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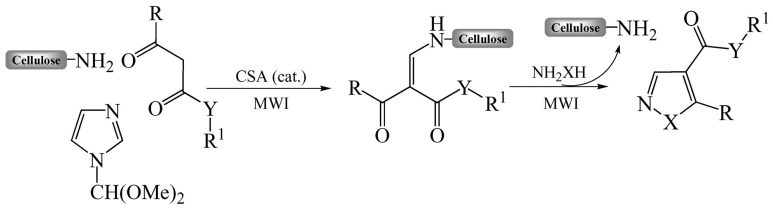

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Cellulose Beads: a New Versatile Solid Support for Microwave-Assisted Synthesis. Preparation of Pyrazole and Isoxazole Libraries

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The synthesis of libraries of substituted pyrazoles and isoxazoles has been developed via in situ generation of polymer-bound enaminones. The synthetic protocol makes use of commercially available aniline cellulose, a low-cost and versatile biopolymer, under very mild conditions. This new support allowed us to carry out reactions in polar solvents under both conventional heating and MW irradiation without degradation of the polymer. The reaction between cellulose-bound enaminone and hydroxylamine or hydrazines to afford the target heterocycles in high yields directly in solution is the key step. The support can be conveniently recycled.

Introduction

The importance of solid-phase organic synthesis is rapidly increasing as the concept of combinatorial chemistry begins to attract growing attention in the drug discovery community.[1] The physical and chemical properties of solid supports play a decisive role in a specific synthesis. The reactivity of a resin depends on both its chemistry and the environment in which the support is used.[2] Moreover, the access of reagents to reactive sites within the polymer matrix is crucial for the success of a synthesis.[3] Resins based on polystyrene cross-linked with divinylbenzene[4] are the most widely used solid supports for organic synthesis; they have a good swelling in low-polarity solvents such as benzene, toluene, halogenated hydrocarbons and tetrahydrofuran (THF), thereby "opening" the matrix. Therefore, they enable a number of reactions to be carried out. However, they do not swell efficiently in aliphatic hydrocarbons or polar solvents, such as methanol and water. Moreover, polystyrene resins suffer thermal and chemical instability and extensive adsorption of reagents. Since "one-bead" screening[5] for biological activity is typically carried out in aqueous solutions, the use of polar solvents in solid-phase synthesis should be a requirement for many of today's supports. A long-standing goal has thus been the development of new resins that ease the shortcomings of PS−DVB.[6,7] The selection of possible supports introduced and tested over the years includes polyamides,[8] polyethylene−polystyrene films,[9] cotton and other carbohydrates,[10] controlled-pore silica glass,[11] poly(ethylene glycol)−polystyrene (PEG−PS and TentaGel) graft resins, poly(ethylene glycol)−polyacrylamide (PEGA) resins,[12] tetraethylene glycol diacrylate/cross-linked polystyrene, and chemically modified polyolefin particles ("ASPECT").[13]

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Results and Discussion

As a part of a general program looking at base matrixes, alternative to both polystyrene and PEG-based resins commonly used in SPOS and PASP applications, we have examined the applicability of various beaded cellulose supports to organic synthesis. Cellulose shows good swelling properties in polar and aqueous solvents, and it is biodegradable, too.[14] Since the early reports by Frank, derivatized cellulose has found widespread applications in the synthesis of peptides and oligonucleotides, both in combinatorial and in high throughput parallel synthesis formats.[15,16]

Classical cellulose sheets have been used in multiple peptide synthesis with the SPOT technique.[17] Recently, several problems related to the use of fibrous and powdered cellulose have been solved by introducing new forms, which are both porous and spherical.[18] These cellulose supports, notably beads, can offer considerably higher loading levels than those obtained with planar supports. Consequently, these new beads appear well suited for the synthesis of libraries and heterogeneous screening assays. Surprisingly, to the best of our knowledge, only a single report has been published. In that paper Steel et al.[19] describe the use of a cellulose-based resin as a scavenger support to remove the bromine excess or acylating agents.

Herein, we wish to report the novel use of a commercially available[20] aniline functionalized cellulose as support for chemical synthesis. The cellulose employed was a modified bead-form containing aminoaryl-ethyl sulfone groups in flexible chains (Figure 1).

To prepare a library of pharmacologically relevant heterocyclic scaffolds with a good level of potential molecular diversity, we decided to use this natural polymer as support for the preparation of a library of highly substituted pyrazoles and isoxazoles. Our choice for these heterocycles was based on their intrinsic interest as a basic structure of several drugs[21] and on the possibility to have substituted scaffolds with

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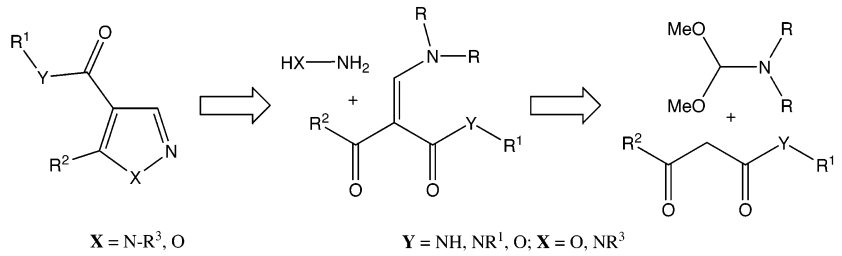
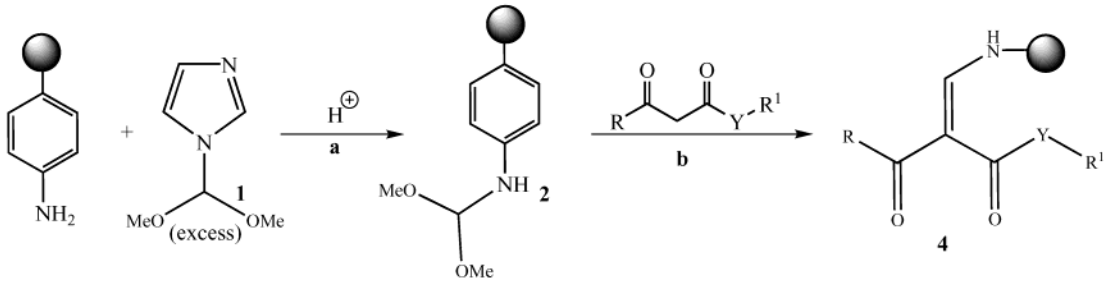
Scheme 1. Retrosynthetic Analysis of a Library of Pyrazoles and Isoxazoles



