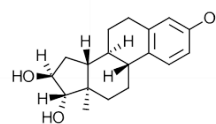
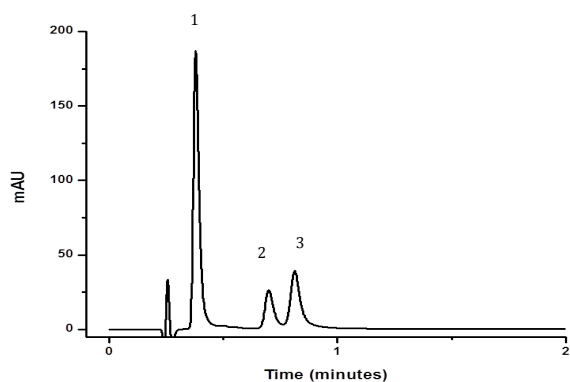


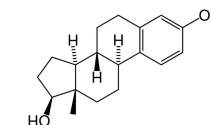
## Flare C18 Mixed-Mode Column: Separation of Estrogens (Steroids)

### HPLC Conditions

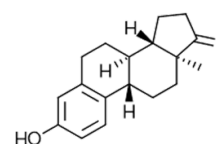
<b>Column Name:</b>	Flare C18 Mixed-Mode
<b>Column Dimensions:</b>	4.6 x 33mm (DA1094-2)
<b>HPLC System:</b>	Agilent 1200
<b>Injection Volume:</b>	1.00µl
<b>Detection:</b>	UV at 280nm
<b>Flow Rate:</b>	1.0ml/min
<b>Mobile Phase:</b>	345ml THF + 655ml H <sub>2</sub> O + 5ml DCM + 0.5ml EDA + 3.2ml Acetic Acid
<b>Temperature:</b>	30°C
<b>Analytes:</b>	1. Estriol (E3) 2. Estradiol (E2) 3. Estrone (E1)



1. Estriol (E3)



2. Estradiol (E2)



3. Estrone (E1)

### Notes

Estrone (E1), Estradiol (E2), and Estriol (E3) are structurally similar naturally occurring female steroids (hormones). There has been recent concern regarding the possibility of a link between the discharge of these steroids in waste water and endocrine disruption in fish and in humans. HPLC is the method of choice to study these hormones. However, most silica C18 columns do not have adequate selectivity to baseline separate these molecules. When possible, long columns are employed resulting in high back pressures and long retention times. In this application note, a short, but highly selective Flare C18 Mixed-Mode Core-Shell column is able to resolve these compounds in less than 1 minute.

### References

1. Purdom et al., Estrogenic Effects of Effluents from Sewage Treatment Works. *Chem. Ecol.* 1994, 8, 275-285
2. Wang et. Al, Determination of Estrogens in Water by HPLC-UV Using Cloud Point Extraction, *Talanta*, 2006, 70, 47-51

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