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Pyridine C3-arylation of nicotinic acids accessible *via* a multicomponent reaction: an entry to all-substituted-3,4-diarylated pyridines†

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An efficient route for the synthesis of penta-substituted/functionalized-3,4-diarylated pyridines, biologically important templates, *via* pyridine C3-arylation of nicotinic acids has been developed. The poly-substituted nicotinic acid precursors were prepared by an established multicomponent condensation approach. This route shows an excellent opportunity for introducing versatile (hetero)aryls and other substituents/functionalities into the pyridine ring. Several of the synthesized compounds exhibited significant anti-proliferative properties.

Introduction

Poly-substituted/functionalized pyridines are omnipresent in natural products, bioactive compounds, and functional materials.[1] In particular, aryl or poly-aryl substituted pyridine derivatives have attracted considerable synthesis attention because they are often found as a valuable structural motif in a wide range of pharmaceutically active compounds (Fig. 1). Etoricoxib that contains the 2,3-diarylpyridine motif is a COX-2 selective inhibitor.[2,3] 2,4,6-Triarylated pyridine compounds exhibit topoisomerase inhibition (Fig. 1).[4] Perampanel is a 1,3,5-triarylated pyridine derivative that acts as an AMPA-receptor antagonist.[5] 5,6-Diheteroarylpyridin-2-carboxamide derivative is a selective A2B adenosine receptor antagonist.[6] 2-Amino-4,6-diarylpyridines have been found to be potent ligands for estrogen receptor.[7] There are numerous bioactive natural products that contain polyarylated pyridines. Nosiheptide[8] has 1,5,6-triarylated 3-hydroxypyridine and promothiocin A[9] has 5,6-diarylated pyridine-1-carboxamide and both are potent antibiotics. 2,6-Diarylpyridine-carboxylic ester is useful as a fluorescent chemosensor.[10]

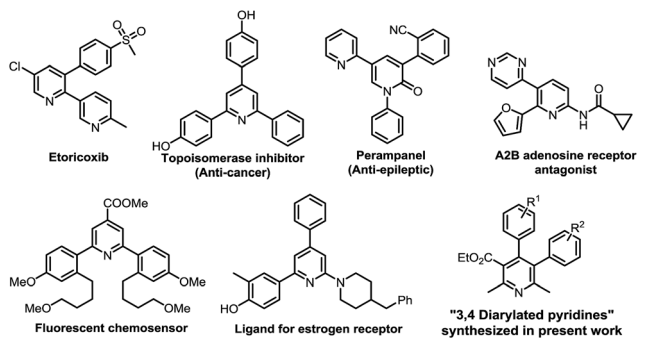

Fig. 1 Selected bioactive agents containing the poly-arylated pyridine structure.

In recent times, as a part of current momentum of exploring new useful organic functional/bioactive materials, the synthesis of compounds that possess not only important heterocyclic scaffold(s) but also structurally resembles in whole molecular skeleton to drugs/bioactive agents has become a valuable research area to organic chemists.[11] In this direction, late stage functionalization of drugs has also attracted significant attention.[12] The incorporation of trimethoxyphenyl moiety in target organic compounds is valuable, since it plays role as an important pharmacophoric motif in exhibiting biological properties[13] and is present in natural products.[14] Keeping these aspects in mind, we were interested[15] in the synthesis of 3,4-diarylated pyridines which contain structural features, 3,4,5-trimethoxyphenyl as an aryl motif, and resemblance to a nifedipine drug[16] and biologically important nicotinic acid.[17]

Over the years, numerous synthetic strategies have been developed to access poly-functionalized pyridines.[18,19] They involve majorly a pyridine-forming step of appropriately functionalized precursors[20] or the sequential introduction of substituents on the preformed pyridine ring.[21] The first strategy is limited to accessibility of particularly-functionalized precursor, while the later strategy includes mainly aromatic substitutions,[22] direct metallation, or metal halogen-exchange reactions.[23] Here we report a novel route for synthesis of all-substituted/functionalized pyridines containing 3,4-diaryl moieties *via* one-pot pyridine-3-arylation of nicotinic acid skeleton that is easily accessible by an established multicomponent reaction approach.

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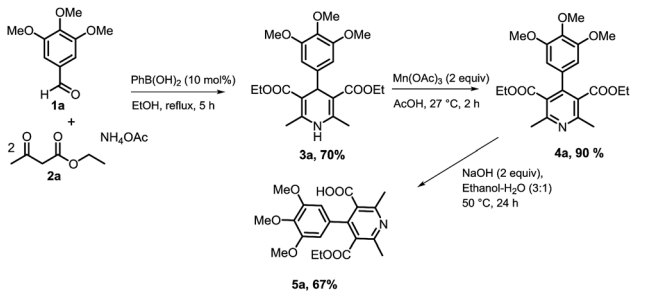
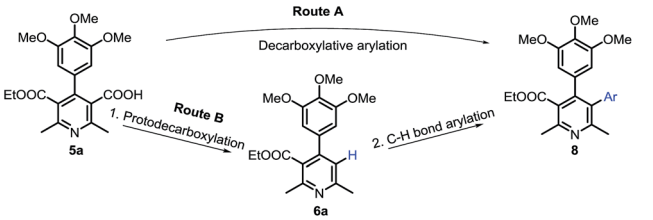
Results and discussion

At the outset, we investigated for preparation of nicotinic acid precursor (**5a**) by a Hantzsch multicomponent condensation of aryl aldehyde (**1a**), ethyl acetoacetate (**2a**) and ammonium acetate to construct dihydropyridine (**3a**), its dehydrogenative aromatization to product **4a**, and mono-hydrolysis of di-ester (Scheme 1). Hantzsch reaction by a reported method was good yielding.[24] Dehydrogenative aromatization reactions utilizing different conditions[25] were performed (see ESI, Table 1†). Highest yield was obtained using $Mn(OAc)_3$ and AcOH at room temperature. Initially, the mono-hydrolysis of pyridine-3,5-diesters **4a** was found to be difficult. Both the ester functionalities underwent simultaneously the hydrolysis. A survey of various conditions revealed that 2 equiv. NaOH in 2 mL of EtOH–H_2O (3 : 1) was most effective for promoting mono-hydrolysis to produce nicotinic acid **5a** (see ESI, Table 2†).

