Microdroplets

International Edition: DOI: 10.1002/anie.201913069
German Edition: DOI: 10.1002/ange.201913069

Chemoselective N-Alkylation of Indoles in Aqueous Microdroplets

*Elumalai Gnanamani, Xin Yan, and Richard N. Zare**

Abstract: *Many reactions show much faster kinetics in microdroplets than in the bulk phase. Most reported reactions in microdroplets mirror the products found in bulk reactions. However, the unique environment of microdroplets allows different chemistry to occur. In this work, we present the first chemoselective N-alkylation of indoles in aqueous microdroplets via a three-component Mannich-type reaction without using any catalyst. In sharp contrast, bulk reactions using the same reagents with a catalyst yield exclusively C-alkylation products. The N-alkylation yield is moderate in microdroplets, up to 53 %. We extended the scope of the microdroplet reaction and obtained a series of new functionalized indole aminals, which are likely to have biological activities. This work clearly indicates that microdroplet reactions can show reactivity quite different from that of bulk-phase reactions, which holds great potential for developing novel reactivities in microdroplets.*

Recent findings have shown that microdroplets provide unique reaction environments that can be used to dramatically enhance reaction rates.[1] The acceleration factors of microdroplet reactions can be many orders of magnitude compared to the corresponding reactions in bulk.[2] Reaction rate accelerations in microdroplets have been demonstrated for carbon–carbon bond formation,[3] carbon–nitrogen bond formation,[4] carbon–oxygen bond formation,[5] deprotection of N-Boc,[6] demetalation,[7] and oxidation–reduction.[8] Microdroplet synthesis has been scaled up to a production rate of about 1–30 mg min^{-1}, which makes it preparative.[3a,8b,9] This tempting feature of microdroplet reactions also stimulates its application in many fields, such as high-throughput reaction screening,[10] preparation of gold nanostructures,[11] and accelerated degradation of pharmaceuticals.[12]

It has become apparent that the environment in microdroplets is strikingly different from that of the corresponding bulk phase.[1,13] Many features of microdroplets may contribute to reaction acceleration, such as the confinement of reagents in small-volume reactors,[13a] the large surface-to-volume ratios of small reactors, the higher density of molecules on the surface of the microdroplets,[3c,13a] solvent evaporation with associated increases in reagent concentrations,[2a,3b] and extreme pH values.[2a,3b,14] Thus far, most of the reported accelerated reactions in microdroplets mirror the products found in the bulk reaction.[1] Exceptions, however, are emerging, such as the phosphorylation of sugars,[13b] the production of gold nanostructures without the addition of a reducing agent,[11] the spontaneous reduction of organic molecules, and the spontaneous generation of hydrogen peroxide in aqueous microdroplets.[13c,d] In another example, our research group previously reported that the Diels–Alder reaction of 3,5-hexadienyl acrylate ester could not occur in microdroplets, with mainly unreacted substrate and a small fraction of hydrolyzed substrate being present under different microdroplet reaction conditions.[2b] In contrast, the desired Diels–Alder product can be easily obtained in aqueous media at high temperature using indium(III) triflate as a catalyst in the bulk phase. In other words, microdroplets inhibit the Diels–Alder reaction.

Herein, we report the first chemoselective N-alkylation of indoles in aqueous microdroplets. Alkylated indoles were obtained by a three-component Mannich-type reaction both in microdroplets and in bulk phase. The conventional bulk reaction produced the C-alkylation product via a traditional Mannich-type reaction between an aldehyde, an amine, and an indole,[20,21] whereas a microdroplet reaction between the same three starting materials produced a new compound resulting from N-alkylation of the indole (Figure 1).

