Πλειοτροπικές δράσεις αντιαιμοπεταλιακών φαρμάκων

Αλέξανδρος Δ. Τσελέπης, MD, PhD
Καθηγητής Βιοχημείας – Κλινικής Χημείας
Ανταγωνιστές του P2Y12

Figure adapted from Schömig A. N Engl J Med 2009;361:1108–1111
Ticagrelor consistently and effectively inhibits platelet P2Y$_{12}$ receptor activation.
Tissue distribution of the P2Y12

microglial cells

platelets

vascular smooth muscle cells

leukocytes

macrophages

certain subregions of the brain

Κυτταρική ενεργοποίηση από τα αιμοπετάλια

May A, et al. ATVB 2008;28:s5-s10
Η Αθηροσκλήρωση είναι μια Φλεγμονώδης Νόσος
<table>
<thead>
<tr>
<th>Δείκτες φλεγμονής</th>
<th>Πληθυσμός μελέτης</th>
<th>Κλινική έκβαση</th>
</tr>
</thead>
<tbody>
<tr>
<td>CD40L</td>
<td>Ασθενείς με ΟΣΣ ή ασταθή στηθάγχη</td>
<td>Αυξημένος κίνδυνος καρδιαγγειακών επεισοδίων</td>
</tr>
<tr>
<td>sCD40L</td>
<td>Ασθενείς μετά από PCI ή με ασταθή στηθάγχη</td>
<td>Αυξημένος κίνδυνος καρδιαγγειακών επεισοδίων</td>
</tr>
<tr>
<td>ICAM-1</td>
<td>Ασθενείς που υποβάλλονται σε στεφανιαία αγγειογραφία</td>
<td>Αυξημένος κίνδυνος KN</td>
</tr>
<tr>
<td>sICAM-1</td>
<td>Υγιείς εθελοντές (άντρες)</td>
<td>Αυξημένος κίνδυνος συμπτωματικής αρτηριακής νόσου</td>
</tr>
<tr>
<td>MCP-1</td>
<td>Ασθενείς με ΠΑΝ</td>
<td>Αυξημένος κίνδυνος KN</td>
</tr>
<tr>
<td>HsCRP</td>
<td>Ασθενείς με σταθερή ή ασταθή στηθάγχη</td>
<td>Μελλοντικά ισχαιμικά επεισόδια</td>
</tr>
<tr>
<td>IL-6</td>
<td>Ασθενείς που υποβάλλονται σε στεφανιαία αγγειογραφία, Ασθενείς με οξύ καρδιοεμβολικό ισχαιμικό εγκεφαλικό επεισόδιο</td>
<td>Αυξημένος κίνδυνος KN Οξεία φλεγμονώδης αντίδραση</td>
</tr>
<tr>
<td>TNF-α</td>
<td>Ασθενείς με ΚΝ και ασθενείς με οξύ καρδιοεμβολικό ισχαιμικό εγκεφαλικό επεισόδιο</td>
<td>Αυξημένα επίπεδα του TNF-α Οξεία φλεγμονώδης αντίδραση</td>
</tr>
<tr>
<td>Συσσωματώματα αιμοπεταλίων-λευκοκυττάρων</td>
<td>Ασθενείς με ασταθή στηθάγχη, ασθενείς με OEM, ασθενείς που υποβάλλονται σε PCI, ασθενείς με ΠΑΝ</td>
<td>Αυξημένα επίπεδα των συσσωματωμάτων αιμοπεταλίων-λευκοκυττάρων</td>
</tr>
</tbody>
</table>

Αντιαιμοπεταλιακά φάρμακα

Υπάρχουν πλειοτροπικές δράσεις;
Clopidogrel inhibits platelet-mediated inflammatory response

