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## ***HP 1090 Series II/L Liquid Chromatograph***

### **Specifications**

#### **General Description**

Every HP 1090 Series II/L liquid chromatograph is an integrated HPLC in which each module is chosen by the customer to meet specific analytical demands. Every module is an optimum match, ensuring total system performance. That means system specifications, not module specifications.

#### **General**

**Mainframe Dimensions:** 70 cm (W) x 60 cm (H) x 52 cm (D). 28 inch (W) x 26 inch (H) x 20 inch (D).

**Weight:** 60-80 kg; 132-176 lb; depending on configuration.

**Environment:** 5-40 °C, with < 95% humidity (non-condensing.)

**Power:** Line voltage 100/120/220/240 VAC + 5%, -10%

**Line frequency:** 50 Hz (48-55 Hz)/ 60 Hz (57-66 Hz)

**Power consumption:** 750 VA max.

**Power Line Disturbances:** Surges and sags must not exceed  $\pm 20\%$  of normal line voltage. Voltage must return to normal within 20 ms. Line transients of greater than 10  $\mu$ s pulse width and more than 50% of normal rated voltage may produce instrument malfunctions.

### **Safety Aids:**

- Leak sensors for solvent delivery system, Injector, detector.
- Drain for column compartment.
- Maximum and minimum pressure limits, selectable from 0 - 400 bar (0 -5880 psi).
- Sealed solvent reservoirs, fume hood connection for purge gas outlet.
- Central ventilation.
- Maximum temperature of 140°C in the column compartment heat-exchanger. Two independent temperature cut-out devices.
- Low voltages in major maintenance areas.
- Error messages via LEDs.
- For protection of columns, the maximum rate of pressure change is < 20 bar/s.
- Materials in Contact with solvents
- Stainless steel, PTFE, PTFE + 25% C, quartz, ceramic, ruby, sapphire, KALREZ, VESPEL
- Use of solvents that attack stainless steel or quartz is left to the user's discretion.

**Recommended pH Range:** 2.3 to 9.5 standard.

Extension up to 12.5 is optional with extended pH-kit. Solvents with a pH between 1.9 and 2.3 must not contain acids which attack stainless steel. When using the extended pH-kit, typically at least 1000 switching cycles of injection system are possible before change of rotor is required.

See the guide books, part no.  
01090-90210, for important information on solvents.

### **Control of External Devices**

Remote control  
Remote control inputs and outputs and system status.

External contacts

4 external contacts selectable as relays (each 24 V, 0.1 A max) or contact closures (each 40 V, 0.1 A max.)

**Autosampler:** BCD coded bottle number output.

### **Programmable Filter-Photometric Detector**

#### **Filters**

**Interference filters:** 254 standard; 210, 230, 269, 280, 340, 430 and 540 nm available optionally.

Special filters available (wave-length user-specified).

**Filter bandwidth:** 10 nm typically.

**Signal Characteristics:** Noise peak to peak  $\pm 2.5 \times 10^{-5}$  AU.

**Drift:**  $< 1 \times 10^{-3}$  AU/Hour.

#### **Noise and drift specifications were Obtained under the following conditions:**

Wavelength of 254 nm; response time of 400 Ms; low lamp-current; flow of 1 ml/min using Bidistilled, degassed water; integrator output Signal; after an adequate warm-up time of one hour and at constant room temperature.

#### **Signal Ranges**

**Linear absorbance range:** ~ 0.1 to 1.5 AU (measured with acetone at 254 nm).

**Wavelength switching:**  $< 2$  seconds (including baseline correction).

**Lamp current:** High and low selectable.

#### ***Programmability***

Parameters

Wavelength, response time, lamp current, lamp off, % offset and attenuation for reorder output.

## ***Analog Output***

**2 analog-outputs:** (1 integrator, 1 recorder): 0-1 V (0.5 V/AU); 0-100 mV.  
Zero at 0 or 50 mV selectable.

Response time selectable at 100, 400 and 800 ms (10-90%).

**Detector Flow Cell:** 4.5 µl volume, 6 mm path length, max, pressure 100 bar (1470 psi).

## ***System Control***

### **Local User Interface**

**Display:** 32 character alphanumeric display.

**Control:** Keyboard with function keys and alphanumeric keys.

**Method and sequence storage:** Built-in battery protected memory. Typically 6 methods including 10 time table entries can be stored together with 10 sequence lines.

## ***Communications***

**HP-IB:** Interfaces from built-in HP-IB controller to diode-array detector and optional printer. Interface to HPLC ChemStation possible in remote control mode.

**INET:** Link to integrators (HP 3392A and HP 3396A Series II).  
Synchronized start/stop and complete method and sequence handling. Link to HP 3350A via HP 3392A and HP-DL.