Nitrogen-containing molecules play a major role in the pharmaceutical, food, and agricultural industries. In particular, indole and its derivatives are important molecules in several natural products, and some have biological activities (Figure 2).[15] For example, delvavirdine (**A**) is a drug used for the treatment of HIV type 1,[16] and yohimbine (**B**) has potential for the treatment of sexual dysfunction as well as type-2 diabetes in both animal and human models.[17] Indole-containing compound **C** has potent anticancer properties against cell lines resistant to paclitaxel,[18] and 5HT$_{2C}$ agonist

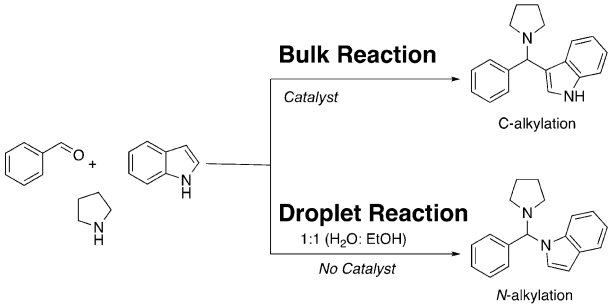


Figure 1. Chemoselective synthesis of alkylated indoles in a three-component Mannich-type reaction. The conventional bulk reaction forms the C-alkylation product whereas the microdroplet reaction produces the N-alkylation product.

[*] E. Gnanamani, X. Yan, R. N. Zare
Department of Chemistry, Stanford University
333 Campus Drive, Stanford, CA 94305-5080 (USA)
E-mail: zare@stanford.edu
E. Gnanamani, R. N. Zare
Department of Chemistry, Fudan University
Shanghai 200438 (China)
X. Yan
Department of Chemistry, Texas A&M University
580 Ross Street, College Station, TX 77843-3255 (USA)

Supporting information and the ORCID identification number(s) for the author(s) of this article can be found under: https://doi.org/10.1002/anie.201913069.

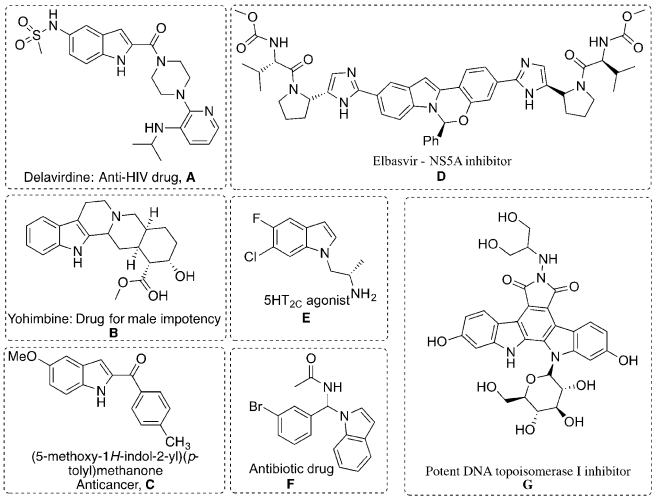


Figure 2. Representative examples of bioactive indole derivatives.

E has potential therapeutic utility for the treatment of obsessive compulsive disorder.[19]

Owing to the importance of indole derivatives (see above), we chose to investigate the synthesis of alkylated indoles via a three-component Mannich-type reaction. Kumar and co-workers[20] have developed a green, three-pot synthesis of indole C-alkylation products that is catalyzed by L-proline (Figure 3a). Other groups studied the same reaction using different catalytic systems, including silver nanoparticles, a Co xanthane complex, or SiO_2–iodine.[21] Typically, several hours of reaction time were required to obtain the products (Figure 3a).[20] Given that reactions are often accelerated in microdroplets, we anticipated that our droplet method would significantly reduce the reaction time. Therefore, we performed a three-component Mannich-type reaction without adding any catalyst to the microdroplets (Figure 3b).

