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A New Approach to the Rapid Parallel Development of Four Neurokinin Antagonists. Part 5. Preparation of ZM374979 Cyanoacid and Selective Crystallisation of ZM374979 Atropisomers

Jeremy S. Parker,* Neil A. Smith, Matthew J. Welham, and William O. Moss

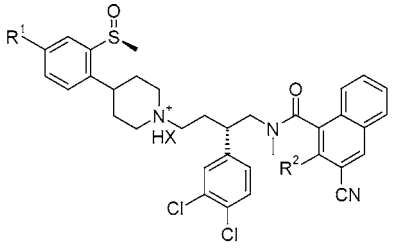
AstraZeneca, Process Research and Development, Avlon Works, Severn Road, Hallen, Bristol BS10 7ZE, UK

Abstract:

ZM374979 cyanoacid was prepared from ZD4974 cyanoester by a selective Grignard reaction followed by selective ester hydrolysis. On conversion of ZM374979 cyanoacid to ZM374979 free base, atropisomerism was observed, necessitating the development of a process for the selective crystallisation of a single atropisomer.

Introduction

Previous contributions in this series have described Zeneca Pharmaceuticals' rapid parallel development approach for the delivery of 1 kg of a series of neurokinin antagonists; ZD6021 **1**, ZD2249 **2**, ZD4974 **3**, and ZM374979 **4**.[1−5]



1 ($R^1 = R^2 = H$, X = Hydrogen Fumarate)
2 ($R^1 = OMe$, $R^2 = H$, X = Hydrogen Fumarate)
3 ($R^1 = H$, $R^2 = OMe$, X = Citrate)
4 ($R^1 = H$, $R^2 = Et$, X = Maleate)

ZM374979 cyanoacid **5** was required for delivery of ZM374979 **4**, and the preparation of compound was undertaken using the rapid development approach.

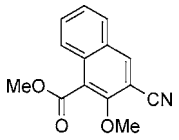
On preparation of ZM374979 **4** crude, it was discovered that this compound exhibits atropisomerism. This property had been observed in previous compounds in this series, but was not previously considered to be a significant issue due to the short half-lives of the atropisomers. For ZM374979 **4**, the half-life was sufficiently long to require the isolation of a single atropisomer for toxicity and clinical studies.

This contribution describes the preparation of ZM374979 cyanoacid **5**, and the challenge of developing a large-scale method for the selective crystallisation of a single atropisomer.

Results and Discussion

Preparation of ZM374979 Cyanoacid 5. ZM374979 **4** was the final compound in the series of neurokinin antagonists prepared by Zeneca Pharmaceuticals. The assembly of this compound was undertaken from three advanced intermediates; ZM374979 cyanoacid **5**, ZD7944 Pip sulfoxide **6**, and ZD6021 *N*-methylamine fumarate **7** (Figure 1).[3]

ZD7944 Pip sulfoxide **6** and ZD6021 *N*-methylamine fumarate **7** had been prepared previously, and large amounts of these materials were available. Thus, the preparation of ZM374979 **4** only required the synthesis of ZM374979 cyanoacid **5**. It was decided synthesise this compound from ZD4974 cyanoester **8**, which was available in multikilogram quantities from the preparation of ZD4974 **3**.



8

The conversion of ZD4974 cyanoester **8** to ZM374979 cyanoacid **5** had previously been demonstrated by Discovery (Scheme 1). ZD4974 cyanoester **8** was demethylated using magnesium iodide[6] to afford ZM374979 naphthol **9**. This was then reacted with triflic anhydride to generate ZM374979 triflate **10**, which was converted to ZM374979 cyanoester **11** using a palladium-catalysed cross-coupling reaction with triethylborane.[7] ZM374979 cyanoester **11** was then converted to ZM374979 cyanoacid **5** using trimethylsilyl iodide.