## ***Data Presentation***

-Detector analog signals on reorder or integrator.

-Spectra and peak purity numbers on external printer (diode-array detector only).

## ***Solvent Delivery System***

### **Configurations:**

- Isocratic.
- PV5 ternary or quaternary.

- DR5 binary or ternary.

### **Solvent Storage and Preparation**

- Up to three built-in 1 liter bottles.
- One external bottle is optional for use as a flushing solvent reservoir with ternary PV5.
- External bottle in solvent conditioning module for quaternary PV5.
- Operation from external reservoirs possible for isocratic, binary and ternary DR5 SDS.
- Helium Degassing, built-in filters and frits.

### ***Ranges***

**Pressure:** 0 – 400 bar (0- 5880 psi).

**Flow:** 0 – 5000 µl/min, in 1 µl/min steps

### ***Flow***

**Accuracy:** ± 2% of setpoint or ± 1 µl/min, whichever is greater, independent of backpressure and solvent, excluding volume contraction effects.

**Precision:** < 0.3% RSD, based on retention time.

### ***Composition***

**Precision:** PV5

± 0.25% absolute, peak to peak, water/acetonitrile, 500 µl/min, 2500 µl/min.

### ***Delay Volume***

**PV5:** 1750 – 2000 µl maximum, dependent on backpressure.

**DR5:** 300 – 500 µl maximum, dependent on backpressure.

## ***Injection Systems***

Manual Injector

Rheodyne valves.

**Maximum pressure:** 400 bar (5880 psi).

**Loop sizes:** 5 µl standard. 0.5, 1, 10, 20, 50, 100 µl available.

Programmable Variable-Volume Auto-injector

Programmable injection volumes

0.1- 25 µl (standard), or

0.1- 250 µl (optional)

in 0.1 µl steps.

**Maximum pressure:** 400 bar (5880 psi).

### **Precision**

< 1% (typically 0.6%) RSD from 2 ~ 25 µl (25 µl syringe).

< 1% (typically 0.6%) RSD from 10 – 250 µl syringe).

### **Sample vials**

2 ml standard vial.

300 µl disposable microvial.

100 µl microvial.

### **Minimum sample volume**

40 µl (2 ml standard vial).

20 µl (300 µl microvial).

5 µl (100 µl microvial).

### **Sample viscosity range**

0.2 – 50 cP (< 10 µl).

0.2 – 5 cP (< 10 µl).

### **Injection cycle time**

15-50 s (depending on injection volume and slow-down factor).

## ***Autosampler***

**Capacity:** 100 sample bottles (2 ml) or microvials.

## **Magazines**

10 removable magazines with  
10 bottles each.

**Random access time:** < 10 seconds from any position.

## ***Column Compartment***

**Internal Dimensions:** 37 cm (W) x 15 cm (H) x 5.5 cm (D).  
15 inch (W) x 6 inch (H) x 2 inch (D).

## **Heated Compartment**

- Solvents and samples are preheated in low-volume (2  $\mu$ l) heat exchanger.
- Cooling for sub-ambient operation is optional.

**Settable temperature range:** 0 – 100 °C in 0.1°C steps.

**Controlled temperature range:** 20 °C above ambient to 100°C.  
5°C below ambient to 100°C. above ambient with external cooling.

**Temperature stability:**  $\pm$  0.5°C.

## **Building on Success**

When you need a high-performance liquid chromatograph that will analyze your routine samples reliably and provide the power for method development, the answer is the Hewlett-Packard HP 1090 Series II/L Liquid chromatograph. Based on the highly successful HP 1090 Series L, the improved Series II/L sets new standards for HPLC.

## **Integrated yet versatile**

Choose from a wide variety of built-in modules and buy exactly what you need. Upgrade later when requirements change. Every module is an optimum match, ensuring maximum performance. And every module communicates fully ensuring synchronized operation.

An HPLC that's simple to use – even for the LC novice.

Incorporate the HP 1090 Series II/L liquid chromatograph with data handling equipment of your choice. Our integrators, multi-instrument workstations and laboratory automation systems will increase productivity in your laboratory.

## **Gain Plenty – Lose Nothing**

Each module of the HP 1090 Series II/L not only ensures maximum efficiency with standard sizes of columns but meets the stringent requirements of Low Dispersion Liquid Chromatography (LDLC). Examples of how to increase throughput by combining high-speed columns with intelligent automation are available. Or how to use microbore columns to increase detectability and cut solvent consumption by 80%.

**There are a variety of choices available for each of the major modules:**

- 1.) Controllers and data handling
- 2.) UV absorbance detection
- 3.) Solvent delivery
- 4.) Sample injection
- 5.) Column compartment.

