

Supplementary Information

**Chemspyd: An Open-Source Python Interface
for Chemspeed Robotic Chemistry and Materials Platforms**

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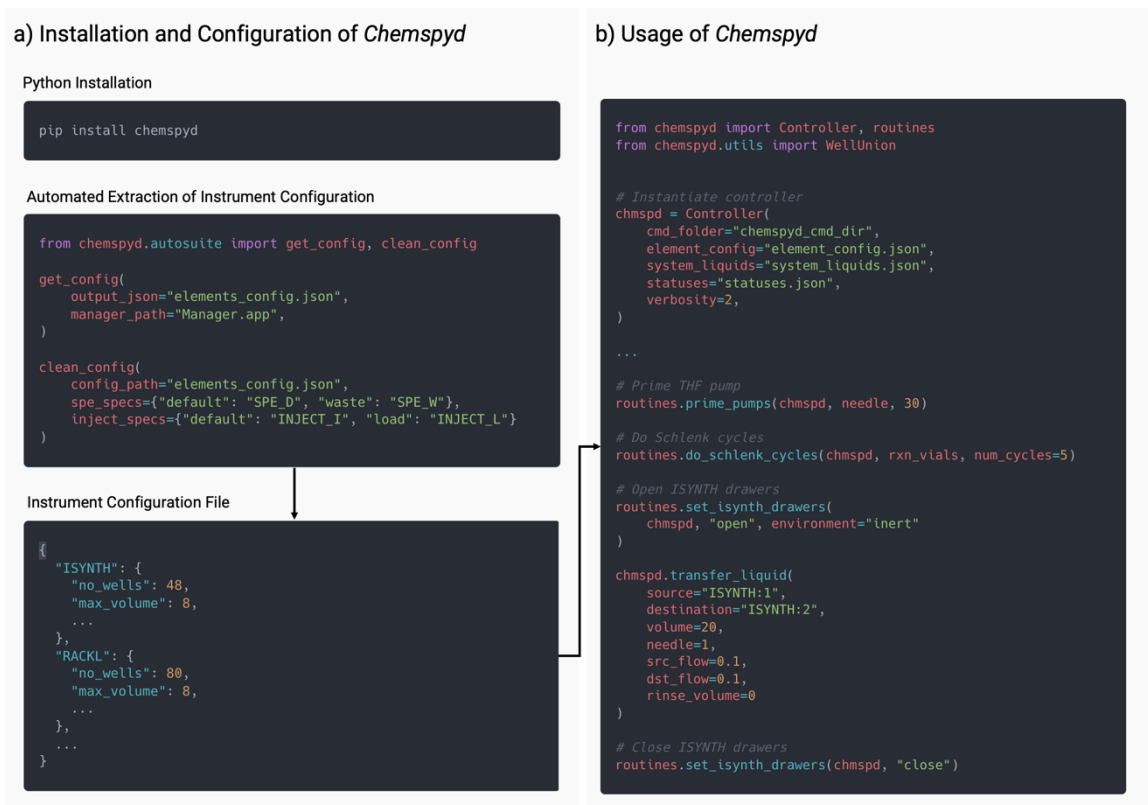


Figure S1. Workflow for installing and using Chemspyd with code examples.

1. Software Architecture

1.1 Communication Protocol between Chemspyd and AutoSuite

The communication between Chemspyd's Executor class and the Manager in AutoSuite proceeds through the dynamic update of four CSV files. The contents of the respective files are summarized in Table S1.

Table S1: Contents of the CSV files used for communication between Chemspyd and the Manager, written in the proprietary AutoSuite software.

File Name	Content	Chemspyd read/write	AutoSuite read/write
response.csv	Instrument operational state (busy / idle / error)	read	write
command.csv	Name and execution parameters of the elementary operation to be performed	read / write	read / write
return.csv	Return values of the performed elementary operation	read	write
status.csv	Dynamically updated collection of experimental metadata (temperature, humidity, pressure)	read	write

As long as the operational state of the instrument is “idle” (as defined in *response.csv*), a command can be submitted to the instrument by *Chemspyd*. If the instrument is “busy”, command execution is paused until the operational state changes to “idle”. *Chemspyd* submits a command to the instrument by providing a command identifier, as well as all required execution parameters, to *command.csv*. Once the *command.csv* is parsed by the Manager, the instrument status (as defined in *response.csv*) is set to “busy”, and the command is executed on the robotic platform. Upon completion of the task, *command.csv* is cleared, the operational state is set to “idle”. If the action generates output values (i.e. measurement values), they are written to *return.csv*, and can be parsed by *Chemspyd*. Instrument metadata are regularly updated in *status.csv*.