As has been previously described for the rapid parallel development approach, scale-up of the Discovery synthesis is the preferred method of delivery, assuming that both safety and robustness could be demonstrated.[1]

An initial evaluation of the Discovery procedures raised significant concerns with the chemistry used. The demethylation reaction used to prepare ZM374979 Naphthol **9** was shown to be extremely solvent sensitive, working only in benzene/diethyl ether. Simple changes such as converting

(1) Moseley, J. D.; Moss, W. O. *Org. Process Res. Dev.* **2003**, *7*, 53.
(2) Moseley, J. D.; Moss, W. O.; Welham, M. J.; Ancell, C. L.; Banister, J.; Bowden, S. A.; Norton, G.; Young, M. J. *Org. Process Res. Dev.* **2003**, *7*, 58.
(3) Parker, J. S.; Bowden, S. A.; Firkin, C. R.; Moseley, J. D.; Murray, P. M.; Welham, M. J.; Wisedale, R.; Young, M. J.; Moss, W. O. *Org. Process Res. Dev.* **2003**, *7*, 67
(4) Ashworth, I. W.; Bowden, M. C.; Dembofsky, B.; Levin, D.; Moss, W. O.; Robinson, E.; Szczur, N.; Virica, J. *Org. Process Res. Dev.* **2003**, *7*, 74.
(5) Bowden, S. A.; Burke, J. N.; Gray, F.; McKown, S.; Moseley, J. D.; Moss, W. O.; Murray, P. M.; Welham, M. J. *Org. Process Res. Dev.* **2004**, *8*, 33−44.
(6) Hauser, F. M.; Sengupta, D.; Corlett, S. A. *J. Org. Chem.* **1994**, *59*, 1967.
(7) Harada, T.; Yoshida, T.; Inoue, A.; Takeuchi, M.; Oku, A. *Synlett* **1995**, 283.

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Published on Web 12/05/2003

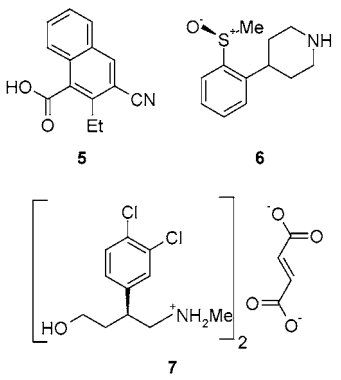
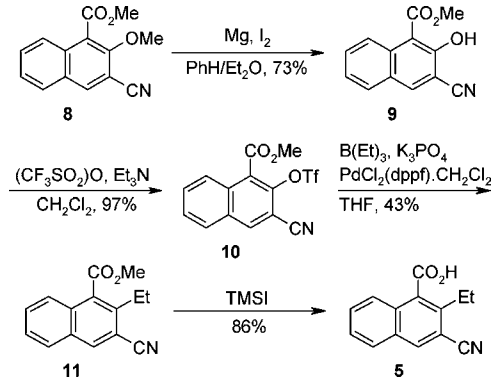


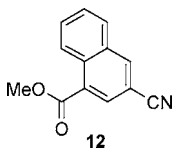
Figure 1.

Scheme 1



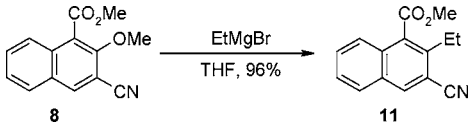
the benzene/diethyl ether system to toluene/diethyl ether resulted in iodination of the methyl group rather than demethylation. The procedure also produced a stoichiometric amount of toxic methyl iodide as the reaction byproduct, which could be accepted using the rapid parallel development approach, but still raised containment and handling concerns. Alternative conditions for this transformation were investigated, including the use of boron trichloride in dichloromethane; however, only partial demethylation was observed.

The other stage with significant problems was the palladium-catalysed cross-coupling reaction of ZM374979 Triflate **10** to prepare ZM374979 ester **11**. Competing β-elimination in the reaction afforded ZD6021 cyanoester **12**, which needed to be separated from the product by flash column chromatography. Finally, the catalyst for the cross-coupling reaction was extremely expensive, which could be accepted using rapid parallel development approach, but remained an issue for the delivery of future campaigns.