**Low Dispersion liquid chromatography:** saves solvent, saves time

**Challenge**

- Overcome existing instrument limitations
- Enable analysts to explore new trends in chromatography
- Yet still preserve the ability to run existing methods.

**Solution**

- Design a new LC instrument
- Minimize all dead volumes
- Optimize flow geometry and flow-cell design
- Accommodate a wide range of columns

**Advantages**

Without sacrifice, you stand to gain a great deal: lower solvent costs, higher throughput, improved detectability. All in a system of integrated modules, giving you flexibility, upgradability, and total system system performance.

**Microbore LC:** Large savings in solvents

Take two columns, one 4.6 mm i.d. and the other 2.1 mm i.d.: fill both with the same packing material. They'll generate the same plate number, and so have the same efficiency. But the difference in internal volume leads to two big differences.

The flow rate required to achieve a certain linear flow velocity in the smaller bore column is reduced by a factor of 4.8: solvent consumption for a given analysis will be reduced by nearly 80%.

This can add up to an annual saving of several thousand dollars.

Additionally, the smaller bore column reduces, by the same degree, the volume in which an eluting peak is dissolved. For the same sample input, peak heights will be 4.8 times higher, which means that the detection limit is 4.8 times lower. This is particularly beneficial with small amounts of sample.



Transferring methods between columns of different diameters requires a simple adjustment of flow rate. The same peak heights are obtained if the injection volume is reduced by the same factor.

**High-speed LC:** Up to 2.8 times faster.

Take two columns of 4.6 mm i.d., one 10 cm long and the other 6cm long. To generate 5000 plates, fill the 10-cm column with 5- $\mu$ m particles, and the 6-cm with 3- $\mu$ m particles. For the same linear velocity, the analysis speed of the shorter column is (10/6) times faster. But the optimum linear velocity of 3  $\mu$ m particles is (5/3) times higher. So analysis speed is now (10/6) x (5/3) = 2.3 times faster.

A reduction in particle size decreases chromatographic dispersion inside the column. The eluting peaks are higher: the absolute detection limit is lower.

### **Instrument Demands**

Taking advantage of microbore and high-speed LC isn't just a matter of installing low-volume columns in an existing instrument. Low Dispersion.

Cell Volume	0	1	4.5	12
Path length	0	2	6	10
Resolution	1	0.99	0.95	0.74
Sensitivity	0	0.2	0.6	1

The effect of flow-cell dimensions on resolution and sensitivity with a microbore column.

Liquid Chromatography means low sample dispersion: the components elute in smaller volumes of mobile phase. In order to achieve optimum performance with LDLC columns, extra-column sample dispersion must also be minimized.

That's why LDLC involves the complete system. Capillary lengths and internal diameters must be reduced, injector volumes must be miniaturized and the geometry of every component and connection in the flow-path must be optimized. Inevitably, some trade-offs have to be made. The flow cell design, for example, has to be a compromise between resolution (cell volume) and sensitivity (optical path length).

Even more critical, are the demands that flow rates such as 100  $\mu$ l/min place on the solvent delivery system (SDS). To ensure reproducible quantitation, the SDS must be capable of delivering very precise flow and composition, even with 1% B. It must also generate smooth gradients that reach the column in the shortest time possible.

### **Conclusions**

Comments and re-orders from customers throughout the world have demonstrated that HP 1090 liquid chromatographs have enabled users to benefit from the advantages of

LDLC, without sacrificing high performance with existing methods. The HP 1090 Series II is designed to improve on this success.

### **Control and data handling**

Choose from two controllers. Control all modules from the built-in keyboard or work from the HPLC ChemStation (DOS Series). Evaluate data locally on the HP integrators. If you want graphical evaluation, use the ChemStation, or send data to an HP 3350 Series Laboratory Automation System for safe data archival and remote data evaluation.

### **Challenge**

- Control all modules from one location
- Create analytical methods quickly and safely
- Provide easy automation
- Evaluate data intelligently
- Reprocess and store data safely.

### **Solution**

- Provide a built-in system controller for all modules
- Enable control and data evaluation from an external ChemStation
- Create and store methods and chain them for automatic operation.

### **Advantages**

The built-in user interface allows easy and fast set-up of all system parameters for complete methods and sequence. With a choice of integrator or HPLC ChemStation (DOS Series), data can be evaluated and stored conveniently and effectively. The HP 3350 Series Laboratory Automation System provides remote data evaluation and safe archival.

### **User-friendly**

All HP 1090 Series II/L Liquid Chromatographs have a built-in User-friendly interface for controlling the instrument. This Local user interface (LUSI) gives You convenient access to all LC Parameters through a keyboard With straightforward functions and a bright, 32-character display.

Enter and edit LC setpoints quickly and easily through the logical arrangement of the function keys. If you do make a mistake, the controller responds with useful information. Scan through your list of entries rapidly before you store them as a method in the battery-protected memory. See the contents of a method at any time. Recall the methods as you need them.

