HDV Ab

Competitive Enzyme Immunoassay for the qualitative determination of antibodies to Hepatitis Delta Virus in human serum and plasma

- for "in vitro" diagnostic use only -

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REF DAB.CE
96 Tests

HDV Ab

A. INTENDED USE

Competitive Enzyme ImmunoAssay (ELISA) for the qualitative determination of antibodies to Hepatitis Delta Virus or HDV in human plasma and sera with a "two-steps" methodology.
The kit is used for the follow-up of patients infected by HDV.
For "in vitro" diagnostic use only.

B. INTRODUCTION

The Hepatitis Delta Virus or HDV is a RNA defective virus composed of a core presenting the delta-specific antigen, encapsulated by HBsAg, that requires the helper function of HBV to support its replication.
Infection by HDV occurs in the presence of acute or chronic HBV infection. When acute delta and acute HBV simultaneously occur, the illness becomes severe and clinical and biochemical features may be indistinguishable from those of HBV infection alone. In contrast, a patient with chronic HBV infection can support HDV replication indefinitely, usually with a less severe illness appearing as a clinical exacerbation.
The determination of HDV specific serological markers (HDV Ag, HDV Ab, HDV IgM and HDV IgG) represents in these cases an important tool to the clinician for the classification of the etiological agent, for the follow up of infected patients and their treatment. The detection of HDV total antibodies allows the classification of the illness and the monitoring of the seroconversion event.

C. PRINCIPLE OF THE TEST

Anti-HDV antibodies, if present in the sample, compete with a virus-specific polyclonal IgG, labeled with peroxidase (HRP), for a fixed amount of rec-HDV coated on the microplate. The test is carried out with a two steps incubation competitive system. First the sample is added to the plate and specific anti HDV antibodies bind to the adsorbed antigen. After washing, an enzyme conjugated antibody to HDV is added and binds to the free portion of the antigen coated. After washing a chromogen/substrate mixture is dispensed. The concentration of the bound enzyme on the solid phase becomes inversely proportional to the amount of anti-HDV antibodies in the sample and its activity is detected by the added chromogen/substrate. The concentration of HDV-specific antibodies in the sample is determined by means of a cut-off value that allows for the semi quantitative detection of anti-HDV antibodies.

D. COMPONENTS

Each kit contains sufficient reagents to perform 96 tests.

1. Microplate: MICROPLATE
8x12 microwell strips coated with recombinant HDV-specific antigen and sealed into a bag with desiccant. Allow the microplate to reach room temperature before opening; reseal unused strips in the bag with desiccant and store at 4°C.

2. Negative Control: CONTROL -
1x2.0ml/vial. Ready to use. Contains goat serum proteins, 100 mM Tris-HCl buffer pH 7.4 +/-0.1, 0.09% Sodium Azide and 0.1% Kathon GC as preservatives. The negative control is colour coded pale yellow.

3. Positive Control: CONTROL +
1x2.0ml/vial. Ready to use. Contains goat serum proteins, high titer anti HDV antibodies, 100 mM Tris-HCl buffer pH 7.4 +/-0.1, 0.09% Sodium Azide and 0.1% Kathon GC as preservatives.
The positive control is colour coded green.

4. Calibrator: CAL ....
n° 1 vial. Lyophilised. To be dissolved with EIA grade water as reported in the label. Contains bovine serum proteins, low titer human antibodies to HDV, 0.2 mg/ml gentamicine sulphate and 0.1% Kathon GC as preservatives.
Note: The volume necessary to dissolve the content of the vial may vary from lot to lot. Please use the right volume reported on the label.

5. Wash buffer concentrate: WASHBUF 20X
1x60ml/bottle. 20x concentrated solution.
Once diluted, the wash solution contains 10 mM phosphate buffer pH 7.0+/-0.2, 0.05% Tween 20 and 0.1% Kathon GC.

6. Enzyme conjugate: CONJ
1x16ml/vial. Ready-to-use solution. Contains 5% bovine serum albumine, 10 mM tris buffer pH 6.8 +/-0.1, Horseradish peroxidase conjugated antibody to HDV in presence of 0.2 mg/ml gentamicine sulphate and 0.1% Kathon GC as preservatives. The component is colour coded red.

7. Chromogen/Substrate: SUBS TMB
1x16ml/vial. Contains a 50 mM citrate-phosphate buffered solution at pH 3.5-3.8, 4% DMSO, 0.03% tetra-methyl-benzidine or TMB and 0.02% hydrogen peroxide of H_2O_2.
Note: To be stored protected from light as sensitive to strong illumination.

8. Sulphuric Acid: H_2SO_4 0.3 M
1x15ml/vial. Contains 0.3 M H_2SO_4 solution.
Attention: Irritant (H315, H319; P280, P302+P352, P332+P313, P305+P351+P338, P337+P313, P362+P363).

Plate sealers n° 2

Instructions for Use n° 1

E. MATERIALS REQUIRED BUT NOT PROVIDED

1. Calibrated Micropipettes in the range 10-1000 ul and disposable plastic tips.
2. EIA grade water (double distilled or deionized, charcoal treated to remove oxidizing chemicals used as disinfectants).
3. Timer with 60 minute range or higher.
4. Absorbent paper tissues.
5. Calibrated ELISA microplate thermostatic incubator (dry or wet) set at +37°C.
6. Calibrated ELISA microwell reader with 450nm (reading) and with 620-630nm (blanking) filters.
7. Calibrated ELISA microplate washer.
8. Vortex or similar mixing tools.

F. WARNINGS AND PRECAUTIONS

1. The kit has to be used by skilled and properly trained technical personnel only, under the supervision of a medical doctor responsible of the laboratory.
2. All the personnel involved in performing the assay have to wear protective laboratory clothes, talc-free gloves and glasses. The use of any sharp (needles) or cutting (blades) devices should be avoided. All the personnel involved should be trained in biosafety procedures, as recommended by the Center for Disease Control, Atlanta, U.S. and reported in the National Institute of Health's publication: "Biosafety in Microbiological and Biomedical Laboratories", ed. 1984.
3. All the personnel involved in sample handling should be vaccinated for HBV and HAV, for which vaccines are available, safe and effective.

4. The laboratory environment should be controlled so as to avoid contaminants such as dust or air-born microbial agents, when opening kit vials and microplates and when performing the test. Protect the Chromogen/Substrate (TMB/H_2O_2) from strong light and avoid vibration of the bench surface where the test is undertaken.
5. Upon receipt, store the kit at +2..8°C into a temperature controlled refrigerator or cold room.
6. Do not interchange components between different lots of the kits. It is recommended that components between two kits of the same lot should not be interchanged.
7. Check that the reagents are clear and do not contain visible heavy particles or aggregates. If not, advise the laboratory supervisor to initiate the necessary procedures.
8. Avoid cross-contamination between serum/plasma samples by using disposable tips and changing them after each sample.
9. Avoid cross-contamination between kit reagents by using disposable tips and changing them between the use of each one.
10. Do not use the kit after the expiration date stated on external (primary container) and internal (vials) labels.
11. Treat all specimens as potentially infective. All human serum specimens should be handled at Biosafety Level 2, as recommended by the Center for Disease Control, Atlanta, U.S. in compliance with what reported in the Institutes of Health's publication "Biosafety in Microbiological and Biomedical Laboratories", ed. 1984.
12. The use of disposable plastic labware is recommended in the preparation of the washing solution or in transferring components into other containers of automated workstations, in order to avoid contamination.
13. Waste produced during the use of the kit has to be discarded in compliance with national directives and laws concerning laboratory waste of chemical and biological substances. In particular, liquid waste generated from the washing procedure, from residuals of controls and from samples has to be treated as potentially infective material and inactivated. Suggested procedures of inactivation are treatment with a 10% final concentration of household bleach for 16-18 hrs or heat inactivation by autoclave at 121°C for 20 min.
14. Accidental spills have to be adsorbed with paper tissues soaked with household bleach and then with water. Tissues should then be discarded in proper containers designated for laboratory/hospital waste.
15. The Sulphuric Acid is an irritant. In case of spills, wash the surface with plenty of water.
16. Other waste materials generated from the use of the kit (example: tips used for samples and controls, used microplates) should be handled as potentially infective and disposed according to national directives and laws concerning laboratory wastes.

G. SPECIMEN: PREPARATION AND RECOMMANDATIONS

1. Blood is drawn aseptically by venepuncture and plasma or serum is prepared using standard techniques of preparation of samples for clinical laboratory analysis. No influence has been observed in the preparation of the sample with citrate, EDTA and heparin.
2. Avoid any addition of preservatives to samples; especially sodium azide as this chemical would affect the enzymatic activity of the conjugate.
3. Samples have to be clearly identified with codes or names in order to avoid misinterpretation of results. When the kit is used for the screening of blood units, bar code labeling and electronic reading is strongly recommended.
4. Haemolysed (red) and visibly hyperlipemic ("milky") samples have to be discarded as they could generate false results. Samples containing residues of fibrin or heavy particles or microbial filaments and bodies should be discarded as they could give rise to false results.
5. Sera and plasma can be stored at +2°..8°C for up to five days after collection. For longer storage periods, samples can be stored frozen at –20°C for several months. Any frozen samples should not be frozen/thawed more than once as this may generate particles that could affect the test result.
6. If particles are present, centrifuge at 2.000 rpm for 20 min or filter using 0.2-0.8u filters to clean up the sample for testing.

H. PREPARATION OF COMPONENTS AND WARNINGS

A study conducted on an opened kit has not pointed out any relevant loss of activity up to 6 re-uses of the device and up to 3 months.

1. Antigen coated microwells:
Allow the microplate to reach room temperature (about 1 hr) before opening the container. Check that the desiccant has not turned dark green, indicating a defect in manufacturing. In this case, call Dia.Pro's customer service.
Unused strips have to be placed back into the aluminium pouch, with the desiccant supplied, firmly zipped and stored at +2°-8°C. When opened the first time, unused strips are stable until the humidity indicator inside the desiccant bag turns from yellow to green.

2. Negative Control:
Ready to use. Mix well on vortex before use.

3. Positive Control:
Ready to use. Mix well on vortex before use.

4. Calibrator:
Low positive control. Add precisely the volume of EIA grade water, reported on its label, to the lyophilized powder; let fully dissolve and then gently mix on vortex.
Note: *The dissolved calibrator is not stable. Store it frozen in aliquots at –20°C. When thawed do not freeze again; discard it.*

5. Wash buffer concentrate:
The whole content of the 20x concentrated solution has to be diluted with EIA grade water up to 1200 ml and mixed gently end-over-end before use. During preparation avoid foaming as the presence of bubbles could impact on the efficiency of the washing cycles.
Note: *Once diluted, the wash solution is stable for 1 week at +2..8° C.*

6. Enzyme conjugate:
Ready to use. Mix well on vortex before use.
Avoid contamination of the liquid with oxidizing chemicals, dust or microbes. If this component has to be transferred, use only plastic, and if possible, sterile disposable containers.

7. Chromogen/Substrate:
Ready to use. Mix well on vortex before use.
Avoid contamination of the liquid with oxidizing chemicals, air-driven dust or microbes. Do not expose to strong light, oxidizing agents and metallic surfaces.
If this component has to be transferred use only plastic, and if possible, sterile disposable container

8. Sulphuric Acid:
Ready to use. Mix well on vortex before use.
Attention! Irritant (H315, H319; P280, P302+P352, P332+P313, P305+P351+P338, P337+P313, P362+P363).

Legenda:
Warning H statements:
H315 – Causes skin irritation.
H319 – Causes serious eye irritation.

Precautionary P statements:
P280 – Wear protective gloves/protective clothing/eye protection/face protection.
P302 + P352 – IF ON SKIN: Wash with plenty of soap and water.
P332 + P313 – If skin irritation occurs: Get medical advice/attention.
P305 + P351 + P338 – IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.
P337 + P313 – If eye irritation persists: Get medical advice/attention.
P362 + P363 – Take off contaminated clothing and wash it before reuse.

I. INSTRUMENTS AND TOOLS USED IN COMBINATION WITH THE KIT

1. Micropipettes have to be calibrated to deliver the correct volume required by the assay and must be submitted to regular decontamination (70% ethanol, 10% solution of bleach, hospital grade disinfectants) of those parts that could accidentally come in contact with the sample or the components of the kit. They should also be regularly maintained in order to show a precision of 1% and a trueness of ±2%.
2. The ELISA incubator has to be set at +37°C (tolerance of ±0.5°C) and regularly checked to ensure the correct temperature is maintained. Both dry incubators and water baths are suitable for the incubations, provided that the instrument is validated for the incubation of ELISA tests.
3. The ELISA washer is extremely important to the overall performances of the assay. The washer must be carefully validated and correctly optimized using the kit controls/calibrator and reference panels, before using the kit for routine laboratory tests. Usually 4-5 washing cycles (aspiration + dispensation of 350ul/well of washing solution = 1 cycle) are sufficient to ensure that the assay performs as expected. A soaking time of 20-30 seconds between cycles is suggested. In order to set correctly their number, it is recommended to run an assay with the kit controls/calibrator and well characterized negative and positive reference samples, and check to match the values reported below in the sections "Validation of Test" and "Assay Performances". Regular calibration of the volumes delivered and maintenance (decontamination and cleaning of needles) of the washer has to be carried out according to the instructions of the manufacturer.
4. Incubation times have a tolerance of ±5%.
5. The ELISA microplate reader has to be equipped with a reading filter of 450nm and with a second filter (620-630nm, strongly recommended) for blanking purposes. Its standard performances should be (a) bandwidth ≤ 10 nm; (b) absorbance range from 0 to ≥ 2.0; (c) linearity to ≥ 2.0; repeatability ≥ 1%. Blanking is carried out on the well identified in the section "Assay Procedure". The optical system of the reader has to be calibrated regularly to ensure that the correct optical density is measured. It should be regularly maintained according to the manufacturer 's instructions.
6. When using an ELISA automated work station, all critical steps (dispensation, incubation, washing, reading, shaking, data handling) have to be carefully set, calibrated, controlled and regularly serviced in order to match the values reported in the sections "Validation of Test" and "Assay Performances". The assay protocol has to be installed in the operating system of the unit and validated as for the washer and the reader. In addition, the liquid handling part of the station (dispensation and washing) has to be validated and correctly set. Particular attention must be paid to avoid carry over by the needles used for dispensing samples and for washing. This must be studied and controlled to minimize the possibility of contamination of adjacent wells due to strongly reactive samples, leading to false positive results. The use of ELISA automated work stations is recommended for blood screening and when the number of samples to be tested exceed 20-30 units per run.
7. Dia.Pro's customer service offers support to the user in the setting and checking of instruments used in combination with the kit, in order to assure full compliance with the requirements described. Support is also provided for the installation of new instruments to be used with the kit.

L. PRE ASSAY CONTROLS AND OPERATIONS

1. Check the expiration date of the kit printed on the external label (primary container). Do not use if expired.
2. Check that the liquid components are not contaminated by visible particles or aggregates. Check that the Chromogen/Substrate is colorless or pale blue by aspirating a small volume of it with a sterile plastic pipette. Check that no breakage occurred in transportation and no spillage of liquid is present inside the box (primary container). Check that the aluminum pouch, containing the microplate, is not punctured or damaged.
3. Dilute all the content of the 20x concentrated Wash Solution as described above.
4. Dissolve the Calibrator as described above and gently mix.
5. Allow all the other components to reach room temperature (about 1 hr) and then mix gently on vortex all liquid reagents.
6. Set the ELISA incubator at +37°C and prepare the ELISA washer by priming with the diluted washing solution, according to the manufacturers instructions. Set the right number of washing cycles as found in the validation of the instrument for its use with the kit.
7. Check that the ELISA reader is turned on or ensure it will be turned on at least 20 minutes before reading.
8. If using an automated work station, turn on, check settings and be sure to use the right assay protocol.
9. Check that the micropipettes are set to the required volume.
10. Check that all the other equipment is available and ready to use.
11. In case of problems, do not proceed further with the test and advise the supervisor.

M. ASSAY PROCEDURE

The assay has to be carried out according to what reported below, taking care to maintain the same incubation time for all the samples in testing.

1. Place the required number of strips in the microplate holder. Leave A1 well empty for the operation of blanking.
Store the other strips into the bag in presence of the desiccant at +2..8°C, sealed.

2. Pipette 100 µl of Negative Control in triplicate, 100 µl Positive Control in single and then 100 µl of samples. Check that controls and samples have been correctly added.
Then incubate the microplate at **+37°C for 60 min.**

3. Wash the microplate as reported in section I.3.

4. In all the wells except A1, pipette 100 µl Enzyme Conjugate. Check that the reagent has been correctly added.
Then incubate the microplate at **+37°C for 60 min.**

Important note: *Be careful not to touch the inner surface of the well with the pipette tip when dispensing the Enzyme Conjugate. Contamination might occur.*

5. Wash the microplate as described.

6. Pipette 100 µl TMB/H_2O_2 mixture in each well, the blank wells included. Check that the reagent has been correctly added. Then incubate the microplate at **room temperature for 20 min**.

Important note: *Do not expose to strong direct light as a high background might be generated.*

7. Pipette 100 µl Sulphuric Acid into all the wells using the same pipetting sequence as in step n° 6 to stop the enzymatic reaction. Addition of the stop solution will turn the negative control and negative samples from blue to yellow.

8. Measure the colour intensity of the solution in each well, as described in section I.5 using a 450nm filter (reading) and a 620-630nm filter (background subtraction, strongly recommended), blanking the instrument on A1.

Important notes:

1. *If the second filter is not available, ensure that no finger prints are present on the bottom of the microwell before reading at 450nm. Finger prints could generate false positive results on reading.*
2. *Reading has should ideally be performed immediately after the addition of the Stop Solution but definitely no longer than 20 minutes afterwards. Some self oxidation of the chromogen can occur leading to a higher background.*
3. *The use of the Calibrator, a low positive control, is not mandatory for the assay as the CAL does not enter into the cut-off calculation. The CAL may be used as a low titer positive control when a laboratory internal quality verification is required by the management. When used for such purpose, dispense 100 ul of it, possibly in duplicate.*

N. ASSAY SCHEME

Controls/Calibrator	100 ul
Samples	100 ul
1st incubation	**60 min**
Temperature	+37°C
Washing step	4-5 cycles
Enzyme Conjugate	100 ul
2nd incubation	**60 min**
Temperature	+37°C
Washing step	4-5 cycles
TMB/H2O2 mix	100 ul
3rd incubation	**20 min**
Temperature	r.t.
Sulphuric Acid	100 ul
Reading OD	450nm & 620nm

An example of dispensation scheme (including CAL) is reported in the table below:

Microplate

	1	2	3	4	5	6	7	8	9	10	11	12
A	BLK	S2										
B	NC	S3										
C	NC	S4										
D	NC	S5										
E	CAL	S6										
F	CAL	S7										
G	PC	S8										
H	S1	S9										

Legenda: BLK = Blank NC = Negative Control
CAL = Calibrator PC = Positive Control S = Sample

O. INTERNAL QUALITY CONTROL

A check is performed on the negative and positive controls any time, and on the Calibrator in addition when the kit is used for the first time, in order to verify whether the expected OD450nm or Co/S values have been matched in the analysis.
Ensure that the following parameters are met:

Parameter	Requirements
Blank well	< 0.100 OD450nm value
Negative Control (NC)	> 1.000 OD450nm after blanking If lower carefully control the washing procedure and decrease the number of cycles or the soaking time coefficient of variation < 30%
Positive Control (PC)	OD450 nm < NC/10
Calibrator (CAL)	PC < OD450nm < (NC+PC)/5

If the results of the test match the requirements stated above, proceed to the next section.
If they don't, do not proceed any further and perform the following checks:

Problem	Check
Blank well > 0.100 OD450nm	that the Chromogen/Substrate solution has not become contaminated during the assay
Negative Control (NC) < 1.000 OD450nm after blanking coefficient of variation > 30%	1. that the washing procedure and the washer settings are as validated in the pre qualification study; 2. that the proper washing solution has been used and the washer has been primed with it before use; 3. that no mistake has been done in the assay procedure (dispensation of positive control instead of negative control); 4. that no contamination of the negative control or of the wells where the control was dispensed has occurred due to positive samples, to spills or to the enzyme conjugate; 5. that micropipettes have not become contaminated with positive samples or with the enzyme conjugate; 6. that the washer needles are not blocked or partially obstructed.
Calibrator OD450nm Outside the range	1. that the procedure has been correctly performed; 2. that no mistake has occurred during its distribution (ex.: dispensation of negative control instead of Calibrator); 3. that the washing procedure and the washer settings are as validated in the pre qualification study; 4. that no external contamination of the calibrator has occurred.
Positive Control OD450nm > NC/10	1. that the procedure has been correctly performed; 2. that no mistake has occurred during the distribution of the control (dispensation of negative control instead of positive control). 3. that the washing procedure and the washer settings are as validated in the pre qualification study; 4. that no external contamination of the positive control has occurred.

If any of the above problems have occurred, report the problem to the supervisor for further actions.

P. RESULTS

The results are calculated by means of a cut-off value determined with the following formula:

$$\text{Cut-Off} = (\text{NC} + \text{PC}) / 5$$

Important note *When the calculation of results is performed by the operating system of an ELISA automated work station, ensure that the proper formulation is used to calculate the cut-off value and generate the correct interpretation of results.*

Q. INTERPRETATION OF RESULTS

Results are interpreted as ratio between the cut-off value and the sample OD450nm or Co/S. Results are interpreted according to the following table:

Co/S	Interpretation
< 0.9	Negative
0.9 – 1.1	Equivocal
> 1.1	Positive

A negative result indicates that the patient has not been infected by HDV.
Any patient showing an equivocal result should be re-tested on a second sample taken 1-2 weeks after the initial sample.
A positive result is indicative of HDV infection and therefore the patient should be treated accordingly.

Important notes:

1. *Interpretation of results should be done under the supervision of the laboratory supervisor to reduce the risk of judgement errors and misinterpretations.*
2. *When test results are transmitted from the laboratory to another facility, attention must be paid to avoid erroneous data transfer.*
3. *Diagnosis of viral hepatitis infection has to be taken by and released to the patient by a suitably qualified medical doctor.*

An example of calculation is reported below.

The following data must not be used instead or real figures obtained by the user.

Negative Control: 2.100 – 2.200 – 2.000 OD450nm
Mean Value: 2.100 OD450nm
Higher than 1.000 – Accepted

Positive Control: 0.100 OD450nm
Lower than NC/10 – Accepted

Cut-Off = (2.100 + 0.100) / 5 = 0.440

Calibrator: 0.300-0.260 OD450nm
Mean value: 0.280 OD450nm
Within the range PC < OD450nm < (NC+PC)/5 – Accepted

Sample 1: 0.020 OD450nm
Sample 2: 1.900 OD450nm
Sample 1 Co/S > 1.1 positive
Sample 2 Co/S < 0.9 negative

R. PERFORMANCES

Evaluation of Performances has been conducted in accordance to what reported in the Common Technical Specifications or CTS (art. 5, Chapter 3 of IVD Directive 98/79/EC).

1. LIMIT OF DETECTION:

In absence of an international standard, the sensitivity of the assay has been calculated by means of the product named Accurun n° 127 supplied by Boston Biomedica Inc. – USA.

The table below reports the OD450nm shown by this preparation when diluted in Fetal Calf Serum to prepare a limiting dilution curve, in three different lots.