1.2 Elementary Actions in *Chemspyd*'s Controller

At the time of publication, *Chemspyd* implements the following elementary actions. Integration of further elementary actions, parallel to more recent module developments from Chemspeed Technologies, is targeted as a community effort.

Table S2. Currently implemented equivalent methods, tasks in *Chemspyd* and *AutoSuite*, respectively.

<i>Chemspyd</i> Controller method	Equivalent <i>AutoSuite</i> task name
transfer_liquid()	Transfer Volumetrically
transfer_solid()	Transfer Gravimetrically
set_drawer()	Set Drawer Valve
set_reflux()	Reflux
set_temperature()	Heat / Cool
set_stir()	Stir
set_vacuum()	Set Vacuum
vial_transport()	Vial Transport
measure_level()	Measure Level
wait()	Wait
unmount_all()	<i>[no direct equivalents]</i>
read_status()	<i>[no direct equivalents]</i>
stop_manager()	<i>[no direct equivalents]</i>

2. Experimental Details

2.1 General

All experiments were performed using the Chemspeed SWING XL platform available in our laboratory at the MatterLab at the University of Toronto, equipped with the following modules (letter labels in Figure S2):

- Liquid Handling System (4-Needle Head **A**, Dilutor with four 10 mL syringe pumps **B**)
- GDU-Pfd Solid Handling System **C**
- GDU-Pfd Fine Dosing Rack (2 x 8 containers) **D**
- ISYNTH Reactor Block **E** (48 x 8 mL reactors, inertization and evaporation block, shaker, temperature control, reflux module)
- MTP Rack for Heating and Magnetic Stirring **F** (two microtiter plates, 24 x 4 mL each)
- Double-Level Holder **G** (80 x 8 mL)
- Double-Level Holder **H** (30 x 20 mL)
- Double-Level Holder **I** (two microtiter plates, 48 x 0.7 mL each)
- Filtration Rack **J** (80 x 8 mL)
- Injection Port **K** (Valco 6-way valve for HPLC injection)
- Gripper for moving vials (**L**) and switching the ISYNTH valves (**M**)

Chemspyd Python scripts for all experiments are provided in the *Gitlab* repository.¹