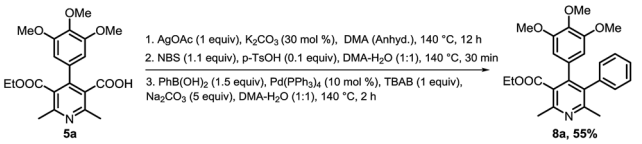
Next, we investigated to explore decarboxylative arylation of nicotinic acid **5a**, with the goal of finding a convenient method for synthesis of penta-substituted/functionalized pyridines containing 3,4-diaryl moieties (Route A, Scheme 2). Several methods of decarboxylative arylation[26] known for other scaffolds were investigated for the reaction of compound **5a**. The desired product was obtained, but the yield more than 8% could not be accomplished by the methods as well as variation in reported conditions. We envisaged that protodecarboxylation,[27] and subsequent C3–H bond arylation could afford C3-arylated pyridine (Route B, Scheme 2). The protodecarboxylation of compound **5a** underwent *via* Ag-carboxylate on treatment with AgOAc (1 equiv.), K_2CO_3 (30 mol%) in DMA (anhyd.) at 140 °C for 12 h and the product **6a** was obtained in 80% yield. Next, the C3–H arylation reactions of compound **6a** following the reported pyridine-arylation methods were performed. Ye developed a Pd-catalyzed C3-selective arylation of unsubstituted pyridine with bromoarenes.[28] Carrying out C3-arylation of substituted/functionalized pyridine **6a** with bromo- or iodotoluene using Ye's conditions provided 3-arylated pyridine (**8a**) in poor yields (8% and 10% yields, respectively). A Pd-catalyzed 3-arylation of pyridine with aryl tosylates was reported by Dai.[29] This methodology did not promote the reaction of compound **6a** with *p*-tolyl tosylate. We explored previously a Pd-catalyzed regioselective C6–H arylation of 3-aminoimidazo[1,2-*a*]pyrazine that underwent *via* concerted metalation–deprotonation process.[30] This method promoted the C3–H arylation of pyridine derivative **6a**, but the method as well as the variation of its conditions could not improve the product's yield more than 20%. Majorly, the substrate remained intact. The poor conversion and yields in the C3-arylation of pyridine **6a** by reported methods might be due to significant steric hindrance by 4-aryl moiety to inhibit the substrate to undergo the C3-palladation with arylpalladium complex. With aim of obtaining the C-3



Scheme 1 Synthesis of nicotinic acid derivative **5a**.

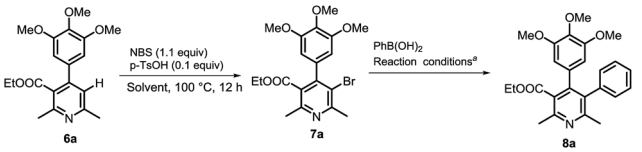


Scheme 2 Strategies towards synthesis of polysubstituted 3,4-diarylated pyridines.



Scheme 3 Optimized reaction condition for one-pot proto-decarboxylation–bromination–Suzuki coupling.

Table 1 Evaluation of reagents and conditions[a]



Entry	Variable	Yield[b]
Catalyst (10 mol%), Na_2CO_3 (5 equiv.), TBAB (1 equiv.), DMA–H_2O (1 : 1)		
1	$Pd(PPh_3)_4$	70
2	$Pd(dppf)Cl_2$	55
3	$Pd(PPh_3)_2Cl_2$	NR
4[c]	$Pd(OAc)_2$ + PPh_3	NR
5[d]	$Pd(PPh_3)_4$	40
$Pd(PPh_3)_4$ (10 mol%), base (5 equiv.), TBAB (1 equiv.), DMA–H_2O (1 : 1)		
6	K_2CO_3	60
7	Cs_2CO_3	55
8	K_3PO_4	40
9[e]	Na_2CO_3	45
$Pd(PPh3)_4$ (10 mol%), Na_2CO_3 (5 equiv.), TBAB (1 equiv.), solvent		
10	EtOH–H_2O (1 : 1)	60
11	DMA	55
12[f]	EtOH	45

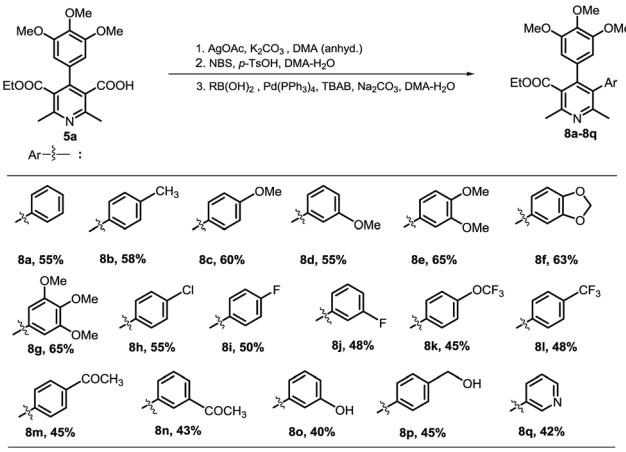
[a] Reagents and conditions: $PhB(OH)_2$ (1.5 equiv.), 100 °C, Ar, 2 h; 0.5 mmol scale. [b] Yield for maximum conversion in optimum time. [c] PPh_3 (40 mol%). [d] $Pd(PPh_3)_4$ (5 mol%). [e] Na_2CO_3 (2.5 equiv.). [f] Reaction temperature: 80 °C.

arylated product, we then investigated for an alternate approach involving protodecarboxylation, C3-bromination and Suzuki coupling sequentially (Scheme 3). C3-Bromination[31] of pyridine **6a** leading to product **7a** was found to be nearly quantitative (95%). Therefore, we were interested to explore one-pot conditions for bromination–Suzuki coupling. A $Pd(OAc)_2$-catalyzed method reported by Liu[32] for Suzuki reaction of *N*-heteroaryl halides was followed in the one-pot reaction of pyridine **6a** with phenylboronic acid, however, the desired product was obtained in 25% yield only. Christakakou's[33] reaction conditions yielded the product **8a** in 40% yield only. Gratifyingly, one-pot bromination and Suzuki coupling of pyridine **6a** with phenylboronic acid using $Pd(PPh_3)_4$ catalyst, Na_2CO_3, TBAB in DMA–H_2O provided 3,4-diaryl-pyridine **8a** in 70% yield (Table 1, entry 1). However, further variation in conditions did not improve yield of the product. Incomplete conversion and inferior yield (15%) were obtained for the reaction carried out in the presence of open air, indicating requirement of non-aerobic conditions for Pd(0)-catalysis. The reaction without TBAB resulted in reduced yield (40%). Other palladium sources were evaluated (Table 1, entries 2–4). $Pd(PPh_3)_4$ was found to be best. Reducing the Pd-catalyst loading (entry 5) was not beneficial. Among various bases investigated, Na_2CO_3 provided best result (entries 6–8). Decreasing the equivalence of sodium carbonate below to 5 equiv. resulted in reduced yield (Table 1, entry 9). DMA or EtOH was found effective solvent for bromination, however, they were inferior for promoting the Suzuki coupling in one-pot. Among various solvents and their mixture with water, DMA–H_2O was found to be most effective for promoting bromination–arylation in one-pot (entries 10–12).

We then investigated protodecarboxylation of nicotinic acid **5a**, bromination and Suzuki coupling sequentially in one-pot. Amazingly, it worked well in one-pot with overall yield of 55% (Scheme 3). The one-pot multi-reactions synthesis of a target molecule is considered as a useful approach in synthetic organic chemistry.[34] The present work illustrates an important example of one-pot three-reaction process.

With the optimized procedure, we next investigated to explore its substrate scope and synthesize various substituted pyridines. We were pleased to find that the method was found to be flexible in introducing into pyridine at C3 a variety of (hetero) aryls (Scheme 4). Aryls containing electron-withdrawing or donating functionalities were incorporated. The biologically relevant (hetero)aryl motifs were also introduced easily at C3-position of pyridine derivatives. In the established route, difficulty for pyridine C3-arylation due to steric hindrance by presence of a multi-substituted aryl (3,4,5-trimethoxyphenyl) at C4-position was circumvented.

