Organic Process Research & Development **2000**, *4,* 477–487

Chemical Development of MDL 103371: An *N*-Methyl-D-Aspartate-Type Glycine Receptor Antagonist for the Treatment of Stroke†

Timothy J. N. Watson,* Stephen W. Horgan, Ramnik S. Shah, Robert A. Farr, Richard A. Schnettler, C. Richard Nevill, Jr., Franz J. Weiberth, Edward W. Huber, Bruce M. Baron, Mark E. Webster,‡ Rajesh K. Mishra,‡ Boyd L. Harrison,§ Phillip L. Nyce,|| Cynthia L. Rand,⊥ and Christian T. Goralski⊥

Aventis Pharmaceuticals, Chemical Development, 2110 East Galbraith Road, Building 51, Cincinnati, Ohio 45215-6300, U.S.A.

Abstract:

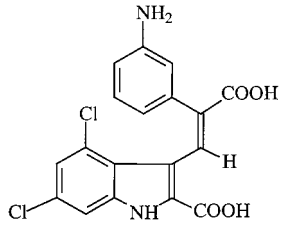
MDL 103371 is a *N*-methyl-D-aspartate (NMDA)-type glycine receptor antagonist for the potential treatment of stroke. Evaluation of five different synthetic routes, which included Stille, Suzuki, enol ether, Knoevenagel, and the Mukaiyama coupling reactions, revealed the Knoevenagel approach superior for preparing large quantities of drug substance for evaluation. The overall process utilized some classical chemistry. Fischer indole cyclization, followed by a Vilsmeier−Haack formylation and a Knoevenagel condensation gave immediate access into the proper carbon framework of the target molecule. A unique hydrogenation cataylst and solvent system for a nitro reduction, followed by a two step acid−base hydrolysis of a nitrile gave the crude product. Purification was accomplished by a potassium salt crystallization followed by a Schiff base formation to give MDL 103371 in nine steps in an overall yield of 38%.

Introduction:

Brain ischemia or concussive injury is accompanied by a dramatic rise in the extracellular concentrations of glutamate and aspartate. These amino acids function as neurotransmitter substances and are capable of activating an ion channel-linked receptor with a high permeability to calcium ions, the *N*-methyl-D-aspartate (NMDA)-type glutamate receptor. A variety of experimental observations link excessive or prolonged glutamate receptor stimulation with cell death occurring following ischemia or traumatic injury. Glutamate antagonists have been shown to be of benefit in animal models of stroke and trauma by greatly increasing neuronal survival, reducing animal mortality, and improving neurological outcome.

Glycine has been shown to be an obligatory co-agonist for the NMDA receptor activating at a site distinct from that utilized by glutamate. Thus, glycine antagonists are noncompetitive antagonists of glutamate and offer a potential mechanistic advantage since they do not have to compete with the ischemia-induced elevated levels of glutamate. We have demonstrated that glycine antagonists can antagonize NMDA receptor-mediated responses in vitro and in vivo. Therefore, glycine antagonists offer a novel mechanism to provide neuroprotection in stroke and trauma.

One such compound, MDL 103371 (**1**), was selected as a potential drug candidate for the treatment of stroke. It became necessary to develop a synthesis of **1** that would provide the multikilogram quantities needed for further development.



1

One can envision MDL 103371 (**1**) as a 3-aminophenylacetic acid linked to a dichloro-indole acid through an *E*-olefin, thus placing the diacid functionality in a *syn* spatial relationship. The FDA's policy[1] in developing geometric isomers states, "Geometric isomers and diastereomers therefore should, with the exception of cases where interconversion occurs, be treated as separate drugs and developed accordingly." In fact, in vivo interconversion from the *E* to *Z* isomer does not take place, and the *Z* isomer was determined to be inactive. Second, MDL 103371 is sensitive to isomerization in the presence of light and in a variety of organic and aqueous solutions. However, to our benefit, the *E* isomer is generally favored, depending on the conditions. Along with developing a synthesis that could prepare kilogram quantities of **1**, we had to address the isomerization and stability issues and develop a purification method to isolate the *E* isomer exclusively.

Route Selection

Potential routes will be discussed in general terms with an emphasis on olefin formation to form the proper carbon framework of the desired system and issues pertaining to

† This paper was presented at the Hope Symposium, Hope College, June 11, 1999, and the Organic Reactions & Processes Gordon Conference July 11-16, 1999, by Timothy J. N. Watson. This paper will follow the outline of the lecture presented and only incorporate the experimentals from the scale-up work.

* To whom correspondence should be addressed. Current Address: Pfizer Process Research & Development, Pfizer Inc., Eastern Point Road, Groton, CT 06340. E-mail: timothy_j_watson@groton.pfizer.com.

‡ Current address: Procter & Gamble, 8700 Mason-Montgomery Road, Mason, OH 45040-9462.

§ Current Address: Associate Director, Chemical Sciences Wyeth-Ayerst Research CN, 8000 Princeton, NJ 08543.

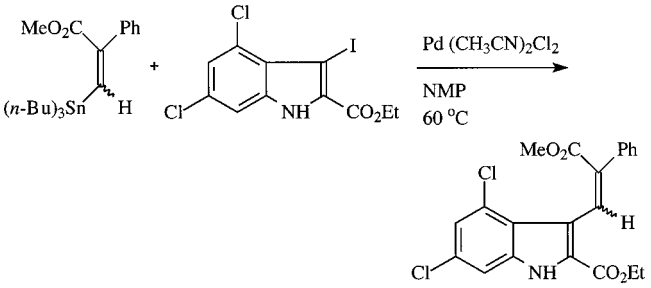
|| Current Address: Vertex Pharmaceuticals Inc., 130 Waverly Street, Cambridge, MA 02139.

⊥ Contract Manufacturing Services R&D, The Dow Chemical Company, 1710 Building, Midland, MI 48674.

(1) www.fda.gov/cder/guidance/stereo.htm.

10.1021/op000286s CCC: $19.00
Published on Web 09/20/2000

Scheme 1



scale-up. The routes discussed were developed for screening a variety of possible drug candidates where the aniline portion of the structure was altered; MDL 103371 (**1**) was the lead compound generated from this work. Chemical Development used the various routes to help prioritize our options. Complete details on the specific analogues will not be discussed.[2,3]

The first potential route utilizing the Stille Coupling[4] (Scheme 1) was easily eliminated. The prerequisite intermediates were coupled in the presence of bis(acetonitrile) palladium dichloride in NMP at elevated temperatures. The only analogue prepared via this route was the phenyl derivative which was obtained in 30% yield after extensive chromatographic purification. In addition to the product purification issues, the use of the iodinated indole and vinyl tin intermediate introduced waste issues and toxic handling hazards.

The second potential route utilized a Suzuki Coupling[5] as shown in Scheme 2. In this approach the indole aldehyde (product from a Vilsmeier−Haack) was protected as the *N*-tosylate. This was treated with an α-bromo-phosphonate ester[6] under Horner-Emmons conditions to give the desired (*Z*)-vinyl bromide in 65% yield after fractional crystallization from cyclohexane. Treatment of the vinyl bromide with a wide variety of arylboronic acids under some modified Suzuki conditions (method A,[7a] method B)[7b−d] gave the desired mixed esters with variable results. It was hoped that the coupled products would possess only the desired *E*-stereochemistry, thus simplifying purification and characterization of the products.

The Suzuki approach was attractive to prepare a wide variety of aryl analogues from a single late stage intermediate. The ability to differentiate between the two ester functionalities added diversity to the analogues. However, from a scale-up perspective it added two steps with a protection and deprotection of the indole. The complex reaction mixtures required chromatography and generally took long times for the reaction to reach completion. Anhydrous Suzuki conditions had to be developed because of the labile nature of the tosylate under standard conditions. This approach received a low priority. Any attempts to directly prepare the desired analogue with the appropriate Horner-Emmons reagent were unsuccessful. Some of the analogues prepared were the phenyl, *p*-chlorophenyl, 3,4-dichlorophenyl, 2- and 3-thiophene and furan, and the *m*-methoxyphenyl.

