960769

# MILLENNIUM APPLICATION BRIEF

## TOPIC: Analysis of Chlorhexidine using an Alliance System

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### INTRODUCTION:

Chlorhexidine(CH) is a common main ingredient in a variety of pharmaceutical formulations. Analysis of chlorhexidine in a pharmaceutical formulation was done on a 2690 separations module using dial-a-mix for solvent blending and mixing. In the past premixed mobile phases were required on traditional HPLC's to allow for a reproducible retention time to be achieved. Using the 2690 and PDA detection analysis times of less than 6 minutes was possible. No degradation products were found in these samples. Figure 1 is a representative chromatogram of the separation. Twenty repetitive injections were done and overlaid as shown in figure 2. A retention time standard deviation of 0.018 minutes. Figure 3 is a system suitability report for those twenty injections showing little, if any, change in retention time. As further proof of the ability of the 2690 to reproducibly deliver this mix of mobile phase ten injections at 69/31 composition was done and the twenty injections at 70/30 and ten at 69/31 are overlaid in figure 4. As can be seen the 2690 was able to reproducibly deliver this mixture of mobile phase using dial-a-mix which helps to significantly reduce solvent handling, waste and variability between batches of premixed mobile phases.

### EXPERIMENTAL:

HPLC Analysis

| | |
|---|---|
| Mobile phase: | A: Phosphate/TEA pH=3 |
| | B: Acetonitrile |
| Flow rate: | 1.5 mL/min (70/30 A:B)-done using dial-a-mix feature on 2690 |
| Detection: | 239nm |
| Column: | Symmetry C-18 (3.9 X 250mm) |
| Temperature: | 40 degrees Celsius |

### FIGURES:

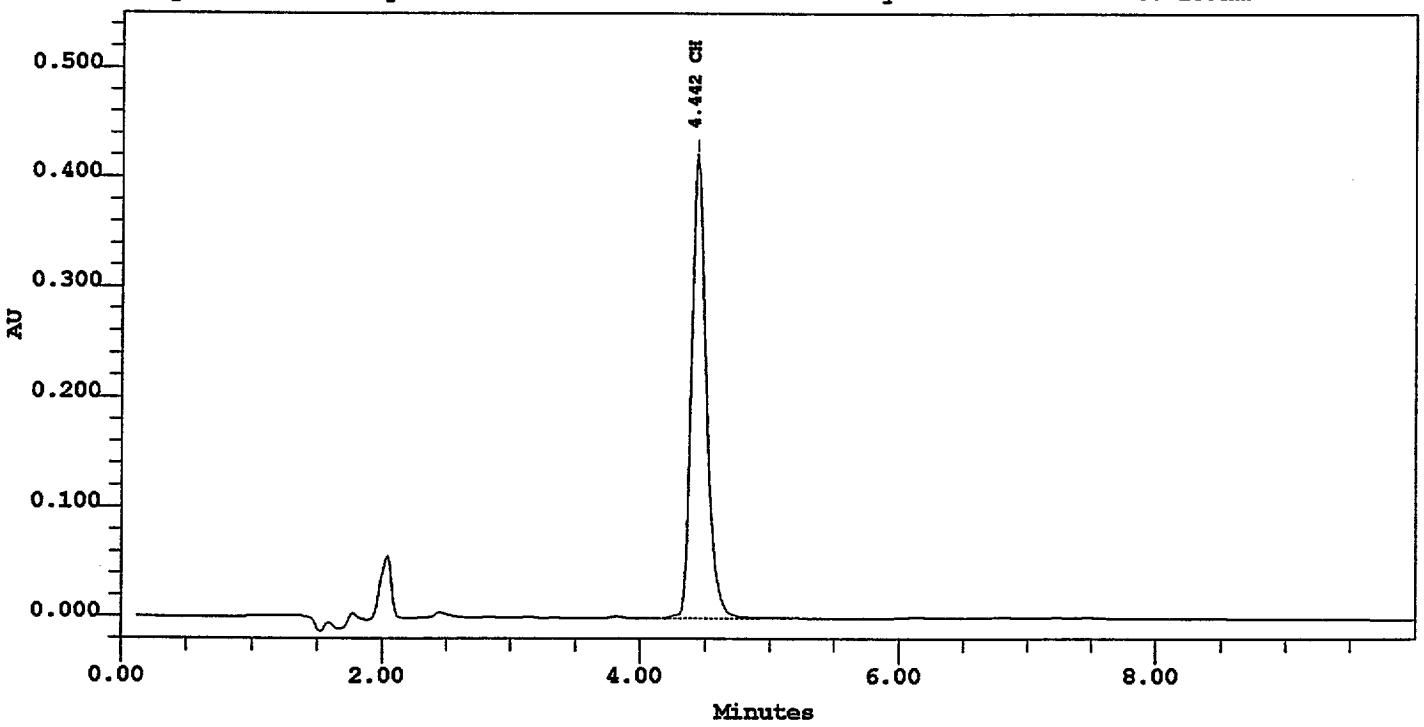
Figure 1: Chromatogram of CH separation
Figure 2: Overlay of twenty injections (20)
Figure 3: Summary report for twenty injections
Figure 4: System suitability report for twenty injections
Figure 5: Overlay of twenty injections at 70/30 and ten injections at 69/31