On survey of literature, we found that Simoni demonstrated an attractive profile of cytotoxicity and apoptosis-inducing activity of 2-(3,4,5-trimethoxyphenyl)-3-(3-hydroxy-4-methoxyphenyl)-pyridine (compound **A**, Fig. 2).[35] Zheng documented that 2-(3-hydroxy-4-methoxyphenyl)-6-(3,4,5-trimethoxyphenyl)-pyridine (**B**) inhibited cell survival and growth, comparable to a clinical agent, combretastatin A-4 (CA-4).[36] These representative classes of compounds reveal that 3,4-diarylated pyridine derivatives



Scheme 4 Substrate scope.[a,b] [a]Substrates, reagents and conditions: **5a** (1 mmol), AgOAc (1 equiv.), K_2CO_3 (30 mol%), DMA (anhyd., 2 mL), 12 h; then, NBS (1.1 equiv.), *p*-TsOH (0.1 equiv.), DMA–H_2O (1 : 1, 4 mL), 30 min; then, $RB(OH)_2$ (1.5 equiv.), $Pd(PPh_3)_4$ (10 mol%), TBAB (1 equiv.), Na_2CO_3 (5 equiv.), 140 °C, 1–12 h; [b]yield for maximum conversion in optimum time.

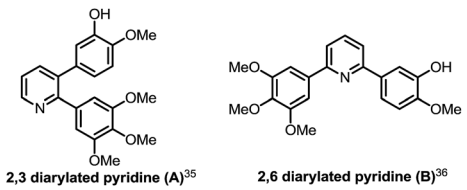
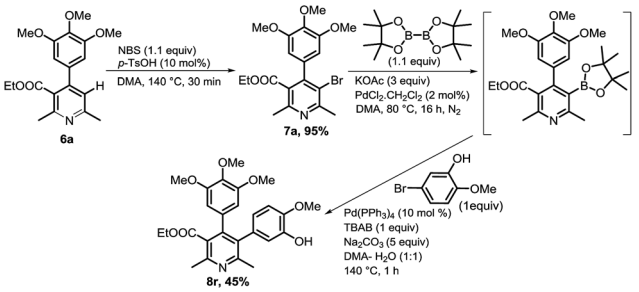


Fig. 2 Reported diarylated pyridines with potent anticancer activity.

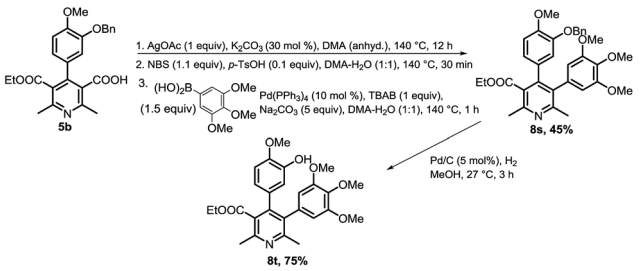
synthesized in the present work have potential of exhibiting versatile bioactivities especially the antiproliferative properties.

We extended our developed method applicable to the synthesis of 3,4-diarylated pyridines with relevant substitutions that are present in CA-4 and the diarylated pyridines synthesized and bio-evaluated by Simoni and Zhang.

Ethyl 5-(3-hydroxy-4-methoxyphenyl)-2,6-dimethyl-4-(3,4,5-trimethoxyphenyl)nicotinate (**8r**) was prepared *via* an approach (Scheme 5) involving C3-bromination of precursor **6a**, boronation[37] of **7a** with bispinacolatodiborane, and Suzuki coupling with 5-bromo-2-methoxyphenol. This prompted us to synthesize compound **8t** with switch in aryl substitutions of



Scheme 5 Synthesis of ethyl 5-(3-hydroxy-4-methoxyphenyl)-2,6-dimethyl-4-(3,4,5-trimethoxyphenyl)nicotinate (**8r**).



Scheme 6 Synthesis of ethyl 4-(3-hydroxy-4-methoxyphenyl)-2,6-dimethyl-5-(3,4,5-trimethoxyphenyl)nicotinate (**8t**).

compound **8r**. We began the preparation of compound **8t** *via* similar strategy, utilizing 5-(ethoxycarbonyl)-4-(3-hydroxy-4-methoxyphenyl)-2,6-dimethylnicotinic acid as the precursor, but bromination did not take place. We anticipated that bromination was problematic due to presence of free hydroxyl group in the precursor. So, benzylation of the hydroxyl group was done to circumvent this problem, and we carried out the synthesis of compound **8t** utilizing benzyloxy-aryl derivative of nicotinic acid (**5b**). One-pot protodecarboxylation–bromination–Suzuki coupling was done to afford compound **8s**, which on debenzylation resulted in compound **8t** (Scheme 6).

A set of significantly varied 3,4-diarylated pyridine derivatives (**8a–8t**) were prepared in the developed approach. Purity (HPLC) of all the compounds were found to be >95%. Next antiproliferative activity was tested. All compounds were screened for % inhibition of cell proliferation in HeLa cells as a representative cancer cell line at 5 μM concentration (see ESI, Table 3†). Several of them were found to exhibit considerable antiproliferative activity. Four most potent compounds (pyridines **8b**, **8f**, **8j**, **8p**) were further evaluated for *in vitro* tubulin polymerization inhibition (see ESI, Fig. 1†). The non-activity revealed that the antitubulin may not be the pathway for exhibiting cytotoxicity.

Conclusions

In conclusion, we have developed an efficient route for synthesis of all-substituted/functionalized pyridines containing 3,4-diaryl moieties. It involves the preparation of nicotinic acids *via* multicomponent condensation and their pyridine C3-arylation *via* one-pot protodecarboxylation–bromination–Suzuki coupling. A wide range of (hetero)aryls, including especially the biologically important aryl-motifs can be easily introduced in this route. The structural features of the products, structural resemblance to pharmacologically important agent as well as drug and the presence of a biologically important motif 3,4,5-trimethoxyphenyl as one aryl ring, indicate that these functionalized 3,4-diarylpyridines have potential application in finding of versatile bioactive agents. Several of the synthesized compounds were found to exhibit significant anti-proliferative activity.

Experimental

Chemistry

General information. ATR & IR (KBr) Microscope spectrometer was used to record Infrared (IR (KBr)) spectra. ^{1}H NMR spectra were taken on a 400 MHz spectrometer. Data were reported in sequence of chemical shifts in ppm from tetramethylsilane as an internal standard in $CDCl_3$/CD_3OD, integration, multiplicity (s = singlet, d = doublet, t = triplet, q = quartet, m = multiplet, dt = doublet of triplet, dd = doublet of doublet, br = broad), and coupling constants (Hz). ^{13}C NMR spectra were recorded on a 100 MHz spectrometer with protons-decoupling. High-resolution mass spectra (HRMS) were recorded on a high-resolution LCMS/MS instrument with "QTOF" mass analyzer. Thin-layer chromatography (TLC) analysis was done using commercially received pre-coated TLC plates (silica gel 60 GF434, 0.43 mm). Column chromatography silica gel 100–200 (silica gel 100–200 mesh, neutral, spherical) was used for purification of products. The starting materials and solvents were used as received from commercial sources without further purification.

Representative experimental procedure for the synthesis of diethyl 2,6-dimethyl-4-(3,4,5-trimethoxyphenyl)-1,4-dihydropyridine-3,5-dicarboxylate (Scheme 1, 3a).[24] 3,4,5-Trimethoxybenzaldehyde **1a** (1 mmol, 196 mg), ethyl acetoacetate **2a** (2 mmol, 260 mg, 2 eq.), NH_4OAc (2 mmol, 154 mg, 2 eq.), and $PhB(OH)_2$ (0.1 mmol, 12 mg, 0.1 eq.) were taken in a round bottom flask and refluxed in EtOH (2 mL) for 5 h. The reaction mixture was poured into ice cold H_2O. It was then extracted with EtOAc (2 × 10 mL). The organic solution was washed with brine, dried over Na_2SO_4, and concentrated under vacuum. The crude product was purified by recrystallization from EtOH, which provided 1,4-dihydropyridine **3a** in 70% yield.