X = N-R^3, O **Y** = NH, NR^1, O; **X** = O, NR^3

Scheme 2. Synthesis of *N*-Formamide Dialkylacetal-Functionalized Cellulose



Reagents and conditions: (**a**) DMF, CSA (cat.), 80 °C, 36 h; (b) DMF, 80 °C, 6 h.

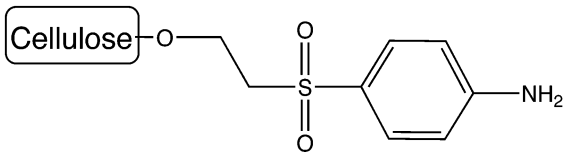


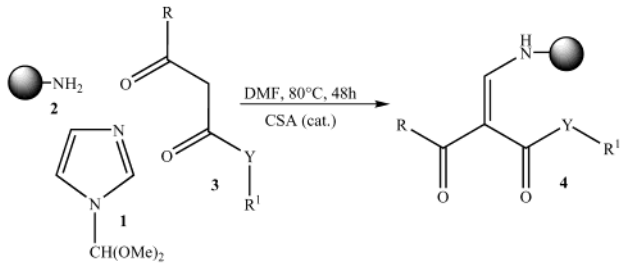
Figure 1. Aniline-functionalized cellulose.

potentially large molecular diversity.[22] For example, substituted pyrazoles have already been prepared in the solid phase[23] and in solution[24] following different synthetic strategies in which hydrazine derivatives cyclize with 1,3-dicarbonyl or α,β-unsaturated carbonyl compounds. We decided to follow the retrosynthetic approach reported in Scheme 1, originally described by Schenone and coworkers,[25] and further employed by a group at Pfizer to prepare a selective NHE-1 inhibitor.[26] The sites for molecular diversity in this approach are the substituents on hydrazine (NR^3), the derivatives of the carboxylic function (ester OR^1 or amide NHR^1), and the substituents on the starting β-keto ester (R^2).

According to Scheme 1, enaminones were identified as convenient starting materials because they can react with different bidentate nucleophiles to give pyrazoles and isoxazoles. We decided to anchor the β-enamino ketones on cellulose support in order to have a versatile rapid synthesis. Consequently, the NH_2 group on cellulose was an attractive point to realize a "catch and release" approach delivering the target libraries with minimal purification steps. Utilizing this functionality, we needed to prepare a formamide acetal on solid support. We decided, therefore, to prepare a modified Bredereck's reagent[27] **1** to convert the amino group of cellulose into the formamide acetal[28] **2** (Scheme 2) to immobilize a β-keto ester or amide.

The cellulose was so treated with an excess of formyl imidazole **1** under acid-catalyzed conditions (camphorsulfonic acid: CSA) in DMF. The mixture was heated at 80 °C for 36 h to afford the functionalized support **2** and then treated with an excess of a β-dicarbonyl compounds in DMF

Scheme 3. Preparation of Cellulose-Bound Enaminone



4 a-c: **Y** = O; **R** = Me; **R**^{1} = $PhCH_2$, Et, Me
4 d-j: **Y** = O; **R** = ^{i}Pr; **R**^{1} = Et, Allyl, Ph, $PhCH_2CH_2$, ^{i}Pr, -CH_2CH_2OH, -CH_2CH_2CN
4 k-m: **Y** = O, **R** = ^{t}BuCH_2; **R**^{1} = $PhCH_2CH_2CH_2$, -CH_2CH_2OH, -CH_2CH_2CN
4 n-r: **Y** = N; **R** = ^{i}Pr; **R**^{1} = Ph, $PhCH_2$, $PhCH_2CH_2$, cyclopentyl, cyclohexyl

Reagents and conditions: (a) DMF, 80 °C, 48 h. Yields are >99% (as determined by colorimetric assays of cellulose beads).

at the same temperature for 6 h to give the corresponding solid-supported β-enaminodiones **4** that were isolated by filtration.[28]

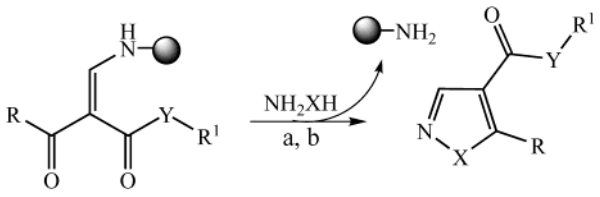
We prepared compound **4** in an "one-pot" reaction, too. Therefore, a mixture of cellulose supported *N*-formylimidazole dimethylacetal **1** and a β-keto ester **3** were mixed and heated for 48 h in DMF in the presence of 10% CSA as catalyst (Scheme 3). The filtration from the beads gave β-enaminodione **4** in good yields (loading measured with respect to that of the starting cellulose).[30]

The synthetic strategy was initially optimized using commercial β-keto esters and subsequently extended to β-keto esters and β-keto amides prepared on solution phase according to the classical malonic ester method.[31]

Treatment of the cellulose-bound enaminones **4a**−**r** with monosubstituted hydrazines or hydroxylamine afforded the corresponding isomerically pure pyrazoles **5** or isoxazoles **6** (Table 1), restoring the starting aniline cellulose.

