Pergamon

Bioorganic & Medicinal Chemistry Letters 12 (2002) 1799–1802

BIOORGANIC & MEDICINAL CHEMISTRY LETTERS

Synthesis of a Novel, Recyclable, Solid-Phase Acylating Reagent

Robert B. Nicewonger,* Lori Ditto, Doug Kerr and Laszlo Varady

Department of Chemistry ArQule, Inc., 19 Presidential Way, Woburn, MA 01801, USA

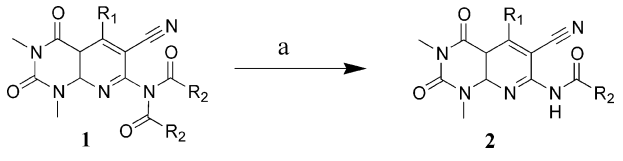
Received 30 November 2001; accepted 20 February 2002

Abstract—In this paper, we describe the synthesis of *N*-(6-cyano-1,3-dimethyl-2,4-dioxo-5-substituted-1,3-dihydropyridino[2,3-*d*]pyrimidin-7-yl)imides **1**. We will show the synthesis of **1** using both conventional heating and microwave techniques. In addition, the imide was attached to polystyrene and this immobilized imide was equally as effective at acylating various primary and secondary amines as its solution-phase counterpart.

The acylation of amines is one of the most common reactions in all of organic chemistry. Among the numerous reagents available for this transformation are acyl halides, anhydrides, and activated esters prepared from an acid and coupling reagents such as EDC, DCC, BOP, PyBOP, HOBt, CDI, HBTU, or HATU.[1] Solid-phase coupling agents have also been developed and are available commercially. These include polymer supported EDC, HOBt, tetrafluorophenol, and a few different carbodiimides.

During the course of a library synthesis, we noticed that *N*-(6-cyano-1,3-dimethyl-2,4-dioxo-5-substituted-1,3-dihydropyridino[2,3-*d*]pyrimidin-7-yl)imides **1** could be deacylated with a macroporous Tris resin (Scheme 1).[2] At this point we realized a potential use of these compounds as acylating reagents and decided to pursue.

We will show the synthesis of this imide using both conventional organic techniques and microwave technology (optimization of the microwave parameters will be discussed in detail) as well as the acylation of several amines. With the appropriate linker, the imide could be bound to a solid-phase support. We will show the synthesis of the solid-phase imide and its reaction with various amines.



1 **2**

Scheme 1. Deacylation of imide **1**. Reagents and conditions: (a) macroporous-Tris amine resin, pyridine/acetonitrile (1:1).

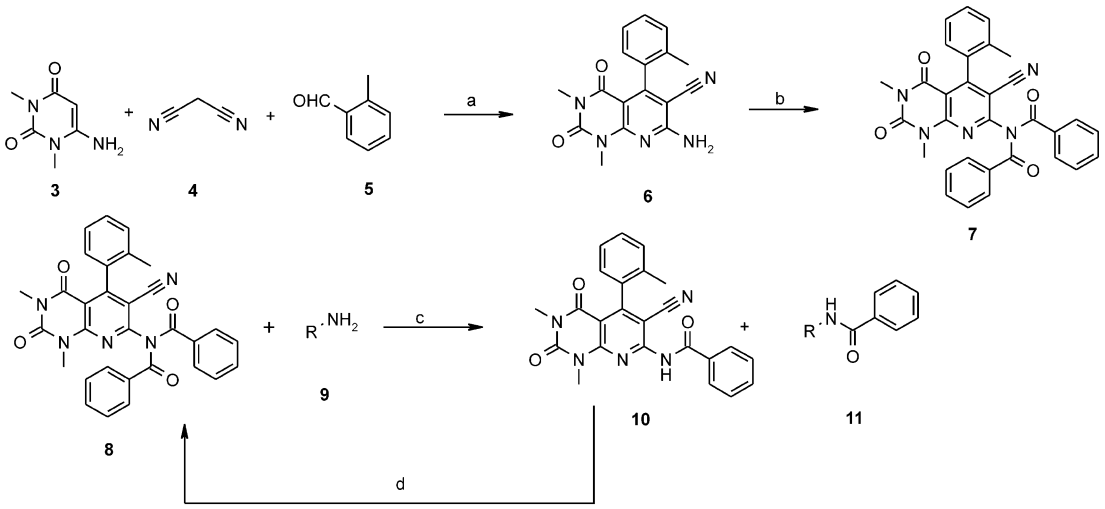
Synthesis of the imide is shown in Scheme 2. Amine **6** was generated from a three component condensation reaction between 6-amino-1,3-dimethyl uracil, cyanomalonate, and *o*-tolualdehyde.[3] This amine was very unreactive toward general electrophiles such as anhydrides, isocyanates, and isothiocyanates. In fact, the only conditions that allowed a reaction to occur at the exocyclic nitrogen was with excess acid chloride in refluxing pyridine for prolonged times (12–48 h). With our recent interest in shortening reaction times using microwave assisted organic synthesis and in optimizing chemical reactions using statistical design of experiments,[4] we thought this reaction would be an ideal candidate to apply both principles.

