Appendix G. Detailed Assessment of the Strength of Evidence

Appendix Table G1. Assessment of strength of evidence domains

| **Key Question** | **Population** | **Test/Assay** | **Outcome** | **Risk of bias** | **Directness** | **Consistency** | **Precision** | **Reporting bias** | **Other Issues/Notes** | **SOE and additional information** |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **1a: What it the analytic validity of tests for genotyping CYP2C19 variants?** | NA | Genotyping for any CYP2C19 variant | NA | NA | NA | NA | NA | NA | Few studies provided information on analytic validity specifically using samples obtained from patient populations relevant to this review. When available, data were limited to test–retest reliability or inter-assay agreement. | SOE was not evaluated. |
| **1b: What is the predictive value of genetic testing for CYP2C19 variants?** | Ischemic heart disease | Genotyping for LOF CYP2C19 variants | Stent thrombosis | Intermediate | Direct | Consistent | Somewhat imprecise; but the lower bound of the confidence interval of the summary effect indicated a 17% increase in risk | Suspected (publication bias and selective outcome reporting) | None | Moderate |
|  |  |  | MACE | Intermediate | Direct | Consistent | Precise | Suspected (publication bias) | None | Moderate |
|  |  |  | Cardiovascular mortality | Intermediate | Direct | Consistent | Imprecise | Suspected (selective outcome reporting) | None | Low |
|  |  |  | All other clinical outcomes | Intermediate | Direct | Consistent or not enough data to assess (single study) | Imprecise | Suspected (selective outcome reporting; in some cases publication bias) | None | Insufficient |
|  |  | Genotyping for GOF CYP2C19 variants | MACE | Intermediate | Direct | Consistent | Somewhat imprecise | Suspected (selective outcome reporting) | None | Low |
|  |  |  | All other clinical outcomes | Intermediate | Direct | Somewhat inconsistent for bleeding events; consistent for all other outcomes | Imprecise | Suspected (selective outcome reporting) | None | Insufficient |
|  | Other patient groups who are candidates for clopidogrel therapy | Genotyping for any CYP2C19 variants | All clinical outcomes | Intermediate | Direct | It was not possible to evaluate consistency because studies were conducted in diverse populations and reported information on different outcomes | Imprecise | Suspected (selective outcome reporting) | None | Insufficient |

| **Key Question** | **Population** | **Test/Assay** | **Outcome** | **Risk of bias** | **Directness** | **Consistency** | **Precision** | **Reporting bias** | **Other Issues/Notes** | **SOE and additional information** |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **1c: What factors affect the predictive value of genetic testing for CYP2C19 variants?** | All patient populations | Genotyping for any CYP2C19 variants | All clinical outcomes | Intermediate | Direct | Inconsistent | Imprecise | Suspected (selective outcome reporting) | In meta-regression analyses we found some evidence of effect modification by ethnicity (East Asians vs. White) for MACE and stent thrombosis. However, this result was based on comparisons across studies, which may be confounded by other study characteristics, and was not corroborated by within-study analyses. | Insufficient |
| **2a: What is the analytic validity of tests for on-clopidogrel platelet reactivity?** | NA | All assays used to measure on-clopidogrel platelet reactivity | NA | NA | NA | NA | NA | NA | Few studies reported information on analytic sensitivity and specificity, possibly reflecting the research community’s belief that there is no good reference standard assay for platelet reactivity. Agreement ranged from poor to moderate and was variable between tests. The highest agreement was observed between applications of the same assay with different concentrations of agonists, rather than between different assays. | SOE was not evaluated. |
| **2b: What is the predictive ability of phenotypic testing for platelet reactivity?** | Ischemic heart disease | LTA | All-cause mortality | Intermediate | Direct | It was not possible to evaluate consistency with respect to the effect size of the association because studies used different metrics and cut-offs for platelet reactivity. Qualitatively, studies were consistent in demonstrating an association between platelet reactivity and the outcomes of interest. | Study-level findings were generally imprecise | Suspected (selective outcome reporting) | Studies used heterogeneous methods to define increased reactivity | Low |
|  |  |  | Cardiovascular mortality | Intermediate | Direct | It was not possible to evaluate consistency with respect to the effect size of the association because studies used different metrics and cut-offs for platelet reactivity. Qualitatively, studies were consistent in demonstrating an association between platelet reactivity and the outcomes of interest. | Study-level findings were generally imprecise | Suspected (selective outcome reporting) | Studies used heterogeneous methods to define increased reactivity. | Low |
|  |  |  | Acute coronary syndromes | Intermediate | Direct | It was not possible to evaluate consistency with respect to the effect size of the association because studies used different metrics and cut-offs for platelet reactivity. Qualitatively, studies were consistent in demonstrating an association between platelet reactivity and the outcomes of interest. | Study-level findings were generally imprecise | Suspected (selective outcome reporting) | Studies used heterogeneous methods to define increased reactivity | Low |