The initial attempts to prepare the *p*-fluoro analogue via the Suzuki coupling with the vinyl bromide and commercially available *p*-fluorophenylboronic acid used method A. However, because of the complexity of the product mixture obtained and the difficulty in purification of the product, a third approach was explored involving a Lewis acid catalyzed condensation reaction of the indole ester with an appropriate enol ether (see Scheme 3). A variety of enol ethers were condensed with the indole to give direct access to the proper carbon framework. This approach was the route that Chemical Development utilized to prepare the first sample of MDL 103371 (more specifics of this approach will be discussed in this section because we did use this work to make a 25 g sample). Reaction of 3-nitrophenylacetate with sodium methoxide and methyl formate, followed by MeI gave the enol ether in modest yield. Upon scaling up this chemistry, we found out the enol ether would slowly decompose at room temperature. The decomposition was difficult to handle on larger scale due to the longer times required to isolate the sample. Nevertheless, if one had pure enol ether (via quickly filtering through silica gel/chromatography), the reaction went very cleanly using TMSOTf, which provided the best yield and conversion to product; however, a full equivalent was required as the resulting formation of methanol quenches the catalyst. Second, if the ethyl ester of the indole was used, transesterification of the esters gave complex reaction mixtures. Reduction of the nitro group was accomplished with $SnCl_2$; however, the removal of the tin salt byproducts gave unacceptable filtration rates (to be discussed in depth in the Discussion). The *E/Z* ratio of the diester product was approximately 80:20. Hydrolysis with LiOH gave crude MDL 103371.

Purification of MDL 103371 could be accomplished by either chromatography of the diester prior to hydrolysis or by crystallization of **1** from 100 parts of methanol after hydrolysis. The chromatography was tedious, and the crystallization was problematic. It became clear from this work that aside from any synthetic approach an improved purification process was desperately needed. As a general comment, the scope of this procedure may be limited to enol ethers having electron-withdrawing or weak electron-donating groups, since the *m*- or *p*-methoxyphenyl- and 2- or

(2) (a) Salituro, F. G.; Baron, B. M.; Harrison, B. L.; Nyce, P. L. U.S. Patent 5,519,048, 21 May 1996. (b) Harrison, B. L.; Nyce, P. L.; Farr, R. A. U.S. Patent 5,563,157, 8 October 1996. (c) Farr, R. A.; Harrison, B. L.; Nyce, P. L. U.S. Patent 5,981,553, 9 November 1999.

(3) Baron, B. M.; Harrison, B. L.; Kehne, J. H.; Schmidt, C. J.; Gross, R. W.; van Giersbergen, P.; White, H. S.; Farr, R.; Siegel, B. W.; Slone, A. L.; Senyah, Y.; McCloskey, T. C.; Fadayel, G.; Taylor, V.; Murawsky, M.; Meikrantz, S.; Nyce, P.; Janowick, D.; Salituro, F. G. Pharmacological characterization of MDL 105,519, a systemically active antagonist of the NMDA receptor-associated glycine recognition site. *Eur. J. Pharmacol.* **1997**, 323, 181−192.

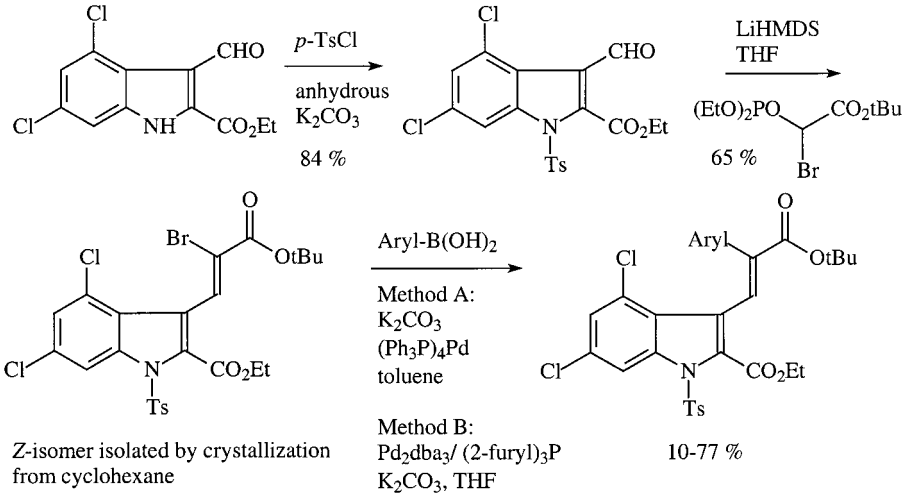
(4) (a) Stille, J. K.; Groh, B. L. *J. Am. Chem. Soc.* **1987**, *109*, 813−817. (b) Stille, J. K. *Angew. Chem., Intd. Ed. Engl.* **1986**, *25*, 508−524.

(5) (a) Maddaford, S. P.; Keay, B. A. *J. Org. Chem.* **1994**, *59*, 6501−6503. (b) Shieh, W.-C, Carlson, J. A. *J. Org. Chem.* **1992**, *57*, 379−381. (c) Miyaura, N.; Ishiyama, T.; Sasaki, H.; Ishikawa, M.; Satoh, M.; Suzuki, A. *J. Am. Chem. Soc.* **1989**, *111*, 314−312.

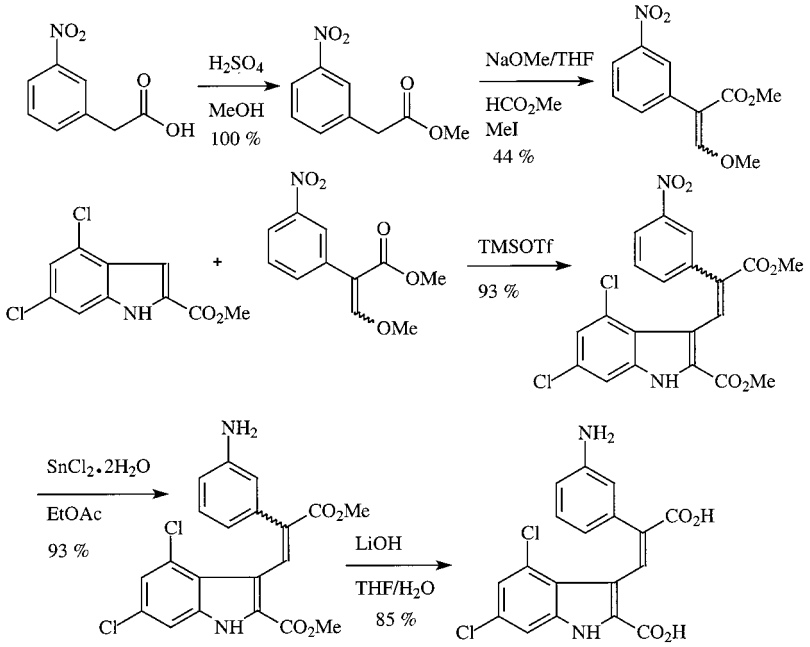
(6) McKenna, Charles E.; Khawli, Leslie A. *J. Org. Chem.* **1986**, *51*, 5467−5471.

(7) (a) Hoshino, Y.; Miyaura, N.; Suzuki, A. *Bull. Chem. Soc. Jpn.* **1988**, *61*, 3008−3010. (b) Farina, V.; Krishnan, B. *J. Am. Chem. Soc.* **1991**, *113*, 9585−9595. (c) Thompson, W. J.; Jones, J. H.; Lyle, P. A.; Thies, J. E. *J. Org. Chem.* **1988**, *53*, 2052−2055. (d) Wallow, T. I.; Novak, B. M. *J. Org. Chem.* **1994**, *59*, 5034−5037.

Scheme 2



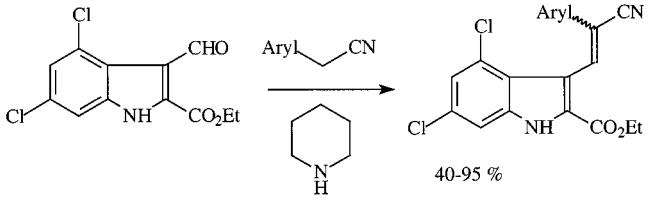
Scheme 3



3-thienyl-substituted enol ethers did not appear to give any coupled products. Some of the analogues prepared were the *p*-F, *p*-Cl, *p*-Br, *p*-I, p-Me, *p*-CF_3, *m*-NO_2, and *o*-Cl derivatives.

Since the enol ether method was not applicable to the synthesis of analogues with electron-donating phenyl groups, the Knoevenagel condensation[8] was investigated (fourth approach shown in Scheme 4). Under classical Knoevenagel condensation conditions arylacetic esters are condensed with aldehydes and ketones in the presence of a catalytic amount of base to give the α,β-unsaturated esters. Any attempts with multiple analogues of arylacetic esters were not successful;

Scheme 4

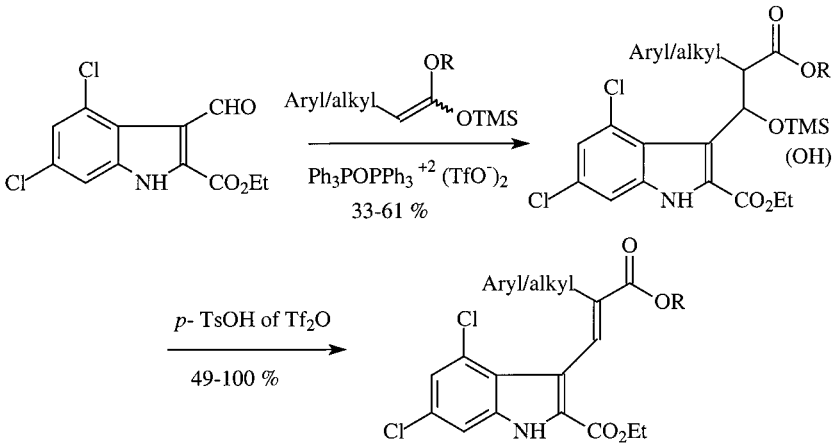


however, the condensation of the appropriate arylacetonitrile with the aldehyde gave the desired results. Depending on the aryl substituents, the condensation was rather sluggish and proceeded in low yield with starting material being recovered.

