

AUTHORIZED DISTRIBUTOR

MZ-Analysentechnik GmbH, Barcelona-Allee 17 • D-55129 Mainz

Tel +49 6131 880 96-0, Fax +49 6131 880 96-20

e-mail: info@mz-at.de, www.mz-at.de

Circular RNA Separation by SEC and RP Columns

Sepax Technologies





Overview



mRNA has continued to gain success as a new therapeutic agent with broad potential for application in biological systems. However, it has fundamental limitations relating to stability and its relatively short half-life. To overcome the shortcomings of linear mRNA, circular RNA is emerging as a new promising alternative modality.

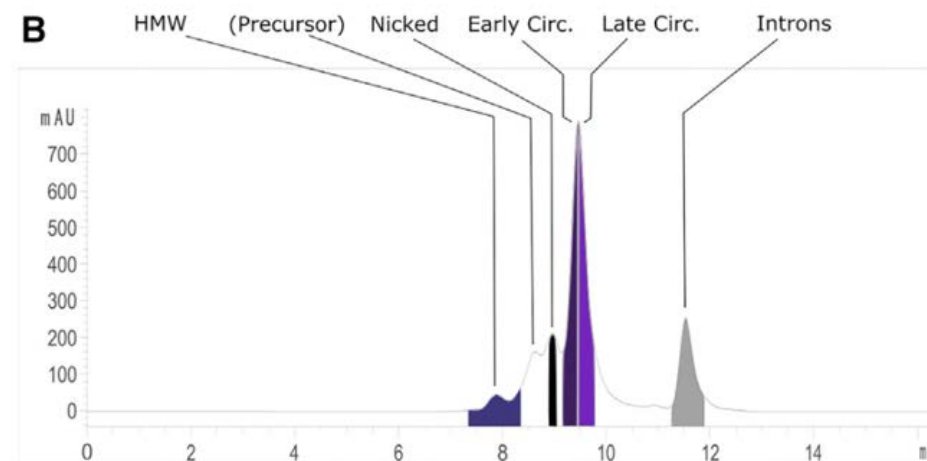
In this application:

Size Exclusion and Reversed Phase HPLC methods are discussed for the separation of the Circular RNA sample. Sepax **SRT SEC-2000** column in 2000 Å large pore size was able to separate its aggregates and fragments. And the Sepax **Bio-C18** column in 300 Å can be used to analyze the precursor, nicked and circular RNA.

Sample information:

- Precursor: Uncircularized RNA, approximately 2800 nt.
- Nicked: Linear RNA, approximately 2500 nt.
- Circular: Circular RNA, approximately 2500 nt.