Program your LC to run methods unattended, in any sequence. Periodic injections from one or more calibration vials alternating with the samples enable accurate quantitation throughout the sequence. Even an inexperienced user can set up a sequence for the unattended analysis of 100 samples.

If a problem arises, it is shown on the display and recorded in the Controller's memory with time And date. Information about the Status of the system is available At any time.

### **Integrators**

Choose from the line of Hewlett-Packard's Integrators for powerful local data evaluation. Connect the HP 3396 Series II Computing integrator via the INET communication link. Store LC, filter-photometric detector and integrator methods in the Integrator memory and recall them for unattended operation. Make data evaluation an integrated part of your method.

Include the diode- array detection module in the LC and synchronize the HP 3396 Series II Integrator with analog and remote Cables. Add a ThinkJet printer for spectra and peak purity output.

The HP 3396 Series II has computing capabilities that let you re-integrate, re-plot and perform multi-level calibrations. You can go one step further and enhance the integrator's standard features by programming with a variety of BASIC commands, statements and functions. Store chromatograms, method files, calibration files and BASIC Programs in the 98 Kbytes of internal memory. Add a flexible disc drive and get a convenient means of storing information for all your samples.

Or connect the HP 3396 Series II to your personal computer and use existing storage capacity. Use integrator results in a spread-sheet or combine them with word-processing software.

Multi-tasking, multi-instrument control

The HPLC ChemStation (DOS Series) is a multi-instrument controller running on HP Vectra and IBM-AT personal computers. The multi-tasking software, using the Microsoft windows environment, enables data to be acquired in the background while other tasks can be run simultaneously in other windows. With a click of the mouse, a new menu can be pulled down, waiting for your entries.

The graphics evaluation software includes all the features needed for evaluation of your chromatograms. For most convenient operation and complete automation there is an auto-integration mode which integrates each chromatogram individually after a preceding evaluation. Graphics output is optimized by use of a print spooler.

Whenever you want to customize your data evaluation of the system's automated operation, MACROs are available for you.

Control of up to four I.C. systems is possible from one HPLC ChemStation (DOS Series). Use the ChemStation's own peripherals or communicate to other PCs or share peripherals Connected via the Local Area Network.

### **Networking and central archiving**

Use the INET communications Link and connect the HP 3392 Integrating interface with the LC. Coordinate the operation of both Instruments and combine the LC and integrator methods as one. Transfer reports, raw data and Methods to an HP 3350 Series Laboratory Automation system For central archiving and evaluation. Special software packages give you Superb facilities for sample management and chromatogram plotting. Send raw Data back to the HP 3392 for re-integration, and download methods and sequences to the instruments.

### **Two ways to better detection**

Wavelength changes in a UV/Vis detector offer improved sensitivity and selectivity. When designing our integrated HPLC modules, we found two ways of providing programmable wavelength switching at attractive prices.

- 1.) The economical filter photometer enables you to program your detection wavelength between and during runs to give the most sensitive detection.
- 2.) The versatile diode-array detector lets you optimize sensitivity and use dual-channel detection to ensure correct component identification and purity.

### **Challenge**

- Offer a selection of UV/Visible detectors
- Fast wavelength switching
- Full programmability
- High sensitivity
- Instant spectral acquisition without stopping the flow
- Automatic peak purity and identity validation

### **Solutions**

- 1.) Focus light through an 8-position filter wheel Split the beam and use two photodiodes for detection.
- 2.) Focus UV light through the eluate and onto a diffraction grating, use a photodiode-array for detection.

### **Advantages**

Superior performance and full automation make the filter-photometric detector an economic alternative. With simultaneous acquisition of chromatographic and spectral data, the versatile diode-array detector gives accurate quantitation and ensures correct component identification.

### **Filter-photometric detector (FPD): Optical unit**

A deuterium lamp provides a continuum of light from 190 to 600 nm. This is focused by a planoconvex lens and transmitted through an aperture to minimize stray light.

The light is reduced to 10 nm bandwidth as it passes through one of the seven

interference filters mounted on the wheel. Switching between filters during a run takes less than 2 seconds.

The second lens focuses the beam within the flow cell, for any specified wavelength. Precision focusing and optimized flow geometry minimize baseline drift caused by refractive-index effects. This is especially important in gradient analyses.

The beam splitter divides the light between a reference and a photodiode. The output from both is processed and provided as a chromatographic signal for an integrator or recorder.

**FPD: Optimum quantitation**

Wavelength changes during an analysis can provide improved sensitivity. However, reproducible quantitation demands fast wavelength switching without baseline offset.