As a preliminary reaction, one equivalent each of benzaldehyde and indole and 1.5 equivalents of pyrrolidine were mixed in a water–ethanol solvent (7:3 v/v) in a syringe and introduced through a fused silica capillary (i.d. 100 μm) at

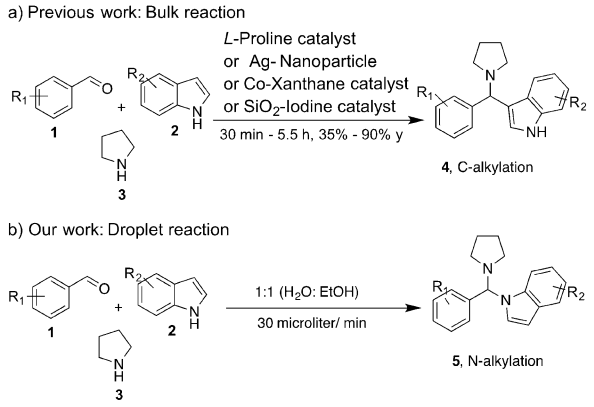


Figure 3. a) Previous work on the synthesis of C-alkylated indoles in the bulk phase using different catalytic systems, including L-proline, silver nanoparticles, a Co xanthane complex, or SiO_2–iodine. b) Our work on the three-component one-step synthesis of the N-alkylation products of indoles in microdroplets without any catalyst.

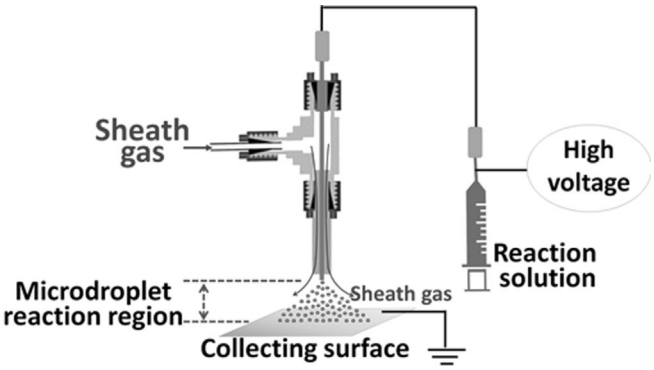


Figure 4. Experimental setup used in the microdroplet synthesis of indole aminals. The charged microdroplets were generated by applying a −10 kV voltage to the bulk solution with assisted nebulizing dry nitrogen gas at 80 psi (a schematic diagram of the collection process is provided in the Supporting Information).

a rate of 30 μL min^{-1} to the spray tip (Figure 4). A potential of −10 kV was applied to the solution to initiate the formation of charged microdroplets. A coaxial sheath gas (dry N_2 operated at 80 psi) flowing around the capillary resulted in better nebulization. We collected the products on a grounded surface for 15 min and subjected them for crude ^{1}H NMR analysis. Surprisingly, instead of the expected C-alkylation product, we observed moderate conversion (30%) into the N-alkylation product (Figure 3b). The molecule has not been reported before, and the structure of this new indole aminal was confirmed by high-resolution mass spectrometry and ^{13}C NMR, ^{1}H NMR, and IR spectroscopy (see the Supporting Information).

Looking at the literature for methods to synthesize the N-alkylated products, we found that the analogous morpholine aminal was synthesized in two steps by Love and Nguyen[22a] and that Joshi and co-workers[22b] had developed a method to synthesize indole aminals by utilizing urea and thiourea nucleophiles.[22] There were no reports on the one-step synthesis of indole aminals from simple amines. This result prompted us to search the literature for examples of biologically relevant N-alkylation products, of which there are several. Indole aminals are present both in elbasvir (**D**), which is a highly potent and selective NS5A inhibitor used for treating the hepatitis C virus, and in a potential antibiotic drug (**F**).[23] Indole hemiaminals, such as the one found in DNA topoisomerase I inhibitor (**G**), are also relevant (Figure 2).[24] Only a few methods are available for preparing these delicate motifs, which involve many steps. Our result encouraged us to optimize further this one-step method for synthesizing indole aminals.

To improve the conversion, the reaction was carried out with 1 equivalent of benzaldehyde, 1.05 equivalents of indole, and 1.5 equivalents of pyrrolidine in ethanol–water (1:1 v/v), which gave 85% conversion of the aldehyde. The better solubility of the starting materials likely caused this behavior. The reaction was scaled up using a higher droplet flux (dual spray source with a total flow rate of 60 μL min^{-1}) and higher concentration (0.066 M, 0.4 mmol) of aldehyde using nitrogen gas. After collecting the reaction product, crude ^{1}H NMR indicated that complete conversion of aldehyde had occurred.

