Optimizing the Capillary GC Separation of Acids, Esters, and Other Flavor Components in Distilled Liquor Products

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ABSTRACT

Distilled liquor products contain a wide range of volatile and non-volatile compounds in an ethanol/water matrix. The most abundant fusel alcohols and esters can be determined by simple split injection, which also minimizes the amount of matrix ethanol and water transferred to the column. However, many additional trace fatty acids and their esters, which often are used to indicate product quality in alcoholic beverages such as whiskey and rum, cannot be determined by this approach. Capillary gas chromatography is a powerful tool for the analysis of these compounds, but the large ranges in volatilities and acidities can make it difficult to quantitate all of the components in a single chromatographic separation. In addition, because the concentrations can vary widely, splitless injection techniques with some type of preconcentration step often are necessary. One example of this is a large volume injection with a venting step, which can be optimized to remove most of the matrix ethanol and water.

Using a bonded polyethylene glycol (PEG) capillary column, flavor compounds in distilled liquor products can be quantitated in a single splitless injection. The Stabilwax™-DA column was selected for this application. To improve peak shape and reproducibility of acidic components on this column, an acidic functionality has been added to the backbone of the PEG stationary phase. This results in less adsorption of acidic components and significantly less peak tailing. In addition, the chromatographic separation of these compounds has been optimized to allow for the rapid analysis of fatty acids, esters, and other trace level components. An optimized configuration of 30m, 0.18mm ID, 0.18µm allows for significantly reduced analysis times. An interesting application is in the determination of flavor compounds in alcoholic beverages such as malt whiskeys and grappas.

PROCEDURE

To optimize the chromatographic conditions for this analysis, a test mixture containing acids, esters, and flavor compounds typically found in alcoholic beverages was prepared (see Figure 1). A computer modeling program, ezGC™, was used to optimize the column configuration, temperature program, and inlet flow for this system. Based on this, an optimized configuration of 30m, 0.18mm ID, 0.18µm was developed. To test the applicability of this column in these dimensions, the critical pair of caproic acid and ethyl laurate was studied. These components can be very difficult to resolve on standard Carbowax®-type columns. This is especially true if peak tailing or broadening occurs, or if one component is present at a significantly higher concentration. The Stabilwax™-DA column achieves baseline resolution of these two compounds within a reasonable analysis time of 13 minutes (Figure 2).

Because alcoholic beverage samples often are injected via splitless mode, the stability of the Stabilwax®-DA column when exposed to aqueous injections is important. We verified stability by performing a splitless injection of the alcoholic beverage test mix, followed by five 1µL injections of water. This process was repeated 10 times, followed by a final injection of the test mix. The final test mix injection can be seen in (Figure 3). Even after repeated splitless injections of 100% water, very little degradation occurs in the peak shapes of the test mix components. Over the course of the study, the variation in the peak retention times was 0.08-0.22% RSD. This includes the polar free fatty acids, which can be difficult to analyze under ideal conditions. The excellent stability of this stationary phase is proven by the reproducibility of the retention times over the course of the water stability study.

ANALYSIS OF MALT WHISKEY

Whiskey is distilled from a fermented mash of grain, such as corn, rye, barley, or wheat. Aging of the whiskey takes place within barrels or casks, and it is during the aging process that whiskey obtains its characteristic color, flavor, and aroma. Factors that influence the flavor of the final product include the characteristics of the grain, recipes, and how the whiskey is distilled. The flavor profile of a whisky contains hundreds of compounds, including fatty acids, esters, alcohols, and aldehydes. An example of a malt whiskey profile, as determined by GC/MS, can be seen in (Figure 4).

ANALYSIS OF GRAPPA

Grappa is the spirit produced from grape marc, or the skins of the grapes after they have been pressed during wine production. This grape marc is fermented and distilled either directly or by water vapor. Grappas generally do not require the same amount of aging as other alcoholic beverages, although, for example, Italian law requires at least six months of aging. Flavored grappas can be produced by adding ingredients such as herbs and fruits. The flavor profile of grappas contains hundreds of compounds at a wide range of concentrations. The chromatographic profile of a sample of grappa can be seen in (Figure 5).