The heterocycle cyclization was carried out to completion by refluxing **4** in ^{i}PrOH in the presence of several hydrazines or hydroxylamines. After 1 h of refluxing, pyrazoles **5a**−**z** were obtained in excellent yields, whereas isoxazoles **6a**−**k** needed 5 h of refluxing. At least 5 equiv of hydrazines or

Table 1. Regiospecific Synthesis of 1,4,5-Trisubstituted Pyrazoles and 4,5-Disubstituted Isoxazoles Using Cellulose-Bound Enaminones **4**[a]



4a-r **5-6**

entry	R	R^1	Y	X	% yield
5a	Me	$PhCH_2$	O	*N*-Ph	99
5b	Me	Et	O	*N*-Ph	99
5c	^{i}Pr	Et	O	*N*-Ph	98
5d	^{i}Pr	allyl	O	*N*-Ph	96
5e	^{i}Pr	$PhCH_2$	O	*N*-Ph	98
5f	^{i}Pr	$PhCH_2CH_2$	O	*N*-Ph	94
5g	^{i}Pr	$Ph(CH_2)_2CH_2$	O	*N*-Ph	94
5h	^{i}Pr	^{i}Pr	O	*N*-Ph	99
5i	^{i}Pr	^{t}BuOCH$_2$(Me)CH	O	*N*-Ph	95
5j	^{i}Pr	Cbz-CH_2CH_2	O	*N*-Ph	99
5k	^{i}Pr	Cbz-prolinol	O	*N*-Ph	98
5l	$PhCH_2$	4-(OH)-cyclohexyl	O	*N*-Ph	95
5m	^{t}BuCH$_2$	$HO(CH_2)_2CH_2$	O	*N*-Ph	96
5n	Et(Ph)CH	$EtCH(OH)CH_2CH_2$	O	*N*-Ph	94
5o	^{i}Pr	$(MeO)PhCH_2$	NH	*N*-Ph	98
5p	^{i}Pr	$PhCH_2CH_2$	NH	*N*-Ph	98
5q	^{i}Pr	Et	NEt	*N*-Ph	99
5r	^{t}BuCH$_2$	cyclopentyl	N	*N*-Ph	97
5s	Me	$PhCH_2$	O	*N*-$CONH_2$	94
5t	^{t}BuCH$_2$	^{t}BuOCH$_2$(Me)CH	O	*N*-$CONH_2$	95
5u	Me	Me	O	*N*-(2,4-Ph$(NO_2)_2$)	97
5v	Me	Et	O	*N*-(5-benzothiazol)	97
5z	Me	Me	O	*N*-(5-(4-Cl-Ph)thiazol	98
6a	Me	Ph	O	O	99
6b	Me	Me	O	O	98
6c	Me	Et	O	O	98
6d	^{t}BuCH$_2$	$CNCH_2CH_2$	O	O	97
6e	^{t}BuCH$_2$	$Ph(CH_2)_2CH_2$	O	O	96
6f	^{i}Pr	Et	O	O	97
6g	^{i}Pr	Et	NEt	O	98
6h	^{i}Pr	$-(CH_2)_2O(CH_2)_2$	N	O	97
6i	Me	4-CF_3−$PhCH_2$	N	O	99
6j	^{t}BuCH$_2$	cyclopentyl	N	O	98
6k	^{t}Bu	cyclohexyl	N	O	99

[a] Reagents and conditions: (a) 6a−k, X = O, ^{i}PrOH, 5 h, reflux; (b) 5a−z, X = NR, ^{i}PrOH, 1 h, reflux (5s and 5t, 20 h reflux).

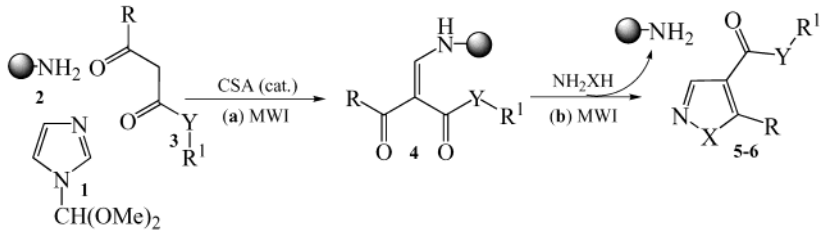
hydroxylamine was necessary to force the reaction to completion. The excess of reagents could be easily removed through an aqueous acidic workup or by using an amine acid resin scavenger. Pyrazoles and isoxazoles were obtained in high yields and excellent purity (>95%, as judged by HPLC/UV220/MS measurements) independently on the structure of the β-keto ester or β-keto amide employed. Using semicarbazides, the cyclization reaction (entryies **5s** and **5t**) requires up to 20 h. The cellulose, differently from the classic polystyrene (PS-DVB) resins, has good swelling properties in ^{i}PrOH. Moreover, at refluxing temperature, cellulose is much more resistant than common synthetic polymeric matrix to thermal shock.

So the cellulose used can be conveniently regenerated in turnover cycle. The procedure described could be, in fact, repeated using recycled cellulose, with high yields of the desired products having excellent levels of purity for at least four cycles, after which yields dropped to 85%.