After purification by silica gel chromatography, the N-alkylation product **5a** was obtained as the sole product in 47% yield with complete conversion of aldehyde. Electron-rich *p*-anisaldehyde gave the analogous N-alkylation product **5b** in 43% isolated yield. Similarly, 3-methyl- and 4-fluorobenzadehyde also gave the corresponding indole aminals **5c** and **5d** in 37% and 51% yield, respectively (Table 1). The scope of the reaction was further expanded to include other indole derivatives such as 3-methylindole (**2e**) and 5-methoxyindole (**2f**). The former gave the aminal (**5e**) in 35% yield while the latter bearing the electron-donating methoxy group gave N-alkylation product **5f** in 32% yield (Table 1, entry 6).

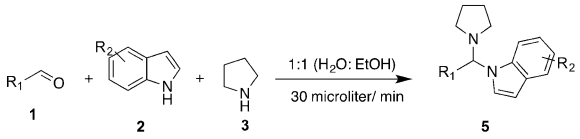
Additionally, the use of a heteroaryl aldehyde (2-thienyl) with indole was also well tolerated giving rise to N-alkylation product **5g** in good yield. The 2-thienylcarboxaldehyde also successfully reacted with 3-methylindole to afford the corresponding product **5h** in 39% yield. Having succeeded in the Mannich-type reaction using microdroplet chemistry with various aromatic aldehydes and nucleophiles, we then investigated the reaction with aliphatic aldehydes to test the generality of this method. Notably, butyraldehyde reacted with indole and 3-methylindole to afford the corresponding N-alkylation products **5i** and **5j** in 48% and 53% yield, respectively. When we attempted to extend this method to acetaldehyde as the electrophilic partner, the reaction failed to give the addition product. This may be caused by the low boiling point of the aldehyde. Similarly, utilizing sterically crowded 2,3-dimethylindole as the nucleophile failed to form the addition product because of its increased steric demand.

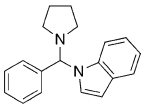
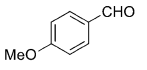
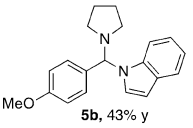
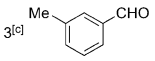
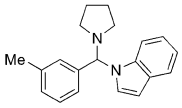
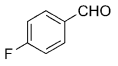
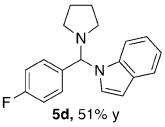
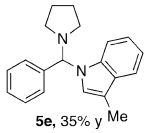
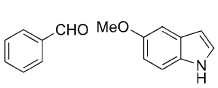
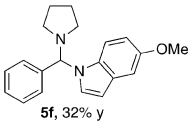
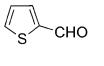
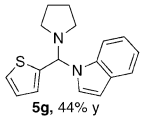
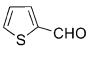
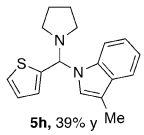
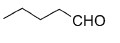
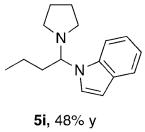
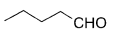
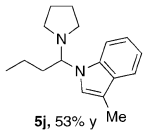
In conclusion, we have described the first example of a one-step chemoselective N-alkylation of indoles. This was accomplished by a three-component reaction in negatively charged aqueous microdroplets without using any catalyst. Instead of the C-alkylation products that were obtained from bulk reactions, N-alkylation products were synthesized under aqueous microdroplet conditions. Functionalized indoles and benzaldehydes were added to the microdroplet reagents, and their corresponding N-alkylation products were also successfully obtained.

At present, yields are only moderate. However, recent work has shown that it is possible to scale up the product amount for some reactions.[25] It remains to be demonstrated whether the yield can be increased by recycling the droplet spray, but this is a topic for future work.