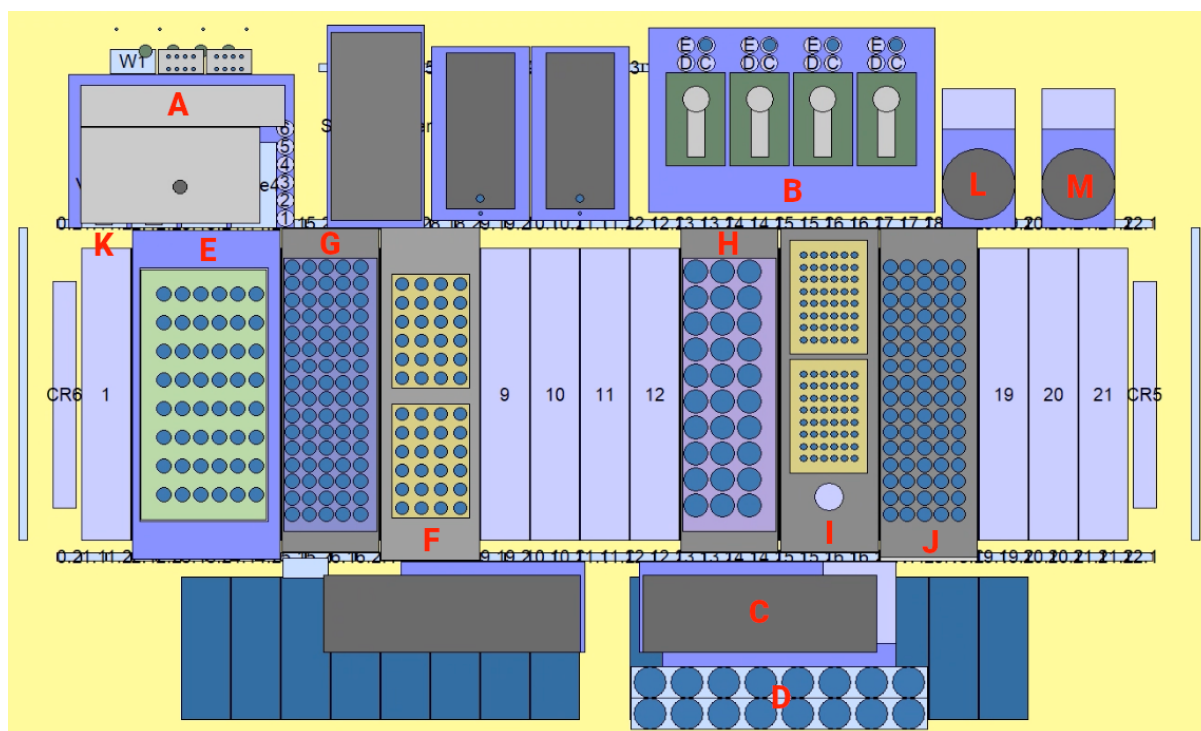


Figure S2. Deck layout of the Chemspeed SWING platform used in this study.

All reagents were purchased from Sigma-Aldrich, Strem or Combi-Blocks, and were used without further purification.

HPLC-MS measurements were performed on a ThermoFisher HPLC-MS system, consisting of a ThermoFisher Vanquish HPLC system equipped with a binary pump, a heated column compartment and a DAD detector, and a ThermoFisher QExactive mass spectrometer. As the stationary phase, a ThermoFisher AccuCore column (100x2.1 mm, 2.6 μm particle size, C18 or Phenylhexyl, respectively) was used. HPLC-MS operation was controlled by our in-house Python software, combined with the proprietary Chromeleon software kit.

Steady-state absorption spectroscopy was performed in a custom-built system described in a previous report from our lab.² UV/Vis spectra were recorded using a white light lamp (Ocean Insight, DH-MINI) combined with a spectrometer (Ocean Insight, QEPro XR). Samples were automatically transferred from an autosampler to the measurement cell using syringe pumps, as described previously.² All measurements were controlled by our in-house Python software.

2.2 Condition Screening for Silver Nanoprism Formation

Silver nanoprisms were synthesized according to the general procedure described in a previous report by Frank *et al.*³ Stock solutions of sodium citrate, silver nitrate, hydrogen peroxide, potassium bromide, and sodium borohydride (for concentrations, see Table S3) were prepared manually, and the respective 20 mL glass vials were placed on the double-level vial holder.

48 parallel nanoprism synthesis experiments were then performed using the ISYNTH reactor. For each experiment, the respective quantities (see below) of all reagents were transferred to the reaction vial, and water was added to obtain a total reaction volume of 6.5 mL. All reaction mixtures were stirred at room temperature for 2.5 min and were then subjected to the automated absorption spectroscopy workflow described in a previous report from our group.²

The 48 sets of reaction conditions were generated through Latin Hypercube sampling (using the implementation in the *SciPy* library⁴). The ranges of allowed target concentrations for each reagent are shown in Table S3. Specific conditions can be found in the same place as the Python code for the experiment.

Table S3: Stock solution concentrations and target concentration ranges of all reagents used for silver nanoprism condition screening.