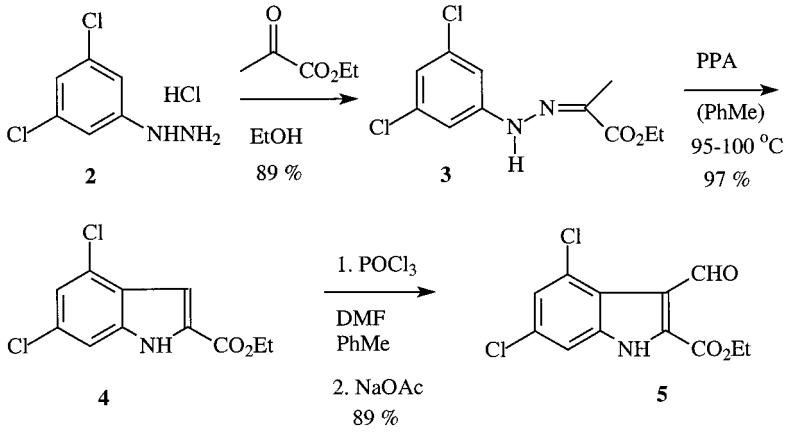
The drawback of this method, with exception of the pyridyl analogues, was that the hydrolysis of the α,β-unsaturated nitriles proceeded only to the amide stage. Conditions for further or direct hydrolysis to the diacids had not been fully investigated during the course of this work. The commercially available *m*-nitrophenylacetonitrile made

(8) (a) Contracted to Wynberg H. Syncom, Holland. (b) Crowell, T. I.; Kim, T.-R. *J. Am. Chem. Soc.* **1973**, *95:20*, 6781−6786. (c) Prout, F. S.; Beaucaire, V. D.; Drykacz, G. R.; Koppes, W. M.; Kuznicki, R. E.; Marlewski, T. A.; Pienkowski, J. J.; Puda, J. M. *J. Org. Chem.* **1973**, *38*, 1512−1517. (d) Crowell, T. I., Peck, D. W. *J. Am. Chem. Soc.* **1953**, *75*, 1075. (e) Cordes, E. H., Jencks, W. P. *J. Am. Chem. Soc.* **1962**, *84*, 826. (f) For more Knoevenagel references, see: House, H. O. *Modern Synthetic Reactions*, 2nd ed.; The Benjamin/Cummings Publishing Company: New York, 1972.

Scheme 5



Scheme 6



this attractive approach to **1** a high priority. Some of the other analogues prepared were the *p*-NO_2, 2-, 3-, and 4-pyridyl derivatives.

To circumvent the problems encountered with the hydrolysis of the nitriles, a fifth approach utilizing the Mukaiyama reaction[9] was investigated (shown in Scheme 5). It was felt that the use of the Mukaiyama reaction, which involves transfer of a silyl group to the oxygen of the aldol, could circumvent the reversibility of the Knoevenagel condensation with the esters and drive the reaction to completion. In addition, the Mukaiyama reaction would also provide a means of preparing analogues from both alkyl- and arylacetic esters containing relatively unactivated methylene groups.

In practice,[10] the reaction gave complex mixtures of *erythro/threo* aldols and their silyl-protected counterparts which were difficult to separate via chromatography. Second, it was difficult to control the retro aldol reaction from taking place during the elimination process, thus giving unreproducible results. This approach received low priority. Some of the analogues prepared were the phenyl, *m*-pyridyl, *m*-phenoxyphenyl, and *m*-methoxyphenyl derivatives.

Chemical Development's initial efforts were focused on the enol ether route and the Knoevenagel condensation. Although the enol ether route was not attractive from a scale-up position, it provided us with problems common to the two routes (nitro reduction and purification). The Knoevenagel route was the one that ultimately provided the solution to preparing larger quantities of MDL 103371. From this point on in the discussion the focus will be directed towards development of the Knoevenagel approach.

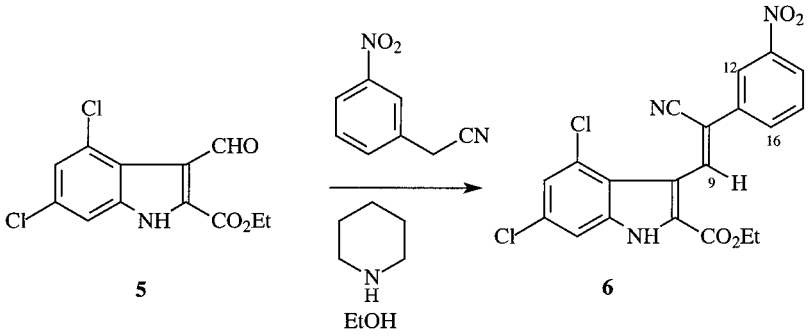
Discussion

Preparation of the indole aldehyde precursor to the Knoevenagel reaction utilized classical chemistry (Scheme 6). 3,5-Dichlorophenylhydrazine (**2**) was treated with ethyl pyruvate in ethanol to give (*E/Z*)-ethyl-2-[(3,5-dichlorophenyl)hydrazono]propanoate (**3**) in 89% yield. The reaction was originally performed with the addition of sulfuric acid as the cataylst; however, it was later found that the hydrochloric acid from the 3,5-dichlorophenylhydrazine hydrochloride was a sufficient acid source. Hydrazone **3** was cyclized under Fischer Indole cyclization[11] conditions with polyphosphoric acid (PPA) to give ethyl-4,6-dichloro-1*H*-indole-2-carboxylate (**4**) in 97% yield. The viscous nature of PPA at ambient temperature made it difficult to stir with a glass stirring rod in a 22 L flask, and therefore toluene was added to reduce

(9) Mukaiyama, T.; Matsui, S.; Kashiwagi, K. *Chem. Lett*. **1989**, 993−996.
(10) (a) Prepared by the method of Emde, H.; Simchen, G. *Synthesis* **1977**, 867−869. (b) Aaberg, A.; Gramstad, T.; Husebye, S. *Tetrahedron Lett.* **1979**, 2263−2264. (c) Hendrickson, J. B.; Schwartzman, S. M. *Tetrahedron Lett*. **1975**, 277−280

(11) Loudon, M. G. *Organic Chemistry*; Addison-Wesley Publishing Company: Reading, MA, 1983.

Scheme 7



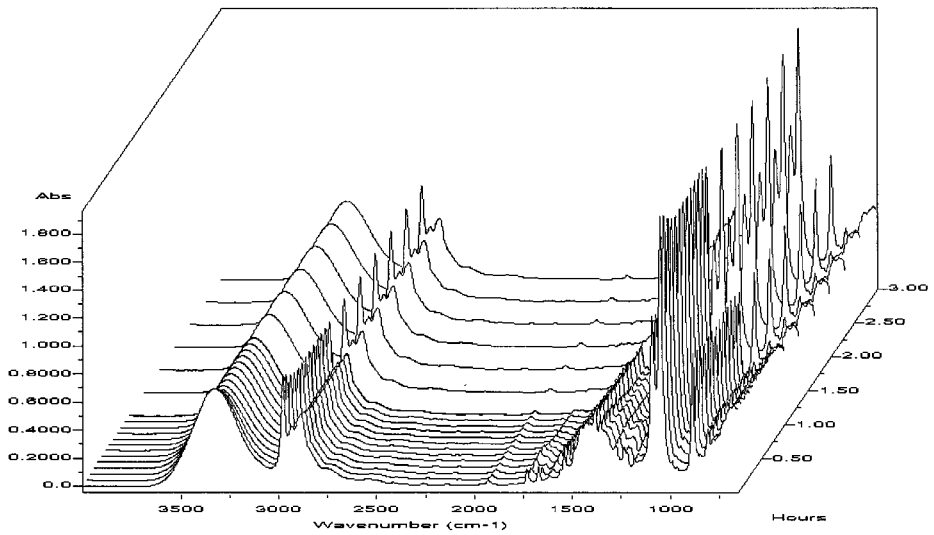


Figure 1. **ReactIR 3D plot of Knoevenagel reaction.**

the viscosity. The toluene did not effect the reaction and could be removed prior to the quench via distillation. In a reactor, however, stirring was found adequate without the need for toluene. The product of the Fischer indole cyclization **4** was subjected to a Vilsmeier−Haack[11] formylation. Originally dichloroethane (DCE) was used with the phosphorus oxychloride to form and isolate the Vilsmeier−Haack intermediate as the dichlorophosphoric acid salt. It was found toluene was an efficient and acceptable replacement for the environmentally undesirable DCE. The Vilsmeier−Haack intermediate was then treated with buffered water to give ethyl-4,6-dichloro-3-formyl-1*H*-indole-2-carboxylate (**5**) in 88% yield. It was later learned that the toluene could be removed via distillation, and the hydrolysis could be performed on the mixture without having to isolate the intermediate via filtration. The entire Fischer indole/Vilsmeier−Haack process was tried in toluene. However, there were impurities formed during the hydrazone formation that could not be removed in intermediates **3**, **4**, or **5**.

