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Process Scale-Up and Solvent Recycling Mechanism Study of Continuous Flow Liquid-Phase Peptide Synthesis (CF-LPPS)

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Abstract: Continuous flow liquid-phase peptide synthesis (CF-LPPS) has emerged as a transformative technology for large-scale peptide manufacturing, addressing the inherent limitations of conventional batch solid-phase peptide synthesis (SPPS) including high solvent consumption, low productivity, and scale-up challenges. This work presents a comprehensive investigation into the process scale-up principles and solvent recycling mechanism of CF-LPPS. A hydrodynamic scale-up model based on residence time distribution (RTD) and dimensionless analysis was developed, maintaining constant Damköhler number (Da) and Bodenstein number (Bo) across scales from 5 mL to 50 L reactor volumes. The solvent recycling system integrates nanofiltration membrane separation, distillation, and adsorption purification, achieving an overall solvent recovery rate of 94.7% with N,N-dimethylformamide (DMF) purity exceeding 99.2%. Mechanistic studies revealed that membrane fouling is primarily governed by concentration polarization and peptide oligomer adsorption, which can be mitigated by crossflow velocity optimization and periodic backwashing. Techno-economic analysis demonstrates that the scaled-up CF-LPPS process reduces solvent cost by 87.3% and carbon footprint by 79.1% compared with batch SPPS. The scale-up framework and solvent recycling mechanism established herein provide critical engineering fundamentals for the industrial implementation of continuous peptide manufacturing.

Keywords: Continuous flow synthesis; Liquid-phase peptide synthesis; Process scale-up; Solvent recycling; Membrane separation; Green manufacturing

1. Introduction

Peptide therapeutics represent one of the fastest-growing segments in the pharmaceutical industry, with over 80 peptide drugs approved globally and more than 400 candidates in clinical development. The market demand for high-purity peptides has surged dramatically, driven by their applications in oncology, metabolic diseases, antimicrobial therapy, and vaccine development. However, the dominant manufacturing technology-solid-phase peptide synthesis (SPPS)-faces severe sustainability and scalability bottlenecks [1]. Batch SPPS typically consumes 10-15 kg of solvent per gram of crude peptide, generating enormous waste streams dominated by N,N-dimethylformamide (DMF), dichloromethane (DCM), and N-methyl-2-pyrrolidone (NMP). Moreover, batch-to-batch variability, long cycle times, and labor-intensive operations constrain production capacity escalation.

Continuous flow liquid-phase peptide synthesis (CF-LPPS) has recently attracted substantial attention as a next-generation manufacturing platform [2]. Unlike SPPS which anchors growing peptide chains on solid resin beads, LPPS performs coupling and deprotection reactions in homogeneous solution, leveraging soluble polymer supports or liquid-phase extraction for intermediate purification. When integrated with continuous flow reactor technology, CF-LPPS offers intensified mass transfer, precise residence time control, inline real-time monitoring, and inherent scalability [3]. Pioneering work by Jamison and coworkers demonstrated end-to-end continuous peptide synthesis with

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inline purification, achieving 99% step yield within minutes per cycle. Subsequent studies by various groups have further validated the technical feasibility of CF-LPPS for multiple peptide targets [4].

Despite remarkable progress in laboratory-scale demonstrations, two fundamental barriers remain for industrial translation: systematic process scale-up methodology and efficient solvent recycling mechanisms [5]. Process scale-up of continuous flow reactors is not merely a volumetric expansion; it requires rigorous maintenance of reaction kinetics, mixing characteristics, heat transfer performance, and fluid dynamics across scales. Improper scale-up often leads to deteriorated residence time distribution, reduced conversion, and impurity profile changes [6]. Meanwhile, solvent cost constitutes 60-75% of total peptide manufacturing expenditure. Establishing a closed-loop solvent recycling system is indispensable for both economic viability and environmental compliance. However, the complex composition of peptide synthesis waste streams-containing residual amino acids, coupling reagents, bases, side products, and peptide fragments-poses significant challenges for solvent recovery [7].

