Supporting Information

Single-neuron comparison of the olfactory receptor response to deuterated and non-deuterated odorants

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Supplementary Information Table of Contents

This document contains Supplemental Figures S1-S7. Supplemental tables of heatmaps showing all cell responses are included as separate worksheets within a single Excel file. In that Excel file, Table S1 contains in heatmap format the tabulated responses of all odorant-responding ORNs, while Table S2 contains a summary of each experiment (i.e. mouse). The calcium imaging traces for any of the cells shown in heatmap format are available upon request.

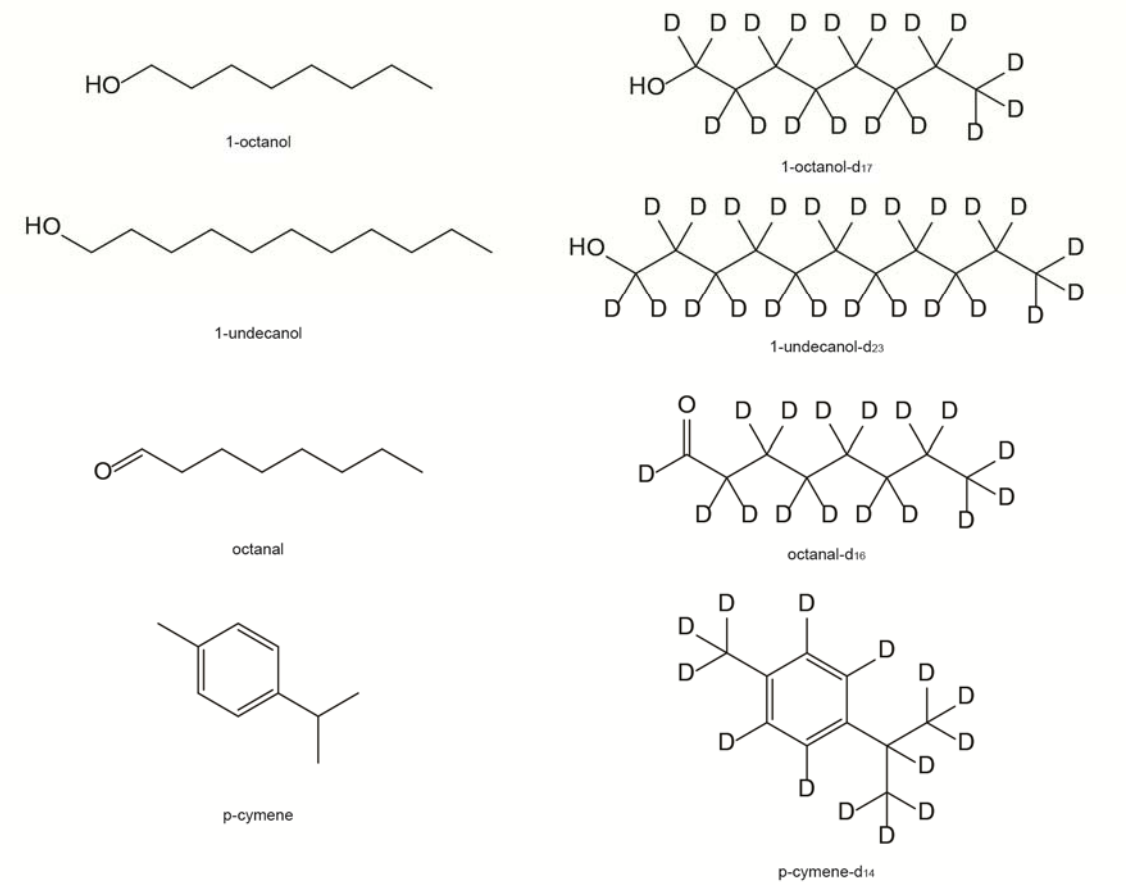

Figure S1. Odorant structures

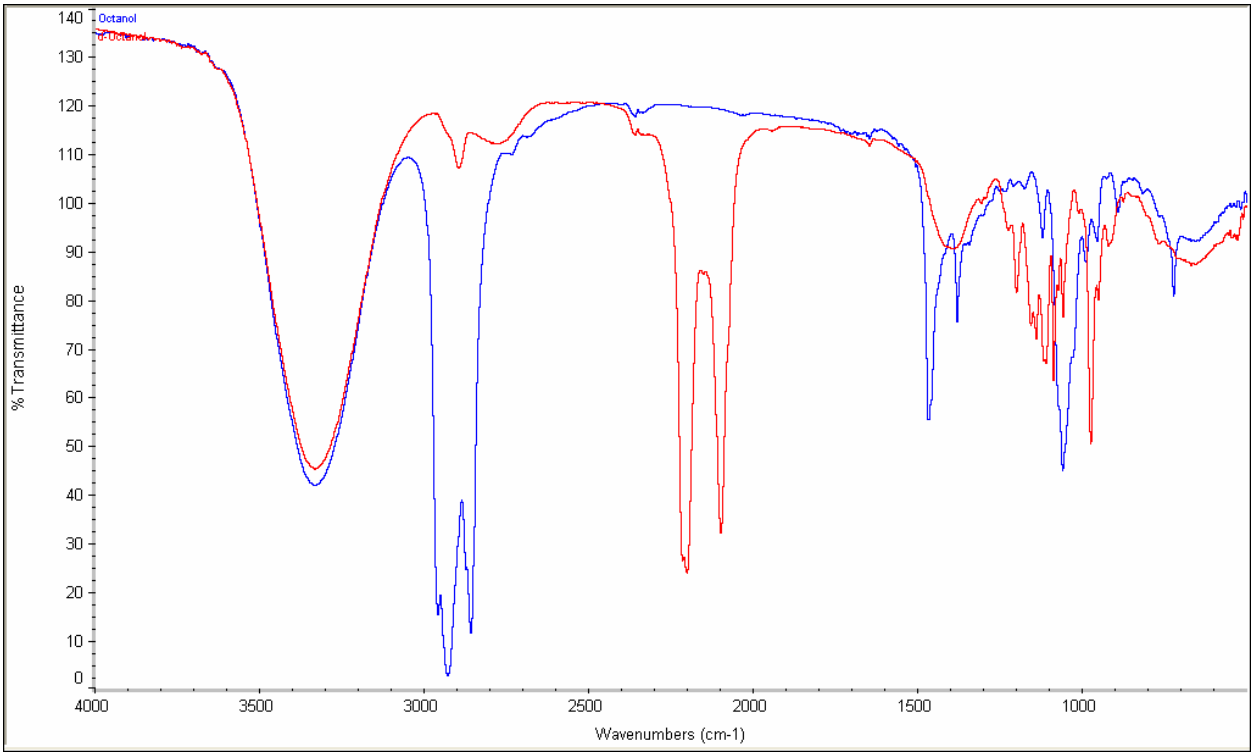

Figure S2a. IR spectra of 1-octanol (blue) and 1-octanol-d_{17} (red)

