BC von Willebrand Reagent

BC VWF REAGENT

Revision bar indicates update to previous version.

CE 0197

For the determination of ristocetin cofactor activity

Intended Use

BC VWF REAGENT is an in vitro diagnostic reagent for the quantitative, WHO-standardized determination of ristocetin cofactor activity of von Willebrand factor as aid to diagnosis and monitoring of congenital or acquired vWF deficiencies in patients with bleeding disorders or at risk for vWF deficiency in human sodium citrated plasma by means of automated turbidimetric methods.

Summary and Explanation

Von Willebrand Disease (VWD) is the most frequent congenital human bleeding disorder and is caused by a reduction or dysfunction of von Willebrand factor (vWF). vWF is a multimeric, high-molecular glycoprotein involved in primary hemostasis, supporting platelet adhesion and aggregation via binding to the platelet glycoprotein Ib (GPIb) receptor under shear stress at the site of injury. Furthermore, vWF is the specific carrier protein of coagulation factor VIII (FVIII), protecting FVIII against inactivation and rapid clearance.

VWD is an autosomal inherited disorder of three different types: type 1 (partial quantitative deficiency of vWF), type 2 (qualitative vWF defects) and type 3 (almost complete deficiency of vWF). VWD type 2 can be further sub-typed into 2A, 2B, and 2M based on their different multimeric pattern and/or altered platelet affinity. Type 2N is characterized by a reduced FVIII-binding activity with otherwise normal functionality. These variants are differentiated on the basis of a series of laboratory tests for vWF activity (e.g. GPIb-binding or ristocetin cofactor assay, collagen binding assay), vWF antigen, platelet function testing (PFA Systems), FVIII activity, platelet count, and vWF multimer analysis[1–6].

Similar clinical and laboratory findings as with congenital VWD may be seen in acquired VWS, a rare but probably underestimated bleeding disorder[7,8]. In patients with aortic stenosis the high molecular multimers are reduced to shear stress, causing an acquired type 2A vWF deficiency, which predisposes patients to bleeding. Such defects also develop with left ventricular assist devices, valve dysfunction, cardiomyopathy or congenital heart defects[9].

The determination of vWF activity in plasma is indicated in the following cases:

- clarifying the cause of a prolonged APTT,
- diagnosing congenital or acquired vWF deficiency states,
- monitoring vWF in patients under therapy (e.g. Desmopressin or vWF concentrates)
- distinguishing between functional defects and quantitative deficiencies (in conjunction with immunochemical methods).

On the other hand, elevated levels of vWF have been reported in response to stress, inflammation and endothelial lesions, and have been related to thrombotic complications such as venous thromboembolism and myocardial infarction[10]. Such increases of vWF levels may obscure the laboratory diagnosis of mild VWD.

Principles of the Procedure

BC VWF REAGENT measures ristocetin cofactor activity as follows: in the presence of ristocetin, the von Willebrand factor (ristocetin cofactor) in the sample causes an agglutination of the stabilized platelets contained in the von Willebrand Reagent. The agglutination process reduces the turbidity at the onset of the reaction. A coagulation analyzer measures the change in optical density and automatically determines the ristocetin cofactor activity of the sample in % of Norm.

Reagents

Note: BC VWF REAGENT can be used on automated coagulation analyzers. Sysmex provides Reference Guides (Application Sheets) for several coagulation analyzers. The Reference Guides (Application Sheets) contain analyzer/assay specific handling and performance information which may differ from that provided in these Instructions for Use. In this case, the information contained in the Reference Guides (Application Sheets) supersedes the information in these Instructions for Use. Please also consult the instruction manual of the instrument manufacturer!