SUMMARY

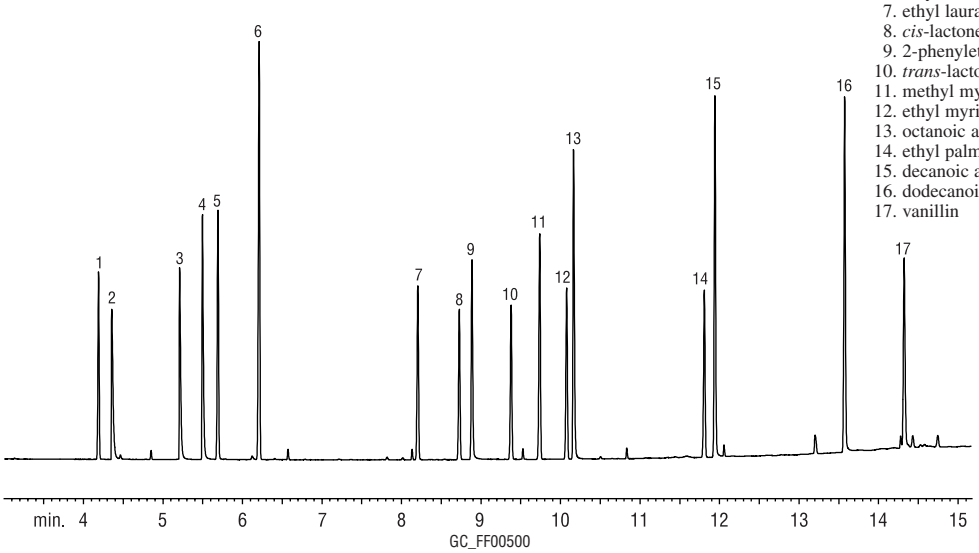
The Stabilwax®-DA column is an excellent choice for the analysis of acids, esters, and other flavor components in alcoholic beverage products. This highly stable column has been optimized for the analysis of acidic compounds, making it possible to analyze a wide range of compounds in a single injection. In addition, the column configuration shown in this article allows fast, efficient analysis of complex products such as malt whiskeys and grappas.

Large volume injection techniques allow for the analysis of a wide range of concentrations in a single run. As shown in this paper, analytes at higher concentrations such as alcohols and esters and trace level flavor compounds can be analyzed simultaneously. Large volume injections on the order of 10-100µL can be used for analyses of this type. The venting step during the large volume injection can be optimized to remove most of the ethanol/water matrix. Since some water will enter the chromatographic column, a stabilized phase such as the Stabilwax®-DA should be used.

Figure #1

Alcoholic beverage test mixture on the Stabilwax-DA™ capillary column.

Peak List	Conc. (ppm)
1. ethyl octanoate	100
2. acetic acid	100
3. propionic acid	100
4. isobutyric acid	100
5. decanol 3	50
6. ethyl decanoate	50
7. ethyl laurate	50
8. *cis*-lactone	100
9. 2-phenylethanol	50
10. *trans*-lactone	100
11. methyl myristate	50
12. ethyl myristate	50
13. octanoic acid	100
14. ethyl palmitate	50
15. decanoic acid	100
16. dodecanoic acid	100
17. vanillin	100

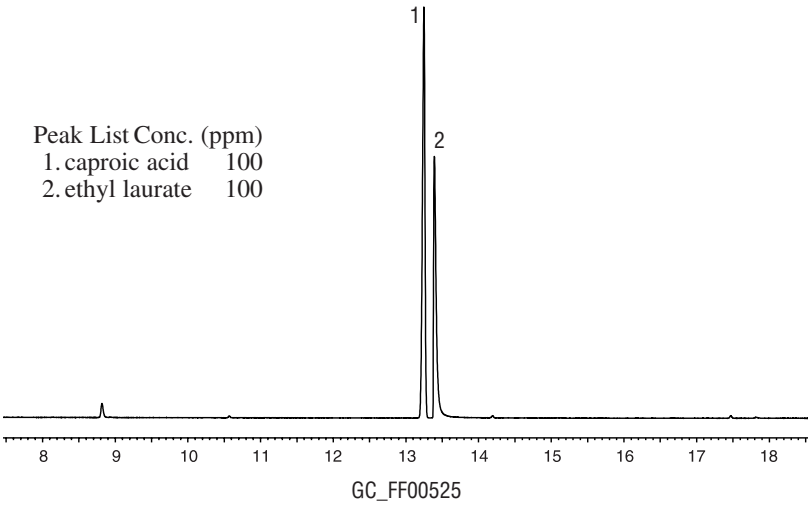


Stabilwax®-DA 30m, 0.18mm ID, 0.18µm (cat.# 550752)