*Ensure the solvent used for sample prep is RNase-free water



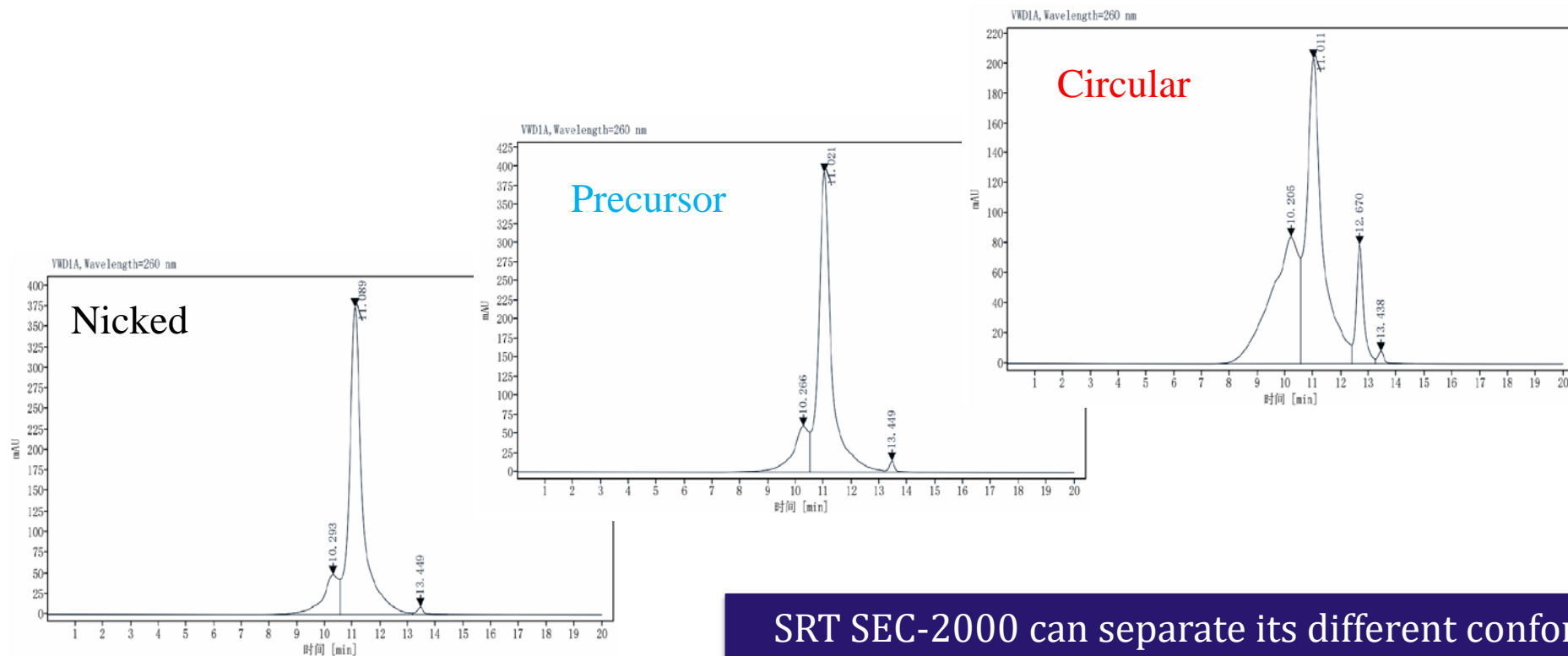
**Picture Source: Alexander, et al. "RNA circularization diminishes immunogenicity and can extend translation duration in vivo." *Molecular cell* 74.3 (2019): 508-520



circRNAs – SEC method



Column: SRT SEC-2000, 5 μm , 2000 \AA , 7.8 \times 300 mm; Part Number: 215980-7830; Mobile Phase: 150 mM PB pH 7.0; Flow Rate: 0.8 mL/min; Detector: UV 260 nm; Column Temperature: RT; Injection Volume: 10 μL , 1 mg/mL; Column Pressure: 58 bar



SRT SEC-2000 can separate its different conformations and effectively separate its aggregates and fragments