Reagent	Description	Storage	Stability
BC von Willebrand Reagent BC VWF REAGENT	Lyophilized reagent containing: • platelets (reconstituted: ~0.018 g/L) • Ristocetin (reconstituted: ~0.5 mmol/L) • EDTA • Stabilizer • Preservative: • Sodium azide (reconstituted: ≤0.25 g/L)	2–8 °C May be used up to the expiry date indicated on the label if stored unopened.	2–8 °C: reconstituted, 2 days; 15 °C[a]: reconstituted, 8 hours

[a] open vial

On-board stability
Information regarding on-board stability is specified in the Reference Guides (Application Sheets) for the different coagulation analyzers.

Warnings and Precautions

For *in-vitro* diagnostic use only.

For laboratory professional use.

According to EU regulation 2017/746, any serious incident that has occurred in relation to the device shall be reported to the manufacturer and the competent authority of the EU Member State through your local distribution representative in which the user and/or patient is established.

Safety data sheets (MSDS/SDS) available upon request.

BC VWF REAGENT
Hazardous ingredient: Sodium azide (0.896 % [w/w]).
H412: Harmful to aquatic life with long lasting effects.
P273: Avoid release to the environment. **P501**: Dispose of contents and container in accordance with all local, regional, and national regulations.

CAUTION! POTENTIAL BIOHAZARD
Each donor or donor unit was tested and found to be negative for human immunodeficiency virus (HIV) 1 and 2, hepatitis B virus (HBV) and hepatitis C virus (HCV) using either tests that are CE marked or FDA approved for this purpose. Because no known test can offer complete assurance of the absence of infectious agents, all human derived products should be handled with appropriate caution.

Contains sodium azide as a preservative. Sodium azide can react with copper or lead plumbing to form explosive metal azides. On disposal, flush reagents with a large volume of water to prevent the buildup of azides. Disposal into drain systems must be in compliance with prevailing regulatory requirements.

Dispose of hazardous or biologically contaminated materials according to the practices of your institution. Discard all materials in a safe and acceptable manner and in compliance with all government requirements.

Summary of Safety and Performance (SSP) is available in the European database on medical devices (see Eudamed public website: https://ec.europa.eu/tools/eudamed). In case Eudamed is not available, SSP can be delivered by the manufacturer on request.

Preparing Reagents

Resuspend the reagent with the amount of distilled water indicated on the label at 15 to 25 °C by mixing (e.g. with an automatic mixer like the Heidolph Mixer "Reax control" set in the range of 75 to 100 %, 2 × 5 seconds). The reagent is then immediately ready to use. Be sure to mix the reagent intensively and to an equal extent for each new reconstitution.

Each time before use (i.e. placement on the instrument) the reagent has to be mixed intensively equal to the initial procedure.

Specimen Collection and Handling

Collecting the Specimen

To obtain the plasma, carefully mix 1 part sodium citrate solution (0.11 mol/L, 3.2 %) with 9 parts venous blood, avoiding the formation of foam. Centrifuge the blood specimen immediately at 1500 × g for no less than 15 minutes at room temperature, then remove the supernatant plasma.

Storing the Specimen

Store at 15 to 25 °C or deep frozen.

Stability of the samples:

at 15 to 25 °C	6 hours
at ≤ −20 °C	1 month

Procedure

Materials Provided

REF	Contents	
OUBD37	BC von Willebrand Reagent BC VWF REAGENT	5 × → 4 mL

For coagulation analyzers without a stirring position, make sure that the reagent is resuspended by mixing at least every 30 minutes.

A new reference curve must be calculated each time there is a change in the device used or a change in the lot of BC VWF REAGENT.

Materials Required but not Provided

Item	Description
REF ORKE41	CONTROL N, Control Plasma N
REF OUPZ17	CONTROL P, Control Plasma P
REF ORKL17	STANDARD PLASMA, Standard Human Plasma
Coagulation analyzers[b], such as:	• AUTOMATED BLOOD COAGULATION ANALYZER CS-2500 (CS-2500 System) • AUTOMATED BLOOD COAGULATION ANALYZER CS-5100 (CS-5100 System)

[b] Availability of analyzers may vary by country.