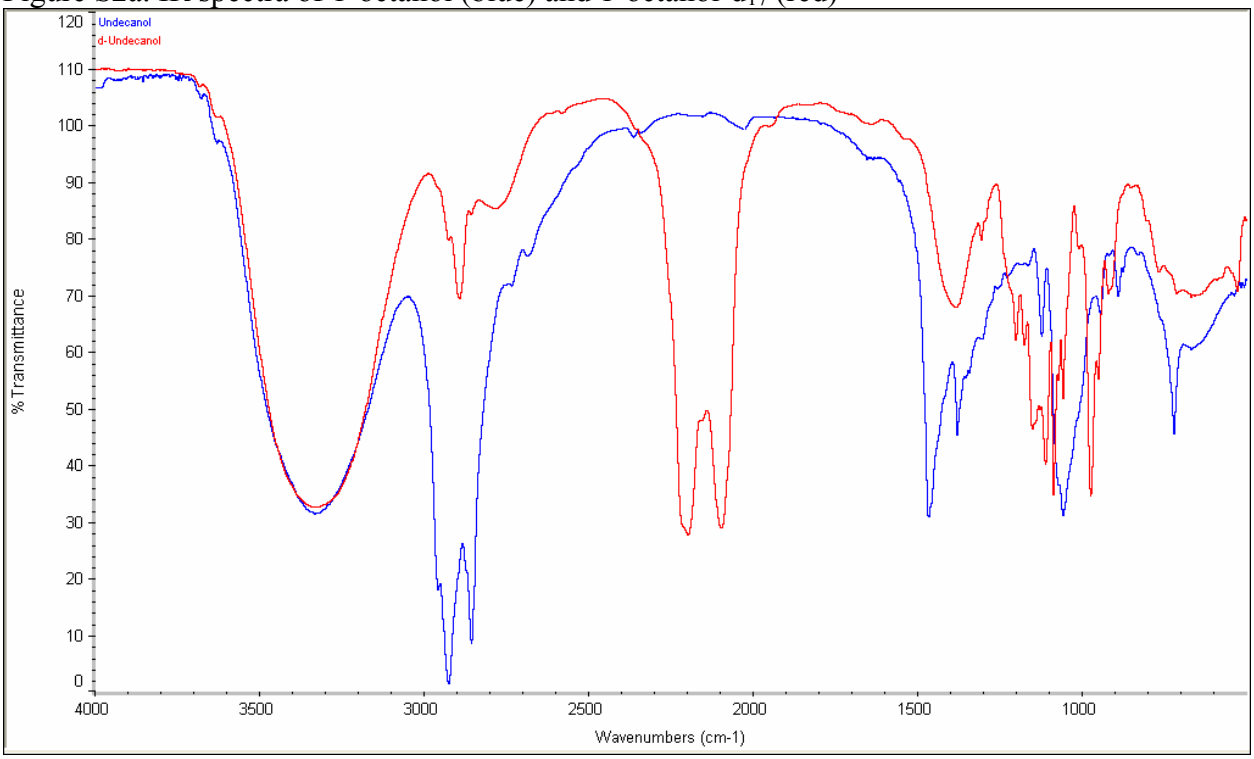

Figure S2b. IR spectra of 1-undecanol (blue) and 1-undecanol-d_{23} (red)

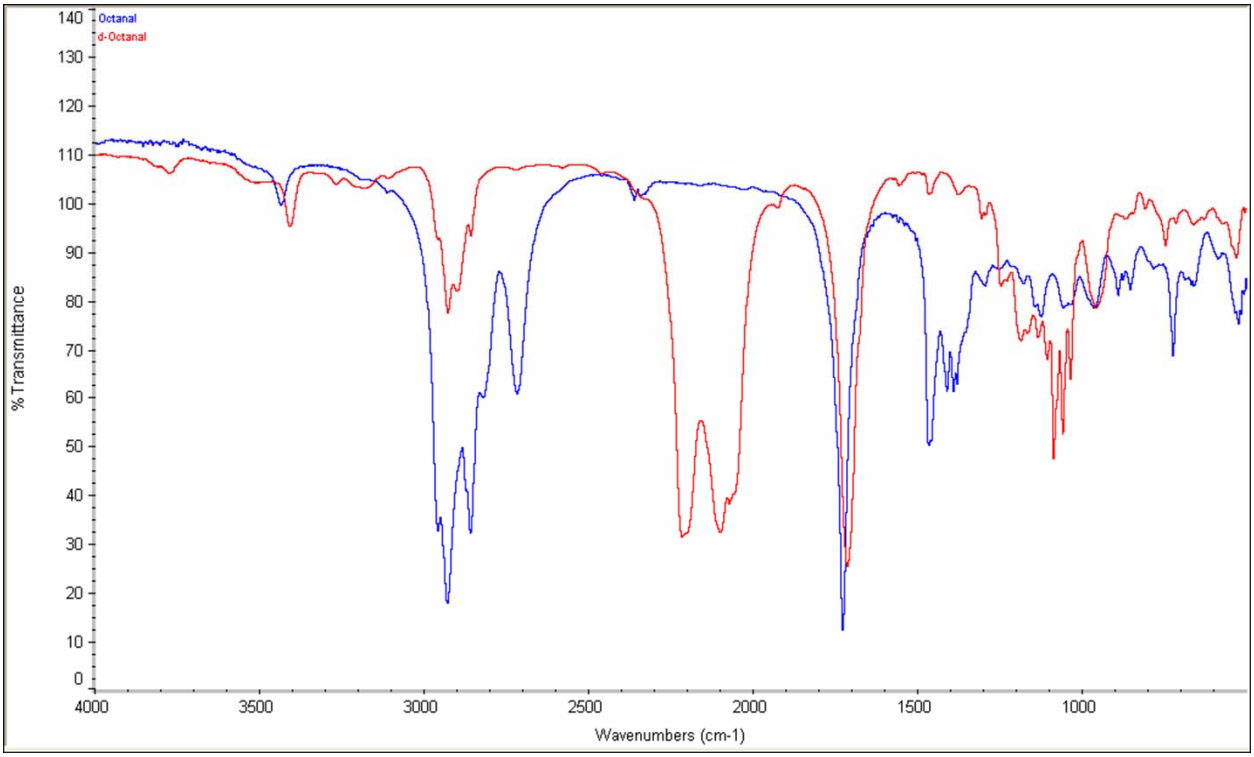

Figure S2c. IR spectra of octanal (blue) and octanal-d16 (red)

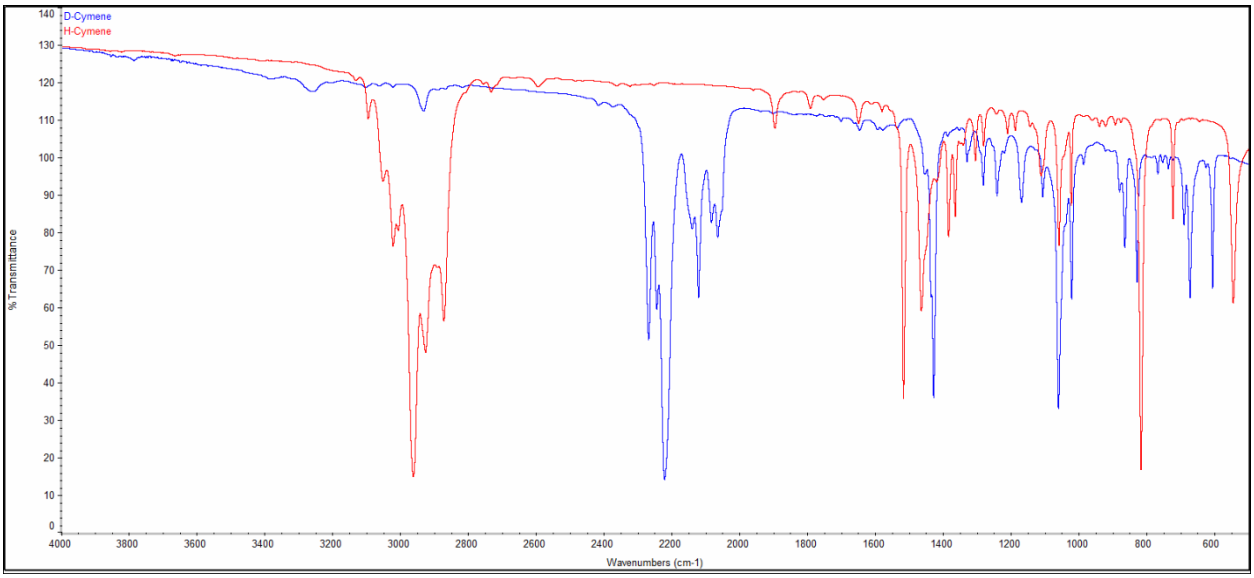

Figure S2d. IR spectra of p-cymene (red) and p-cymene-d14 (blue)