Co/S values

	DAB.CE	Lot # 1102	DAB.CE	Lot # 0103	DAB.CE	Lot # 0403
Accurun # 127	OD450 nm	Co/S value	OD450 nm	Co/S value	OD450 nm	Co/S value
1x	0.171	3.0	0.163	2.9	0.156	2.8
2x	0.167	2.7	0.176	2.6	0.179	2.5
4x	0.230	2.2	0.220	2.1	0.202	2.2
8x	0.298	1.7	0.285	1.6	0.271	1.6
16x	0.417	1.2	0.405	1.1	0.402	1.1
32x	0.514	1.0	0.490	0.9	0.482	0.9
64x	0.717	0.7	0.700	0.7	0.705	0.6
128x	1.063	0.5	1.006	0.5	1.015	0.4
CTRL (-)	2.484	////////	2.261	////////	2.114	////////

2. DIAGNOSTIC SPECIFICITY AND SENSITIVITY

The diagnostic performances were evaluated in a clinical trial conducted by the Department of Gastro-Hepatology, Prof. M.Rizzetto, S.Giovanni Battista hospital, Torino, Italy, on more than 400 samples against a reference kit.
Negative, positive and potentially interfering samples were examined in the trial.
Both plasma, derived with different standard techniques of preparation (citrate, EDTA and heparin), and sera have been used to determine the specificity. No false reactivity due to the method of specimen preparation has been observed.
Results are briefly reported in the tables below:

Sensitivity	> 98 %
Specificity	> 98 %

3. PRECISION

The mean values obtained from a study conducted on two samples of different anti-HDV antibody reactivity, examined in 16 replicates in three separate runs for three lots of product, is reported below:

DAB.CE: lot #1102

Negative Control (N = 16)

Mean values	1st run	2nd run	3rd run	Average value
OD 450nm	2.342	2.428	2.433	2.401
Std.Deviation	0.113	0.106	0.122	0.114
CV %	4.8	4.4	5.0	4.7

Calibrator (N = 16)

Mean values	1st run	2nd run	3rd run	Average value
OD 450nm	0.298	0.289	0.286	0.291
Std.Deviation	0.023	0.027	0.026	0.025
CV %	7.7	9.3	9.1	8.7
Co/S	1.6	1.7	1.7	1.7

DAB.CE: lot #0103

Negative Control (N = 16)

Mean values	1st run	2nd run	3rd run	Average value
OD 450nm	2.208	2.237	2.246	2.230
Std.Deviation	0.105	0.108	0.108	0.107
CV %	4.7	4.8	4.8	4.8

Calibrator (N = 16)

Mean values	1st run	2nd run	3rd run	Average value
OD 450nm	0.269	0.277	0.266	0.271
Std.Deviation	0.026	0.024	0.025	0.025
CV %	9.8	8.5	9.5	9.3
Co/ S	1.7	1.7	1.7	1.7

DAB.CE: lot # 0403

Negative Control (N = 16)

Mean values	1st run	2nd run	3rd run	Average value
OD 450nm	2.246	2.221	2.182	2.216
Std.Deviation	0.097	0.103	0.118	0.106
CV %	4.3	4.6	5.4	4.8

Calibrator (N = 16)

Mean values	1st run	2nd run	3rd run	Average value
OD 450nm	0.286	0.273	0.280	0.280
Std.Deviation	0.027	0.023	0.026	0.025
CV %	9.3	8.5	9.1	9.0
Co/S	1.6	1.7	1.6	1.6

The variability shown in the tables did not result in sample misclassification.

All the IVD Products manufactured by the company are under the control of a certified Quality Management System approved by an EC Notified Body. Each lot is submitted to a quality control and released into the market only if conforming with the EC technical specifications and acceptance criteria.

Manufacturer:
Dia.Pro Diagnostic Bioprobes Srl
Via G. Carducci n° 27 – Sesto San Giovanni (MI) – Italy

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5. LIMITATIONS

Bacterial contamination or heat inactivation of the specimen may affect the absorbance values of the samples with consequent alteration of the level of the analyte.
This test is suitable only for testing single samples and not pooled ones.
Diagnosis of an infectious disease should not be established on the basis of a single test result. The patient's clinical history, symptomatology, as well as other diagnostic data should be considered.

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MINISTERIO DE SANIDAD, CONSUMO Y BIENESTAR SOCIAL

ENAC
CERTIFICACIÓN
Nº 39/C-SG055

LA AGENCIA ESPAÑOLA DE MEDICAMENTOS Y PRODUCTOS SANITARIOS
THE AGENCIA ESPAÑOLA DE MEDICAMENTOS Y PRODUCTOS SANITARIOS

otorga el certificado número
grants the certificate no.

2013 11 0039 EN

según la norma
in accordance with the standard

UNE-EN ISO 13485:2018

(EN ISO 13485: 2016 & ISO 13485: 2016)

Productos Sanitarios: Sistemas de Gestión de Calidad – Requisitos para fines reglamentarios
Medical devices – Quality management systems - Requirements for regulatory purposes

a la empresa
to the company

Dia.Pro Diagnostic Bioprobes S.r.l.

Sede social y de fabricación/ *Headquarters and manufacturing facility*
Via G. Carducci, 27-20099-Sesto San Giovanni-Milano-Italy

Para las siguientes actividades / *For the following activities:*

Diseño, desarrollo y producción de productos sanitarios para diagnóstico *in vitro*:

Reactivos y productos reactivos, calibradores y materiales de control para Inmunología Infecciosa y Técnicas de Biología Molecular

Design, development and manufacturing of "in vitro" medical devices:
Reagents, reagent products, calibrators and control materials for infectious immunology and molecular biology techniques.

Modificaciones de alcance: N/A
Fecha de validez/ *Date of validity:* **Desde/** *From:* **18-12-2018** **Hasta/***To:* **17-12-2021**
Certificación inicial/ *Initial certification date:* **27-11-2013**
Renovación / *Renewal of certification date:* **18-12-2018**

Madrid, 18 de diciembre de 2018
DIRECTORA DE LA AGENCIA ESPAÑOLA DE MEDICAMENTOS Y PRODUCTOS SANITARIOS

agencia española de medicamentos y productos sanitarios

Fdo. Mª Jesús Lamas Díaz

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Fecha de la firma: 18/12/2018
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ENAC
CERTIFICACIÓN
Nº 39/C-SG055

ANEXO I / *ANNEX I*

CERTIFICADO UNE-EN ISO 13485:2018/ *UNE-EN ISO 13485:2018 CERTIFICATE*

Modificaciones del alcance / *Scope modifications:*

Fecha/*Date*	**Descripción de la modificación**/ *Modification description*
18-12-2018	Cambio en la descripción del tipo de técnica en el ámbito tecnológico (inmunología infecciosa y técnicas de biología molecular). Cambio del nivel de detalle en la descripción del ámbito tecnológico *Change in the description of the method of analysis in the technological scope (infectious immunology and molecular biology techniques).* *Change in the level of detail of the technological scope description.*

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CERTIFICADO DE EXAMEN CE DE DISEÑO
de acuerdo con el Anexo IV, punto 4, de la Directiva 98/79/CE

EC DESIGN-EXAMINATION CERTIFICATE
in accordance with Annex IV, Section 4, Directive 98/79/EC

PRÓRROGA/EXTENSION — Fecha inicial/ Initial date: 11/12/2003
Fecha de última prórroga/ Last extension date: 27/11/2013

Certificado nº/Certificate no	Fecha de validez/Date of validity	ON nº/NB no
2003 12 0393 ED	Desde/From 19/11/2018 Hasta/To 18/11/2023	0318

A favor de /In favour of:

Fabricante/Manufacturer:
Nombre/Name: DIA. Pro Diagnostic Bioprobes S.r.l.
Dirección/Address: Via G. Carducci, 27 -20099- Sesto San Giovanni – Milano (Italy).
Representante autorizado ante la UE/Authorized EU representative:
Nombre/Name: Idem Dirección/Address: Idem

Para el producto/For the product:

Categoría/Category: Productos Sanitarios para **Diagnóstico "In Vitro"** / In Vitro Diagnostic Medical Devices
Grupo genérico/Generic group: Diagnóstico de enfermedades infecciosas / Diagnostic of infectious diseases
Tipo/Type: Especificados en Anexos de este Certificado/Specified in Annexes to this Certificate.

Elaborado en/In the facilities:

Dia. Pro Diagnostic Bioprobes S.r.l.
Via G. Carducci, 27 -20099- Sesto San Giovanni – Milano (Italy).

Este certificado debe ir acompañado por el certificado CE de Sistema de Garantía de Calidad Total Nº 2003 12 0388 CT/ This certificate must be accompanied by the EC Full Quality Assurance System Certificate Nº 2003 12 0388 CT.

Este certificado es consecuencia de la evaluación de la documentación técnica del diseño contenida en el expediente Nº 2003 05 0240, y garantiza que el diseño de los productos descritos cumple los requisitos de la Directiva/ This certificate is issued on the assessment of the design documentation contained in dossier Nº 2003 05 0240, and guarantees that the design of the described products fulfil the requirements of the Directive.

Madrid, 19 de noviembre de 2018
DIRECTORA DE LA AGENCIA ESPAÑOLA DE MEDICAMENTOS Y PRODUCTOS SANITARIOS

agencia española de medicamentos y productos sanitarios

Fdo. Mª Jesús Lamas Díaz

Firmado digitalmente por: Agencia Española de Medicamentos y Productos Sanitarios
Fecha de la firma: 19/11/2018
Localizador: GJEC8290C8

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CORREO ELECTRÓNICO
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ORGANISMO NOTIFICADO 0318

CERTIFICADO DE EXAMEN CE DE DISEÑO
de acuerdo con el Anexo IV, punto 4, de la Directiva 98/79/CE

EC DESIGN-EXAMINATION CERTIFICATE
in accordance with Annex IV, Section 4, Directive 98/79/EC

PRÓRROGA/EXTENSION — Fecha inicial/ Initial date: 11/12/2003
Fecha de última prórroga/ Last extension date: 27/11/2013

Certificado nº/Certificate no	Fecha de validez/Date of validity	ON nº/NB no
2003 12 0393 ED	Desde/From 19/11/2018 Hasta/To 18/11/2023	0318

A favor de/In favour of:

Fabricante/Manufacturer:
Nombre/Name: Dia. Pro Diagnostic Bioprobes S.r.l.
Dirección/Address: Via G. Carducci, 27 -20099- Sesto San Giovanni – Milano (Italy).
Representante autorizado ante la UE/Authorized EU representative:
Nombre/Name: Idem Dirección/Address: Idem

Tipo de producto / Device type: Reactivos y productos reactivos, calibradores y materiales de control para el diagnóstico de enfermedades infecciosas / Reagents, and reagent products, calibrators and control materials for diagnostic of human infectious diseases.

Clasificación/Classification: Lista A, Anexo II / List A, Annex II

Reactivos y productos reactivos para la determinación, confirmación y cuantificación en muestras humanas de marcadores de infección por Hepatitis D, mediante técnicas de Inmunoabsorción enzimática (ELISA)/ Reagents and reactive products for the determination, confirmation and quantification in human specimens of markers of Hepatitis D infection, by Enzyme-linked immunosorbent assay (ELISA) [NANDO: IVD 0203]

HDV Ab ELISA cualitativo / ELISA qualitative

- DAB.CE (96 tests)

Este certificado ampara todas las marcas de estos productos incluidas por el fabricante en su declaración de conformidad. / This certificate covers all trademarks of these products included by the manufacturer in his declaration of conformity.

Madrid, 19 de noviembre de 2018
DIRECTORA DE LA AGENCIA ESPAÑOLA DE MEDICAMENTOS Y PRODUCTOS SANITARIOS

agencia española de medicamentos y productos sanitarios

Fdo. Mª Jesús Lamas Díaz

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ORGANISMO NOTIFICADO 0318

LORNE LABORATORIES

EC DECLARATION OF CONFORMITY

Lorne Laboratories Ltd declares that the following in vitro diagnostic reagent:

Product name	Catalogue number
CRP Latex kit	850100A

has been classified as non List A, non List B (Directive 98/79/EC, Annex II) and complies with the essential requirements and provisions of Directive 98/79/EC of the European Parliament and of the Council (also SI 2002 No.618 which transposes the requirements of Directive 98/79/EC).

and is in conformity with the national standards transposing harmonised standards:

- BS EN 980:2008
- BS EN ISO 13485:2012
- BS EN 13612:2002
- BS EN 13640:2002
- BS EN 13641:2002
- BS EN ISO 14971:2012
- BS EN ISO 18113, parts 1&2

The conformity assessment procedure performed was in accordance with Annex III of Directive 98/79/EC.

This declaration of conformity is issued under the sole responsibility of Lorne Laboratories Ltd and is valid from 13 April 2016.

Eddy Velthuis
Technical Director

File No A12241;
ISO 13485:2003; ISO 9001:2008

Lorne Laboratories Limited
Unit 1 Cutbush Park Industrial Estate
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Email: info@lornelabs.com
www.lornelabs.com

LORNE LABORATORIES

EC DECLARATION OF CONFORMITY

Lorne Laboratories Ltd declares that the following in vitro diagnostic reagent:

Product name	Catalogue number
ASO Latex kit	031100A

has been classified as non List A, non List B (Directive 98/79/EC, Annex II) and complies with the essential requirements and provisions of Directive 98/79/EC of the European Parliament and of the Council (also SI 2002 No.618 which transposes the requirements of Directive 98/79/EC).

and is in conformity with the national standards transposing harmonised standards:

- BS EN 980:2008
- BS EN ISO 13485:2012
- BS EN 13612:2002
- BS EN 13640:2002
- BS EN 13641:2002
- BS EN ISO 14971:2012
- BS EN ISO 18113, parts 1&2

The conformity assessment procedure performed was in accordance with Annex III of Directive 98/79/EC.

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Eddy Velthuis
Technical Director

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LORNE
LABORATORIES

EC DECLARATION OF CONFORMITY

Lorne Laboratories Ltd declares that the following in vitro diagnostic reagent:

Product name	Catalogue number
RF Latex kit	830100A

has been classified as non List A, non List B (Directive 98/79/EC, Annex II) and complies with the essential requirements and provisions of Directive 98/79/EC of the European Parliament and of the Council (also SI 2002 No.618 which transposes the requirements of Directive 98/79/EC).

and is in conformity with the national standards transposing harmonised standards:

- BS EN 980:2008
- BS EN ISO 13485:2012
- BS EN 13612:2002
- BS EN 13640:2002
- BS EN 13641:2002
- BS EN ISO 14971:2012
- BS EN ISO 18113, parts 1&2

The conformity assessment procedure performed was in accordance with Annex III of Directive 98/79/EC.

This declaration of conformity is issued under the sole responsibility of Lorne Laboratories Ltd and is valid from 13 April 2016.

Eddy Velthuis
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File No A12241:
ISO 13485:2003; ISO 9001:2008

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LORNE LABORATORIES LTD.

GREAT BRITAIN

CE

RAPID LATEX KIT

DIRECTIONS FOR USE

ASO Latex Kit: For Detection Of Anti-Streptolysin O (ASO) In Serum.

SUMMARY

In acute streptococcal infections, the toxic immunogenic exoenzyme streptolysin O (ASO) is produced in response to streptolysin O antigens liberated by haemolytic streptococci of groups A, C and G. Information on extent and degree of infection can be obtained by measuring serum ASO levels. Elevated ASO levels have also been found in patients suffering from scarlet fever, acute rheumatoid arthritis, tonsillitis, and other streptococcal infections as well as in healthy carriers.

PRINCIPLE

When used by the recommended techniques, latex particles in the reagent will agglutinate (clump) in the presence of anti-streptolysin O antibodies. No agglutination generally indicates the absence of anti-streptolysin O antibodies (see **Limitations**).

KIT DESCRIPTION

Lorne ASO Latex Kit is a serologic test for the detection of ASO antibodies. All the reagents are supplied at optimal dilution for use with all recommended techniques without the need for further dilution or addition. For lot reference number and expiry date see **Vial Labels**.

STORAGE

Do not freeze. Reagent vials should be stored at 2 - 8°C on receipt. Prolonged storage at temperatures outside this range may result in accelerated loss of reagent reactivity.

SPECIMEN COLLECTION

Specimens should be drawn with or without anticoagulant using an aseptic phlebotomy technique. If testing is delayed specimens can be stored at 2-8°C for 7 days or for up to 3 months at or below –20°C. Specimens must be free from bacterial contamination, fibrin, gross lipemia and gross haemolysis.

PRECAUTIONS

1. The kit is for *in vitro* diagnostic use only.
2. Do not use kit past expiration date (see **Vial and Box Labels**).
3. Protective clothing should be worn when handling the reagents, such as disposable gloves and a laboratory coat.
4. Materials used to produce the kit were tested at source and found to be negative for HIV 1+2 and HBsAg using approved microbiological tests. However no known tests can guarantee that products derived from human or animal sources are free from infectious agents. Care must be taken in the use and disposal of each vial and its contents.

DISPOSAL OF KIT REAGENT AND DEALING WITH SPILLAGES

For information on disposal of kit reagent and decontamination of a spillage site see **Material Safety Data Sheets**, available on request.

CONTROLS AND ADVICE

1. ASO Positive and Negative Controls must be tested in parallel with each batch of tests. Tests must be considered invalid if controls do not show expected results.
2. All the reagents must be allowed to reach 18-25°C before use.
3. Shake the reagents well before use to ensure homogeneity.
4. Do not interchange components between different kits.
5. Use of kit and interpretation of results must be carried out by properly trained and qualified personnel in accordance with the requirements of the country where the kit is in use.
6. The user must determine the suitability of the kit for use in other techniques.

KIT COMPONENTS SUPPLIED

1) ASO Latex Reagent 5.0 mL: Latex particles coated with streptolysin O, pH, 8.2 containing a preservative.
2) ASO Positive Control (1.0 mL, Red cap): Human serum with an ASO concentration > 200 IU/mL containing a preservative.
3) ASO Negative Control (1.0 mL, Blue cap): Animal serum containing a preservative.
4) Pipette-Stirrers.
5) Disposable agglutination Slides.

REAGENTS AND MATERIALS REQUIRED

a) Small glass / plastic test tubes.
b) Serological pipettes.
c) Graduated container.
d) 9 g/L saline solution.

RECOMMENDED QUALITATIVE TECHNIQUE

1. Allow the reagents and samples to reach room temperature. The sensitivity of the test may be reduced at low temperatures.
2. Place 50 µL of the sample and one drop of each Positive and Negative controls into separate circles on the slide test.
3. Swirl the ASO-latex reagent gently before using and add one drop (50 µL) next to the sample to be tested.
4. Mix the drops with a stirrer, spreading them over the entire surface of the circle. Use different stirrers for each sample.
5. Swirl the slide gently and after 2 minutes read the results macroscopically. False positive results could appear if the test is read after more than two minutes.

INTERPRETATION OF QUALITATIVE RESULTS

1. **Positive:** Agglutination of latex particles constitutes a positive result and within the accepted limitations of the test procedure, indicates a level of ASO in the specimen > 200 I.U/ml.
2. **Negative:** No visible agglutination of latex particles constitutes a negative result and within the accepted limitations of the test procedure, indicates a level of ASO in specimen < 200 I.U/mL.

RECOMMEDED SEMI-QUANTITATIVE TECHNIQUE

1. The semi-quantitative test can be performed in the same way as the quantitative technique using dilutions of the serum in 9 g/L saline solution.
2. Make doubling dilutions of specimen as follows:

Dilution	Serum	Saline
1/2	100 µl undiluted serum	100 µl
1/4	100 µl 1/2 diluted serum	100 µl
1/8	100 µl 1/4 diluted serum	100 µl

3. Test the specimen dilutions in the same way as for the quantitative technique above.
4. Agglutination of the sera indicates:

Dilution	ASO Levels (I.U. / ml)
1/2	400 (200 x 2)
1/4	800 (200 x 4)
1/8	1600 (200 x 8)

5. Normal levels of ASO in adults is < 200 I.U/ml.

RESULTS

The titre is expressed as the reciprocal of the highest dilution showing macroscopic agglutination, e.g. if this occurs in the 1/8 dilution, the titre is 1600.

INTERPRETATION OF SEMI-QUANITATIVE RESULTS

Positive results may indicate an acute streptococcal infection. In which case the test should be repeated at weekly intervals to determine the progression of infection.

STABILITY OF THE REACTIONS

Slide tests should be interpreted immediately after the 2-minute period to avoid the possibility that a negative result may be incorrectly interpreted as positive due to drying of the reagent.

LIMITATIONS

1. False positive results may be obtained in conditions such as scarlet fever, acute rheumatoid arthritis, tonsillitis and other streptococcal infections as well as in healthy carriers.
2. Hemoglobin (≤ 10 g/L), bilirubin (≤ 20 mg/dL), lipemia (≤ 10 g/L), rheumatoid factors (≤ 300 IU/mL) do not interfere. Other substances may interfere[7].
3. Early infections in children from 6 months to 2 years may cause false negative results.
4. A single ASO determination does not produce much information about the actual state of the disease. Titrations at biweekly intervals during 4 or 6 weeks are advisable to follow the disease evolution.
5. Clinical diagnosis should not be made on findings of a single test result, but should integrate both clinical and laboratory data.
6. False positive or false negative results may also occur due to:
   a) Contamination of test materials
   b) Improper storage of test materials or omission of reagents
   c) Deviation from the recommended techniques

SPECIFIC PERFORMANCE CHARACTERISTICS

1. The kit has been characterised by all the procedures mentioned in the **Recommended Techniques**.
2. Prior to release, each lot of Lorne ASO Latex Kit is tested by **Recommended Techniques** to ensure suitable reactivity.
3. Analytical sensitivity: 200 (± 50) IU/mL, under the described assay conditions.
4. Prozone effect: No prozone effect was detected up to 1500 IU/mL.
5. Diagnostic sensitivity: 98 %.
6. Diagnostic specificity: 97 %.

DISCLAIMER

1. The user is responsible for the performance of the kit by any method other than those mentioned in the **Recommended Techniques**.
2. Any deviations should be validated prior to use using established laboratory procedures.

BIBLIOGRAPHY

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2. Ahmed Samir et al. Pediatric Annals 1992; 21: 835-842.
3. Spaun J et al. Bull Wld Hlth Org 1961; 24: 271-279.
4. The association of Clinical Pathologists 1961. Broadsheet 34.
5. Picard B et al. La Presse Medicale 1983; 23: 2-6.
6. Klein GC. Applied Microbiology 1971; 21: 999-1001.
7. Young DS. Effects of drugs on clinical laboratory test. 4th ed. AACC Press, 1995.

AVAILABLE KIT SIZES

Kit Size	Catalogue Number
100 Tests Per Kit: 5.0 ml ASO Latex 1.0 ml Positive Control 1.0 ml Negative Control	031100A

For the availability of other sizes, please contact:

Lorne Laboratories Limited
Unit 1 Cutbush Park Industrial Estate
Danehill
Lower Earley
Berkshire, RG6 4UT
England
Tel: +44 (0) 118 921 2264
Fax: +44 (0) 118 986 4518
E-mail: info@lornelabs.com

TABLE OF SYMBOLS

LOT	Batch Number	IVD	*In-vitro* Diagnostic
REF	Catalogue Reference		Store At
	Expiry Date		Manufacturer
	Read Pack Insert		

LORNE LABORATORIES LTD.
GREAT BRITAIN

CE

RAPID LATEX KIT
DIRECTIONS FOR USE

CRP Latex Kit: For Detection Of C-Reactive Protein (CRP) In Serum.

SUMMARY

C-Reactive Protein (CRP) usually appears in serum of individuals in response to inflammatory conditions and tissue necrosis and disappears when causative conditions subside. It is routinely found in cases of bacterial infection, active rheumatic fever and many malignant diseases and is often seen in association with rheumatoid arthritis, viral infections and tuberculosis. CRP has also been detected in patients following blood transfusions and surgical operations as well as in patients with burns, pemphigus vulgaris and other bullaous lesions.

PRINCIPLE

When used by the recommended techniques, latex particles in the reagent will agglutinate (clump) in the presence of CRP. No agglutination generally indicates absence of CRP (see **Limitations**).

KIT DESCRIPTION

Lorne CRP Latex Test Kit is for the detection of CRP. Test reagent consists of latex particles coated with rabbit Anti-CRP (IgG). All the latex reagents are supplied at optimal dilution for use with all recommended techniques without need for further dilution or addition. For lot reference number and expiry date see **Vial Labels**.

STORAGE

Do not freeze. Reagent vials should be stored at 2 - 8°C on receipt. Prolonged storage at temperatures outside this range may result in accelerated loss of reagent reactivity.

SPECIMEN COLLECTION

Specimens should be drawn with or without anticoagulant using an aseptic phlebotomy technique. If testing is delayed specimens can be stored at 2-8°C for 7 days or for up to 3 months at or below –20°C. Specimens must be free from bacterial contamination, fibrin, gross lipaemia and gross haemolysis.

PRECAUTIONS

1. The kit is for *in vitro* diagnostic use only.
2. Do not use kit past expiration date (see **Vial and Box Label**).
3. Protective clothing should be worn when handling the reagents, such as disposable gloves and a laboratory coat.
4. Materials used to produce the kit were tested at source and found to be negative for HIV 1+2 and HBsAg using approved microbiological tests. However, no known tests can guarantee that products derived from human or animal sources are free from infectious agents. Care must be taken in the use and disposal of each vial and its contents.

DISPOSAL OF KIT REAGENT AND DEALING WITH SPILLAGES

For information on disposal of kit reagent and decontamination of a spillage site see **Material Safety Data Sheets**, available on request.