Inj.:	1µL splitless (hold 0.5 min.) at conc. shown in peak list, in ethyl acetate, 4mm ID splitless liner w/wool (cat.# 20814-202.1)
Inj. temp.:	240°C
Carrier gas:	hydrogen
Make-up gas:	nitrogen
Linear velocity:	28psi @ 240°C
Oven temp.:	70°C to 240°C at 12°C/min. (hold 3 min.)
Det.:	FID

Figure #2

Complete resolution of caproic acid and ethyl laurate can be achieved in 13 minutes.

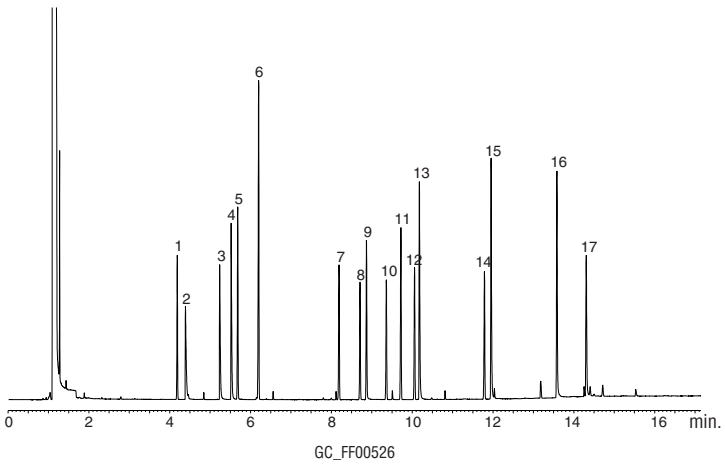


Stabilwax®-DA 30m, 0.18mm ID, 0.18µm (cat.# 550752)

Inj.:	1µL splitless (hold 0.5 min.) at conc. shown in peak list, in ethyl acetate, 4mm ID splitless liner w/wool (cat.# 20814-202.1)
Inj. temp.:	240°C
Carrier gas:	hydrogen
Make-up gas:	nitrogen
Linear velocity:	28psi @ 240°C
Oven temp.:	80°C to 230°C at 5°C/min.
Det.:	FID

Figure #3

Injection of the test mixture following fifty 1µL injections of water.



Peak List	Conc. (ppm)
1. ethyl octanoate	100
2. acetic acid	100
3. propionic acid	100
4. isobutyric acid	100
5. decanol 3	50
6. ethyl decanoate	50
7. ethyl laurate	50
8. *cis*-lactone	100
9. 2-phenylethanol	50
10. *trans*-lactone	100
11. methyl myristate	50
12. ethyl myristate	50
13. octanoic acid	100
14. ethyl palmitate	50
15. decanoic acid	100
16. dodecanoic acid	100
17. vanillin	100

Stabilwax®-DA 30m, 0.18mm ID, 0.18µm (cat.# 550752)

Inj.:	1µL splitless (hold 0.5 min.) at conc. shown in peak list, in ethyl acetate, 4mm ID splitless liner w/wool (cat.# 20814-202.1)
Inj. temp.:	240°C
Carrier gas:	hydrogen
Make-up gas:	nitrogen
Linear velocity:	28psi @ 240°C
Oven temp.:	70°C to 240°C at 12°C/min. (hold 3 min.)
Det.:	FID

Figure #4

Flavor profile of malt whiskey by GC/MS using a large volume injection technique and a Stabilwax®-DA capillary column.