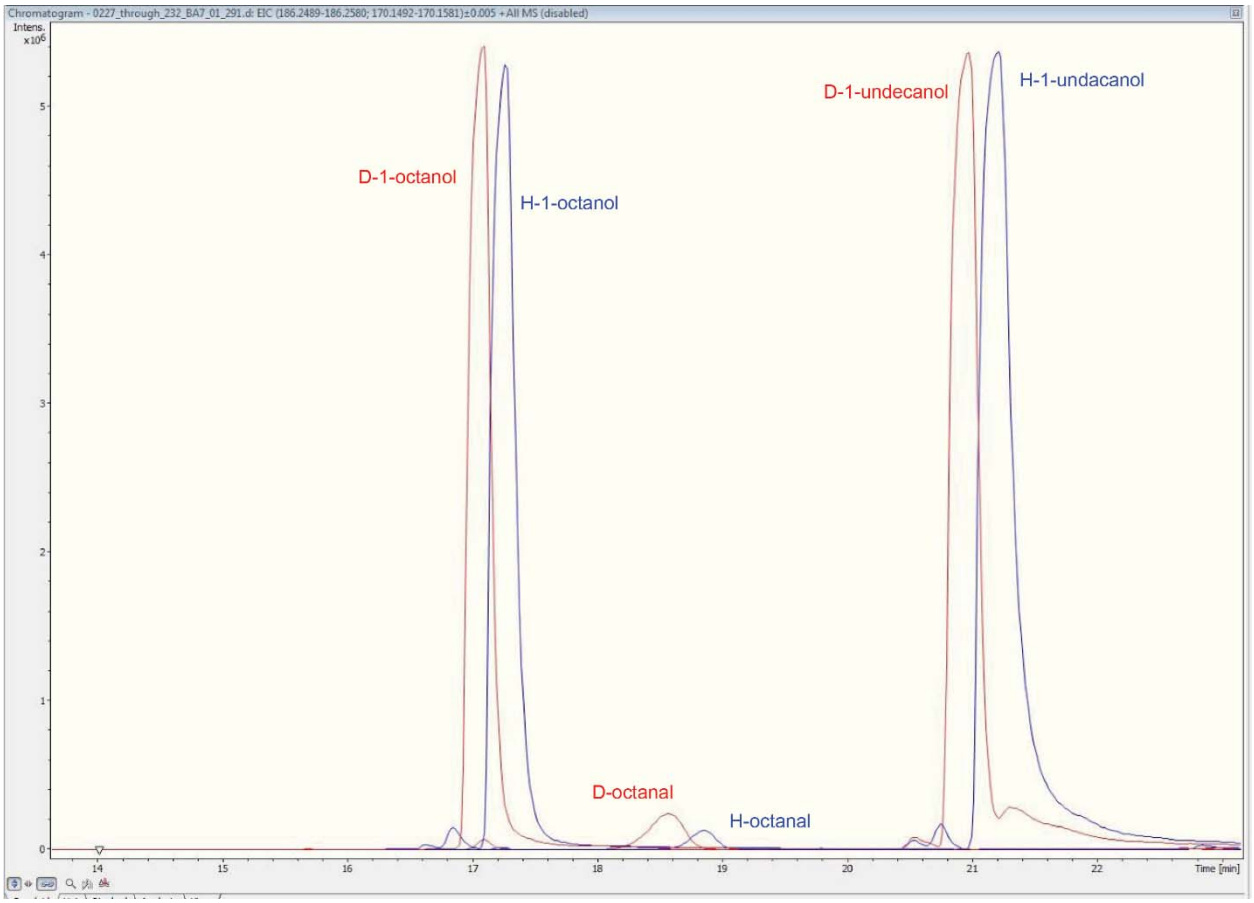

Figure S3. HPLC-MS base-peak chromatogram for the deuterated and non-deuterated 1-octanol, 1-undecanol, and octanal (injected as mixture). The differences in the HPLC retention times of the isotopomers of 1-octanol, 1-undecanol, and octanal are shown. Y-axis represents the intensity of the most abundant peak in the mass spectrum. (Conditions: Column, Thermo-Scientific Aclaim^{TM} 120; C18 3µm 120A 2.1X100mm, Dionex Ultimate-3000, High-resolution Bruker's maXis-II ETD ESI-QqTOF), using a gradient system (A-solution: H_2O with 0.1% formic acid) and B-solution (acetonitrile with 0.1% formic acid) at 0.2 mL/min.

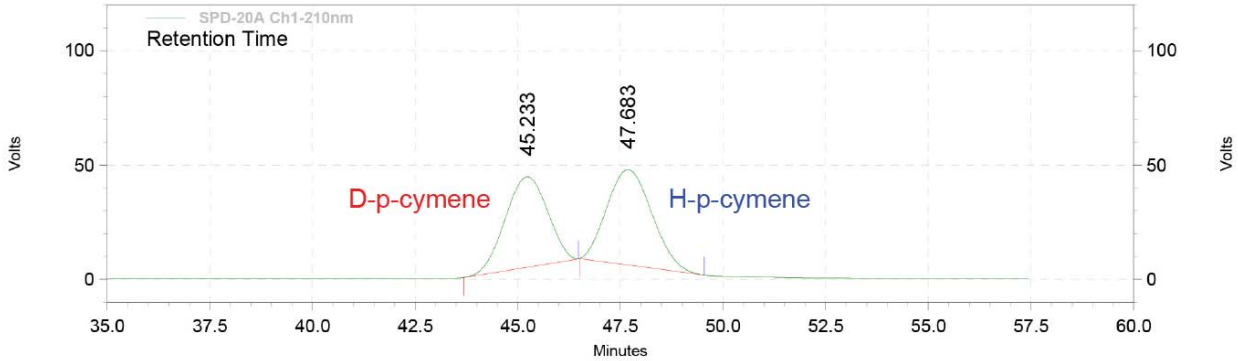

Figure S4. HPLC trace for deuterated and non-deuterated *p*-cymene. (Conditions: Agela Technologies Durachell C18 5µm column, 100Å, 4.6X150mm. Mobile phase: 50% methanol in water, flow rate 0.4 mL/min, UV detection)

Sample Information

Analyzed by	: Admin
Analyzed	: 5/1/2015 6:01:31 PM
Sample Type	: Unknown
Level #	: 1
Sample Name	: octanol
Sample ID	: octanol
IS Amount	: [1]=1
Sample Amount	: 1
Dilution Factor	: 1
Vial #	: 15
Injection Volume	: 1
Data File	: C:\GCMSsolution\Data\users\Min\Olfactory antagonis
Org Data File	: C:\GCMSsolution\Data\users\Min\Olfactory antagonis
Method File	: C:\GCMSsolution\Data\users\Min\Olfactory antagonis
Org Method File	: C:\GCMSsolution\Data\users\Min\Olfactory antagonis
Report File	:
Tuning File	: C:\GCMSsolution\System\Tune1\tune 02 24 2015.qgt
[Comment]	
octanol	
Modified by	: Admin
Modified	: 5/1/2015 6:15:00 PM

Peak Report TIC

Peak#	R.Time	I.Time	F.Time	Area	Area%	Height
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				40226381	100.00	10824589

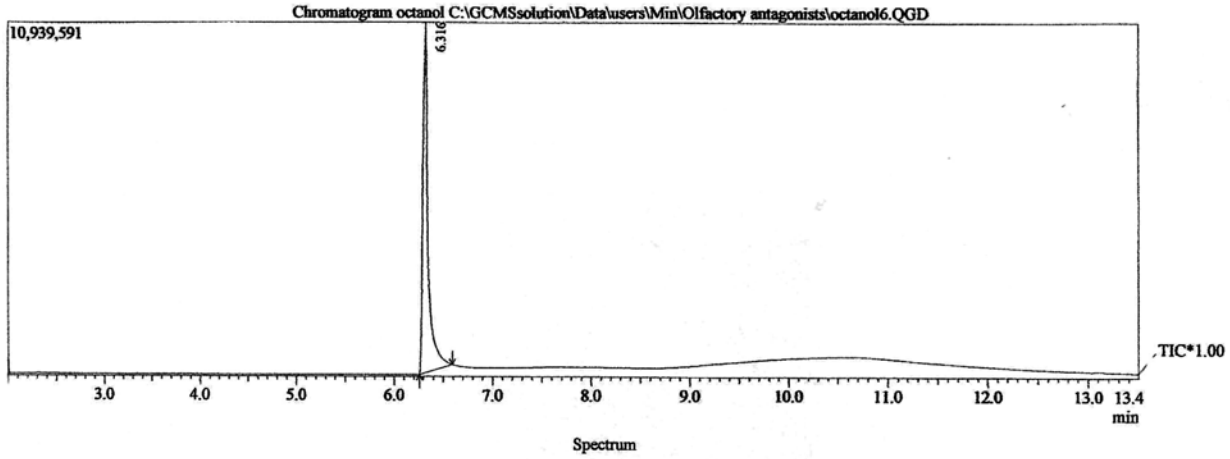


Figure S5a. Gas Chromatography trace for non-deuterated 1-octanol
Printed GC traces were scanned and the contrast was increased in Adobe Photoshop to improve visibility.