CONTROLS AND ADVICE

1. It is recommended the CRP Positive and Negative Controls be tested in parallel with each batch of tests. Tests must be considered invalid if controls do not show expected results.
2. All the reagents must be allowed to reach 18-25°C before use.
3. Shake the reagents well before use to ensure homogeneity.
4. Do not interchange components between different kits.
5. Use of kit and interpretation of results must be carried out by properly trained and qualified personnel in accordance with the requirements of country where kit is in use. The user must determine the suitability of the kit for use in other techniques.

KIT COMPONENTS SUPPLIED

1) CRP Latex Reagent (5 mL): Latex particles coated with goat IgG anti-human CRP, pH, 8.2, containing a preservative.
2) CRP Positive Control (Red cap, 1 mL): Human serum with a CRP concentration > 20 mg/L containing a preservative.
3) CRP Negative Control (Blue cap, 1 mL): Animal serum containing a preservative.
4) Pipette stirrers.
5) Disposable agglutination slide.

MATERIALS AND EQUIPMENT REQUIRED

a) Serological Pipettes.
b) Mechanical rotator capable with adjustable speed of 80-100 rpm.
c) Vortex mixer.
d) Small Glass or Plastic Test Tubes.
e) Distilled or Deionised Water.
f) 9 g/L saline solution.

RECOMMENDED QUALITATIVE TECHNIQUE

1. Allow the reagents and samples to reach room temperature. The sensitivity of the test may be reduced at low temperatures.
2. Place 50 µL of the sample (Note 1) and one drop of each Positive and Negative controls into separate circles on the slide test.
3. Swirl the CRP-latex reagent gently before using and add one drop (50 µL) next to the samples to be tested.
4. Mix the drops with a stirrer, spreading them over the entire surface of the circle. Use different stirrers for each sample.
5. Place the slide on a rotary shaker at 80-100 r.p.m. for 2 minutes. False positive results could appear if the test is read after more than two minutes.

INTERPRETATION OF QUALITATIVE RESULTS

1. **Positive:** Visible agglutination of latex particles constitutes a positive result and within the accepted limitations of the test procedure, indicates a level of CRP in the specimen > 6 mg/l.
2. **Negative:** No visible agglutination of latex particles constitutes a negative result and within the accepted limitations of the test procedure, indicates a level of CRP in the specimen < 6 mg/l.

RECOMMENDED SEMI-QUANTITATIVE TECHNIQUE

1. The semi-quantitative test can be performed in the same way as the qualitative test using dilutions of the serum.
2. Make doubling dilutions of the specimen using 9 g/L saline solution as follows:

Dilution	Serum	Saline
1/2	100 µl undiluted serum	100 µl
1/4	100 µl 1/2 diluted serum	100 µl
1/8	100 µl 1/4 diluted serum	100 µl
1/16	100 µl 1/8 diluted serum	100 µl

3. Test the specimen dilutions in the same way as for the quantitative technique above.
4. Agglutination of the sera indicates:

Dilution	CRP Levels (mg / l)
1/2	12 (6 x 2)
1/4	24 (6 x 4)
1/8	48 (6 x 8)
1/16	96 (6 x 16)

5. Normal levels of CRP in adults are < 6 mg/l.

NOTES

1. High CRP concentration samples may give false negative results (pro-zone effect). Re-test the sample again using a drop of 20 µl.

RESULTS

The titre is expressed as the reciprocal of the highest dilution showing macroscopic agglutination: e.g. if this occurs in dilution 1/8, the titre is 48.

INTERPRETATION OF SEMI-QUANTITATIVE RESULTS

The elevation of CRP levels above normal indicates tissue damage, inflammation, or both with greater reliability. Regular monitoring of CRP levels is often used as a means of assessing disease activity and of guiding therapy. CRP determination is considered of greater practical significance than other indicators of inflammatory disease. Erythrocyte sedimentation rate (ESR) may become elevated as a result of non-inflammatory conditions. In these circumstances inflammatory disease may be excluded if CRP is absent.

STABILITY OF THE REACTIONS

Slide tests should be interpreted immediately after the 2-minute rotation period to avoid the possibility that a negative result may be incorrectly interpreted as positive due to drying of the reagent.

LIMITATIONS

1. Reactions read beyond the two-minute interval may be invalid.
2. The results obtained from this assay must be considered a part of the differential diagnosis and medical history of the patient.
3. There is no relationship between the strength of reactivity and C-reactive protein levels.
4. Hemoglobin (≤ 10 g/L), bilirubin (≤ 20 mg/dL) and lipemia (≤ 10 g/L), do not interfere. Rheumatoid factors (≥ 100 IU/mL), interfere. Other substances may interfere[7].
5. False positive or false negative results may also occur due to:
   a) Contamination of test materials
   b) Improper storage of test materials or omission of reagents
   c) Deviation from the recommended techniques

SPECIFIC PERFORMANCE CHARACTERISTICS

1. The kit has been characterised by all the procedures mentioned in the **Recommended Techniques**.
2. Prior to release, each lot of Lorne CRP Latex Test Kit is tested by **Recommended Techniques** to ensure suitable reactivity.
3. The CRP-latex sensitivity is calibrated to the Reference Material ERM-DA 472/IFCC.
4. **Analytical sensitivity:** 6 (5-10) mg/L, under the described assay conditions.
5. **Prozone effect:** No prozone effect was detected up to 1600 mg/L (Note 1).
6. **Diagnostic sensitivity:** 95.6 %.
7. **Diagnostic specificity:** 96.2 %.

DISCLAIMER

1. The user is responsible for the performance of the kit by any method other than those mentioned in the **Recommended Techniques**.
2. Any deviations should be validated prior to use using established laboratory procedures.

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7. Young DS. Effects of drugs on clinical laboratory test, 4th ed. AACC Press, 1995.

AVAILABLE KIT SIZES

Kit Size	Catalogue Number
100 Tests Per Kit	850100A

For the availability of other sizes, please contact:

Lorne Laboratories Limited
Unit 1 Cutbush Park Industrial Estate
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TABLE OF SYMBOLS

LOT	Batch Number	IVD	*In-vitro* Diagnostic
REF	Catalogue Reference		Store At
	Expiry Date		Manufacturer
	Read Pack Insert		

LORNE LABORATORIES LTD.

GREAT BRITAIN

CE

RAPID LATEX KIT

DIRECTIONS FOR USE

RF Latex kit: For Detection Of Rheumatoid Factor (RF).

SUMMARY

Rheumatoid factors are a group of antibodies directed to determinants in the Fc portion of the IgG molecule. Although rheumatoid factors are found in a number of rheumatoid disorders, such as systemic lupus erythematosis (sLE) and Sjögren's syndrome, as well as in non-rheumatoid conditions, it's central role lies in aiding in the diagnosis of rheumatoid arthritis

PRINCIPLE

When used by recommended techniques, latex particles in reagent will agglutinate (clump) in presence of rheumatoid factor (RF). No agglutination generally indicates absence of RF (see **Limitations**).

KIT DESCRIPTION

Lorne RF Latex Kit is for the detection of rheumatoid factor. The latex reagent is a suspension of polystyrene latex particles coated with human gamma globulins, which agglutinate in the presence of Rheumatoid Factor (RF). All latex reagents are supplied at optimal dilution for use with all recommended techniques without the need for further dilution or addition. For lot reference number and expiry date see **Vial Labels**.

STORAGE

Do not freeze. Reagent vials should be stored at 2 - 8°C on receipt. Prolonged storage at temperatures outside this range may result in accelerated loss of reagent reactivity.

SPECIMEN COLLECTION

Specimens should be drawn without anticoagulant using an aseptic phlebotomy technique. If testing is delayed, fresh serum can be stored at 2-8°C for 7 days or for up to 3 months at or below –20°C. Specimens must be free from bacterial contamination, fibrin, gross lipeamia and gross haemolysis.

PRECAUTIONS

1. The kit is for *in vitro* diagnostic use only.
2. Do not use kit past expiration date (see **Vial and Box Labels**).
3. Protective clothing should be worn when handling the reagents, such as disposable gloves and a laboratory coat.
4. All the reagents must be allowed to reach 18-25°C before use.
5. Materials used to produce the kit were tested at source and found to be negative for HIV 1+2 and HBsAg using approved microbiological tests. However, no known tests can guarantee that products derived from human or animal sources are free from infectious agents. Care must be taken in the use and disposal of each vial and its contents.

DISPOSAL OF KIT REAGENT AND DEALING WITH SPILLAGES

For information on disposal of kit reagent and decontamination of a spillage site see **Material Safety Data Sheets**, available on request.

CONTROLS AND ADVICE

1. It is recommended the RF Positive and Negative Controls be tested in parallel with each batch of tests. Tests must be considered invalid if controls do not show expected results.
2. All the reagents must be allowed to reach 18-25°C before use.
3. Shake the reagents well before use to ensure homogeneity.
4. Do not interchange components between different kits.
5. Use of kit and interpretation of results must be carried out by properly trained and qualified personnel in accordance with the requirements of country where kit is in use. The user must determine the suitability of the kit for use in other techniques.
6. Results obtained with a latex method do not compare with those obtained with the Rose Waaler test. Differences in the results between methods do not reflect differences in the ability to detect rheumatoid factors.

KIT COMPONENTS SUPPLIED

1) RF Latex Reagent (5 mL): Latex particles coated with human γ-globulin, pH, 8.2, and a preservative.
2) RF Positive Control (Red cap, 1 mL): Human serum with a RF concentration > 30 IU/mL and a preservative.
3) RF Negative Control (Blue cap, 1 mL): Animal serum and a preservative.
4) Pipette-Stirrers.
5) Reusable Agglutination Slide (18 each).

MATERIALS AND EQUIPMENT REQUIRED

a) Glass Test Tubes (10 x 75 mm or 12 x 75 mm).
b) Pasteur and Graduated Pipettes.
c) Vortex mixer.
d) Mechanical rotator with adjustable speed of 80-100 rpm.

RECOMMENDED QUALITATIVE TECHNIQUE

1. Allow the reagents and samples to reach room temperature. The sensitivity of the test may be reduced at low temperatures.
2. Place 50 µL of the sample and one drop of each Positive and Negative controls into separate circles on the slide test.
3. Shake the RF-latex reagent vigorously or use a vortex mixer before use and add one drop (50 µL) next to the sample to be tested.
4. Mix the drops with a stirrer, spreading them over the entire surface of the circle. Use different stirrers for each sample.
5. Place the slide on a rotary shaker at 80-100 r.p.m. for 2 minutes. False positive results could appear if the test is read after more than two minutes.

INTERPRETATION OF QUALITATIVE RESULTS

1. **Positive:** Visible agglutination of latex particles constitutes a positive result and within the accepted limitations of the test procedure, indicates a level of RF in the specimen > 8 IU/ml.
2. **Negative:** No visible agglutination of latex particles in a milky liquid constitutes negative result and within accepted limitations of test procedure, indicates level of < 8 IU/ml RF in specimen.

RECOMMENDED SEMI-QUANTITATIVE TECHNIQUE

1. The semi-quantitative test can be performed in the same way as the qualitative test using dilutions of the serum.
2. Make doubling dilutions of serum specimen in 9 g/L saline as follows:

Dilution	Serum	Saline
1/2	100 µl undiluted serum	100 µl
1/4	100 µl 1/2 diluted serum	100 µl
1/8	100 µl 1/4 diluted serum	100 µl
1/16	100 µl 1/8 diluted serum	100 µl

3. Test the specimen dilutions in the same way as for the quantitative technique above.
4. Agglutination of the sera indicates:

Dilution	RF Levels (I.U/mL)
1/2	16 (8 x 2)
1/4	32 (8 x 4)
1/8	64 (8 x 8)
1/16	128 (8 x 16)

5. Normal levels of RF in adults is < 8 IU/mL.

RESULTS

The titre is expressed as the reciprocal of the highest dilution showing macroscopic agglutination: e.g. if this occurs in dilution 1/8, the titre is (8 x 8 IU/mL) = 64 IU/mL.

STABILITY OF THE REACTIONS

Slide tests should be interpreted straight after the 2-minute rotation period to avoid the possibility that a negative result may be incorrectly interpreted as positive due to drying of the reagent.

LIMITATIONS

1. Using a latex test system, positive results are not always found with every case of clinically defined rheumatoid arthritis, the number of positives reported using various types of latex reagent range from 70% to over 90%.
2. The incidence of false positive results is about 3-5 %. Individuals suffering from infectious mononucleosis, hepatitis, syphilis as well as elderly people may give positive results.
3. Diagnosis should not be solely based on the results of latex method but also should be complemented with a Rose Waaler test along with the clinical examination.
4. Haemoglobin (≤ 10 g/L), bilirubin (≤ 20 mg/dL) and lipaemia (≤ 10 g/L) do not interfere. Other substances may interfere[6].
5. False positive or false negative results may also occur due to:
   a) Contamination of test materials
   b) Improper storage of test materials or omission of reagents
   c) Deviation from the recommended techniques

SPECIFIC PERFORMANCE CHARACTERISTICS

1. The kit has been characterised by all the procedures mentioned in the **Recommended Techniques**.
2. The RF latex sensitivity is calibrated against the RF International Calibrator from the WHO (WHO 64/2 Rheumatoid Arthritis Serum).
3. Analytical sensitivity: 8 (6-16) IU/mL, under the described assay conditions.
4. Prozone effect: No prozone effect was detected up to 1500 IU/mL.
5. Diagnostic sensitivity: 100 %.
6. Diagnostic specificity: 100 %.
   The diagnostic sensitivity and specificity have been obtained using 139 samples compared with the same method of a competitor.

DISCLAIMER

1. The user is responsible for the performance of the kit by any method other than those mentioned in the **Recommended Techniques.**
2. Any deviations should be validated prior to use using established laboratory procedures.

BIBLIOGRAPHY

1. Robert W Dorner et al. Clinica Chimica Acta 1987; 167: 1 – 21.
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AVAILABLE KIT SIZES

Kit Size	Catalogue Number
100 Tests Per Kit	830100A

For the availability of other sizes, please contact:

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TABLE OF SYMBOLS

LOT	Batch Number	IVD	*In-vitro* Diagnostic
REF	Catalogue Reference		Store At
	Expiry Date		Manufacturer
	Read Pack Insert		

Declaration of Conformity

HL-7-0692DC DOI 2015/08 (1)

In Application of the Council Directive 98/79/EC on the approximation of the laws of the Member States relating to *In Vitro* Diagnostic Medical Devices & CE marking.

Declaration of conformance to applicable sections of Annex I - Essential Requirements and Annex III (EC Declaration of Conformity) imposed by sections 2 to 5. The below listed products are not classified under Annex II Lists A or B. Access to the appropriate technical files will be made available to the appropriate body in the event this is required.

Product Code	Description	GMDN Classification Code
5250H	Manual D-Dimer	47346

I, the undersigned declare that the devices registered against the above GMDN Classification Code conforms to the said Directives.

Full Name: M.J. Stephenson Title: Managing Director

Signed: Date: 25 Aug 2015

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Manual D-Dimer

REF 5250
REF 5250H

CE IVD

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HL-2-0554P 2016/01 (14)

Manual D-Dimer
Instructions for use

en

INTENDED PURPOSE

The Manual D-Dimer kit is intended for carrying out immunoturbidimetric based haemostasis assays.

Manual D-Dimer is a latex agglutination test for semi-quantitative determination of fibrin D-dimer. D-dimer containing moieties are formed by plasmin degradation of Factor XIIIa cross-linked fibrin. Elevated levels of D-dimer are found in clinical conditions such as deep vein thrombosis (DVT), pulmonary embolism (PE) and disseminated intravascular coagulation (DIC)[1,2]. D-dimer levels rise during pregnancy and high levels are associated with complications[3]. Manual D-Dimer utilizes a monoclonal antibody reacting with fibrin D-dimer or fragment D of fibrin but not with intact fibrinogen[4], allowing D-dimer determination in human plasma. Serum samples suited for FDP analysis can also be used. No international standard for D-dimer exists at present. For numerical values Helena has aligned the Manual D-Dimer with commercially available enzyme-linked immunosorbent assay (ELISA) tests.

WARNINGS AND PRECAUTIONS

The reagents contained in this kit are for in vitro diagnostic use only – DO NOT INGEST. Wear appropriate personal protective equipment when handling all kit components. Refer to the product safety declaration for the link to appropriate hazard and precautionary statements where applicable. Dispose of components in accordance with local regulations.

Blood products have been screened and found negative (unless otherwise stated on the kit box or vial) for the presence of:
Hepatitis B Antigen (HbsAg)
HIV 1 antibody
HIV 2 antibody
HCV antibody
However they should be handled with the same precautions as a human patient sample.

COMPOSITION

Component	Content	Description	Preparation
Manual D-Dimer Latex	1 x 1.7 mL (REF 5250) 1 x 0.85 mL (REF 5250H)	Suspension of latex beads. Beads are coated with anti D-dimer monoclonal antibody.	Agitate the latex by repeatedly inverting the vial for 5 seconds immediately before use. If latex mass collects at the neck of the vial or is visible in the latex suspension, vortex vial for 10 seconds, to ensure any settled material is resuspended.
Positive Control Plasma	1 x 1 mL	Lyophilized human plasma enriched with fibrin D-dimer.	Reconstitute with 1 mL Saline Solution. Allow to stand for 20 minutes and mix well before use, do not shake.
Negative Control Plasma	1 x 1 mL	Lyophilized human plasma.	Reconstitute with 1 mL of Saline Solution. Allow to stand for 20 minutes, and mix well before use, do not shake.
Saline Solution	2 x 8 mL (REF 5250) 1 x 8 mL (REF 5250H)	Buffered saline, pH 7.3. Contains 0.2 g/L sodium azide.	The reagent is ready for use as packaged.
Test Cards	16 x 6 sample (REF 5250) 8 x 6 sample (REF 5250H)		
Mixing Sticks	50 (REF 5250) 25 (REF 5250H)		

Each kit contains instructions for use.

ITEMS REQUIRED BUT NOT PROVIDED

1. Pipettes for 20, 100 and 1000 μL volumes
2. Pipette tips
3. 3 mL test tubes
4. Timer

STORAGE, SHELF-LIFE AND STABILITY

The reagents should be stored at $2-8°C$ and used prior to the expiration date recorded on the box. Control plasmas, once reconstituted, may be stored at $2-8°C$ for one week or one month at -20°C. The rubber stopper in the control vials may be discarded after vial opening.

SAMPLE COLLECTION AND PREPARATION

Plastic or siliconized glass should be used throughout. Blood (9 parts) should be collected into 3.2% or 3.8% sodium citrate anticoagulant (1 part). Separate plasma after centrifugation at 1500 x g for 15 minutes[5]. Plasma should be kept at $2-8°C$ or $18-24°C$. Testing should be completed within 24 hours of sample collection, or plasma can be stored frozen at -20°C or -70°C for 24 months[6]. Thaw quickly at 37°C prior to testing. Do not keep at 37°C for more than 5 minutes.

PROCEDURE

Allow all vials to warm to room temperature before use, minimum 10 minutes.

A. Qualitative method

1. Place 20 μL of sample, positive and negative control plasmas in circles on a test card.
2. Place 20 μL of Manual D-Dimer latex suspension in a nearby area of each circle.
3. Quickly mix the sample and Manual D-Dimer latex using clean mixing sticks for each sample. Start the timer.
4. Rock the test card gently and read agglutination between 180 and 200 seconds. Positive (+) or negative (-) agglutination are compared to results obtained using the controls. The positive control is merely qualitative and should not be further diluted. Non-agglutinated latex means the sample is normal and no further testing is required.

B. Semi-quantitative method

Performed only on samples tested positive.

1. Serially dilute 100 μL of sample 1:2, 1:4 and 1:8 with 100 μL Saline Solution using small test tubes.
2. Mark the positions of sample dilutions on the test card and mix with latex suspension as per Qualitative method.
3. The D-Dimer concentration may be determined from the table at "Reference Values".

INTERPRETATION OF RESULTS

1. Agglutination occurs within 180-200 seconds for samples containing more than 250 ng/mL D-dimer. The mean level of D-dimer in a healthy population is between about 8 and 135 ng/mL, and neat plasma from normal, healthy individuals should not agglutinate. The negative predictive value of the Manual D-Dimer for thrombosis is high[7]. The circulatory half-life of D-dimer is about 12 hours. Elevated D-dimer levels can therefore persist for some time after the active process has ceased. In clinical studies on normal subjects, patients with phlebographically confirmed DVT, patients with DIC and patients with pre-eclampsia (Pre-EC) the following results were obtained; titres indicate the highest dilution at which the sample shows agglutination.

Sample	Number of patients	Negative D-Dimer	Plasma dilution: Undiluted	1:2	1:4	1:8
Normal	101	100	1*	-	-	-
DVT	48	3	10*	7*	14*	14*
DIC	29	0	3	3	4	19
Pre-EC	6	2	1	3	-	-

* The agglutination was inhibited by addition of the D-dimer specific antibody (0.2 mg/mL) but not with a non-related antibody PAM-1.

2. The results may be reported either in D-dimer units or in fibrinogen equivalent units (FEU). 1 ng/mL of D-dimer is equivalent to approximately 2 ng/mL of FEU[6].
3. Agglutination will be more pronounced and appears more rapidly at higher D-dimer concentrations.

Should you require quantitative D-Dimer analysis, Helena Biosciences Europe supplies the following kits for use on photo-optical instruments:

REF 5551 / 5552 / 5554	Auto Blue D-Dimer 400
REF 5601 / 5501	Auto Red D-Dimer 700

LIMITATIONS

1. A negative Manual D-Dimer test does not completely rule out thrombosis. A negative predictive value for patients with suspected DVT has been found to be 94% with the Manual D-Dimer kit[8]. Detection of elevated levels of D-dimer should be used with other clinical information in forming a diagnosis.
2. Agglutination with samples containing normal D-dimer levels may occasionally occur due to non-specificity.
3. A small number of samples, when mixed with the latex, may exhibit white flakes which should not be confused with agglutination.

QUALITY CONTROL

The positive and negative controls provided should be used for quality control of each kit. It is recommended that both positive and negative controls are tested each time the kit is used. If either positive or negative control fails to elicit the appropriate response, patient results obtained on that occasion should not be used and the kit should be discarded or returned to Helena Biosciences Europe.

REFERENCE VALUES

Agglutination occurs within 180 to 200 seconds for samples containing more than 250 ng/mL D-dimer. By testing serially diluted plasmas, semi-quantitative results can be obtained.

D-Dimer Level (ng/mL) ng/mL	Undiluted	1:2	1:4	1:8
< 250	-	-	-	-
250-500	+	-	-	-
500-1000	+	+	-	-
1000-2000	+	+	+	-
> 2000	+	+	+	+

Agglutination may be more pronounced and appears more rapidly at higher D-dimer concentrations. If you wish to express the results in fibrinogen equivalent units (FEU) multiply the D-dimer levels in the table above by 2, e.g. <250 ng/mL becomes <500 FEU. As discussed in several reports[7,8], some commercial latex tests do not have the claimed sensitivity when compared to commercial ELISA.

PERFORMANCE CHARACTERISTICS

Specificity

The monoclonal antibody used in this device is specific for D-dimer by virtue of the screening method used for hybridoma selection[4]. A hybridoma, secreting antibodies which reacted positively with purified D-dimer but not with whole fibrinogen or fragment D of fibrinogen, was selected. No crossreactivity with fibrinogen or des-AA-fibrinogen was observed when either analyte was substituted for plasma in the assay. Plasma from 16 patients with rheumatoid arthritis was assayed and 14 of these were found to be non-agglutinating with the Manual D-Dimer test. The two agglutinations could be inhibited by addition of the D-dimer specific monoclonal antibody but not with addition of a monoclonal of the same subgroup, IgG1k (PAM-1). This suggests that the Manual D-Dimer is insensitive to rheumatoid factor disturbances.

Reproducibility

Three plasma samples were selected to test reproducibility of the assay. Each sample was tested 10 times on each of three different days. In each case, where a positive result was obtained, the sample was titrated. The results in D-dimer (ng/mL):

Reproducibility

Sample	D-Dimer Level	Result
Normal	< 250 ng/mL	Negative
Intermediate	2000 ng/mL	Titer 1:8
High	> 16,000 ng/mL	Titer 1:64

Accuracy

The Manual D-Dimer kit was compared to another commercially available D-dimer latex. Both products gave a negative reaction when tested on 25 normal specimens. When 30 patient plasma samples were tested with ELISA and Manual D-Dimer, all samples with ELISA values above 225 ng/mL showed agglutination with Manual D-Dimer.

BIBLIOGRAPHY

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Manual D-Dimer
Fiche technique

fr

UTILISATION

Le kit Manual D-Dimer est destiné à la réalisation des analyses de l'hémostase par immunoturbidimétrie.