Representative experimental procedure for the synthesis of diethyl 2,6-dimethyl-4-(3,4,5-trimethoxyphenyl)pyridine-3,5-dicarboxylate (Scheme 1, 4a).[25a] Diethyl 1,4-dihydro-2,6-dimethyl-4-(3,4,5-trimethoxyphenyl)pyridine-3,5-dicarboxylate (**3a**) (1 mmol, 253 mg), manganese triacetate (2 mmol, 536 mg, 2 eq.) and acetic acid (5 mL) were taken in a round bottom flask. The reaction mixture was stirred at room temperature till the completion of reaction. Manganese diacetate was filtered through celite bed and the mixture was poured into ice-cold water. Ice-cold aqueous $NaHCO_3$ solution was added dropwise to neutralize the mixture. It was then extracted with dichloromethane (2 × 10 mL) and dried over anhydrous sodium sulfate. The solution was concentrated under reduced pressure. The resulting crude mixture on crystallisation from ethanol provided pure product **4a** in 90% yield.

Representative experimental procedure for the synthesis of 2,6-dimethyl-5-(ethoxycarbonyl)-4-(3,4,5-trimethoxyphenyl)nicotinic acid (Scheme 1, 5a). Aqueous solution of KOH (2 mmol, 112 mg, 2 eq.) in 0.5 mL water was added to a solution of compound **4a** (1 mmol, 417 mg) in ethanol (1.5 mL). The mixture was refluxed (80 °C) till completion of the reaction as indicated by TLC (7 h). The solvent was evaporated and the crude mass obtained was redissolved in methanol. It was then neutralised to pH 7 with dropwise addition of ice-cold methanolic HCl. The organic solution was concentrated under reduced pressure. The column chromatographic purification of crude mass on silica gel eluting with MeOH–EtOAc (1 : 9) provided the product **5a** in 67% yield.

Representative experimental procedure for synthesis of 2,6-dimethyl-3-(ethoxycarbonyl)-5-phenyl-4-(3,4,5-trimethoxyphenyl)-pyridine (Scheme 4, entry 1) (8a). Monoacid **5a** (1 mmol, 389 mg), AgOAc (1 mmol, 167 mg, 1 eq.) and K_2CO_3 (0.30 mmol, 41.5 mg, 0.3 eq.) were taken under nitrogen in an oven-dried sealed tube equipped with a rubber septum and magnetic bar. DMA (anhyd., 2 mL) was added under nitrogen. The tube was then sealed. The mixture was stirred at 140 °C. Upon completion of reaction as indicated by TLC (12 h), *N*-bromosuccinimide (1.1 mmol, 196 mg, 1.1 equiv.), *p*-TsOH (0.1 mmol, 17 mg, 0.1 eq.) and water (2 mL) were added in the reaction tube. After completion of the bromination reaction after 30 min, sodium carbonate (5 mmol, 530 mg, 5 eq.), tetrabutyl ammonium bromide (1 mmol, 322 mg, 1 eq.), phenylboronic acid (1.5 mmol, 182 mg, 1.5 eq.) and $Pd(PPh_3)_4$ (0.1 mmol, 115 mg, 0.1 eq.) were added to the reaction tube under nitrogen. The tube was then sealed. The mixture was stirred at 140 °C till the completion of reaction monitored by TLC (2 h). Then, resultant mixture was allowed to cool to room temperature, diluted with ethyl acetate (20 mL). The organic layer was washed with aqueous solution of ammonia (3 × 5 mL) and brine (5 mL), dried over Na_2SO_4 and the organic layer was concentrated under reduced pressure. The column chromatographic purification of crude mass on silica gel eluting with EtOAc–hexane (1 : 3) provided 2,6-dimethyl-3-(ethoxycarbonyl)-5-phenyl-4-(3,4,5-trimethoxyphenyl)pyridine **8a** in 55% overall yield.

Products (**8b–8t**) were also prepared following this representative procedure.

Diethyl 2,6-dimethyl-4-(3,4,5-trimethoxyphenyl)-1,4-dihydropyridine-3,5-dicarboxylate (3a).[38] Yellow solid; 293 mg, 70%; mp 140 °C; ^{1}H NMR (400 MHz, $CDCl_3$): δ 6.52 (s, 2H), 5.77 (s, 1 NH), 4.98 (s, 1H), 4.17–4.08 (m, 4H), 3.79 (s, 9H), 2.34 (s, 6H), 1.25 (t, J = 7.1 Hz, 6H) ppm; ^{13}C{^{1}H} NMR (100 MHz, $CDCl_3$): δ 167.7, 152.6, 143.9, 143.4, 105.0, 103.9, 60.8, 59.8, 55.9, 19.6, 14.4 ppm; IR (KBr): ν_{max} 3354, 2987, 1650, 1232, 1123, 1006 cm^{-1}.

Diethyl 2,6-dimethyl-4-(3,4,5-trimethoxyphenyl)pyridine-3,5-dicarboxylate (4a). White solid; 375 mg, 90%; mp 130 °C; ^{1}H NMR (400 MHz, $CDCl_3$): δ 6.51 (s, 2H), 4.08 (q, J = 7.1 Hz, 4H), 3.86 (s, 3H), 3.83 (s, 6H), 2.60 (s, 6H), 0.99 (t, J = 7.1 Hz, 6H) ppm; ^{13}C{^{1}H} NMR (100 MHz, $CDCl_3$): δ 168.0, 155.4, 152.9, 145.7, 138.1, 131.9, 126.8, 105.5, 61.5, 60.9, 56.2, 22.8, 13.7 ppm; IR (KBr): ν_{max} 2987, 1719, 1585, 1232, 1123, 1006 cm^{-1}; HRMS (ESI) m/z: calcd for $C_{22}H_{28}NO_7$ $[M + H]^+$ 418.1866, found: 418.1858.

2,6-Dimethyl-5-(ethoxycarbonyl)-4-(3,4,5-trimethoxyphenyl) nicotinic acid (5a). Off white solid; 260 mg, 67%; mp charred at 225 °C; ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.03 (s, 1H), 6.58 (s, 2H), 4.14 (q, J = 7.2 Hz, 2H), 3.88 (s, 3H), 3.87 (s, 6H), 2.60 (s, 3H), 2.59 (s, 3H), 1.06 (t, J = 7.2 Hz, 3H) ppm; ^{13}C{^{1}H} NMR (100 MHz, $CDCl_3$): δ 169.2, 158.7, 155.0, 153.3, 148.2, 138.2, 134.3, 125.8, 120.8, 105.1, 61.4, 60.9, 56.2, 24.5, 22.8, 13.8 ppm; IR (KBr): ν_{max} 3423, 2970, 1718, 1582, 1249, 1122, 1010 cm^{-1}; HRMS (ESI) m/z: calcd for $C_{20}H_{24}NO_7$ $[M + H]^+$ 390.1553, found: 390.1547.

