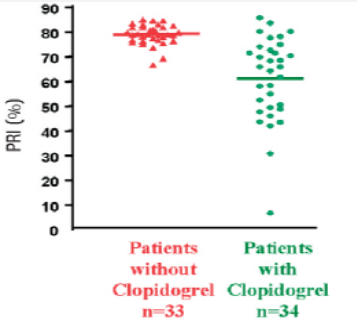
PLT VASP/P2Y12 Test kit

Principle of the assay

The **PLT VASP/P2Y12 Test kit** is qualified to monitor specific platelet ADP receptor (P2Y12) antagonists.
The blood sample is first incubated with PGE1 alone or with PGE1 + ADP for 10 minutes. Afterwards, the cells are permeabilized, and the phosphorylated VASP is labelled by indirect immunostaining with a specific monoclonal antibody (clone 16C2). The platelets are counterstained with an anti-CD61 specific antibody. Dual color flow cytometry analysis allows to compare the two tested conditions and to evaluate for each sample the capacity of ADP to inhibit VASP phosphorylation.



A platelet reactivity index (PRI) is calculated using corrected mean fluorescence intensities (MFIc) in the presence of PGE1 alone (PGE1) or PGE1 and ADP simultaneously (PGE1+ ADP).

Aleil et al. J. Thromb Haemost. 2005, 3, 85-92

Indications

- Monitoring of specific platelet ADP receptor antagonists
- Thienopyridine resistance (Clopidogrel, Ticlopidin)
- Risk assessment of coronary subacute stent thrombosis (SAT).

Key Features

- **Assay:** Standardized Flow cytometric assay
- **Type:** Dual color, no compensation necessary
- **Time:** 30 min protocol
- **Validated on:** BD FACSCalibur and Beckman Coulter Instrument type XL and XL MCL
- **Sample type:** Whole blood
- **Sample Volume:** 30 µL
- **Sample Stability:** Up to 48 hours at room temperature
- **Assay Range**: 0 - 100% of PRI
- **Correlation:** with aggregometry (gold standard): r= 0.72, p < 0.0001

- **No interference of ASA, Reopro and Integrilin**

- **For in vitro diagnostic use**

Scientific information **PLT VASP/P2Y12** ….over

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Platelet Vasodilator Stimulated Phosphoprotein (PLT VASP)

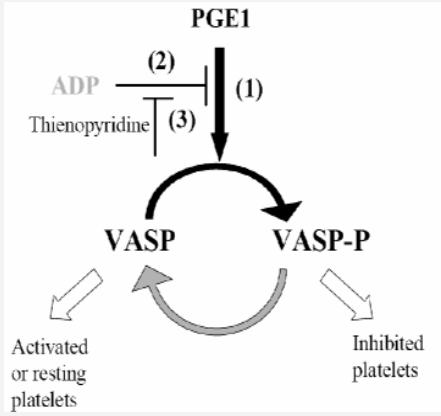
Introduction

VASP is an intracellular platelet protein, which is non-phosphorylated at basal state. The phosphorylation of VASP is dependent on the level of activation of the platelet P2Y12 receptor, which is targeted by thienopyridine derivatives.

VASP phoshorylation is regulated by the cAMP (cyclic Adenosine Monophosphate) cascade. PGE1 (Prostaglandin E1) activates this cascade **(1)**, whereas it is inhibited by ADP (Adenosine Diphosphate) through P2Y12 receptors **(2)**.

VASP phosphorylation correlates with the P2Y12 receptor inhibition, whereas its non-phosphorylation state correlates with the active form of P2Y12 receptor.

Inter-individual variability and resistance to thienopyridines have been widely described. The effect of thienopyridines **(3)** can be measured by the persistence of VASP phosphorylation induced by PGE1 even with the simultaneous addition of ADP.



Diagnostic relevance

The phosphorylation of vasodilator-stimulated phosphoprotein (VASP) depends on the level of activation of the platelet P2Y12 receptor. The P2Y12 receptor is targeted by thienopyridine derivatives, like clopidogrel (Plavix®, Iscover®), and ticlopidine (Ticlid®).

Interindividual variability of the inhibitory effect of thienopyridine derivatives on platelet functions may lead to incomplete P2Y12 receptor inhibition, so-called “thienopyridine resistance”.

Thienopyridine resistance has been observed in some patients with ischemic cardiovascular disease. Moreover, it has been described as a risk factor for subacute stent thrombosis (SAT).

Measurement of the VASP phosphorylation state has high sensitivity and specificity for thienopyridine treatment resistance and good correlation with the platelet aggregation measured within 2 hours. VASP phosphorylation analysis may be useful to assess the risk for coronary subacute stent thrombosis (SAT).

References

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- High stability of blood samples for flow cytometric analysis of VASP phosphorylation to measure the clopidogrel responsiveness in patients with coronary artery disease. Aleil et al. Thromb Haemost 2005, 94(4):886-887.
- Resistance to thienopyridines: clinical detection of coronary stent thrombosis by monitoring of vasodilator-stimulated phosphoprotein phosphorylation. Barragan et al. Cathet Cardiovasc Intervent 2003, 59: 295-302.
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