

Retention time normalization kit: Calibration peptides for time resolved mass spectrometry

Quick Reference

This Quick Reference Card provides abbreviated procedures you can refer to when you use the **RT-Kit-WR**.

This assay kit contains synthetic peptides in a pre-pooled mix for retention time re-calibration.

For general chemical safety information, background information, and detailed procedures, refer to the Biognosys Reagents Chemistry Reference Guide (available at www.biognosys.ch/rt-kit/)

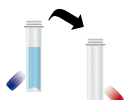
Running the Protocol

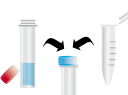
Follow the procedures shown below. Modify the procedures if you determine that alternative steps are required for your sample.

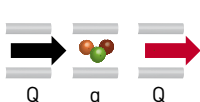
The MRM assays for these peptides can be found on the backside of this Quick Reference or in a Excel document **RT-Kit-WR.xls** at www.biognosys.ch/rt-kit/. Please refer to description in this document for instructions how to set up an instrument method.

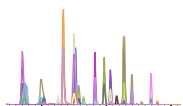
Please see our Forum on www.biognosys.ch/rt-kit/ for FAQs and support.

RT-Kit Quick Guide

1  1. Add **50 µl** Dissolution Buffer (blue cap) to the Standard Mixture tube (red cap)
2. Vortex to mix, sonicate 5 min. if possible.
3. This solution is the 10x *Standard Mixture*

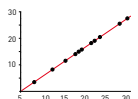
2  Dissolve *Standard Mixture* 10 times (1:10) in *Buffer A* or peptide sample.

3  Use transitions for *Standard Mixture* as indicated in **RT-Kit-WR.xls**. Use the gradient that you plan to use for your analytical sample measurements. Run LC-MS experiment in non-scheduled MRM mode.

4  Extract retention times for each standard in *Standard Mixture* with the software of your choice.

5 At www.biognosys.ch/RT-Kit you find documentation and a link to our iRT calculator. With our iRT-calculator you can:

- Convert assay retention times into iRT
- Convert iRT into real retention times

6  Check linearity of recalibration and get either your retention times or *iRT* values for each assay.

Transitions for standard mixture (Prod. Nr. 1007B-50001) are indicated below as quick reference. See documentation at www.biognosys.ch/rt-kit/ for full documentation and recommended machine specific settings.

Q1	Q3	Intensity	Charge	ID	Transition- ID	Nominal Sequence*
487.53	860.42	100	2	RT-Kit-WR_a	RT-Kit-WR_a1	LGGNETQVR
487.53	503.29	53	2	RT-Kit-WR_a	RT-Kit-WR_a2	LGGNETQVR
487.53	803.40	50	2	RT-Kit-WR_a	RT-Kit-WR_a3	LGGNETQVR
645.19	800.45	100	2	RT-Kit-WR_b	RT-Kit-WR_b1	AGGSSEPVTGLADK
645.19	604.33	17	2	RT-Kit-WR_b	RT-Kit-WR_b2	AGGSSEPVTGLADK
645.19	1016.53	16	2	RT-Kit-WR_b	RT-Kit-WR_b3	AGGSSEPVTGLADK
684.22	819.39	100	2	RT-Kit-WR_c	RT-Kit-WR_c1	VEATFGVDESANK
684.22	966.45	56	2	RT-Kit-WR_c	RT-Kit-WR_c2	VEATFGVDESANK
684.22	663.30	46	2	RT-Kit-WR_c	RT-Kit-WR_c3	VEATFGVDESANK
547.62	817.44	100	2	RT-Kit-WR_d	RT-Kit-WR_d1	YILAGVESNK
547.62	633.32	54	2	RT-Kit-WR_d	RT-Kit-WR_d2	YILAGVESNK
547.62	704.36	52	2	RT-Kit-WR_d	RT-Kit-WR_d3	YILAGVESNK
670.24	928.42	100	2	RT-Kit-WR_e	RT-Kit-WR_e1	TPVISGGPYER
670.24	1041.50	48	2	RT-Kit-WR_e	RT-Kit-WR_e2	TPVISGGPYER
670.24	841.38	39	2	RT-Kit-WR_e	RT-Kit-WR_e3	TPVISGGPYER
684.26	956.45	100	2	RT-Kit-WR_f	RT-Kit-WR_f1	TPVITGAPYER
684.26	855.40	59	2	RT-Kit-WR_f	RT-Kit-WR_f2	TPVITGAPYER
684.26	1069.53	44	2	RT-Kit-WR_f	RT-Kit-WR_f3	TPVITGAPYER
699.75	855.44	100	2	RT-Kit-WR_g	RT-Kit-WR_g1	GDLDAASYAPVR
699.75	926.47	68	2	RT-Kit-WR_g	RT-Kit-WR_g2	GDLDAASYAPVR
699.75	605.34	58	2	RT-Kit-WR_g	RT-Kit-WR_g3	GDLDAASYAPVR
727.27	1066.48	100	2	RT-Kit-WR_h	RT-Kit-WR_h1	DAVTPADFSEWSK
727.27	533.75	65	2	RT-Kit-WR_h	RT-Kit-WR_h2	DAVTPADFSEWSK
727.27	584.27	23	2	RT-Kit-WR_h	RT-Kit-WR_h3	DAVTPADFSEWSK
623.23	598.37	100	2	RT-Kit-WR_i	RT-Kit-WR_i1	TGFIIDPGGVIR
623.23	713.40	30	2	RT-Kit-WR_i	RT-Kit-WR_i2	TGFIIDPGGVIR
623.23	826.48	28	2	RT-Kit-WR_i	RT-Kit-WR_i3	TGFIIDPGGVIR
637.25	626.40	100	2	RT-Kit-WR_k	RT-Kit-WR_k1	GTFIIDPAAIVR
637.25	854.51	66	2	RT-Kit-WR_k	RT-Kit-WR_k2	GTFIIDPAAIVR
637.25	741.43	60	2	RT-Kit-WR_k	RT-Kit-WR_k3	GTFIIDPAAIVR
777.41	904.49	100	2	RT-Kit-WR_l	RT-Kit-WR_l1	FLLQFGAQGSPLFK
777.41	1051.56	91	2	RT-Kit-WR_l	RT-Kit-WR_l2	FLLQFGAQGSPLFK
777.41	504.32	80	2	RT-Kit-WR_l	RT-Kit-WR_l3	FLLQFGAQGSPLFK

* Nominal sequences are sequences that have the same mass and identical ion-series like the chemical sequences except for terminal b- or y-ions.

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