a simple and standardized kit
qPCR analysis of circulating platelet-derived microRNAs

Cardiovascular Diseases

Neurodegenerative Diseases

Musculoskeletal Diseases

microRNA Biomarkers of Platelet Function

stability for life.
Circulating microRNAs are a novel class of blood-borne biomarkers. They are secreted from virtually any cell in the human body and distributed to other cells via the circulation. Local pathophysiological processes in tissues can be detected using circulating microRNAs, and used for diagnosis and treatment monitoring of age-associated diseases.

The thrombomiR™ kit enables simple and standardized analysis of microRNA biomarkers for platelet function.

**thrombomiR™ kit applications**

- **Monitor the drug effects** on platelet function, reactivity and hemostasis
- **Diagnosis of platelet-related disorders**

**Unique features of the thrombomiR™ kit:**

- **Works with frozen sample material** (serum or plasma)
- **Responds to all platelet-activating signals** (ADP, Collagen, etc.)
- **Measures a platelet-signal generated in-vivo** thus complementing results from ex vivo platelet-function tests (LTAs, VASP, ...)

The thrombomiR™ test should be used with platelet-poor plasma. Alternatively, serum can be used if coagulation time has been kept constant.
How does it work?

**all-in-one RT-qPCR kit**
the thrombomiRTM kit contains all necessary reagents for:

1. RNA extraction
2. cDNA synthesis
3. Preparation of qPCR Mix
4. Real time qPCR analysis
5. Data analysis: proprietary software

Which type of samples can be used?

### Platelet miRNA content in different blood components

<table>
<thead>
<tr>
<th>Blood Component</th>
<th>miRNA Content</th>
</tr>
</thead>
<tbody>
<tr>
<td>PRP</td>
<td>platelet miRNA content decreases</td>
</tr>
<tr>
<td>Serum</td>
<td>leukocyte miRNA release during coagulation</td>
</tr>
<tr>
<td>Conventional Plasma</td>
<td>residual platelets</td>
</tr>
<tr>
<td>PPP</td>
<td>baseline miRNA release</td>
</tr>
</tbody>
</table>

The thrombomiRTM test should be used with platelet-poor plasma. Alternatively, serum can be used if coagulation time has been kept constant. Visit our website at [www.tamirna.com/sample-requisites](http://www.tamirna.com/sample-requisites) for further information.
Assay format

- **Low sample volume:** 200 µL human plasma/serum
- **Platelet function analysis based on the thrombomiR™ signature:**
  11 thrombomiRs™ and 5 controls/sample
- **Reduced hands-on time:** primer coated 96-well plates
- **High throughput:** one kit allows analysis of up to 48 samples
  (6 samples/plate, 8 plates/kit)
- **Fast and simple data analysis:** thrombomiR™ software included

### microRNAs included in the thrombomiR™ kit

<table>
<thead>
<tr>
<th>microRNA</th>
<th>platelet enrichment *</th>
<th>myocardial infarction **</th>
<th>T2D complications ***</th>
<th>tissue / cellular origin</th>
</tr>
</thead>
<tbody>
<tr>
<td>hsa-miR-126-3p</td>
<td>+++</td>
<td></td>
<td>●</td>
<td>platelets &amp; endothelium</td>
</tr>
<tr>
<td>hsa-miR-223-3p</td>
<td>+++</td>
<td></td>
<td>●</td>
<td>platelets &amp; hematopoietic cells</td>
</tr>
<tr>
<td>hsa-miR-191-5p</td>
<td>+++</td>
<td></td>
<td></td>
<td>ubiquitous</td>
</tr>
<tr>
<td>hsa-miR-24-3p</td>
<td>++</td>
<td></td>
<td></td>
<td>platelets &amp; epithelial cells</td>
</tr>
<tr>
<td>hsa-miR-21-5p</td>
<td>++</td>
<td></td>
<td></td>
<td>platelets</td>
</tr>
<tr>
<td>hsa-miR-28-3p</td>
<td>++</td>
<td></td>
<td></td>
<td>platelets &amp; hematopoietic cells</td>
</tr>
<tr>
<td>hsa-miR-320a</td>
<td>++</td>
<td></td>
<td></td>
<td>ubiquitous</td>
</tr>
<tr>
<td>hsa-miR-150-5p</td>
<td>+</td>
<td></td>
<td>●</td>
<td>platelets &amp; hematopoietic cells</td>
</tr>
<tr>
<td>hsa-miR-197-3p</td>
<td>+</td>
<td></td>
<td></td>
<td>platelets</td>
</tr>
<tr>
<td>hsa-miR-27b-3p</td>
<td>+</td>
<td></td>
<td>●</td>
<td>ubiquitous</td>
</tr>
<tr>
<td>hsa-miR-122-5p</td>
<td>–</td>
<td></td>
<td></td>
<td>liver</td>
</tr>
</tbody>
</table>

* Kaudewitz D et al 2016   ** platelet associated disorders   *** negative control

### Key publications

Clinical utility of the thrombomiR™ kit

- Platelet microRNAs are released from cells upon activation.
- Release is independent of the activation pathway (e.g. ADP, collagen, etc.).
- MicroRNAs are protected from degradation in serum/plasma due to vesicular encapsulation.

Correlation between ex vivo platelet aggregation tests and platelet-derived microRNAs.

Correlation between microRNA levels and results of VerifyNow test (A) and the VASP assay (B) in patients on dual antiplatelet therapy for 30 days post acute coronary syndrome. PRU denotes P2Y12 reaction units (y axis). Higher PRU values reflect higher P2Y12-mediated platelet reactivity.


Low levels of thrombomiRs in serum are associated with lower risk of cardiovascular death.

High baseline levels ("upper third") of miR-197 and miR-223 are associated with reduced survival (due to cardiovascular death) in a cohort of 873 patients, of which 340 are cases with acute coronary syndrome and 533 cases of stable angina pectoris.

From Schulte, C., et al. PLoS ONE, 10(12), pp. 1–12, Figure 1
Low levels of thrombomiRs in serum are associated with lower risk of cardiovascular death.

Correlation between ex vivo platelet aggregation tests and platelet-derived microRNAs.

Clinical utility of the thrombomiR™ kit

High baseline levels ("upper third") of miR-197 and miR-223 are associated with reduced survival (due to cardiovascular death) in a cohort of 873 patients, of which 340 are cases with acute coronary syndrome and 533 cases of stable angina pectoris.

From Schulte, C., et al. PLoS ONE, 10(12), pp. 1–12., Figure 1

Correlation between microRNA levels and results of VerifyNow test (A) and the VASP assay (B) in patients on dual anti platelet therapy for 30 days post acute coronary syndrome. PRU denotes P2Y12 reaction units (y axis). Higher PRU values reflect higher P2Y12-mediated platelet reactivity.