Ethyl-4,6-dichloro-3-formyl-1*H*-indole-2-carboxylate (**5**) was treated with 3-nitrophenylacetonitrile (Scheme 7) under Knoevenagel[8] conditions using catalytic piperidine in ethanol to give ethyl-(*Z*)-4,6-dichloro-3-[2-cyano-2-(3-nitrophenyl)-ethenyl]-1*H*-indole-2-carboxylate (**6**) exclusively as the *Z* isomer in 91% yield (large NOEs from H-9 to H-12 and H-16 are consistent with (*Z*)-olefin geometry). Product began to crystallize from the hot mixture approximately 15 min after reaching refluxing temperature. After the mixture cooled, the product **6** was isolated with greater than 99% purity. The insoluble nature of **6** most likely helps drive the reaction to completion and eliminates any "retro aldol" condensation from taking place. However, because of the insolubility of nitrile **6,** we initially could not find an efficient way to monitor the progress of the reaction.

The availability of new instrumentation allowed us to investigate the Knoevenagel reaction using in situ FTIR monitoring techniques (ReactIR)[12] The data generated is summarized in three figures , including reaction profile 3D plot (Figure 1), ConcIRT analysis of identified components (Figure 2), and reaction profile of identified components (Figure 3).

(12) In situ FTIR reaction monitoring was performed using a ReactIR MP Mobile Reaction Analysis System (ASI Applied Systems, Millersville, MD) equipped with a 9-bounce, SiComp probe. The probe was inserted into glass round-bottomed flasks via a 24/40 airtight adapter and submersed into the reaction mixtures. (a) First, the progress of the reaction was monitored by FTIR in real time by collecting data (4000−650 cm^{-1}) at predetermined intervals (typically every 0.5−2 min) for the duration of the reaction. The set of absorbance spectra was then plotted as a function of time to generate a ReactIR 3D Plot profile for each reaction sequence. (b) Second, each ReactIR 3D Plot that was generated was then analyzed using ConcIRT, a post-experiment algorithm which tracks changes in absorbance profiles that occur over time as a reaction proceeds. Even with complex reaction mixtures that have overlapping IR bands, ConcIRT has the capability of "extracting" absorption bands of the individual species in a reaction mixture, and thus can provide the individual IR spectrum for each "identified" component, along with the corresponding reaction concentration profiles.

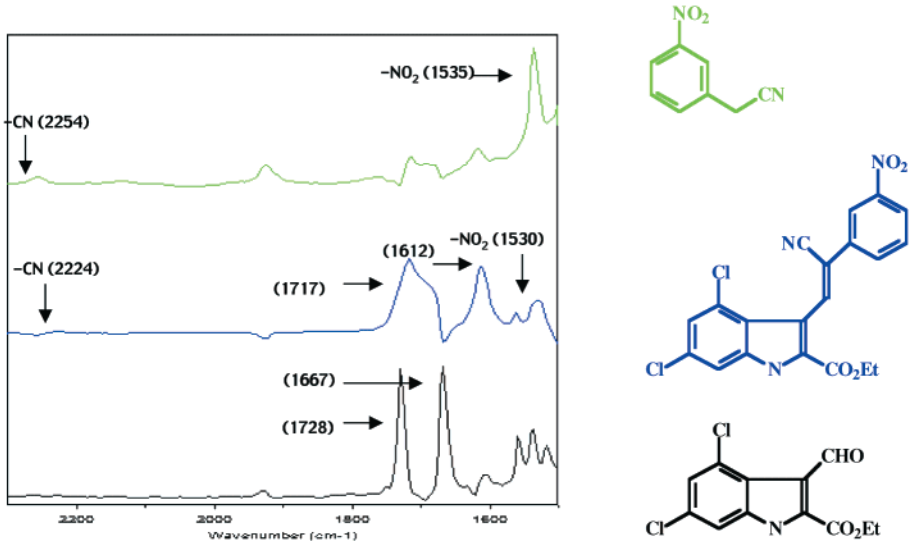


Figure 2. **ConcIRT analysis of the Knoevenagel reaction.**

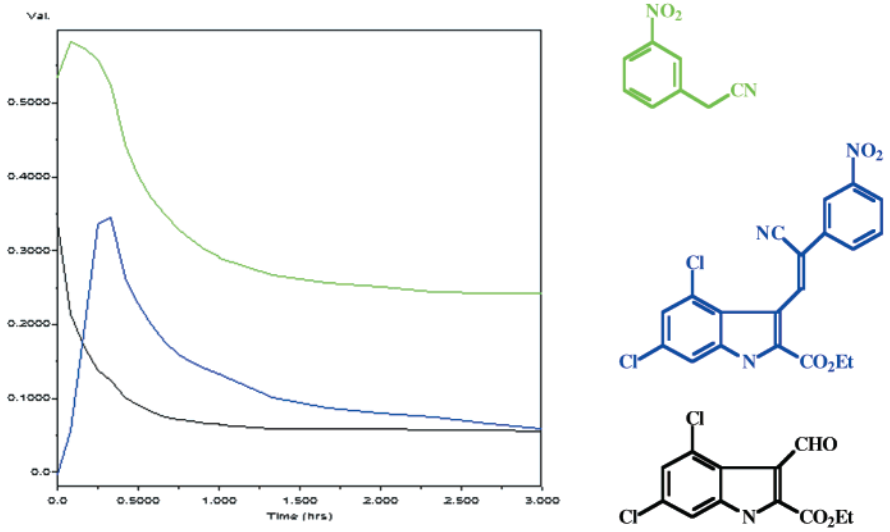


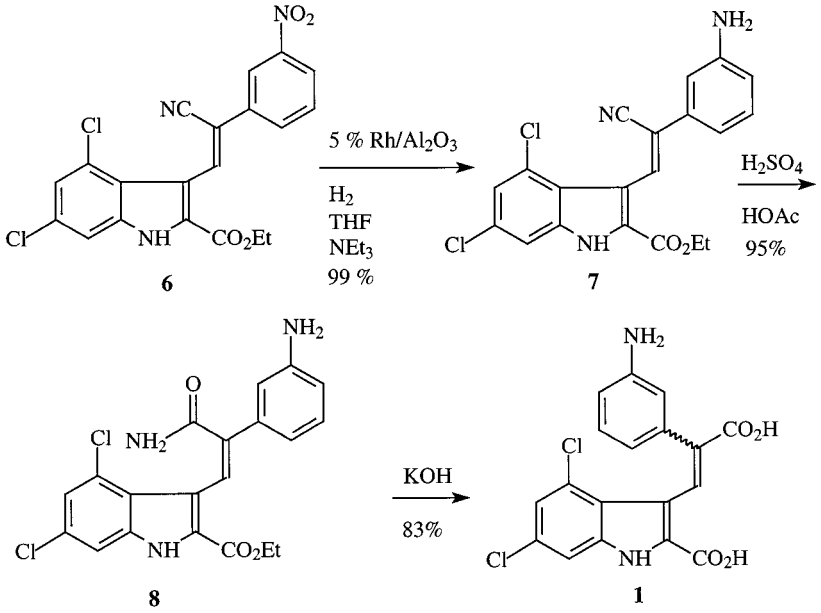
Figure 3. **ConcIRT component profiles of Knoevenagel reaction.**

The ConcIRT algorithm looked at the changing bands in the IR plot and identified three components in the reaction mixture (Figure 2). Upon closer analysis, those components were the two starting materials and product (as compared to blank runs of each component). Upon inspection of the components' concentration versus time (Figure 3) plot it became clear that the reaction was over in about 1.5 h. The first 15 min were used to heat the reaction to reflux. During that time, the 3-nitrophenylacetonitrile was not completely in solution as evident from Figure 3 (solution concentration increases during heat up, then decreases as it is consumed). The concentrations of ethyl-4,6-dichloro-3-formyl-1*H*-indole-2-carboxylate (**5**) and 3-nitrophenylacetonitrile began to decrease, and ethyl-(*Z*)-4,6-dichloro-3-[2-cyano-2-(3-nitrophenyl)ethenyl]-1*H*-indole-2-carboxylate (**6**) began to appear and increase in the first 30 min. However, due to the insoluble nature of **6**, the apparent concentration decreases as it drops out of solution due to the inability of the ReactIR probe to take an IR of a suspended solid. Nevertheless, one can easily monitor the disappearance of the two starting materials to determine the progress of the reaction. It may be interesting to note that it is not evident if the *Z* isomer is formed exclusively or if the *E* isomer is equilibrated to the thermodynamically favored *Z* isomer.