Waters Corporation

Sample Information

| | | | |
|---|---|---|---|
| SampleName: | CH Sample1 | | |
| Vial: | 2 | Sample Type: | Unknown |
| Injection: | 3 | Volume: | 50.00 |
| Channel: | 996 | Run Time: | 20.0 min |
| Date Acquired: | 03/24/96 07:12:07 PM | Date Processed: | 06/26/96 12:34:22 PM |
| Scale Factor: | 1.00 | Dilution: | 1.00000 |
| Acq Meth Set: | PG | | |
| Processing Method: | PG | | |

**SampleName: CH Sample1 Comments: Chlorhexidine analysis Channel Name: 239nm**

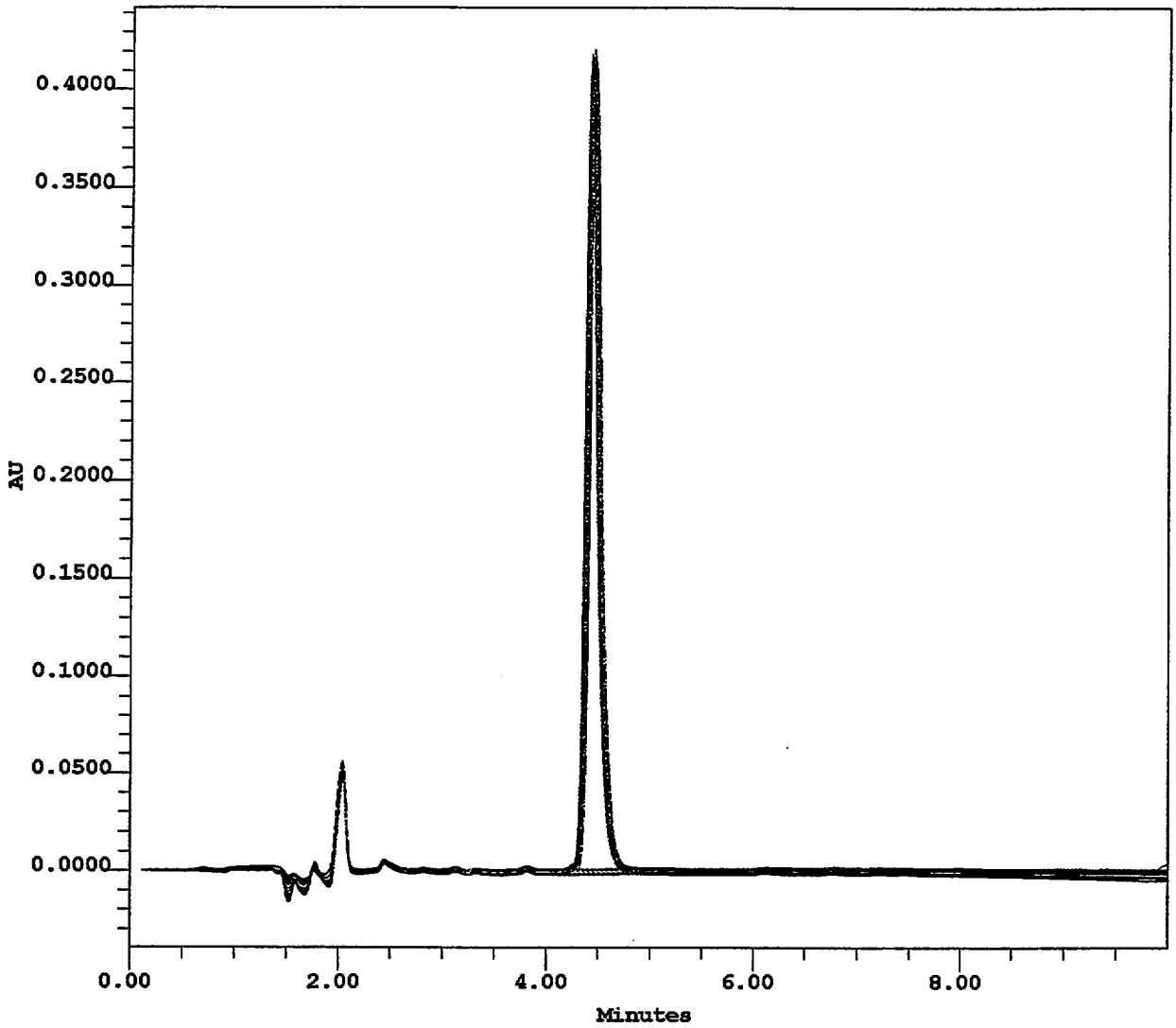



*Peak Results*

| # | Name | Ret Time (min) | Area (uV*sec) | Height (uV) |
|---|---|---|---|---|
| 1 | CH | 4.442 | 3368628 | 422768 |



fig. 1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Minutes



fig 2.

*Peak Results*

| # | SampleName | Vial | Inj | Channel | Peak Name | RT (min) | Area (uV*sec) |
|---|---|---|---|---|---|---|---|
| 1 | CH Sample1 | 2 | 1 | 996 | CH | 4.455 | 3357778 |
| 2 | CH Sample1 | 2 | 2 | 996 | CH | 4.443 | 3360567 |