Reagent	Stock Solution [mol L ⁻¹]	Minimum Conc. [mol L ⁻¹]	Maximum Conc. [mol L ⁻¹]
Sodium Citrate	1.0·10 ⁻²	0	1.5·10 ⁻⁵
Silver Nitrate	5.0·10 ⁻⁴	0	1.0·10 ⁻⁶
Hydrogen Peroxide	2.0·10 ⁻¹	0	2.0·10 ⁻⁴
Potassium Bromide	2.5·10 ⁻⁵	0	2.5·10 ⁻⁸
Sodium Borohydride	1.0·10 ⁻²	0	1.0·10 ⁻⁵

2.3 Condition Screening for Buchwald-Hartwig Couplings

A systematic, combinatorial screen of different categorical reaction variables was performed for the Buchwald-Hartwig coupling between 4-methylaniline and 3-bromopyridine (Figure S3), which was adapted from work by Ahneman *et al.*⁵ Specifically, all permutations of three Pd sources, eight ligands and two bases (2 x 8 x 3 = 48) were evaluated in an automated fashion.



Figure S3. General reaction scheme for the Buchwald-Hartwig coupling.

Parallel experiments were performed using the ISYNTH reactor block. In preparation of the experiments, Pd sources were dosed into the respective vials using an automated powder dosing station (Mettler Toledo QUANTOS, for quantities see Table S4). Pre-loaded vials were manually transferred to the ISYNTH reactor block. For all other components, stock solutions in DMSO were prepared manually (for concentrations see Table S4), inertized, and the corresponding 20 mL vials were loaded to the double-level holder. Reaction vials in the ISYNTH block were inertized by five consecutive evacuate–backfill (“Schlenk”) cycles. Stock solutions of all reagents (ligand, nucleophile, electrophile, internal standard, base) were added under a constant stream of inert gas. The reactors were closed, and all reactions were heated to 60 °C under reflux for 16 h. After cooling to room temperature, an aliquot of the reaction mixture (0.10 mL) was filtered through a short plug of celite on the filtration rack and eluted with acetonitrile (3.0 mL). After gravity filtration, the filtrates were subjected to automated analysis by HPLC-MS, using the on-deck HPLC injection valve. The HPLC separation gradient is given in Table S5.

The relative quantities of 3-(4-methylphenylamino)pyridine were determined by integrating the peaks of the product and the internal standard in the UV/Vis chromatogram. Calibration curves to determine the actual product yields were not recorded.

Table S4: Reactants and reagents used for screening the conditions of the Buchwald-Hartwig coupling of 4-methylaniline and 3-bromopyridine.

Type	Name	Stock Solution		Mass
		Concentration [mol L ⁻¹]	Volume [mL]	
Nucleophile	4-Methylaniline	1.0	0.4	---
Electrophile	3-Bromopyridine	0.5	0.8	---
Pd Source	Pd(OAc) ₂	---	---	9.0
	Pd ₂ (dba) ₃	---	---	18.3
	PdCl ₂	---	---	7.1
Ligand	<i>t</i> -BuXPhos	0.025	2.4	---
	P(<i>o</i> -tol) ₃	0.025	2.4	---
	JohnPhos	0.025	2.4	---
	P(<i>t</i> -Bu) ₃	0.025	2.4	---
	XPhos	0.025	2.4	---
	RuPhos	0.025	2.4	---
	BrettPhos	0.025	2.4	---
Base	MTBD	0.75	0.8	---
	BTMG	0.75	0.8	---
Standard	TMB	1.0	0.4	---

MTBD: 7-Methyl-1,5,7-triazabicyclo(4.4.0)dec-5-ene. BTMG: 2-tert-Butyl-1,1,3,3-tetramethylguanidine. TMB: 1,3,5-Trimethoxybenzene.

Table S5: HPLC gradient used for analysis of crude reaction mixtures from the Buchwald-Hartwig coupling. Polar eluent: water + 0.1% formic acid. Nonpolar eluent: Acetonitrile + 5% THF + 0.1% formic acid.