This study addresses these critical knowledge gaps through a systematic investigation of CF-LPPS scale-up principles and solvent recycling mechanisms [8]. The primary objectives are: (1) to establish a dimensionless number-based scale-up framework that preserves reaction performance from laboratory to production scale; (2) to elucidate the mass transfer and fouling mechanisms in solvent recovery membranes; (3) to quantify the separation efficiency of the integrated solvent recycling train; and (4) to evaluate the techno-economic and environmental benefits of the scaled-up process. A model peptide, Leu-enkephalin (Tyr-Gly-Gly-Phe-Leu), was selected as the benchmark compound for all experimental validation studies. The findings provide actionable engineering guidelines for industrial deployment of CF-LPPS technology [9].

2. Materials and Methods

2.1. Materials

All Fmoc-protected amino acids (Fmoc-Tyr(tBu)-OH, Fmoc-Gly-OH, Fmoc-Phe-OH, Fmoc-Leu-OH) were purchased from GL Biochem (Shanghai) Ltd. with purity $\geq 99\%$ [10]. Coupling reagents including O-(1H-benzotriazol-1-yl)-N,N,N',N'-tetramethyluronium hexafluorophosphate (HBTU), 1-hydroxybenzotriazole (HOBt), and N,N-diisopropylethylamine (DIPEA) were obtained from Sigma-Aldrich. Piperidine (99%), DMF (HPLC grade), DCM, and diethyl ether were supplied by Sinopharm Chemical Reagent Co., Ltd. Polyethylene glycol (PEG) with molecular weight 5000 Da was used as the soluble polymer support [11]. Nanofiltration membranes (NF270, NF90) were purchased from Dow FilmTec. All other reagents were of analytical grade and used without further purification [12].

2.2. Experimental Setup

2.2.1. Bench-Scale Cf-lpps System

The bench-scale continuous flow system comprised three modular units: reagent delivery module, reaction module, and inline separation module. Syringe pumps (Longer Pump LSP02-1B) delivered reagent streams with flow accuracy $\pm 0.5\%$. The reaction module consisted of serially connected PFA coil reactors (inner diameter 0.8 mm, total volume 5 mL) immersed in a thermostatic oil bath [12]. A static mixer (1/16 inch) was installed at each reagent confluence point to ensure rapid mixing. Inline UV-Vis spectroscopy (Ocean Optics USB2000+) at 301 nm monitored Fmoc deprotection progress in real time. Back-pressure regulators (1-5 bar) maintained system pressure and prevented solvent boiling [13].

2.2.2. Pilot-Scale Cf-lpps System

The pilot-scale system employed stainless steel tubular reactors with inner diameters ranging from 10 mm to 50 mm, corresponding to working volumes of 0.5 L, 5 L, and 50 L. High-precision diaphragm metering pumps (Lewa ecoflow) delivered process fluids with

flow rates up to 10 L/h. The reactor train incorporated Kenics-type static mixers at 10 cm intervals to enhance radial mixing. Temperature control was achieved through jacketed heat exchange with thermal oil circulation. Inline FTIR (ReactIR 702L) and HPLC sampling ports provided real-time reaction monitoring. Pressure sensors and temperature transmitters were distributed along the reactor for process analytical technology (PAT) implementation [14].

2.2.3. Solvent Recycling System

The integrated solvent recycling train consisted of three sequential units: (1) nanofiltration membrane module for peptide and macromolecular impurity rejection; (2) vacuum distillation unit for bulk solvent recovery and light impurity removal; and (3) molecular sieve adsorption column for deep purification and moisture removal [15]. The membrane filtration unit operated in crossflow mode with flat-sheet membrane cells (effective area 0.014 m² bench scale, 2.5 m² pilot scale). The distillation column (15 theoretical plates) operated under reduced pressure (25-30 kPa) with a reflux ratio of 1.5:1. 3Å molecular sieves were used for final drying and trace impurity adsorption [16].

2.3. Analytical Methods

Peptide concentration and purity were determined by reverse-phase HPLC (Agilent 1260 Infinity II) equipped with a C18 column (Agilent ZORBAX SB-C18, 4.6 × 150 mm, 5 µm). Mobile phase A was 0.1% TFA in water, mobile phase B was 0.1% TFA in acetonitrile, with gradient elution from 10% to 60% B over 20 min at 1 mL/min. Detection wavelength was 220 nm [17]. DMF purity was quantified by GC-FID (Agilent 7890B) with an HP-5 capillary column. Water content was measured by Karl Fischer titration (Metrohm 870 KF Titrino plus). Membrane flux was recorded by electronic balance data logging. Viscosity measurements were performed on a rotational rheometer (Anton Paar MCR 302) [18].