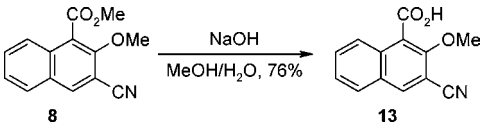


In light of the concerns raised by the evaluation of the Discovery route, it was decided to investigate alternative methods of preparing ZM374979 cyanoacid **5**. A survey of the literature on the displacement of aryl methoxy groups with alkyl groups yielded a number of references by Meyers and co-workers, on the use of *o*-oxazoline groups activating

Scheme 2



Scheme 3



the nucleophilic aromatic substitution of methoxy groups by Grignard reagents.[8] As a speculative reaction, it was decided to investigate the analogous reaction of ZD4974 cyanoester **8** with ethylmagnesium bromide to see whether the methyl ester would act in a manner similar to that of the oxazoline group in activating the nucleophilic aromatic substitution reaction. This reaction was successful, converting ZD4974 cyanoester **8** to ZM374979 cyanoester **11** (Scheme 2), with no significant reaction at either the ester or nitrile groups. It is likely that the electron-withdrawing character of both the ester and nitrile groups promotes the nucleophilic aromatic substitution such that this reaction occurs exclusively.

Development work optimised the Grignard reagent stoichiometry to 1.2 equiv. Higher charges led to other products, assumed to result from reaction at the nitrile or ester functions, whilst lower charges resulted in residual ZD4974 cyanoester **8**, which would be carried through subsequent stages.

ZM374979 cyanoester **11** has a low melting point and high solubility in a range of organic solvents, which hindered the development of a satisfactory crystallisation process. Trituration with isohexane promoted crystallisation of the crude oil, but recoveries were poor. Eventually, flash column chromatography was selected for purification of the ZM374979 cyanoester **11**, and the produced crude material was purified to afford 2.13 kg of product, with a 95% purity. The yield for the reaction, including chromatography, was 96%.

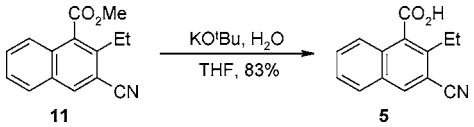
The final reaction required to produce ZM374979 cyanoacid **5** was the hydrolysis of ZM374979 cyanoester **11**. This transformation had been demonstrated by Discovery using trimethylsilyl iodide as both demethylating reagent and solvent. This reagent is highly corrosive, and its minimisation or elimination was the key objective of the initial development work. The trimethylsilyl iodide was reduced to a stoichiometric amount and the reaction conducted in dichloromethane; however, poor conversion was observed using these conditions.

The use of sodium hydroxide in aqueous methanol had been used in the analogous reaction for the preparation of ZD4974 cyanoacid **13** (Scheme 3).

These conditions were investigated for the conversion of ZM374979 cyanoester **11** to ZM374979 cyanoacid **5**, but the reaction gave a number of products, presumably arising from the competing hydrolysis of the nitrile group. Lithium hydroxide was investigated as an alternative base, but this reaction gave a similar outcome.

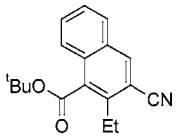
(8) Reuman, M.; Meyers, A. I. *Tetrahedron* **1985**, *41*, 837

Scheme 4



A survey of the literature identified the use of "anhydrous hydroxide"; a procedure developed by Gassman and co-workers for the hydrolysis of tertiary amides[9] and hindered esters,[10] as a method for the preparation of ZM374979 cyanoacid **5**. The procedure involves the in situ generation of one mole equivalent of hydroxide ion from one mole equivalent of water reacting with potassium *tert*-butoxide. The hydroxide ion initiates the desired hydrolysis, which is completed by the action of a second equivalent of *tert*-butoxide ion, but the absence of additional water stops undesired side reactions.

Application of the standard conditions, adding 2−4 mol equiv of base and 1 mol equiv of water at the start of the reaction, afforded a mixture comprising 70% ZM374979 cyanoacid **5** with 3% residual ZM374979 cyanoester **11**. No nitrile hydrolysis was observed, but 20% of a nonpolar impurity was detected which was shown to be ZM374979 *tert*-butylester **14**. Increasing the charge of potassium *tert*-butoxide resulted in a faster reaction but did not reduce the level of this impurity.