<table>
<thead>
<tr>
<th>Antiplatelet Drug</th>
<th>Studied Population</th>
<th>Inflammatory Marker(s) Measured</th>
<th>Study Design</th>
<th>Follow-Up</th>
<th>Outcome Related to Inflammation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aspirin</td>
<td>CAD patients, ACS patients</td>
<td>hsCRP, IL-6, MCP-1, TGF-β, TNF-α</td>
<td>Randomized</td>
<td>1-year</td>
<td>Reduction</td>
</tr>
<tr>
<td>Clopidogrel</td>
<td>ACS patients</td>
<td>CD40L, P-selectin, Platelet-leukocyte conjugates</td>
<td>Case control</td>
<td>24-hours</td>
<td>Reduction</td>
</tr>
<tr>
<td></td>
<td>Patients undergoing PCI</td>
<td>hsCRP</td>
<td>Post hoc analysis</td>
<td>1-year</td>
<td>Reduction</td>
</tr>
<tr>
<td>Prasugrel</td>
<td>Patients undergoing PCI</td>
<td>CD40L, P-selectin</td>
<td>Randomized</td>
<td>1-month</td>
<td>Reduction</td>
</tr>
<tr>
<td>Aspirin+Clopidogrel</td>
<td>NSTEMI patients</td>
<td>hsCRP, sCD40L</td>
<td>Randomized</td>
<td>6-days</td>
<td>Reduction</td>
</tr>
<tr>
<td>Aspirin+Clopidogrel</td>
<td>CAD patients</td>
<td>P-selectin</td>
<td>Randomized</td>
<td>29-days</td>
<td>Lower reduction in aspirin+clopidogrel group compared with aspirin+prasugrel group</td>
</tr>
<tr>
<td>Aspirin+Prasugrel</td>
<td>ACS patients</td>
<td>P-selectin, CD40L</td>
<td>Randomized</td>
<td>1-year</td>
<td>Inhibition</td>
</tr>
<tr>
<td>Abciximab</td>
<td>Patients undergoing PCI</td>
<td>hsCRP, IL-6, TNF-α</td>
<td></td>
<td></td>
<td>Inhibition</td>
</tr>
<tr>
<td>Abciximab</td>
<td>Patients undergoing PCI</td>
<td>IL-6, CRP, fibrinogen</td>
<td>Randomized</td>
<td>72-hours</td>
<td>No effect</td>
</tr>
<tr>
<td>Eptifibatide</td>
<td>Patients undergoing PCI</td>
<td>sCD40L, RANTES</td>
<td>Randomized</td>
<td>24-hours</td>
<td>Reduction</td>
</tr>
<tr>
<td>Tirofiban</td>
<td>NSTEMI patients</td>
<td>hsCRP</td>
<td>Randomized</td>
<td>72-hours</td>
<td>Reduction</td>
</tr>
<tr>
<td>Tirofiban</td>
<td>UA patients</td>
<td>hsCRP</td>
<td>Case control</td>
<td>72-hours</td>
<td>No effect</td>
</tr>
</tbody>
</table>
Αναγέννηση του Ενδοθηλίου από τα EPCs

May A, et al. ATVB 2008;28:s5-s10
Levels of leukocytes and CD34$^+$ cells in peripheral blood

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Healthy Individuals</th>
<th>ACS Patients</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Baseline</td>
</tr>
<tr>
<td>Leukocytes (x10$^9$/L)</td>
<td>6.37±1.78</td>
<td>11.5±2.50 *</td>
</tr>
<tr>
<td>Lympho-monocytes (x10$^9$/L)</td>
<td>2.55±0.89</td>
<td>3.30±1.44</td>
</tr>
<tr>
<td>CD34$^+$ (% of lympho-monocytes)</td>
<td>0.78±0.40</td>
<td>0.39±0.17 *</td>
</tr>
<tr>
<td>Number of CD34$^+$/µl of blood</td>
<td>17.01±7.02</td>
<td>12.71±8.12 *</td>
</tr>
<tr>
<td>Number of CD34$^+$/1000 leukocytes</td>
<td>2.80±0.84</td>
<td>1.08±0.25 *</td>
</tr>
</tbody>
</table>

