Chemistry–A European Journal
Communication
doi.org/10.1002/chem.202005191
Chemistry Europe
European Chemical Societies Publishing

C–H Functionalization |Hot Paper|

Novel Arylindigoids by Late-Stage Derivatization of Biocatalytically Synthesized Dibromoindigo

Christian Schnepel,[a, b] Veronica I. Dodero,[a] and Norbert Sewald*[a]

Abstract: Indigoids represent natural product-based compounds applicable as organic semiconductors and photoresponsive materials. Yet modified indigo derivatives are difficult to access by chemical synthesis. A biocatalytic approach applying several consecutive selective C–H functionalizations was developed that selectively provides access to various indigoids: Enzymatic halogenation of L-tryptophan followed by indole generation with tryptophanase yields 5-, 6- and 7-bromoindoles. Subsequent hydroxylation using a flavin monooxygenase furnishes dibromoindigo that is derivatized by acylation. This four-step one-pot cascade gives dibromoindigo in good isolated yields. Moreover, the halogen substituent allows for late-stage diversification by cross-coupling directly performed in the crude mixture, thus enabling synthesis of a small set of 6,6′-diarylindigo derivatives. This chemoenzymatic approach provides a modular platform towards novel indigoids with attractive spectral properties.

Indole is a widespread heterocycle found in many natural products.[1] For instance, indigo dyes derived from indole have been applied in textile dyeing for thousands of years due to their outstanding spectral properties.[2,3] Indigo (**1**) developed to a bulk chemical in the last century whereas its C6-brominated analogue, 6,6′-dibromoindigo (6,6′-**2**), the major component of the high-value pigment Tyrian purple, still remains a rarity (Scheme 1 A). Thanks to tremendous efforts by several research groups traditional indigoids recently turned into focus as natural product-based, non-toxic materials for sustainable organic electronics.[4–6] Topical studies examining the photoswitching abilities of *N*,*N*′-aryl-substituted indigos pointed to a useful strategy to tailor their photochemical properties.[7–9] Synthetic routes towards **1** were developed by Baeyer and by Heumann and paved the way to multi-ton production, yet its dibrominated counterpart **2** has never entered an industrial scale production.[10]

Today biocatalysis offers a versatile methodology to address selectivity issues, e.g., arising from similarly reactive C–H moieties.[11] Recent advancements on enzyme discovery, engineering as well as tremendous efforts on process development open up elaborate transformations that can be carried out under mild conditions, often with excellent selectivities.[12–14] Immense progress has been achieved in enzyme-catalyzed C–H functionalization.[15] Especially oxyfunctionalization is a paramount approach to activate C–H bonds by using biocatalysts that are capable of utilizing molecular oxygen. Manifold biocatalytic approaches in this rapidly evolving field, especially on the use of P450 enzymes, were extensively reviewed, giving a wide overview on the current state of the art.[16–19] The first fermentative synthesis of indigo was reported by Ensley and later by Lee et al. using a dioxygenase for indole hydroxylation. Tryptophanase that originated from endogenous tryptophan catabolism was exploited to obtain indole.[20,21] Heme-dependent monooxygenases (MOs) were later evolved towards C3-hydroxylation of indole.[22–24] Flitsch et al. used formation of indigo-derived pigments for detecting activity of MO mutants.[25,26] Biocatalytic functionalization of unprotected indole was achieved using engineered myoglobin variants which catalyze non-native carbene transfer in whole cells.[27] Besides using heme-dependent enzymes, also flavin-dependent MOs play a key role in oxyfunctionalization. Accordingly a flavin monooxygenase from *Methylophaga* sp. (mFMO) was established for the biotechnological production of indigo and indirubin.[28,29] Nevertheless, the synthesis of valuable halogenated indigos has remained on analytical or small preparative scale, particularly due to the low efficiency of halogenases as a severe bottleneck. In more recent studies, Tischler and co-authors reported on the conversion of haloindoles using a small array of styrene MOs.[30] One-pot synthesis of indigoids either in bacteria or plant as the hosts was recently achieved: By introducing a tryptophan halogenase into the host strain along with a hydroxylase, production of indigoids from tryptophan was feasible, omitting the need for costly substituted indole substrates through exploiting the cellular metabolism.[31,32] However, product titers remained low and the isolation of the pigment from the cultivation broth can become a tedious procedure. Moreover, structural modifications that can be, for ex-

[a] Dr. C. Schnepel, Dr. V. I. Dodero, Prof. Dr. N. Sewald
Organische und Bioorganische Chemie, Fakultät für Chemie
Universität Bielefeld, Universitätsstraße 25, 33615 Bielefeld (Germany)
E-mail: norbert.sewald@uni-bielefeld.de

[b] Dr. C. Schnepel
Present address: School of Chemistry
Manchester Institute of Biotechnology, The University of Manchester
131 Princess Street, Manchester, M1 7DN (UK)

Supporting information and the ORCID identification number(s) for the author(s) of this article can be found under: https://doi.org/10.1002/chem.202005191.