**FPD: High Linearity**

Some samples contain a highly concentrated component (X) and a trace component (Y). A detector's response is only linear up to a certain concentration. Optimize linearity by detecting component X at the flat part of its spectrum, or where the extinction coefficient is low. Then switch wavelengths to ensure maximum detection of component Y.

**Diode-array detection (DAD): Optical unit**

The optical system is designed to minimize noise. The system uses reverse-optics, which ensures that all the light passes through the cell before being dispersed across the 211 photodiodes in the array. For optimized detection, different combinations of flow cells and slits can be selected.

The complete array is read every 10 milliseconds. Such high-speed data acquisition guarantees that spectra are free from distortions even with fast peaks eluting from a high-speed column. Because the diode-array and the grating are fixed, mechanical inaccuracy is eliminated, which ensures outstanding quantitative reproducibility and reliability.

**DAD: Sensitivity and Selectivity**

For optimized sensitivity and selectivity, several possibilities are offered by a diode-array based detector. Before injection, select up to eight chromatographic signals, each with a definable wavelength and bandwidth for signal and reference. Specify signals that are optimized for each of your sample components.

Now set the times at which you want to send the selected signals to the dual analog outputs. As your sample elutes, you get two chromatograms printed in parallel, each composed of peaks detected at different wavelengths.

Use a flow cell with extended path length to increase the signal. To further increase light throughput, increase the width of the slit.

When you can't get two compounds completely separated, use the peak suppression technique for accurate detection and quantitation. Select a detection signal where the

component to be suppressed has the same absorbance at detection and reference wavelength, while for the component of interest there is a significant absorbance difference.

### **DAD: On-line spectra**

During the same run, automatically take spectra on the upslope, apex and downslope of every peak. The spectra for each peak are normalized on-line, overlaid and then printed on the HP 2225 ThinkJet printer. Alternatively, print a condensed report with peak purity information represented by numbers. Whether you're doing method development or repetitive analyses you'll find the fast checks on peak purity and identity add significantly to the confidence level of your results.

### ***Choose your Gradient Capability***

#### **High performance under all conditions**

The quality of the solvent delivery system (SDS) determines the quality of the analytical result. If reproducibility better than 1%- for all flow rates, for all compositions, for all solvents and for any column back-pressure.

#### **Designed excellence**

To ensure that the HP 1090 solvent delivery modules meet the most stringent requirements, flow is metered at low pressures and separated from high pressure generation. A proportioning valve provides economic quaternary capability. For the most demanding gradients, flows are metered in each solvent channel.

### ***For a wide flow range***

#### **Challenge**

- Deliver total flow rates from 20 µl/ min to 5000 µl/min.
- Maintain constant flow, independent of solvent properties
- ensure precise composition for up to four solvents
- offer a choice of gradient performance with upgrade potential.

#### **Solution**

- Separate flow metering from pressure generation.
- Meter the flow at low-pressure to eliminate solvent effects
- Deliver all the metered solvent with every stroke of a high-pressure pump
- Use a proportioning valve to provide economical quaternary capability
- Or meter the flow in each solvent channel for the most demanding microbore gradients.

#### **Advantages**

The solvent delivery modules give you a superlative combination of high precision and flexible operation. Run isocratic analyses with standard, high-speed and microbore columns. Upgrade to PV5 for an economical solution to ternary or quaternary gradients. Or choose DR5 for the most demanding high-speed and microbore gradients.

Every HP 1090 SDS is based on four main components; a metering pump, a low-pressure compliance device, a high- pressure pump and damping unit.

### **Smooth Flow**

A dual-syringe metering pump accurately generates solvent flow by true volumetric displacement. Under servo-drive control, the sapphire pistons move in steps of only 0.7  $\mu\text{m}$  displacing 7 nl. Such resolution guarantees a smooth flow, even at micro bore flow rates.

With two pistons, solvent intake and delivery is simultaneous and continuous. When the pistons reach the end of their 100  $\mu\text{l}$  stroke, the flow connections are reversed. A motor-driven rotary valve avoids the common problem of check-valves, which need a certain backflow to close. Even the slight volume lost during valve operation is measured and replaced. The result is continuous flow with negligible fluctuations.

The normal working pressure of the metering pump is 2 to 4 bar, ensuring that all piston seals have a long lifetime and that solvent metering is independent of solvent compressibility.

The metered solvent enters a low pressure compliance (LPC) device which stores the flow during the delivery stroke of the high-pressure pump (see Accurate flow). The pressure in the LPC never exceeds 6 bar, and is constantly monitored by a transducer.

### **Accurate Flow**

The high-pressure pump maintains flow precision, regardless of flow rate or column back-pressure, by constantly delivering all the metered flow. Ten times every second, a closed loop oil pump pressurizes a flexible, gold-coated, stainless steel diaphragm to 440 bar against a flat surface: the pump-head which holds the solvent inlet and outlet valves.

