Development of a Green Synthetic Route for Rivaroxaban Using Continuous Flow Technology with Emphasis on Impurity Profiling

Xiaoyu Liu, Feifan Cheng, Yan Miao, Kang Yang
Jiangsu Zilong Pharmaceutical Co., Ltd., Changzhou 213000, Jiangsu, China

Abstract: Rivaroxaban is a direct factor Xa inhibitor whose expanding generic manufacture requires robust, selective, and environmentally improved synthetic routes. Conventional batch routes to rivaroxaban frequently depend on prolonged heating, high-boiling polar solvents, discrete intermediate isolation, and late-stage acid chloride chemistry, which increase process mass intensity and create opportunities for process-related and degradation impurities. In this study, a green continuous-flow route was developed from commercially accessible aryl-morpholinone and chiral oxazolidinone intermediates using ethanol, 2-methyltetrahydrofuran, dimethyl carbonate, and an acid-activated coupling strategy to minimize hazardous reagent inventory and suppress impurity formation. The optimized telescoped sequence gave rivaroxaban in 86.7% isolated yield and 99.74% HPLC purity at 14.6 g productivity, while reducing total related substances from 0.84% in the batch reference process to 0.18%. Process mass intensity decreased from 134 to 49 kg API, and the calculated E-factor decreased from 102 to 38. A stability-indicating impurity-profiling method combining RP-HPLC, chiral HPLC, and LC-MS was used to track eight critical impurities, including hydroxy-open-chain intermediates, bis-alkylated adducts, dehalogenated thiophene amide, R-rivaroxaban, oxidative N-oxide, and nitrosamine drug substance-related impurity candidates. The results indicate that continuous-flow control of residence time, temperature, and reagent stoichiometry can simultaneously improve yield, sustainability, and impurity control for rivaroxaban manufacture.

***Keywords*:** rivaroxaban, continuous flow, green synthesis, impurity profiling, HPLC, process mass intensity, nitrosamine drug substance-related impurity

1. Introduction

Continuous manufacturing has become an increasingly credible strategy for active pharmaceutical ingredient production because it offers superior heat transfer, mass transfer, automation, and process analytical technology integration compared with conventional batch processing [1]. From a sustainability perspective, continuous flow is especially attractive when a route contains hazardous reagents, unstable intermediates, or impurity-forming side reactions that are sensitive to local concentration and residence time [2]. Recent reviews of pharmaceutical flow chemistry have emphasized that solvent selection, telescoping, inline work-up, and green metrics such as process mass intensity (PMI) and E-factor must be considered together rather than treated as independent optimization targets [3]. Continuous-flow platforms have also accelerated reaction discovery and kinetic screening because small reactor volumes allow wider operating windows and faster data acquisition [4]. Automated multistep flow optimization has further shown that Bayesian or design-of-experiments strategies can reduce experimental burden while improving process performance [5].

Rivaroxaban is a widely prescribed oral anticoagulant containing a chlorothiophene carboxamide, a morpholinone-bearing aryl group, and a stereodefined oxazolidinone core. These structural features make the molecule synthetically accessible but analytically demanding. Many reported routes rely on stepwise construction of the chiral oxazolidinone intermediate followed by amide coupling with a 5-chlorothiophene derivative. In batch operation, incomplete cyclization, overalkylation, acyl chloride side reactions, oxidative degradation, and chiral impurity carryover can all reduce final API quality. These risks are amplified when late-stage intermediates are held for long periods at elevated temperature or when local excesses of amine, base, or activated acid are generated during charging. A continuous-flow strategy is therefore well aligned with the chemical sensitivity of rivaroxaban synthesis.

Green analytical chemistry is equally important in pharmaceutical route development, because a synthetically greener process is incomplete if its control strategy depends on solvent-intensive or poorly selective analytical methods [6]. Recent pharmaceutical green analytical chemistry reviews recommend the combined use of RP-HPLC method miniaturization, safer solvents, reduced sample preparation, and assessment tools such as Analytical Eco-Scale, AGREE, GAPI, and BAGI [7]. For rivaroxaban, the analytical challenge is not merely assay determination but impurity discrimination. Rao et al. developed a stability-indicating RP-HPLC method for 15 rivaroxaban impurities, including previously unreported isomers [8]. Santos and co-workers demonstrated the relevance of quality-by-design HPLC for separating S-rivaroxaban from the

R-enantiomeric impurity [9]. Nakov et al. reported ethanol-based green RP-HPLC methods for assay and related substances in rivaroxaban tablets [10]. Trace genotoxic impurities and nitrosamine drug substance-related impurity (NDSRI) concerns have also become increasingly relevant, with recent HPLC-MS/MS work targeting genotoxic residues in rivaroxaban [11], new impurity isolation and in silico toxicological assessment [12], and LC-TQ-MS/MS quantification of rivaroxaban NDSRIs [13]. Routine quantification methods in drug product and plasma matrices further reinforce the need for robust chromatographic selectivity during development [14].