Time	Nonpolar Eluent [%]	Flow Rate [mL min ⁻¹]
0.00	5	0.40
2.00	5	0.40
6.75	100	0.40
7.40	100	0.40
7.45	5	0.40

2.2 Kinetic Monitoring of an Amide Coupling

Chemspyd's ability to dynamically control experiments is demonstrated by tracking the kinetics of the amide coupling between 2-methoxybenzoic acid and naphthalene-2-methanamine. The synthesis procedure was adapted from a previous report by Liu *et al.*⁶

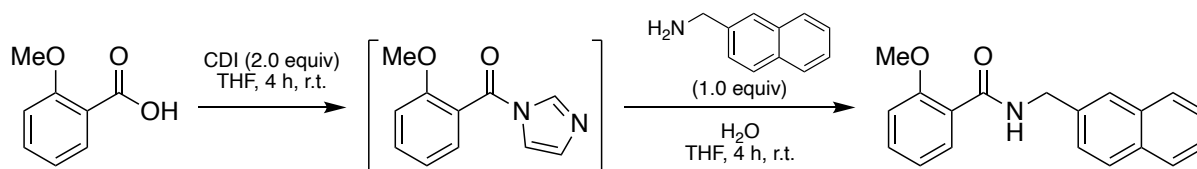


Figure S4. Reaction scheme of the two-step amide coupling.

Reactions were performed in 24 x 4 mL vials in multi-well plates on the heat-stir rack of our Chemspeed SWING platform. Stock solutions of all reactants and reagents in THF were prepared manually (for concentrations see Table S6), and the corresponding 20 mL vials were loaded onto the double-level holder. To each vial was added 2-methoxybenzoic acid with 1,3,5-trimethoxybenzene as the internal standard (0.54 mL stock solution, 0.27 mmol, 1.0 equiv.) and CDI (2.16 mL stock solution, 0.54 mmol, 2.0 equiv.). The reactions were stirred at room temperature for 4 h, and aliquots for HPLC-MS analysis were taken from vials 1–12 in 15-minute intervals (for details see below). Afterwards, water (100 μ L, 5.5 mmol, 10 equiv.) was added to each vial, and the mixtures were stirred at room temperature for 1.5 h to quench excess CDI. Naphthalene-2-methanamine (0.54 mL, 0.27 mmol, 1.0 equiv.) was added to all vials, and the mixtures were stirred at room temperature for an additional 6 h. In this time period, aliquots for HPLC-MS analysis were taken from vials 13–24 every 15 minutes.

Throughout the entire experiment, HPLC aliquots were taken by sampling 150 μ L from a single, reactor vial. The sampling of aliquots was distributed evenly over all reaction vials (1–12 for the first coupling step, 13–24 for the second coupling step), to minimize the effects of mass loss through iterative removal of reaction volumes. Each aliquot was transferred to a well on the 48 x 0.7 mL MTP plate, and sodium methoxide (0.5 mL, 0.375 mmol, 2.5 equiv.) was added to derivatize the reaction intermediate to the stable, HPLC-detectable methyl ester.⁶ After thorough mixing, the sample was injected to the HPLC-MS instrument.

Relative concentrations of 2-methoxybenzoic acid, naphthalene-2-methanamine, methyl 2-methoxybenzoate (derivatized intermediate) and the reaction product are reported as the ratio between the integrated UV chromatogram peak and the internal standard peak, normalized by the maximum obtained value for this ratio.

Table S6: Stock solution concentrations of all reagents used for kinetic analysis of the amide coupling reaction.

Compound	Stock Solution Concentration [mol L ⁻¹]
1,3,5-Trimethoxybenzene	0.5
2-Methoxybenzoic acid	0.5
CDI	0.25
Naphthalen-2-methanamine	0.5
Sodium methoxide	0.075

Table S7: HPLC gradient used for analysis of the amide coupling reaction aliquots. Polar eluent: water + 0.1% formic acid. Nonpolar eluent: Acetonitrile + 5% THF + 0.1% formic acid.

Time	Nonpolar Eluent [%]	Flow Rate [mL min ⁻¹]
0.0	5	0.65
0.5	5	0.65
3.5	100	0.65
6.3	100	0.65
6.5	5	0.65

3. References

- 1 Chempyid: An Open-Source Python Interface for Chemspeed Robotic Platforms, <https://gitlab.com/aspuru-guzik-group/self-driving-lab/instruments/chempyid>.
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- 4 Latin Hypercube Sampling in Scipy, <https://docs.scipy.org/doc/scipy-1.9.2/reference/generated/scipy.stats.qmc.LatinHypercube.html>.
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