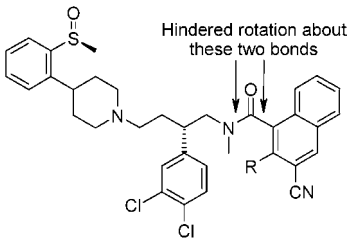
14

It was reasoned that the ZM374979 *tert*-butylester **14** was formed by a transesterification reaction between the *tert*-butoxide anion and the ZM374979 cyanoester **11** competing with the reversible attack of the hydroxide anion. Adding the base portionwise to maintain a low concentration in the reaction mixture successfully reduced the impurity to 7% (Scheme 4). The product could then be separated from the impurity by a simple acid/base workup, yielding material with excellent purity.

This procedure was used to produce 1.59 kg of product, with a 100% purity. The yield for the reaction was 83%.

Atropisomerism. The assembly of ZM374979 **4** from ZM374979 cyanoacid **5**, ZD7944 Pip sulfoxide **6**, and ZD6021 *N*-methylamine fumarate **7** has been described previously.[3] A significant challenge in this work, was the isolation of a single atropisomer of ZM374979 **4**.

Atropisomerism occurs due to restricted rotation about a single bond, with BINAP and related compounds being notable examples of molecules displaying this property. The area remains relatively obscure, with the last major review of the topic being published in 1983.[11] Atropisomerism has been observed in a number of molecules, including synthetically challenging targets such as vancomycin.[12] There have also been limited reports of manufactured pharmaceuticals displaying atropisomerism.[13,14]



1 (R = H)
3 (R = OMe)
4 (R = Et)

Figure 2.

Table 1. **Interconversion times for neurokinin antagonist and naphthamide atropisomers**

	neurokinin antagonists		naphthamides	
R	compound	$T_{1/2}$	compound	$T_{1/2}$
H	**1**	<5 min	**15**	1 s
OMe	**3**	5 h	**16**	19 h
Et	**4**	41 h	**17**	84 h

The interconversion time, or half-life of atropisomers, is an important physical property of these compounds. Molecules that rapidly interconvert (having short half-lives) cannot be easily isolated as single atropisomers. However, molecules that interconvert slowly (having long half-lives) allow isolation of single atropisomers. For pharmaceutical compounds displaying atropisomerism, the activity normally resides in a single atropisomer.[14]

Isolating a single enantiomer is a significant technical challenge, and possible approaches include preparative scale chromatography, asymmetric synthesis, selective crystallisation, resolution, and dynamic crystallisation. However, with only limited information available in the literature on this topic, an additional challenge was added to the delivery of ZM374979 **4**.

In the series of neurokinin antagonists prepared by Zeneca Pharmaceuticals, the atropisomerism occurs due to restricted rotation about the amide C−N bond and the aryl C−C bond (Figure 2).

As each compound was prepared by Discovery, the atropisomer interconversion time ($T_{1/2}$) was determined (Table 1). These results compare well to similar work by Clayden and co-workers, who measured the interconversion time for a comparable series of naphthamides **15**−**17**.[15]

ZD6021 **1** (and the analogous ZD2249 **2**) have a low interconversion time; thus, separation of the atropisomers is not possible. ZD4974 **3** and ZM374979 **4** atropisomers, with a slower rate of interconversion, can be separated by HPLC (Figure 3). This analytical work shows the compounds exist

(9) Gassman, P. G.; Hodgson, P. K. G.; Balchunis, R. J. *J. Am. Chem. Soc.* **1976**, *98*, 1275.
(10) Gassman, P. G.; Schenk, W. N. *J. Org. Chem.* **1977**, *42*, 918.
(11) Oki, M. *Top. Stereochem.* **1983**, *14*, 1.
(12) Williams, D. H. *Nature* **1999**, *397*, 567 and references therein.
(13) Friary, R. J.; Spangler, M.; Osterman, R.; Schulman, L.; Schwerdt, J. H. *Chirality* **1996**, *8*, 364.
(14) Chenard, B. L.; Devries, K. M.; Welch, W. M. WO9838187, 1998.
(15) Ahmed, A.; Bragg, R. A.; Clayden, J.; Lai, L. W.; McCarthy, C.; Pink, J. H.; Westlund, N.; Yasin, S. A. *Tetrahedron* **1998**, *54*, 13277.