We decided to optimize the timing of the protocol using microwave irradiation because the key steps of the synthesis were endothermic reactions. In recent years, the concept of speeding up resin-bound chemistry by microwave activation has created a lot of interest from both the academic and industrial communities.[32−34] In a significant number of publications, rate accelerations and very high loading for several solid-phase protocols have been reported, with reaction times being reduced in some cases from hours to a few minutes.[35]

Thus we reinvestigated the "one-pot" Bredereck-type condensation (Scheme 4) under microwave irradiation. Cellulose-bound enaminone synthesis can be effectively performed in high yields within 15 min using a self-tunable microwave synthesizer at 80 °C.[36] The reaction was carried out in an open vessel to allow the removal of the formed methanol from the equilibrium.[37] Even the further cyclization was successful carried out in ^{i}PrOH under microwave irradiation, giving high yields of the desired pyrazoles and

Scheme 4. Microwave-Assisted Synthesis of Pyrazoles and Isoxazoles Library



Reagents and condition: (a) DMF, 80 °C, 15 min. (b) ^{i}PrOH, 82 °C, 15 min. Reaction regulated via a temperature sensor in a MW oven. All reactions were carried out utilizing controlled single-mode microwave irradiation under open-vessel conditions. For details, see text. The yield of isolated compound after workup was >95%, and the purity was >98%, as determined by HPLC. For a graphical representation of all compounds **5**−**6**, see Figure S in the Supporting Information.

isoxazoles after 15 min of heating at 82 °C, again working in an open flask.[38] Moreover, using the MW-assisted protocol, the cellulose-supported aniline could be recycled up to 10 times without any reduction in yields and purity.[39]

During the development of this protocol, we used two colorimetric tests to verify the progress of the reactions. The free amine on cellulose was monitored using the "β-naphthol test".[40] This rather sensitive assay enables the detection of even small amounts of free aromatic amines on the cellulose, and thereby, a negative test indicates that the enaminone was probably present on the solid support. The presence on the resin (and consequently, the delivery from the resin) of enaminones **4** could be monitored also with an "iron III test" [41] that was able to do an intensely colored complex (rust-brown) with enolizable ketones. Finally, chemical identity was established by ^{1}H NMR and HPLC/UV/MS measurements, and a selection of these samples were further corroborated by comparing their ^{1}H NMR data with the spectra obtained from conventional solution-phase experiments.

In summary, we have described the versatility of the use of cellulose for preparing libraries of substituted pyrazoles and isoxazoles under very mild conditions and through a microwave strategy. Thermal drawbacks of solid-supported chemistry, such as degradation of the polymer support caused by heating for long times, are so avoided, and at least for the compounds we have prepared, cellulose appears to be more convenient than the most common synthetic resins. Furthermore, the low cost of the cellulose beads may permit carrying out a solid-phase synthesis on amacro scale, too.

Experimental Section

General Procedure for Conventional and Microwave-Assisted Pyrazole and Isoxazole Synthesis. 1. Conventional Thermal Protocol. Cellulose-Bound Enaminone. To a suspension of Perloza VT-100 (1 g, 0.5 mmol) swollen in DMF was added a solution of *N*-formylimidazole dimethylacetal (0.43 mg, 3 mmol), benzyl acetoacetate (0.58 g, 0.52 mL, 3 mmol), and camphosulfonic acid (43.0 mg, 10% w/w) in DMF (15 mL). The resulting mixture was heated to 80 °C for 48 h (until negative β-naphthol colorimetric test). After cooling, the cellulose sample was collected by filtration using a sintered glass funnel. The cellulose was thoroughly washed with alternating portions of DMF (3 × 10 mL), MeOH (3 × 10 mL), and Et_2O (3 × 10 mL). The cellulose was dried under reduced pressure. An FTIR spectrum of this modified cellulose sample showed a strong carbonyl absorption bands at 1690, 1640, and 1573 cm^{-1}, indicating successful solid support capture of the β-keto ester.[42]

Cyclization. The cellulose sample from the above experiment (**4a**) was suspended in dry ^{i}PrOH (5 mL) in a round-bottom flask with a reflux condenser. To the supported β-enaminone, a dry 2-propanol solution (10 mL) of phenylhydrazine hydrochloride (0.43 g, 3 mmol) or hydroxylamine hydrochloride (0.21 g, 3 mmol) and NEt_3 (0.34 g, 0.46 mL, 3.3 mmol) was added. The mixture was heated at reflux until disappearance of carbonyl function on the support bound material (negative to an $FeCl_3$ colorimetric test). After cooling to room temperature, the cellulose was then collected using a Buchner funnel and successively washed with ^{i}PrOH (3 × 10 mL). Before storing, the bead cellulose was washed with MeOH (3 × 3 mL) and dry Et_2O (3 × 5 mL) and dried in a vacuum. All of the alcoholic layers were combined and concentrated in vacuo, and the residue was dissolved in CH_2Cl_2 (20 mL). Methylene chloride solution was sequentially washed with 5% HCl aqueous solution, saturated Na_2CO_3 aqueous solution, and brine and dried over anhydrous Na_2SO_4. The solvent was evaporated to dryness under reduced pressure to give the pure title pyrazole **5a** (0.14 g, 99% yield) or isoxazole **6a** (0.11 g, 99% yield).

2. Microwave Irradiation Procedure. Cellulose-Bound Enaminone. To a suspension of Perloza VT-100 (1.0 g, 0.5 mmol) swollen in DMF was added a solution of *N*-formylimidazole dimethylacetal (0.43 g, 3 mmol), benzyl acetoacetate (0.58 g, 0.52 mL, 3 mmol), and camphosulfonic acid (43.0 mg, 10% w/w) in DMF (15 mL). The open flask was irradiated at 80 °C (by modulation of the power) for 15 min in a self-tuning single mode CEM Discover Focused Synthesizer. The mixture was cooled rapidly to room temperature by passing compressed air through the microwave cavity for 3 min. The reaction progress was monitored by a β-naphthol colorimetric test. After cooling to room temperature, the cellulose sample was collected by filtration using a sintered glass funnel. The cellulose was thoroughly washed with alternating portions of DMF (3 × 10 mL), MeOH (3 × 10 mL), and Et_2O (3 × 10 mL). The cellulose sample was dried under reduced pressure. An FTIR spectrum (KBr pellet) of this modified cellulose sample showed strong carbonyl absorption bands at 1690, 1645, and 1573 cm^{-1}, indicating successful solid support capture of the β-keto ester.