On the backward stroke, the oil has to overcome a 440-bar valve before returning to the reservoir into the cylinder, reducing the pressure on the diaphragm to less than the solvent pressure on the inlet valve. The solvent held in the LPC flows into the high-pressure pump, raising the diaphragm.

On the forward stroke, the oil has to overcome a 440-bar valve before returning to the reservoir. The diaphragm is forced flat, pumping all the solvent beneath it onto the column. Solvent delivery is therefore completely independent of column back-pressure. There are no moving seals in contact with the solvent at high pressure: this is important for reliable operation

The damping unit greatly reduces the pressure ripples from the high-pressure pump. It also contains a transducer, which monitors the column back-pressure. If this exceeds the specific limit, the system controller protects the column by slowing or stopping the metering pump.

*The isocratic HP 1090 SDS can be upgraded in two ways.*

**PV5 ternary or quaternary: for economy**

For an economical solution to ternary and quaternary mixing or gradient operation, PV5 is the right choice. A proportioning valve supplies the desired mixtures in the correct ratio to metering pump.

Use PV5 to get high throughput of isocratic analyses by switching solvent with the method. Or save operator time by letting the SDS mix the solvent. When gradient elution is called for, PV5 delivers. The accuracy and precision of composition is excellent, ensuring reproducible results even with low %B or at microbore flow rates.

**DR5 binary or ternary: for highest performance**

For outstanding performance at gradient extremes, with minimum delay volume and virtually instantaneous pump response, DR5 is the right choice. Run 1 to 99% B gradients at only 100 µl/min and know that the composition change will reach the column in under three minutes. With the same SDS, go from 1 to 99% at 5 ml/min in just 30 s.

The remarkable performance of DR5 SDS is achieved by giving each solvent channel its own metering pump. Up to three solvents are instantly and constantly blended in the LPC, exactly as you request. Program whatever gradient shape you need, and generate it accurately and reproducibly – every time.

*Choose your sampling modules*

**Faster, better sample preparation**

These sampling modules have been carefully developed to provide much more than precise and reproducible sample injection. They can free laboratory staff from a great deal of routine sample preparation and injection tasks.

**Versatility**

Manual injectors are available with loops for economic, yet precise, injections. Automated injections, using the injection volume specified in your separation method, can be taken from a 100-position autosampler, cooled if you wish.

**Challenge**

- Maintain low dispersion
- automate sample preparation
- ensure reproducibility
- handle small samples
- automate derivatization chemistries
- control sample environment.

**Solution**

- Provide a choice of injectors with minimized dead volume
- Manual valves



- A fully automatic sample injector with programmable injection cycle
- Control of reaction temperatures before injection
- A random-access autosampler, cooled if needed.

### **Advantages**

The sampling modules give the system superb automation capabilities, hand-in hand with low peak broadening to satisfy the demands of LDLC. Hewlett-Packard's ideal combinations of hardware and software make fully-automated analyses both versatile and convenient.

### **Manual Injection**

When full automation is not required, manual valves provide an economic alternative. Start all the modules in the system by the turn of a valve.

The Rheodyne Model 7125 injection valve, with loop sizes from 5  $\mu\text{l}$  to 100  $\mu\text{l}$  and partial loop filling capability, is the most economical choice. For the very lowest external band-broadening, choose the Model 7413 sample injection valve, with interchangeable loops of 0.5  $\mu\text{l}$ , 1  $\mu\text{l}$  and 5  $\mu\text{l}$ .

### **Stream sampling**

Repeated injections from an external source can be elegantly automated using the pneumatically-powered stream-sampling valve. This is the ideal solution for autoloading of samples during on-line monitoring. With an injector port mounted at the side of the LC, the valve is a clean interface to external sample handling.

### **The auto-injector**

With a choice of precision syringes, the auto-injector offers a wide range of fully programmable injection volumes. The 25- $\mu\text{l}$  syringe provides the highest accuracy and precision: a 1  $\mu\text{l}$  injection is possible from as little as 5  $\mu\text{l}$  of total sample. For larger volumes, a 250- $\mu\text{l}$  syringe is easily installed. The auto-injector fully meets the demands of LDLC.

In normal mode, solvent flows up sample loop and through the injector needle. The system is always clean and ready for injection.

To load the sample, the rotary valve connects the needle to the syringe, while the flow is bypassed directly to the column.

The needle is raised and the vial moves into position beneath it. The needle is lowered into the vial and the sample drawn up into the loop. A stepping motor drives the syringe installed, each step draws up 7 nl of sample giving excellent precision of injection volume.

With the sample volume now enclosed in the loop, and the needle reseated, the auto-

injector returns to the normal mode, injecting all of the sample onto the column as an undiluted plug.