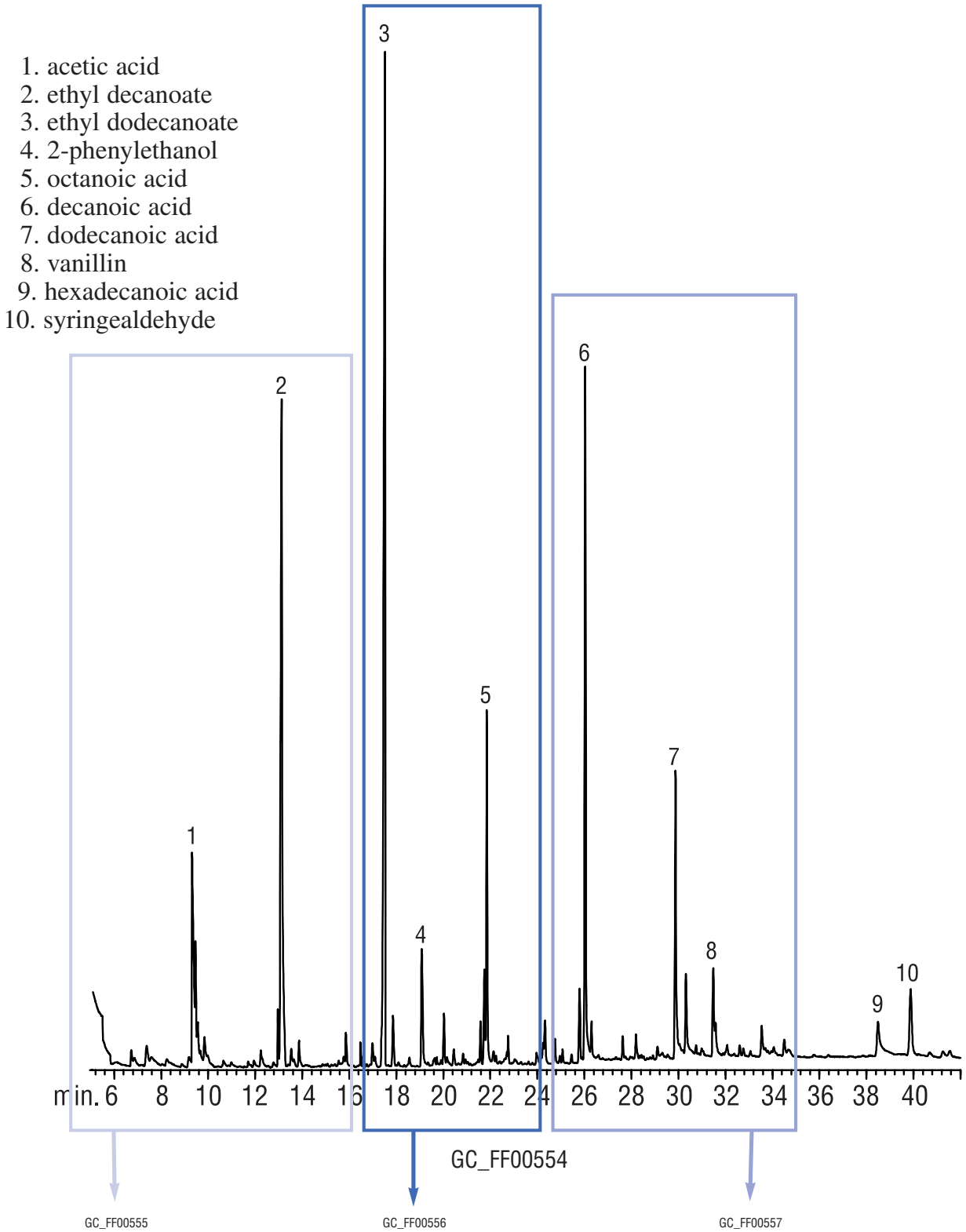
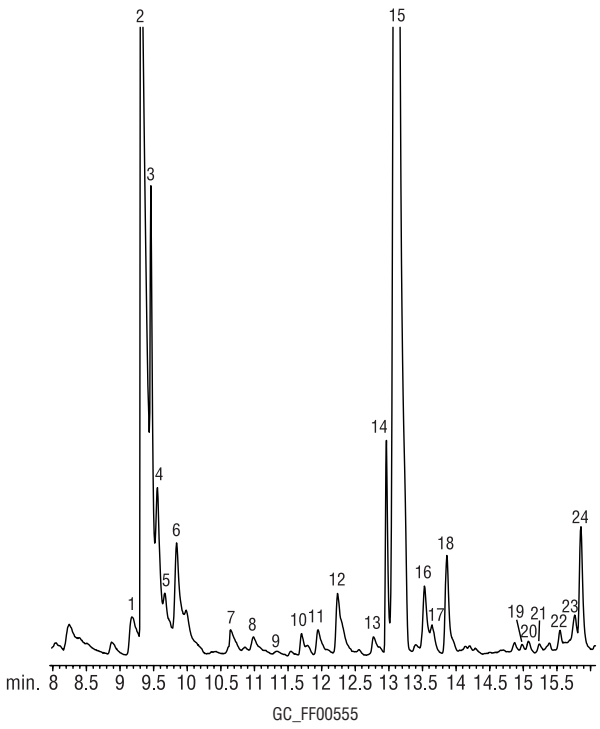
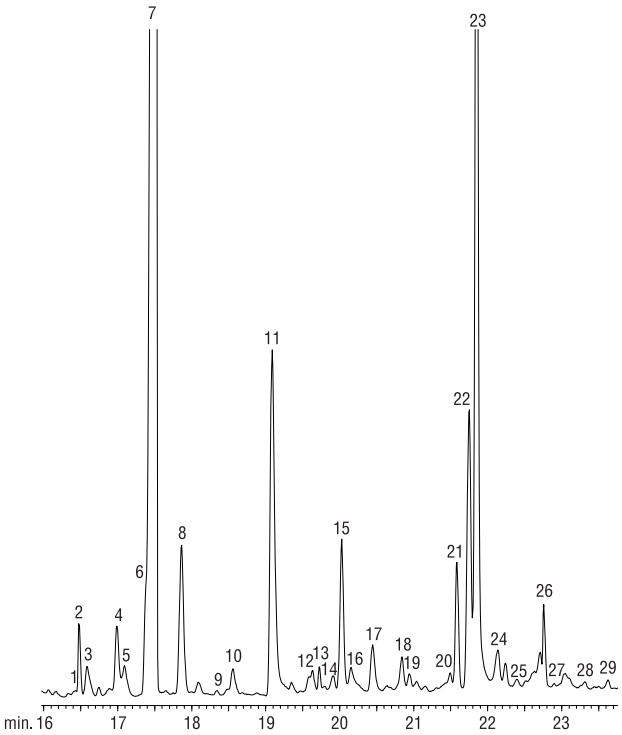