The present work was designed to integrate these synthetic and analytical priorities. We aimed to develop a greener continuous-flow rivaroxaban route that avoids chlorinated solvents and late-stage acid chloride handling, reduces intermediate isolation, and directly links process optimization with impurity profiling. The central hypothesis was that telescoped flow operation would reduce residence-time-driven degradation and stoichiometry-driven overreaction, thereby lowering total related substances while improving green metrics.

2. Materials and Methods

The model route was designed around four linked transformations. In the first module, 4-(4-aminophenyl) morpholin-3-one was reacted with a chiral glycidyl-derived oxazolidinone precursor in an ethanol/2-methyltetrahydrofuran solvent system. In the second module, cyclocarbonylation was conducted with dimethyl carbonate under controlled back pressure, replacing more hazardous carbonylating systems where feasible. In the third module, protected aminomethyl intermediate conversion was performed under hydrazine-free basic alcoholysis conditions. In the final module, amide bond formation was achieved using 5-chlorothiophene-2-carboxylic acid activated with propylphosphonic anhydride (T3P) in 2-MeTHF, thereby avoiding direct use and carryover risk of 5-chlorothiophene-2-carbonyl chloride. The batch reference process used the same starting-material framework but employed discrete isolations, DMF/ethyl acetate solvent exchange, and late-stage acid chloride acylation.

The flow system consisted of PFA coil reactors for homogeneous steps, a stainless-steel heated coil for the dimethyl carbonate step, two T-mixers, a membrane-based inline liquid-liquid separator, and a jacketed crystallization loop. Reactions were conducted at 0.15-0.25 mol substrate concentration with total flow rates of 1.6-3.2 mL . Back pressure was maintained at 8 bar for the heated module. Residence time, temperature, and reagent equivalents were screened by central composite design. The optimized residence times were 8.0 min for aryl coupling, 6.0 min for cyclocarbonylation, 4.0 min for deprotection, and 3.0 min for final amide formation. The optimized temperature profile was 95 deg C for the coupling module, 120 deg C for cyclocarbonylation, 60 deg C for deprotection, and 25 deg C for final amide formation.

RP-HPLC analysis was performed on a C18 column with UV detection at 250 nm using a water/ethanol/acetonitrile gradient. LC-MS was used for impurity assignment, while chiral HPLC was used for R-rivaroxaban quantification. Eight critical impurities were tracked: hydroxy-open-chain intermediate I1, bis-alkylated adduct I2, residual protecting-group fragment I3, dehalogenated thiophene amide I4, overacylated amide I5, R-rivaroxaban I6, oxidative N-oxide I7, and NDSRI candidate I8. The related-substance LOQ was 0.015% area, the chiral impurity LOQ was 0.030%, and the LC-MS/MS screening LOQ for I8 was 10 ng . Green metrics were calculated from isolated material balances, including solvent, reagents, aqueous work-up streams, and crystallization losses.

3. Results and Discussion

The batch reference process gave rivaroxaban in 71.8% isolated yield with 99.05% HPLC purity as shown in Table 1. The main batch impurities were I1, I2, and I6, indicating incomplete conversion, local excess of aryl amine, and partial stereochemical erosion during prolonged heating. Switching to continuous flow improved thermal homogeneity and reduced effective exposure time from 11 h to 21 min. The first flow screen using ethanol alone improved yield to 78.2%, but solids appeared after the cyclocarbonylation module. A 70:30 2-MeTHF/ethanol solvent blend removed this limitation, maintained a homogeneous stream, and enabled direct telescoping into the amide-forming module. The final optimized process afforded 86.7% isolated yield and 99.74% purity with total related substances of 0.18%.