Sample Information

Analyzed by	: Admin
Analyzed	: 4/30/2015 1:12:06 PM
Sample Type	: Unknown
Level #	: 1
Sample Name	: d-octanol
Sample ID	: d-octanol
IS Amount	: [1]=1
Sample Amount	: 1
Dilution Factor	: 1
Vial #	: 16
Injection Volume	: 1
Data File	: C:\GCMSsolution\Data\users\Min\Olfactory antagonis
Org Data File	: C:\GCMSsolution\Data\users\Min\Olfactory antagonis
Method File	: C:\GCMSsolution\Data\users\Min\Olfactory antagonis
Org Method File	: C:\GCMSsolution\Data\users\Min\Olfactory antagonis
Report File	:
Tuning File	: C:\GCMSsolution\System\Tune1\tune 02 24 2015.qgt
[Comment]	
d-octanol	
Modified by	: Admin
Modified	: 4/30/2015 1:25:37 PM

Peak Report TIC

Peak#	R.Time	I.Time	F.Time	Area	Area%	Height
1	6.125	6.075	6.417	42459436	98.02	10625411
2	8.956	8.917	9.000	857590	1.98	497723
				43317026	100.00	11123134

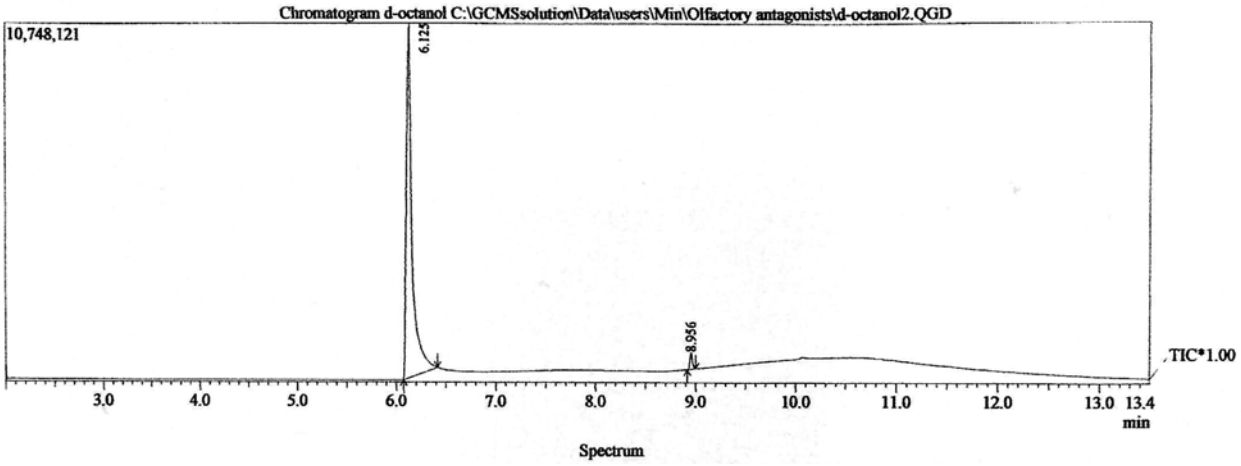
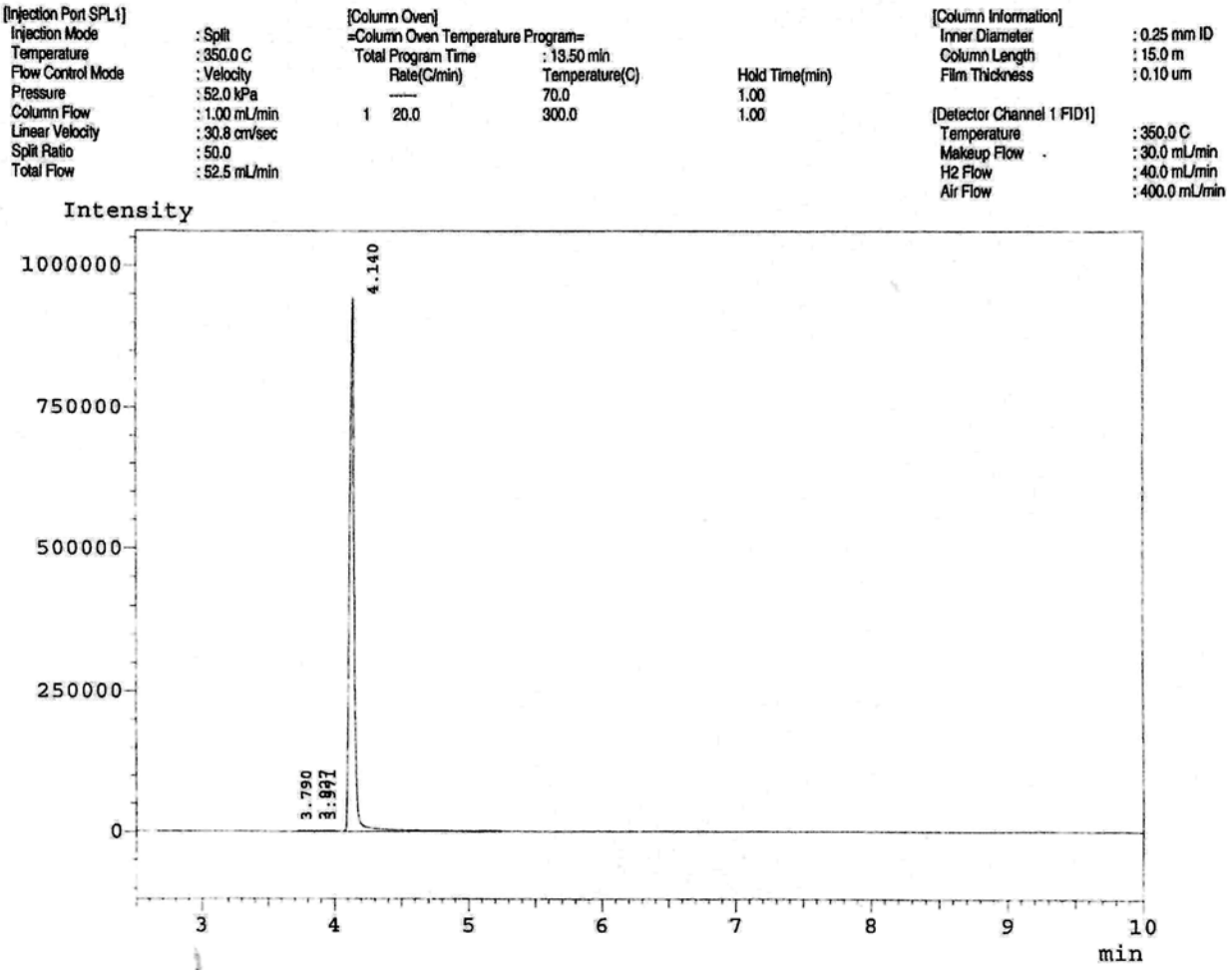
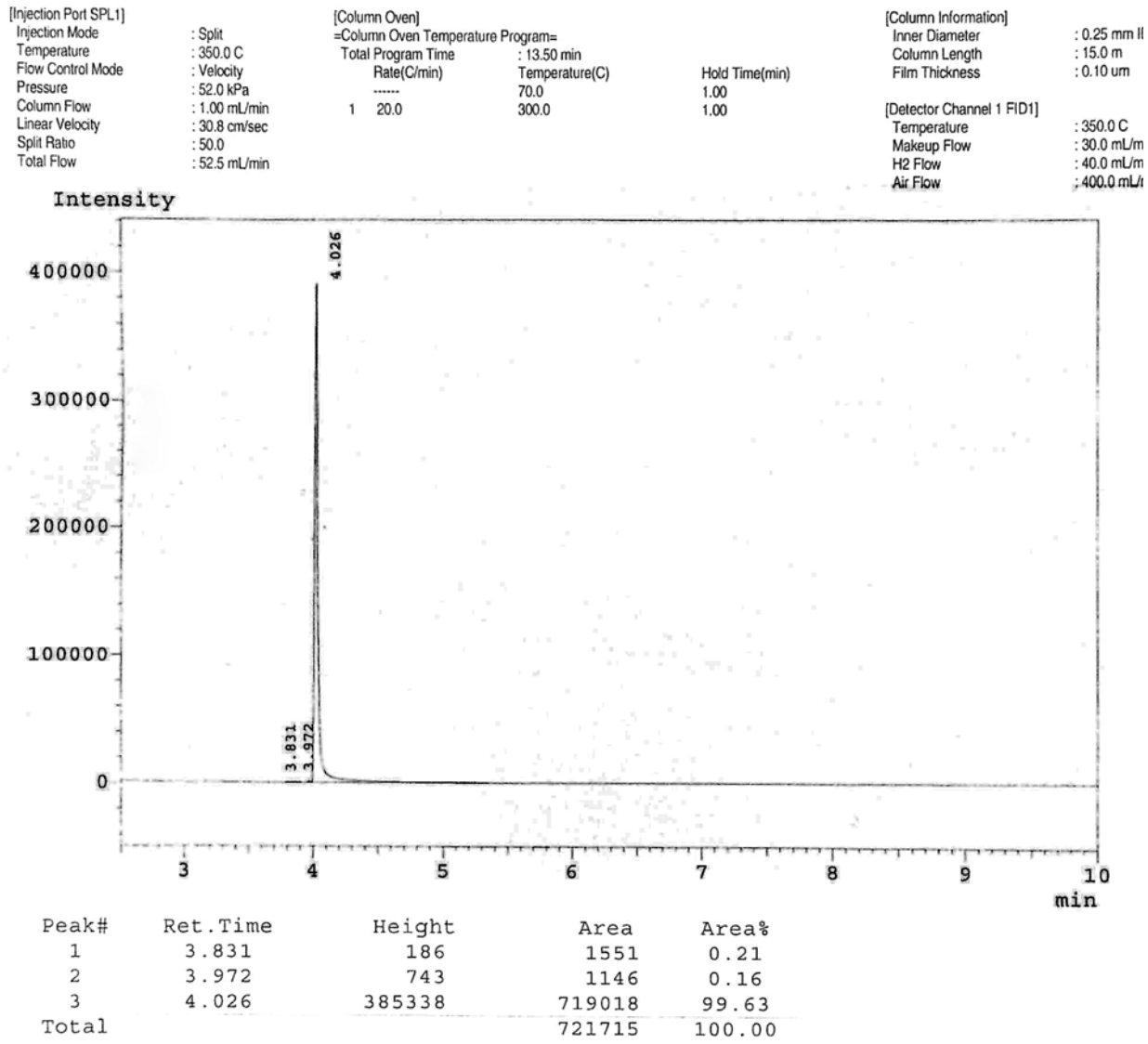