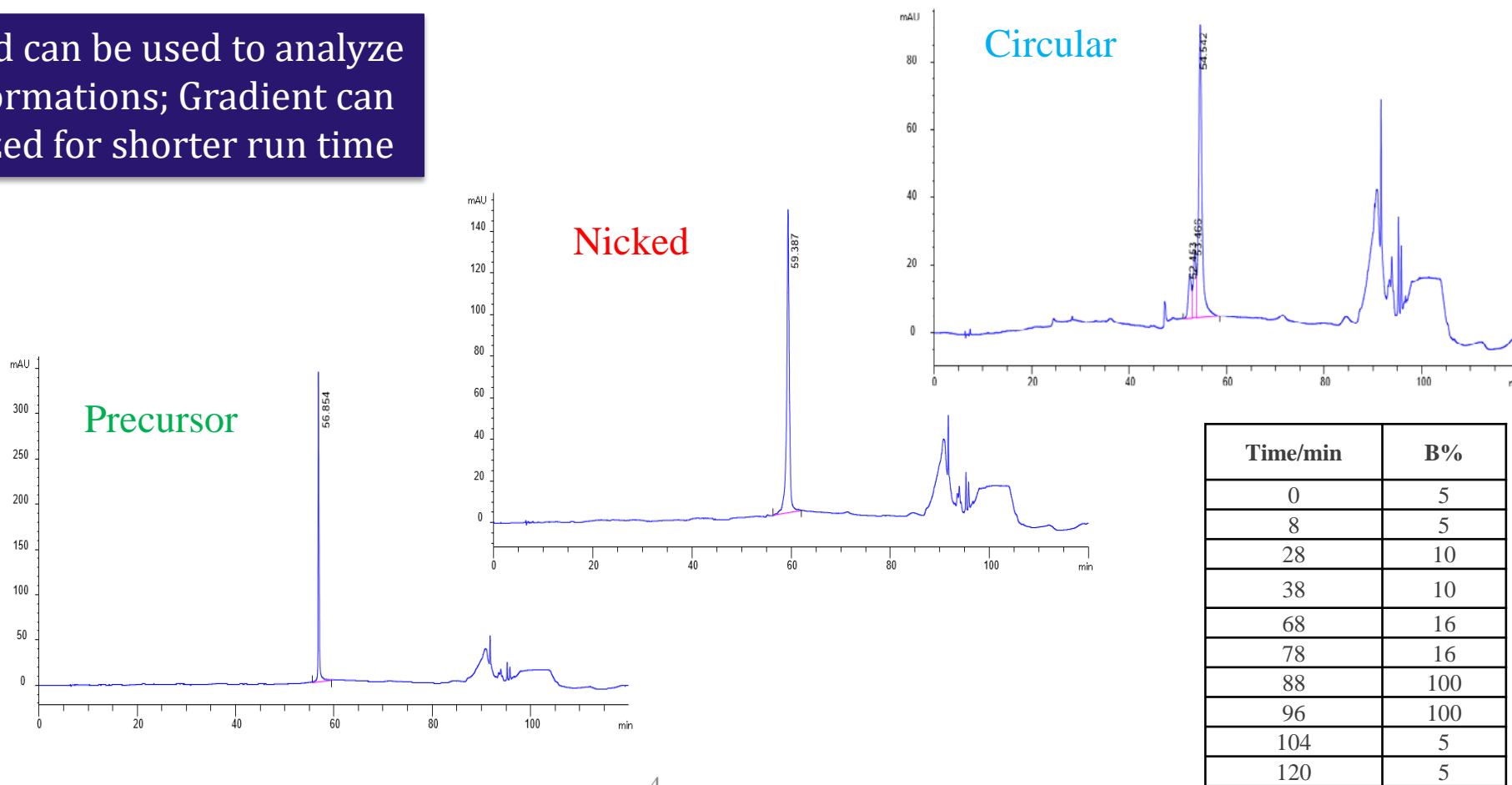


circRNAs – RP method



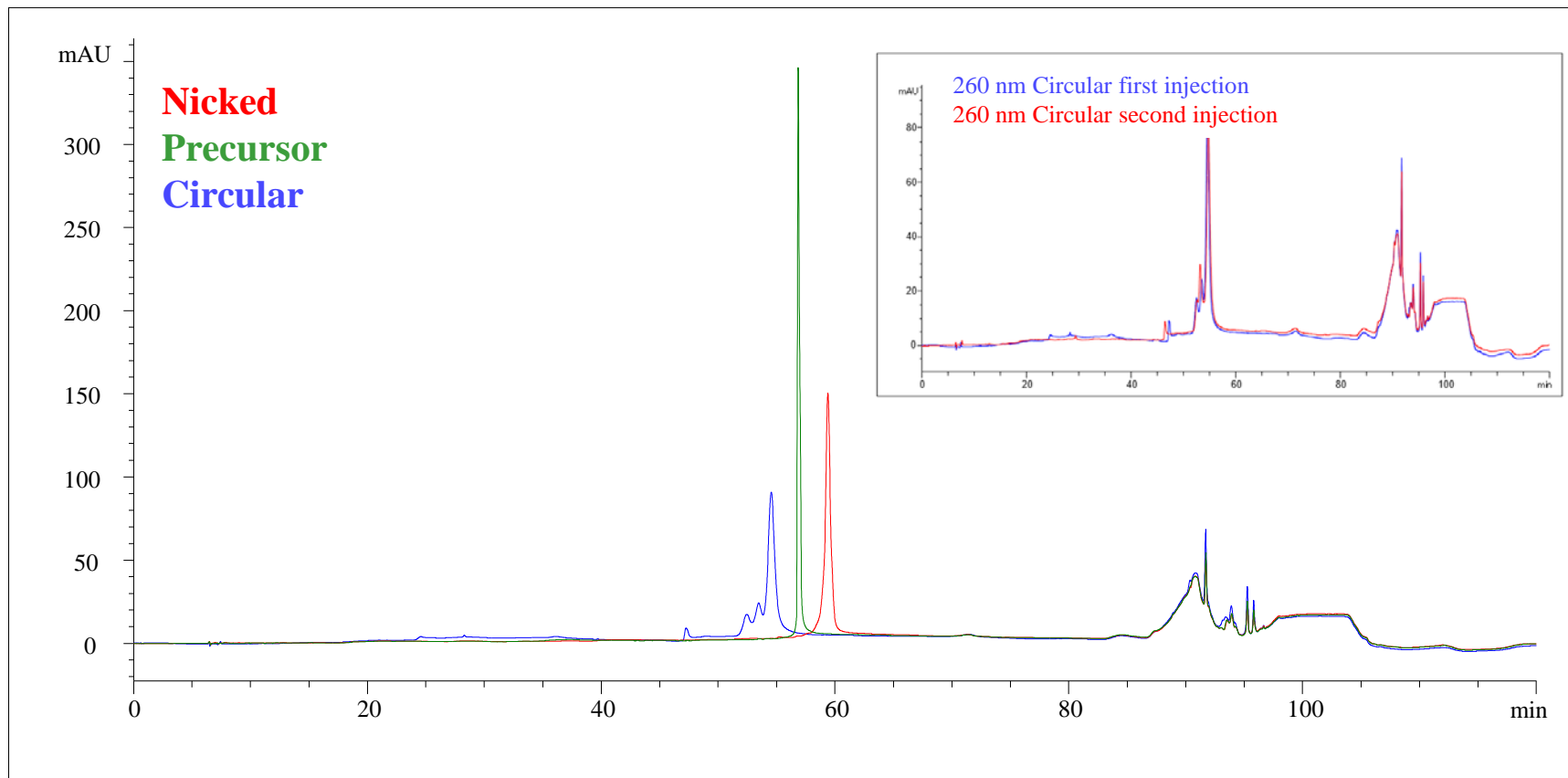
Column: circRNA-specific column Bio-C18, 5 μm , 300 \AA , 4.6 \times 250 mm; Part Number: 106185-4625;
Mobile Phase: A: 100 mM TEAA, B: 95% ACN + 5% A; Flow Rate: 0.5 mL/min; Detector: UV at 260 nm;
Column Temperature: 50 $^{\circ}\text{C}$; Injection Volume: 2 μL , 1 mg/mL; Column Pressure: 57 bar

Bio-C18 RP method can be used to analyze the different conformations; Gradient can be further optimized for shorter run time





circRNAs - RP method overlays





Literature Review – circRNA Separation



App ID	Paper Title
<u>LR202202962</u>	RNA Circularization Diminishes Immunogenicity and Can Extend Translation Duration In Vivo
<u>LR2022021136</u>	Design and application of circular RNAs with protein-sponge function