Manual D-Dimer est un essai d'agglutination au latex pour analyse semi-quantitative des D-dimères de fibrine. Les fragments contenant des D-dimères résultent de la dégradation par la plasmine de la fibrine stabilisée avec le facteur XIIIa. On trouve des concentrations élevées de D-dimères dans le cadre de pathologies cliniques du type thrombose veineuse profonde (TVP), embolie pulmonaire (EP) et coagulation intravasculaire disséminée (CIVD)[1,2]. Les taux de D-dimères augmentent pendant la grossesse et des taux élevés sont également observés en cas de complications[3]. Manual D-Dimer exploite l'effet d'un anticorps monoclonal qui rentre en réaction avec les D-dimères ou fragments D de la fibrine mais pas avec le fibrinogène intact[4], permettant ainsi d'analyser les D-dimères dans le plasma humain. On peut également utiliser des échantillons de sérum appropriés pour analyse PDF. Aucune norme internationale n'existe actuellement pour les D-dimères. Pour obtenir des valeurs numériques, Helena a aligné la méthode Manual D-Dimer sur la technique de dosage immunoenzymatique commercialisée (ELISA).

AVERTISSEMENTS ET PRÉCAUTIONS

Les réactifs du kit sont à usage diagnostique in vitro uniquement – NE PAS INGÉRER. Porter un équipement de protection individuelle approprié lors de la manipulation de tous les composants du kit. Consulter la fiche de données de sécurité du produit pour obtenir le lien vers les phrases de risque et les conseils de prudence le cas échéant. Éliminer les composants conformément aux réglementations locales.

Un dépistage des produits sanguins a été réalisé et a donné un résultat négatif (sauf indication contraire sur la boîte de kit ou sur le flacon) quant à la présence de :
Antigène de l'hépatite B (AgHBs)
Anticorps anti-VIH 1
Anticorps anti-VIH 2
Anticorps anti-VHC
Cependant, ils doivent être manipulés avec les mêmes précautions que celles prises pour les échantillons patients humains.

COMPOSITION

Composant	Contenu	Description	Préparation
Manual D-Dimer Latex	1 x 1.7 mL (REF 5250) 1 x 0.85 mL (REF 5250H)	Suspension de particules de latex. Les particules sont enduites d'anticorps monoclonaux.	Agitez le latex en renversant plusieurs fois la fiole pendant 5 secondes immédiatement avant utilisation. Si des amas de latex s'amoncellent dans le cou du flacon ou sont présents dans la suspension de latex, vortexer le flacon pendant 10 secondes pour s'assurer que le dépôt est remis en suspension.
Positive Control Plasma	1 x 1 mL	Plasma de contrôle positif. Plasma humain lyophilisé enrichi en D-dimères.	Reconstituer en ajoutant 1 mL de Saline Solution. Laisser reposer 20 minutes et bien mélanger avant utilisation (ne pas agiter).
Negative Control Plasma	1 x 1 mL	Plasma de contrôle négatif. Plasma humain lyophilisé.	Reconstituer en ajoutant 1 mL de Saline Solution. Laisser reposer 20 minutes et bien mélanger avant utilisation (ne pas agiter).
Saline Solution	2 x 8 mL (REF 5250) 1 x 8 mL (REF 5250H)	Tampon salin de pH 7,3. Contient 0,2 g/L d'azide de sodium.	Le réactif est prêt à l'emploi.
Test Cards	16 x 6 échantillon (REF 5250) 8 x 6 échantillon (REF 5250H)		
Mixing Sticks	50 (REF 5250) 25 (REF 5250H)		

Chaque kit contient une fiche technique.

MATÉRIEL NÉCESSAIRE NON FOURNI

1. Pipettes à 20, 100 et 1000 μL
2. Pointes de pipette
3. Tubes de dosage de 3 mL
4. Minuterie

CONSERVATION, DURÉE DE VIE UTILE ET STABILITÉ

Les réactifs doivent être conservés à $2-8°C$ et utilisés avant la date de péremption mentionnée sur la boîte. Une fois reconstitués, il est possible de conserver les plasmas de contrôle une semaine à $2-8°C$ ou un mois à -20 °C. Le bouchon en caoutchouc dans les fioles de contrôle doit être jeté après ouverture de la fiole.

PRÉLÈVEMENT ET PRÉPARATION DES ÉCHANTILLONS

Utiliser tout au long du prélèvement du plastique ou du verre siliconé. Mélangez 9 volumes de sang et 1 volume de citrate de sodium à 3,2% ou 3,8%. Séparer le plasma après centrifugation à 1500 x g pendant 15 minutes[5]. Conserver le plasma entre $2-8°C$ ou $18-24°C$. L'analyse doit être terminée dans les 24 heures suivant le prélèvement de l'échantillon, sinon, il est possible de congeler le plasma à -20°C ou -70°C à 24 mois[6]. Décongeler rapidement à 37°C avant de réaliser l'analyse. Ne pas laisser à 37°C plus de 5 minutes.

PROCÉDURE

Attendez que toutes les fioles se réchauffent à température ambiante au moins 10 minutes avant de les utiliser.

A. Méthode qualitative

1. Placez 20 μL d'échantillon et de plasmas contrôles positifs et négatifs en cercles sur une carte d'essai.
2. Placez 20 μL de Manual D-Dimer latex en suspension à proximité de chacun des cercles formés.
3. Mélangez rapidement l'échantillon et le Manual D-Dimer latex avec un bâtonnet propre pour chaque échantillon. Mettez la minuterie en marche.
4. Agitez doucement la carte d'essai et mesurez l'agglutination au bout de 180-200 secondes. Comparez les résultats positifs (+) et négatifs (-) avec les contrôles. Le contrôle positif est d'ordre purement qualitatif et une dilution supplémentaire est donc inutile. En l'absence d'agglutination du latex, l'échantillon est jugé normal et n'a donc pas besoin d'être testé de nouveau.

B. Méthode semi-quantitative

Effectuée sur les seuls échantillons au résultat positif.

1. Utilisez 100 μL d'échantillon pour effectuer une dilution en série de 1:2, 1:4 et 1:8 avec 100 μL de solution saline dans de petits tubes de dosage.
2. Marquez la position des dilutions sur la carte d'essai et mélangez avec le latex en suspension comme indiqué au Méthode qualitative.
3. La concentration en D-dimères pourra être établie par référence au tableau "Valeurs de Référence".

INTERPRÉTATION DES RÉSULTATS

1. L'agglutination intervient dans les 180-200 secondes pour les échantillons contenant plus de 250 ng/mL de D-dimères. Le taux moyen de D-dimères dans une population saine va de 8 à 135 ng/mL et le plasma pur provenant d'individus sains ne devrait donc pas s'agglutiner. Manual D-Dimer a une valeur prédictive négative élevée pour la thrombose[7]. La demi-vie des D-dimères dans le sang circulant est de 12 heures environ. Des taux élevés de D-dimères peuvent donc persister pendant un certain temps après cessation du processus actif. Des études cliniques portant sur des sujets sains, des patients avec un diagnostic de thrombose veineuse profonde confirmé par phlébographie, des patients avec CIVD et des patients avec prééclampsie (Pré-EC) ont mis en évidence les résultats suivants (les titres correspondent au taux le plus élevé de dilution auquel l'échantillon démontre des signes d'agglutination).

Échantillon	Nombre de patients	Négatif au test Manual D-Dimer	Dilution du plasma: Non dilué	1:2	1:4	1:8
Normal	101	100	1*	-	-	-
TVP	48	3	10*	7*	14*	14*
CIVD	29	0	3	3	4	19
Pré-EC	6	2	1	3	-	-

* L'agglutination a été inhibée par addition d'anticorps spécifiquement dirigés contre les D-dimères (0,2 mg/mL), mais l'ajout de l'anticorps PAM-1 indépendant est resté sans effet.

2. Les résultats peuvent être exprimés soit en D-dimères, soit en unités équivalent fibrinogène (UEF). 1 ng/mL de D-dimères équivaut à environ 2 ng/mL de UEF[6].
3. L'agglutination peut être plus prononcée et plus rapide pour des concentrations de D-dimères assez élevées.

Si vous avez besoin d'une détermination quantitative des D-dimères, Helena Biosciences Europe fournit les kits suivants à utiliser avec les instruments photo-optiques :

REF 5551 / 5552 / 5554	Auto Blue D-Dimer 400
REF 5601 / 5501	Auto Red D-Dimer 700

LIMITES

1. Un taux négatif en Manual D-Dimer n'élimine pas totalement l'hypothèse de thrombose. La trousse d'analyse Manual D-Dimer a donné une valeur prédictive négative de 94% pour des patients avec suspicion de thrombose veineuse profonde[8]. La détection de taux élevés de D-dimères doit donc être confirmée par d'autres données cliniques pour arriver à un diagnostic.
2. Un phénomène d'agglutination des échantillons contenant des taux normaux de D-dimères peut occasionnellement être observé en raison de la non-spécificité de la réaction.
3. Un petit nombre d'échantillons peuvent, après mélange avec le latex, présenter des flocons blancs qui ne doivent toutefois pas être confondus avec un phénomène d'agglutination.

CONTRÔLE QUALITÉ

Les contrôles positifs et négatifs fournis doivent être utilisés pour contrôle de la qualité de chaque trousse. Il est recommandé de tester les contrôles positifs et négatifs à chaque nouvelle utilisation de la trousse. Si l'un des contrôles (positif ou négatif) ne fournit pas la réponse attendue, les résultats obtenus pour ce test ne doivent pas être exploités et la trousse utilisée doit être délaissée ou renvoyée à Helena Biosciences Europe.

VALEURS DE RÉFÉRENCE

L'agglutination intervient dans les 180-200 secondes pour les échantillons contenant plus de 250 ng/mL de D-dimères. Une dilution du plasma en série permet d'obtenir des résultats semi-quantitatifs.

Taux de D-dimères (ng/mL) ng/mL	Non dilué	1:2	1:4	1:8
< 250	-	-	-	-
250-500	+	-	-	-
500-1000	+	+	-	-
1000-2000	+	+	+	-
> 2000	+	+	+	+

L'agglutination peut être plus prononcée et plus rapide pour des concentrations de D-dimères assez élevées. Si vous souhaitez exprimer les résultats en unités équivalent fibrinogène (UEF), multipliez par 2 les taux de D-dimères du tableau ci-dessus, par exemple, <250 ng/mL devient <500 UEF. Différents rapports[7,8] ont montré que certaines analyses commerciales au latex n'ont pas la sensibilité alléguée par rapport à la technique ELISA commercialisée.

CARACTÉRISTIQUES DE PERFORMANCES

Spécificité

L'anticorps monoclonal utilisé dans cet appareil est spécifiquement dirigé contre les D-dimères en raison de la méthode de dépistage employée pour la sélection d'hybridomes[4]. Un hybridome sécrétant des anticorps réagissant positivement aux D-dimères purifiés mais non au fibrinogène intègre ou au fragment D du fibrinogène a été sélectionné. Aucune forme de réactivité croisée avec le fibrinogène ou le monomère de fibrine de type I n'a été observée lorsque l'un ou l'autre des échantillons à analyser a été substitué au plasma dans cet essai. Le plasma de 16 patients atteints de polyarthrite rhumatoïde a été analysé et 14 de ces échantillons n'ont montré aucun signe d'agglutination avec le test Manual D-Dimer. Les deux cas d'agglutination ont pu être inhibés par addition de l'anticorps monoclonal spécifiquement dirigé contre les D-dimères, mais l'ajout d'un monoclonal du même sous-groupe, IgG1k (PAM-1) est resté sans effet. Ce résultat laisse à penser que Manual D-Dimer n'est pas sensible aux troubles liés au facteur rhumatoïde.

Reproductibilité

Trois échantillons de plasma ont été sélectionnés pour un test de reproductibilité. Chaque échantillon a été testé 10 fois par jour sur trois jours. Dans chaque cas, en cas de résultat positif, l'échantillon a été titré. Voici les résultats en D-dimères (ng/mL) :

Reproductibilité

Échantillon	Taux de D-dimères	Résultat
Normal	< 250 ng/mL	Négatif
Intermédiaire	2000 ng/mL	Titre 1:8
Haut	> 16,000 ng/mL	Titre 1:64

Exactitude

La trousse Manual D-Dimer a été comparée à une autre méthode d'analyse des D-dimères au latex parmi celles disponibles en commerce. Les deux produits ont donné une réaction négative suite à l'analyse de 25 specimens normaux. Lors de l'analyse de 30 échantillons de plasma de patients avec ELISA et le test Manual D-Dimer, tous les échantillons dépassant les 225 ng/mL avec ELISA ont montré des signes d'agglutination avec Manual D-Dimer.

BIBLIOGRAPHIE

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2. Declerck PV *et al.* (1987) Fibrinolytic response and fibrin fragment D dimer in patients with deep vein thrombosis. *Thromb. Haemostas.* 58:1025-1029
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6. Clinical and Laboratory Standards Institute (2011) Quantitative D-dimer for the Exclusion of Venous Thromboembolic Disease: Approved Guideline. CLSI: H59-A
7. Hansson PO *et al.* (1994) Can laboratory testing improve screening strategies for deep vein thrombosis at an emergency unit? *J. Intern. Med.* 235:143-151
8. Bounameaux H *et al.* (1989) Measurement of plasma D-dimer for diagnosis of deep venous thrombosis. *Am. J Clin. Path.* 91:82-85

Manual D-Dimer
Anleitung

de

VERWENDUNGSZWECK

Das Manual D-Dimer Kit ist für immunturbidimetrische Bestimmungen vorgesehen.

Manual D-Dimer ist ein Latex-Agglutinationstest für den semiquantitativen Nachweis von Fibrin D-Dimeren. [illegible] Entstehen Fibrin, das durch den Faktor XIIIa quervernetzt ist, entstehen D-dimer enthaltende Spaltprodukte. [illegible] D-dimer werden bei klinischen Zuständen wie tiefer Venenthrombose (TVT), Pulmonalembolie (PE) und disseminierter intravasaler Koagulation (DIC)[1,2] beobachtet. D-dimer Werte steigen während der Schwangerschaft an [illegible][3]. Manual D-Dimer bedient sich eines monoklonalen Antikörpers, der mit dem Fibrin D-Dimer oder dem Fibrin Fragment D, nicht aber mit intaktem Fibrinogen reagiert[4], und dadurch den Nachweis des D-dimers im Humanplasma ermöglicht. [illegible] für die FDP-Analyse geeignete Serum-Proben herangezogen werden. Gegenwärtig gibt es keinen internationalen Standard für D-dimere. Für numerische Werte hat Helena das Manual D-Dimer an im Handel erhältlichen [illegible] immunosorbent Assay (ELISA)-Tests angeglichen.

WARNHINWEISE UND VORSICHTSMASSNAHMEN

Die in diesem Kit enthaltenen Reagenzien sind ausschließlich für die Verwendung zur in-vitro-Diagnostik vorgesehen. NICHT VERSCHLUCKEN. Tragen Sie beim Umgang mit sämtlichen Komponenten des Kits geeignete Schutzausrüstung. [illegible] Entsorgen Sie die Komponenten gemäß den örtlichen Vorschriften.

Die Blutprodukte wurden untersucht und sind für folgende Stoffe ohne Befund (sofern nicht anderweitig auf der Verpackung oder den Ampullen angegeben):
Hepatitis-B-Antikörper (HbsAg)
HIV-Antikörper 1
HIV-Antikörper 2
HCV-Antikörper
Sie sind jedoch mit den gleichen Vorkehrungen zu behandeln wie Proben von menschlichen Patienten.

ZUSAMMENSETZUNG

Komponente	Inhalt	Beschreibung	Vorbereitung
Manual D-Dimer Latex	1 x 1,7 mL (REF 5250) 1 x 0,85 mL (REF 5250H)	Suspension aus Latexbeads. Die Beads sind mit dem Anti-D-dimer monoklonalen Antikörper [illegible]	Mischen Sie das Latex, indem Sie das Fläschchen unmittelbar vor dem Gebrauch wiederholt für jeweils 5 Sekunden umdrehen. [illegible]
Positive Control Plasma	1 x 1 mL	Positives Kontroll-Plasma. Lyophilisiertes, mit dem Fibrin D-dimer angereichertes Humanplasma.	Mit 1 mL Saline Solution rekonstituieren. 20 Minuten stehen lassen und vor Gebrauch gut mischen, aber nicht schütteln.
Negative Control Plasma	1 x 1 mL	Negatives Kontroll-Plasma. Lyophilisiertes Humanplasma.	Mit 1 mL Saline Solution rekonstituieren. 20 Minuten stehen lassen und vor Gebrauch gut mischen, aber nicht schütteln.
Saline Solution	2 x 8 mL (REF 5250) 1 x 8 mL (REF 5250H)	Gepufferte Kochsalzlösung mit einem pH von 7,3. Enthält 0,2 g/L Natriumazid.	Das Reagenz ist gebrauchsfertig verpackt.
Test Cards	16 x 6 probe (REF 5250) 8 x 6 probe (REF 5250H)		
Mixing Sticks	50 (REF 5250) 25 (REF 5250H)		

Jedes Kit enthält eine Gebrauchsanweisung.

ERFORDERLICHE, ABER NICHT MITGELIEFERTE ARTIKEL

1. Pipetten für 20, 100 und 1000 μL
2. Volumina Pipettenspitzen
3. 3 mL Reagenzgläser
4. Timer

LAGERUNG, HALTBARKEIT UND STABILITÄT

Die Reagenzien sollten bei $2-8°C$ gelagert und vor Ablauf des aufgedruckten Verfallsdatums verwendet werden. [illegible]

PROBENENTNAHME UND VORBEREITUNG

Nur Plastik oder Silikonglas verwenden. Blut (9 Teile) sollte in 3,2% oder 3,8% Natriumcitrat als Antikoagulans (1 Teil) [illegible]. 15 Minuten bei 1500 g zentrifugieren und Plasma abpipettieren[5]. [illegible]

VORGEHENSWEISE

Lassen Sie alle Fläschchen, vor dem Gebrauch für mindestens 10 Minuten auf Raumtemperatur kommen.

A. Qualitative Methode

1. Tragen Sie jeweils 20 μL der Probe sowie der positiven und negativen Kontrollplasmen [illegible]
2. Tragen Sie unmittelbar neben jeden Kreis jeweils 20 μL der Manual D-Dimer Latex-Suspension auf.
3. Vermischen Sie Proben und Manual D-Dimer Latex rasch mit sauberen Mischstäbchen [illegible] Timer.
4. Schütteln Sie die Testkärtchen vorsichtig und lesen Sie die Agglutination zwischen 180 und 200 Sekunden ab. [illegible]

B. Semiquantitativen Methode

Wird nur an positiv getesteten Proben ausgeführt.

1. Verdünnen Sie 100 μL der Probe seriell 1:2, 1:4 und 1:8 mit 100 μL Kochsalzlösung in kleinen Reagenzgläsern.
2. Markieren Sie die Positionen der Probenverdünnungen auf dem Testkärtchen und mischen Sie mit der Latex-Suspension wie Qualitative Methode.
3. Die D-Dimer Konzentration kann aus der Tabelle in "Referenzwerte" bestimmt werden.

INTERPRETATION DER ERGEBNISSE

1. Bei Proben mit mehr als 250 ng/mL D-dimer tritt die Agglutination innerhalb von 180 - 200 Sekunden ein. [illegible]

Proben	Patientenanzahlen	Negative D-Dimer Resultate	Plasmaverdünnung: Unverdünnt	1:2	1:4	1:8
Normal	101	100	1*	-	-	-
DVT	48	3	10*	7*	14*	14*
DIC	29	0	3	3	4	19
Pre-EC	6	2	1	3	-	-

* Die Agglutination wurde durch die Zugabe eines D-dimer spezifischen Antikörpers (0,2 mg/mL) gehemmt, jedoch nicht durch den nicht-verwandten Antikörper PAM-1.

2. Die Ergebnisse können entweder in D-dimer Einheiten oder als Fibrinogen Equivalent Units (FEU) angegeben werden. 1 ng/mL D-dimer entspricht in etwa 2 ng/mL FEU[6].
3. Bei höheren D-Dimer Konzentrationen tritt die Agglutination deutlicher hervor und setzt schneller ein.

Falls Sie eine quantitative D-Dimer-Analyse benötigen, bietet Helena Biosciences Europe die folgenden Kits zur Verwendung auf fotooptischen Geräten an:

REF 5551 / 5552 / 5554	Auto Blue D-Dimer 400
REF 5601 / 5501	Auto Red D-Dimer 700

EINSCHRÄNKUNGEN

1. Ein negativer Manual D-Dimer Test schließt Thrombosen nicht vollständig aus. [illegible]
2. Eine Agglutination von Proben mit normalen D-dimer Werten kann [illegible]
3. Bei einigen wenigen Proben kann es beim Mischen mit Latex zu weißen Ausflockungen kommen, [illegible] Agglutination verwechselt werden sollten.

QUALITÄTSKONTROLLE

Die mitgelieferten Positiv- und Negativkontrollen sollten für die Qualitätskontrolle jedes Kits herangezogen werden. [illegible]

REFERENZWERTE

Bei Proben mit mehr als 250 ng/mL D-Dimer tritt die Agglutination innerhalb von 180 bis 200 Sekunden ein. [illegible] verdünnten Plasmen können semiquantitative Ergebnisse erzielt werden.

D-Dimer Wert (ng/mL) ng/mL	Unverdünnt	1:2	1:4	1:8
< 250	-	-	-	-
250-500	+	-	-	-
500-1000	+	+	-	-
1000-2000	+	+	+	-
> 2000	+	+	+	+

Bei höheren D-dimer Konzentrationen tritt die Agglutination deutlicher hervor und setzt schneller ein. [illegible] <500 FEU. Wie aus mehreren Berichten ersichtlich[7,8], [illegible] ELISA nicht die entsprechende Sensitivität auf.

LEISTUNGSMERKMALE

Spezifität

[illegible]

Reproduzierbarkeit

[illegible]

Reproduzierbarkeit

Probe	D-Dimer Wert	Ergebnis
Normal	< 250 ng/mL	Negativ
Mäßig	2000 ng/mL	Titer 1:8
Hoch	> 16,000 ng/mL	Titer 1:64

Genauigkeit

Der Manual D-Dimer Kit wurde mit einem anderen handelsüblichen erhältlichen D-dimer Latex verglichen. Beide Produkte ergaben eine negative Reaktion im Test bei 25 normalen Proben. Beim Testen von Plasmaproben von 30 Patienten mittels ELISA und Manual D-Dimer zeigten alle Proben mit ELISA Werten über 225 ng/mL eine Agglutination mit dem Manual D-Dimer.

LITERATURVERZEICHNIS

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2. Declerck PV *et al.* (1987) Fibrinolytic response and fibrin fragment D dimer in patients with deep vein thrombosis. *Thromb. Haemostas.* 58:1025-1029
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Manual D-Dimer
Istruzioni per l'uso

it

SCOPO PREVISTO

Il kit Manual D-Dimer è concepito per l'esecuzione di dosaggi di emostasi mediante test immunoturbidimetrico.

Manual D-Dimer è un test di agglutinazione del lattice per la determinazione semiquantitativa del D-dimero della fibrina. [illegible] Livelli elevati di D-dimero sono stati riscontrati in condizioni cliniche quali la trombosi venosa profonda (DVT), l'embolia polmonare (PE) e la coagulazione intravascolare disseminata (DIC)[1,2]. I livelli di D-dimero aumentano durante la gravidanza e livelli molto elevati sono associati a complicazioni[3]. [illegible] nel plasma umano. È possibile utilizzare anche campioni di siero adatti all'analisi FDP. Non esistono attualmente standard internazionali per il D-dimero. Per i valori numerici Helena ha allineato Manual D-Dimer a test con immunosorbente legato all'enzima (ELISA) disponibili in commercio.

AVVERTENZE E PRECAUZIONI

I reagenti contenuti in questo kit sono destinati esclusivamente alla diagnostica *in vitro* - NON INGERIRE. Indossare un'adeguata attrezzatura protettiva personale durante la manipolazione di tutti i componenti del kit. Per conoscere i relativi simboli precauzionali [illegible] alla dichiarazione di sicurezza del prodotto. Smaltire i componenti conformemente alle normative locali vigenti.

I prodotti emàtici sono stati sottoposti a screening e trovati negativi (salvo diversa indicazione sulla confezione del kit o sulla fiala) per la presenza di:
Antigene dell'epatite B (HBsAg)
Anticorpo HIV 1
Anticorpo HIV 2
Anticorpo HCV
Questi prodotti devono tuttavia essere manipolati con le stesse misure precauzionali adottate per un campione paziente umano.