Figure S5b. Gas Chromatography trace for deuterated 1-octanol



Peak#	Ret.Time	Height	Area	Area%
1	3.790	521	3626	0.16
2	3.927	1152	3262	0.14
3	3.971	1403	3477	0.15
4	4.140	912141	2298538	99.55
Total			2308903	100.00

Figure S5c. Gas Chromatography trace for non-deuterated 1-undecanol



Peak#	Ret.Time	Height	Area	Area%
1	3.831	186	1551	0.21
2	3.972	743	1146	0.16
3	4.026	385338	719018	99.63
Total			721715	100.00

Figure S5d. Gas Chromatography trace for deuterated 1- undecanol

Sample Information

Analyzed by	: Admin
Analyzed	: 12/1/2015 6:59:05 PM
Sample Type	: Unknown
Level #	: 1
Sample Name	: octanal-12-1-2
Sample ID	: octanal-12-1-2
Tray#	: Tray2
IS Amount	: [1]=1
Sample Amount	: 1
Dilution Factor	: 1
Vial #	: 1
Injection Volume	: 10
Data File	: C:\GCMSsolution\Data\users\Min\Olfactory antagonist
Org Data File	: C:\GCMSsolution\Data\users\Min\Olfactory antagonist
Method File	: C:\GCMSsolution\Data\users\Min\Olfactory antagonist
Org Method File	: C:\GCMSsolution\Data\users\Min\Olfactory antagonist
Report File	:
Tuning File	: C:\GCMSsolution\System\Tune1\tune 11 30 2015.qgt
Modified by	: Admin
Modified	: 12/1/2015 7:12:42 PM

Peak Report TIC

Peak#	R.Time	I.Time	F.Time	Area	Area%	Height
1	2.419	2.400	2.442	24661	0.21	19426
2	4.980	4.825	5.017	11610208	99.54	1917703
3	7.883	7.858	7.900	4476	0.04	3193
4	8.745	8.717	8.783	9540	0.08	4642
5	11.436	11.392	11.492	14391	0.12	4585
				11663276	100.00	1949549

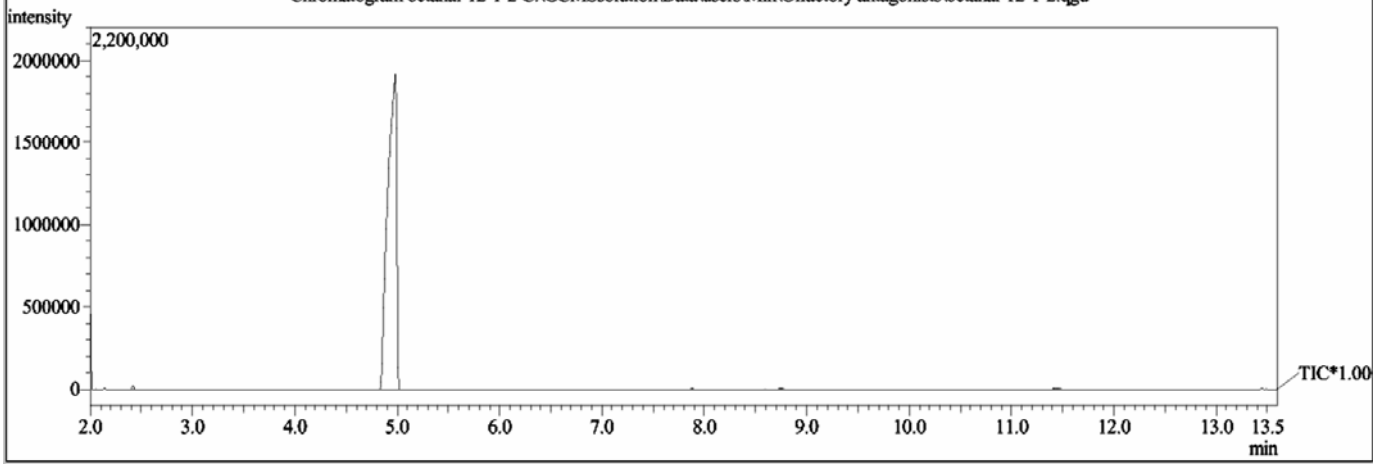


Figure S5e. Gas Chromatography trace for non-deuterated octanal

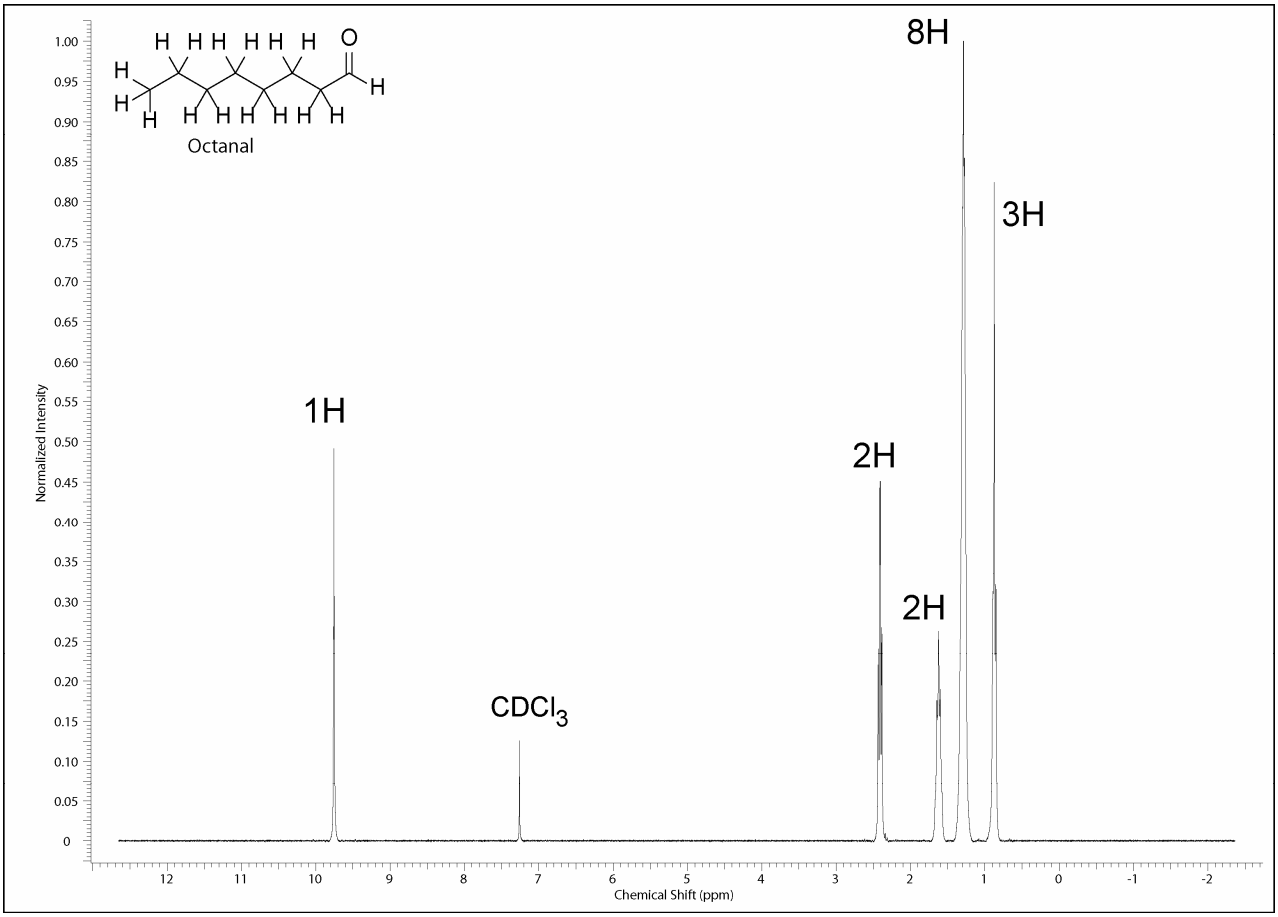


Figure S5e (cont'd). 1H-NMR spectrum of non-deuterated octanal used in calcium imaging experiments.