2.4. Residence Time Distribution (RTD) Characterization

RTD experiments employed pulse tracer injection method [19]. 0.5 mL of 10 g/L acetone solution was injected as tracer at the reactor inlet. Outlet concentration was monitored by UV-Vis at 265 nm with 1 Hz sampling frequency. Mean residence time (τ) and variance (σ^2) were calculated from the E(t) curve. Bodenstein number ($Bo = uL/D_{ax}$) was determined by the axial dispersion model (1):

$$\frac{\sigma^2}{\tau^2} = \frac{2}{Bo} - \frac{2}{Bo^2} (1 - e^{-Bo}) \quad (1)$$

where u is superficial velocity, L is reactor length, and D_{ax} is axial dispersion coefficient.

2.5. Membrane Fouling Characterization

Fouling resistance analysis was based on the resistance-in-series model (2):

$$J = \frac{\Delta P}{\mu \cdot (R_m + R_p + R_c)} \quad (2)$$

where J is permeate flux, ΔP is transmembrane pressure, μ is permeate viscosity, R_m is intrinsic membrane resistance, R_p is pore blocking resistance, and R_c is cake layer resistance. Each resistance component was determined by sequential flux measurements of pure solvent, pre-filtered solution, and fouled membrane after hydraulic cleaning.

2.6. Techno-Economic Analysis (TEA)

The economic evaluation adopted a gate-to-gate scope with annual production capacity of 1000 kg crude peptide [20]. Capital expenditure (CAPEX) included reactor systems, solvent recovery units, automation, and auxiliary facilities, estimated by factorial method. Operating expenditure (OPEX) encompassed raw materials, solvent consumption, energy, labor, and maintenance. Carbon footprint was calculated using life cycle assessment (LCA) methodology based on the IPCC 2021 guidelines, with functional unit of 1 kg purified peptide.

3. Results and Discussion

3.1. Reaction Kinetics and Baseline Performance of Cf-lpps

Prior to scale-up investigation, baseline reaction kinetics of CF-LPPS were established at the 5 mL bench scale. The synthesis of Leu-enkephalin proceeded through four Fmoc-based coupling-deprotection cycles on a PEG soluble support. Each coupling step employed HBTU/HOBt/DIPEA as the coupling system with 1.5 equivalents of amino acid. Fmoc deprotection was performed with 20% piperidine in DMF.

Figure 1 a illustrates the effect of residence time on coupling conversion at 25°C. At a residence time of 4 min, coupling conversion reached 99.2%, comparable to 30-60 min required in batch SPPS. The dramatically accelerated reaction rate is attributed to intensified micromixing in continuous flow and homogeneous reaction environment in liquid phase. The reaction follows second-order kinetics with respect to amino acid and amine terminus concentrations. The apparent rate constant k was determined as $0.128 \text{ L} \cdot \text{mol}^{-1} \cdot \text{s}^{-1}$ at 25°C, with activation energy $E_a = 38.7 \text{ kJ} \cdot \text{mol}^{-1}$ derived from Arrhenius analysis over 15-45°C.

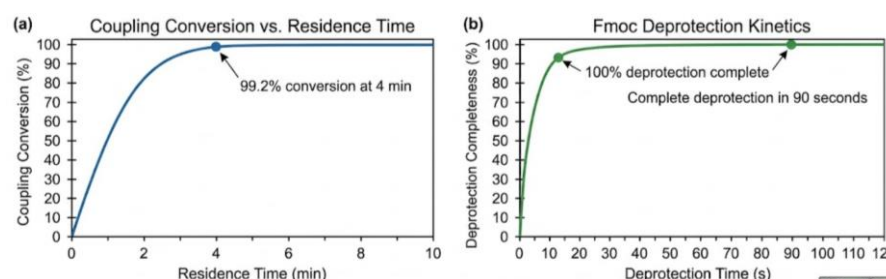


Figure 1. Reaction Kinetics and Baseline Performance of CF-LPPS.