|  |  |  | Stent thrombosis | Intermediate | Direct | It was not possible to evaluate consistency with respect to the effect size of the association because studies used different metrics and cut-offs for platelet reactivity. Qualitatively, studies were consistent in demonstrating an association between platelet reactivity and the outcomes of interest. | Study-level findings were generally imprecise | Suspected (selective outcome reporting) | Studies used heterogeneous methods to define increased reactivity | Low |
|  |  |  | Stroke | Intermediate | Direct | It was not possible to evaluate consistency with respect to the effect size of the association because studies used different metrics and cut-offs for platelet reactivity. Qualitatively, studies were consistent in demonstrating an association between platelet reactivity and the outcomes of interest. | Study-level findings were generally imprecise | Suspected (selective outcome reporting) | Studies used heterogeneous methods to define increased reactivity | Low  (for lack of association) |
|  |  |  | MACE | Intermediate | Direct | It was not possible to evaluate consistency with respect to the effect size of the association because studies used different metrics and cut-offs for platelet reactivity. Qualitatively, studies were consistent in demonstrating an association between platelet reactivity and the outcomes of interest. | Study-level findings were generally imprecise | Suspected (selective outcome reporting) | Studies used heterogeneous methods to define increased reactivity | Low |
|  |  |  | All other clinical outcomes | Intermediate | Direct | It was not possible to evaluate consistency with respect to the effect size of the association because studies used different metrics and cut-offs for platelet reactivity. Qualitatively, studies were consistent in demonstrating an association between platelet reactivity and the outcomes of interest. | Study-level findings were generally imprecise | Suspected (selective outcome reporting) | Clinical and population heterogeneity or small number of studies limited our ability to draw conclusions. Studies used heterogeneous methods to define increased reactivity. | Insufficient |
|  |  | VerifyNow | All-cause mortality | Intermediate | Direct | Consistent | Somewhat imprecise | Suspected (selective outcome reporting) | None | Low  (for lack of association) |
|  |  |  | Cardiovascular mortality | Intermediate | Direct | Consistent | Imprecise | Suspected (selective outcome reporting) | None | Moderate |
|  |  |  | Acute coronary syndromes | Intermediate | Direct | Qualitatively consistent | Imprecise | Suspected (selective outcome reporting) | None | Low |
|  |  |  | Stent thrombosis | Intermediate | Direct | Consistent | Imprecise | Suspected (publication bias and selective outcome reporting) | None | Low  (for lack of association) |
|  |  |  | MACE | Intermediate | Direct | Somewhat inconsistent with regards to the magnitude of the effect size, but consistent with regards to the direction of effects | Imprecise | Suspected (publication bias and selective outcome reporting) | None | Moderate |
|  |  |  | Bleeding events (major and all levels of severity combined) | Intermediate | Direct | Inconsistent for major events; consistent for all events | Imprecise for major events; precise for all events | Suspected (mainly selective outcome reporting) | None | Low  (for lack of association) |
|  |  |  | All other clinical outcomes | Intermediate | Direct | Few studies were available for each outcome of interest; results were somewhat inconsistent (when 2 or more studies were available) | Imprecise | Suspected (mainly selective outcome reporting) | Clinical heterogeneity and small number of studies limited our ability to draw conclusions | Insufficient |
|  |  | VASP assay | Cardiovascular mortality | Intermediate | Direct | Somewhat inconsistent (estimates from individual studies indicated different directions of effect) | Imprecise | Suspected (mainly selective outcome reporting) | None | Insufficient |
|  |  |  | Acute coronary syndromes | Intermediate | Direct | Consistent | Imprecise | Suspected (mainly selective outcome reporting) | None | Low  (for lack of association) |
|  |  |  | Stent thrombosis | Intermediate | Direct | Consistent | Imprecise | Suspected (mainly selective outcome reporting) | None | Low |
|  |  |  | MACE | Intermediate | Direct | Inconsistent | Somewhat imprecise; but the lower bound of the confidence interval of the summary effect indicated a 21% increase in risk | Suspected (mainly selective outcome reporting) | None | Low |
|  |  |  | All other clinical outcomes | Intermediate | Direct | Few studies were available for each outcome of interest; results were somewhat inconsistent (when ≥2 studies were available) | Imprecise | Suspected (mainly selective outcome reporting) | Few studies reported information. Clinical heterogeneity or small number of studies limited our ability to draw conclusions. | Insufficient |
|  |  | PFA-100 | MACE | Intermediate | Direct | Qualitatively consistent | Imprecise | Not suspected | Heterogeneity in the methods used to define increased reactivity precluded definitive conclusions. Studies generally indicated an association between increase platelet reactivity and composite clinical outcomes. | Low |