Although the reduction of ethyl-(*Z*)-4,6-dichloro-3-[2-(3-aminophenyl)2-cyanoethenyl]-1*H*-indole-2-carboxylate (**7**) with $SnCl_2$ went in high chemical purity, the yield of the reaction varied (67−97%) due to emulsion problems during the work-up. Attempted filtration of the emulsion through a pad of Celite prior to separation of the aqueous and organic layers was tedious due to poor flow rate and difficulty in extracting all of aniline **7** from the filter cake. Because of these problems, as well as the toxicity issues surrounding the use of tin reagents, alternative reaction conditions were sought for the reduction of nitro-nitrile **6**.

There is a veritable plethora of methods for reduction of aromatic nitro compounds to the corresponding anilines; however, nitro-nitrile **6** demands a very selective reducing agent because of the breadth of functionality present in the molecule (i.e., the nitrile, the aryl chlorides, and the ester moieties). A widely used reagent for the reduction of nitro

Scheme 8



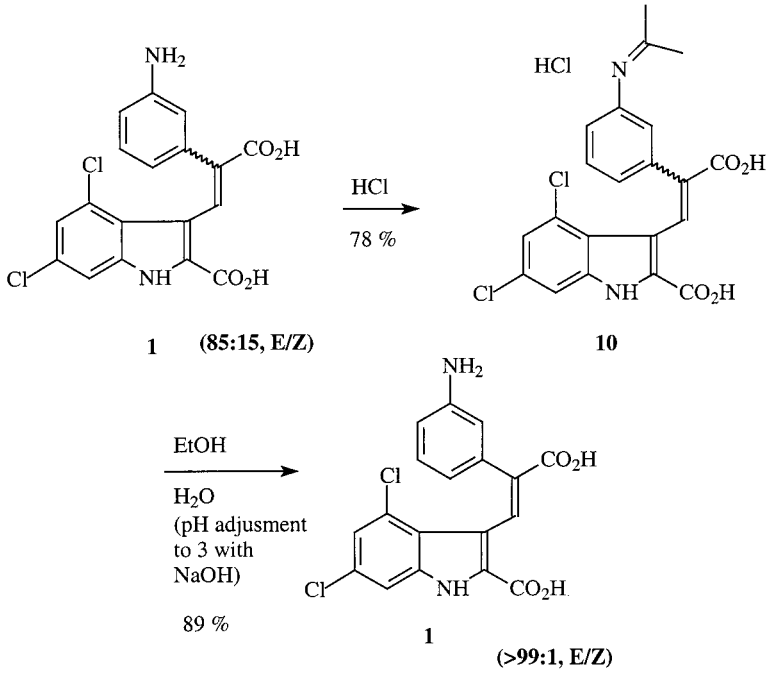
compounds to amines is iron.[13] Most of these reactions use an iron salt in conjunction with iron metal to accomplish the reduction. Thus, nitro-nitrile **6** was treated with Fe/$FeSO_4$[14] in various solvent systems (EtOH/H_2O, THF/H_2O, and EtOAc/H_2O) heated at reflux. All of the reactions gave similar results—little to no reaction with poor chemoselectivity for formation of aniline **7**. The use of Fe/NH_4Cl[15] in MeOH/H_2O heated at reflux also showed little reaction. An attempt was made to reduce nitro-nitrile **6** with $Cu(acac)_2$/$NaBH_4$ in EtOH/THF,[16] but the reaction did not go to completion and gave several products. A general problem with these attempted reductions was the poor solubility of nitro-nitrile **6** in most organic solvents. It was found that **6** was soluble in DMSO and THF (~1 g of nitro-nitrile **6** per 10−15 mL of THF at ambient temperature) and slightly soluble in EtOAc but insoluble in chlorinated solvents, MeCN, alcohols,[17] heptane, or toluene.

The next approach to reduce aromatic nitro-nitrile **6** was via catalytic hydrogenation (Scheme 8). The choice of catalyst was deemed to be crucial to the success or failure of this approach because of the various functionality present in nitro-nitrile **6**. The use of Pd/C with hydrazine had been reported to reduce aromatic nitro groups in the presence of aliphatic nitriles;[18] however, under these conditions, nitro-nitrile **6** afforded aniline **7** contaminated with two unidentified by-products. Use of Pd/Pb/C with hydrazine gave several products. Because it was thought palladium catalysts were possibly causing the indole ring of nitro-nitrile **6** to undergo hydro-dehalogenation, PtO_2 and Pt/C were examined. Reduction of nitro-nitrile **6** with PtO_2 under one atmosphere of hydrogen gave two products (TLC), one of which appeared to be desired product aniline **7**. A more interesting result was obtained using Pt/C with hydrazine to reduce nitro-nitrile **6**. A single product was obtained which was initially thought to be aniline **7**; however, careful comparison of this new product with aniline **7** by TLC and NMR revealed they were not the same. The new product was identified as the hydroxylamine analogue. It is interesting to note that this new product is not formed when hydrogen (1 atm) is substituted for hydrazine, or if hydrazine is present but the catalyst is omitted. The hydroxylamine was stable at ambient temperature under inert atmosphere as a solid, but when it was dissolved in either THF or EtOAc and allowed to stand at ambient temperature, it decomposed into primarily one new unidentified compound.

The best conditions found to date for the reduction of nitro-nitrile **6** to aniline **7** use Rh/Al_2O_3 and Et_3N under an atmosphere of hydrogen in THF at ambient temperature for ~20 h. Under these conditions a near quantitative yield of aniline **7** is obtained. It was not ascertained if Et_3N was absolutely crucial for the reaction.[19] The reaction was monitored by HPLC, and an intermediate product could be seen, which was converted to product if the reaction was allowed to run to completion. This intermediate product is thought to be hydroxylamine since it had the same HPLC retention time as a previously identified hydroxylamine sample. Varying the pressure of hydrogen from atmospheric to 40 psi resulted in complete reaction in <16 h with no loss of product purity. However, upon scaling the reaction

(13) Hodgson, H. H.; Whitehurst, J. S. *J. Chem. Soc.* **1945**, 202.
(14) Hodgson, H. H,; Hathway, D. E. *J. Chem. Soc.* **1944**, 538.
(15) Ramadas, K.; Srinivasan, N. *Synth. Commun.* **1992**, *22*, 3189; b) Tsuji, K.; Nakamura, K.; Konishi, N.; Okumura, H.; Matsuo, M. *Chem. Pharm. Bull.* **1992**, *40*, 2399.
(16) Kizil, M.; Lampard, C.; Murphy, J. A. *Tetrahedron Lett.* **1996**, *37*, 2511. For other $NaBH_4$−metal salts systems see: He, Y.; Zhao, H.; Pan, X.; Wang, S. *Synth. Commun.* **1989**, *19*, 3047 and references therein.
(17) The solubility of **6** in methanol was not checked due to concerns of transesterification.
(18) Adger, B. M.; Young, R. G. *Tetrahedron Lett.* **1984**, *25*, 5219. The use of Raney nickel with hydrazine affords concomitant reduction of the nitrile to the corresponding amine but leaves aromatic bromides and chlorides unchanged (also see: Leggetter, B. E.; Brown, R. K. *Can. J. Chem.* **1960**, *38*, 2363).
(19) Amine was added to the reaction to retard possible hydrodechlorination of the indole. An initial experiment done without Et_3N had a significant by-product produced, which is now thought to be hydroxylamine based upon HPLC retention times, so it may be possible to perform the reaction without Et_3N.

Scheme 9



1 **(85:15, E/Z)** **10**

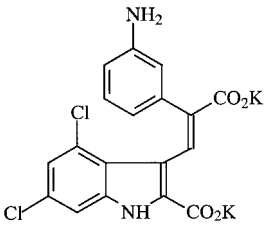
1 **(>99:1, E/Z)**

with an increase in pressure and temperature, one began to see a *des*-chloro analogue.[20] This impurity is difficult to remove in any of the remaining steps, thus contaminating the final product.