To find optimal microwave reaction conditions, we used design of experiments (DoE). To generate our model, we chose a quadratic design with three variables—equivalents of benzoyl chloride, microwave power, and time of reaction. The worksheet of experiments that were performed is shown in Table 1. Examination of the results show that for product formation, there is a quadratric dependence on equivalents of acid chloride and also a strong positive interaction between power and time (i.e., the reaction could be done at low power for a long time or high power for a short time).[5] Maximum product formation was predicted and found to occur with 12.4 equiv acid chloride at 300 W for 2 min.

Imide **8** was then reacted with amines in solvents such as dichloromethane, THF, or acetonitrile to generate benzoyl amides **11** (Scheme 2). Table 2 shows the results from a variety of amines (see Fig. 1). Note that no reaction occurred with anilines or sterically hindered

*Corresponding author. Tel.: +1-781-994-0496; fax: +1-781-376-6019; e-mail: rnicewonger@arqule.com

0960-894X/02/$ - see front matter
PII: S0960-894X(02)00272-X



Scheme 2. Synthesis of imide **8** and use as an acylating reagent. Reagents and conditions: (a) isopropanol, reflux, 24 h; (b) benzoyl chloride, pyridine, reflux, 36 h or microwave irradiation 300 W, 2 min; (c) dichloromethane, rt; (d) silica gel purification, then benzoyl chloride, pyridine, reflux, 36 h or microwave irradiation 300 W, 2 min.

amines even at 50 °C for 24 h. Secondary amines reacted sluggishly, but could be pushed by heating to 50 °C overnight with selective *N*-acylation in the presence of hydroxyl groups (see entries **17**–**19**).

It should also be noted that the imides were stable to prolonged treatment in methanol (3 days at 50 °C). Primary amines and piperazines reacted quickly at room temperature. The by-product amide **10** of the reaction could be isolated, reacylated with an acid chloride and used in further acylations reactions.

The resin-bound version of **1** was constructed as shown in Scheme 3. The three component condensation of aminouracil, dicyanomalonate, and 4-carbomethoxybenzaldehyde generated ester **27**. Hydrolysis of the ester proved challenging—even under reflux with conventional heating, no product was observed after 7 days. We again turned to microwave irradiation conditions—165 °C for 20 min proved successful. The high temperature was necessary to get the ester into solution. Standard peptide coupling conditions afforded the resin-bound amine **29**. The same reaction conditions used to make the imide in solution were applied to the solid phase—12.4 equiv acid chloride, 300 W of microwave power for 2 min using pyridine as a solvent. (Note that if thermal conditions were applied here—excess acid chloride, 12–36 h in refluxing pyridine—the resin turned deep brown and could not be thoroughly washed. Even after extensive Soxhlet extraction, leachables were present in high amounts.) The loading of the resin was 0.37 mmol/g as determined by reaction with tryptamine **26**.

