



Lab Highlights

LAH 0118

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FLOW RATE EFFECTS UPON TRACE ENRICHMENT WITH SEP-PAK® CARTRIDGES

Too often we assume that since off-line sampling devices like SEP-PAK^R cartridges are a simple tool they, therefore, require relatively little skill or attention to detail to use successfully. SEP-PAK^R cartridges, however, do require attention to detail for successful operation.

How often have you observed a researcher taking 10 ml of sample and "squirting" the volume through the SEP-PAK^R cartridge. Unfortunately "squirting" can be at a flow rate ranging from 200 ml/min to 30 ml/min, and 30 ml/min is the maximum recommended flow rate.

THE EXPERIMENT. We decided to investigate the effect of flow rate to illustrate the potential problems with real samples. We chose to "Trace-Enrich" riboflavin from 10 ml of an aqueous solution. A new SEP-PAK^R cartridge was used (properly conditioned*) for each experiment. Four individual flow rates were chosen: gravity flow, 1.0 ml/min, 10 ml/min and 27 ml/min.

Remember that dispensing 10 ml through a SEP-PAK^R cartridge takes 10 min at 1 ml/min, 1 min at 10 ml/min, and 22 seconds at 27 ml/min. The "squirt" is significantly faster than this last flow rate.

After the trace enrichment, the riboflavin was eluted from the SEP-PAK^R cartridge by washing with 2 ml of methanol, and the effluent was analyzed for riboflavin by LC. The table below indicates riboflavin recovery. The appearance of the yellow band of riboflavin as it traveled through the column was recorded also.

RECOVERY OF RIBOFLAVIN

<u>Flow Rate</u>	<u>% Recovery</u>	<u>Visual Observation of Yellow Band (Riboflavin) on SEP-PAK® cartridge</u>
Gravity	100%	very narrow (≈ 0.1 cm)
1 ml/min	100%	narrow (≈ 0.2 cm)
10 ml/min	100%	broad (≈ 0.4 cm)
27 ml/min	95%	very broad--cartridge totally yellow

CONCLUSION. SEP-PAK^R cartridge performance in trace enrichment can be flow rate dependent over the range which we recommend for operation. Flow rate through SEP-PAK^R cartridge is a variable which should be controlled (remember the relationship of plate height, "h", vs. linear velocity, "u", in LC). This is not surprising since band broadening is greater at higher flow rates. The implication to SEP-PAK^R cartridge usage is that results could be irreproducible and/or inaccurate if flow rate is not controlled with a "proper" range. In this example 1 ml/min to 10 ml/min would be considered "proper" as it resulted in 100% recovery. Each sample will probably be unique.

* See SEP-PAK^R cartridge Care and Use Manual

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