in combination with a beta-blocker.

Carcinogenesis, mutagenesis, impairment of fertility:

Since apoptosis is a naturally occurring means of eliminating genetically altered, potentially malignant cells, prolonged administration of drugs that inhibit apoptosis could be anticipated to increase the incidence of cancer. A variety of studies have in fact indicated that the use of calcium antagonists is associated with an increased incidence of cancer.[42-44] However, a meta-analysis performed to compare the risk of serious adverse events (including cancer) associated with the use of all formulations of isradipine found no evidence for increased risk of serious adverse events in patients receiving isradipine as monotherapy for hypertension.[45]

Treatment of male rats for 2 years with 2.5, 12.5, or 62.5 mg/kg/day isradipine admixed with the diet (approximately 6, 31, and 156 times the maximum recommended daily dose based on a 50 kg man) resulted in dose dependent increases in the incidence of benign Leydig cell tumors and testicular hyperplasia relative to untreated control animals. These findings, which were replicated in a subsequent experiment, may have been indirectly related to an effect of isradipine on circulating gonadotropin levels in the rats; a comparable endocrine effect was not evident in male patients receiving therapeutic doses of the drug on a chronic basis. Treatment of mice for two years with 2.5, 15, or 80 mg/kg/day isradipine in the diet (approximately 6, 38, and 200 times the maximum recommended daily dose based on a 50 kg man) showed no evidence of oncogenicity. There was no evidence of mutagenic potential based on the results of a battery of mutagenic tests. No effect on fertility was observed in male and female rats treated with up to 60

mg/kg/day isradipine.

Pregnancy:

Isradipine was administered orally to rats and rabbits during organogenesis. Treatment of pregnant rats with doses of 6, 20, or 60 mg/kg/day produced a significant reduction in maternal weight gain during treatment with the highest dose (150 times the maximum recommended human daily dose) but with no lasting effects on the mother or the offspring. Treatment of pregnant rabbits with doses of 1,3, or 10 mg/kg/day (2.5, 7.5, and 25 times the maximum recommended human daily dose) produced decrements in maternal body weight gain and increased fetal resorption at the two higher doses. There was no evidence of embryotoxicity at doses which were not maternotoxic and no evidence of teratogenicity at any dose tested. In a peri/postnatal administration study in rats, reduced maternal body weight gain during late pregnancy at oral doses of 20 and 60 mg/kg/day. Isradipine was associated with reduced birth weights and decreased peri and postnatal pup survival.

There are no adequate and well controlled studies in pregnant women. The use of isradipine during pregnancy should only be considered if the potential benefit outweighs potential risks.

Nursing mothers:

It is not known whether isradipine is excreted in human milk. Because many drugs are excreted in human milk, and because of the potential for adverse effects of isradipine on nursing infants, a decision should be made as to whether to discontinue nursing or discontinue the drug, taking into account the importance of the drug to the mother.

Pediatric use:

Safety and effectiveness in pediatric patients have not been established.

VIIb.Drug Interaction Potential

The drug interaction potential of isradipine is also summarized in the *Dynacirc* drug package insert:

Hydrochlorothiazide: A study in normal healthy volunteers has shown that concomitant administration of isradipine and hydrochlorothiazide does not result in altered pharmacokinetics of either drug. In a study in hypertensive patients, addition of isradipine to existing hydrochlorothiazide therapy did not result in any unexpected adverse effects, and isradipine had an additional antihypertensive effect.

Propranolol: In a single dose study in normal volunteers, co-administration of propranolol had a small effect on the rate but no effect on the extent of isradipine bioavailability. Significant increases in AUC (27%) and Cmax (58%) and decreases in tmax (23%) of propranolol were noted in this study. However, concomitant administration of 5 mg b.i.d. isradipine and 40 mg b.i.d.

propranolol to healthy volunteers under steady-state conditions had no relevant effect on either drug's bioavailability. AUC and Cmax differences were <20% between isradipine given singly and in combination with propranolol, and between propranolol given singly and in combination with isradipine.

Cimetidine: In a study in healthy volunteers, a one-week course of cimetidine at 400 mg b.i.d. with a single 5 mg dose of isradipine on the sixth day showed an increase in isradipine mean peak plasma concentrations (36%) and significant increase in area under the curve (50%). If isradipine therapy is initiated in a patient currently receiving cimetidine, careful monitoring for adverse reactions is advised and downward dose adjustment may be required.

Rifampicin: In a study in healthy volunteers, a six-day course of rifampicin at 600 mg/day followed by a single 5 mg dose of isradipine resulted in a reduction in isradipine levels to below detectable limits. If rifampicin therapy is required, isradipine concentrations and therapeutic effects are likely to be markedly reduced or abolished as a consequence of increased metabolism and higher clearance of isradipine.

Warfarin: In a study in healthy volunteers, no clinically relevant pharmacokinetic or pharmacodynamic interaction between isradipine and racemic warfarin was seen when two single oral doses of warfarin (0.7 mg/kg body weight) were administered during 11 days of multiple-dose treatment with 5 mg b.i.d. Isradipine. Neither racemic warfarin nor isradipine binding to plasma proteins *in vivo* was altered by the addition of the other drug.

Digoxin: The concomitant administration of isradipine and digoxin in a single-dose pharmacokinetic study did not affect renal, nonrenal and total body clearance of digoxin.

Fentanyl Anesthesia: Severe hypotension has been reported during fentanyl anesthesia with concomitant use of a beta blocker and a calcium channel blocker. Even though such interactions have not been seen in clinical studies with isradipine, an increased volume of circulating fluids might be required if such an interaction were to occur.

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