In this work, we have provided a new method for synthesizing indole aminals. All of the structures of the new molecules were confirmed. The fact that we observed N-alkylation rather than C-alkylation demonstrates that strikingly different reactivity can occur in microdroplets compared to that in bulk solution. Based on our previous experience in microdroplet chemistry,[13c,d] water droplets can produce hydroxyl radicals and hydrogen peroxide. These reactive oxygen species may catalyze the reaction to selectively obtain the N-alkylated products. Support for this contention is provided by the work of Heaney and Ley,[26] who showed that indole could be deprotonated by hydroxide anion, although this process required the use of dimethyl sulfoxide as a solvent. It is expected that the concentration of the hydroxide anion is enhanced on the periphery of the aqueous microdroplet.[27] The product that we observe might

Table 1: Scope of the microdroplet reaction with various indoles and aldehydes.[a,b]



Entry	Aldehyde **1**	**2**	Product
1			 **5a,** 47% y
2			 **5b,** 43% y
3[c]			 **5c,** 37% y
4			 **5d,** 51% y
5			 **5e,** 35% y
6			 **5f,** 32% y
7			 **5g,** 44% y
8			 **5h,** 39% y
9			 **5i,** 48% y
10			 **5j,** 53% y

[a] All reactions were performed using the microdroplet method with 0.4 mmol of aldehyde, 0.42 mmol of indole, and 0.6 mmol of pyrrolidine in water–ethanol solvent (1:1 v/v). [b] Yields of isolated products. [c] < 10% of the aldehyde was recovered.

then result from the reaction of the indole N-anion with the iminium ion derived from the aldehyde–pyrrolidine condensation.[28] Further detailed mechanistic investigations to understand the chemoselective formation of the N-alkylation product and work toward larger-scale reactions are currently under way.

Acknowledgements

We thank Prof. Hao Chen for valuable suggestions and Dr. Vijaya Lakshmi Kanchustambham for help with recording mass spectra. This work was supported by the Scientific Research Startup Foundation (Grant IDH1615113) of Fudan University and the United States Air Force Office of Scientific Research through a Basic Research Initiative grant (AFOSR FA9550-16-1-0113).

Conflict of interest

The authors declare no conflict of interest.

Keywords: chemoselectivity · indoles · liquid microdroplets · N-alkylation · reaction acceleration