Figure #4 (cont.)



GC_FF00555

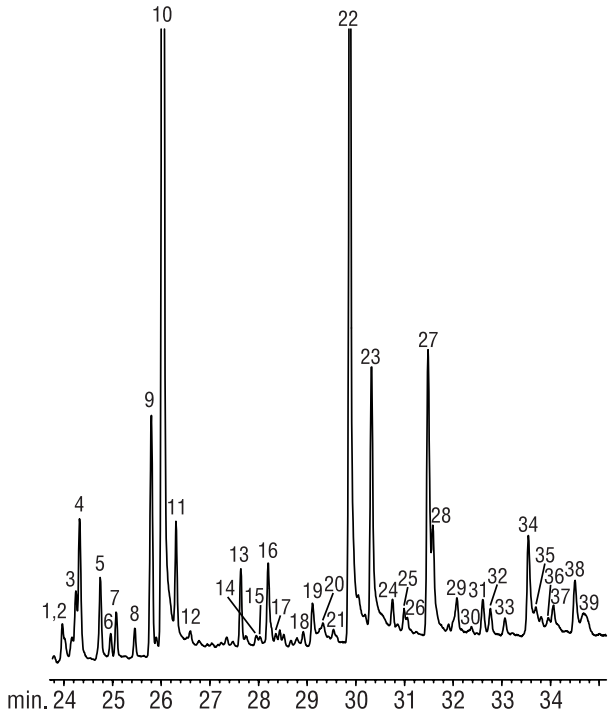
1. ethyl octanoate
2. acetic acid
3. siloxane
4. 1-hydroxy-2,3-butadione
5. 1-hydroxy-2-propanone acetate
6. furfural
7. formic acid
8. propionic acid
9. isobutyric acid
10. dimethyl sulfoxide
11. 5-methyl furfural
12. methyl decanoate + unknown
13. butyric acid
14. siloxane
15. ethyl decanoate
16. furfuryl alcohol + isoamyl octanoate
17. isovaleric acid + 2-methylbutyric acid
18. diethyl succinate
19. 3-methyl-2(5H)-furanone (possible)
20. valeric acid
21. ethyl undecanoate
22. isobutyl decanoate
23. 2(5H)-furanone
24. unknown
25. diethyl pentanedioate