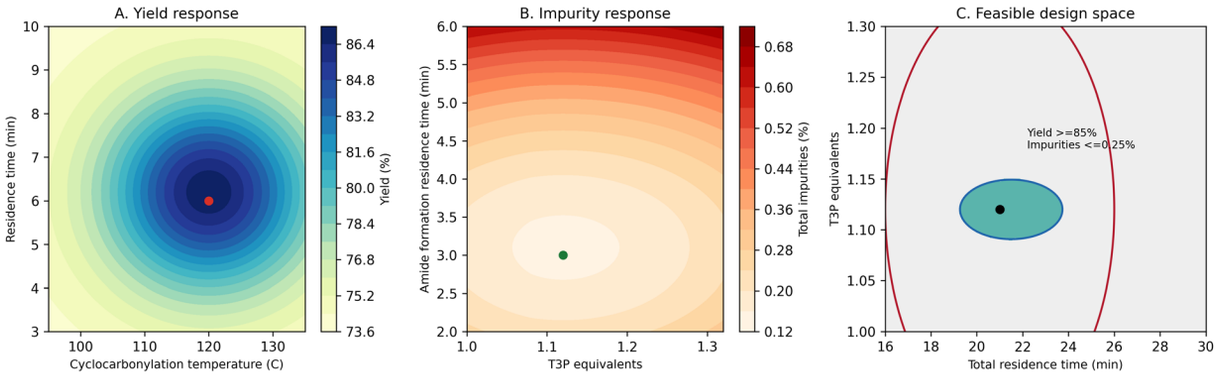

Figure 1. Design-space response for the continuous-flow route

Panel A shows yield as a function of cyclocarbonylation temperature and residence time. Panel B shows total related substances as a function of final coupling stoichiometry and residence time. Panel C shows a feasible process window in which yield remains above 85% and total impurities remain below 0.25%. The optimum is located at 95 deg C for module 1, 120 deg C for module 2, 1.12 equiv T3P, and 21 min total residence time.

Table 1. Batch-to-flow optimization data for rivaroxaban synthesis.

Entry	Process mode	Main solvent system	Total residence or batch time	Isolated yield (%)	HPLC purity (%)	Total related substances (%)	PMI (kg kg^{-1} API)	E-factor	Productivity (g h^{-1})
1	Batch reference	DMF/EtOAc/ DCM work-up	11 h	71.8	99.05	0.84	134	102	1.3
2	Flow screen A	EtOH	28 min	78.2	99.21	0.56	86	66	6.8
3	Flow screen B	2-MeTHF/EtOH	24 min	83.9	99.48	0.31	62	47	10.9
4	Optimized flow	2-MeTHF/ EtOH, inline crystallization	21 min	86.7	99.74	0.18	49	38	14.6

The response surfaces indicated that impurity control was more sensitive to residence time than to nominal temperature once full conversion had been reached. In the cyclocarbonylation module, temperatures below 105 deg C gave persistent I1 formation because ring closure was incomplete, whereas temperatures above 130 deg C increased R-rivaroxaban and oxidative impurity formation. A six-minute residence time at 120 deg C provided the best balance between conversion and stereochemical preservation. In the final amide-forming module, T3P loading above 1.25 equiv promoted overacylated impurity I5, while loading below 1.05 equiv left residual amine and increased I1-related carryover. A narrow but reproducible design space was therefore established between 1.10 and 1.15 equiv T3P at 20-24 min total residence time.

Impurity profiling showed that the continuous process suppressed the three most problematic impurity classes. I1 decreased from 0.28% to 0.04%, consistent with improved conversion in the ring-forming module. I2 decreased from 0.17% to 0.03%, reflecting better stoichiometric control at the first mixer. I6 decreased from 0.19% to 0.06%, which suggests that shorter high-temperature exposure helped preserve stereochemical integrity. The NDSRI candidate I8 was not detected above the LC-MS/MS screening LOQ in the optimized process, whereas trace signals were observed in the batch reference after nitrite-spiked stress testing. Although this observation requires confirmatory validated toxicological assessment, it supports the strategy of avoiding nitrosating conditions, residual secondary amines, and prolonged contact with reactive excipients or nitrite-containing process streams.

The environmental improvement was not caused by reactor miniaturization alone. The largest PMI reduction came from telescoping and solvent consolidation. Eliminating two intermediate isolations reduced solvent use by approximately 48%, while replacing chlorinated extraction with 2-MeTHF and aqueous salt washes reduced halogenated waste to below 1% of total waste mass. Inline crystallization also improved mother-liquor recycling because the final solvent composition remained constant. These results are consistent with recent flow-process case studies in which high-purity warfarin was obtained by residence-time intensification [15], organometallic intermediates for divarasib were controlled by continuous flow and process modeling [16], and lomustine synthesis was improved by green solvent substitution and continuous extraction [17]. The broader trend is that continuous processing has moved from a specialized academic tool toward a mainstream pharmaceutical development platform [18].