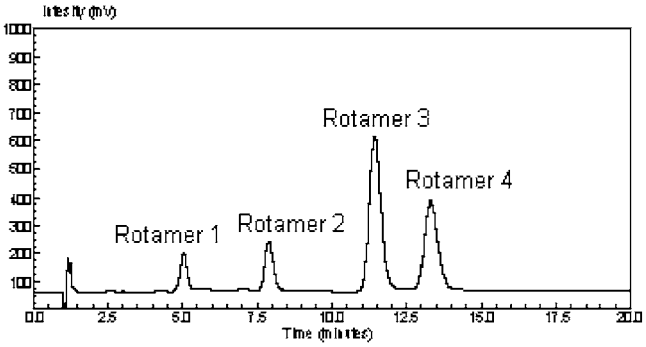
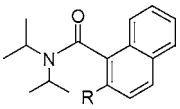


Figure 3. **HPLC analysis of ZM374979 4 atropisomers.**



15 (R = H)
16 (R = OMe)
17 (R = Et)

as a mixture of four atropisomers, which were named rotamers 1−4 on the basis of the order of elution on the HPLC system.

By comparing the neurokinin antagonists to similar systems reported by Clayden and co-workers,[16] a model of the four possible atropisomers (and the methods for their interconversion) has been postulated (Figure 4).

The first significant efforts to address the atropisomer issue were undertaken for the preparation of ZD4974 **3**. It was impossible to determine the active atropisomer of this compound, due to interconversion in vivo; thus, the target was to produce material that contained as near to equal proportions of the atropisomers as possible.

Early work by Discovery showed that, in solution, ZD4974 **3** (both free base and amine salts) atropisomers would interconvert to an equilibrium composition, with the ratio being dependent on the solvent used. On crystallisation, the amine salt counterion was found to have a significant effect on the atropisomer composition in the solid state (Table 2).

Initial development work focused on identifying robust crystallisation conditions that would afford a solid with good filtration properties and in a high yield. Crystallisation of the citrate salt from ethanol was shown to give 76% yield, and careful control of crystallisation conditions produced a form amenable to fast deliquoring. The ZD4974 **3** Citrate salt isolated by Discovery had contained a mixture of atropisomers similar to that found in solution, and this was judged to be an acceptable ratio for toxicological and clinical trials.

Thus, 1.77 kg of crude material was crystallised from ethanol as the citrate salt, affording 1.22 kg of ZD4974 **3** citrate pure, with a product purity of 97.3%. The yield for this stage was 66%. This material was then analysed to determine the atropisomer ratio (Table 3).

The difference in atropisomer ratio between the Discovery and Development materials was attributed to the former being amorphous, whereas the latter was crystalline. In an attempt

(16) Clayden, J.; Pink, J. H. *Angew. Chem., Int. Ed.* **1998**, *37*, 1937.

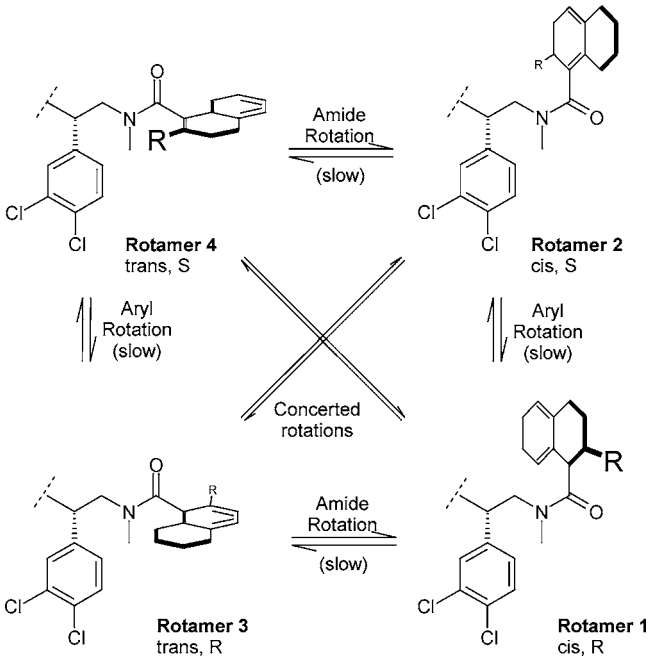


Figure 4. **Postulated atropisomers and methods of interconversion.**

Table 2. **Ratio of rotamers for salts of ZD4974 3 isolated by Discovery**

salt	rotamer 1	rotamer 2	rotamer 3	rotamer 4
citrate	6.0	13.0	53.0	28.0
fumarate	6.8	22.5	55.3	15.6
malonate	0.9	5.8	93.3	0.0

Table 3. **Ratio of rotamers of ZD4974 3 citrate pure isolated by development**

salt	rotamer 1	rotamer 2	rotamer 3	rotamer 4
citrate	1.1	1.5	89.2	8.3

to produce material with a more even distribution of atropisomers, the crystalline ZD4974 **3** citrate pure was redissolved and evaporated. However, this work resulted in an oil, rather than a solid.