Please note that the applications on other analyzers can be validated by the instrument manufacturer in accordance with the requirements of the REGULATION (EU) 2017/746 under their responsibility as long as the intended purpose and performance are not modified.

Internal Quality Control

Normal range: CONTROL N

Pathological range: CONTROL P

Two controls should be measured with each reagent filling, each calibration and at least every 8 hours during each day of testing (one in the normal range and one in the pathological range). The controls should be treated like the samples. Measurements are to be carried out as duplicates. The results obtained must lie within the range indicated for the controls in the lot-specific table of assigned values. If a measured control value lies outside the allowable range, the coagulation analyzer, reagent and reference curve should be checked. Do not release patient results until the cause of deviation has been identified and corrected.

Results

The ristocetin cofactor activity is indicated in % of Norm.

Limitations

The manufacturer has validated use of these reagents on various analyzers to optimize product performance and meet product specifications. Please note that the applications on other analyzers can be validated by the instrument manufacturer in accordance with the requirements of the REGULATION (EU) 2017/746 under their responsibility as long as the intended purpose and performance are not modified. User defined modifications are not supported by the manufacturer as they may affect performance of the system and assay results. It is the responsibility of the user to validate modifications to these instructions or use of the reagents on analyzers other than those included in Application Sheets or these Instructions for Use.

It has been reported that two sequence variants in the VWF gene can cause artificial low VWF:RCo activity values[11].

Results of this test should always be interpreted in conjunction with the patient's medical history, clinical presentation and other findings.

Expected Values

Reference intervals vary from laboratory to laboratory depending on the population served and the technique, method, equipment and reagent lot used. Therefore, each laboratory must establish its own reference intervals or verify them whenever one or more of the aforementioned variables are changed. (For additional information on determining the reference interval, see CLSI Document EP28 A3c[12].)

Results below the determined reference interval can be considered an indication of von Willebrand Syndrome.

In a study carried out on ostensibly healthy subjects (n = 185) using the BCS® System, the following data were obtained (5^{th} to 95^{th} percentile): **58 to 172 % to , Median: 99 %** (blood group 0: 49 to 142 %, Median: 79 %; blood group non-0: 66 to 183 %, Median: 121 %).

There is a connection between blood group (AB0 type), age and von Willebrand factor concentration[13].

Performance Characteristics

Measuring Range

The measuring range depends on the individual application of the assay due to instrument related conditions. Application specific performance data is listed in the respective Reference Guides of the instruments.

Precision

Over a five day period, precision studies (one run per day in replicates of eight) were performed by assaying normal and pathological control plasmas. For normal control plasmas (n = 80), the within-run coefficient of variation (CV) ranged from 8.0 to 9.6 %, while the total CV ranged from 8.0 to 10.3 %. For pathological control plasma (n = 80), the within-run CV ranged from 6.1 to 16.2 %, while the total CV ranged from 7.6 to 16.9 %.

Other system specific results are given in the respective Reference Guides (Application Sheets).

The reproducibility was assessed by the manufacturer for VWF Ristocetin Cofactor activity with BC VWF REAGENT based on publicly available proficiency testing information in 2019/2020. The overall reproducibility median CV% was found to be <22 % including lot, instrument, laboratory and operator variability factors.

Method Comparison

A comparative study was carried out with BC VWF REAGENT and another commercially available reagent. A total of 70 patient plasma samples (35 normal and 35 pathological) were tested using both methods. The regression analysis produced a correlation coefficient of 0.94, a y-axis intercept of −4.4 % and a slope of 0.97.

Technical Assistance

For customer support, contact your local technical support provider or distributor.

Current Version of Application Sheets

BC VWF REAGENT can be used in combination with various automated coagulation analyzers. Sysmex provides Reference Guides/Application Sheets for the coagulation analyzers listed in section "Materials Required but not Provided", page 3 under the dedicated link below:

sysmex-ifu.com/ag

As the manufacturer continuously monitors the product performance and safety, the users are required to ensure that they work with the correct revision of the instructions for the product lots in use. Please periodically review the availability of new electronic labeling revisions to ensure safe use of the product.