| 3 | CH Sample1 | 2 | 3 | 996 | CH | 4.442 | 3368628 |
| 4 | CH Sample1 | 2 | 4 | 996 | CH | 4.445 | 3361721 |
| 5 | CH Sample1 | 2 | 5 | 996 | CH | 4.440 | 3358346 |
| 6 | CH Sample1 | 2 | 6 | 996 | CH | 4.422 | 3352490 |
| 7 | CH Sample1 | 2 | 7 | 996 | CH | 4.420 | 3351919 |
| 8 | CH Sample1 | 2 | 8 | 996 | CH | 4.423 | 3351279 |
| 9 | CH Sample1 | 2 | 9 | 996 | CH | 4.413 | 3350162 |
| 10 | CH Sample1 | 2 | 10 | 996 | CH | 4.405 | 3347209 |
| 11 | CH Sample 2 | 3 | 1 | 996 | CH | 4.412 | 3352885 |
| 12 | CH Sample 2 | 3 | 2 | 996 | CH | 4.420 | 3346730 |
| 13 | CH Sample 2 | 3 | 3 | 996 | CH | 4.442 | 3348195 |
| 14 | CH Sample 2 | 3 | 4 | 996 | CH | 4.423 | 3350461 |
| 15 | CH Sample 2 | 3 | 5 | 996 | CH | 4.438 | 3350464 |
| 16 | CH Sample 2 | 3 | 6 | 996 | CH | 4.460 | 3350794 |
| 17 | CH Sample 2 | 3 | 7 | 996 | CH | 4.463 | 3338322 |
| 18 | CH Sample 2 | 3 | 8 | 996 | CH | 4.455 | 3337006 |
| 19 | CH Sample 2 | 3 | 9 | 996 | CH | 4.452 | 3352842 |
| 20 | CH Sample 2 | 3 | 10 | 996 | CH | 4.463 | 3337343 |