<u>LR2022021128</u>	Engineered circular ADAR-recruiting RNAs increase the efficiency and fidelity of RNA editing in vitro and in vivo
<u>LR2022021104</u>	Intratumoral Delivered Novel Circular mRNA Encoding Cytokines for Immune Modulation and Cancer Therapy
<u>LR2022021101</u>	RNA circles with minimized immunogenicity as potent PKR inhibitors
<u>LR2022021052</u>	Engineering circular RNA for potent and stable translation in eukaryotic cells



Order Info

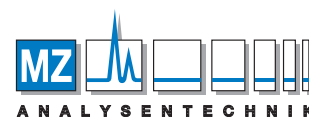


Product	Particle Size	Pore Size	Column Size	PN
SRT SEC-2000	5 μm	2000 \AA	7.8 \times 300 mm	215980-7830
Bio-C18	5 μm	300 \AA	4.6 \times 250 mm	106185-4625

Check our website for more products and application data

www.sepax-tech.com

SEC | Affinity | IEX | HIC | RP | Service



AUTHORIZED DISTRIBUTOR

MZ-Analysentechnik GmbH, Barcelona-Allee 17 • D-55129 Mainz

Tel +49 6131 880 96-0, Fax +49 6131 880 96-20

e-mail: info@mz-at.de, www.mz-at.de

SR1002