In light of this result, it was decided to use the crystalline ZD4974 **3** Citrate Pure, despite the bias in atropisomer composition. It was reasoned that the atropisomers would interconvert in vivo, reverting to the solution equilibrium ratio.

For delivery of ZM374979 **4**, Discovery identified a single active atropisomer, rotamer 4. Due to the high interconversion time, it was necessary to develop a process to isolate this single atropisomer, so that it could be evaluated in toxicological and clinical trials. This represented a significant challenge for the Process Development Department, as the only known method of preparing rotamer 4 in greater than 95% purity had been the use of preparative HPLC.

With the isolation of ZD4974 **3** Citrate Pure containing high levels of a single atropisomer, it was reasoned that a similar approach could be used for the isolation of ZM374979 **4** rotamer 4. With the tight time deadlines of the rapid parallel development approach, an intensive program of screening

Table 4. **Organic acids and solvents screened during selective atropisomer crystallisation work**

organic acid	solvent
phthalic	alcohols
succinic	dimethyl sulfoxide
citric	tetrahydrofuran
malonic	dichloromethane
oxalic	acetonitrile
mesaconic	ethyl acetate
fumaric	toluene
L-tartaric	acetone
maleic	

solvents and organic bases (Table 4) was initiated to identify conditions which would favour the isolation of ZM374979 **4** rotamer 4.

A major concern with all the work in this area, was being able to produce material containing a consistent ratio of atropisomers for crystallisation studies. This problem was overcome by stirring the ZM374979 **4** in the crystallisation solvent at elevated temperature prior to salt formation. Typical ZM374979 **4** Crude contains the atropisomers in a ratio of 5:15:45:35 (rotamer 1:rotamer 2:rotamer 3:rotamer 4), which on equilibrium results in a ratio of 10:10:40:40. Profiling studies showed that 48 h at 60 °C would ensure that the equilibration ratio was reached, providing a consistent point from which crystallisation studies could be started.

The screen of organic acids and solvents identified maleic acid and methanol as the only combination that produced selectivity for rotamer 4, affording a 20% yield in 94.6% purity. Thus, attention focused on this result, which had used 9 volumes (relative to input weight) of solvent and 1 equivalent of acid.

Reducing the amount of solvent in the crystallisation improved the yield but reduced the atropisomer selectivity, with the amount of rotamer 3 in the isolated solid increasing. The use of anti-solvents (water, methyl *tert*-butyl ether, or isohexane) was investigated, which increased the crystallisation yield, but afforded material with the same atropisomer ratio as observed at equilibrium in solution.

Reducing the isolation temperature was investigated, but this led to reduced atropisomer selectivity, rather than the anticipated improvement in yield.

The amount of maleic acid used was also investigated, but reducing the acid charge inhibited crystallisation of the salt, leading to longer crystallisation times and a lower yield.

Seeding the crystallisation was found to improve the crystallisation, producing smaller crystals with better atropisomer selectivity. The use of this technique also improved consistency between experiments and was adopted for all experiment in this area.

Finally, the purity of input material was investigated. ZM374979 **4** free base, which had been purified by flash column chromatography, was found to give an improved yield in the crystallisation compared to unpurified material. Thus, the ZM374979 **4** free base produced was purified before crystallisation.[3]

The combination of these improvements afforded a robust process, which consistently afforded 23% of ZM374979 **4** maleate rotamer 4 in >97% purity. An isolated yield of 23% represents 58% of the available atropisomer at equilibrium.