GC_FF00556

1. di(ethyleneglycol) butyl ether
2. siloxane
3. methyl dodecanoate
4. phenylethyl acetate
5. methylcyclopentenolone
6. hexanoic acid
7. ethyl dodecanoate
8. isoamyl decanoate + guaiacol
9. dodecyl acetate (possible)
10. whiskey lactone (1)
11. 2-phenylethanol
12. heptanoic acid
13. siloxane
14. dimethoxybenzene or 4-methylguaiacol
15. whiskey lactone (2)
16. dodecanol
17. unknown
18. phenol
19. methyl tetradecanoate
20. nerolidol
21. diethyl malate
22. ethyl tetradecanoate
23. octanoic acid
24. unknown
25. *p*-cresol
26. siloxane
27. diethyl octanedioate
28. monomethyl succinate
29. 3,5-dimethyl-2,4(5H) furandione

Figure #4 (cont.)



GC_FF00557

1. nonanoic acid
2. diethyl 2-hydroxyglutarate
3 unknown
4. tetradecanol
5. 4-vinylguaiacol
6. diethyl nonanedioate
7. methyl hexadecanoate
8. ethyl γ-lactone 2-hydroxyglutarate
9. ethyl hexadecanoate
10. decanoic acid
11. ethyl 9-hexadecanoate
12. triacosan
13. unknown
14. phthalide
15. diethylphthalate
16. hexadecanol
17. 4-hydroxycinnamic acid (decomp.)
18. methyl stearate
19. benzoic acid
20. methyl 8-octadecenoate
21. ethyl stearate
22. dodecanoic acid
23. hydroxymethylfurfural
24. ethyl linoleate
25. 4-allyl-2,6-dimethoxyphenol
26. diisobutyl phthalate
27. vanillin
28. sinapic acid (decomp.)
29. 2-phenylethyl decanoate + 2 unknowns
30. 2-propenyl-2,6-dimethoxyphenol
31. ethyl vanillate
32. acetovanillone
33. vanillin methyl ketone
34. tetradecanoic acid
35. ethyl homovanillate
36. propiovanillone
37. fatty acid ester
38. (similar to 4-allyl-2,6-dimethoxyphenol)
39. unknown

Figure 4 conditions:

Stabilwax®-DA 30m, 0.18mm ID, 0.18µm (cat.# 550752)

Inj.:	10µL large volume injection (splitless), at 10µL/min.
Std. conc.:	neat
Gerstel CIS Injector:	35°C (hold 2 min.), to 300°C @ 10°C/sec. (hold 5 min.)
Helium vent flow:	600mL/min with 1.8 min. vent end time
Carrier gas:	helium
Linear velocity:	45cm/sec.
Oven temp.:	60°C (hold 2 min.) to 100°C @ 20°C/min., to 240°C @ 5°C/min. (hold 10 min.)
Det.:	MSD
Transfer line temp.:	240°C
Quadrupole temp.:	150°C
MS source temp.:	230°C
Scan range:	30–400amu
Ionization:	70eV
Mode:	EI

Chromatogram courtesy of Kevin MacNamara, Ph.D., Irish Distilleries, Ltd.

Figure #5

Flavor profile of grappa by GC/MS using a large volume injection technique and a Stabilwax®-DA capillary column.