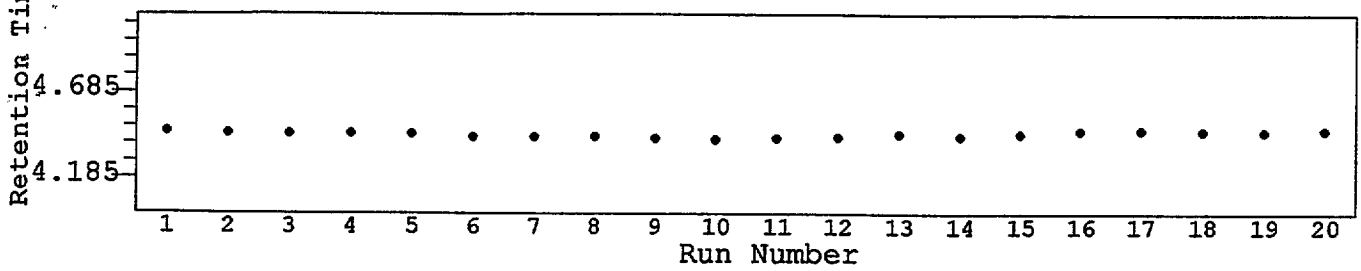
-- Peak Results Summary --

RT Min: 4.405 min
RT Max: 4.463 min
RT Average: 4.437 +/- 0.0183 min
RT %RSD: 0.411
Area Min: 3337006 uV*sec
Area Max: 3368628 uV*sec
Area Average: 3351257 +/- 7982.7 uV*sec
Area %RSD: 0.238