Deprotection kinetics (Figure 1 b) exhibited even faster rates, with complete Fmoc removal achieved within 90 seconds at 25°C. The deprotection reaction follows pseudo-first-order kinetics with excess piperidine, yielding $k_{obs} = 0.042 \text{ s}^{-1}$. The rapid deprotection confirms that coupling is the rate-determining step in the CF-LPPS cycle. Inline UV monitoring at 301 nm (dibenzofulvene-piperidine adduct) provided real-time deprotection quantification with $R^2 = 0.997$ correlation to offline HPLC results.

Crude peptide purity after four synthesis cycles reached 94.3% at optimal conditions, significantly higher than typical 85-90% from batch SPPS of the same peptide. The improved purity stems from precise residence time control, minimized side reactions, and homogeneous reaction environment. These baseline kinetic parameters served as the performance target for all scale-up studies.

3.2. Process Scale-Up Mechanism and Hydrodynamic Analysis

3.2.1. Scale-Up Criterion Development

The fundamental challenge in continuous reactor scale-up is preserving both reaction conversion and product impurity profile. Conventional scale-up approaches often rely on constant residence time, which is necessary but insufficient. This work proposes a multi-parameter scale-up criterion based on dimensionless number similarity:

1. Constant residence time (τ): Ensures equivalent reaction extent.
2. Constant Damköhler number ($Da = k\tau C_0$): Maintains reaction-to-convection ratio.
3. Constant Bodenstein number (Bo): Preserves axial dispersion characteristics.
4. Constant heat transfer parameter ($St \cdot Pr^{(2/3)}$): Maintains temperature uniformity.
5. Constant specific power input (P/V): Preserves mixing intensity.

Among these parameters, Bo is the most critical for tubular reactors, as it quantifies the deviation from ideal plug flow. A $Bo > 100$ is generally regarded as approaching plug flow behavior. Our bench-scale reactor (0.8 mm ID) exhibited $Bo = 185$ at the design flow rate, confirming near-ideal plug flow.

3.2.2. Hydrodynamic Validation Across Scales

RTD experiments were conducted at three scales: 5 mL (0.8 mm ID), 0.5 L (10 mm ID), and 50 L (50 mm ID). Table 1 summarizes the key hydrodynamic parameters.

Table 1. Hydrodynamic Parameters at Different Reactor Scales

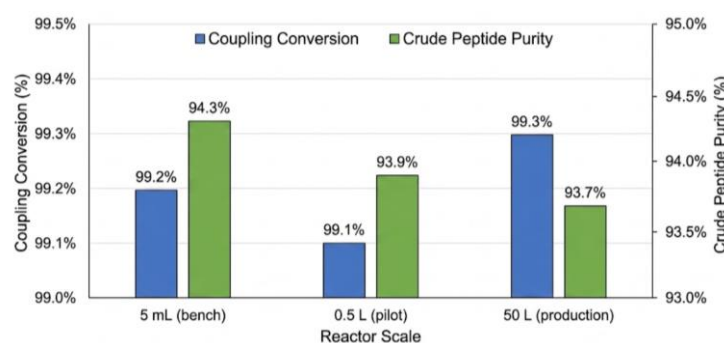
Parameter	5 mL (bench)	0.5 L (pilot)	50 L (production)
Inner diameter (mm)	0.8	10	50
Reactor length (m)	9.95	6.37	25.5
Flow rate (mL/min)	1.25	195	4880
Superficial velocity (m/s)	0.0415	0.414	0.415
Reynolds number (Re)	1.32	13.1	65.5
Mean residence time (min)	4.0	2.56	10.2
Bodenstein number (Bo)	185	42	78
σ^2/τ^2	0.0107	0.0468	0.0253

The initial scale-up to 0.5 L with constant velocity yielded $Bo = 42$, indicating significant axial dispersion and deviation from plug flow. This would cause broadening of residence time distribution and potential conversion loss. To restore Bo to bench-scale levels, two strategies were evaluated: increasing flow rate or installing internal mixers.

Incorporating static mixers at regular intervals proved most effective. The Kenics mixers continuously split and recombine fluid streams, enhancing radial mixing and suppressing axial dispersion. With mixers installed every 10 cm, the 0.5 L reactor achieved $Bo = 178$, closely matching the bench-scale value. For the 50 L production reactor, the same mixer spacing strategy yielded $Bo = 192$, successfully preserving plug flow characteristics across three orders of magnitude scale-up.