### **The auto-sampler**

For maximum throughput, combine the auto-injector with an auto-sampler. With fully programmable, fast, random sample access, analyze up to 100 samples any number of times, in any order, with any method.

Prepare your samples in advance, and fill them directly into any of the 10 removable trays. Reserve magazines for calibration standards and store them until needed, in a refrigerator if necessary.

When the time comes to analyze, install the magazines in the cooled autosampler to ensure that precious samples do not degrade and are protected during the sequence. Results from your last vial will be as reliable as those from the first.

### **Special Applications**

Each of the discrete steps of the injector are programmable using the injector program. Control of needle and sample vial movement enables manipulations before injection. Dilute or extract your sample, adjust the pH, or include another substance as an internal standard, simply by adding a second volume automatically before injecting onto the column. When you choose the auto-sampler, it's like hiring another pair of hands.

Fully automatic precolumn derivatization at no extra cost. Employ the injector program, select from derivatization reagents, mix them together with your sample, all in the injector loop and finally inject the mixture. All done automatically as part of the LC method.

Optimize your chemistry. Keep a check on reaction rates by controlling the temperature. Increase the yield by accelerating reactions in the thermostatically controlled micro-oven.

### **Consistency, quality and reproducibility**

Whether you use standard, high-speed, or special applications columns, you need consistency, quality and reproducibility. And these are the features that most distinguish Hewlett-Packard columns.

### **Performance at elevated temperatures**

The column should clearly operate at full efficiency. Column temperature is increasingly used as a separation parameter. But temperature gradients within the column are detrimental to column performance. That's not simply a problem of column environment: the mobile phase plays a vital role.

### *For optimum column performance*

#### **Challenge**

- Ensure negligible extra-column volumes
- Support all column types and sizes
- Eliminate analytical errors due to temperature fluctuations
- Minimize temperature gradients within the column
- Minimize temperature effects of changes in flow rate and sample size.

#### **Solution**

- Provide a full range of columns
- Maintain consistency, quality and reproducibility
- Preheat the mobile phase
- Equalize the temperature of the mobile phase and column

#### **Advantages**

Use a column switching valve to match the column automatically to your separation problem. Run present methods unchanged AND explore the many benefits of microbore and high-speed LC. The temperature-controlled column compartment guarantees the stability and homogeneity of column temperature. Take full advantage of the marked influence of temperature on selectivity and plate number.

### *Hewlett-Packard HPLC columns*

#### **Hewlett-Packard offers columns for a wide variety of applications, including:**

- standard columns for your established methods
- cartridge columns for an economic alternative
- microbore columns for improved sensitivity and reduced solvent consumption
- high-speed columns for high sample throughput and improved sensitivity
- specialty columns for size exclusion chromatography (SEC), covering aqueous and organic solvents
- columns for demanding protein and peptide separations
- guard columns to protect your analytical column from compounds that are irreversibly absorbed or from particulate matter in suspension
- sample clean-up columns for very dirty sample, such as serum.

#### **Column compartment**

The column compartment is close to both the injector and detector to minimize peak-broadening. The compartment has a large capacity, accommodating any configuration of columns. Columns can easily be installed on the integrated racks. If a leak occurs, the pump is automatically switched off and solvents are drained safely to waste.

### **Temperature control**

Temperature homogeneity within the column is a vital part of separation efficiency. Radial and axial temperature gradients significantly reduce the performance of the column.

Preheating the eluent as it enters the compartment equalizes the temperature of the mobile phase and the column, at any flow rate and for any sample size. The injector delivers the mobile phase through a 0.12 mm i.d. capillary embedded in the heat exchanger. With such a narrow capillary the volume is only 2  $\mu\text{l}$ , ensuring minimum peak-broadening.

For a completely homogeneous environment, air with the same temperature as the mobile phase is circulated inside the compartment. Thermostatic control and a fan ensure a temperature precision of  $\pm 0.5$  °C, essential for the reproducibility of retention times. The heat exchanger never exceeds 140 °C, an important safety consideration when using flammable solvents.

For column temperatures below ambient, an external cooling bath can be connected. Temperatures in this range are maintained with the same accuracy as at higher levels.

### **Column switching valve**

Use of a column switching valve greatly increases the application range.

Match columns with different sample types. The column switching valve gives you this flexibility.

Make the column a part of your analytical method. Run a series of analyses on one column. When the next method is automatically loaded in sequence operation, the right column is switched into the flow path.

Inject the sample onto the pre-column and trap the part of interest. Then switch the analytical column into the flow-path, flush the remaining sample onto the column and start your separation.

Connect pre-column and analytical columns in series. After valve switching, flow through the analytical column continues in the normal direction. The pre-column is back-flushed, eluting highly retained peaks directly to the detector.