2,6-Dimethyl-3-(ethoxycarbonyl)-5-phenyl-4-(3,4,5-trimethoxyphenyl)pyridine (8a). White solid; 232 mg, 55%; mp 120 °C; ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.20–7.19 (m, 5H), 6.56 (s, 1H), 6.42 (s, 1H), 4.15–4.12 (m, 2H), 3.95 (s, 3H), 3.84 (s, 3H), 3.59 (s, 3H), 2.57 (s, 3H), 2.26 (s, 3H), 1.03 (t, J = 7.1 Hz, 3H) ppm; ^{13}C{^{1}H} NMR (100 MHz, $CDCl_3$): δ 168.9, 157.5, 155.1, 152.3, 151.7, 148.4, 142.3, 135.7, 133.6, 130.6, 127.9, 127.6, 126.8, 126.3, 122.7, 108.3, 61.2, 61.1, 60.9, 56.1, 24.2, 23.0, 13.7 ppm; IR (KBr): ν_{max} 2978, 2935, 2850, 1726, 1586, 1485, 1264, 1096 cm^{-1}; HRMS (ESI) m/z: calcd for $C_{25}H_{28}NO_5$ $[M + H]^+$ 422.1967, found: 422.1958.

2,6-Dimethyl-3-(ethoxycarbonyl)-5-(*p*-tolyl)-4-(3,4,5-trimethoxyphenyl)pyridine (8b). White solid; 252 mg, 58%; mp 123 °C; ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.07 (d, J = 7.1 Hz, 2H), 7.00 (d, J = 7.7 Hz, 2H), 6.54 (s, 1H), 6.44 (s, 1H), 4.15–4.09 (m, 2H), 3.94 (s, 3H), 3.84 (s, 3H), 3.60 (s, 3H), 2.57 (s, 3H), 2.29 (s, 3H), 2.28 (s, 3H) 1.02 (t, J = 7.1 Hz, 3H) ppm; ^{13}C{^{1}H} NMR (100 MHz, $CDCl_3$): δ 168.9, 157.5, 155.1, 152.1, 151.8, 148.6, 142.2, 136.3, 133.7, 132.6, 130.5, 128.4, 127.8, 126.3, 122.8, 108.2, 61.1, 61.0, 60.9, 56.1, 24.2, 23.1, 21.2, 13.7 ppm; IR (KBr): ν_{max} 2925, 2854, 1724, 1587, 1488, 1263, 1087 cm^{-1}; HRMS (ESI) m/z: calcd for $C_{26}H_{30}NO_5$ $[M + H]^+$ 436.2124, found: 436.2124.

2,6-Dimethyl-3-(ethoxycarbonyl)-5-(4-methoxyphenyl)-4-(3,4,5-trimethoxyphenyl)pyridine (8c). White solid; 271 mg, 60%; mp > 200 °C; ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.12 (d, J = 7.8 Hz, 2H), 6.75 (d, J = 8.4 Hz, 2H), 6.53 (s, 1H), 6.44 (s, 1H), 4.15–4.09 (m, 2H), 3.94 (s, 3H), 3.84 (s, 3H), 3.77 (s, 3H), 3.58 (s, 3H), 2.58 (s, 3H), 2.30 (s, 3H), 1.02 (t, J = 7.1 Hz, 3H) ppm; ^{13}C{^{1}H} NMR (100 MHz, $CDCl_3$): δ 168.9, 158.3, 157.6, 155.1, 152.1, 151.8, 148.7, 142.3, 133.7, 131.7, 127.9, 127.4, 126.4, 122.7, 113.1, 108.2, 61.2, 61.1, 60.9, 56.1, 55.1, 24.3, 23.1, 13.7 ppm; IR (KBr): ν_{max} 2930, 2850, 1725, 1586, 1488, 1262, 1245, 1096 cm^{-1}; HRMS (ESI) m/z: calcd for $C_{26}H_{30}NO_6$ $[M + H]^+$ 452.2073, found: 452.2068.

2,6-Dimethyl-3-(ethoxycarbonyl)-5-(3-methoxyphenyl)-4-(3,4,5-trimethoxyphenyl)pyridine (8d). White solid; 248 mg, 55%; mp 116 °C; ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.13 (dd, J = 7.8 Hz, J = 7.7 Hz, 1H), 6.84–6.78 (m, 2H), 6.74–6.71 (m, 1H), 6.55 (s, 1H), 6.46 (s, 1H), 4.15–4.12 (m, 2H), 3.95 (s, 3H), 3.84 (s, 3H), 3.65 (s, 3H), 3.63 (s, 3H), 2.57 (s, 3H), 2.29 (s, 3H), 1.03 (t, J = 7.1 Hz, 3H) ppm; ^{13}C{^{1}H} NMR (100 MHz, $CDCl_3$): δ 168.9, 158.9, 157.6, 154.9, 152.3, 151.7, 148.5, 142.3, 137.0, 133.5, 128.5, 127.7, 126.3, 123.3, 122.6, 115.9, 112.9, 108.2, 61.2, 61.1, 61.0, 56.1, 55.1, 24.2, 22.9, 13.8 ppm; IR (KBr): ν_{max} 2927, 2852, 1724, 1586, 1462, 1274, 1259, 1088 cm^{-1}; HRMS (ESI) m/z: calcd for $C_{26}H_{30}NO_6$ $[M + H]^+$ 452.2073, found: 452.2068.

5-(3,4-Dimethoxyphenyl)-2,6-dimethyl-3-(ethoxycarbonyl)-4-(3,4,5-trimethoxyphenyl)pyridine (8e). Pale yellow solid; 313 mg, 65%; mp 135 °C; ^{1}H NMR (400 MHz, $CDCl_3$): δ 6.84–6.79 (m, 2H), 6.74 (d, J = 8.2 Hz, 1H), 6.55 (s, 1H), 6.46 (s, 1H), 4.18–4.11 (m, 2H), 3.95 (s, 3H), 3.85 (s, 3H), 3.84 (s, 3H), 3.67 (s, 3H), 3.62 (s, 3H), 2.56 (s, 3H), 2.29 (s, 3H), 1.05 (t, J = 7.1 Hz, 3H) ppm; ^{13}C{^{1}H} NMR (100 MHz, $CDCl_3$): δ 168.9, 157.7, 154.8, 152.1, 151.8, 148.7, 147.9, 147.7, 142.3, 133.6, 128.2, 127.5, 126.4, 123.1, 122.5, 114.1, 110.3, 108.3, 61.2, 61.1, 60.9, 56.1, 55.7, 24.3, 22.9, 13.8 ppm; IR (KBr): ν_{max} 2924, 2853, 1726, 1586, 1463, 1248, 1086 cm^{-1}; HRMS (ESI) m/z: calcd for $C_{26}H_{32}NO_7$ $[M + H]^+$ 482.2179, found: 482.2181.

