Assay method findings for post-infusion monitoring of Jivi[1]

Assay method	Activator	aPTT reagent	Key findings
Chromogenic	All	All	Accurate results obtained for all reagents Low interlaboratory variability for all reagents Effective in measuring Jivi FVIII activity
One Stage	Ellagic acid	Actin® FSL, Siemens	Accurate results obtained Effective in measuring Jivi FVIII activity
		Actin® FS, Siemens	Overestimated Jivi FVIII activity, especially at lower concentrations
		SynthAFax, Instrumentation Laboratory	Accurate results obtained Effective in measuring Jivi FVIII activity
	Colloidal silica	SynthASil, Instrumentation Laboratory	Accurate results obtained Effective in measuring Jivi FVIII activity
	Silica	Pathromtin®, Siemens	Accurate results obtained Effective in measuring Jivi FVIII activity
		APTT-SP, Instrumentation Laboratory	Underestimated Jivi FVIII activity by <18% at all concentrations Interlaboratory variability ≥75%
		STA®-PTT-A, Stago	Underestimated Jivi FVIII activity by <25% at all concentrations Interlaboratory variability ≥75%
	Kaolin	C.K. Prest®, Stago	Increased variability versus other one-stage agents Jivi FVIII activity overestimated, especially at lower concentrations

Key findings

FVIII activity following Jivi administration can be accurately measured with most common ellagic acid- and silica-based one-stage and chromogenic reagents. However, the silica-based assays, APTT-SP and PTT-A, underestimated results and should be avoided when measuring plasma samples that contain Jivi.[1]

If you use a reagent that was not included in this field test analysis, it is recommended that you evaluate your samples in advance

aPTT, activated partial thromboplastin time; rAHF-PFM, antihemophilic factor (recombinant) plasma/albumin-free method.

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FIELD STUDY: A PUBLICATION SUMMARY

Factor VIII activity of Jivi® is accurately measured with most commonly used assays: Results from an international laboratory study

1. Church N *et al*. Haemophilia 2018; 24(5): 823–832.

THE STUDY

Objective
To provide conclusions on expected assay performance, and guidance on effective PEGylated extended half-life FVIII measurement, by assessing the accuracy of common FVIII assays (predominantly one-stage) in the field[1]

Methods
52 clinical laboratories were given blinded FVIII-deficient plasma samples spiked with three pre-defined levels of Jivi and recombinant FVIII comparator, to assess the accuracy of their routinely used one-stage assays and when available, chromogenic assays[1]

Results
Two silica-based reagents, APTT-SP and PTT-A, underestimated Jivi levels at all concentrations. Otherwise, most one-stage and chromogenic assays accurately estimated FVIII levels. See back cover for results summary[1]

Conclusion
Clinical laboratories should identify and avoid specific inappropriate one-stage FVIII assay reagents, such as APTT-SP and PTT-A[1]