Table 1. Experiments generated by CCF design

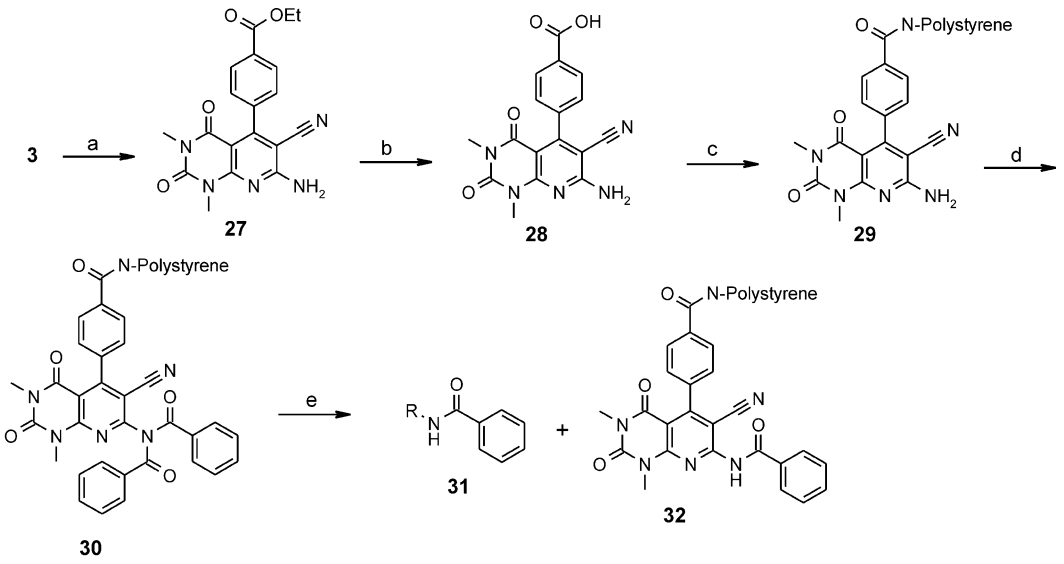
Experiment number[a]	Benzoyl chloride (equiv)	Power (W)	Time (min)	Relative reaction yield[b]
1	2	30	2	5
2	20	30	2	29
3	2	300	2	53
4	20	300	2	100
5	2	30	10	35
6	20	30	10	95
7	2	300	10	0
8	20	300	10	10
9	2	165	6	28
10	20	165	6	40
11	11	30	6	52
12	11	300	6	32
13	11	165	2	94
14	11	165	10	32
15	11	165	6	77
16	11	165	6	41
17	11	165	6	62

[a]Reactions were carried out by adding benzoyl chloride to a solution of **6** in pyridine. All samples were irradiated using a Discover single-mode microwave instrument from CEM.
[b]Relative reaction yield was determined by measuring the HPLC area of the product amide. The largest peak is designated as 100. Values for each of the other experiments is calculated as a ratio of peak area divided by the maximum peak area.

Table 2. Reaction conversions[a] for solution-phase benzoylation of amines **12**–**24**

Amine	% Conversion in 2 h at rt	% Conversion o/n at rt	% Conversion o/n at 50 °C
12	0	0	0
13	0	0	0
14	0	0	0
15	0	0	0
16	0	0	0
17	0	30	56
18	14	56	95
19	35	65	72
20	100	nr	nr
21	93	100	nr
22	100	nr	nr
23	100	nr	nr
24	100	nr	nr

[a]Aliquots were removed from reactions at given times and diluted with DMSO. Conversion was measured by the disappearance of starting material. Percentages were calculated by measuring the HPLC peak area of the remaining starting material at the given time divided by the HPLC peak area of the starting material peak with no acylating reagent present.



Scheme 3. Synthesis of polymer bound imide and use as an acylating reagent. Reagents and conditions: (a) 4-carbomethoxybenzaldehyde, cyanomalonate, isopropanol, reflux, 24 h; (b) microwave irradiation, 165 °C, 20 min 1 M HCl/MeOH (3:1); (c) nova aminomethyl polystyrene, HATU, NMM, DMF; (d) 12.4 equiv benzoyl chloride, pyridine, microwave irradiation 300 W, 2 min; (e) RNH_2, dichloromethane, 2–24 h, 25–50 °C.