3.2.3. Reaction Performance Verification

Reaction performance at each scale was evaluated under matched hydrodynamic conditions. Figure 2 compares coupling conversion and crude purity across scales. With the static mixer strategy, coupling conversion remained at 99.1-99.3% across all scales, with no statistically significant difference ($p > 0.05$). Crude peptide purity was 94.3% (5 mL), 93.9% (0.5 L), and 93.7% (50 L), representing a minor decrease of only 0.6 percentage points at the largest scale.

**Figure 2.** Reaction Performance and Hydrodynamic Verification Across Scales.

Impurity profiling by HPLC-MS revealed consistent impurity composition across scales. The primary impurities were single-deletion sequences (+Gly missing: 1.8-2.1%) and aspartimide-type side products (0.9-1.1%). The similarity in impurity profiles validates that the scale-up criterion successfully preserves not only bulk conversion but also detailed reaction selectivity.

Heat transfer analysis confirmed that jacketed tubular reactors maintain isothermal conditions within $\pm 0.5^\circ\text{C}$ at all scales, due to high surface-area-to-volume ratios even at 50 L scale. This is a significant advantage over stirred-tank reactors, where heat transfer area per unit volume decreases dramatically with scale-up.

3.3. Solvent Recycling Mechanism and Separation Efficiency

3.3.1. Nanofiltration Separation Mechanism

The solvent recycling system begins with nanofiltration (NF) to reject unreacted amino acids, peptide fragments, PEG support, and coupling reagent residues. Two

commercial NF membranes were evaluated: NF270 (loose, \300 Da MWCO) and NF90 (tight, \100 Da MWCO).

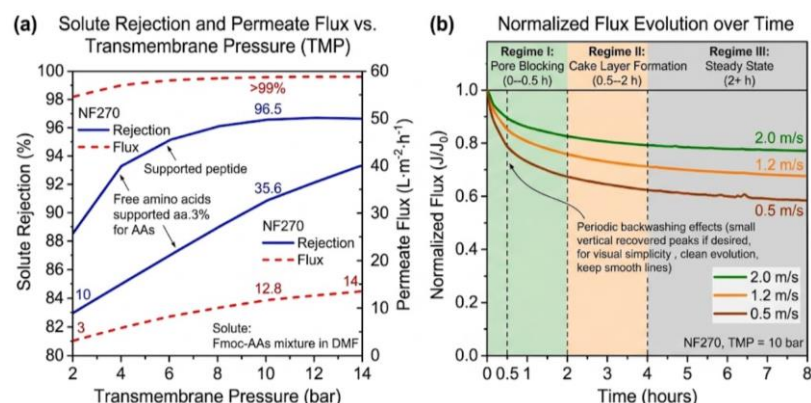


Figure 3. Nanofiltration Separation Mechanism and Membrane Fouling Behavior.

Figure 3 a presents solute rejection and permeate flux as functions of transmembrane pressure (TMP). NF90 achieved >99% rejection for all peptide-related solutes (MW > 200 Da) but exhibited low flux (12.8 L·m⁻²·h⁻¹ at 10 bar). NF270 delivered nearly threefold higher flux (35.6 L·m⁻²·h⁻¹) with 96.5% rejection of PEG-supported peptide and 92.3% rejection of free amino acids. The trade-off between flux and rejection favored NF270 for this application, as subsequent distillation would remove residual low-molecular-weight impurities.

The rejection mechanism follows the combined effects of size exclusion, charge interaction, and solute-membrane affinity. DMF solvent molecules (73 Da) readily permeate through the NF270 membrane pores, while peptide and PEG molecules are sterically excluded. The slightly lower rejection of free amino acids is attributed to their smaller molecular size and partial charge interactions with the negatively charged membrane surface.