COMPOSIZIONE

Componente	Contiene	Descrizione	Preparazione
Manual D-Dimer Latex	1 x 1,7 mL (REF 5250) 1 x 0,85 mL (REF 5250H)	Sospensione di sfere di lattice. Le sfere sono ricoperte di anticorpo monoclonale.	Immediatamente prima dell'uso, agitare il lattice per 5 secondi capovolgendo ripetutamente il vial. Se la massa di lattice si raccoglie intorno al collo della fiala oppure è visibile nella sospensione, agitare la fiala su vortex per 10 secondi per assicurarsi che nessun materiale sia risospeso.
Positive Control Plasma	1 x 1 mL	Plasma di controllo positivo. Plasma umano liofilizzato arricchito con D-dimero della fibrina.	Ricostituire con 1 mL di Saline Solution. Lasciare riposare per 20 minuti e miscelare accuratamente prima dell'uso, non scuotere.
Negative Control Plasma	1 x 1 mL	Plasma di controllo negativo. Plasma umano liofilizzato.	Ricostituire con 1 mL di Saline Solution. Lasciare riposare per 20 minuti e miscelare accuratamente prima dell'uso, non scuotere.
Saline Solution	2 x 8 mL (REF 5250) 1 x 8 mL (REF 5250H)	Soluzione tampone a pH 7,3. Contiene 0,2 g/L di sodio azide.	Il reagente è in confezione pronta all'uso.
Test Cards	16 x 6 campione (REF 5250) 8 x 6 campione (REF 5250H)		
Mixing Sticks	50 (REF 5250) 25 (REF 5250H)		

Ogni kit contiene un Istruzioni per l'uso.

MATERIALI NECESSARI, MA NON IN DOTAZIONE

1. Pipette da 20, 100 e 1000 µL
2. Puntali per pipette
3. Provette da 3 mL
4. Cronometro

CONSERVAZIONE, VITA UTILE E STABILITÀ

I reagenti devono essere conservati a una temperatura compresa tra "2 – "8°C ed utilizzati prima della data di scadenza riportata sulla confezione. I plasmi di controllo, dopo la ricostituzione, devono essere a "2 – "8°C per una settimana o un mese a -20°C. Il tappo di gomma dei vial dei controlli può essere gettato dopo l'apertura del vial.

RACCOLTA E PREPARAZIONE DEI CAMPIONI

Nel corso dell'intera procedura è necessario utilizzare plastica o vetro siliconizzato. Il sangue (9 parti) deve essere raccolto in sodio citrato al 3,2% o al 3,8% come anticoagulante (1 parte). Separare il plasma in seguito a centrifugazione a 1500 x g per 15 minuti[5]. [illegible]

PROCEDURA

[illegible]

A. Metodo qualitativo

[illegible]

B. Metodo semiquantitativo

[illegible]

INTERPRETAZIONE DEI RISULTATI

[illegible]

Campione	Numero di pazienti	Risultato negativo di D-Dimer	Diluizione del plasma			
			Non diluito	1:2	1:4	1:8
Normale	101	100	1*	-	-	-
DVT	48	3	10*	7*	14*	14*
DIC	29	0	3	3	4	19
Pre-EC	6	2	1	3	-	-

[illegible]

REF 5551 / 5552 / 5554 — Auto Blue D-Dimer 400
REF 5601 / 5501 — Auto Red D-Dimer 700

LIMITAZIONI

[illegible]

CONTROLLO QUALITÀ

[illegible]

VALORI DI RIFERIMENTO

[illegible]

Livello di D-dimero (ng/mL) ng/mL	Non diluito	1:2	1:4	1:8
< 250	[illegible]	[illegible]	[illegible]	[illegible]
250-500	[illegible]	[illegible]	[illegible]	[illegible]
500-1000	[illegible]	[illegible]	[illegible]	[illegible]
1000-2000	[illegible]	[illegible]	[illegible]	[illegible]
> 2000	[illegible]	[illegible]	[illegible]	[illegible]

[illegible]

CARATTERISTICHE PRESTAZIONALI

Specificità

[illegible]

Riproducibilità

[illegible]

Campione	Livello di D-dimero	Risultato
Normale	< 250 ng/mL	Negativo
Intermedio	3000 ng/mL	Titolo 1:8
Superiore	> 16.000 ng/mL	Titolo 1:64

Accuratezza

[illegible]

BIBLIOGRAFIA

[illegible]

Manual D-Dimer
Instrucciones de uso

es

USO PREVISTO

El uso previsto del kit Manual D-Dimer es realizar ensayos de hemostasia basados en la inmunoturbidimetría.

Manual D-Dimer es un ensayo de aglutinación por látex para la determinación semicuantitativa del dímero D de la fibrina. Los fragmentos que contienen dímero D se forman por la acción de la plasmina al degradar la fibrina estabilizada por el factor XIIIa. Niveles elevados de dímero D pueden observarse en patologías clínicas tales como la trombosis venosa profunda (DVT), el embolismo pulmonar (PE) y la coagulación intravascular diseminada (DIC)[1,2]. Los niveles de dímero D aumentan durante el embarazo, estando asociados los niveles altos a complicaciones[3]. En Manual D-Dimer se utiliza un anticuerpo monoclonal que reacciona con el dímero D o el fragmento D de la fibrina pero no con el fibrinógeno[4], lo que permite la determinación del dímero D en el plasma humano. También pueden utilizarse muestras de suero adecuadas para el análisis FDP. Actualmente no existe un patrón internacional para el dímero D. En cuanto a los valores numéricos, Helena ha ajustado Helena D dimer a los ensayos inmunoabsorbentes ligados a enzimas (ELISA) disponibles en el mercado.

ADVERTENCIAS Y PRECAUCIONES

Los reactivos que contiene este kit son sólo para uso de diagnóstico *in vitro*. NO INGERIR. Lleve el equipo de protección personal adecuado cuando utilice todos los componentes del kit. Consulte la declaración de seguridad del producto para saber más sobre las indicaciones adecuadas de advertencia y riesgo. Desechar los componentes de conformidad con las normativas locales.

La sangre se ha sometido a pruebas que han resultado negativas (a menos que se indique lo contrario en la caja del kit o en el vial) de la presencia de:
Antígeno de la hepatitis B (HBsAg)
Anticuerpos del VIH 1
Anticuerpos del VIH 2
Anticuerpos del VHC
Sin embargo, deben manipularse con las mismas precauciones que una muestra de un paciente.

COMPOSICIÓN

Componente	Contiene	Descripción	Preparación
Manual D-Dimer Latex	1 x 1,7 mL (REF 5250) 1 x 0,85 mL (REF 5250H)	Suspensión de perlas de látex. Las perlas están recubiertas con anticuerpo monoclonal anti-dímero-D.	Agitar el látex invirtiendo repetidamente el vial durante 5 segundos justo antes de usarlo. Si la masa de látex se acumula en el cuello del vial o es visible en la suspensión de látex, agite el vial en vortex durante 10 segundos para asegurarse de que ningún material sedimentario quede resuspendido.
Positive Control Plasma	1 x 1 mL	Plasma de control positivo. Plasma humano liofilizado enriquecido con dímero D de fibrina.	Reconstituir con 1 mL de Saline Solution. Dejar reposar durante 20 minutos y mezclar bien antes de usar, no agitar.
Negative Control Plasma	1 x 1 mL	Plasma de control negativo. Plasma humano liofilizado.	Reconstituir con 1 mL de Saline Solution. Dejar reposar durante 20 minutos y mezclar bien antes de usar, no agitar.
Saline Solution	2 x 8 mL (REF 5250) 1 x 8 mL (REF 5250H)	Solución salina tamponada, pH 7,3, con 0,2 g/L de azida sódica.	El reagente está listo para ser utilizado directamente del paquete.
Test Cards	16 x 6 muestra (REF 5250) 8 x 6 muestra (REF 5250H)		
Mixing Sticks	50 (REF 5250) 25 (REF 5250H)		

Cada kit contiene instrucciones de uso.

ARTÍCULOS NECESARIOS NO SUMINISTRADOS

1. Pipetas para 20, 100 y 1000 µL
2. Puntas de pipeta
3. Tubos de ensayo de 3 mL
4. Temporizador

ALMACENAMIENTO, CADUCIDAD Y ESTABILIDAD

Los reactivos deben conservarse a una temperatura entre "2 – "8°C y ser utilizados antes de la fecha de caducidad que figura en el envase. Plasmas control, una vez reconstituidos pueden conservarse a entre "2 – "8°C durante una semana o un mes a -20°C. Tras la apertura de los viales de control puede desecharse el tapón de caucho.

RECOGIDA Y PREPARACIÓN DE LAS MUESTRAS

Debe usarse siempre plástico o vidrio siliconizado. Debe recogerse sangre (9 partes) en el anticoagulante citrato sódico al 3,2% o al 3,8% (1 parte). Separar el plasma después de la centrifugación a 1500 x g durante 15 minutos[5]. El plasma debe conservarse a "2 – "8°C o "18 – "24°C. Las pruebas deberían terminarse en 24 horas desde la recogida de las muestras o el plasma puede conservarse congelado a -20°C o -70°C durante 24 mes[6]. Descongelar rápidamente a "37°C antes de realizar la prueba. No conservar a "37°C durante más de 5 minutos.

PROCEDIMIENTO

Antes de su uso deberá esperarse a que los viales alcancen la temperatura ambiente (mín. 10 minutos).

A. Método cualitativo

1. Aplicar en círculos 20 µL de los plasmas de muestra, de control positivo y de control negativo sobre una tarjeta de ensayo.
2. Aplicar 20 µL de la suspensión de Manual D-Dimer Latex en las proximidades de cada círculo.
3. Mezclar rápidamente la muestra y el Manual D-Dimer Latex utilizando un palillo de mezclar limpio para cada muestra. Poner en marcha el temporizador.
4. Balancear suavemente la tarjeta de ensayo y leer la aglutinación al cabo de 180 - 200 segundos. La aglutinación positiva (+) o negativa (-) se compara con los resultados obtenidos con los controles. El uso del control positivo es puramente cualitativo y no debe diferir nada. Un látex no aglutinado significa que la muestra es normal, por lo que no es necesario realizar más ensayos.

B. Método semicuantitativo

Para realizar titulaciones en muestras que han dado positivo:

1. Realizar diluciones seriadas de 100 µL de la muestra a 1:2, 1:4 y 1:8 con 100 µL de solución salina utilizando tubos de ensayo pequeños.
2. Mezclar los reactivos de las diluciones de la muestra en la tarjeta de ensayo y mezclar con suspensión de látex de acuerdo con el Método cualitativo.
3. La concentración de dímero D puede determinarse con la tabla de la sección "Valores de Referencia".

INTERPRETACIÓN DE LOS RESULTADOS

1. La aglutinación se produce al cabo de 180 - 200 segundos en aquellas muestras que contienen más de 250 ng/mL de dímero D. El nivel medio de dímero D en una población sana se encuentra entre 8 y 135 ng/mL aprox. y el plasma puro de sujetos normales sanos no debe aglutinarse. El valor negativo predictivo de Manual D-Dimer para la trombosis es elevado[7]. La semivida circulatoria del dímero D es de aproximadamente 12 horas. Por ello los niveles elevados de dímero D pueden persistir durante algún tiempo después de cesar el proceso activo. En estudios clínicos con sujetos normales, pacientes con DVT confirmada flebográficamente, pacientes con DIC y pacientes con pre-eclampsia (Pre-EC) se obtuvieron los siguientes resultados; los títulos indican la máxima dilución a la cual la muestra presenta aglutinación.

Muestra	Número de pacientes	Resultado negativo con D-Dimer	Dilución del plasma			
			Sin diluir	1:2	1:4	1:8
Normal	101	100	1*	-	-	-
DVT	48	3	10*	7*	14*	14*
DIC	29	0	3	3	4	19
Pre-EC	6	2	1	3	-	-

* La aglutinación se vio inhibida por la adición del anticuerpo específico del dímero D (0,2 mg/mL) pero no por la de un anticuerpo PAM-1 no relacionado.

2. Los resultados pueden expresarse en unidades de dímero D o en unidades equivalentes de fibrinógeno (FEU). 1 ng/mL de dímero D equivale a aprox. 2 ng/mL de FEU[8].
3. La aglutinación puede ser más pronunciada y aparece más rápidamente con concentraciones mayores de dímero D.

Si necesita un análisis cuantitativo de D-Dímero, Helena Biosciences Europe ofrece los siguientes kits para uso en instrumentos foto-ópticos:

REF 5551 / 5552 / 5554 — Auto Blue D-Dimer 400
REF 5601 / 5501 — Auto Red D-Dimer 700

LIMITACIONES

1. Un resultado de Manual D-Dimer negativo no excluye totalmente una trombosis. Con el kit Manual D-Dimer el valor predictivo negativo para pacientes con sospecha de DVT obtenido ha sido del 94%[7]. Para establecer un diagnóstico deberá tenerse en cuenta la detección de niveles elevados de dímero D junto con el resto de la información clínica.
2. A veces puede producirse una aglutinación en muestras que contienen niveles de dímero D normales causada por una inespecificidad.
3. Algunas muestras cuando son mezcladas con látex pueden presentar copos blancos que no deben confundirse con una aglutinación.

CONTROL DE CALIDAD

Para el control de calidad de cada kit se utilizarán los controles positivo y negativo suministrados. Se recomienda valorar tanto el control positivo como el negativo cada vez que se utiliza el kit. Si el control positivo o el negativo no proporciona una respuesta adecuada, deberán descartarse los resultados del paciente obtenidos en esa ocasión y el kit deberá ser eliminado o devuelto a Helena Biosciences Europe.

VALORES DE REFERENCIA

La aglutinación se produce a los 180 - 200 segundos en aquellas muestras que contienen más de 250 ng/mL de dímero D. Si se analizan plasmas diluidos en serie, pueden obtenerse resultados semicuantitativos.

Nivel de dímero D (ng/mL) ng/mL	Sin diluir	1:2	1:4	1:8
< 250	-	-	-	-
250-500	+	-	-	-
500-1000	+	+	-	-
1000-2000	+	+	+	-
> 2000	+	+	+	+

La aglutinación puede ser más pronunciada y aparece más rápidamente con concentraciones mayores de dímero D. Si desea expresar los resultados en unidades equivalentes de Fibrinógeno (FEU) deberá multiplicar por 2 los niveles de dímero D de la tabla anterior, p.ej., <250 ng/mL = <500 FEU. Como se ha debatido en varios informes[6,8], algunos ensayos de látex comerciales no presentan la sensibilidad declarada cuando se comparan con el método ELISA comercial.

CARACTERÍSTICAS FUNCIONALES

Specificità

L'anticorpo monoclonale utilizzato in questo dispositivo è specifico per il D-dimero in virtù del metodo di screening utilizzato per la selezione dell'ibridoma[4]. È stato selezionato un ibridoma che secerne anticorpi che reagiscono con il D-dimero purificato ma non con l'intero fibrinogeno o con il frammento D del fibrinogeno. [illegible] aggiungendo l'anticorpo monoclonale specifico del D-dimero ma non un monoclonale dello stesso sottogruppo, IgG1k (PAM-1). Ciò suggerisce che il Manual D-Dimer non è sensibile ai disturbi provocati dal fattore reumatoide.

Riproducibilità

Per verificare la riproducibilità del test, sono stati selezionati tre campioni di plasma. Ogni campione è stato testato 10 volte al giorno, per 3 giorni. In ciascun caso, quando è stato ottenuto un risultato positivo, il campione è stato titolato. Di seguito sono riportati i risultati in D-dimero (ng/mL).

Reproducibilidad

Muestra	Nivel de dímero D	Resultado
Normale	< 250 ng/mL	Negativo
Intermedio	3000 ng/mL	Titolo 1:8
Superiore	> 16,000 ng/mL	Titolo 1:64

Accuratezza

Il kit Manual D-Dimer è stato confrontato con un altro lattice D-dimero disponibile in commercio. Entrambi i prodotti hanno dato una reazione negativa quando testati su 25 campioni normali. Testando con ELISA e Manual D-Dimer 30 campioni di plasma da paziente, tutti i campioni aventi valori ELISA maggiori di 225 ng/mL hanno mostrato agglutinazione con Manual D-Dimer.

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Тест-система "Д-димер, ручной метод"
инструкция

ru

НАЗНАЧЕНИЕ

[illegible]

ПРЕДУПРЕЖДЕНИЯ И МЕРЫ ПРЕДОСТОРОЖНОСТИ

[illegible]

Антитела к вирусу гепатита С (HCV)
[illegible]

СОСТАВ

Компоненты	Состав набора	Описание	Приготовление реагентов
Д-димер латексный реагент	1 x 1,7 мл (Кат. № 5250) 1 x 0,85 мл (Кат. № 5250H)	[illegible]	[illegible]
Д-димер позитивный контроль	1 x 1 мл	Лиофилизированная обогащенная Д-димером плазма человека.	Добавьте 1,0 мл солевого буфера во флакон с контрольной плазмой. Осторожно перемешайте и оставьте при комнатной температуре в течение 20 минут, перемешайте [illegible] перед использованием.
Д-димер негативный контроль	1 x 1 мл	Лиофилизированная плазма человека.	Добавьте 1,0 мл солевого буфера во флакон с контрольной плазмой. Осторожно перемешайте и оставьте [illegible]
Буфер солевой	2 x 8 мл (Кат. № 5250) 1 x 8 мл (Кат. № 5250H)	Солевой буфер, pH 7,3. Содержит 0,2 г/л азида натрия.	Реагент поставляется в готовом для использования виде.
Карты тестовые	16 x 6 образец (Кат. № 5250) 8 x 6 образец (Кат. № 5250H)		
Палочки перемешивающие	50 (Кат. № 5250) 25 (Кат. № 5250H)		

Каждый набор содержит инструкцию по применению.

НЕОБХОДИМЫЕ КОМПОНЕНТЫ, НЕ ВКЛЮЧЕННЫЕ В КОМПЛЕКТ ПОСТАВКИ

1. Пипетка на 20, 100 и 1000 мкл
2. Наконечники для пипеток
3. Пробирки объемом 3 мл
4. Секундомер

ХРАНЕНИЕ, СРОК ГОДНОСТИ И УСТОЙЧИВОСТЬ

[illegible]

ОТБОР И ПОДГОТОВКА ОБРАЗЦОВ

[illegible]

ПРОЦЕДУРА

[illegible]

ИНТЕРПРЕТАЦИЯ РЕЗУЛЬТАТОВ

[illegible]

ОГРАНИЧЕНИЯ

[illegible]

КОНТРОЛЬ КАЧЕСТВА

[illegible]

НОРМАЛЬНЫЕ ПОКАЗАТЕЛИ

[illegible]

ЭКСПЛУАТАЦИОННЫЕ ХАРАКТЕРИСТИКИ

[illegible]

ЛИТЕРАТУРА

[illegible]

Declaration of Conformity

HL-7-0664DC DOI 2015/08 (1)

In Application of the Council Directive 98/79/EC on the approximation of the laws of the Member States relating to *In Vitro* Diagnostic Medical Devices & CE marking.

Declaration of conformance to applicable sections of Annex I - Essential Requirements and Annex III (EC Declaration of Conformity) imposed by sections 2 to 5. The below listed products are not classified under Annex II Lists A or B. Access to the appropriate technical files will be made available to the appropriate body in the event this is required.

Product Code	Description	GMDN Classification Code
5267L	Thromboplastin L	55983

I, the undersigned declare that the devices registered against the above GMDN Classification Code conforms to the said Directives.

Full Name: M.J. Stephenson Title: Managing Director

Signed: Date: 06 Aug 2015

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Thromboplastin L

REF 5265HL
REF 5265L
REF 5267L

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HL-2-3035P 2015/10 (1)

Thromboplastin L
Instructions for use

en

INTENDED PURPOSE

The Thromboplastin L kit is intended for carrying out clot based haemostasis assays.

The first standardised one-stage prothrombin time test was developed by Dr. Armand Quick in 1935. It has now become the basic coagulation screening test for the diagnosis of congenital and acquired deficiencies of clotting factors from the extrinsic pathway (factors II, V, VII and X)[1,2]. It is also used for the induction and monitoring of oral anticoagulant therapy[1,2] and can be used to assess the protein synthesis capability of the liver in chronic or acute hepatic disorders.
Thromboplastin L is of rabbit brain origin but resembles human preparations in its low International Sensitivity Index (ISI). The ISI of Thromboplastin L is approximately 1.1 and is calibrated against the WHO international reference preparation[3]. Thromboplastin L is particularly suited to the monitoring of oral anticoagulant therapy and, in conjunction with the appropriate factor deficient plasma, the measurement of factor activity in the extrinsic pathway. Tissue thromboplastin, in the presence of calcium ions, is an activator which initiates the extrinsic pathway of coagulation. When a mixture of tissue thromboplastin and calcium ions is added to normal citrated plasma, the clotting mechanism is activated, leading to a fibrin clot. If a deficiency exists within the extrinsic pathway, the time required for clot formation will be prolonged depending on the severity of the deficiency.

WARNINGS AND PRECAUTIONS

The reagents contained in this kit are for in vitro diagnostic use only – DO NOT INGEST. Wear appropriate personal protective equipment when handling all kit components. Refer to the product safety declaration for the link to appropriate hazard and precautionary statements where applicable. Dispose all components in accordance with local regulations.

COMPOSITION

Composition	Content	Description	Preparation
Thromboplastin L	2 x 5 mL (REF 5265HL) 8 x 5 mL (REF 5265L) 10 x 10 mL (REF 5267L)	Lizard Rabbit Brain Thromboplastin containing Calcium Chloride, stabilisers and preservatives.	The liquid, calcified thromboplastin is ready-to-use. No further solution is required to carry out standard PT Assays. The contents of the vial should be mixed well before use. (5 minutes on roller).

Each kit contains Instructions For Use.
Each kit contains lot specific reference values issued.

ITEMS REQUIRED BUT NOT PROVIDED

The below products can be used in conjunction with Thromboplastin L:

REF 5515	ISI Calibrant Plasma Set
REF 5490	INR Reference Set

STORAGE, SHELF-LIFE AND STABILITY

Unopened reagents are stable until the given expiry date when stored under conditions indicated on the vial or kit label.

Thromboplastin L: Opened vials are stable for 2 months at 2 – 8°C, 5 days at 15°C (on-board Sysmex CA-1500) and 8 hours at 37°C (on-board AC-4 including reagent container and cap). A shift-use stability of 7 days (Sysmex CA-1500) can be achieved.
DO NOT FREEZE. Large clumps of particles or changes in expected values may indicate product deterioration.

SAMPLE COLLECTION AND PREPARATION

Plasma or citrated-blood should be used throughout. Blood (9 parts) should be collected into 3.2% or 3.8% sodium citrate anticoagulant (1 part). Separate plasma after centrifugation at 1500 x g for 15 minutes. Plasma should be kept at 18 – 24°C. Testing should be completed within 4 hours of sample collection, or plasma can be stored frozen at -20°C for 2 weeks or -70°C for 6 months. Thaw quickly at 37°C prior to testing. Do not keep at 37°C for more than 5 minutes[4].

PROCEDURE

For accurate INR reporting, it is recommended to determine the laboratory specific ISI of the reagent with the testing system in use. The Helena Biosciences Europe ISI Calibrant Plasma Set (REF 5515) is recommended for this purpose[3]. This should be performed for each new reagent batch. The Helena Biosciences Europe INR Reference Set (REF 5490) should be used to check for shifts in the local system ISI which have been noted with changes in laboratory temperature and pipet instrument servicing, amongst other local variances.

Manual Method

1. Mix sufficient Thromboplastin L to complete the anticipated testing for the day and incubate at 37°C for no more than 4 hours.
2. Pre-warm 0.1 mL of the test plasma at 37°C for 2 minutes.
3. Add 0.2 mL of freshly mixed thromboplastin reagent to the plasma while simultaneously starting a stopwatch.
4. Note the time for clot formation to the nearest 0.1 seconds.

Automated Method

Refer to the appropriate instrument operation manual for detailed instructions or contact Helena Biosciences Europe for instrument specific application guides.

INTERPRETATION OF RESULTS

Results should be reported to the nearest 0.1 seconds and duplicates should agree within 5% of each other. %PT values can be interpolated from the calibration graph (%PT at PT Calibration plasma's various measured clot time), which should be a straight line when plotted on log-log graph paper.