The hydrolysis to the acid was accomplished via the amide in a two-step sequence. First, treatment of ethyl-(*Z*)-4,6-dichloro-3-[2-(3-aminophenyl)2-cyanoethenyl]-1*H*-indole-2-carboxylate (**7**) with a 1:1 mixture of sulfuric and acetic acid gave the amide (one time purification and isolation yield of 95%). Many acid−acid, acid−water combinations were screened only to produce decomposition and multiple product mixtures. If the sulfuric acid−acetic acid hydrolysis temperature was kept at 50 °C, the *Z* geometry of the olefin was retained. However, if the temperature of the hydrolysis were increased to 90 °C, then mixtures of *E/Z* isomers were obtained in variable ratios depending on the time at the elevated temperatures. Nevertheless, the product amide-aniline **8** was subjected to potassium hydroxide to give the desired crude MDL 103371 in 83% yield. The ratio of the crude product **1** was approximately 80:20 *E/Z* and could not be equilibrated any further to the desired *E* isomer under many conditions. Second, subjecting different mixtures of the *E/Z* to the basic hydrolysis conditions consistently drove the mixture to approximately 80:20 ratio of *E/Z* isomers, the apparent thermodynamic equilibrium ratio under these given conditions. Much to our surprise, the basic hydrolysis produced small amounts of the *des*-chloro impurity.[20] This was confirmed by subjecting a clean sample of **1** to prolonged basic hydrolysis conditions and monitoring the formation of the *des*-chloro analogue as compared to an authentic sample. The mechanism of this unfortunate phenomenon was not investigated nor is it fully understood.

Purification

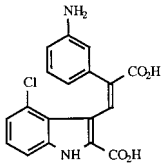
Two purification techniques were ultimately identified. The first was found by allowing the potassium hydroxide hydrolysis reaction mixture to slowly cool overnight. Crystals of the dipotassium salt **9** slowly precipitated from the reaction medium. This solid could be isolated by filtration and was determined to have roughly 93% of the *E* isomer and 4% of the *Z* isomer. The salt could be neutralized with hydrochloric acid to give back the diacid under mild conditions which did not to affect this ratio.



9

The second purification was the result of parallel crystallization studies. Crystallization of a 85:15 *E/Z* mixture from acetone gave crystals of the acid-catalyzed Schiff base **10** (Scheme 9) in greater than a 99.9:0.1% *E/Z* ratio. (This was a sample that had not been purified as the dipotassium salt, **9**; however, **9** had been over-acidified from the work-up of potassium hydrolysis, and thus had HCl present. Typical *E/Z* ratios before the dipotassium salt purification were on the order of 85:15 *E/Z*.) The Schiff base was easily converted

(20) The *des*-chloro isomer was isolated by preparative chromatography and characterized to be the following structure. The other possible des-chloro isomers were not detected with LC/MS techniques.



back to MDL 103371 under mild conditions which did not affect the desired ratio. The product was isolated in the absence of light in a Nutsche filter and stored appropriately without further isomerization. Although the *des*-chloro analogue could not be fully removed by either of the two purification methods, it could be minimized by repeated Schiff base crystallization.

Conclusions

The preparation of kilogram quantities of MDL 103371 utilized some classical chemistry. Fischer indole cyclization, followed by a Vilsmeier−Haack formylation and a Knoevenagel condensation gave immediate access into the proper carbon framework of the target molecule. A unique hydrogenation catalyst and solvent system followed by a two-step acid−base hydrolysis gave the crude product. Purification was accomplished by potassium salt crystallization followed by Schiff base formation to give MDL 103371 in nine steps (including the purification step) in an overall yield of 38%. All of the steps gave yields of greater than 80% (except the Schiff base, 78%) with easily isolated solids. (Two batches of 1.3 and 2.0 kg of MDL 103371 were prepared in Cincinnati.)

For the process to be complete, a better understanding of the hydrogenation and the basic hydrolysis would be necessary to control *des*-chloro formation. Because the project was terminated, these issues were not investigated and are still not fully understood.

Experimental Section

General. All ^{1}H- and ^{13}C NMR spectra were performed on a Varian XL-300 spectrophotometer at 300 and 75 MHz, respectively. All GC data were collected on a Hewlett-Packard 5890 GC with an HP 5972 mass selective detector. The GC column was an HP-5 (cross-linked 5% Ph Me silicone, 30 m × 0.25 mm × 0.25 μm film thickness); injector temperature − 250 °C; detector temperature − 280 °C; temperature program − 50 °C for 1 min then heated to 300 °C at the rate of 20 °C/min. HPLC analysis was done on a Thermo Separations P2000 HPLC with a Spectra 100 detector and a Spectra Physics SP4270 integrator The column used was a Waters Symmetry C18 column (3.9 × 150 mm); the wavelength of detection was at 230 nm; the mobile phase used for the chromatography to monitor the synthesis of (*E*)-4,6-dichloro-3-[2-(3-aminophenyl)-2-carboxyethenyl]-1*H*-indole-2-carboxylic acid and (*E*)-4,6-dichloro-3-[2-carboxy-2-[3-[(1-methylethylidene)amino]ethenyl]-1*H*-indole-2-carboxylic acid monohydrochloride was 70% 0.05 M NaH_2PO_4 buffer pH 3.0/30% CH_3CN at a flow rate of 1.0 mL/min. The mobile phase used to monitor the synthesis of ethyl (*E/Z*)-4,6-dichloro-3-[3-amino-2-aminophenyl)-3-oxo-1-propenyl]-1*H*-indole-2-carboxylate and ethyl (*Z*)-4,6-dichloro-3-[2-(3-aminophenyl)2-cyanoethenyl]-1*H*-indole-2-carboxylate was 40% 0.05% aqueous TFA/60% CH_3CN at a flow rate of 1.0 mL/min. Thin-layer chromatography was done using 5 × 10 cm Merck 60F-254 plates.

(*E/Z*)- Ethyl 2-[(3,5-Dichlorophenyl)hydrazono]propanoate, 3. A mixture of 2.0 kg (9.36 mol) of 3,5-dichlorophenylhydrazine hydrochloride (Acros) and 12 L of EtOH was charged to a 22-L three-necked flask fitted with a stirrer, thermometer, condenser, and a continuous N_2 purge. The mixture was stirred at 50 °C for 1 h (there were insolubles present). A total of 1.08 kg (9.37 mol) of ethyl pyruvate was added in one portion. The temperature rose from 45 to 54 °C over ∼5 min. The reaction mixture was allowed to stir at ambient temperature. The reaction was monitored by GC (t_R of **2** = 9.2 min; t_R of **3** = 11.3 min) and reached completion after 4 h. The insolubles were filtered off and washed with 1 L of EtOH. The solvent was removed from the filtrate at 40 °C/50 Torr to give a residue. The solid obtained was air-dried to a constant weight to give 2.282 kg, 89% yield, of (*E/Z*)- ethyl 2-[(3,5-dichlorophenyl)-hydrazono]propanoate, **3**. ^{1}H NMR (DMSO-d_6) δ 10.1 (bs, 1 H), 7.21 (s, 2 H), 6.98 (s, 1 H), 4.21 (q, 2 H, J = 7.2 Hz), 2.05 (s, 3 H), 1.26 (t, 3 H, J = 7.1 Hz). ^{13}NMR (DMSO-d_6) 164.7, 147.0, 135.1, 134.7, 119.6, 112.1, 60.6, 14.1, 12.1. Anal. Calcd for $C_{11}H_{12}Cl_2N_2O_2$: C, 48.02; H, 4.40; N, 10.18. Found: C, 47.43; H, 4.25; N, 9.95. MS *m/z*: (M^+) calcd 274.0, obsd (M − H) 273.8. IR (2% in KBr) υ_{max}/cm^{-1} = 1698, 1583.

Ethyl 4,6-Dichloro-1*H*-indole-2-carboxylate, 4. A mixture of 53 kg of polyphosphoric acid, 14 L of toluene, and 2.83 kg (10.3 mol) of ethyl 2-[(3,5-dichlorophenyl)hydrazono]propanoate, **3** was charged to a 20 gallon glass-lined reactor under a continuous N_2 purge. The mixture was heated to 95−100 °C (*NOTE*: At ∼70 °C HCl gas was given off). The progress of the reaction was monitored by GC (t_R of **4** = 11.8 min, t_R of **3** = 11.3 min) and found to be complete after 2 h. The reaction mixture was cooled to 80 °C, and toluene was removed at 100 Torr. The reaction mixture was discharged from the reactor and quenched into a 30 gallon Hastelloy reactor which contained 50 kg of ice and 10 L of ice water. The resulting slurry was stirred at 35−50 °C for 30 min. The product was filtered off, washed acid free with 2 × 20 L H_2O, then air-dried to give 2.57 kg, 97% yield, of ethyl 4,6-dichloro-1*H*-indole-2-carboxylate, **4**. ^{1}H NMR (DMSO-d_6) δ 12.46 (bs, 1 H), 7.45 (s, 1 H), 7.28 (s, 1 H), 7.11 (s, 1 H), 4.41 (q, 2 H, J = 7.1 Hz), 1.34 (t, 3 H, J = 7.1 Hz). ^{13}NMR (DMSO-d_6) 160.8, 137.8, 129.4, 129.3, 127.1, 124.5, 120.2, 111.5, 105.5, 61.0, 14.2. Anal. Calcd for $C_{11}H_9Cl_2NO_2$: C, 51.19; H, 3.51; N, 5.43. Found: C, 51.34; H, 3.38; N, 5.34. MS *m/z*: (M^+) calcd 257.0, obsd (M − H) 256.5 IR (2% in KBr) υ_{max}/cm^{-1} = 1698.