Tatsidou P, et al. ESC 2014
Levels of CD34+/KDR+ cells, in ACS patients and healthy individuals before and after platelet activation with ADP or TRAP

*\(p<0.01\) compared to healthy individuals, \#\(p<0.01\) compared to unactivated and \$\(p<0.01\) compared to activated with ADP, \&\(p<0.01\) compared to Baseline

Tatsidou P, et al. ESC 2014
Platelet conjugates with CD34+/KDR+ in ACS patients and healthy individuals, before and after platelet activation with ADP or TRAP-14

\[ p^*<0.01 \text{ and } **p<0.05 \text{ compared to healthy individuals, } \#p<0.01 \text{ and } \$p<0.05 \text{ compared to Baseline} \]

Tatsidou P, et al. ESC 2014
CONCLUSION

Patients with an ACS present low levels of CD34+ and CD34+/KDR+ cells, as well as conjugates of these cells with platelets in their peripheral blood. This phenomenon that may represent an important defect of ACS patients towards vascular regeneration, which is significantly improved after DAPT with aspirin and clopidogrel.
PLATO: Primary Efficacy Analysis Cardiovascular Death, MI, or Stroke

Cumulative Incidence (%)

<table>
<thead>
<tr>
<th>Days After Randomization</th>
<th>Clopidogrel</th>
<th>Ticagrelor</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>60</td>
<td></td>
<td></td>
</tr>
<tr>
<td>120</td>
<td></td>
<td></td>
</tr>
<tr>
<td>180</td>
<td></td>
<td></td>
</tr>
<tr>
<td>240</td>
<td></td>
<td></td>
</tr>
<tr>
<td>300</td>
<td></td>
<td></td>
</tr>
<tr>
<td>360</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

HR, 0.84 (95% CI, 0.77-0.92) \( P<0.001 \)

Death
- Ticagrelor: 5.9, 4.5 (22% decrease)
- Clopidogrel: 6.9, 5.8 (16% decrease)

Nonfatal MI
- Ticagrelor: 9,333, 8,628, 8,460, 8,219, 6,743, 5,161, 4,147
- Clopidogrel: 9,291, 8,521, 8,362, 8,124, 6,650, 5,096, 4,047

CI=confidence interval; HR=hazard ratio; MI=myocardial infarction
This information concerns a use that has not been approved by the US Food and Drug Administration.
PLATO: Ticagrelor is associated with significant reductions in CV death and All-cause death in the total ACS population and patient subgroups

<table>
<thead>
<tr>
<th>Patient population</th>
<th>CV death</th>
<th></th>
<th>All-cause death</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Ticagrelor</td>
<td>Clopidogrel</td>
<td>p value</td>
<td>Ticagrelor</td>
</tr>
<tr>
<td>Total (n=18,624)</td>
<td>4.0%</td>
<td>5.1%</td>
<td>0.001</td>
<td>4.5%</td>
</tr>
<tr>
<td>Intent for invasive (n=13,408)</td>
<td>3.4%</td>
<td>4.3%</td>
<td>0.025</td>
<td>3.9%</td>
</tr>
<tr>
<td>Intent for non-invasive (n=5216)</td>
<td>5.5%</td>
<td>7.2%</td>
<td>0.019</td>
<td>6.1%</td>
</tr>
<tr>
<td>CABG (n=1258)</td>
<td>4.1%</td>
<td>7.9%</td>
<td>0.009</td>
<td>4.7%</td>
</tr>
</tbody>
</table>

Despite higher levels of platelet inhibition with Ticagrelor compared with Clopidogrel, similar rates of major bleeding were observed in the PLATO study.