5-(Benzo[*d*][1,3]dioxol-5-yl)-2,6-dimethyl-3-(ethoxycarbonyl)-4-(3,4,5-trimethoxyphenyl)pyridine (8f). White solid; 293 mg, 63%; mp 115 °C; ^{1}H NMR (400 MHz, $CDCl_3$): δ 6.73–6.67 (m,

3H), 6.52 (s, 1H), 6.49 (s, 1H), 5.92 (s, 2H), 4.16–4.09 (m, 2H), 3.94 (s, 3H), 3.83 (s, 3H), 3.63 (s, 3H), 2.58 (s, 3H), 2.34 (s, 3H), 1.02 (t, J = 7.1 Hz, 3H) ppm; $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$): δ 168.8, 157.6, 155.3, 152.2, 151.8, 148.6, 146.9, 146.3, 142.2, 133.7, 129.4, 127.3, 126.3, 124.3, 122.5, 111.1, 108.1, 107.7, 100.8, 61.2, 61.1, 60.9, 56.1, 24.3, 23.1, 13.7 ppm; IR (KBr): ν_{max} 2925, 2851, 1723, 1586, 1481, 1455, 1262, 1233, 1123, 1081 cm^{-1}; HRMS (ESI) m/z: calcd for $C_{26}H_{28}NO_7$ $[M + H]^+$ 466.1866, found: 466.1866.

2,6-Dimethyl-3-(ethoxycarbonyl)-4,5-bis(3,4,5-trimethoxyphenyl)-pyridine (8g). Yellow semisolid; 332 mg, 65%; ^1H NMR (400 MHz, $CDCl_3$): δ 6.57 (s, 1H), 6.51 (s, 2H), 6.47 (s, 1H), 4.18–4.12 (m, 2H), 3.95 (s, 3H), 3.85 (s, 3H), 3.80 (s, 3H), 3.68 (s, 9H), 2.56 (s, 3H), 2.31 (s, 3H), 1.06 (t, J = 7.1 Hz, 3H) ppm; $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$): δ 168.8, 157.7, 154.6, 152.4, 152.2, 151.6, 148.6, 142.3, 136.8, 133.4, 131.1, 127.6, 126.4, 122.2, 108.3, 108.2, 61.3, 61.11, 61.09, 60.9, 56.1, 56.0, 24.2, 22.8, 13.9 ppm; IR (KBr): ν_{max} 2931, 2852, 1726, 1586, 1464, 1262, 1127, 1103, 1008 cm^{-1}; HRMS (ESI) m/z: calcd for $C_{28}H_{34}NO_8$ $[M + H]^+$ 512.2284, found: 512.2280.

5-(4-Chlorophenyl)-2,6-dimethyl-3-(ethoxycarbonyl)-4-(3,4,5-trimethoxyphenyl)pyridine (8h). White solid; 250 mg, 55%; mp > 200 °C; ^1H NMR (400 MHz, $CDCl_3$): δ 7.19–7.14 (m, 4H), 6.54 (s, 1H), 6.42 (s, 1H), 4.15–4.12 (m, 2H), 3.94 (s, 3H), 3.84 (s, 3H), 3.59 (s, 3H), 2.58 (s, 3H), 2.31 (s, 3H), 1.03 (t, J = 7.2 Hz, 3H) ppm; $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$): δ 168.7, 157.7, 155.3, 152.5, 151.6, 148.2, 142.3, 134.2, 133.5, 132.7, 132.0, 127.8, 126.41, 126.36, 122.6, 108.3, 61.2, 61.1, 60.9, 56.1, 24.2, 22.9, 13.7 ppm; IR (KBr): ν_{max} 2934, 2851, 1724, 1585, 1483, 1261, 1084, 1005, 827 cm^{-1}; HRMS (ESI) m/z: calcd for $C_{25}H_{27}NO_6$ $[M + H]^+$ 456.1570, found: 456.1588.

2,6-Dimethyl-3-(ethoxycarbonyl)-5-(4-fluorophenyl)-4-(3,4,5-trimethoxyphenyl)pyridine (8i). White solid; 220 mg, 50%; mp 145 °C; ^1H NMR (400 MHz, $CDCl_3$): δ 7.20–7.17 (m, 2H), 6.91 (dd, J = 8.5 Hz, J = 8.4 Hz, 2H), 6.55 (s, 1H), 6.41 (s, 1H), 4.17–4.10 (m, 2H), 3.95 (s, 3H), 3.84 (s, 3H), 3.59 (s, 3H), 2.58 (s, 3H), 2.30 (s, 3H), 1.03 (t, J = 7.2 Hz, 3H) ppm; $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$): δ 168.8, 161.7 (d, J_{C-F} = 244 Hz), 157.7, 155.2, 152.4, 151.7, 148.3, 142.3, 133.7, 132.3 (d, $J_{C-C-C-F}$ = 8 Hz), 131.6 (d, $J_{C-C-C-C-F}$ = 4 Hz), 126.7, 126.3, 122.6, 114.6 (d, J_{C-C-F} = 21 Hz), 108.3, 61.2, 61.1, 60.9, 56.1, 24.2, 23.1, 13.8 ppm; IR (KBr): ν_{max} 2959, 2928, 2870, 1716, 1586, 1487, 1257, 1093, 1082, 1006 cm^{-1}; HRMS (ESI) m/z: calcd for $C_{25}H_{27}FNO_5$ $[M + H]^+$ 440.1873, found: 440.1860.

2,6-Dimethyl-3-(ethoxycarbonyl)-5-(3-fluorophenyl)-4-(3,4,5-trimethoxyphenyl)pyridine (8j). White solid; 211 mg, 48%; mp 114 °C; ^1H NMR (400 MHz, $CDCl_3$): δ 7.18–7.13 (m, 1H), 6.98–6.96 (m, 1H), 6.91–6.86 (m, 1H), 6.54 (s, 1H), 6.44 (s, 1H), 4.15–4.10 (m, 2H), 3.95 (s, 3H), 3.84 (s, 3H), 3.62 (s, 3H), 2.58 (s, 3H), 2.30 (s, 3H), 1.03 (t, J = 7.2 Hz, 3H) ppm; $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$): δ 168.7, 162.2 (d, J_{C-F} = 243 Hz), 157.8, 155.3, 152.6, 151.6, 148.1, 142.3, 137.9 (d, $J_{C-C-C-F}$ = 8 Hz), 133.6, 129.0 (d, $J_{C-C-C-F}$ = 8 Hz), 126.6, 126.4, 126.3, 122.4, 117.6 (d, J_{C-C-F} = 21 Hz), 113.7 (d, J_{C-C-F} = 21 Hz), 108.3, 61.2, 61.1, 61.0, 56.1, 24.2, 23.1, 13.8 ppm; IR (KBr): ν_{max} 2955, 2927, 2852, 1724, 1584, 1482, 1260, 1129, 1084, 1014 cm^{-1}; HRMS (ESI) m/z: calcd for $C_{25}H_{27}FNO_5$ $[M + H]^+$ 440.1873, found: 440.1869.

2,6-Dimethyl-3-(ethoxycarbonyl)-5-(4-(trifluoromethoxy)phenyl)-4-(3,4,5-trimethoxyphenyl)pyridine (8k). White solid; 227 mg, 45%; mp 130 °C; ^1H NMR (400 MHz, $CDCl_3$): δ 7.24 (d, J = 7.6 Hz, 2H), 7.06 (d, J = 8.1 Hz, 2H), 6.57 (s, 1H), 6.36 (s, 1H), 4.16–4.11 (m, 2H), 3.95 (s, 3H), 3.85 (s, 3H), 3.62 (s, 3H), 2.58 (s, 3H), 2.27 (s, 3H), 1.03 (t, J = 7.1 Hz, 3H) ppm; $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$): δ 168.7, 157.8, 155.3, 152.6, 151.6, 148.0, 142.3, 134.5, 133.6, 132.1, 126.4, 126.3, 122.5, 120.4 (q, J_{C-F} = 255 Hz), 120.1, 108.3, 61.2, 61.1, 60.9, 56.1, 24.1, 23.1, 13.8 ppm; IR (KBr): ν_{max} 2935, 2850, 1725, 1586, 1484, 1251, 1161, 1082, 1005 cm^{-1}; HRMS (ESI) m/z: calcd for $C_{26}H_{27}F_3NO_6$ $[M + H]^+$ 506.1790, found: 506.1787.