[1] X. Yan, R. M. Bain, R. G. Cooks, *Angew. Chem. Int. Ed.* **2016**, *55*, 12960 – 12972; *Angew. Chem.* **2016**, *128*, 13152 – 13166.
[2] a) S. Banerjee, R. N. Zare, *Angew. Chem. Int. Ed.* **2015**, *54*, 14795 – 14799; *Angew. Chem.* **2015**, *127*, 15008 – 15012; b) S. Banerjee, E. Gnanamani, X. Yan, R. N. Zare, *Analyst* **2017**, *142*, 1399 – 1402.
[3] a) T. Müller, A. Badu-Tawiah, R. G. Cooks, *Angew. Chem. Int. Ed.* **2012**, *51*, 11832 – 11835; *Angew. Chem.* **2012**, *124*, 12002 – 12005; b) M. Girod, E. Moyano, D. I. Campbell, R. G. Cooks, *Chem. Sci.* **2011**, *2*, 501 – 510; c) Y. Li, X. Yan, R. G. Cooks, *Angew. Chem. Int. Ed.* **2016**, *55*, 3433 – 3437; *Angew. Chem.* **2016**, *128*, 3494 – 3498.
[4] a) Y.-H. Lai, S. Sathyamoorthi, R. M. Bain, R. N. Zare, *J. Am. Soc. Mass Spectrom.* **2018**, *29*, 1036 – 1043; b) A. K. Badu-Tawiah, D. I. Campbell, R. G. Cooks, *J. Am. Soc. Mass Spectrom.* **2012**, *23*, 1461 – 1468; c) X. Yan, R. Augusti, X. Li, R. G. Cooks, *ChemPlusChem* **2013**, *78*, 1142 – 1148; d) R. M. Bain, C. J. Pulliam, R. G. Cooks, *Chem. Sci.* **2015**, *6*, 397 – 401.
[5] R. M. Bain, C. J. Pulliam, S. A. Raab, R. G. Cooks, *J. Chem. Educ.* **2016**, *93*, 340 – 344.
[6] P. W. Fedick, R. M. Bain, K. Bain, T. F. Mehari, R. G. Cooks, *Int. J. Mass Spectrom.* **2018**, *430*, 98 – 103.
[7] J. K. Lee, H. G. Nam, R. N. Zare, *Q. Rev. Biophys.* **2017**, *50*, 1 – 7.
[8] a) X. Yan, H. Cheng, R. N. Zare, *Angew. Chem. Int. Ed.* **2017**, *56*, 3562 – 3565; *Angew. Chem.* **2017**, *129*, 3616 – 3619; b) X. Yan, Y.-H. Lai, R. N. Zare, *Chem. Sci.* **2018**, *9*, 5207 – 5211; c) J. K. Lee, S. Banerjee, H. G. Nam, R. N. Zare, *Q. Rev. Biophys.* **2015**, *48*, 437 – 444.
[9] Z. Wei, X. Zhang, J. Wang, S. Zhang, X. Zhang, R. G. Cooks, *Chem. Sci.* **2018**, *9*, 7779 – 7786.
[10] H. S. Ewan, K. Iyer, S.-H. Hyun, M. Wleklinski, R. G. Cooks, D. H. Thompson, *Org. Process Res. Dev.* **2017**, *21*, 1566 – 1570.
[11] J. K. Lee, D. Samanta, H. G. Nam, R. N. Zare, *Nat. Commun.* **2018**, *9*, 1562.
[12] Y. Li, Y. Liu, H. Gao, R. Helmy, W. P. Wuelfing, C. J. Welch, R. G. Cooks, *Chem. Eur. J.* **2018**, *24*, 7349 – 7353.
[13] a) Z. Zhou, X. Yan, Y.-H. Lai, R. N. Zare, *J. Phys. Chem. Lett.* **2018**, *9*, 2928 – 2932; b) I. Nam, J. K. Lee, H. G. Nam, R. N. Zare, *Proc. Natl. Acad. Sci. USA* **2017**, *114*, 12396 – 12400; c) J. K. Lee, D. Samanta, H. G. Nam, R. N. Zare, *J. Am. Chem. Soc.* **2019**, *141*, 10585 – 10589; d) J. K. Lee, K. L. Walker, H. S. Han, J. Kang, F. B. Prinz, R. M. Waymouth, H. G. Nam, R. N. Zare, *Proc. Natl. Acad. Sci. USA* **2019**, *116*, 19294 – 19298.
[14] G. J. Van Berkel, F. Zhou, J. T. Aronson, *Int. J. Mass Spectrom. Ion Processes* **1997**, *162*, 55 – 67.
[15] a) R. J. Sundberg, *The Chemistry of Indoles*, Academic Press, New York, **1996**; b) T. R. Garbe, M. Kobayashi, N. Shimizu, N. Takesue, M. Ozawa, H. Yukawa, *J. Nat. Prod.* **2000**, *63*, 596 – 598; c) D. Faulkner, *Nat. Prod. Rep.* **1999**, *16*, 155 – 198; d) R. Contractor, I. J. Samudio, Z. Estrov, D. Harris, J. A. McCubrey, S. H. Safe, M. Andreeff, M. Konopleva, *Cancer Res.* **2005**, *65*, 2890 – 2898.