|  |  |  | All other clinical outcomes | Intermediate | Direct | Few studies were available for other outcomes; results were somewhat inconsistent when ≥2 studies were available for the same outcome | Imprecise | Suspected (mainly selective outcome reporting) | Few of the available studies reported information on other outcomes | Insufficient |
|  |  | All other assays | All clinical outcomes | Intermediate | Direct | Qualitatively inconsistent | Imprecise | Suspected (mainly selective outcome reporting) | Few studies were available for each assay; when ≥2 studies were available for the same outcome they used heterogeneous metrics or thresholds to define increased reactivity or used different agonists for ex vivo stimulation of platelets | Insufficient |
|  | Other patient groups who are candidates for clopidogrel therapy | All assays used to measure on-clopidogrel platelet reactivity | All clinical outcomes | Intermediate | Direct | NA (single study available for each population) | Imprecise | Suspected (mainly selective outcome reporting) | 6 studies, using diverse assays to measure reactivity, were available in clinically heterogeneous populations.  Studies were fairly small. | Insufficient |
| **2c: What factors affect the predictive value of phenotypic testing for platelet reactivity?** | All patient populations | All assays used to measure on-clopidogrel platelet reactivity | All clinical outcomes | Intermediate | Direct | Inconsistent (for factors evaluated by ≥2 studies) | Imprecise | Suspected (mainly selective outcome reporting) | 7 studies provided information on effect modification; no factor was assessed by more than 3 studies. Effect modification by study-level factors could not be assessed for most assays–outcome pairs; when such analysis was possible (for VerifyNow MACE), results indicated substantial uncertainty. | Insufficient |
| **3a: What is the comparative effectiveness of alternative test-and-treat strategies** | Ischemic heart disease | Genetic testing for CYP2C19 variants or phenotypic testing for platelet reactivity (all assays assessed) | All clinical outcomes | Low- Intermediate | Direct | NA (single study was available for each population/treatment strategy of interest) | Imprecise | Not suspected | Repurposed RCTs reported on non-random subsets of the populations included in the parent trials and were not specifically designed to assess effect modification; a single well-conducted RCT directly comparing test-based vs. non-test-based treatment had short followup and did not report any major clinical events. | Insufficient |
|  |  | Phenotypic testing for platelet reactivity | All clinical outcomes | Intermediate -High | Direct | Generally qualitatively consistent (RCTs produced consistent results between them; the NRCS produced inconsistent results with those of RCTs for cardiovascular mortality) | Imprecise | Suspected (mainly selective outcome reporting) | None | Insufficient |
|  | Atrial fibrillation | Genetic testing for CYP2C19 variants or phenotypic testing for platelet reactivity (all assays assessed) | All clinical outcomes | Intermediate | Direct | NA (single study) | Imprecise | Not suspected | 1 study providing information on effect modification by CYP2C19 status was identified | Insufficient |
|  |  | Phenotypic testing for platelet reactivity | All clinical outcomes | NA | NA | NA | NA | NA | No studies were identified | Insufficient |
|  | Other patient populations | Genetic testing for CYP2C19 variants or phenotypic testing for platelet reactivity (all assays assessed) | All clinical outcomes | Intermediate | Direct | NA (single study) | Imprecise | Not suspected | 1 study provided information on treatment effect modification in a mixed population of patients with atherothrombotic disease (cardiovascular, cerebrovascular, or peripheral arterial) along with asymptomatic individuals at risk for atherothrombotic disease | Insufficient |
|  |  | Phenotypic testing for platelet reactivity | All clinical outcomes | NA | NA | NA | NA | NA | No studies were identified | Insufficient |
| **3b: What factors modify the comparative effectiveness of alternative test-and-treat strategies?** | All patient populations | Genetic testing for CYP2C19 variants or phenotypic testing for platelet reactivity (all assays assessed) | All clinical outcomes | Intermediate | Direct | NA (each study assessed different effect modifiers) | Imprecise | Suspected (mainly selective outcome reporting) | None | Insufficient |
| **4: What are the harms of testing? What are the harms of test-directed treatment?** | All patient populations | Genetic testing for CYP2C19 variants | All clinical outcomes | Low- Intermediate (for the harms of test-directed treatment) | Direct (for the harms of test-directed treatment) | A single study was available for each population/treatment strategy of interest | Imprecise | Not suspected | No studies provided direct information on the harms of testing per se | Insufficient, both for harms of test-directed treatment and for harms of testing per se |
|  |  | Phenotypic testing for platelet reactivity (all assays assessed) | All clinical outcomes | Intermediate -High | Indirect | NA (single study) | Imprecise | Suspected (mainly selective outcome reporting) | 1 study reported delays in PCI due to repeat testing. No studies provided direct information on the harms of testing per se. | Insufficient, both for harms of test-directed treatment and for harms of testing per se |

CI = confidence interval; GOF = gain-of-function; LOF = loss-of-function; MACE = major adverse cardiovascular events; NA = not applicable; PCI = percutaneous coronary intervention; RR = relative risk; SOE = strength of evidence.