Cyclization. The cellulose sample from the above experiment (**4a**) was suspended in dry ^{i}PrOH (5 mL) in a round-

bottomed flask with a reflux condenser. To the supported β-enaminone, a dry 2-propanol solution (10 mL) of phenylhydrazine hydrochloride (0.43 g, 3 mmol) or hydroxylamine hydrochloride (0.21 g, 3 mmol) and NEt_3 (0.34 g, 0.46 mL, 3.3 mmol) was added. Then the suspension was placed into an open flask, and the reaction mixture was exposed to microwave irradiation as above for 15 min at 82 °C and monitored for the disappearance of carbonyl function on the support-bound material (negative $FeCl_3$ colorimetric test). After cooling to room temperature, the cellulose was then collected using a Buchner funnel and successively washed several times with ^{i}PrOH (3 × 10 mL). Before storing, bead cellulose was washed with MeOH (3 × 3 mL) and dry Et_2O (3 × 5 mL) and dried in a vacuum. All of the alcoholic layers were combined and concentrated in a vacuum, and the residue was dissolved in CH_2Cl_2 (20 mL). Methylene chloride solution was sequentially washed with 5% HCl aqueous solution, saturated Na_2CO_3 aqueous solution, and brine and dried over anhydrous Na_2SO_4. The solvent was evaporated to dryness under reduced pressure to give the pure target pyrazole **5a** (0.14 g, 99% yield) or isoxazole **6a** (0.11 g, 99% yield).

5-Methyl-1-phenyl-1-*H*-pyrazole-4-carboxylic Acid Benzyl Ester 5a. Pale yellow oil, 99% yield. ^{1}H NMR δ (ppm): 2.57 (s, 3H), 5.32 (s, 2H), 7.31−7.52 (m, 10H), 8.07 (s, 1H). ^{13}C NMR δ (ppm): 11.9, 65.6, 112.5, 119.1, 125.4, 128.0, 128.1, 128.5, 128.6, 129.2, 136.2, 141.9, 143.7, 163.5. MS (ESI + ve ion): 293.6 $(M + H)^+$. HPLC purity 99%. Anal. Calcd for $C_{18}H_{16}N_2O_2$ (292.12): C, 73.95; H, 5.52; N, 9.58. Found: C, 73.98; H, 5.49; N, 9.57.

5-Methylisoxazole-4-carboxylic acid benzyl ester 6a. Pale yellow oil (99% yield). ^{1}H NMR δ (ppm): 2.35 (s, 3H), 5.30 (s, 2H), 7.36−7.39 (m, 5H), 7.73 (s, 1H). ^{13}C NMR δ (ppm): 10.8, 71.7, 100.9, 128.2, 128.7, 141.3, 150.1, 158.1, 169.8. HPLC analysis: 99%. MS (ESI + ve ion): 218.3 $(M + H)^+$. Anal. Calcd for $C_{12}H_{11}NO_3$ (217.22): C, 66.35; H, 5.10; N, 6.45. Found: C, 66.31; H, 5.04; N, 6.42.

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Supporting Information Available. Synthetic procedures and characterization of new compounds (PDF). This material is available free of charge via Internet at http://pubs.acs.org/.

References and Notes

(1) For an excellent book and reviews, see: (a) Nicolau, K. C. *Handbook of Combinatorial Chemistry*; Wiley-VCH: Weinheim, 2002. (b) Zaragozza Dorwald, F. *Solid-Phase Synthesis*; Wiley-VCH: Weinheim, 2000. (c) Jung, G. *Combinatorial Chemistry*; Wiley-VCH: Weinheim, 1999. (d) DeWitt, S. H.; Czarnik, A. W. *A Pratical Guide to Combinatorial Chemistry*; American Chemistry Society: Washington, 1997. (d) Hudson, D. *J. Comb. Chem.* **1999**, *1*, 333−360, 403−457. (e) Dolle, R. E. *J. Comb. Chem.* **2001**, *3*, 477−517. (f) Dolle, R. E. *J. Comb. Chem.* **2002**, *4*, 370−418 and references therein.

(2) (a) Ellman, J. A. *Acc. Chem. Res.* **1996**, *29*, 132. (b) Fruchtel, J. S.; Jung, G. *Angew. Chem., Int. Ed. Engl.* **1996**, *35*, 17. (c) Gerritz, S. W.; Trump, R. P.; Zuercher, W. J *J. Am. Chem. Soc.* **2000**, *122*, 6357. (d) Li, W.; Yan, B. *J. Org. Chem.* **1998**, *63*, 4092. (e) Franzen, R. G. *J. Comb. Chem.* **2000**, *2*, 195.

(3) (a) Santini, R.; Griffith, M. C.; Qi, M. *Tetrahedron Lett.* **1998**, *39*, 8951. (b) Wilson, M. E.; Paech, K.; Zhou, W. J.; Kurth, M. J. *J. Org. Chem.* **1998**, *63*, 5094.

(4) (a) Gutte, B.; Merrifield, R. B. *J. Biol. Chem.* **1971**, *246*, 1922. (b) Crowley, J.; Rapoport, H. *Acc. Chem. Res.* **1976**, *9*, 135. (c) Bayer, E.; Rapp, W. *Poly(Ethylene Glycol)-Chemistry: Biotechnical and Biomedical Applications*; Harris, J. M., Ed.; Plenum Press: New York, 1992; Chapter 20. (d) Svec, F.; Frechet, J. M. J. *Science* **1996**, *273*, 205. (e) Hodge, P. *Chem. Soc. Rev.* **1997**, *26*, 417. (f) Sherrington, D. C. *Chem. Commun.* **1998**, 2275. (g) Ley, S. V.; Baxendale, I. R.; Bream, R. M.; Jackson, P. S.; Leach, A. G.; Longbottom, D. A.; Nesi, M.; Scott, J. S.; Storer, R. I.; Taylor, S. J. *J. Chem. Soc., Perkin Trans. 1* **2000**, *23*, 3815−4195.