[16] L. J. Scott, C. M. Perry, *Drugs* **2000**, *60*, 1411 – 1444.
[17] a) K. E. Andersson, *Pharmacol. Rev.* **2001**, *53*, 417 – 450; b) A. H. Rosengren et al., *Science* **2009**, *327*, 217 – 220.
[18] S. Mahboobi, H. Pongratz, H. Hufsky, J. Hockemeyer, M. Frieser, A. Lyssenko, D. H. Paper, J. Bürgermeister, F.-D. Bhömer, H.-H. Fiebig, S. Baasner, T. J. Beckers, *J. Med. Chem.* **2001**, *44*, 4535 – 4553.
[19] M. Boes, F. Jenck, J. R. Martin, J.-L. Moreau, A. J. Sleight, J. Wichmann, U. Widmer, *J. Med. Chem.* **1997**, *40*, 2762 – 2769.
[20] A. Kumar, M. K. Gupta, M. Kumar, *Green Chem.* **2012**, *14*, 290 – 295.
[21] a) U. C. Rajesh, R. Kholiya, S. Pavan, D. S. Rawat, *Tetrahedron Lett.* **2014**, *55*, 2977 – 2981; b) L. D. Mohit, K. B. Pranjal, *Curr. Organocatal.* **2016**, *3*, 84 – 89; c) M. Kumar, K. Soni, B. Satpati, C. S. Gopinath, S. Deka, *Chem. Commun.* **2016**, *52*, 8737 – 8740.
[22] a) B. E. Love, B. T. Nguyen, *Synlett* **1998**, 1123 – 1125; b) K. Nimavat, K. Popat, S. Vasoya, H. S. Joshi, *J. Indian Chem. Soc.* **2003**, *80*, 711.
[23] a) C. A. Coburn, P. T. Meinke, W. Chang, C. M. Fandozzi, D. J. Graham, B. Hu, Q. Huang, S. Kargman, J. Kozlowski, R. J. Liu, A. McCauley, A. A. Nomeir, R. M. Soll, J. P. Vacca, D. Wang, H. Wu, B. Zhong, D. B. Olsen, S. W. Ludmerer, *ChemMedChem* **2013**, *8*, 1930 – 1940; b) H. Li, K. M. Belyk, J. Yin, Q. Chen, A. Hyde, Y. Ji, S. Oliver, M. T. Tudge, L.-C. Campeau, K. R. Campos, *J. Am. Chem. Soc.* **2015**, *137*, 13728 – 13731, and references therein; c) P. Nimavat, J. Vasoya, *J. Indian Chem. Soc.* **2003**, *80*, 711 – 713; d) R. Tiwari, G. Chhabra, *Asian J. Chem.* **2010**, *22*, 5987 – 5992.
[24] A. Akao, S. Hiraga, T. Iida, A. Kamatani, M. Kawasaki, T. Mase, T. Nemoto, N. Satake, S. A. Weissman, D. M. Tschaen, K. Rossen, D. Petrillo, R. A. Reamer, R. P. Volante, *Tetrahedron* **2001**, *57*, 8917 – 8923.
[25] C. Liu, J. Li, H. Chen, R. N. Zare, *Chem. Sci.* **2019**, *10*, 9367 – 9373.
[26] H. Heaney, S. V. Ley, *Org. Synth.* **1974**, *54*, 58.
[27] J. K. Beattie, A. M. Djerdjev, *Angew. Chem. Int. Ed.* **2004**, *43*, 3568 – 3571; *Angew. Chem.* **2004**, *116*, 3652 – 3655.
[28] We thank Roderick W. Bates, Division of Chemistry, Nanyang Technological University, Singapore, for suggesting this possibility.

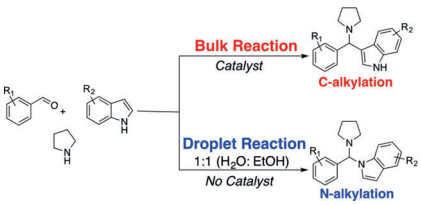
Manuscript received: October 14, 2019
Revised manuscript received: December 12, 2019
Accepted manuscript online: December 14, 2019
Version of record online: ■■ ■■, ■■■■

Communications

Microdroplets

E. Gnanamani, X. Yan,
R. N. Zare* ——————— ■■■–■■■

Chemoselective N-Alkylation of Indoles in Aqueous Microdroplets



The chemoselective N-alkylation of indoles in aqueous microdroplets proceeds in the absence of any catalyst. This approach provides a new synthetic method for the synthesis of indole aminals.