2,6-Dimethyl-3-(ethoxycarbonyl)-5-(4-(trifluoromethyl)phenyl)-4-(3,4,5-trimethoxyphenyl)pyridine (8l). White solid; 235 mg, 48%; mp 147 °C; ^1H NMR (400 MHz, $CDCl_3$): δ 7.47 (d, J = 8 Hz, 2H), 7.35 (d, J = 7.3 Hz, 2H), 6.57 (s, 1H), 6.38 (s, 1H), 4.18–4.12 (m, 2H), 3.95 (s, 3H), 3.85 (s, 3H), 3.61 (s, 3H), 2.58 (s, 3H), 2.28 (s, 3H), 1.04 (t, J = 7.1 Hz, 3H) ppm; $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$): δ 168.7, 157.8, 155.4, 152.8, 151.6, 147.9, 142.3, 139.7, 133.6, 131.1, 128.8 (q, J_{C-C-F} = 32 Hz), 126.4, 126.2, 124.52, 124.49, 124.2 (q, J_{C-F} = 270 Hz), 122.4, 108.4, 61.3, 61.1, 61.0, 56.1, 24.2, 23.1, 13.8 ppm; IR (KBr): ν_{max} 2935, 1725, 1586, 1322, 1260, 1121, 1084, 1065, 1005 cm^{-1}; HRMS (ESI) m/z: calcd for $C_{26}H_{27}F_3NO_5$ $[M + H]^+$ 490.1841, found: 490.1841.

5-(4-Acetylphenyl)-2,6-dimethyl-3-(ethoxycarbonyl)-4-(3,4,5-trimethoxyphenyl)pyridine (8m). White solid; 208 mg, 45%; mp 121 °C; ^1H NMR (400 MHz, $CDCl_3$): δ 7.81 (d, J = 8 Hz, 2H), 7.34 (d, J = 7.3 Hz, 2H), 6.56 (s, 1H), 6.41 (s, 1H), 4.19–4.12 (m, 2H), 3.95 (s, 3H), 3.85 (s, 3H), 3.60 (s, 3H), 2.58 (s, 3H), 2.57 (s, 3H), 2.27 (s, 3H), 1.05 (t, J = 7.1 Hz, 3H) ppm; $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$): δ 198.0, 168.7, 157.8, 155.3, 152.8, 151.6, 148.0, 142.3, 141.1, 135.3, 133.5, 130.9, 127.7, 126.5, 126.3, 122.5, 108.4, 61.3, 61.11, 61.06, 56.1, 26.6, 24.3, 23.1, 13.8 ppm; IR (KBr): ν_{max} 2924, 2853, 1725, 1683, 1586, 1462, 1262, 1136, 1085, 1005 cm^{-1}; HRMS (ESI) m/z: calcd for $C_{27}H_{30}NO_6$ $[M + H]^+$ 464.2073, found: 464.2082.

5-(3-Acetylphenyl)-2,6-dimethyl-3-(ethoxycarbonyl)-4-(3,4,5-trimethoxyphenyl)pyridine (8n). White solid; 199 mg, 43%; mp 129 °C; ^1H NMR (400 MHz, $CDCl_3$): δ 7.84 (s, 1H), 7.80–7.77 (m, 1H), 7.46 (d, J = 6.9 Hz, 1H), 7.32 (dd, J = 7.7 Hz, J = 7.6 Hz 1H), 6.58 (s, 1H), 6.43 (s, 1H), 4.19–4.14 (m, 2H), 3.96 (s, 3H), 3.86 (s, 3H), 3.62 (s, 3H), 2.56 (s, 3H), 2.46 (s, 3H), 2.27 (s, 3H), 1.07 (t, J = 7.1 Hz, 3H) ppm; $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$): δ 198.1, 168.7, 157.8, 155.1, 152.6, 151.6, 148.0, 142.3, 136.4, 136.2, 135.4, 133.6, 131.2, 127.9, 126.6, 126.48, 126.45, 122.5, 108.4, 61.3, 61.1, 61.0, 56.1, 26.6, 24.2, 22.9, 13.8 ppm; IR (KBr): ν_{max} 2934, 2850, 1723, 1684, 1584, 1261, 1137, 1083, 1012 cm^{-1}; HRMS (ESI) m/z: calcd for $C_{27}H_{30}NO_6$ $[M + H]^+$ 464.2073, found: 464.2064.

2,6-Dimethyl-3-(ethoxycarbonyl)-5-(3-hydroxyphenyl)-4-(3,4,5-trimethoxyphenyl)pyridine (8o). Pale yellow solid; 175 mg, 40%; mp 169 °C; ^1H NMR (400 MHz, $CDCl_3$): δ 7.05 (s, 1H), 6.79–6.72 (m, 2H), 6.64 (d, J = 7.9 Hz, 1H), 6.53 (s, 1H), 6.48 (s, 1H), 4.14–4.08 (m, 2H), 3.94 (s, 3H), 3.83 (s, 3H), 3.61 (s, 3H), 2.55 (s, 3H), 2.27 (s, 3H), 1.01 (t, J = 7.1 Hz, 3H) ppm; $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$): δ 168.8, 157.4, 155.5, 154.9, 152.3, 151.7, 148.8, 142.3, 137.1, 133.3, 128.8, 127.6, 126.5, 122.9, 117.8, 114.2, 108.2, 61.3,

61.1, 61.0, 56.1, 23.8, 22.6, 13.7 ppm; IR (KBr): ν_{max} 3395, 2936, 2851, 1726, 1592, 1448, 1280, 1133, 1096 cm^{-1}; HRMS (ESI) *m/z*: calcd for $C_{25}H_{28}NO_6$ $[M + H]^+$ 438.1916, found: 438.1916.

2,6-Dimethyl-3-(ethoxycarbonyl)-5-(4-(hydroxymethyl)phenyl)-4-(3,4,5-trimethoxyphenyl)pyridine (8p). White solid; 203 mg, 45%; mp 152 °C; ^1H NMR (400 MHz, $CDCl_3$): δ 7.23–7.17 (m, 4H), 6.54 (s, 1H), 6.44 (s, 1H), 4.64 (s, 2H), 4.15–4.12 (m, 2H), 3.94 (s, 3H), 3.84 (s, 3H), 3.59 (s, 3H), 2.56 (s, 3H), 2.26 (s, 3H), 1.03 (t, J = 7.1 Hz, 3H) ppm; $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$): δ 168.7, 157.5, 155.2, 152.3, 151.7, 148.5, 142.2, 139.4, 134.9, 133.6, 130.8, 127.5, 126.4, 126.2, 122.7, 108.2, 64.9, 61.2, 61.1, 60.9, 56.1, 24.2, 22.9, 13.8 ppm; IR (KBr): ν_{max} 3395, 2980, 2936, 2850, 1725, 1588, 1488, 1267, 1136, 1094 cm^{-1}; HRMS (ESI) *m/z*: calcd for $C_{26}H_{30}NO_6$ $[M + H]^+$ 452.2073, found: 452.2073.