INR values can be calculated using the following formula: $INR = (PT\ Test\ Patient / Mean\ Normal\ PT\ Time)^{ISI}$

For clear guidance on the indications for and management of patients on warfarin, please refer to The British Society for Haematology, for their most current edition of 'Guidelines on oral anticoagulation with warfarin'. At time of printing this is the 2011 fourth edition[5].

LIMITATIONS

The use of serial dilutions of a reference plasma for the %PT curve is not recommended as this can lead to discrepancies caused by the low fibrinogen in the reference plasma dilutions which are not reflected in patient samples having predominantly normal fibrinogen levels. Helena Biosciences Europe advise use of the 5504R %PT/Direct INR kit for this purpose.

QUALITY CONTROL

Each laboratory should establish a quality control program. Normal and abnormal control plasmas should be tested prior to each batch of patient samples, to ensure satisfactory instrument and operator performance. If controls do not perform as expected, patient results should be considered invalid.
Helena Biosciences Europe supplies the following controls available for use with this product.

REF 5186	Routine Control N
REF 5187	Routine Control A
REF 5183	Routine Control SA
REF 5490	INR Reference Set

REFERENCE VALUES

Reference values can vary between laboratories depending on the techniques and systems in use. For this reason each laboratory should establish its own reference ranges. This is particularly important for local ISI calibration. Using the Sysmex series of instruments, normal values ranging from 11.50 - 14.60 seconds, 0.930 - 1.169 INR, 79.10 - 112.80 %PT are typical.

PERFORMANCE CHARACTERISTICS

The following performance characteristics have been determined by Helena Biosciences Europe or their representatives using a Sysmex CA-1500 coagulation instrument. Each laboratory should establish its own performance data.

Reproducibility

Sample	Routine Control N		Routine Control A		Routine Control SA	
	SD	*CV (%)*	*SD*	*CV (%)*	*SD*	*CV (%)*
Reproducibility	0.07	0.59	0.24	1.09	0.45	1.11
Between-run	0.10	0.83	0.16	0.75	0.49	1.20
Between-day	0.04	0.32	0.06	0.27	0.25	0.62
Within-device / Laboratory	0.12	1.07	0.29	1.35	0.72	1.75

Interferences

Helena Thromboplastin L is insensitive to Heparin levels of up to 2 U/mL. Using a 5% interference threshold, there is no significant interference from Haemoglobin at concentrations up to 10 g/L. Using a 5% interference threshold, there is no significant interference from Bilirubin at concentrations up to 0.5 g/L for Thromboplastin L. Lipid interference testing demonstrates that lipid levels do not directly affect the clot time of the reagent up to 3.75g/L. Lipid concentrations in excess of the present clot detection.

Method Comparison

Comparison of clot time in seconds and INR values were determined using Thromboplastin L and Thromboplastin LI on 268 samples. The following correlations were obtained:

Thromboplastin L (Seconds) = 0.9911x + 0.1038 $r^2 = 0.9941$ n = 268
Thromboplastin L (INR) = 0.9853x + 0.0261 $r^2 = 0.9569$ n = 268

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Thromboplastin L
Fiche technique

fr

UTILISATION

Le kit Thromboplastin L est destiné à la réalisation des analyses de l'hémostase basées sur la formation de caillots.

La première méthode de détermination standardisée du temps de prothrombine en une étape a été développée en 1935 par le Dr. Armand Quick. Cette méthode de Quick constitue désormais l'analyse de base de la coagulation servant à diagnostiquer des déficiences des facteurs de coagulation, congénitales ou acquises, à partir de la voie extrinsèque (facteurs II, V, VII et X)[1,2]. Elle sert aussi à l'induction et au monitorage des thérapies avec anticoagulants oraux[1,2] et elle peut être utilisée pour évaluer la capacité de synthèse des protéines du foie chez les patients souffrants de troubles hépatiques chroniques ou aigus.
Le Thromboplastin L provient de cerveaux de lapin mais il ressemble au BCT humain en raison de son indice de sensibilité international (ISI) faible. L'ISI du Thromboplastin L est d'environ 1,1 et est étalonné en comparaison avec la préparation internationale de référence de l'OMS[3]. Le Thromboplastin L convient tout particulièrement au monitorage des thérapies avec anticoagulants oraux et, utilisé conjointement au plasma carencé en un facteur approprié, à la détermination de l'activité du facteur de la voie extrinsèque. La thromboplastine tissulaire, en présence d'ions calcium, est un activateur qui démarre la voie extrinsèque de la coagulation. Quand un mélange de thromboplastine tissulaire et d'ions calcium est ajouté à un plasma citraté normal, le processus de coagulation, qui doit conduire à la production d'un caillot fibreux, s'active. Si la voie extrinsèque présente une anomalie, le temps nécessaire à la formation du caillot est allongé suivant la gravité du trouble de la coagulation.

AVERTISSEMENTS ET PRÉCAUTIONS

Les réactifs du kit sont à usage diagnostique in vitro uniquement – NE PAS INGÉRER. Porter un équipement de protection individuelle approprié lors de la manipulation de tous les composants du kit. Consulter la fiche de données de sécurité du produit pour obtenir le lien vers les phrases de risque et les conseils de prudence le cas échéant. Éliminer les composants conformément aux réglementations locales.

COMPOSITION

Composant	Contient	Description	Préparation
Thromboplastin L	2 x 5 mL (REF 5265HL) 8 x 5 mL (REF 5265L) 10 x 10 mL (REF 5267L)	Thromboplastine liquide de cerveaux de lapin contenant du chlorure de calcium, des stabilisateurs et des conservateurs.	La Thromboplastine liquide calcifiée est prête à l'emploi. Aucun calcium supplémentaire n'est nécessaire pour réaliser des déterminations standard du TP. Le contenu du flacon doit être bien mélangé avant utilisation (5 minutes sur un mélangeur à rouleaux).

Chaque kit contient une fiche technique.
Chaque kit contient valeurs de référence spécifiques du lot.

MATÉRIEL NÉCESSAIRE NON FOURNI

Les produits ci-dessous peuvent être utilisés en conjonction avec la Thromboplastin L :

REF 5515	ISI Calibrant Plasma Set
REF 5490	INR Reference Set

CONSERVATION, DURÉE DE VIE UTILE ET STABILITÉ

Les facteurs de réactif non ouverts sont stables jusqu'à la date de péremption indiquée s'ils sont conservés dans les conditions indiquées sur l'étiquette du kit ou du flacon.

Thromboplastin L : Les flacons ouverts sont stables pendant 2 mois à 2 °C – 8 °C, pendant 5 jours à 15 °C (à bord du Sysmex CA-1500) et pendant 8 heures à 37 °C (à bord de l'AC-4, récipient de réactif et capuchon inclus). Il est possible d'obtenir une stabilité de période de travail de 7 jours (Sysmex CA-1500).
NE PAS CONGELER. La présence de grumeaux de particules ou un écart par rapport aux valeurs prévues indique une détérioration du produit.

PRÉLÈVEMENT ET PRÉPARATION DES ÉCHANTILLONS

Utiliser tout au long du prélèvement du plasma ou du verre silicone. Mélanger 9 volumes de sang et 1 volume de citrate de sodium à 3,2% ou 3,8%. Séparer le plasma après centrifugation à 1500 x g pendant 15 minutes. Conserver le plasma entre 18 à 24°C. L'analyse doit être terminée dans les 4 heures suivant le prélèvement de l'échantillon ; sinon, il est possible de congeler le plasma 2 semaines à -20°C ou 6 mois à -70°C. Décongeler rapidement à 37°C avant de réaliser l'analyse. Ne pas laisser à 37°C plus de 5 minutes[4].

PROCÉDURE

Pour obtenir un RNI (rapport normalisé international) précis, il est recommandé à chaque laboratoire de déterminer l'ISI spécifique du réactif avec le système d'analyse utilisé. Il est conseillé d'utiliser le ISI Calibrant Plasma Set Helena Biosciences Europe (REF 5515) pour cela[3,4]. Cette opération doit être réalisée pour chaque nouveau lot de réactif. Le kit RNI de référence Helena Biosciences Europe (REF 5490) doit être utilisé pour vérifier l'existence d'un décalage de l'ISI du système local déterminé en raison d'une variation de la température du laboratoire, suite à une opération de maintenance réalisée sur l'instrument ou toute autre variable locale.

Méthode Manuelle

1. Mélanger du Thromboplastin L en quantité suffisante pour réaliser les analyses prévues dans la journée et incuber à 37 °C pendant 4 heures au maximum.
2. Préchauffer 0,1 mL du plasma à 37°C pendant 2 minutes.
3. Ajouter 0,2 mL de réactif de thromboplastine fraîchement mélangé au plasma et démarrer à ce moment un chronomètre.
4. Relever le temps de formation du caillot en arrondissant au dixième de seconde.

Méthodes Automatisées

Consulter le manuel d'utilisation de l'analyseur approprié pour obtenir des instructions détaillées ou contacter Helena Biosciences Europe pour obtenir des notes d'application spécifiques à l'instrument.

INTERPRÉTATION DES RÉSULTATS

Les résultats doivent être indiqués en arrondissant au dixième de seconde et l'écart maximal entre aux est de 5%. Il est possible d'interpoler les valeurs de %TP à partir de la courbe d'étalonnage (%TP des plasmas d'étalonnage du TP par rapport au temps de coagulation mesuré), qui doit correspondre à une ligne droite quand elle est représentée sur du papier logarithmique.

La formule suivante permet de calculer les valeurs du RNI : $RNI = (Temps\ TP\ patient / Temps\ TP\ moyen\ normal)^{ISI}$

Pour obtenir des informations claires quant aux indications et à la prise en charge des patients sous warfarine, consulter la British Society for Haematology pour obtenir la dernière édition des Guidelines on oral anticoagulation with warfarin (Recommandations relatives à l'anticoagulation orale avec la warfarine). Au moment de l'impression du présent document, il s'agit de la quatrième édition de 2011[5].

LIMITES

Il est déconseillé de réaliser des dilutions du plasma de référence pour la courbe de %TP, car cela risquerait d'entraîner des divergences dues au faible taux de fibrinogène présent dans les dilutions du plasma de référence, ce qui ne serait pas représentatif des échantillons patients qui ont principalement des taux de fibrinogène normaux. Helena Biosciences Europe conseille l'utilisation du kit 5504R %PT/Direct INR dans ce but.

CONTRÔLE QUALITÉ

Chaque laboratoire doit établir un programme de contrôle qualité. Les plasmas de contrôle, normaux et anormaux, doivent être testés avant chaque lot d'échantillons patients afin de s'assurer que l'instrument et l'opérateur offrent des performances satisfaisantes. Si les contrôles ne donnent pas les résultats prévus, les résultats du patient doivent être considérés comme non valables.
Helena Biosciences Europe distribue les contrôles suivants à utiliser avec ce produit.

REF 5186	Routine Control N
REF 5187	Routine Control A
REF 5183	Routine Control SA
REF 5490	INR Reference Set

VALEURS DE RÉFÉRENCE

Les valeurs de référence peuvent varier d'un laboratoire à l'autre suivant les techniques et les systèmes utilisés. C'est pour cette raison que chaque laboratoire doit déterminer ses propres plages de référence. Ceci est particulièrement important pour l'étalonnage de l'ISI local. Avec les instruments de la série Sysmex, l'intervalle type des valeurs normales est de 11,50 - 14,60 secondes, 0,930 - 1,169 INR, 79,10 - 112,80 %PT.

CARACTÉRISTIQUES DE PERFORMANCES

Helena Biosciences Europe ou ses mandataires ont déterminé les caractéristiques de performance suivantes en utilisant un instrument de coagulation Sysmex CA-1500. Chaque laboratoire doit établir ses propres données de performance.

Reproductibilité

Échantillon	Routine Control N		Routine Control A		Routine Control SA	
	SD	*CV (%)*	*SD*	*CV (%)*	*SD*	*CV (%)*
Répétabilité	0,07	0,59	0,24	1,09	0,45	1,11
Inter-séries	0,10	0,83	0,16	0,75	0,49	1,20
Inter-jours	0,04	0,32	0,06	0,27	0,25	0,62
Inter-dispositif/laboratoire	0,12	1,07	0,29	1,35	0,72	1,75

Interférences

Thromboplastin L (ISI tuile) d'Helena ne présente pas d'interférences avec un taux d'héparine jusqu'à 2 U/mL. En utilisant un seuil d'interférence de 5 %, il n'y a pas d'interférences significatives de l'hémoglobine à une concentration jusqu'à 10 g/L. En utilisant un seuil d'interférence de 5 %, il n'y a pas d'interférences significatives de la bilirubine à une concentration jusqu'à 0,5 g/L pour la Thromboplastin L. Une évaluation de l'interférence des lipides montre que les taux de lipides n'affectent pas le temps de coagulation du réactif jusqu'à 3,75 g/L. Une concentration en lipides dépassant cette limite empêche la détection du caillot.

Comparaison de la méthode

Des comparaisons du temps de coagulation en secondes et en valeurs de RNI ont été déterminées en utilisant les produits Thromboplastin L et Thromboplastin LI avec 268 échantillons. Les corrélations suivantes ont été obtenues :

Thromboplastin L (secondes) = 0,9911x + 0,1038 $r^2 = 0,9941$ n = 268
Thromboplastin L (INR) = 0,9853x + 0,0261 $r^2 = 0,9569$ n = 268

BIBLIOGRAPHIE

1. Quick AJ (1935) A Study of the Coagulation Defect in Hemophilia and Jaundice. *Am. J. Med. Sci.* 190: 501.
2. Biggs R (1976) Human Blood Coagulation, Haemostasis and Thrombosis. 2nd Edition. Blackwell Scientific Publications, London.
3. Hirsh J, Poller L, Deykin D, Levine J, Dalen JE (1989) Optimal Therapeutic Range for Oral Anticoagulants. *Chest.* 95: 5S-11S.
4. Poller L (1986) Laboratory Control of Anticoagulant Therapy. *Sem. Thromb. Haemostasis.* 12: 13-19.
5. World Health Organisation (1994) Expert Committee on Biological Standards. *Technical Series.* 700: 19.
6. Clinical and Laboratory Standards Institute (2008) Collection, Transport and Processing of Blood Specimens for Testing Plasma-Based Coagulation Assays and Molecular Haemostasis Assays. Approved Guideline. 5th edn. CLSI: H21-A5.
7. Poller L, Triplett DA, Hirsh J, Carroll J, Clarke K (1995) The value of plasma calibrants in correcting coagulometer effects on International Normalised Ratios (INR): An international multicentre study. *Amer. J. Clin. Pathol.* 103: 358-365.
8. Poller L, Triplett DA, Hirsh J, Carroll J, Clarke K (1995) A comparison of lyophilised artificially depleted plasmas and lyophilised plasmas from warfarin treated patients in correcting for coagulometer effects on International Normalised Ratios. *Amer. J. Clin. Pathol.* 103: 366-371.
9. Keeling D (2011) Guidelines on Oral Anticoagulation with warfarin: Forth Edition. *British Journal of Haematology.* 154(3): 311-324.

Thromboplastin L
Anleitung

de

VERWENDUNGSZWECK

Das Thromboplastin L Kit ist für koagulometrische Gerinnungstests vorgesehen.

Die erste standardisierte Prothrombinzeit als Einstufen-Test wurde 1935 von Dr. Armand Quick entwickelt. Mittlerweile ist er der Standard-Screenertest in der Gerinnung zur Diagnose eines vererbten oder erworbenen Gerinnungsfaktormangels des extrinsischen Systems (Faktor II, V, VII und X)[1,2]. Er wird auch zur Einstellung und Überwachung von oraler Antikoagulationstherapien[1,2] verwendet und kann bei chronischen und akuten Lebererkrankungen zur Beurteilung der Funktionstüchtigkeit der Leber bei der Proteinsynthese herangezogen werden.
Thromboplastin L wird aus Kaninchenhirn gewonnen, ähnelt aber mit seinem niedrigen Internationalen Empfindlichkeitsindex (ISI) humanen Blutgerinnungsmitteln. Der ISI von Thromboplastin L liegt bei ca. 1,1 und ist gegen WHO Internationale Referenz Präparation kalibriert[3]. Thromboplastin L ist besonders für die Überwachung einer Antikoagulationstherapie geeignet und, in Verbindung mit dem entsprechenden Faktor-Mangelplasma, für die Messung der Faktor-Aktivität im extrinsischen System. Gewebethromboplastin ist in Anwesenheit von Calciumionen ein Aktivator der das extrinsische Gerinnungssystem startet. Gibt man eine Mischung von Gewebethromboplastin und Calciumionen zu normalem Citratplasma, wird die Gerinnungskaskade aktiviert und es bildet sich ein Fibringerinnsel. Besteht innerhalb des extrinsischen Systems ein Mangel, verlängert sich, je nach Schwere dieses Mangels, die Zeit bis zur Ausbildung eines Gerinnsels.

WARNHINWEISE UND VORSICHTSMASSNAHMEN

Die in diesem Kit enthaltenen Reagenzien sind ausschließlich für die Verwendung zur In-vitro-Diagnostik vorgesehen – NICHT VERSCHLUCKEN. Tragen Sie beim Umgang mit sämtlichen Komponenten des Kits geeignete Schutzbekleidung. Beachten Sie gegebenenfalls die Hinweise auf entsprechende Gefahren- und Vorsichtsmaßnahmen in der Produktsicherheitserklärung. Entsorgen Sie die Komponenten gemäß den örtlichen Vorschriften.

ZUSAMMENSETZUNG

Komponente	Inhalt	Beschreibung	Vorbereitung
Thromboplastin L	2 x 5 mL (REF 5265HL) 8 x 5 mL (REF 5265L) 10 x 10 mL (REF 5267L)	Flüssiges Thromboplastin aus Hasenhirn mit Calciumchlorid, Stabilisatoren und Konservierungsstoffen.	Das flüssige, kalzifizierte Thromboplastin ist einsatzbereit. Für die Durchführung von standardmäßigen PT-Analysen ist kein zusätzliches Calcium erforderlich. Der Inhalt der Ampulle muss vor der Verwendung gut gemischt werden (5 Minuten auf dem Walzenmischer).

Jedes Kit enthält eine Gebrauchsanweisung.
Jedes Kit enthält chargenspezifischen Referenzwerten.

ERFORDERLICHE, ABER NICHT MITGELIEFERTE ARTIKEL

Die folgenden Produkte können in Kombination mit Thromboplastin L verwendet werden:

REF 5515	ISI Calibrant Plasma Set
REF 5490	INR Reference Set

LAGERUNG, HALTBARKEIT UND STABILITÄT

Ungeöffnete Reagenzien sind unter den auf Verpackung oder Etiketten angegebenen Lagerbedingungen bis zum aufgedruckten Verfallsdatum stabil.

Thromboplastin L: Offene Ampullen sind 2 Monate bei 2 – 8 °C, 5 Tage bei 15°C im Sysmex CA-1500) und 8 Stunden bei 37°C (im AC-4 mit Reagenzienbehälter und Deckel) stabil. Eine Stabilität von 7 Tagen im Schichtbetrieb (Sysmex CA-1500) kann erreicht werden.
NICHT EINFRIEREN. Große Verklumpungen oder Veränderungen im Testergebnissen können auf eine Verfall des Produkts hinweisen.

PROBENENTNAHME UND VORBEREITUNG

Nur Plastik oder Silikonglas verwenden. Blut (9 Teile) sollte in 3,2% oder 3,8 % Natriumcitrat als Antikoagulant (1 Teil) entnommen werden. 15 Minuten bei 1500 g zentrifugieren und Plasma abpipettieren. Plasma bei 18 – 24 °C lagern. Proben sollte innerhalb von 4 Stunden verarbeitet oder bei -20°C bis 2 Wochen oder -70°C bis 6 Monate gelagert werden. Vor dem Testen schnell bei 37°C auftauen. Nicht länger als 5 Minuten bei 37°C erwärmen[4].

VORGEHENSWEISE

Um einen korrekten INR zu erhalten wird empfohlen, den laborspezifischen ISI für das Reagenz mit dem verwendeten Testsystem zu bestimmen. Helena Biosciences Europe empfiehlt zu diesem Zweck das Plasma-Kalibrator-Set (REF 5515)[3,4]. Das sollte mit jeder neuen Reagenzien-Charge durchgeführt werden. Das Helena Biosciences Europe INR Reference Set (REF 5490) dient zur Überprüfung von Verschiebungen im ISI vor Ort, die unter anderem durch Veränderungen der Labortemperatur und nach Wartungsarbeiten an den Geräten festgestellt werden sind.

Manuelle Methode

1. Mischen Sie eine ausreichende Menge von Thromboplastin L an, um die für den Tag geplanten Tests durchzuführen und inkubieren Sie es nicht länger als 4 Stunden bei 37°C.
2. 0,1 mL des zu testenden Plasmas bei 37°C 2 Minuten vorwärmen.
3. 0,2 mL des frisch zubereiteten Thromboplastin-Reagenz zum Plasma pipettieren, dabei gleichzeitig Stoppuhr drücken.
4. Die Zeit bis zur Gerinnselbildung bis auf 0,1 Sekunden genau stoppen.

Automatisierte Methoden

Siehe die Bedienungsanleitung des entsprechenden Geräts für genaue Anweisungen oder wenden Sie sich an Helena Biosciences Europe für spezielle anwendungsbezogene Hinweise.

INTERPRETATION DER ERGEBNISSE

Ergebnisse sollten bis auf 0,1 Sekunden genau gemessen werden und Doppelbestimmungen nicht mehr als 5 % abweichen. % TPZ-Werte können ausgelesen von der Kalibrierkurve interpoliert werden (% TPZ des TP-Kalibrationsplasmas zu gemessener Gerinnungszeit) und sollten auf doppeltlogarithmischem Millimeterpapier eine gerade Linie ergeben.

INR-Werte können anhand der folgenden Formel berechnet werden: $INR = (PT\ Zeit\ Patient / Mittelwert\ normale\ PT Zeit)^{ISI}$

Für klare Leitlinien zu den Indikationen für und der Verwaltung von Patienten unter Warfarin siehe die aktuelle Ausgabe der "Guidelines on oral anticoagulation with warfarin" der British Society for Haematology. Zum Zeitpunkt der Drucklegung ist das die 4. Ausgabe, 2011[5].

EINSCHRÄNKUNGEN

Die Verwendung einer Serienverdünnung von Referenzplasma für die %PT-Kurve wird nicht empfohlen, da dies zu Diskrepanzen aufgrund von niedrigen Fibrinogen in den Referenzplasma-Verdünnungen führen kann, die nicht den Patientenproben entspricht, die in der Regel normale Fibrinogen-Werte haben. Helena Biosciences Europe empfiehlt zu diesem Zweck das 5504R %PT/Direct INR Verwenden.

QUALITÄTSKONTROLLE

Jedes Labor muss für eine eigene Qualitätssicherung sorgen. Vor jeder Testserie mit Patientenproben normale und pathologische Kontrollplasmen getestet werden, um eine zufrieden stellende Gerätestellung und Bedienung zu gewährleisten. Liegen die Kontrollen außerhalb den Normbereichs, sind die Patientenergebnisse nicht zu verwenden.
In Verbindung mit diesem Produkt bietet Helena Biosciences Europe die folgenden Kontrollen an:

REF 5186	Routine Control N
REF 5187	Routine Control A
REF 5183	Routine Control SA
REF 5490	INR Reference Set

REFERENZWERTE

Referenzwerte können je nach Technik und verwendetem System von Labor zu Labor unterschiedlich sein. Aus diesem Grund sollte jedes Labor seine eigenen Referenzwerte ermitteln. Dies ist insbesondere für die ISI-Kalibrierung von Bedeutung. On wichtig. Mit der Instrumentenreihe Sysmex liegen die typischen Normwerte zwischen 11,50 - 14,60 Sekunden, 0,930 - 1,169 INR, 79,10 - 112,80 %PT.

LEISTUNGSMERKMALE

Die folgenden Leistungsergebnisse wurden von Helena Biosciences Europe oder in deren Auftrag mit einem Sysmex CA-1500 Gerinnungsgerät ermittelt. Jedes Labor muss seine eigenen Werte ermitteln.