(5) (a) Lam, K. S.; Lebl, M.; Krchnak, V. *Chem. Rev.* **1997**, *97*, 411−448. (b) Jung, G. *Combinatorial Chemistry*; Wiley-VCH: Weimheim, **1999**; Chapter 1, pp 15−16 and references therein (17, 18, 95−97). (c) Smith, H. K.; Bradley, M. *J. Comb. Chem.* **1999**, *1*, 326−332. (d) St. Hilaire, P. M.; Willert, M.; Juliano, M.; Juliano, L.; Meldal, M. *J. Comb. Chem.* **1999**, *1*, 509−523. (e) Rosse, G.; Kueng, E.; Page, M. G. P.; Schauer-Vukasinovic, V.; Giller, T.; Lahm, H.-W.; Hunziker, P.; Schlatter, D. *J. Comb. Chem.* **2000**, *2*, 461−466. Liu, R.; Marik, J.; Lam, K. S. *J. Am. Chem. Soc.* **2002**, *124*, 7678−7680. (f) Pastor, J. J.; Fernandez, I.; Rabanal, F.; Giralt, E. *Org. Lett.* **2002**, *4*, 3831−3833. (g) St. Hilaire, P. M.; Alves, L. C.; Herrera, F.; Renil, M.; Sanderson, S. J.; Mottram, J. C.; Coombs, G. H.; Juliano, M. A.; Juliano, L.; Arevalo, J.; Meldal, M. *J. Med. Chem.* **2002**, *45*, 1971−1982.

(6) (a) Atherton, D.; Clive, D. L. J.; Sheppard, R. C. *J. Am. Chem. Soc.* **1975**, *97*, 6584−6585. (b) Bayer, E. *Angew. Chem., Int. Ed. Engl.* **1991**, *30*, 113−129. (b) Meldal, M. *Tetrahedron Lett.* **1992**, *33*, 3077−3088. (c) Kempe, M.; Barany, G. *J. Am. Chem. Soc.* **1996**, *118*, 7083−7093.

(7) (a) Arshady, R. *J. Chromatogr.* **1991**, *586*, 181−197, 199. (b) Mendoca, A. J.; Xiao, X. Y. *Med. Res. Rev.* **1999**, *19*, 451. (d) Toy, P. H.; Janda, K. D. *Tetrahedron Lett.* **1999**, *40*, 6329−6332. (e) Garibay, P.; Toy, P. H.; Hoeg-Jensen, T.; Janda, K. D. *Synlett* **1999**, *9*, 1438−1440. (f) Vaino, A. R.; Goodin, D. B.; Janda, K. D. *J. Comb. Chem.* **2000**, *2*, 330−336. (g) Reger, T. S.; Janda, K. D. *J. Am. Chem. Soc.* **2000**, *122*, 6929−6934. (h) Toy, P. H.; Reger, T. S.; Garibay, P.; Garno, J. C.; Liu, G.-Y.; Janda, K. D. *J. Comb. Chem.* **2001**, *3*, 117−124. (i) Brummer, O.; Clapham, B.; Janda, K. D. *Tetrahedron Lett.* **2001**, *42*, 2257. (l) Moss, J.; Dickerson, T. J.; Janda, K. D. *Tetrahedron Lett.* **2002**, *43*, 37−40. JandaJel resins are commercially available through the Aldrich Chemical Co.

(8) (a) Atherton, E.; Clive, D. L. J.; Sheppard, R. C. *J. Am. Chem. Soc.* **1975**, *97*, 6584−6585. (b) Arshady, R.; Atherton, E.; Clive, D. L. J.; Sheppard, R. C. *J. Chem. Soc., Perkin Trans. 1* **1981**, 529−537.

(9) Berg, R. H.; Almdal, K.; Batsberg Pedersen, W.; Holm, A.; Tam, J. P.; Merrifield, R. B. *J. Am. Chem. Soc.* **1989**, *111*, 8024−8026.

(10) (a) Eichler, J.; Bienert, M.; Stierandova, A.; Lebl, M. *Peptide Res.* **1991**, *4*, 296−307. (b) Englebretsen, D. R.; Harding, D. R. K. *Int. J. Pept. Protein Res.* **1994**, *43*, 546−554.

(11) Buttner, K.; Zahn, H.; Fischer, W. H. In *Peptides: Chemistry and Biology*, Proceedings of the Tenth American Peptide Symposium; Marshall, G. R., Ed.; Escom Science Publishers: Leiden, The Netherlands, 1988; pp 210−211.

(12) (a) Rapp, W.; Zhang, L.; Bayer, E. In *Solid-Phase Synthesis*, Epton, R., Ed; SPCC (UK) Ltd.: Birmighan, U.K., 1990; p 205. (b) Medal, M. *Tetrahedron Lett.* **1992**, *33*, 3077−3080.

(13) The PEG-grafted resins, TentaGel S, and PEG-*cross*-linked resins, such as PEGA, POE-POP, *J*anda*J*el, CLEAR, and SPOCC, are aqueous compatible but they are too expensive, sometime not commercially available and with low loading values. (a) Miranda, L. P.; Lubell, W. D.; Halkes, K. M.; Groth, T.; Grøtli, M.; Rademann, J.; Gotfredsen, C. H.; Meldal, M. *J. Comb. Chem.* **2002**, *4*, 523−529.

(14) Chesney, A.; Barwell, P.; Stonehouse, D. F.; Steel, P. G. *Green Chem.* **2000**, *2*, 57−62.