Sample Information

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Analyzed	: 12/1/2015 7:20:29 PM
Sample Type	: Unknown
Level #	: 1
Sample Name	: d-octanal-column-2
Sample ID	: d-octanal-column-2
Tray#	: Tray2
IS Amount	: [1]=1
Sample Amount	: 1
Dilution Factor	: 1
Vial #	: 2
Injection Volume	: 10
Data File	: C:\GCMSsolution\Data\users\Min\Olfactory antagonist
Org Data File	: C:\GCMSsolution\Data\users\Min\Olfactory antagonist
Method File	: C:\GCMSsolution\Data\users\Min\Olfactory antagonist
Org Method File	: C:\GCMSsolution\Data\users\Min\Olfactory antagonist
Report File	:
Tuning File	: C:\GCMSsolution\System\Tune1\tune 11 30 2015.qgt
Modified by	: Admin
Modified	: 12/1/2015 7:34:06 PM

Peak Report TIC

Peak#	R.Time	I.Time	F.Time	Area	Area%	Height
1	2.425	2.400	2.450	23299	0.31	18564
2	4.755	4.642	4.800	7513494	99.52	1537978
3	11.433	11.400	11.492	13101	0.17	4255
				7549894	100.00	1560797

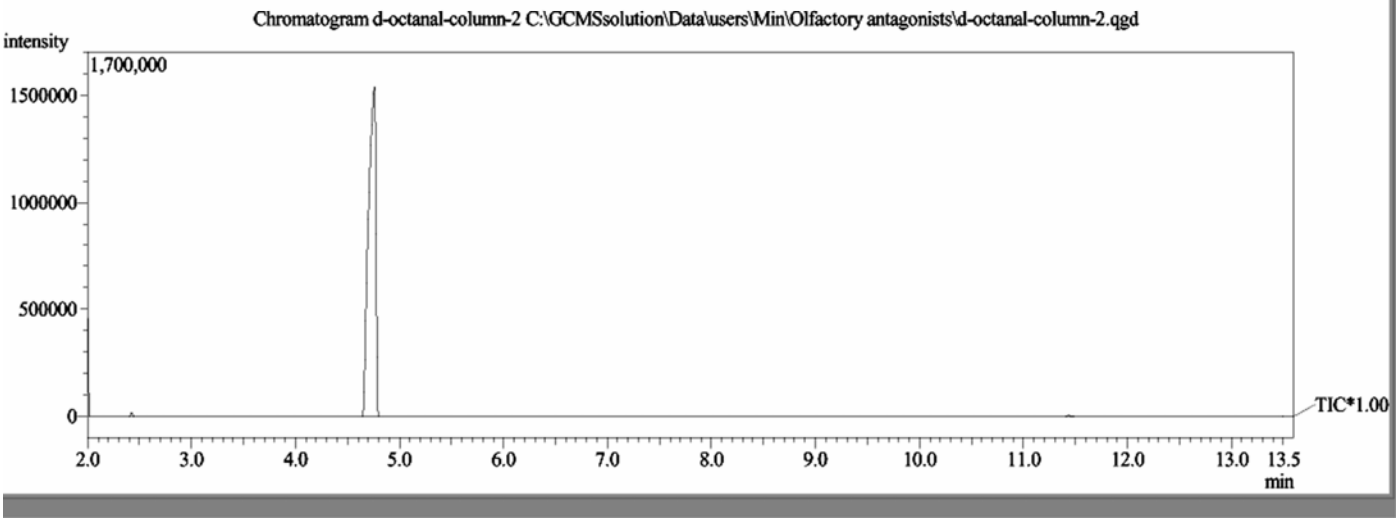
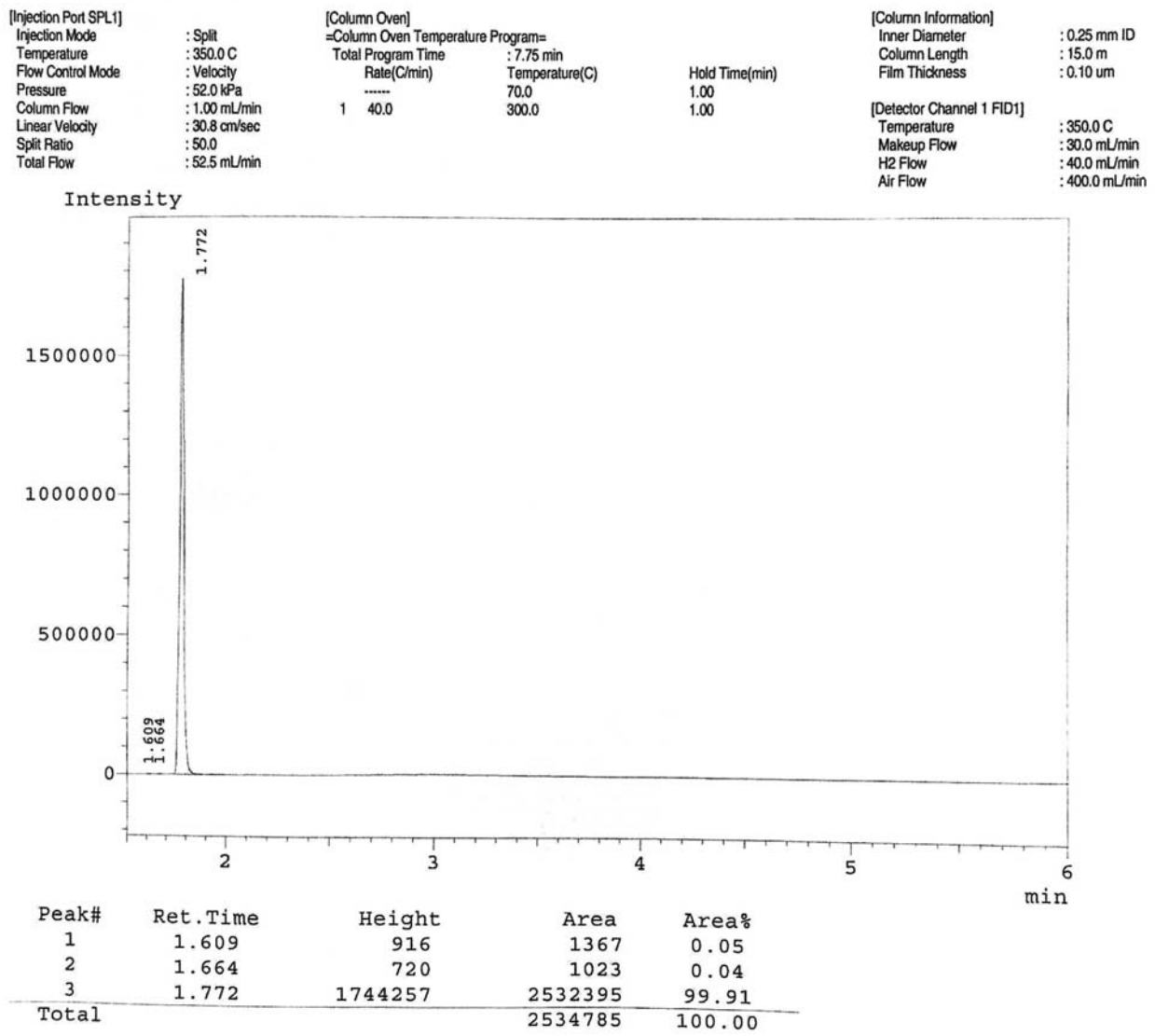
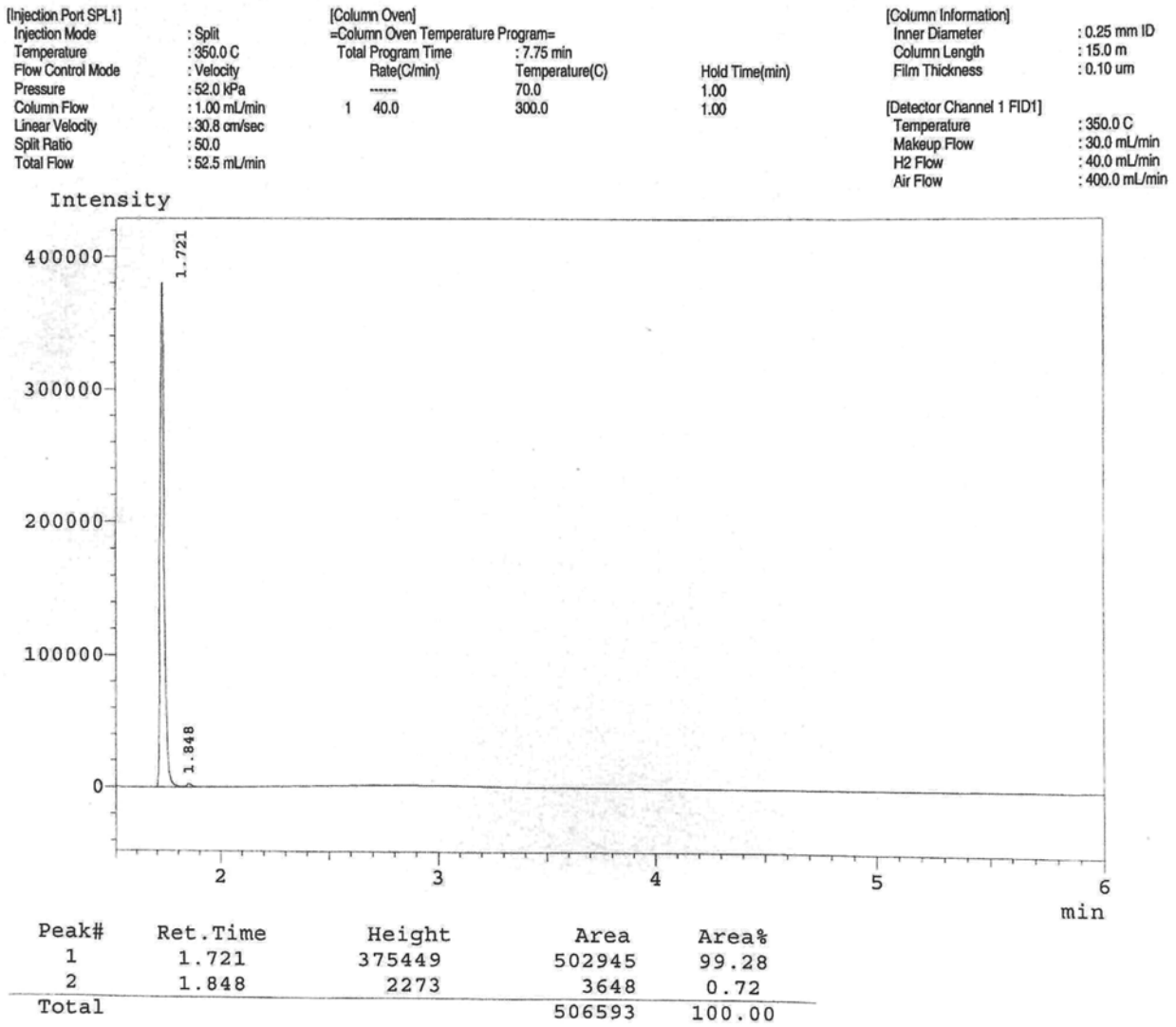