Reproduzierbarkeit

Probe	Routine Control N		Routine Control A		Routine Control SA	
	SD	*CV (%)*	*SD*	*CV (%)*	*SD*	*CV (%)*
Wiederholbarkeit	0,07	0,59	0,24	1,09	0,45	1,11
Bei verschiedenen Analysen	0,10	0,83	0,16	0,75	0,49	1,20
An verschiedenen Tagen	0,04	0,32	0,06	0,27	0,25	0,62
Selbes Gerät/Labor	0,12	1,07	0,29	1,35	0,72	1,75

Störungen

Helena Thromboplastin L ist unempfindlich gegenüber Heparinwerten bis zu 2 Einheiten/mL. Mit einer Störschwelle von 5 % gibt es keine besondere Interferenz durch Hämoglobin bei Konzentrationen von bis zu 10 g/L. Mit einer Störschwelle von 5 % gibt es keine besonderen Interferenz durch Bilirubin bei Konzentrationen von bis zu 0,5 g/L des Thromboplastin L. Lipidstörfaktorentests zeigen, dass die Lipidmenge der Gerinnungszeit des Reagenz bis 3,7 g/L nicht direkt beeinflusst. Höhere Lipidkonzentrationen verhindern die Gerinnungsbestimmung.

Methodenvergleich

Thromboplastin L und Thromboplastin LI wurden an 268 Proben angewandt. Dabei wurden die Gerinnungszeiten in Sekunden verglichen und die INR-Werte bestimmt. Es ergaben sich folgende Korrelationen:

Thromboplastin L (Sekunden) = 0,9911x + 0,1038 $r^2 = 0,9941$ n = 268
Thromboplastin L (INR) = 0,9853x + 0,0261 $r^2 = 0,9569$ n = 268

LITERATURVERZEICHNIS

1. Quick AJ (1935) A Study of the Coagulation Defect in Hemophilia and Jaundice. *Am. J. Med. Sci.* 190: 501.
2. Biggs R (1976) Human Blood Coagulation, Haemostasis and Thrombosis. 2nd Edition. Blackwell Scientific Publications, London.
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9. Keeling D (2011) Guidelines on Oral Anticoagulation with warfarin: Forth Edition. *British Journal of Haematology.* 154(3): 311-324.

Thromboplastin L
Istruzioni per l'uso

it

SCOPO PREVISTO

Il kit Thromboplastin L è concepito per l'esecuzione di dosaggi di emostasi basati sulla presenza di coaguli.

Il primo test del tempo di protrombina standardizzato venne messo a punto dal Dr. Armand Quick nel 1935. Attualmente, questo test è ovviamente il metodo base di screening della coagulazione per la diagnosi di deficienze congenite ed acquisite dei fattori di coagulazione del percorso estrinseco (fattore II, V, VII e X)[1,2]. Questo test viene adoperato anche per l'induzione e il monitoraggio della terapia anticoagulante orale[3,4] e può essere impiegato per valutare la capacità di sintesi proteica del fegato in disordini epatici cronici o acuti.

Il kit Thromboplastin L è realizzato a partire da cervello di coniglio, ma rassomiglia a BCT umano in termini di basso indice di sensibilità internazionale (ISI). L'ISI del kit Thromboplastin L è approssimativamente pari a 1,1 ed è calibrato rispetto alla preparazione di riferimento internazionale dell'OMS[5]. Il kit Thromboplastin L è particolarmente indicato per il monitoraggio della terapia anticoagulante orale e, in combinazione con plasma carente del fattore appropriato, per la misurazione dell'attività del fattore nel percorso estrinseco. In presenza di ioni di calcio, la tromboplastina insolubile è un attivatore che dà inizio al percorso di coagulazione estrinseco. Quando una miscela di tromboplastina insolubile e di ioni di calcio viene aggiunta a normale plasma citrato, si attiva il meccanismo di coagulazione che porta alla formazione di un coagulo di fibrina. Qualora sussista una deficienza all'interno del percorso estrinseco, il tempo richiesto per la formazione del coagulo risulterà prolungato in funzione della gravità della deficienza.

AVVERTENZE E PRECAUZIONI

I reagenti contenuti in questo kit sono destinati esclusivamente alla diagnostica in vitro - NON INGERIRE. Indossare un'adeguata attrezzatura protettiva personale durante la manipolazione di tutti i componenti del kit. Per conoscere i relativi simboli precauzionali e di pericolo, fare riferimento alla dichiarazione di sicurezza del prodotto. Smaltire i componenti contaminanti alla normativa locale vigente.

COMPOSIZIONE

Componente	Contiene	Descrizione	Preparazione
Thromboplastin L	2 x 5 mL (REF 5265HL) 8 x 5 mL (REF 5265L) 10 x 10 mL (REF 5267L)	Tromboplastina liquida di cervello di coniglio contenente cloruro di calcio, stabilizzanti e conservanti.	La tromboplastina calcica liquida è pronta all'uso. Per eseguire dosaggi PT standard non è necessario alcun calcio. Il contenuto della fiala deve essere miscelato accuratamente prima dell'uso (5 minuti su un rullo).

Ogni kit contiene un foglietto per l'uso.

Ogni kit contiene un inserto recante i valori di riferimento specifici per il lotto.

MATERIALI NECESSARI, MA NON IN DOTAZIONE

In combinazione con la Thromboplastin L è possibile utilizzare i seguenti prodotti:

REF 5519	ISI Calibrant Plasma Set
REF 5490	INR Reference Set

CONSERVAZIONE, VITA UTILE E STABILITÀ

I reagenti non aperti sono stabili fino alla data di scadenza indicata se conservati nelle condizioni riportate sull'etichetta o sull'astuccio del kit.

Thromboplastin L: Le fiale aperte sono stabili per 2 mesi se una temperatura compresa tra 2 e 8°C, per 5 giorni a 15°C (Sysmex CA-1500 on-board) e per 6 ore a 37°C (ACL® on-board compresa il contenitore del reagente e il tappo). È possibile ottenere una stabilità di fino a 7 giorni (Sysmex CA-1500).
NON CONGELARE. Eventuali consistenti di particelle o variazioni nei valori previsti possono essere indice di deterioramento del prodotto.

RACCOLTA E PREPARAZIONE DEI CAMPIONI

Nel corso dell'intera procedura è necessario utilizzare plastica o vetro siliconizzato. Il sangue (9 parti) deve essere raccolto in sodio citrato al 3,2% o al 3,8% come anticoagulante (1 parte). Esamina il plasma in seguito a centrifugazione a 1500 x g per 15 minuti. Il plasma deve essere conservato a 18 – 24°C. I test devono essere completati entro 4 ore dalla raccolta dei campioni. In alternativa, il plasma può essere conservato congelato a -20°C per 2 settimane o a -70°C per 6 mesi. Decongelare rapidamente a 37°C prima di eseguire i test. Non conservare a 37°C per oltre 5 minuti[6].

PROCEDURA

Per un riferimento accurato dell'INR si raccomanda di determinare l'ISI specifico del laboratorio per il reagente con il sistema di test in uso. A tale scopo si raccomanda il kit Calibrant Plasma Set (REF 5519) di Helena Biosciences Europe[7,8]. Questa procedura deve essere eseguita per ogni nuovo lotto di reagente. L'INR Reference Set (REF 5490) di Helena Biosciences Europe deve inoltre essere utilizzato per rilevare eventuali spostamenti dell'ISI del sistema locale ottenuti in concomitanza con cambiamenti della temperatura del laboratorio e in seguito a manutenzione delle strumenta, tra le altre variazioni locali.

Metodo Manuale

1. Miscelare un quantitativo di Thromboplastin L sufficiente a completare i test previsti per la giornata e incubare a +37°C per un massimo di 4 ore.
2. Preriscaldare 0,1 mL di plasma di prova a 37°C per 2 minuti.
3. Aggiungere al plasma 0,2 mL di reagente a base di tromboplastina appena miscelato, azionando contemporaneamente un cronometro.
4. Annotare il tempo di formazione del coagulo con un'approssimazione a 0,1 secondi.

Metodo Automatico

Fare riferimento al manuale utente dello strumento appropriato per soluzioni dettagliate oppure contattare Helena Biosciences Europe per le note applicative specifiche dello strumento.

INTERPRETAZIONE DEI RISULTATI

I risultati devono essere inclusi con un'approssimazione a 0,1 secondi e le ripetizioni devono corrispondere con una tolleranza del 5%. I valori di %PT possono essere interpolati sul grafico di calibrazione (%PT dei plasmi di calibrazione PT vs. tempo di coagulazione rilevato), ma si raccomanda una stima a doppia scala logaritmica, dove ogniuno sotto forma di linea retta.

I valori di INR possono essere calcolati usando la seguente formula:

INR = (Tempo di PT Paziente / Tempo di PT normale medio)[9]

Per una guida intera sulle indicazioni per la gestione dei pazienti con warfarina fare riferimento a The British Society for Haematology's dosing guidelines più aggiornata delle 'Linee guida sull'anticoagulazione orale con warfarina'. Al momento della stampa questa è la quarta edizione dal 2011[9].

LIMITAZIONI

Si sconsiglia l'impiego di diluizioni serial di un plasma di riferimento per la curva %PT, che infatti possono dare luogo a discrepanze elevate al bassi livelli di fibrinogeno nelle diluizioni del plasma di riferimento, che non compaiono invece nei campioni dei pazienti con livelli di fibrinogeno prevalentemente normali. Helena Biosciences Europe consiglia di utilizzare a questo scopo il kit SSIAR %PT/INR diretta o tal fin.

CONTROLLO QUALITÀ

Ogni laboratorio deve definire un programma di controllo qualità. I plasmi di controllo normali e anormali devono essere testati prima di ogni lotto di campioni di pazienti, per garantire un livello prestazionale soddisfacente sia per quanto riguarda lo strumento che per l'operatore. Qualora i controlli non funzionassero come previsto, i risultati relativi ai pazienti dovranno essere considerati non validi.
Helena Biosciences Europe mette a disposizione i seguenti controlli utilizzabili con questo prodotto:

REF 5186	Routine Control N
REF 5187	Routine Control A
REF 5183	Routine Control SA
REF 5490	INR Reference Set

VALORI DI RIFERIMENTO

Per la sicurezza del paziente, è necessario che il sistema sia monitorato continuamente da un operatore qualificato. Per tale motivo ciascun laboratorio dovrà stabilirsi i propri range di riferimento. Ciò è particolarmente importante per la calibrazione dell'ISI locale. Con l'impiego della gamma di strumenti Sysmex, i valori normali che variano tra 11,50 - 14,60 secondi; 0,930 - 1,160 INR; 79,10 - 112,80 %PT sono ritenuti tipici.

CARATTERISTICHE PRESTAZIONALI

Le seguenti caratteristiche prestazionali sono state determinate da Helena Biosciences Europe o dai propri rappresentanti con l'utilizzo di uno strumento di coagulazione Sysmex CA-1500. Ciascun laboratorio dovrà pertanto elaborare i propri dati prestazionali.

Riproducibilità

Campione	Routine Control N		Routine Control A		Routine Control SA	
	SD	*CV (%)*	*SD*	*CV (%)*	*SD*	*CV (%)*
Ripetibilità	0,07	0,59	0,24	1,09	0,45	1,11
Tra le serie	0,10	0,83	0,16	0,75	0,49	1,20
Tra i giorni	0,04	0,32	0,06	0,27	0,25	0,62
All'interno del dispositivo/laboratorio	0,12	1,07	0,29	1,35	0,72	1,75

Interferenze

La Thromboplastin L Helena non è sensibile ai livelli di eparina di oltre 3 U/mL. Utilizzando una soglia di interferenza del 5% non risulta esservi alcuna significativa interferenza da parte dell'emoglobina a concentrazioni fino a 10 g/l. Utilizzando una soglia di interferenza del 5%, non risulta esservi alcuna significativa interferenza da parte della bilirubina a concentrazioni fino a 0,5 g/l per la Thromboplastin L. I test per la interferenza dei lipidi dimostrano che i livelli di lipidi non influiscono direttamente il tempo di coagulazione del reagente fino a 3,75 g/l. Concentrazioni lipidiche superiori a questo valore impediscono il rilevamento del coagulo.

Confronto dei metodi

Si è eseguito un confronto su 268 campioni tra il tempo di coagulazione in secondi e i valori INR utilizzando la Thromboplastina L e la thromboplastina LS. Si sono ottenute le seguenti correlazioni:

Thromboplastin L (secondi) = 0,9811x + 0,1838	$r^2 = 0,9941$	n = 268
Thromboplastin L (INR) = 0,9353x + 0,0261	$r^2 = 0,9500$	n = 268

BIBLIOGRAFIA

1. Quick AJ (1935) A Study of the Coagulation Defect in Hemophilia and Jaundice. *Am. J. Med. Sci*, 189: 501.
2. Biggs R (1976) Human Blood Coagulation, Haemostasis and Thrombosis, 2nd Edition, Blackwell Scientific Publications, London.
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5. World Health Organisation (1984) Expert Committee on Biological Standards, *Technical Series*, 700: 19.
6. Clinical and Laboratory Standards Institute (2008) Collection, Transport and Processing of Blood Specimens for Testing Plasma-Based Coagulation Assays and Molecular Haemostasis Assays; Approved Guideline, 5th edn. CLSI: H21-A5.
7. Poller L., Triplett DA, Hirsh J, Carroll J, Clarke K (1995) The value of plasma calibrants in correcting coagulometer effects on International Normalised Ratios (INR): An international multicentre study. *Amer J. Clin. Pathol*, 103: 358-365.
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9. Keeling D (2011) Guidelines on Oral Anticoagulation with warfarin- Forth Edition. *British Journal of Haematology*, 154(3): 311-324.

Thromboplastin L
Instrucciones de uso

es

USO PREVISTO

El uso previsto del kit Thromboplastin L es realizar ensayos de hemostasia basados en la coagulación.

La primera prueba estandarizada de la protrombina en una sola etapa fue desarrollada por el Dr. Armand Quick en 1935. Ahora se ha convertido en la prueba básica de cribado de la coagulación para el diagnóstico de deficiencias congénitas y adquiridas de factores de coagulación de la vía extrínseca (factores II, V, VII y X)[1,2]. Se usa también para la inducción y monitorización del tratamiento anticoagulante oral[3,4] y puede usarse para valorar la capacidad de síntesis de proteínas del hígado en trastornos hepáticos crónicos o agudos.

La Thromboplastin L, basada su origen en cerebro de conejo, pero se parece a la BCT humana en su bajo índice de Sensibilidad Internacional (ISI). El ISI de Thromboplastin L es de aproximadamente 1,1 y se calibra contra el preparado de referencia internacional de la OMS[5]. La prueba de Thromboplastin L está especialmente adaptada a la monitorización del tratamiento anticoagulante oral y, conjuntamente con el plasma deficiente en el factor apropiado, la medición de la actividad de los factores en la vía extrínseca. La tromboplastina insolu, en presencia de iones calcio, es un activador que inicia la vía extrínseca de la coagulación. Cuando se añade una mezcla de tromboplastina insolu e iones calcio al plasma normal citratado, se activa el mecanismo de coagulación, conduciendo a un coágulo de fibrina. Si se produce una deficiencia dentro de la vía extrínseca, el tiempo necesario para la formación de coágulos se prolongará dependiendo de la intensidad de la deficiencia.

ADVERTENCIAS Y PRECAUCIONES

Los reactivos que contiene este kit son sólo para uso de diagnóstico in vitro. NO INGERIR. Lleve el equipo de protección personal adecuado cuando utilice todos los componentes del kit. Consulte la declaración de seguridad del producto para saber más sobre las indicaciones adecuadas de advertencia y riesgo. Deseche los componentes de conformidad con las normativas locales.

COMPOSICIÓN

Componente	Contiene	Descripción	Preparación
Thromboplastin L	2 x 5 mL (REF 5265HL) 8 x 5 mL (REF 5265L) 10 x 10 mL (REF 5267L)	Tromboplastina líquida de cerebro de conejo que contiene cloruro de calcio, estabilizadores y conservantes.	La tromboplastina líquida calcificada está lista para su uso. No es necesario más calcio para realizar pruebas estándar de PT. Los contenidos del vial se deben mezclar bien antes de utilizarlos (5 minutos en el balancín).

Cada kit contiene instrucciones de uso.

Cada kit contiene valores de referencia específicos insertados del lote.

ARTÍCULOS NECESARIOS NO SUMINISTRADOS

Los siguientes productos se puede utilizar junto con la Thromboplastin L:

REF 5519	ISI Calibrant Plasma Set
REF 5490	INR Reference Set

ALMACENAMIENTO, CADUCIDAD Y ESTABILIDAD

Los reactivos no abiertos son estables hasta la fecha de caducidad indicada cuando se conservan en las condiciones indicadas en el vial o en la etiqueta del kit.

Thromboplastin L: Los viales abiertos permanecen estables durante 2 meses a una temperatura de entre 2 – 8°C, 5 días a una temperatura mayor a 15°C (en el Sysmex CA1500), y 6 horas a una temperatura mayor a 33°C (en el ACL incluido el contenedor del reactivo y el tapón). Se puede conseguir estabilidad multiuso de 7 días (Sysmex CA-1500).
NO CONGELAR. Los grumos grandes de partículas o los cambios en los valores esperados pueden indicar deterioro del producto.

RECOGIDA Y PREPARACIÓN DE LAS MUESTRAS

Debe usarse siempre plástico o vidrio siliconizado. Debe recogerse sangre (9 partes) en el anticoagulante citrato sódico al 3,2% o al 3,8% (1 parte). Separe el plasma después de la centrifugación a 1500 x g durante 15 minutos. El plasma debe conservarse a 18 – 24°C. Las pruebas deben terminarse en 4 horas desde la recogida de las muestras o el plasma puede conservarse congelado a -20°C durante 2 semanas o -70°C durante 6 meses. Descongelar rápidamente a 37°C antes de realizar la prueba. No conservar a 37°C durante más de 5 minutos[6].

PROCEDIMIENTO

Para la comunicación exacta del INR, se recomienda determinar la ISI específica del laboratorio del reactivo con el sistema de prueba en uso. Se recomienda el ISI Calibrant Plasma Set de Helena Biosciences Europe (REF 5519) para este fin[7,8]. Esto debe realizarse para cada nuevo lote de reactivos. Debe usarse el Set de Referencia de INR de Helena Biosciences Europe (REF 5490) para comprobar las desviaciones en el ISI del sistema local que se han observado con cambios en la temperatura del laboratorio y el mantenimiento posterior, entre otras variaciones locales.

Método Manual

1. Mezclar la cantidad suficiente de Thromboplastin L para completar la prueba anticipada para el día e incubar a +37°C durante un período de tiempo no superior a 4 horas.
2. Precalentar 0,1 mL del plasma de prueba a 37°C durante 2 minutos.
3. Añada 0,2 mL de reactivo de tromboplastina recién mezclado al plasma mientras pone en marcha simultáneamente un cronómetro.
4. observa el tiempo hasta la formación del coágulo procurando anotar en la décima 0,1 de segundo más próxima.

Método Automatizado

Consulte el manual del usuario del instrumento adecuado para instrucciones detalladas o póngase en contacto con Helena Biosciences Europe para notas de aplicación específicas del instrumento.

INTERPRETACIÓN DE LOS RESULTADOS

Los resultados deben comunicarse en los 0,1 segundos más próximos y las pruebas duplicadas deben estar de acuerdo entre sí dentro del 5%. Los valores de %TP pueden interpolarse a partir del gráfico de calibración (%TP de los plasmas de calibración del TP frente al tiempo de coagulo medido) que debe ser una línea recta cuando se representa en un papel de gráficos log-log.

Los valores de INR pueden calcularse usando la siguiente fórmula:

INR = (Tiempo de TP Paciente / Tiempo de TP normal medio)[9]

Si necesita una guía para las indicaciones y tratamiento de pacientes con warfarina, consulte The British Society for Haematology y su última edición de 'Guidelines on oral anticoagulation with warfarin' que, en el momento de impresión de este documento, se encuentra en su cuarta edición de 2011[9].

LIMITACIONES

El uso de diluciones seriadas de un plasma de referencia para la curva de %TP no se recomienda, porque puede llevar a discrepancias importantes por el bajo fibrinógeno en las diluciones del plasma de referencia, que no se refleja en las muestras de pacientes que tienen fundamentalmente niveles normales de fibrinógeno. Helena Biosciences Europe recomienda utilizar el kit SSIAR %PT/INR directa o tal fin.

CONTROL DE CALIDAD

Cada laboratorio debe establecer un programa de control de calidad. Los controles normales y anormales deben establecerse antes de cada lote de muestras del paciente, para asegurar un funcionamiento adecuado del instrumento y el operador. Si los controles no se realizan como se esperaba, los resultados del paciente deben considerarse inválidos.
Helena Biosciences Europa suministra los siguientes controles disponibles para usar con este producto:

REF 5186	Routine Control N
REF 5187	Routine Control A
REF 5183	Routine Control SA
REF 5490	INR Reference Set

VALORES DE REFERENCIA

Los valores de referencia pueden variar entre los laboratorios dependiendo de las técnicas y sistemas usados. Por esta razón, cada laboratorio debe establecer su propio intervalo normal. Esto es especialmente importante para la calibración local del ISI. Con la serie de instrumentos Sysmex, es frecuente que los valores normales estén entre 11,50 - 14,60 segundos; 0,930 - 1,160 INR; 79,10 - 112,80 %PT.

CARACTERÍSTICAS FUNCIONALES

Las siguientes características de rendimiento han sido determinadas por Helena Biosciences Europa o sus representantes usando un instrumento de coagulación Sysmex CA-1500. Cada laboratorio debe establecer sus propios datos de rendimiento.

Reproducibilidad

Muestra	Routine Control N		Routine Control A		Routine Control SA	
	SD	*CV (%)*	*SD*	*CV (%)*	*SD*	*CV (%)*
Repetibilidad	0,07	0,59	0,24	1,09	0,45	1,11
Entre series	0,10	0,83	0,16	0,75	0,49	1,20
Entre días	0,04	0,32	0,06	0,27	0,25	0,62
En el dispositivo/laboratorio	0,12	1,07	0,29	1,35	0,72	1,75

Interferencias

Helena Thromboplastin L no detecta niveles de heparina inferiores a 2 U/mL Con un umbral de interferencia del 5%, no hay interferencias significativas de hemoglobina en concentraciones de hasta 10 g/L. Con un umbral de interferencia del 5%, no hay interferencias significativas de bilirrubina en concentraciones de hasta 0,5 g/L para Thromboplastin L. La interferencia de lípidos demuestra que los niveles de lípidos no afectan directamente al tiempo de coagulación del reactivo hasta 3,75 g/L. Las concentraciones de lípidos superiores evitan detectar la coagulación.

Comparación del método

La comparación del tiempo de coagulación y los valores INR se determinó con tromboplastina L y tromboplastina LS en 268 muestras. Se obtuvieron las siguientes correlaciones:

Thromboplastin L (segundos) = 0,9811x + 0,1838	$r^2 = 0,9941$	n = 268
Thromboplastin L (INR) = 0,9353x + 0,0261	$r^2 = 0,9500$	n = 268

BIBLIOGRAFÍA

1. Quick AJ (1935) A Study of the Coagulation Defect in Hemophilia and Jaundice. *Am. J. Med. Sci*, 189: 501.
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Тест-система "Жидкий тромбопластин"
инструкция

ru

НАЗНАЧЕНИЕ

Комплект Тест-система "Жидкий тромбопластин" предназначен для выполнения ряда анализов гемостаза, на основе крайнего случая.

Впервые стандартизированный лабораторный тест для определения протромбинового времени был предложен д-ром Армандом Квиком в 1935г. Сейчас он стал основным скрининговым тестом для диагностики врожденного и приобретенного дефицитов факторов свертывания крови по внешнему пути (фактора II, V, VII и X)[1,2]. Он также используется для мониторинга терапии оральными антикоагулянтами[3,4], а также может применяться и для оценки белок синтетической функции печени при ее острых либо хронических заболеваниях.

Реагент приготовлен на экстракте мозга кролика и по значению своего МНИ (Международного Индекса Чувствительности) согласован со Стандартом Тромбопластина (ВСТ – Potin Comparative Thromboplastin). МНИ Жидкого тромбопластина примерно равен 1,1 и откалиброван относительно международного референса ВОЗ[5]. Жидкий тромбопластин особенно подходит для мониторинга терапии прямыми антикоагулянтами и для измерения активности факторов внешнего пути свертывания крови с соответствующей (фактор-дефицитной плазмой). Тканевый тромбопластин в присутствии ионов кальция является активатором внешнего пути свертывания крови. При добавлении смеси тканевого тромбопластина с кальцием к нормальной цитратной плазме активируется механизм ее свертывания, приводящий к образованию фибринового сгустка. Если у пациента имеет место дефицит факторов внешнего пути, то время, необходимое для образования сгустка будет удлиняться прямо пропорционально степени дефицита фактора.