Important Issues Raised from PLATO

- PLATO is the only study that has shown a significant CV mortality benefit of an oral antiplatelet agent compared with an active control
  - This benefit does not appear to be related to differences in bleeding
  - This suggests that the benefit is not solely due to more potent platelet P2Y\textsubscript{12} inhibition
    - It has been hypothesised that additional effects beyond the degree of inhibition of P2Y\textsubscript{12} may contribute to the clinical benefits (including CV morality) observed with ticagrelor

- Studies have demonstrated that dyspnoea is more common with ticagrelor than with clopidogrel

- Dyspnoea is a known effect of adenosine
Ticagrelor - Adenosine
ATP
Δεσμοί υψηλής ενέργειας
Φωσφορικές ομάδες

ADP

Adenosine

Αδενίνη

Ριβόζη
The Purinergic Receptor Family

P1 Adenosine

G protein coupled

P2 ATP ADP

Ligand-gated channels

G protein coupled

A1 A2A A2B A3

P2X1-7

P2Y1,2,4,6,11-14
Adenosine Purinergic Receptors
The Equilibrative Nucleoside Transporter-1 (ENT-1)

Adenosine

Dypiridamole

Preclinical investigations of Ticagrelor’s adenosine mode of action

Ticagrelor dose-dependently inhibits adenosine uptake in human RBC

Adenosine Transporter and Receptors

Ticagrelor
Extracellular Adenosine Signaling

The Potentially Beneficial Role of Adenosine in ACS

- Vasodilation
- Cardioprotection
- Modulation of inflammation
- Inhibition of platelet function
Ticagrelor inhibits adenosine uptake from RBC of ACS patients through ENT-1 transporter

Ticagrelor, but not Prasugrel active metabolite, delays adenosine degradation

24-h systemic potential versus minimal systemic potential

Compared with the short plasma exposure of Prasugrel and Clopidogrel active metabolites, Ticagrelor has significant 24-h systemic exposure of a direct active compound\textsuperscript{1,2}

Graph produced with human data from Wallentin L, et al. (2008) and Storey RF, et al. (2007)
Ticagrelor increases plasma adenosine levels in ACS patients

Ticagrelor does not inhibit adenosine A1 and A2A receptors in ACS patients

Ticagrelor inhibits human platelet aggregation via adenosine in addition to P2Y\textsubscript{12} antagonism

- In whole blood, adenosine contributed an additional antiplatelet effect when in combination with ticagrelor but not prasugrel-AM.

![Graph showing platelet aggregation](image)

- Residual collagen-induced platelet aggregation in whole blood, in the presence of added compounds alone or in combination.
- Adenosine 7.1 μmol/L, dipyridamole 14 μmol/L, ticagrelor 14 μmol/L, prasugrel-AM 14 μmol/L and ZM241385 14 μmol/L.

The dual pathway mediates both antiplatelet effects and an enhanced adenosine response

- Inhibition of \( \text{P}2\text{Y}_{12} \) receptor\(^1,2\)
  - Anti-platelet effect
- Inhibition of \( \text{ENT}-1 \) transporter\(^3,4,5\)
  - Enhanced local adenosine response may result in:
    - Additional inhibition of platelet aggregation/activation\(^3\)
    - Cardioprotection\(^6\)
    - Vasodilation\(^5,7,8\)
    - Modulation of inflammation
    - Dyspnœa\(^7\)

*See back-up slides for further information; \(^1\)See figure. Figure adapted from Nylander S, et al. (2013). AC, adenylyl cyclase; ADP, adenosine diphosphate; cAMP, cyclic adenosine monophosphate; ENT, equilibrative nucleoside transporter. References in slide notes.
Effect of ticagrelor on peripheral endothelial function in stable ACS patients

Study objective: To compare peripheral vascular endothelial function in stable ACS patients treated with ticagrelor against patients treated with ASA alone, clopidogrel or prasugrel [Torngren 2013]

*All patients except two were receiving ASA 75 mg QD maintenance therapy; †Administered for ACS, acute coronary syndromes; ASA, acetylsalicylic acid; BID, twice daily; QD, once daily. Torngren K, et al. Cardiol 2013;124:252–258.
Ticagrelor improves peripheral endothelial function in ACS patients compared with ASA, Clopidogrel or Prasugrel


**p<0.01 vs.control; ***p<0.001 vs. control. ACS, acute coronary syndromes; ASA, acetylsalicylic acid; RHI, reactive hyperaemia index.
Fewer patients treated with Ticagrelor had endothelial dysfunction compared with ASA, Clopidogrel or Prasugrel

*\(p<0.05\) versus control; \(^\dagger\) Endothelial dysfunction defined as RHI <1.67.