(15) (a) Frank, R.; Heikens, W.; Heisterberg-Moutsis, G.; Blocker, H. *Nucleic Acids Res.* **1983**, *11*, 4365−4377. (b) Frank, R.; Doring, R. *Tetrahedron* **1988**, *44*, 6031.

(16) (a) Volkmer-Engert, R.; Hoffmann, B.; Schneider-Mergener, J. *Tetrahedron Lett.* **1997**, *38*, 1029−1032. (b) Heine, N.; Germeroth, L.; Schneider-Mergener, J.; Wenschuh, H. *Tetrahedron Lett.* **2001**, 42227−230.

(17) Frank, R. *Tetrahedron* **1992**, *48*, 9217−9232.

(18) Sepharose beads have been used in the solid-phase synthesis of peptides. See: Tegge, W.; Frank, R. *J. Pept. Res.* **1997**, *49*, 355−362.

(19) Chesney, A.; Steel, P.; Stonehouse, P. G. *J. Comb. Chem.* **2000**, *2*, 434−437.

(20) Aniline-functionalized cellulose was obtained from Iontosorb, Czech Republic (Usti nad Labem). The content of amino aryl groups in Iontosorb AV can be regulated according to the customer's demands in the range 0.1−2.8 mmol/g.

(21) (a) Shim, J.-Y.; Welsh, W. J.; Cartier, E.; Edwards, J. L.; Howlett, A. C. *J. Med. Chem.* **2002**, *45*, 1447−1458. (b) Pinto, D. J. P.; Orwat, M. J.; Wang, S.; Fevig, J. M.; Quan, M. L.; Amparo, E.; Cacciola, J.; Rossi, K. A.; Alexander, R. S.; Smallwood, A. M.; Lettgen, J. M.; Liang, L.; Aungst, B. J.; Wright, M. R.; Knabb, R. M. *J. Med. Chem.* **2001**, *44*, 566−578. (c) Selwood, D. L.; Brummel, D. G.; Budworth, J.; Burtin, G. E.; Campbell, R. O.; Chana, S. S.; Charles, I. G.; Fernandez, P. A.; Glen, R. C.; Goggin, M. C.; Hobbs, A. J.; Kling, M. R.; Liu, Q.; Madge, D. J. *J. Med. Chem.* **2001**, *44*, 78−99. (d) Stauffer, S. R.; Coletta, C. J.; Tedesco, R.; Nishiguchi, G.; Carlson, K.; Sun, J.; Katzenellenbogen, B. S.; Katzenellenbogen, J. A. *J. Med. Chem.* **2000**, *43*, 4934−4947. (e) Kikuchi, K.; Hibi, S.; Yoshimura, H.; Tai, K.; Hida, T.; Tokuhara, N.; Yamauchi, T.; Nagai, M. *Bioorg. Med. Chem. Lett.* **2000**, *10*, 619−622. (f) Baraldi, P. G.; Cozzi, P.; Geroni, C.; Mongelli, N.; Romagnoli, R.; Spalluto, G. *Bioorg. Med. Chem. Lett.* **1999**, *7*, 251−262.

(22) De Luca, L.; Giacomelli, G.; Riu, A. *J. Org. Chem.* **2001**, *66*, 6823.

(23) (a) Shen, D.-M.; Shu, M.; Chapman, K. T. *Org. Lett.* **2000**, *2*, 2789−2792. (b) Stauffer, S. R.; Katzenellenbogen, J. A. *J. Comb. Chem.* **2000**, *2*, 318−329. (c) Groshe, P.; Höltzel, A.; Walk, T. B.; Trautwein, A. W.; Jung, G. *Synthesis* **1999**, 1961−1978. (d) Spivey, A. C.; Diaper, C. M.; Adams, H.; Rudge, A. J. *J. Org. Chem.* **2000**, *65*, 5253−5263. (d) Marzinzik, A. L.; Felder, E. R. *J. Org. Chem.* **1998**, *63*, 723−727.

(24) (a) Donohue, B. A.; Michelotti, E. L.; Reader, J. C.; Reader, V.; Stirling, M.; Tice, C. M. *J. Comb. Chem.* **2002**, *4*, 23−32. (b) Stauffer, S. R.; Huang, Y.; Coletta, C. J.; Tedesco, R.; Katzenellenbogen, J. A. *Bioorg. Med. Chem.* **2001**, *9*, 141−150. (c) Franzen, R. G. J. *J. Comb. Chem.* **2000**, *2*, 195−214. (d) Molteni, V.; Hamilton, M. H.; Mao, L.; Crane, C. M.; Termin, A. P.; Wilson, D. M. *Synthesis* **2002**, 1669−1674.

(25) Menozzi, G.; Mosti, L.; Schenone, P. J. *Heterocyclic Chem.* **1987**, *24*, 1669−1674.

(26) Guzman-Perez, A.; Wester, R. T.; Allen, M. C.; Brown, J. A.; Buchholz, A. R.; Cook, E. R.; Day, W. W.; Hamanaka, E. S.; Kennedy, S. P.; Knight, D. V.; Kowalczyk, P. J.; Marala, R. B.; Mularski, C. J.; Novomisle, W. A.; Ruggeri, R. B.; Tracey, W. R.; Hill, R. J. *Bioorg. Med. Chem. Lett.* **2001**, *11*, 803−807.

(27) Its preparation is straightforward from imidazole and trimethylorthoformate. See: Brown, R. S. *J. Org. Chem.* **1980**, *45*, 4038.

(28) Bienaymé, H. *Tetrahedron Lett.* **1998**, *39*, 4255−4258.

(29) The reaction progress was followed by a colorimetric test, by FT-IR spectroscopy (formation of the enaminone signals), and by nitrogen elemental analysis (through a comparison with starting cellulose). For colorimetric assay, see refs 40 and 41.