The liquors isolated during this process contain >70% of the available ZM374979 **4**, and isolation of a further crop of rotamer 4 from this mixture was viewed as important to improve throughput. Equilibration of the ZM374979 **4** maleate was found to be possible using conditions identical to those used for ZM374979 **4** Free Base. Thus, the liquors were heated at 60 °C for 72 h, affording the previously observed ratio of 10:10:40:40 (rotamer 1:rotamer 2:rotamer 3:rotamer 4). The concentration of the liquors was adjusted as part of this process to 9 volumes (relative to input weight) of solvent by evaporation of solvent or addition of further ZM374979 **4** free base and maleic acid. Crystallisation of the equilibrated solution afforded a 23% yield of ZM374979 **4** maleate rotamer 4 in >96% purity. In combination, the crystallisation and liquor equilibration/crystallisation processes afforded ZM374979 **4** maleate rotamer 4 in a 40% yield.

The combined procedure was judged to be fit for purpose for Large Scale Laboratory operations, thus for the first crystallisation, 830 g of ZM374979 **4** free base was crystallised to afford 237 g of ZM374979 **4** maleate rotamer 4 with a purity of 94.7%. The yield was 23% as expected. On scaling-up, the crystallisation was found to be much slower than that observed in the laboratory, which may explain the lower purity. The majority of the additional impurity was ZM374979 **4** maleate rotamer 3.

The isolation of 237 g of ZM374979 **4** maleate rotamer 4 met the initial demand for material for preliminary toxicity, and the purity was judged fit for the intended purpose. It was thus decided not equilibrate the liquors and attempt isolation of a second crop of material.

At this time, the neurokinin antagonist project was put on hold, pending toxicology results. Thus, no further work was undertaken into the selective atropisomer crystallisation or into finding the reasons for the slower crystallisation and lower purity on scale-up. Should development have continued, the introduction of a dynamic process was envisaged, involving the controlled crystallisation of ZM374979 **4** maleate rotamer 4 with simultaneous reequilibration of the free base liquors.

However, the developed process afforded ZM374979 **4** maleate rotamer 4 in excellent purity, representing a significant achievement, considering the tight time schedule and scientific challenge involved.

Conclusions

ZM374979 cyanoacid **5** was prepared from ZD4974 cyanoester **8** using Zeneca Pharmaceuticals' rapid parallel development approach. Investigation of the Discovery conditions for this transformation showed that the chemistry was unsuitable for Large Scale Laboratory preparation. Thus, alternative conditions were introduced using a selective Grignard reaction and a selective ester hydrolysis.

On conversion of ZM374979 cyanoacid **5** to ZM374979 **4**, atropisomerism was observed. A process for the selective crystallisation of a single atropisomer was developed, which was effective on a large scale, affording ZM374979 **4** maleate

rotamer 4 in 95% purity. The developed process represents a significant contribution to the delivery of single atropisomers in the pharmaceutical industry.

Adverse toxicology results for all the compounds (ZD6021 **1**, ZD2249 **2**, ZD4979 **3**, and ZM374979 **4**) in this section of the neurokinin antagonist project, resulted in the termination of development effort in this area. However, the project had allowed extensive opportunities to demonstrate the rapid parallel development approach, scaling-up more than 60 processes to 20−100-L scale.[1−5]

Experimental Section

General Procedures. Melting points were determined using a Griffin melting point apparatus (aluminium heating block) and are uncorrected. ^{1}H NMR spectra were recorded on a Varian Inova 400 MHz spectrometer with chemical shifts given in ppm relative to TMS at $\delta = 0$. The reaction mixtures and products were analysed by reverse phase HPLC on a Hewlett-Packard 1100, according to the following conditions: column, Waters Spherisorb S5ODS1, 250 mm × 4.6 mm i.d.; eluent, 560:440 acetonitrile:water with 0.1% v/v trifluoroacetic acid; flow rate, 1.0 mL/min; wavelength, 235 nm; injection volume, 5 μL; column temperature, 25 °C. HPLC purities were % w/w against a standard of known purity as determined by ^{1}H NMR. Analytical TLC was carried out on commercially prepared plates coated with 0.25 mm of self-indicating Merck Kieselgel 60 F_{254} and visualised by UV light at 254 nm. Preparative scale silica gel flash chromatography was carried out by standard procedures using Merck Kieselgel 60 (230−400 mesh).