Background

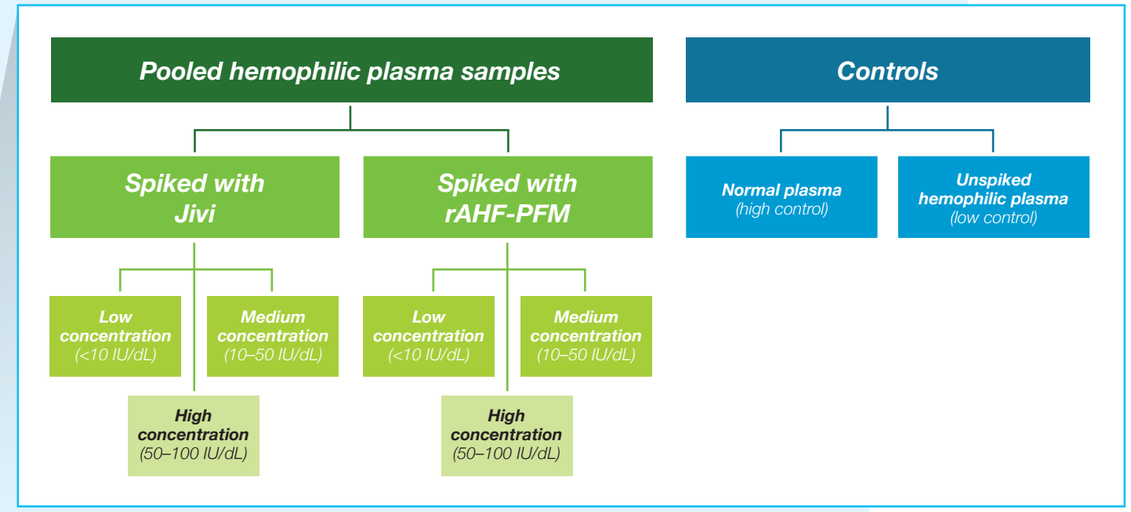
- Jivi is a site-specifically PEGylated, extended half-life rFVIII[1]
- FVIII activity can be measured with one-stage or chromogenic assays[1]
- Discrepancies in results using different assays or reagents have been recognized[1]

Study methods

52 clinical laboratories analyzed blinded plasma samples spiked with different pre-defined levels of Jivi and rAHF-PFM, and control plasma samples (see study design below). Laboratories were based across North America (n=25), Europe (n=26), and Israel (n=1). The study consisted of two parts, each assessing:[1]

- **Part 1 - In-house assays**
  36 laboratories were included and used their standard procedures, reagents and standards (calibrators) to measure activity of Jivi and rAHF-PFM in the samples with one-stage assays; 13 laboratories used both one-stage and chromogenic assays and 3 laboratories used only chromogenic assays[1]
- **Part 2 - Effect of giving guidance to laboratories on one-stage assay reagents**
  Laboratories were given Pathromtin® (in 51 laboratories) and SynthASil (in 52 laboratories), both aPTT silica-based one-stage assay reagents, and asked to follow each manufacturer's instructions[1]

Design[1]*



* Low, medium, and high concentrations were 4.3, 37.5, and 86.5 IU/dL, respectively; the high control (normal plasma) contained 87.8 IU/dL FVIII.

Endpoints[1]

- **Primary:**
  FVIII spiked recovery (defined as 100x measured FVIII level/nominal FVIII level)†
- **Secondary:**
  Measured FVIII levels

Results

- **Chromogenic assays: Accurate FVIII activity results were obtained for Jivi[1]**
  - Mean FVIII recovery for Jivi was similar to that of rAHF-PFM, and interlaboratory variability was low for all three chromogenic assay reagents used[1]
- **One-stage assays: Accurate FVIII activity results were achieved following Jivi administration with most, but not all, reagents[1]**
  - Accurate results were obtained with the ellagic acid-based reagent Actin® FSL, as well as silica-based reagents SynthASil and Pathromtin®[1]
  - Increased variability was seen with the kaolin-based reagent C.K. Prest® vs. other one-stage reagents[1]
    - Actin® FS was found to overestimate FVIII recovery, particularly at low concentrations[1]
    - Two silica-based reagents – APTT-SP and PTT-A – were found to underestimate FVIII recovery (<18% and <25 %, respectively)[1]
      - The one-stage assay interlaboratory variability for recoveries measured with these two reagents was ≥75%, compared with other one-stage reagents that generally was <25%[1]
      - **These results indicate that APTT-SP and PTT-A are likely to contribute to overall interlaboratory variability*, demonstrating the importance of using appropriate one-stage assay reagents when measuring Jivi activity**[1]

It is important that clinical laboratories test the accuracy of their local procedures before measuring FVIII in patients treated with Jivi[1]

† Accuracy was assessed in terms of spiked recoveries falling within the acceptance criteria (geometric mean of 80–125% of the target value).