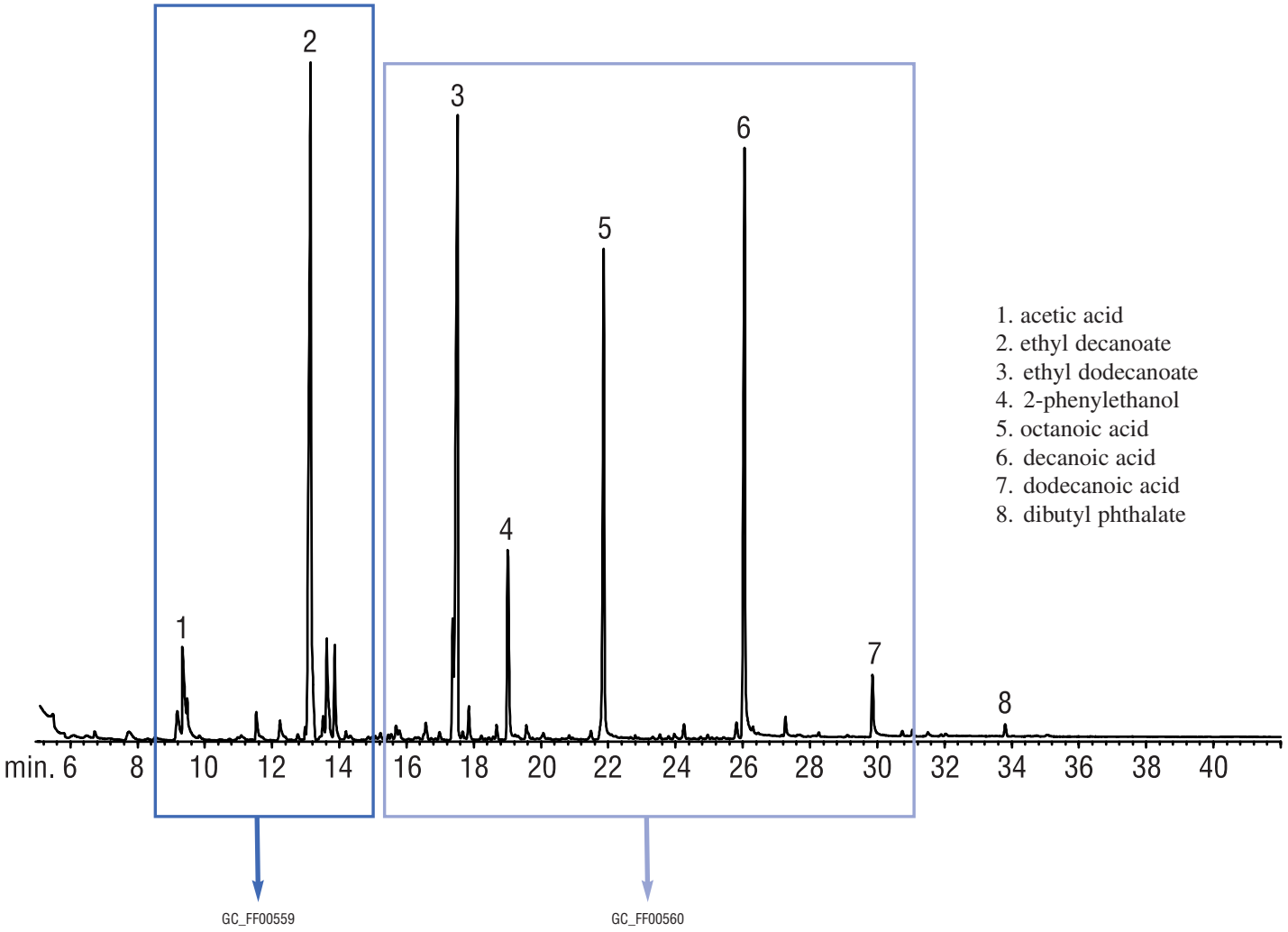
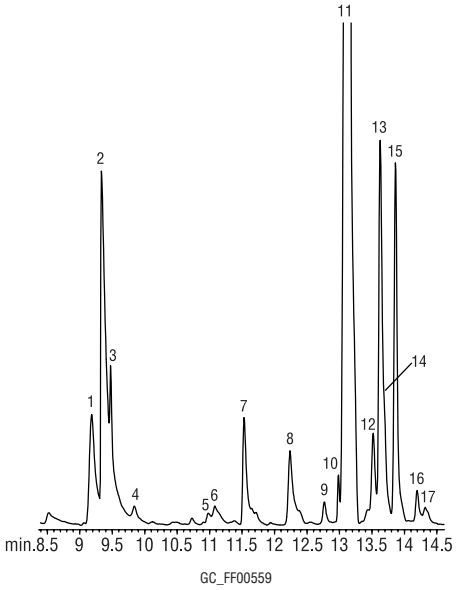
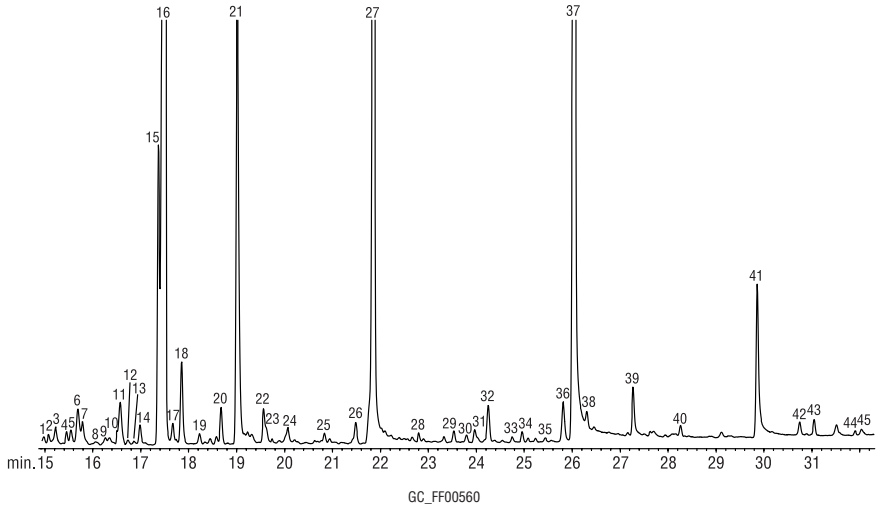