Ethyl 4,6-Dichloro-3-formyl-1*H*-indole-2-carboxylate, 5. A mixture of 2.28 kg (8.85 mol) of ethyl 4,6-dichloro-1*H*-indole-2-carboxylate, **4**, 5.8 L of 1,2-dichloroethane, and 972 g (13.3 mol) of DMF was charged to a 22-L three-necked flask fitted with a stirrer, thermometer, condenser, and continuous N_2 purge. [NOTE: Toluene was used to replace dichloroethane (DCE) in future experiments (same volume). Because the full analytical data was collected on the DCE prepared sample, the experimental will reflect that exact experiment. The toluene process was developed by our Dow Chemical colleagues.] The mixture was stirred to a smooth slurry at 24 °C and 2.04 kg (13.3 mol) of $POCl_3$ was added over 2 min. The temperature rose from 24 to 58 °C. The

solution was heated to and held at reflux (88 °C) for 18 h [90 °C for toluene]. The progress of the reaction was monitored by GC (t_R of **5** = 13.3 min, t_R of **4** = 11.8 min) and typically ~8% of MDL 102317 (**4**) remained. The slurry was cooled to and held at 25 °C for 30 min. The precipitate was filtered off and washed with 2 × 2 L of 1,2-dichloroethane and 2 L of Et_2O [Et_2O was replaced with ethanol in the toluene process]. The filter cake was slurried in 39 L of 1 M NaOAc for 3 h. The product was filtered off and washed with 2 × 20 L of H_2O, 5 L of Et_2O, then air-dried for 3 d to give 2.21 kg, 88% yield, of ethyl 4,6-dichloro-3-formyl-1*H*-indole-2-carboxylate, **5**. ^{1}H NMR (DMSO-d_6) δ 13.00 (bs, 1 H), 10.60 (s, 1 H), 7.53 (s, 1 H), 7.37 (s, 1 H), 4.45 (q, 2 H, J = 7.1 Hz), 1.39 (t, 3 H, J = 7.1 Hz). (DMSO-d_6) 160.5, 148.7, 139.2, 137.5, 134.6, 131.9, 131.3, 130.0, 127.2, 124.2, 122.3, 122.2, 120.3, 116.9, 115.8, 114.4, 112.0, 61.5, 14.0.Anal. Calcd for $C_{12}H_9Cl_2NO_3$: C, 50.38; H, 3.17; N, 4.90. Found: C, 50.23; H, 3.06; N, 4.85. MS *m*/*z*: (M^+) calcd 285, obsd (M + H) 286.4. IR (2% in KBr) υ_{max}/cm^{-1} = 1730, 1667.

Ethyl (*Z*)-4,6-Dichloro-3-[2-cyano-2-(3-nitrophenyl)-ethenyl]-1*H*-indole-2-carboxylate, 6. A mixture of 59 L of EtOH, 2.21 kg (7.76 mol) of ethyl 4,6-dichloro-3-formyl-1*H*-indole-2-carboxylate, 1.26 kg (7.76 mol) of 3-nitrophenylacetonitrile, and 205 g (2.4 mol) of piperidine was charged to a 30 gallon Hastelloy reactor under a continuous N_2 purge. The resulting mixture was heated to and held at reflux (79 °C) for 70 h. The reaction mixture was cooled to 25 °C. The precipitate was filtered off and washed with 2 × 5 L of EtOH. The product was air-dried to give 3.03 kg, 91% yield, of ethyl (*Z*)-4,6-dichloro-3-[2-cyano-2-(3-nitrophenyl)ethenyl]-1*H*-indole-2-carboxylate, **6**. ^{1}H NMR (DMSO-d_6) δ 12.95 (bs, 1 H), 8.63 (s, 1 H), 8.49 (dd, 1 H, J = 1.9, 1.9), 8.32 (dd, 1 H, J = 8.1, 1.9), 8.23 (dd, 1 H, J = 8.0, 1.9), 7.85 (dd, 1 H, J = 8.1, 8.0), 7.54 (d, 1 H, J = 1.8), 7.36, (d, 1 H, J = 1.8), 4.35 (q, 2 H, J = 7.1 Hz), 1.26 (t, 3 H, J = 7.1 Hz). ^{13}NMR (DMSO-d_6) 160.4, 148.4, 138.9, 137.2, 134.5, 131.7, 131.2, 129.7. 127.9, 126.4, 123.7, 122.0 (2), 120.1, 115.9, 155.7, 114.2, 111.9, 61.42, 13.9. Large NOEs from the H-9 to both H-12 and H-16 are consistent with the *Z* geometry. Anal. Calcd for $C_{20}H_{13}Cl_2N_3O_4$: C, 55.83; H, 3.05; N, 9.77. Found: C, 55.69; H, 3.07; N, 9.66. MS *m*/*z*: (M^+) calcd 429, obsd (M − H) 428.6. IR (2% in KBr) υ_{max}/cm^{-1} = 1708.

Ethyl (*Z*)-4,6-Dichloro-3-[2-(3-aminophenyl)2-cyanoethenyl]-1*H*-indole-2-carboxylate, 7. A mixture of 871 g (2.03 mol) of ethyl (*Z*)-4,6-dichloro-3-[2-cyano-2-(3-nitrophenyl)-ethenyl]-1*H*-indole-2-carboxylate, 13.5 L of THF, 175 g of 5% Rh/Al_2O_3, and 244 mL of triethylamine was charged to a 5 gallon autoclave under a N_2 blanket. The H_2 pressure was increased to about 50 psi and maintained at this pressure throughout the reaction. The reaction was slightly exothermic and the temperature was maintained at 23−36 °C by occasional cooling. The progress of the reaction was monitored by HPLC (t_R of **7** = 2.9 min, t_R of **6** = 14.8 min, t_R of hydroxylamine intermediate = 4.4 min) and found to be complete after 6 h (*NOTE*: H_2 uptake ceased after ~5 h). The catalyst was filtered off on a bed of dicalite and washed with 2 × 1 L of THF. The filtrate was concentrated at 35 °C/50 Torr and the solid obtained was dried at 30 °C/50 Torr for 18 h to give 813.0 g, 99% yield, of ethyl (*Z*)-4,6-dichloro-3-[2-(3-aminophenyl)2-cyanoethenyl]-1*H*-indole-2-carboxylate, **7**. ^{1}H NMR (DMSO-d_6) δ 12.95 (bs, 1 H), 8.09 (s, 1 H), 7.51 (d, 1 H, J = 1.7), 7.31 (d, 1 H, J = 1.7), 7.15 (dd, 1 H, J = 8.0,8.1), 6.93 (dd, 1 H, J = 1.7,1.7), 6.87(dd, 1 H, J = 8.2, 1.7), 6.65, (dd, 1 H, J = 8.2, 1.7), 5.39 (s, 2 H), 4.32 (q, 2 H, J = 7.1 Hz), 1.26 (t, 3 H, J = 7.1 Hz). ^{13}NMR (DMSO-d_6)160.7, 149.7, 137.4, 134.5, 133.8, 129.9, 129.8, 127.3, 126.9, 122.3, 122.0, 118.6, 117.7, 115.1, 115.0, 113.0, 111.8, 110.8, 61.3, 14.0. Anal. Calcd for $C_{20}H_{15}Cl_2N_3O_2$: C, 60.02; H, 3.78; N, 10.50. Found: C, 60.10; H, 3.81; N, 10.29. MS *m*/*z*: (M^+) calcd 399.0, obsd (M + H) 400.4. IR (2% in KBr) υ_{max}/cm^{-1} = 1708.