The IFU version number is visible on each product box label. Sysmex ensures that all products lots bearing the same IFU version number are compatible with the electronic labeling provided via sysmex-ifu.com.

References

1. Keesler DA, Flood VH. Current issues in diagnosis and treatment of von Willebrand disease. Res Pract Thromb Haemost. 2018;1-8. 2017.
2. Sharma R, Flood VH. Advances in the diagnosis and treatment of Von Willebrand disease. Blood 2017; 130:2386-2391.
3. De Jong A, Eikenboom J. Developments in the diagnostic procedures for von Willebrand disease. J Thromb Haemost 2016; 14:449-60.
4. Boender J, Kruip MJ, Leebeek FW. A diagnostic approach to mild bleeding disorders. J Thromb Haemost 2016; 14:1507-16.
5. Laffan MA, Lester W, O'Donnell JS, et al. The diagnosis and management of von Willebrand disease: a United Kingdom Haemophilia Centre Doctors Organization guideline approved by the British Committee for Standards in Haematology. Br J Haematol 2014;167:453-65.
6. Lawrie AS, Mackie IJ, Machin SJ, et al.Evaluation of an automated platelet-based assay of ristocetin cofactor activity. Haemophilia. 2011;17:252-6.
7. Mital A. Acquired von Willebrand Syndrome. Adv Clin Exp Med 2016; 25:1337-44.
8. Geisen U, Zieger B, Nakamura L, et al. Comparison of Von Willebrand factor (VWF) activity VWF:Ac with VWF ristocetin cofactor activity VWF:RCo. Thromb Res 2014; 134:246-50.
9. Yasar SJ, Abdullah O, Fay W, et al. Von Willebrand factor revisited. J Interv Cardiol 2018; 31:360-367.
10. Gragnano F, Sperlongano S, Golia E, et al.The Role of von Willebrand Factor in Vascular Inflammation: From Pathogenesis to Targeted Therapy. Mediators Inflamm 2017; 2017:5620314.
11. Bodo I, Eikenboom J, Montgomery R, Patzke J, Schneppenheim R, Di Paola J. Platelet-dependent von Willebrand factor activity. Nomenclature and methodology: communication from the SSC of the ISTH. J Thromb Haemost 2015; 13: 1345-50.
12. CLSI. Defining, Establishing, and Verifying Reference Intervals in the Clinical Laboratory, Approved Guidline – Third Edition. CLSI document **EP28-A3C** [ISBN 1-56238-682-4]. CLSI, 950 West Valley Road, Suite 2500, Wayne, PA 19087-1898 USA, 2010.
13. Gill JC, Endres-Brooks J, Bauer PJ et al. The effect of AB0 blood group on the diagnosis of von Willebrand disease. Blood: 1987; 69:1691-5.

Definition of Symbols

The following symbols may appear on the product labeling:

	Do not reuse		Use By
LOT	Batch Code	REF	Catalogue Number
	Caution		Manufacturer
EC REP	Authorized representative in the European Community	CH REP	Authorized representative in Switzerland
	Contains sufficient for <n> tests		Biological Risks
IVD	*In Vitro* Diagnostic Medical Device		Temperature Limitation
	Consult instruction for Use		Non-sterile
CE	CE marking of conformity	CE 0197	CE marking of conformity with notified body ID number. Notified body ID number can vary.
CONTENTS	Contents	→	Reconstitution volume
LEVEL	Level		Keep away from sunlight and heat
WARNING	Warning	DANGER	Danger
RxOnly	Prescription device (US only)	UDI	Device Identification (UDI) barcode
REACH xx/xx/xx	REACH Authorization Number		

Legal Information

Siemens Healthcare Diagnostics Products GmbH
Emil-von-Behring-Str. 76
35041 Marburg
Germany
siemens-healthineers.com