Preparation of ZM374979 Cyanoester 11. ZD4974 cyanoester **8** (45.00 g, 190.0 mmol, 1.00 equiv) was dissolved in anhydrous tetrahydrofuran (300 mL), purged with nitrogen for 15 min and cooled in a cold water bath. Ethylmagnesium bromide (1 M in THF, 220 mL, 220 mmol, 1.16 equiv) was added dropwise over 1.5 h, and stirring continued for a further 15 min. An HPLC check confirmed that all starting material had been consumed. The reaction mixture was slowly transferred into saturated ammonium chloride solution (500 mL), keeping the temperature below 30 °C. The two-phase mixture was stirred for a further 30 min before separating. The aqueous phase was extracted with MTBE (2 × 150 mL). The organic extracts were combined and concentrated in vacuo to leave the product as a dark oil. The oil was dissolved in dichloromethane (100 mL) and passed through a short silica column (250 g SiO_2), eluting with dichloromethane. The column fractions were monitored by TLC (20% ethyl acetate/isohexane). The product-containing fractions were concentrated in vacuo to leave the product initially as a light oil which crystallised on standing. The product was dried in vacuo at 50 °C to a constant weight, affording ZM374949 cyanoester **11** as a white, crystalline solid (41.1 g, 92.0%). HPLC purity 95%; t_R 12.5 min; mp 77−79 °C; δ_H (400 MHz, $CDCl_3$) 1.36 (t 3H $J = 7$ Hz), 2.96 (q 2H $J = 7$ Hz), 4.06 (s 3H), 7.56 (td 1H $J = 7$, 4 Hz), 7.65 (td 1H $J = 7$, 4 Hz), 7.75 (d 1H $J = 7$ Hz), 7.86 (d 1H $J = 7$ Hz), 8.28 (s 1H); δ_C (100 MHz, $CDCl_3$) 15.44, 26.55, 52.64, 110.77, 117.70, 124.93, 127.28, 128.55, 130.12, 130.72, 131.46, 131.81, 136.71, 139.18, 168.68.

Preparation of ZM374979 Cyanoacid 5. Potassium *tert*-butoxide (4.46 g, 40.0 mmol, 1.00 equiv) was dissolved in anhydrous THF (80 mL) under an inert atmosphere and cooled in a cold water bath. Water (0.72 mL, 40.0 mmol, 1.00 equiv) was added dropwise to the solution and stirred for a further 15 min. A solution of ZM374979 cyanoester **11** (10.00 g, 40.0 mmol, 1.00 equiv) was added over 15 min and stirred for a further hour. Three additional charges of potassium *tert*-butoxide (4.46 g, 40.0 mmol, 1.00 equiv) followed by an hour hold were made to the reaction mixture. The reaction mixture was added slowly to saturated ammonium chloride solution (100 mL) and stirred. The layers were allowed to separate, and the organic phase was retained. The aqueous phase was adjusted to pH 1 by the addition of concentrated HCl and extracted with MTBE (2 × 50 mL). The combined organic extracts were concentrated in vacuo to leave an oil. The product was dissolved in dichloromethane (100 mL) and extracted with 1 M NaOH (100 mL). The aqueous phase was extracted with dichloromethane (100 mL) and the organic phase retained. The aqueous phase was adjusted to pH 1 by the addition of concentrated HCl and extracted with dichloromethane (50 mL). The combined organic extracts were washed with brine (100 mL), and the solvent was removed in vacuo to afford ZM374979 cyanoacid **5** as a white solid (8.20 g, 87.0%). HPLC purity 100%; t_R 5.7 min; mp 140−142 °C; δ_H (400 MHz, DMSO-d_6) 1.11 (t 3H $J = 7$ Hz), 2.94 (q 2H $J = 7$ Hz), 7.69 (td 1H $J = 7$, 4 Hz), 7.80 (td 1H $J = 7$, 4 Hz), 7.88 (d 1H $J = 7$ Hz), 8.10 (d 1H $J = 7$ Hz), 8.67 (s 1H); δ_C (100 MHz, DMSO-d_6) 15.26, 25.77, 109.62, 117.65, 124.71, 127.37, 128.78, 130.27 130.36, 130.40, 133.29, 136.39, 136.80, 169.03.

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