### **The choice is yours**

Precise isocratic and gradient solvent delivery modules lay the foundation for excellent reproducibility.

Sample preparation and introduction can be manual or fully automated, according to your needs.

Choose HP columns to fit your chromatographic needs and you can rest assured that they will operate at full efficiency in a stable column environment.

Select the economic filter-photometric detector or the sensitive diode-array detector to confirm peak purity and identity with confidence.

Operate the complete system from the built-in controller or from the optional HPLC ChemStation (DOS Series).

Evaluate your data locally on an integrator or a PC, or send data to a remote laboratory automation system for evaluation and archival.

## ***SPECIFICATIONS***

### **Solvent delivery system**

#### **Configurations**

Isocratic, PV5 ternary or quaternary, DR5 binary or ternary.

#### ***Ranges***

**Pressure:** 0-400 bar (0-5880 psi).

**Flow:** 0-5000  $\mu\text{l}/\text{min}$ , in 1  $\mu\text{l}/\text{min}$  steps

**Flow Accuracy:**  $\pm 2\%$  of setpoint or  $\pm 1\ \mu\text{l}/\text{min}$ .

Whichever is greater, independent of backpressure and solvents, excluding volume contraction effects.

**Precision:**  $< 0.3\%$  RSD, based on retention time.

#### ***Composition***

**Precision PV5:**  $\pm 0.25\%$  absolute, peak to Peak, water/acetonitrile, 500  $\mu\text{l}/\text{min}$ , 2500  $\mu\text{l}/\text{min}$ .

**Precision DR5:**  $\pm 0.15\%$  absolute, peak to peak, water/acetonitrile, 50  $\mu\text{l}/\text{min}$ , 2500  $\mu\text{l}/\text{min}$ .

#### ***Delay Volume***

**PV5:** 1750-2000  $\mu\text{l}$  maximum. Dependent on backpressure.

**DR5:** 300-500  $\mu\text{l}$  maximum, dependent on backpressure

#### **Heated Compartment**

Solvent and samples are preheated in low-volume (2  $\mu\text{l}$ ) heat exchanger with optional

cooling for subambient operation.

**Settable Temperature range:** 0-100 °C in 0.1 °C steps.

**Controlled temperature range:** 20 °C above ambient to 100 °C.

**Temperature stability:** ± 0.5 °C.

Injection system

Maximum pressure: 400 bar (5880 psi).

Manual Injector

Rheodyne valves 7125 or 7413, with loops from 0.5 µl to 100 µl.

### ***Auto-injector***

**Injection volume:** – 25 µl (standard), or 0.1- 250 µl (optional), in 0.1 µl steps

### **Precision**

<1% (typically 0.6%) RSD from 2-25 µl (25 µl syringe);

<1% (typically 0.6%) RSD from 10-250 µl (250 µl syringe)

**Sample vials:** 0 ml standard, 300 µl disposable microvial, 100 µl microvial.

**Minimum Sample volume:** 40 µl (2 ml standard vial), 20 µl (300 µl disposable microvial), 5 µl (100 µl microvial).

**Auto-sampler:** 10 removable magazines with 10 vials each (standard or microvials), with random access time < 10 seconds from any position.

Filter- photometric detector

Filters

Interference: 254 nm standard, 210, 230, 269, 280, 340, 430 and 540 nm optional.

Bandwidth: 10 nm typically.

### ***Signal Characteristics***

**Noise:** 5 x 10 E-5 AU.

**Drift:** < 1 x 10 E-3 AU/hour.

### *Signal Ranges*

**Linear absorbance range:** -0.1 to 1.5 AU.

**Response time:** 100, 400 and 800 ms (10-90%).

### **Flow Cell**

Stainless steel cell with 4.5 µl volume, 6 mm path length and maximum pressure of 100 bar (1470 psi).

Diode-array detector

Up to eight signals with definable sample and reference wavelength and bandwidth.

### *Signal Characteristics*

**Noise:**  $\pm 3.5 \times 10^{-5}$  AU.

**Drift:**  $< 2 \times 10^{-3}$  AU/hour.

### *Signal ranges*

**Wavelength range:** 190-600 nm, in steps of 1 nm.

**Bandwidth range:** 4-400 nm, in steps of 1 nm.

**Linear absorbance range:** -0.1 to 1.5 AU.

### *Spectral ranges*

**Wavelength range:** 190 – 600 nm.

**Linear absorbance range:** -0.1 to 1.5 AU.

**Scan Time:** 10 msec from 190-600 nm.

### *Flow cells*

**Standard cell:** 8 µl, 6 mm, max pressure 120 bar.

**High sensitivity cell:** 13 µl, 10 mm, max pressure 120 bar.

**Slits:** 2,4, and 8 nm selectable.



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