Figure S5f. Gas Chromatography trace for deuterated octanal



Peak#	Ret.Time	Height	Area	Area%
1	1.609	916	1367	0.05
2	1.664	720	1023	0.04
3	1.772	1744257	2532395	99.91
Total			2534785	100.00

Figure S5g. Gas Chromatography trace for non-deuterated p-cymene



Peak#	Ret.Time	Height	Area	Area%
1	1.721	375449	502945	99.28
2	1.848	2273	3648	0.72
Total			506593	100.00

Figure S5h. Gas Chromatography trace for deuterated p-cymene

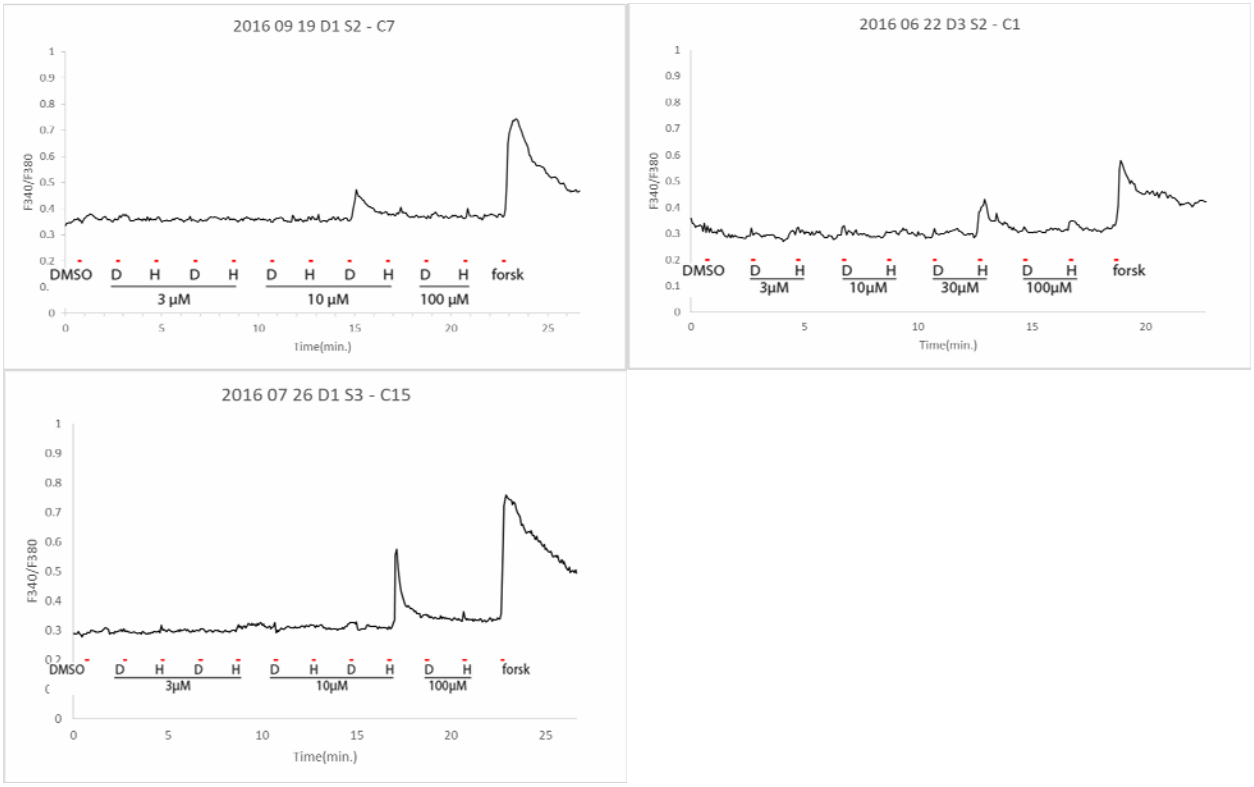
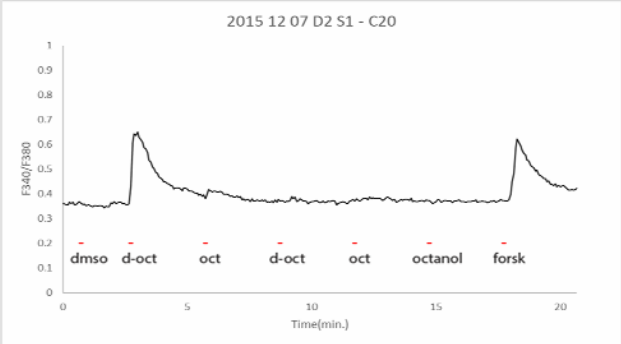