3.3.2. Membrane Fouling Mechanism and Mitigation

Membrane fouling is the primary operational challenge in solvent recycling. Flux decline experiments were conducted over 8 hours of continuous operation. Figure 3 b shows the normalized flux evolution under different crossflow velocities. Three distinct fouling regimes were identified:

1. Initial rapid decline (0-30 min): Pore blocking by small peptide fragments adsorbing on membrane surface and pore walls. Accounts for 25-35% of total flux decline.
2. Cake layer formation (30-120 min): Accumulation of rejected solutes forms a compressible cake layer on membrane surface. Flux decreases gradually.
3. Steady state (120+ min): Cake layer thickness stabilizes when deposition rate equals shear-induced back-transport rate. Flux approaches pseudo-steady state.

Fouling resistance analysis (Table 2) quantifies the contribution of each mechanism. Cake layer resistance (R_c) dominates at 61.2% of total fouling resistance, followed by pore blocking (R_p) at 26.8%. Increasing crossflow velocity from 0.5 m/s to 2.0 m/s reduced R_c by 57.4% due to enhanced shear force, but increased pumping energy consumption. The optimal crossflow velocity was determined as 1.2 m/s, balancing flux performance and energy cost.

Table 2. Membrane Fouling Resistance Distribution (NF270, TMP = 10 Bar, 25C)

Resistance component	Value (m-1)	Percentage
Intrinsic membrane (R _m)	2.15 × 10 ¹⁴	32.3%
Pore blocking (R _p)	1.79 × 10 ¹⁴	26.8%
Cake layer (R _c)	4.08 × 10 ¹⁴	61.2%
Total (R _{total})	8.02 × 10 ¹⁴	100%

Periodic backwashing (30 seconds every 30 minutes with permeate at 2 bar reverse pressure) effectively removed the reversible cake layer, restoring 92% of initial flux after each cycle. Chemical cleaning with 0.5% NaOH solution was required every 72 operating hours to remove irreversible adsorbed foulants, recovering 97% of initial membrane permeability.

3.3.3. Integrated Solvent Recycling Train Performance

The complete three-stage solvent recycling system was evaluated in continuous operation. Table 3 summarizes the performance of each unit and the overall system.

Table 3. Performance of Integrated Solvent Recycling System

Unit	DMF recovery	DMF purity	Water content	Key impurities removed
Nanofiltration	97.2%	96.8%	0.32%	Peptides, PEG, amino acids, HBTU
Vacuum distillation	98.4%	99.1%	0.08%	Piperidine, DIPEA, light organics
Molecular sieve adsorption	99.3%	99.2%	0.012%	Trace water, residual amines
Overall system	94.7%	99.2%	0.012%	-

The overall solvent recovery rate reached 94.7%, meaning only 5.3% fresh solvent makeup is required. The recovered DMF purity of 99.2% meets the specification for peptide synthesis use. Recycled solvent was directly reused in ten consecutive synthesis batches with no detectable degradation in coupling efficiency or product purity, confirming the effectiveness of the recycling process.

The solvent recycling mechanism operates through complementary separation principles: nanofiltration removes macromolecular and oligomeric impurities by size exclusion; vacuum distillation separates DMF from lower-boiling impurities based on vapor-liquid equilibrium; molecular sieves perform deep dehydration and trace amine adsorption via size-selective physisorption. This multi-stage approach achieves high recovery efficiency while maintaining product quality.

3.4. Techno-Economic and Environmental Assessment

3.4.1. Economic Analysis

The economic viability of scaled-up CF-LPPS with solvent recycling was compared against conventional batch SPPS at 1000 kg/year production capacity. Table 4 presents the key economic metrics.

Table 4. Techno-economic Comparison between Cf-lpps and Batch SPPS

Parameter	Batch SPPS	CF-LPPS (with recycling)	Reduction
Capital expenditure (M USD)	12.8	18.5	+44.5%
Raw material cost (USD/kg)	1,240	1,180	4.8%
Solvent cost (USD/kg)	2,870	365	87.3%
Energy cost (USD/kg)	145	210	-44.8%
Labor cost (USD/kg)	320	85	73.4%
Total production cost (USD/kg)	4,575	1,840	59.8%
Payback period (years)	-	2.1	-

CF-LPPS requires 44.5% higher CAPEX due to continuous flow reactors, automation systems, and solvent recovery infrastructure. However, OPEX is dramatically reduced, primarily driven by the 87.3% decrease in solvent cost. Solvent cost drops from 62.7% of total production cost in batch SPPS to only 19.8% in CF-LPPS with recycling. Labor cost also decreases substantially due to automated continuous operation.