Figure #5 (cont.)





1. ethyl octanoate
2. acetic acid
3. siloxane
4. furfural
5. propionic acid
6. benzaldehyde + linalool
7. isobutyric acid
8. methyl decanoate
9. butyric acid
10. siloxane
11. ethyl decanoate
12. isoamyl octanoate
13. isovaleric acid + 2-methylbutyric acid
14. susquiterpene (shoulder)
15. diethyl succinate
16. ethyl 9-decenoate
17. α-terpineol

1. (possible 3-methyl-2(5H)-furanone)
2. valeric acid + susquiterpene
3. linalool oxide
4. susquiterpene
5. isobutyl decanoate
6. 1-decanol
7. β-citronellol
8. susquiterpene
9. methyl salicylate + unknown
10. siloxane
11. methyl dodecanoate
12. hexyl octanoate + 2-tridecanone
13. *trans*-2, *trans*-4-decadienol
14. 2-phenylethyl acetate
15. hexanoic acid
16. ethyl dodecanoate
17. unknown
18. isoamyl decanoate
19. benzyl alcohol
20. unknown
21. 2-phenylethanol
22. 2-ethylhexanoic acid
23. heptanoic acid
24. dodecanol
25. phenol
26. γ-nonalactone
27. octanoic acid
28. siloxane
29. ethyl cinnamate
30. decalactone + unknown
31. nonanoic acid
32. phenylethyl hexanoate
33. diethyl nonanedioate
34. methyl hexadecanoate
35. dimethyl naphthalene
36. ethyl hexadecanoate
37. decanoic acid
38. ethyl 9-hexadecenoate
39. geranic acid
40. phenylethyl octanoate
41. dodecanoic acid
42. ethyl linoleate
43. diisobutyl phthalate
44. ethyl linolenate
45. phenylethyl decanoate

Figure 5 conditions:

Stabilwax®-DA 30m, 0.18mm ID, 0.18µm (cat.# 550752)

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Carrier gas:	helium
Linear velocity:	45cm/sec.
Oven temp.:	60°C (hold 2 min.) to 100°C @ 20°C/min., to 240°C @ 5°C/min. (hold 10 min.)
Det.:	MSD
Transfer line temp.:	240°C
Quadrupole temp.:	150°C
MS source temp.:	230°C
Scan range:	30–400amu
Ionization:	70eV
Mode:	EI

Chromatogram courtesy of Kevin MacNamara, Ph.D., Irish Distilleries, Ltd.