Figure S6. Calcium imaging recordings of uncategorized cells. During the testing of the 23,812 forskolin-positive cells, three cells responded in such a way as not to fit into any of the four categories to which were assigned the 1,610 responding cells. Two of these were observed for 1-undecanol (top) and one for octanal (bottom).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F340/F380
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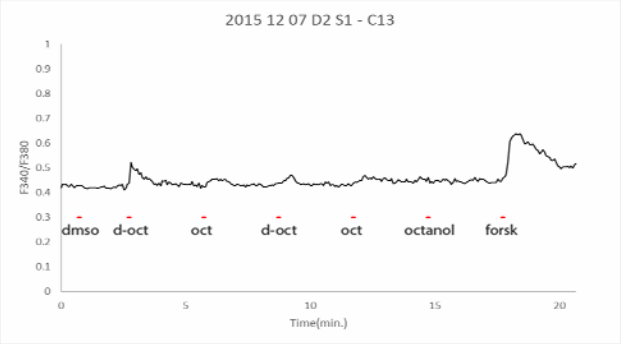
Time(min.)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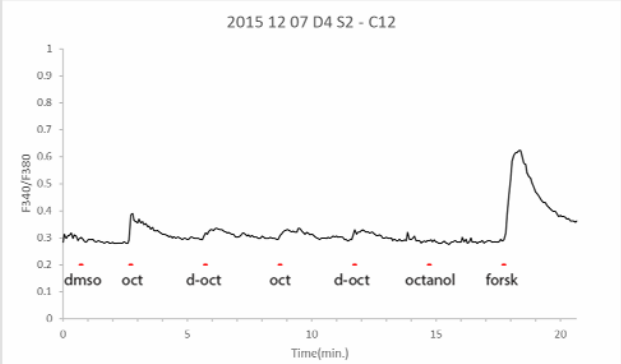
F340/F380
dmso d-oct oct d-oct oct octanol forsk
Time(min.)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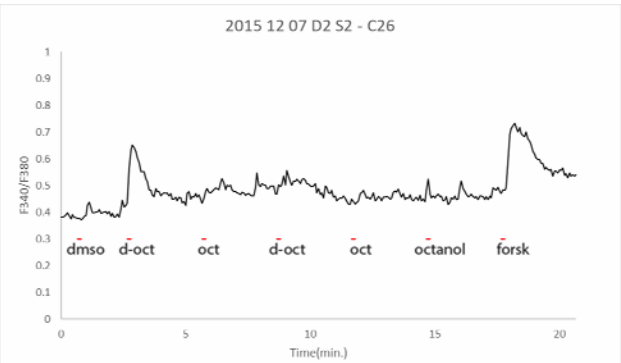
Time(min.)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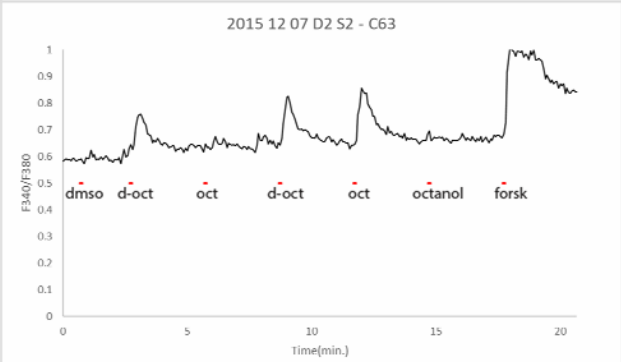
F340/F380
dmso d-oct oct d-oct oct octanol forsk
Time(min.)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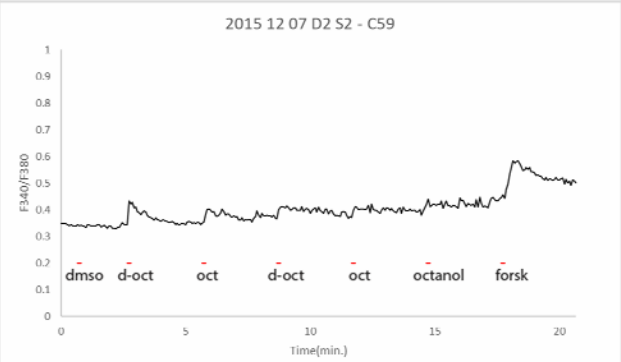
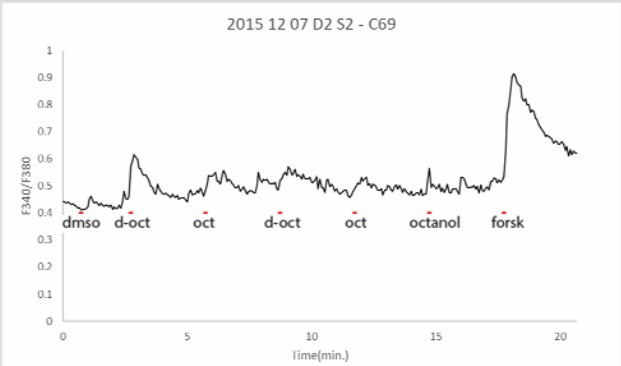
F340/F380
dmso d-oct oct d-oct oct octanol forsk
Time(min.)


2015 12 07 D2 S2 - C59
F340/F380
dmso d-oct oct d-oct oct octanol forsk
Time(min.)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F340/F380
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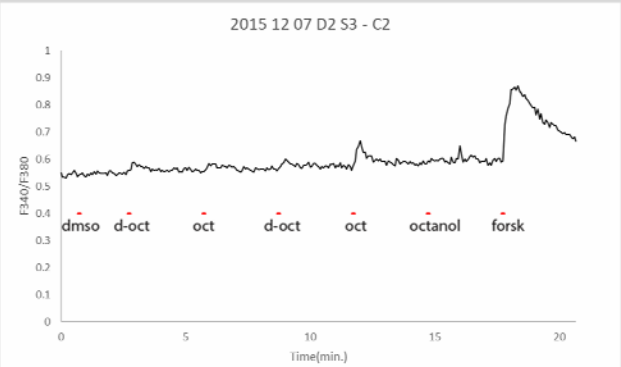
Time(min.)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F340/F380
dmso d-oct oct d-oct oct octanol forsk
Time(min.)

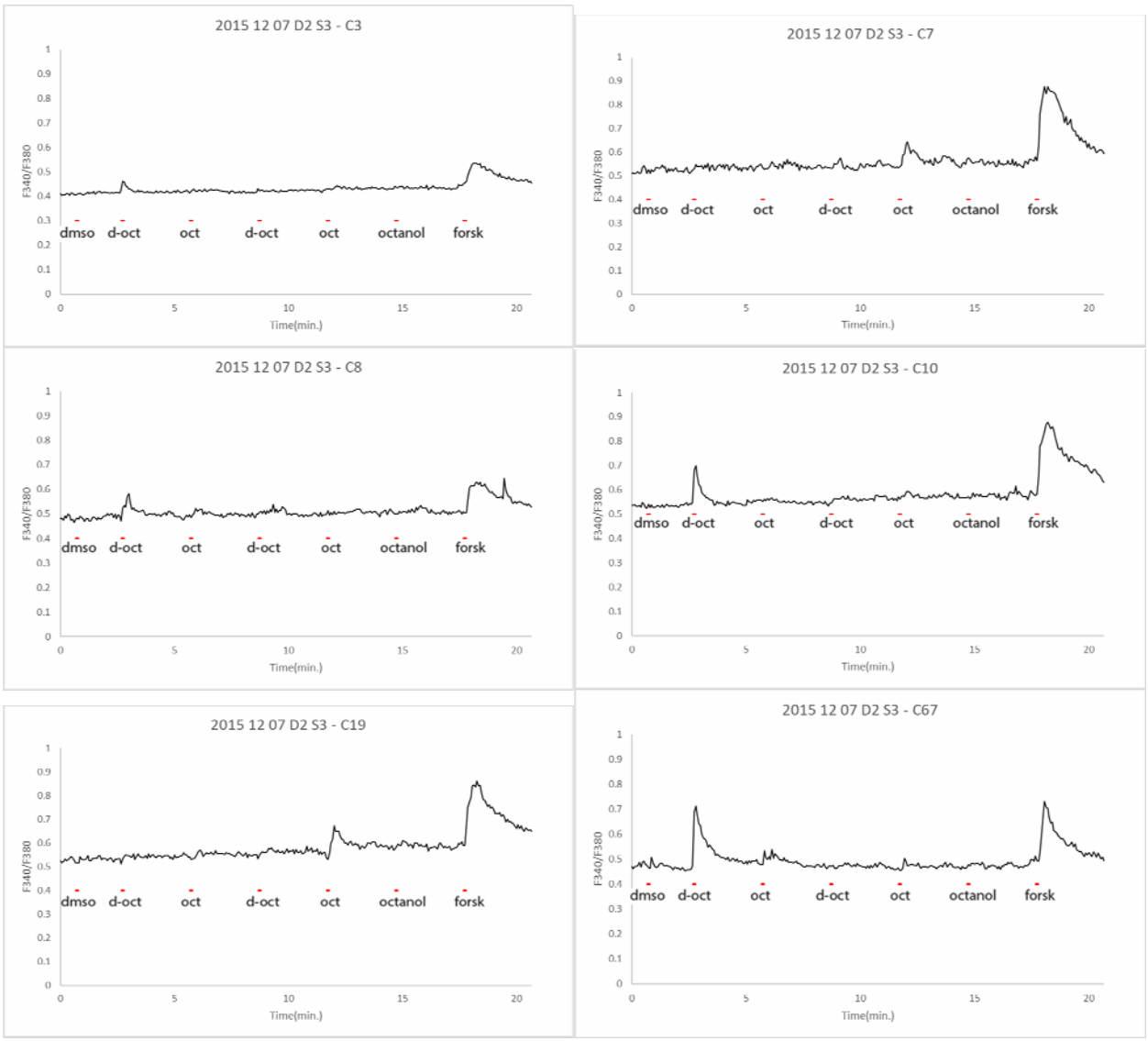

Figure S7. Calcium imaging recordings of UbI7 cells showing ambiguous responses. During the testing of the forskolin-positive UbI7 cells, 14 cells gave ambiguous responses and thus were excluded from the H vs. D analysis. (d-oct: deuterated octanal, oct: non-deuterated octanal)