fig. 3

*Control Chart for: CH, Channel: 996*




*Peak Results*

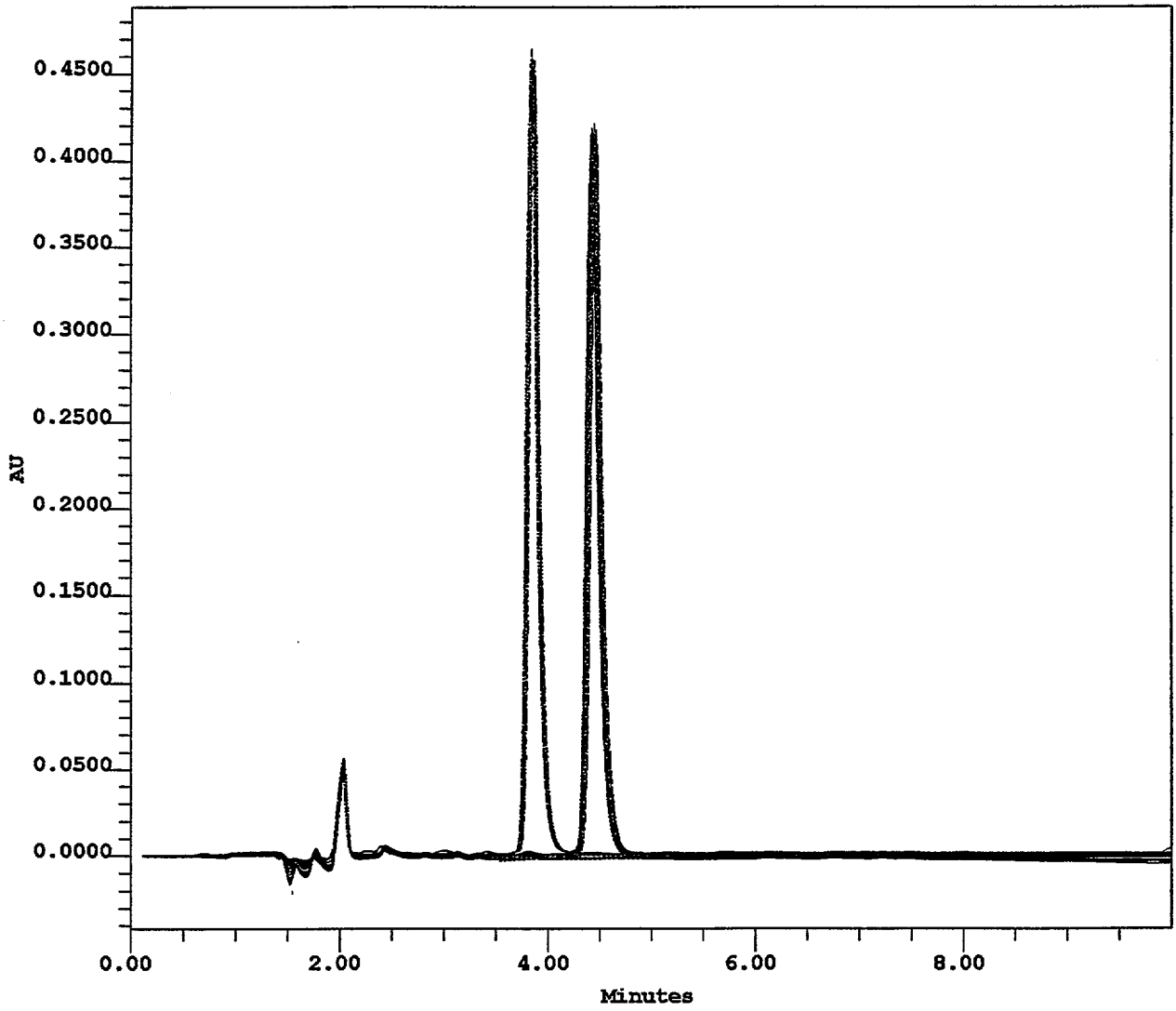
| # | SampleName | Vial | Inj | Channel | Name | RT (min) |
|---|---|---|---|---|---|---|
| 1 | CH Sample1 | 2 | 1 | 996 | CH | 4.455 |
| 2 | CH Sample1 | 2 | 2 | 996 | CH | 4.443 |
| 3 | CH Sample1 | 2 | 3 | 996 | CH | 4.442 |
| 4 | CH Sample1 | 2 | 4 | 996 | CH | 4.445 |
| 5 | CH Sample1 | 2 | 5 | 996 | CH | 4.440 |
| 6 | CH Sample1 | 2 | 6 | 996 | CH | 4.422 |
| 7 | CH Sample1 | 2 | 7 | 996 | CH | 4.420 |
| 8 | CH Sample1 | 2 | 8 | 996 | CH | 4.423 |
| 9 | CH Sample1 | 2 | 9 | 996 | CH | 4.413 |
| 10 | CH Sample1 | 2 | 10 | 996 | CH | 4.405 |
| 11 | CH Sample | 3 | 1 | 996 | CH | 4.412 |
| 12 | CH Sample | 3 | 2 | 996 | CH | 4.420 |
| 13 | CH Sample | 3 | 3 | 996 | CH | 4.442 |
| 14 | CH Sample | 3 | 4 | 996 | CH | 4.423 |
| 15 | CH Sample | 3 | 5 | 996 | CH | 4.438 |
| 16 | CH Sample | 3 | 6 | 996 | CH | 4.460 |
| 17 | CH Sample | 3 | 7 | 996 | CH | 4.463 |
| 18 | CH Sample | 3 | 8 | 996 | CH | 4.455 |
| 19 | CH Sample | 3 | 9 | 996 | CH | 4.452 |
| 20 | CH Sample | 3 | 10 | 996 | CH | 4.463 |

-- Peak Results Summary --

RT Min: 4.405 min
RT Max: 4.463 min
RT Average: 4.437 +/- 0.0183 min
RT Std. Deviation: 0.0183 min



fig 4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4.00
6.00
8.00
Minutes



fig 5