Hints for the Capillary Chromatographer

The Importance of Inlet Sleeve Deactivation

Chromatographersfrequently call with questions about sleeve deactivation. Due to the popularity of this topic, we have decided to rerun the article on "The Importance of Inlet Sleeve Deactivation "from September 1990s issue of the Restek Advantape.

Your entire sample pathway must be inert
Capillary columns are inherently more inert than packed columns. However, capillary chromatographers still experience breakdown and adsorption of active compounds. Inertness of the sample pathway is particularly important with certain pesticides, herbicides, drugs, amines and acids. If the column itself is inert, then what is the source of the problem? One major area that most chromatographers overlook is the inlet. The inertness of the inlet sleeve is very important since it is part of the sample pathway and can often be the source of adsorption or breakdown. If the sleeve is not deactivated properly, adsorption can take place in the sleeve and alter the composition of the sample reaching the column. Also, when analyzing compounds at low levels, adsorption or decomposition of some components in the sleeve results in poor quantitation and even misidentification.

Is **it important to deactivate the sleeve?**
For most samples, the answer to this question is a resounding yes. We tested the adsorptive characteristics of an undeactivated versus a deactivated sleeve (Figure 1). Endrin, an active pesticide, was injected into an untreated and a properly deactivated inlet sleeve. The undeactivated sleeve showed 98% breakdown of a 50pg injection of endrin into its respective degradation products, endrin aldehyde and endrin ketone. The deactivated sleeve showed only 6% breakdown.

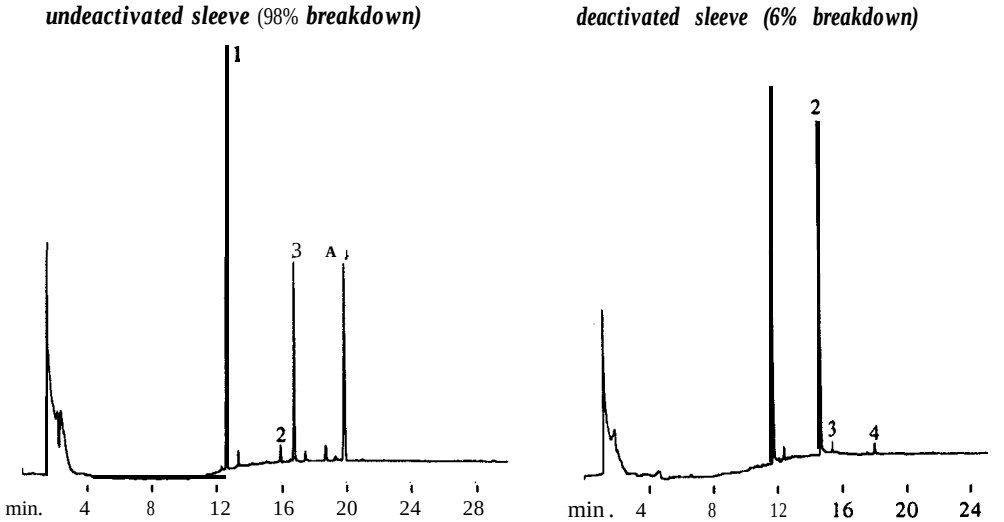
In addition to breakdown, an improperly deactivated sleeve can cause irreversible adsorption. To demonstrate this effect, we compared the response factors of three active compounds, 2,4-dinitrophenol, pentachlorophenol, and benzidine on both undeactivated and properly deactivated sleeves. Table I shows that the response factors for all three compounds are lower on the undeactivated sleeve than on the deactivated sleeve.

Table I
Average response factors relative to naphthalene of 3 injections.

	2,4-dinitrophenol	pentachlorophenol	benzidine
deactivated	0.248	0.240	0.327
undeativated	0.185	0.188	0.234

Why do response factors change for active compounds?
A priming effect occurs when improperly deactivated sleeves are used for adsorptive samples. We injected 1 .0ul of an endrin standard onto a column and calculated the percentage of breakdown. This first injection primed the sleeve with endrin. When we injected the endrin standard onto the column again, the result was less endrin breakdown. This procedure can be

Figure 1 - Comparison of endrin breakdown on an undeactivated and a deactivated Cyclosplitter@.