2,6-Dimethyl-3-(ethoxycarbonyl)-5-(3-pyridinyl)-4-(3,4,5-trimethoxyphenyl)pyridine (8q). White solid; 177 mg, 42%; mp 107 °C; ^1H NMR (400 MHz, $CDCl_3$): δ 8.48 (s, 1H), 8.42 (dd, J = 4.8 Hz, J = 1.5 Hz, 1H), 7.53 (d, J = 7.6 Hz, 1H), 7.15 (dd, J = 7.6 Hz, J = 4.9 Hz, 1H), 6.58 (s, 1H), 6.43 (s, 1H), 4.18–4.09 (m, 2H), 3.96 (s, 3H), 3.86 (s, 3H), 3.64 (s, 3H), 2.57 (s, 3H), 2.30 (s, 3H), 1.03 (t, J = 7.1 Hz, 3H) ppm; $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$): δ 168.6, 157.9, 155.5, 152.9, 151.8, 151.3, 147.9, 147.7, 142.3, 138.0, 133.9, 131.7, 126.4, 123.8, 122.6, 122.5, 108.4, 61.3, 61.1, 61.0, 56.1, 24.3, 23.1, 13.8 ppm; IR (KBr): ν_{max} 2978, 2936, 2848, 1725, 1586, 1465, 1385, 1267, 1139, 1095, 1003 cm^{-1}; HRMS (ESI) *m/z*: calcd for $C_{24}H_{27}N_2O_5$ $[M + H]^+$ 423.1920, found: 423.1918.

2,6-Dimethyl-3-(ethoxycarbonyl)-5-(3-hydroxy-4-methoxyphenyl)-4-(3,4,5-trimethoxyphenyl)pyridine (8r). White solid; 210 mg, 45%; mp 148 °C; ^1H NMR (400 MHz, $CDCl_3$): δ 6.85 (s, 1H), 6.68–6.65 (m, 2H), 6.51 (s, 1H), 6.49 (s, 1H), 5.61 (s, 1H), 4.14–4.09 (m, 2H), 3.93 (s, 3H), 3.84 (s, 3H), 3.83 (s, 3H), 3.62 (s, 3H), 2.57 (s, 3H), 2.32 (s, 3H), 1.01 (t, J = 7.1 Hz, 3H) ppm; $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$): δ 168.8, 157.5, 155.2, 152.1, 151.9, 148.8, 145.6, 144.8, 142.2, 133.7, 128.9, 127.4, 126.3, 122.7, 122.6, 117.0, 109.9, 108.1, 61.1, 61.0, 60.9, 56.1, 55.8, 24.2, 23.0, 13.7 ppm; IR (KBr): ν_{max} 3410, 2935, 2838, 1721, 1586, 1488, 1250, 1087, 1015 cm^{-1}; HRMS (ESI) *m/z*: calcd for $C_{26}H_{30}NO_7$ $[M + H]^+$ 468.2022, found: 468.2013.

4-(3-(Benzyloxy)-4-methoxyphenyl)-2,6-dimethyl-3-(ethoxycarbonyl)-5-(3,4,5-trimethoxyphenyl)nicotinate (8s). Yellow solid; 250 mg, 45%; mp 120 °C; ^1H NMR (400 MHz, $CDCl_3$): δ 7.46 (d, J = 8.6 Hz, 2H), 7.38 (dd, J = 8.6 Hz, J = 7.1 Hz, 2H), 7.32 (dd, J = 7.2 Hz, J = 7.0 Hz 1H), 6.98 (s, 1H), 6.83 (s, 1H), 6.49 (s, 3H), 4.04–4.02 (m, 2H), 3.96 (s, 3H), 3.81 (s, 3H), 3.67 (s, 6H), 2.57 (s, 3H), 2.34 (s, 3H), 1.01 (t, J = 7.2 Hz, 3H) ppm; $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$): δ 168.8, 157.9, 154.7, 152.7, 149.4, 148.8, 147.1, 136.9, 136.7, 135.8, 133.6, 129.3, 128.6, 127.9, 127.4, 126.7, 122.7, 114.8, 113.4, 106.9, 71.1, 61.3, 60.9, 56.2, 56.0, 24.3, 22.9, 13.9 ppm; IR (KBr): ν_{max} 2936, 2840, 1715, 1586, 1501, 1255, 1122, 1079 cm^{-1}; HRMS (ESI) *m/z*: calcd for $C_{33}H_{36}NO_7$ $[M + H]^+$ 558.2492, found: 558.2476.

2,6-Dimethyl-3-(ethoxycarbonyl)-4-(3-hydroxy-4-methoxyphenyl)-5-(3,4,5-trimethoxyphenyl)pyridine (8t). White solid; 350 mg, 75%; mp > 200 °C; ^1H NMR (400 MHz, $CDCl_3$): δ 6.94 (s, 1H), 6.82 (s, 1H), 6.51–6.48 (m, 3H), 5.78 (br s, 1H), 4.18–4.13 (m, 2H), 3.96 (s, 3H), 3.80 (s, 3H), 3.67 (s, 6H), 2.56 (s, 3H), 2.34 (s, 3H), 1.12 (t, J = 7.1 Hz, 3H) ppm; $^{13}C\{^1H\}$ NMR (100 MHz, $CDCl_3$): δ 168.7, 157.8, 154.6, 152.6, 148.8, 146.5, 144.7, 136.8, 136.0, 132.6, 129.9, 126.8, 122.7, 115.6, 112.2, 106.9, 61.3, 60.9, 56.1, 56.0, 24.2, 22.8, 13.9 ppm; IR (KBr): ν_{max} 3453, 2961, 2935, 2840, 1725, 1587, 1499, 1260, 1125, 1083, 1018 cm^{-1}; HRMS (ESI) *m/z*: calcd for $C_{26}H_{30}NO_7$ $[M + H]^+$ 468.2022, found: 468.2015.

Biology

Cell-based screening assay. Synthesized penta-substituted/functionalized-3,4-diarylated pyridine compounds **8a–8t** used in this study were dissolved in 100% DMSO (cell culture grade, Himedia). The compounds were serially diluted in Dulbecco's Modified Eagle (DMEM) cell culture medium to maintain the DMSO concentration less than 0.1% for analyzing its cell proliferation inhibiting potency in HeLa cell line. Briefly, 5×10^3 cells were seeded in 96 well plate.[15b] After 16 h compounds (5 μM) were added and incubated with cells for 24 h. Subsequently, cells were fixed with TCA and processed for sulforhodamine B (SRB) assay.[39]

Effects of diarylated pyridine compounds on *in vitro* tubulin assembly. Tubulin was purified from goat brain using 1 M glutamate as described earlier.[40,41] Tubulin concentration was determined by Bradford method.[42] Purified tubulin (12 μM) in PEM buffer (25 mM PIPES pH 6.8, 3 mM $MgCl_2$ and 1 mM EGTA) was incubated in the absence and presence of 20 μM of compounds (**8b**, **8f**, **8j** and **8p**) for 10 min on ice and then, DMSO (final concentration 10%) and 1 mM GTP was added to the reaction mixtures. Subsequently, the assembly kinetics was monitored at 37 °C by 90° light scattering (350 nm) using Spectramax M2^e. The extent of inhibition of polymerization was measured after 30 min of assembly. The light scattering data of only compounds (20 μM) were also recorded and subtracted from their respective data set. Three independent set of experiments were performed.

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