Ethyl (*E*/*Z*)-4,6-Dichloro-3-[3-amino-2-aminophenyl)-3-oxo-1-propenyl]-1*H*-indole-2-carboxylate, 8: A mixture of 1.52 kg (3.79 mol) of ethyl (*Z*)-4,6-dichloro-3-[2-(3-aminophenyl)2-cyanoethenyl]-1*H*-indole-2-carboxylate, **7**, and 4.85 L of acetic acid was charged to a 22-L three-necked flask fitted with a stirrer, thermometer, dropping funnel, and continuous N_2 purge. The mixture was stirred to a smooth slurry and a total of 4.85 L of H_2SO_4 was added over 3−5 min. The temperature rose from 24 to 90 °C. The reaction temperature was held at 90 °C. The progress of the reaction was monitored by HPLC (t_R of ethyl (*Z*)-4,6-dichloro-3-[2-(3-aminophenyl)2-cyanoethenyl]-1*H*-indole-2-carboxylate, **7**—5.8 min, t_R of ethyl (*E*/*Z*)-4,6-dichloro-3-[3-amino-2-aminophenyl)-3-oxo-1-propenyl]-1*H*-indole-2-carboxylate, **8**—1.2 min) and found to be complete after 1 h. The reaction mixture was cooled to 50 °C and quenched by pouring the reaction mixture into 22 kg of ice. The pH was adjusted to 4.9 by addition of 33 L of 6 M NaOH while maintaining a reaction temperature of 40−50 °C. The precipitate which formed was filtered off and washed with 3 × 10 L of H_2O. The product was air-dried to remove a majority of the H_2O, but the MDL 46757 obtained was used as a water wet pulp in the subsequent step. The yield for the reaction was 1.68 kg, 106% yield, of crude ethyl (*E*/*Z*)-4,6-dichloro-3-[3-amino-2-aminophenyl)-3-oxo-1-propenyl]-1*H*-indole-2-carboxylate, **8**. This was used without further purification.

(*E*)-4,6-Dichloro-3-[2-(3-aminophenyl)-2-carboxyethenyl]-1*H*-indole-2-carboxylic acid, 1. A mixture of 44.5 L of 6 M KOH and 1.34 kg (3.2 mol) of ethyl (*E*/*Z*)-4,6-dichloro-3-[3-amino-2-aminophenyl)-3-oxo-1-propenyl]-1*H*-indole-2-carboxylate (*NOTE*: 1.55 kg of water wet pulp was used) was charged to a 20 gallon glass-lined reactor. The mixture was heated to and held at reflux (108 °C) for 3.5 h (~1h after complete solution). The progress of the reaction was *carefully* monitored by HPLC (t_R of (*E*)-4,6-dichloro-3-[2-(3-aminophenyl)-2-carboxyethenyl]-1*H*-indole-2-carboxylic acid **1** = 7.5 min, t_R of *Z* isomer of MDL 103371, **1** = 9.9 min, t_R of **8** = 13.6 min, t_R of *Z* isomer of *des*-chloro impurity = 3.0 min) and found to be 76% (*E*)-4,6-dichloro-3-[2-(3-aminophenyl)-2-carboxyethenyl]-1*H*-indole-2-carboxylic acid, and 20% *Z* isomer. The reaction mixture was cooled to 25 °C over 2 h and held at this temperature for 20 min. The precipitate was filtered off and not washed. The

di-potassium salt, **9** was analyzed by HPLC and found to be 93% (*E*)-4,6-dichloro-3-[2-(3-aminophenyl)-2-carboxyethenyl]-1*H*-indole-2-carboxylic acid, 4% *Z* isomer, and 0.9% *des*-chloro. The dipotassium salt, **9** of (*E*)-4,6-dichloro-3-[2-(3-aminophenyl)-2-carboxyethenyl]-1*H*-indole-2-carboxylic acid was dissolved in 35 L of H_2O, and the pH was 12.8. The pH of the solution was adjusted to 3.2 by addition of 3.1 L of 3 M HCl while maintaining a reaction temperature of about 25 °C. The resulting slurry was stirred for 10 min. The product was filtered off and washed with 2 × 10 L of H_2O. The material was dried at 50 °C/50 Torr to give 1.05 kg, 83% yield, of crude (*E*)-4,6-dichloro-3-[2-(3-aminophenyl)-2-carboxyethenyl]-1*H*-indole-2-carboxylic acid, **1**.

Purification via (*E*)-4,6-Dichloro-3-[2-carboxy-2-[3-[(1-methylethylidene)amino]ethenyl]-1*H*-indole-2-carboxylic Acid Monohydrochloride (10). A mixture of 1017 g (2.6 mol) of crude (*E*)-4,6-dichloro-3-[2-(3-aminophenyl)-2-carboxyethenyl]-1*H*-indole-2-carboxylic acid and 14 L of acetone was charged to a 22-L three-necked flask fitted with a stirrer, thermometer, condenser, and continuous N_2 purge. The mixture was heated to and held at reflux (56 °C) for 30 min. The resulting turbid solution was clarified through a dicalite bed directly into a 22-L three-necked flask, and the filter cake was washed with 2 L of acetone. The filtrate was heated to and held at 50 °C. A total of 200 mL of concentrated HCl was added over 5 min. The reaction mixture was allowed to cool to rt over 2 h. The precipitate which formed was filtered off and washed with 2 × 1 L of acetone. The solid was dried at 25 °C/50 Torr to give 989.2 g, 78% yield of (*E*)-4,6-dichloro-3-[2-carboxy-2-[3-[(1-methylethylidene)amino]ethenyl]-1*H*-indole-2-carboxylic acid monohydrochloride, **10**. The product contained <0.1% of *Z* isomer by HPLC analysis.

A mixture of 2.66 kg (5.68 mol) of (*E*)-4,6-dichloro-3-[2-carboxy-2-[3-[(1-methylethylidene)amino]ethenyl]-1*H*-indole-2-carboxylic acid monohydrochloride, **10** and 26.6 L of H_2O was charged to a 35-L glass flask fitted with a stirrer. The mixture was stirred for 20 min. The pH was adjusted from 1.3 to 6.8 over 30 min using 3.1 L of 3 M NaOH. The resulting solution was clarified through sintered glass using 1 L of H_2O to wash. The pH of the filtrate was adjusted to 3.1 using 3.0 L of 3 M HCl. The resulting slurry was stirred for 15 min. The product was filtered off, washed with 4 × 5 L of H_2O, then dried for 24 h at 60 °C/50 Torr to give 1.97 kg, 89% yield, of (*E*)-4,6-dichloro-3-[2-(3-aminophenyl)-2-carboxyethenyl]-1*H*-indole-2-carboxylic acid, **1**. ^{1}H NMR (DMSO-d_6) δ 12.10 (s, 1 H), 7.98 (s, 1 H), 7.35 (s, 1 H), 7.18 (s, 1H,), 6.62 (dd, 1 H, J = 8.0,8.0 Hz), 6.25 (m, 2 H), 6.08 (dd, 1H, J = 8.0 Hz). ^{13}NMR (DMSO-d_6) 168.5, 162.1, 147.5, 137.8, 137.1, 136.6, 133.0, 128.9, 127.6, 127.5, 126.9, 122.8, 120.9, 117.7, 116.1, 115.8, 113.0, 111.3. Anal. Calcd for $C_{18}H_{12}Cl_2N_2O_4$ 0.13 H_2O: C, 54.93; H, 3.14; N, 7.12. Found: C, 54.88; H, 3.51; N, 0.6.95. Karl Fischer Analysis: 0.6% H_2O. MS *m*/*z*: (M^+) calcd 390.0, obsd (M + H) 391.1.

Purification of a Sample Containing *des*-Chloro Impurity. A mixture of 517 g (*E*)-4,6-dichloro-3-[2-(3-aminophenyl)-2-carboxyethenyl]-1*H*-indole-2-carboxylic acid (5.8% *des*-chloro impurity, 5.4% *Z* isomer, ~10% H_2O) and 12.5 L of methanol was charged to a 22-L three-necked flask fitted with a stirrer, thermometer, condenser, and a continuous N_2 purge. The mixture was heated to and held at reflux for 30 min. The resulting turbid solution was clarified through dicalite washing with 1 L of methanol. The filtrate was concentrated at atmospheric pressure to a volume of ~5 L. The resulting slurry was cooled to 25 °C and filtered. The filter cake was washed with 2 × 300 mL of methanol. A recovery of 307.1 g was obtained at this point with an HPLC analysis of 5.8% *des*-chloro impurity with essentially no *Z* isomer. The 307.1 g of (*E*)-4,6-dichloro-3-[2-(3-aminophenyl)-2-carboxyethenyl]-1*H*-indole-2-carboxylic acid and 3.5 L of methanol was charged to a 12-L three-necked flask fitted with a stirrer, thermometer, condenser, and a continuous N_2 purge. The mixture was heated to and held at reflux for 18 h. The slurry was cooled to 25 °C and filtered, washing with 2 × 300 mL of methanol. The (*E*)-4,6-dichloro-3-[2-(3-aminophenyl)-2-carboxyethenyl]-1*H*-indole-2-carboxylic acid was dried at 45 °C/50 Torr. A yield of 241.3 g, 47% recovery, was obtained with an HPLC analysis of 0.9% *des*-chloro impurity with essentially no *Z* isomer.

Received for review May 19, 2000.

OP000286S