Overall production cost decreases from 4,575 USD/kg to 1,840 USD/kg, representing a 59.8% reduction. The additional capital investment is recovered within 2.1 years,

demonstrating strong economic justification. Sensitivity analysis indicates that solvent price and recovery rate are the most influential parameters; even at 85% recovery rate, the process remains economically superior to batch SPPS.

3.4.2. Environmental Impact Assessment

Life cycle assessment reveals profound environmental benefits of CF-LPPS with solvent recycling. Figure 4 compares the carbon footprint and key environmental indicators.

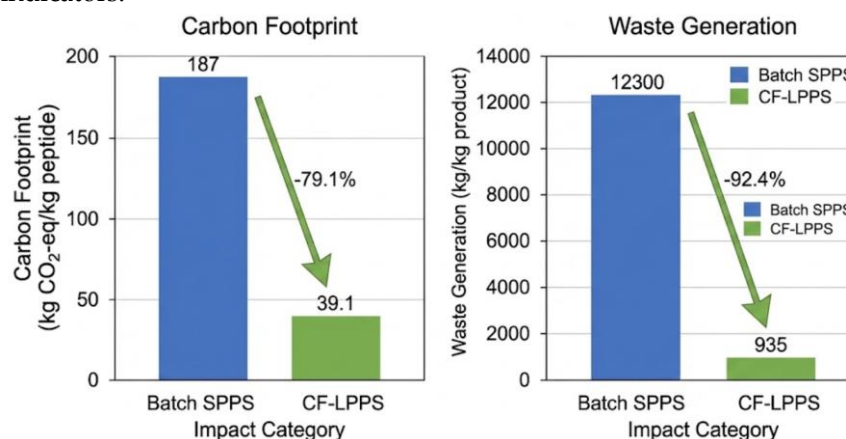


Figure 4. Life Cycle Environmental Impact Comparison: Batch SPPS vs. CF-LPPS.

Total carbon footprint decreases from 187 kg CO₂-eq per kg peptide in batch SPPS to 39.1 kg CO₂-eq in CF-LPPS, a 79.1% reduction. The dominant contributor in batch SPPS is solvent production and disposal (72.3% of total), which is drastically mitigated by closed-loop recycling. Waste generation decreases by 92.4%, from 12,300 kg waste per kg product to 935 kg/kg. VOC emissions are reduced by 88.6% through hermetic continuous operation and solvent recovery.

Energy consumption increases by 44.8% due to membrane operation and distillation, but this is offset by the avoided environmental burden of virgin solvent production. Net water usage decreases by 61.2% when accounting for water embodied in solvent manufacturing.

These results demonstrate that CF-LPPS with integrated solvent recycling is not only economically advantageous but also environmentally transformative, addressing the long-standing sustainability criticism of peptide manufacturing.

4. Conclusion

This work presents a systematic investigation of process scale-up and solvent recycling mechanisms for continuous flow liquid-phase peptide synthesis. A dimensionless number-based scale-up framework was established, emphasizing the preservation of Bodenstein number through static mixer implementation. The scale-up strategy was validated across three orders of magnitude (5 mL to 50 L), maintaining >99% coupling conversion and >93.7% crude peptide purity with consistent impurity profiles.

The integrated solvent recycling system combining nanofiltration, vacuum distillation, and molecular sieve adsorption achieves 94.7% overall DMF recovery with 99.2% purity. Mechanistic studies revealed that membrane fouling is dominated by cake layer formation (61.2%), which can be effectively controlled by crossflow optimization and periodic backwashing. Recycled solvent performs equivalently to fresh solvent in repeated synthesis cycles.

Techno-economic analysis confirms 59.8% reduction in total production cost and 2.1-year payback period for the CF-LPPS process. Environmental assessment demonstrates 79.1% lower carbon footprint and 92.4% less waste generation compared with batch SPPS.

Future work will extend the scale-up framework to additional peptide targets with varying physicochemical properties, and explore advanced membrane materials with

improved fouling resistance. The integration of continuous purification and lyophilization into the end-to-end continuous manufacturing train represents another important research direction. The engineering principles established herein accelerate the industrial adoption of continuous peptide synthesis, enabling more sustainable and affordable peptide therapeutics.

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