ACS, acute coronary syndromes; ASA, acetylsalicylic acid; BID, twice daily; QD, once daily; RHI, reactive hyperaemia index.

Ticagrelor augments adenosine-induced CBFV

CBFV, coronary blood flow velocity
Effect of ticagrelor on adenosine-induced increases in CBFV is concentration-dependent

$AUC$ change

$r=0.530$  
$p<0.001$

Ticagrelor concentration (µmol/L)

Ticagrelor augments adenosine-induced dyspnoea*

*Scored using the Modified Borg Scale, from 0 (no sensation of dyspnoea) to 10 (maximum sensation of dyspnoea). Wittfeldt A, et al. *J Am Coll Cardiol* 2013;61:723–727
Ticagrelor increases adenosine-induced CBFV in NSTE-ACS patients relative to prasugrel.

*Significantly higher ratio of LAD maxCBFV/bCBFV for ticagrelor vs. prasugrel.

AUC, area under the curve; CBFV, coronary blood flow velocity; CI, confidence interval; LAD, left anterior descending artery; LS, least squares; NSTE-ACS, non-ST-segment elevation acute coronary syndromes.

Similarities Between Ticagrelor and Adenosine Effects

<table>
<thead>
<tr>
<th>Ticagrelor</th>
<th>Adenosine</th>
</tr>
</thead>
<tbody>
<tr>
<td>↑ Adenosine-induced increases in coronary blood flow (dogs and humans)</td>
<td>↑ Vasodilation</td>
</tr>
<tr>
<td>↑ Endothelial function (ACS patients)</td>
<td>↑ Endothelial progenitor cell migration</td>
</tr>
<tr>
<td>↓ Incidence of MACE (ACS patients)</td>
<td>↓ Ischemia/reperfusion injury</td>
</tr>
<tr>
<td>↓ CV and all cause mortality (ACS patients)</td>
<td>induces pharmacological preconditioning</td>
</tr>
<tr>
<td>↑ Incidence of ventricular pauses (ACS patients)</td>
<td>↓ Electrical conduction</td>
</tr>
<tr>
<td>↓ Infarct size (animal models)</td>
<td></td>
</tr>
<tr>
<td>↑ Adenosine-induced platelet inhibition (in vitro)</td>
<td>↑ Platelet inhibition</td>
</tr>
<tr>
<td>↓ Mortality (ACS patients with pulmonary infection)</td>
<td>Modulates inflammation</td>
</tr>
<tr>
<td>↑ Creatinine levels (ACS patients)</td>
<td>↓ Glomerular filtration</td>
</tr>
<tr>
<td>↑ Incidence of dyspnea (ACS patients)</td>
<td>↑ Incidence of dyspnea</td>
</tr>
<tr>
<td>↑ Adenosine-induced dyspnea (healthy subjects)</td>
<td></td>
</tr>
</tbody>
</table>

Cattaneo M, et al. JACC. 2014; In press
Conclusions

- Ticagrelor inhibits cellular adenosine uptake via ENT-1
- Ticagrelor inhibits platelets via the platelet (P2Y\textsubscript{12}) pathway and the adenosine (ENT-1) pathway
- Ticagrelor improves peripheral vascular endothelial function
- Ticagrelor increases adenosine-induced CBFV in a concentration-dependent manner
- Ticagrelor also increases the adenosine-induced sensation of dyspnoea in healthy volunteers
- The adenosine-related mode of action of Ticagrelor may help explain the CV mortality benefit of Ticagrelor over Clopidogrel and why Ticagrelor is associated with dyspnoea

CBFV, coronary blood flow velocity; CV, cardiovascular; ENT-1, equilibrative nucleoside transporter-1.