(30) Surprisingly, for analogous reactions in solution phase, both primary amine and aniline was found unreactive (ref 28).

(31) (a) Oikawa, Y.; Yoshioka, T.; Sugano, K.; Yonemitsu, O. *Org. Synth. Coll. VII* **1990**, 359−361. (b) Raillard, S. P.; Chen, W.; Sullivan, E.; Bajjalieh, W.; Bhandari, A.; Baer, T.; A. *J. Comb. Chem.* **2002**, *4*, 470−474 and references therein.

(32) (a) Galema, S. A. *Chem. Soc. Rev.* **1997**, *26*, 233−238. (b) Caddick, S. *Tetrahedron* **1995**, *51*, 10403−10432. (c) Strauss, C. R.; Trainor, R. W. *Aust. J. Chem.* **1995**, *48*, 1665−1692. (d) De la Hoz, A.; Diaz-Ortis, A.; Moreno, A.; Langa, F. *Eur. J. Org. Chem.* **2000**, 3659−3673. Gedye, R. N.; Wei, J. B. *Can. J. Chem.* **1998**, *76*, 525−532. (e) Gedye, R. N.; Wei, J. B. *Can. J. Chem.* **1998**, *76*, 525−532. (f) Larhed, M.; Hallberg, A. *Drug Discovery Today* **2001**, *6*, 406−416.

(33) (a) Kuhnert, N. *Angew. Chem., Int. Ed.* **2002**, *41*, 11, 1863−1866. (b) Strauss, C. R. *Angew. Chem., Int. Ed.* **2002**, *41*, 19, 3589−3590.

(34) For reviews on microwave-assisted combinatorial chemistry, see, for example, the following: (a) Lew, A.; Krutzik, P. O.; Hart, M. E.; Chamberlin, A. R. *J. Comb. Chem.* **2002**, *4*, 95−105. (b) Kappe, C. O. *Curr. Opin. Chem. Biol.* **2002**, *6*, 314−320.

(35) (a) Yu, H.-M.; Chen, S.-T.; Wang, K.-T. *J. Org. Chem.* **1992**, *57*, 4781−4784. (b) Larhed, M.; Lindeberg, G.; Hallberg, A. *Tetrahedron Lett.* **1996**, *37*, 8219−8222. (c) Hoel, A. M. L.; Nielsen, J. *Tetrahedron Lett.* **1999**, *40*, 3941−3944. (d) Yu, A.-M.; Zhang, Z.-P.; Yang, H.-Z.; Thang, C.-Y.; Liu, Z. *Synth. Commun.* **1999**, *29*, 1595−1599. (e) Chandrasekhar, S.; Padmaja, M. B.; Raza, A. *Synlett* **1999**, 1597−1599. (f) Combs, A. P.; Saubern, S.; Rafalski, M.; Lam, P. Y. S. *Tetrahedron Lett.* **1999**, *40*, 1623−1626. (g) Kuster, G.; Scheeren, H. W. *Tetrahedron Lett.* **2000**, *41*, 515−519. (h) Alterman, M.; Hallberg, A. *J. Org. Chem.* **2000**, *65*, 7984−7989. (i) Scharn, D.; Wenschuh, H.; Reineke, U.; Schneider-Mergener, J.; Germeroth, L. *J. Comb. Chem.* **2000**, *2*, 361−369. (l) Stadler, A.; Kappe, C. O. *Tetrahedron* **2001**, *57*, 3915−3920. (n) Glass, B. M.; Combs, A. P. *High-Throughput Synthesis. Principles and Practices*; Sucholeiki, I., Ed.; Marcel Dekker: New York, 2001; pp 123−128. (o) Stadler, A.; Kappe, C. O. *Eur. J. Org. Chem.* **2001**, 919−925. (p) Strohmeier, G. A.; Kappe, C. O. *J. Comb. Chem.* **2002**, *4*, 154−161.

(36) The MW experiments were performed in a self-tuning single mode CEM Discover Focused Synthesiser apparatus. The instrument continuously adjusted the applied wattage to maintain the desired temperature. Reactions were conducted in appropriate 25-mL open vessels. The experiments were performed using a flask equipped with a reflux condenser mounted outside the apparatus.

(37) In a closed-vessel system, the process is significantly less effective.

(38) The volatile reaction product (H_2O) is removed rapidly from the reaction mixture and, therefore, from the equilibrium.

(39) The synthetic strategies herein developed on cellulose beads can be in principle applied to other amine-functionalized solid supports, such as polystyrene (PS), *J*anda*J*el (JJ), TentaJel (TJ), and PEGA. However, only *N*-methyl aminomethylated resin is able to perform the synthesis of supported enaminone. The subsequent cyclocondensation reaction with hydroxylamine or hydrazine has to be performed in ^{i}PrOH/THF (2/1) for increasing the swelling of PS resin. Pyrazoles **5a**−**c** and isoxazoles **6a**−**c** prepared with PS support were recovered after aqueous workup in ∼70−75% yield (purity >94% as judged by HPLC analysis). The reaction times were comparable both under conventional heating and under microwave irradiation. At last, using recycled PS support, the purity of final compound decreased to 80% (65% for PS subjected at conventional heating), so that in the second cycle, it was necessary to purify the heterocycle by chromatography or preparative HPLC. Moreover, no increase in purity was observed using a microwave irradiation.

(40) The aniline cellulose support was first treated with an acid aqueous solution of $NaNO_2$ rather than with a basic aqueous solution of β-naphthol. The cellulose bead immediately changed color from white to red in the presence of free aniline amine.

(41) Iron(III) was able to form an intensely colored complex (rust-brown) with enolizable ketones. Therefore, this old colorimetric assay is suitable and sensitive to detect the presence (absence) of β-dicarbonyl compounds on solid support.

(42) See experimental part in ref 25.

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