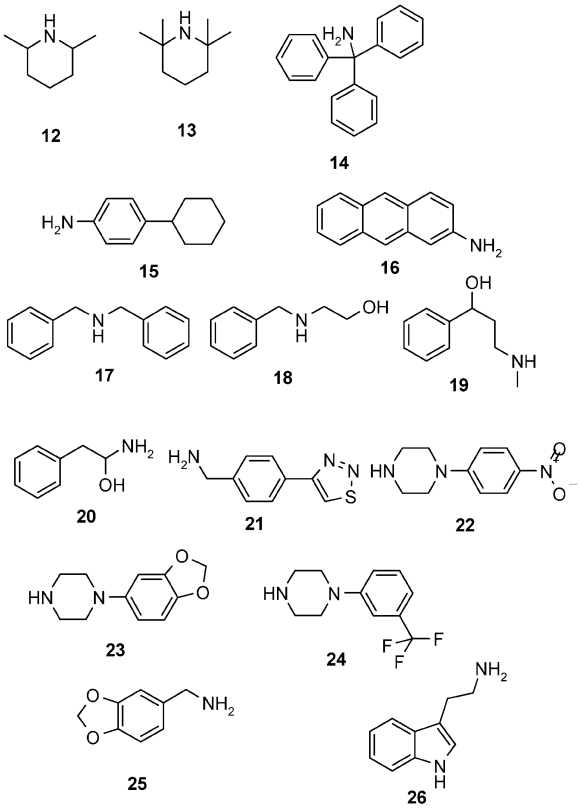


Figure 1. Amines used in the acylation study.

Compound **30** was used to acylate the same amines as in the previous solution-phase examples. Table 3 shows the results. Resin bound imide **30** was equally effective at acylating primary amines and piperazines. Just as its solution phase counterpart, **30** did not acylate anilines. The by-product of the acylation is resin bound amide **32**, which could be washed and reactivated for acyl donation by repeating the microwave acylation conditions. After one such cycle, the loading was measured and found to be 0.34 mmol/g, only slightly lower than the original 0.37 mmol/g.

Table 3. Reaction conversions[a] for solid-phase benzoylation of selected amines

Amine	Conversion o/n at rt
15	0
16	0
21	89
22	98
23	93
24	96
25	88
26	96

[a]Conversion was measured by the disappearance of starting material. Percentages were calculated by measuring the HPLC peak area of the remaining starting material at a given time divided by the HPLC peak area of the starting material peak with no acylating resin present.

In conclusion, we have demonstrated a synthesis of *N*-(6-cyano-1,3-dimethyl-2,4-dioxo-5-substituted-1,3-dihydropyridino[2,3-*d*]pyrimidin-7-yl)imides **1** using both conventional and microwave heating. These imides function as acylating reagents in solution. Primary amines and piperazines reacted quickly at room temperature, while more hindered secondary amines required more time and heat. Anilines were unreactive. A solid-phase imide was also prepared and shown to be equally effective as an acylation reagent. This resin bound acylating reagent was shown to be recyclable after washing with DMF and reactivation using novel microwave conditions.

References and Notes

1. (a) For a more complete list of coupling reagents, refer to: Larock, R. C. *Comprehensive Organic Transformations: A Guide to Functional Group Preparations*; VCH: New York, 1989; p. 972. (b) March, J. *Advanced Organic Chemistry:*

Reactions, Mechanisms, and Structure; John Wiley & Sons: New York, 1985; p. 371, and references cited therein.
2. Nicewonger, R. B.; Ditto, L.; Varady, L. *Tetrahedron Lett*. **2000**, *41*, 2323.
3. For a two-step sequence to make the amine, refer to: Bhuyan, P.; Boruah, R. C.; Sandhu, J. S. *J. Org. Chem*. **1990**, *55*, 568.
4. (a) Nicewonger, R. B.; Kerr, D.; Varady, L. *Book of Abstracts, Part 2*, 222nd National Meeting of the American Chemical Society, Chicago, IL, Aug 26–30, 2001; American Chemical Society: Washington, DC, 2001; ORGN 528. (b) Kerr, D.; Nicewonger, R.B.; Varady, L. *Book of Abstracts, Part 2*, 222nd National Meeting of the American Chemical Society, Chicago, IL, Aug 26–30, 2001; American Chemical Society: Washington, DC, 2001; ORGN 291.
5. For all DoE experiments, we used Modde 6.0 software package from Umetrics, Inc.