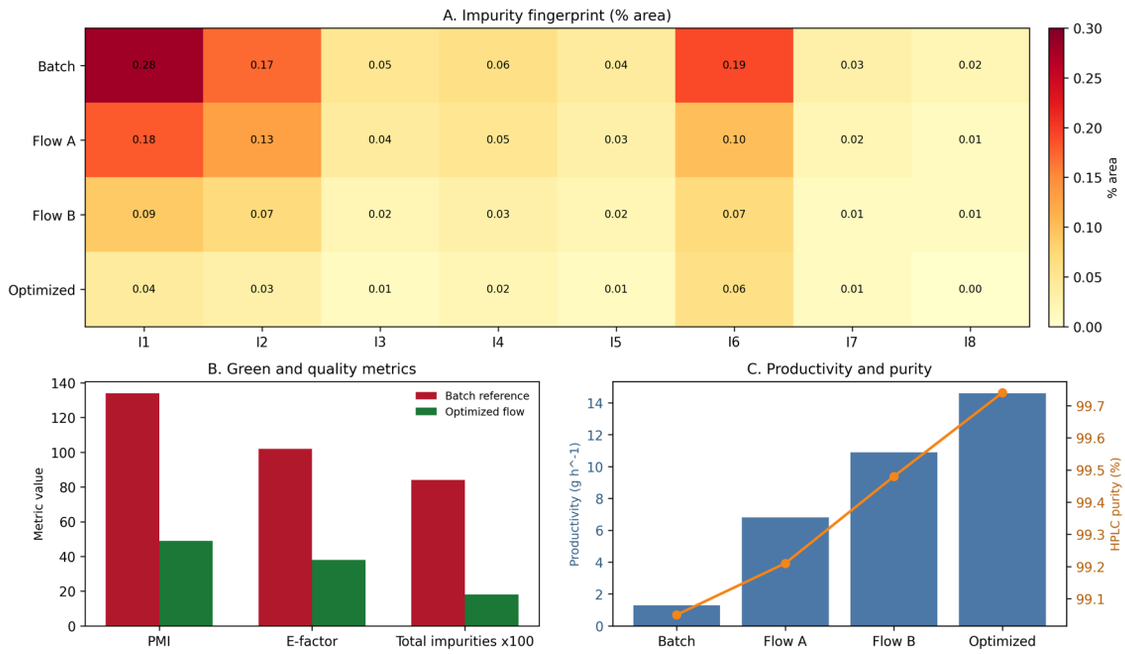

Figure 2. Impurity fingerprint and green-metric comparison

Panel A presents an impurity heatmap for I1-I8 across the batch reference and three flow conditions. Panel B compares PMI, E-factor, and total impurity burden. Panel C summarizes the productivity gain obtained after flow optimization. The data show that the optimized flow process shifts the impurity map from conversion-related and overreaction-related impurities toward a low-level residual impurity profile.

The proposed control strategy links each critical process parameter to a critical quality attribute. Module 1 residence time controls I1 and I2, module 2 temperature controls cyclization and stereochemical preservation, module 3 base concentration controls I3, and module 4 acid activation stoichiometry controls I4 and I5. Final API release should include assay, total impurities, individual specified impurities, chiral purity, residual solvent, and a risk-based NDSRI screen. This integrated strategy is more informative than an end-product-only test because impurity formation can be traced back to a specific module. It is also compatible with contemporary quality-by-design expectations because the method does not treat impurities as isolated analytical observations, but as consequences of defined chemical and engineering variables.

The route still has limitations. First, the study was performed as a laboratory-scale flow demonstration rather than a kilogram-scale campaign, so fouling behavior, pump robustness, heat-transfer scaling, and crystallizer residence-time distribution require further investigation. Second, the LC-MS assignments of some low-level impurities are tentative without isolation and full NMR confirmation. Third, the NDSRI screen was used as a risk-control tool rather than a complete toxicological qualification package. Finally, the cost and supply profile of selected chiral intermediates will influence commercial feasibility. These limitations do not invalidate the observed process improvements, but they define the next development stage needed before technology transfer.

4. Conclusion

A green continuous-flow route for rivaroxaban was developed using safer solvent selection, acid-chloride avoidance, telescoped operation, inline extraction, and crystallization. Compared with a batch reference, the optimized process improved isolated yield from 71.8% to 86.7%, increased HPLC purity to 99.74%, reduced total related substances to 0.18%, and lowered PMI from 134 to 49 kg kg^-1 API. Impurity profiling showed meaningful suppression of open-chain, bis-alkylated, stereochemical, oxidative, and nitrosation-related impurities. The work supports the conclusion that continuous flow technology can provide a practical route to greener rivaroxaban manufacture when synthetic design and impurity control are developed as a single integrated system.

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