15m, 0.53mm ID, 1.0um Rtx-5 (cat. #10252), splitless injection
Oven temp.: 150"C to 275°C @ 4"C/min. ,hold 15 min.
Inj. temp.: 250°C
Det. temp.: 300°C
Carrier gas: helium
Linear velocity: 2 lcm/sec.
ECD sensitivity: 16 x 10"' AFS

repeated until the percentage breakdown remains constant for three injections. If you are running calibration curves and your numbers are jumping around in this manner, your inlet sleeve may not be adequately deactivated.

How **do you know that your sleeve has** been properly deactivated?
The only true test of a sleeve's inertness is in its actual performance with active compounds. A visual inspection of the sleeve will not tell you that the sleeve has been deactivated. In fact, most instrument manufacturers do not deactivate their inlet sleeves.

Are you using a home-brewed sleeve deactivation recipe? Many chromatographers are aware that their sleeves are not deactivated and develop many different recipes to treat their sleeves. The most common treatment is with dimethyldichlorosilane (DMDCS). The use of this material presents several problems. It reacts with moisture to produce HCl vapors, so treatments with DMDCS must-be done in a well-ventilated area. Also, DMDCS, if not properly end capped, can react with humidity in air, resulting in weakened effectiveness.

We examined the quality of in-house deactivation procedures by comparing several home-brew recipes commonly used. We looked at acid treatment, DMDCS treatment, acid and DMDCS treatment, and finally our own proprietary deactivation procedure. We evaluated each of these procedures in duplicate using a CyclosplitteP. The resulting endrin breakdown from these five treatments is shown in Table II. The results clearly show that all deactivation techniques are not equal and a highly inert surface requires a procedure combining both acid and DMDCS treatments.

Table II
% endrin breakdown with various deactivations

Treatment	Run #l	Run #2
Acid only	**36%**	**45%**
DMDCS only	20.8%	18.3%
Acid and DMDCS	19.8%	18.5%
Restek's Old Deactivation*	10.0%	15%
Restek's New Deactivation	2.2%	1.2%

% endrin breakdown calculation:

Σ areas = endrin area + endrin aldehyde area + endrin ketone area

$$\% \text{ total breakdown} = 100 \times \frac{\text{aldehyde area + ketone area}}{\Sigma \text{ area}}$$

* Prior to April 1991.

Will glass wool or beads in your sleeve cause adsorption and breakdown?
Yes. Many chromatographers who analyze dirty samples use a plug of glass wool or beads in their sleeve to act as a filter. However, both wool and beads greatly increase the surface area that the sample contacts and can be a source of adsorption or breakdown, It is critical that the wool or beads be properly deactivated. Even if your wool is deactivated, active sites can be created as the fibers break when inserting the wool into the sleeve. If you plan to use wool, be sure that it is thoroughly deactivated and use care when inserting it into the sleeve.

Can sleeves be cleaned without redeactivation?
Sleeves only need to be redeactivated if the deactivation layer is removed during cleaning. Daily GC use deposits septa particles, sample residue, and pyrolizates on the sleeve surface, contaminating your inlet. One way to prolong the lifetime of your deactivated sleeve is to wash out the sleeve with methanol (since methanol does not swell septa or make them sticky). Nylon tube brushes and pipe cleaners are also helpful for removing small septa fragments from dirty sleeves. ***However, be careful to avoid scratching the liner with the inner metal core of the brush. You*** can then use stronger solvents such as methylene chloride, hexane, or the solvent your sample is diluted in, to remove additional contamination. These mild procedures will not require redeactivation.

If the contamination has been pyrolized onto the sleeve, solvent rinsing or other simple cleaning procedures may still leave a residue. The only effective procedures to remove this residue are strong acids, bases, detergents, or baking in a muffle furnace. If you use these harsh procedures, you will remove the deactivation layer, requiring redeactivation.

Analyzing active compounds is a difficult assignment. If you properly deactivate your inlet sleeve you will find many of your analysis problems solved. For more information, please call our technical service group at 800-356-1688, ext. 4. ■

Suggestions?

Is *there a topic you would like to see covered in "Hints for the Capillary Chromatographer* ***If so,*** *please call our technical service department toll-free at 800-356-l 688 (ext. 4).*