ПРЕДУПРЕЖДЕНИЯ И МЕРЫ ПРЕДОСТОРОЖНОСТИ

Содержащиеся в данном наборе реагенты предназначены только для in vitro диагностики – НЕ ПРОГЛАТЫВАТЬ ВНУТРЬ! При работе со всеми компонентами набора используйте соответствующие средства индивидуальной защиты. В случае необходимости за информацией о безопасности изделия для ознакомления с потенциальными признаками опасного воздействия и сведениями о типах предупредительных надписей компонентов и их меры необходимо в соответствии с местными правилами.

СОСТАВ

Компоненты	Состав набора	Описание	Приготовление реагента
Жидкий тромбопластин	2 x 5 мл (Кат.№ 5265HL) 8 x 5 мл (Кат.№ 5265L) 10 x 10 мл (Кат.№ 5267L)	Тромбопластин - кальцийсодержащий суспензия реагента из экстракта ткани мозга кролика, растворителя кальция и стабилизаторов	Жидкий калиброванный тромбопластин, готовый для использования. Для проведения теста ПВ дополнительных кальций не требуется. Содержимое флакона перемешать до использования (5 мин. на ролике).

Каждый набор содержит инструкцию по применению.

Вкладыш с указанием референсных значений и различными анализаторами и методами исследования, характерными для данного теста.

НЕОБХОДИМЫЕ КОМПОНЕНТЫ, НЕ ВКЛЮЧЕННЫЕ В КОМПЛЕКТ ПОСТАВКИ

Продукция ниже, которая может использоваться совместно с Тест-системой «Тромбопластин»:

Кат.№ 5519	Калибратор МНИ
Кат.№ 5490	Контроль на МНО

ХРАНЕНИЕ, СРОК ГОДНОСТИ И УСТОЙЧИВОСТЬ

Неоткрытые флаконы с реагентами хранятся до истечения срока годности в условиях, указанных на этикетке.

Жидкий тромбопластин	Вскрытые флаконы годны к применению в течение 2 месяцев при температуре хранения 2-8°С, 5 дней при 15°С (на полу Sysmex CA-1500) и 6 часов при 37°С (на полу AC-4, используя контейнер для реагентов с крышкой). Стабильность до 7 дней (Sysmex CA-1500 - 7 дней). НЕ ЗАМОРАЖИВАТЬ! Признаки ухудшения качества реагента: большое скопление частиц или отличие от ожидаемых значений могут означать ухудшение свойств реактива.

ОТБОР И ПОДГОТОВКА ОБРАЗЦОВ

Для работы следует использовать только пластиковую или силиконированную стеклянную посуду. Кровь забирается в пробирку с цитратным антикоагулянтом (3,2% или 3,8% цитрат натрия в соотношении 9 к 1. После центрифугирования при 1500 g в течение 15 минут использованием других параметров должна проводиться лабораторией), получению плазмы необходимо отделить от оформленных элементов крови. Плазму следует хранить при температуре 18 – 24°С. Тестирование должно быть проведено в течение 4 часов после забора образцов, либо плазму можно заморозить и хранить при температуре -20°С в течение 2 недель или при -70°С в течение 6 месяцев. Перед проведением исследования плазму следует быстро разморозить при 37°С. Не держать плазму при 37°С более 5 минут[6].

ПРОЦЕДУРА

Для установления более точного МНО рекомендуется определить международное для лаборатории значение МИН при использовании «реагент-прибор». МИНа для жидкого тромбопластина Helena Biosciences Europe рассчитывает для этих целей применять Калибратор МИН (Кат.№ REF 5519)[7,8]. Подобная процедура должна быть проведена для каждой новой партии реагентов. Контроль на МНО (Кат.№ REF 5490) используется для контроля изменений в «реагенты-МИН» при изм. вер. значение новых температурных характеристик в лаборатории и использования приборов, а также изменений температуры в лаборатории и прочих локальных факторов.

Ручной Метод

1. Взять необходимое количество жидкого тромбопластина, необходимого для проведения тестирования текущего дня и инкубировать при 37°С не более 4 часов.
2. Прогреть 0,1 мл тестируемой плазмы при 37°С в течение 2 минут.
3. Добавить 0,2 мл приготовленного рабочего пенного реагента и включить секундомер одновременно с этим.
4. Зафиксировать время, необходимое для образования сгустка с точностью до 0,1 сек.

Автоматизированный Метод

Для проведения тестирования на автоматических коагулометрах, необходимо обратиться к руководству пользователя и к соответствующей аппликации, которая доступна у производителя коагулометра и компании Хелена

ИНТЕРПРЕТАЦИЯ РЕЗУЛЬТАТОВ

Результаты должны быть представлены с точностью до 0,1 сек и разница в повторах не должна превышать 5%. Значение %ПВ можно определить по калибровочному графику графика зависимости %ПВ от измеренного времени), который должен быть строго линейным в билогарифмических координатах при построении графика на калиброванных с логарифмическими масштабом на обеих осях.

МНО может быть вычислено по формуле:

МНО= (ПВ пациента/среднее ПВ нормы)[9]МИН

Для более подробной информации по мониторингу пациентов на Варфарине обратитесь к рекомендациям Британского Общества по Гематологии – «Рекомендации по антикоагулянтной терапии варфарином» 4-е издание 2011г., на время печати этого документа[9].

ОГРАНИЧЕНИЯ

Для получения правила зависимости (%ПТ) от времени свертывания не рекомендуется использовать серии разведений нормальной плазмы, поскольку это может привести к неточным значениям, вследствие содержания фибриногена в больших разведениях референсной плазмы, что не соответствует образцам пациентов, имеющих, в основном, нормальный уровень фибриногена. Компания Хелена рекомендует использовать калибратор-кит - номер SSIAR для %ПВ, а именно определения МНО.

КОНТРОЛЬ КАЧЕСТВА

Каждая лаборатория должна установить программу контроля качества. Перед измерением каждой партии образцов пациентов необходимо протестировать нормальную и патологическую плазму, чтобы убедиться в удовлетворительной работе оборудования и оператора. Если контрольные измерения не совпадают с ожидаемыми значениями, то полученные данные пациентов следует считать недостоверными.
Компания Хелена рекомендует следующие контрольные материалы:

Кат.№ 5186	Контроль качества, норма
Кат.№ 5187	Контроль качества, умеренно выраженная патология
Кат.№ 5183	Контроль качества, высокая патология
Кат.№ 5490	Контроль на МНО

НОРМАЛЬНЫЕ ПОКАЗАТЕЛИ

Референсные значения могут варьировать между лабораториями в зависимости от используемых методов и анализаторов. В связи с этим каждая лаборатория должна определить свою собственную область референсных значений. Эта крайне важно для установки локального МИН тромбопластина. Диапазон нормальных значений пациентов 11,50 – 14,60 сек.; 0,930 – 1,160 INR; 79,10 – 112,80 %ПТ с использованием коагулометров Sysmex.

ЭКСПЛУАТАЦИОННЫЕ ХАРАКТЕРИСТИКИ

Компания Хелена или ее дистрибьюторами проведены следующие характеристики эффективности. Каждая лаборатория должна определить свои собственные диапазоны нормальных значений.
При использовании аппаратов-анализаторов коагулометров и реагентов Хелена были определены следующие коэффициенты надежности (CV):

Воспроизводимость

Образец	Контроль качества, норма		Контроль качества, умеренно выраженная патология		Контроль качества, высокая патология	
	SD	*CV (%)*	*SD*	*CV (%)*	*SD*	*CV (%)*
Воспроизводимость	0,07	0,59	0,24	1,09	0,45	1,11
в пределах аналитической серии	0,10	0,83	0,16	0,75	0,49	1,20
между разными аналитическими сериями	0,04	0,32	0,06	0,27	0,25	0,62
между партиями/реагентами	0,12	1,07	0,29	1,35	0,72	1,75

Тест-система "Жидкий тромбопластин" не чувствителен к гепарину в концентрации до 2 Ед/мл. Используя 5% допустимое отклонение, реагент не чувствителен к гемоглобину в концентрации до 10 г/л. Используя 5% допустимое отклонение, реагент не чувствителен к билирубину в концентрации до 0,5 г/л. По оценке чувствительности к уровню липидов до 3,75 г/л оказывающие влияние на время свертывания. Концентрации липидов выше этого уровня препятствуют обнаружению сгустка.

Сравнение со стандартами:

Сравнение между МНО, 268 образцов, измеренных с помощью Тест-системы "Тромбопластин" и Тест-системы "Жидкий тромбопластин" дали следующие статистические соотношения:

Жидкий Тромбопластин (сек) = 0,9811x + 0,1838	$r^2 = 0,9941$	n = 268
Жидкий Тромбопластин (МНО) = 0,9353x + 0,0261	$r^2 = 0,9500$	n = 268

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Declaration of Conformity

HL-7-0511DC DOI 2015/08 (4)

In Application of the Council Directive 98/79/EC on the approximation of the laws of the Member States relating to In Vitro Diagnostic Medical Devices & CE marking.

Declaration of conformance to applicable sections of Annex I - Essential Requirements and Annex III (EC Declaration of Conformity) imposed by sections 2 to 5. The below listed products are not classified under Annex II Lists A or B. Access to the appropriate technical files will be made available to the appropriate body in the event this is required.

Product Code	Description	GMDN Classification Code
5376	Clauss Fibrinogen 100	55997

I, the undersigned declare that the devices registered against the above GMDN Classification Code conforms to the said Directives.

Full Name: M.J. Stephenson

Title: Managing Director

Signed:

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Clauss Fibrinogen 100

REF 5376
REF 5376H

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HL-2-0592P 2016/01 (11)

Clauss Fibrinogen 100
Instructions for use

en

INTENDED PURPOSE

The Clauss Fibrinogen 100 kit is intended for carrying out clot based haemostasis assays.

Clauss[1] developed a simple method for the quantitative determination of fibrinogen by measuring the clotting time of dilute plasma after the addition of Thrombin (>30 units/mL). This clot time is proportional to the fibrinogen concentration. Levels of fibrinogen can increase as a result of inflammation, pregnancy or oral contraceptive use[2]. Decreased levels can be found in certain states such as liver disease and DIC. Congenital deficiencies include afibrinogenaemia (no detectable fibrinogen), hypofibrinogenaemia (<1 mg/mL) and dysfibrinogenaemia (abnormal fibrinogen molecules). Clauss Fibrinogen 100 is intended for the quantitative determination of fibrinogen in human plasma.

WARNINGS AND PRECAUTIONS

The reagents contained in this kit are for *in vitro* diagnostic use only – DO NOT INGEST. Wear appropriate personal protective equipment when handling all kit components. Refer to the product safety declaration for the link to appropriate hazard and precautionary statements where applicable. Dispose of components in accordance with local regulations.

Blood products have been screened and found negative (unless otherwise stated on the kit box or vial) for the presence of:

Hepatitis B Antigen (HbsAg)
HIV 1 antibody/ HIV 2 antibody
HCV antibody

However they should be handled with the same precautions as a human patient sample.

COMPOSITION

Component	Content	Description	Preparation
Thrombin 100 NIH/mL	5 x 2 mL (REF 5376) 5 x 1 mL (REF 5376H)	Contains approximately 200 (REF 5376) / 100 (REF 5376H) units Bovine thrombin with stabilisers.	Reconstitute each vial with: 2 mL (REF 5376) 1 mL (REF 5376H) of purified water or kaolin suspension depending on manual or instrument specific methodology. Take care when pipetting to avoid contamination. Swirl gently and allow to stand for 15 minutes. Mix well immediately before use. Do not shake.
Fibrinogen Calibrator	2 x 1 mL (REF 5376) 1 x 1 mL (REF 5376H)	Contains 1 mL of lyophilised normal human plasma assayed for fibrinogen levels.	Reconstitute each vial with exactly 1 mL of purified water. Swirl gently and allow to stand for 10 minutes. Mix gently before use. Do not shake.
Owren's Buffer	2 x 25 mL (REF 5376) 1 x 25 mL (REF 5376H)	Contains 25 mL of buffer which contains barbital and sodium chloride and sodium azide as preservative.	The buffer is ready for use as packaged.
Kaolin Suspension 0.5 g/L	2 x 5 mL (REF 5376) 1 x 5 mL (REF 5376H)	Contains 5 mL of kaolin suspension at 0.5 g/L.	Mix vigorously immediately before use. Ensure no sedimentation is visible.

Each kit contains instructions for use.
Each kit contains lot specific reference values insert.

ITEMS REQUIRED BUT NOT PROVIDED

Helena Biosciences Europe fibrinogen assay reagents are suitable for use on any coagulation analyser capable of performing Clauss Fibrinogen assays, but for use with the AC-K, C-1, C-2, C-4]. Refer to the Instrument Operators' Manual and/or application note for appropriate instructions.

The reagents contained in this kit are also available as separate items:

REF 5374	Clauss Fibrinogen (Thrombin only)
REF 5378	Clauss Fibrinogen (Thrombin only)
REF 5379	Fibrinogen Calibrator
REF 5375	Owren's Buffer
REF 5376S	Kaolin Suspension

STORAGE, SHELF-LIFE AND STABILITY

Unopened vials are stable until the given expiry date when stored under conditions indicated on the vial or kit label.

Thrombin 100 NIH/mL	Once reconstituted, the reagent is stable for 8 hours at 2 – 8°C, 1 week at 2 –8°C or 1 month at -20°C.
Fibrinogen Calibrator	Once reconstituted, the reagent is stable for 4 hours at 2 –8°C.
Owren's Buffer	Store at 2 – 8°C once opened.
Kaolin Suspension	Store at 2 – 8°C once opened.

SAMPLE COLLECTION AND PREPARATION

Plastic or siliconised glass should be used throughout. Blood (9 parts) should be collected into 3.2% or 3.8% sodium citrate anticoagulant (1 part). Separate plasma after centrifugation at 1500 x g for 15 minutes. Plasma should be kept at 2 – 8°C or 18 – 24°C. Testing should be completed within 4 hours of sample collection, or plasma can be stored frozen at -20°C for 2 weeks or -70°C for 6 months. Thaw quickly at 37°C prior to testing. Do not keep at 37°C for more than 5 minutes[3].

PROCEDURE

Manual Method

Prepare all reagents as instructed under "composition".

1. Standard Curve Preparation:
   a. A new standard curve should be used when reagent lot numbers change or if expected values in quality control testing change. Prepare the following dilutions in Owren's Buffer (mix without shaking):

Tube	Dilution	Fibrinogen Calibrator (mL)	Buffer (mL)
1	1 + 4	0.2	0.8
2	1 + 9	0.1	0.9
3	1 + 19	0.1	1.9
4	1 + 29	0.1	2.9
5	1 + 39	0.1	3.9

2. Patient Sample Preparation:
   a. Prepare 1+9 dilutions of the patient plasma or control plasma in Owren's Buffer.
   b. Mix without shaking.
3. Testing:
   a. Perform all tests in duplicate.
   b. Pipette 0.2 mL of standard, patient or control dilution into a reaction tube and incubate at 37°C for 2 minutes.
   c. Add 0.1 mL of thrombin (~15 – 30 IU).
   d. Determine the clot time to the nearest 0.1 seconds.
   e. Plot mean Standard Clot Time (Y-axis) versus Fibrinogen Level (X-axis) on the Fibrinogen graph paper. A straight line should be obtained. Assign the Fibrinogen Calibrator reference value to the 1+9 dilution to allow direct interpolation of patient and control values from the standard curve.

Automated Method

Refer to the appropriate instrument Operator Manual for detailed instructions or contact Helena Biosciences Europe for instrument specific application guides.

INTERPRETATION OF RESULTS

Expected values for fibrinogen in healthy adults are 150-350 mg/dL (1.5-3.5 g/L)[4,5].

LIMITATIONS

Heparin levels >0.6 units/mL and fibrinolytic degradation products >100 mg/mL may cause falsely low fibrinogen quantitation. Analysers fail outside the standard curve values for the salient samples. re-assay using an appropriate dilution to bring values into the standard range.

QUALITY CONTROL

Each laboratory should establish a quality control program. Normal and abnormal control plasmas should be tested prior to each batch of patient samples, to ensure satisfactory instrument and operator performance. If controls do not perform as expected, patient results should be considered invalid. Helena Biosciences Europe supply the following controls available for use with this product:

REF 5301	Specialty Assayed Control N
REF 5302	Specialty Assayed Control A
REF 5186	Routine Control N
REF 5187	Routine Control A
REF 5183	Routine Control SA

REFERENCE VALUES

Reference values can vary between laboratories depending on the techniques and systems in use. For this reason each laboratory should establish its own normal range.

PERFORMANCE CHARACTERISTICS

Helena Biosciences Europe or their representatives have determined the following performance characteristics as a guideline. Each laboratory should establish its own performance data. The manual Clauss Fibrinogen is designed to give a linear calibration from 1.5 – 6.5 g/L.

Reproducibility

Intra-assay precision *Fibrinogen (g/L)*	*n*	*CV (%)*	Inter-assay precision *Fibrinogen (g/L)*	*n*	*CV (%)*
1.24	5	4.88	1.02	10	6.17
3.01	5	3.58	3.62	10	3.75

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Clauss Fibrinogen 100
Fiche technique

fr

UTILISATION

Le kit Clauss Fibrinogen 100 est destiné à la réalisation des analyses de l'hémostase basées sur la formation de caillots.

Clauss[1] a développé une méthode simple de détermination quantitative du fibrinogène en mesurant le temps de coagulation de plasma dilué après l'ajout de Thrombin (>30 unités NIH/mL). Ce temps de coagulation est proportionnel à la concentration en fibrinogène. Le taux de fibrinogène peut augmenter en raison d'une inflammation, d'une grossesse ou de la prise de contraceptifs oraux[2]. Des taux diminués sont trouvés dans certaines pathologies comme une maladie du foie ou une CIVD. L'afibrinogénémie (pas de fibrinogène détectable), l'hypofibrinogénémie (<1 mg/mL) et la dysfibrinogénémie (molécule de fibrinogène anormales) sont des anomalies congénitales. Le kit de Fibrinogène Clauss 100 est utilisé pour la détermination quantitative du fibrinogène dans le plasma humain.

AVERTISSEMENTS ET PRÉCAUTIONS

Les réactifs du kit sont à usage diagnostique *in vitro* uniquement – NE PAS INGERER. Porter un équipement de protection individuelle approprié lors de la manipulation de tous les composants du kit. Consulter la fiche de données de sécurité du produit pour obtenir le lien vers les phrases de risque et les conseils de prudence le cas échéant. Éliminer les composants conformément aux réglementations locales.

Un dépistage des produits sanguins a été réalisé et a donné un résultat négatif (sauf indication contraire sur la boîte du kit ou sur le flacon) quant à la présence de :

Antigène de l'hépatite B (AgHBs)
Anticorps anti-VIH 1
Anticorps anti-VIH 2
Anticorps anti-VHC

Cependant, ils doivent être manipulés avec les mêmes précautions que celles prises pour les échantillons patients humains.

COMPOSITION

Composant	Contient	Description	Préparation
Thrombine 100 NIH/mL	5 x 2 mL (REF 5376) 5 x 1 mL (REF 5376H)	Chaque flacon contient environ 200 (REF 5376) / 100 (REF 5376H) unités de Thrombin bovine additionnés de stabilisateurs.	Reconstituer chaque flacon avec : 2 mL (REF 5376) 1 mL (REF 5376H) d'eau distillée ou de Kaolin Suspension en fonction de la méthode utilisée, manuelle ou spécifique de l'instrument. Faire attention lors du pipetage pour éviter toute contamination. Remuer doucement et laisser reposer 15 minutes. Bien mélanger juste avant utilisation. Ne pas agiter.
Fibrinogen Calibrator	2 x 1 mL (REF 5376) 1 x 1 mL (REF 5376H)	Chaque flacon contient 1 mL de plasma humain normal lyophilisé dosé pour les taux de fibrinogène.	Reconstituer chaque flacon avec exactement 1,0 mL d'eau distillée. Remuer doucement et laisser reposer 10 minutes. Mélanger doucement avant utilisation. Ne pas agiter.
Owren's Buffer	2 x 25 mL (REF 5376) 1 x 25 mL (REF 5376H)	Chaque flacon contient 25 mL de tampon contenant du barbital et du chlorure de sodium additionné d'azide de sodium comme conservateur.	Le tampon est prêt à l'emploi.
Kaolin Suspension: 0.5 g/L	2 x 5 mL (REF 5376) 1 x 5 mL (REF 5376H)	Contient 5 mL de Kaolin Suspension à 0.5 g/L.	Mélanger vigoureusement juste avant utilisation. S'assurer qu'il n'y a pas de sédimentation visible.

Chaque kit contient une fiche technique.
Chaque kit contient valeurs de référence spécifiques du lot.

MATÉRIEL NÉCESSAIRE NON FOURNI

Les Helena Biosciences Europe réactifs de dosage du fibrinogène sont appropriés pour être utilisé avec un analyseur de la coagulation pouvant réaliser le dosage du fibrinogène Clauss (ne doit pas être utilisé avec AC-K, C-1, C-2, C-4). Se référer au manuel d'utilisation de l'instrument et/ou aux notes d'application pour avoir des instructions appropriées.

Les réactifs du kit sont aussi disponibles séparément:

REF 5374	Clauss Fibrinogen (Thrombin only)
REF 5378	Clauss Fibrinogen (Thrombin only)
REF 5379	Fibrinogen Calibrator
REF 5375	Owren's Buffer
REF 5376S	Kaolin Suspension

CONSERVATION, DURÉE DE VIE UTILE ET STABILITÉ

Les flacons non ouverts sont stables jusqu'à la date de péremption indiquée s'ils sont conservés dans les conditions indiquées sur l'étiquette du kit ou du flacon.

Thrombin: 100 NIH/mL	Une fois reconstitué, le réactif est stable 8 heures à température ambiante, 1 semaine entre 2 – 8°C ou 1 mois à -20°C.
Fibrinogen Calibrator	Une fois reconstitué, le réactif est stable 4 heures entre 2 – 8°C.
Owren's Buffer	Conserver entre 2 – 8°C une fois ouvert.
Kaolin Suspension	Conserver entre 2 – 8°C une fois ouvert.

PRÉLÈVEMENT ET PRÉPARATION DES ÉCHANTILLONS

Utiliser tout au long du prélèvement du plastique ou du verre siliconé. Mélanger 9 volumes de sang et 1 volume de citrate de sodium à 3.2% ou 3.8%. Séparer le plasma après centrifugation à 1500 x g pendant 15 minutes. Conserver le plasma entre 2 – 8°C ou 18 – 24°C. L'analyse doit être terminée dans les 4 heures suivant le prélèvement de l'échantillon. sinon, il est possible de congeler le plasma 2 semaines à -20°C or 6 mois à -70°C. Décongeler rapidement à 37°C avant de réaliser l'analyse. Ne pas laisser à 37°C plus de 5 minutes[3].

PROCEDURE

Méthode Manuelle

Préparer tous les réactifs en suivant les indications du paragraphe "Composition".

1. Préparation de la courbe d'étalonnage:
   a. Il faut utiliser une nouvelle courbe d'étalonnage lorsque vous utilisez un nouveau numéro de lot de réactif ou si les valeurs dans le contrôle qualité changent. Préparer les dilutions suivantes du Owren's Buffer (Mélanger sans agiter):

Tube	Dilution	Étalon Fibrinogène (mL)	Tampon (mL)
1	1 + 4	0.2	0.8
2	1 + 9	0.1	0.9
3	1 + 19	0.1	1.9
4	1 + 29	0.1	2.9
5	1 + 39	0.1	3.9

2. Préparation de l'échantillon patient:
   a. Préparer des dilutions 1+9 du plasma du patient ou du plasma de contrôle avec du Owren's Buffer.
   b. Mélanger sans agiter.
3. Analyse:
   a. Réaliser toutes les analyses en double.
   b. Pipeter 0.2 mL de dilution étalon, patient ou contrôle dans un tube à essai et incuber à 37°C pendant 2 minutes.
   c. Ajouter 0.1 mL de Thrombin (~15 – 30°C).
   d. Déterminer le temps de coagulation en arrondissant au dixième de seconde.
   e. Tracer une courbe avec le temps moyen de coagulation de l'étalon (en ordonnée) et le taux de fibrinogène (en abscisse) sur le papier millimétré du fibrinogène. Vous devez obtenir une ligne droite. Assigner la valeur de référence de l'Fibrinogen Calibrator à la dilution 1+9 afin de réaliser une interpolation directe des valeurs patient et contrôle à partir de la courbe d'étalonnage.

Méthodes Automatisées

Consulter le manuel d'utilisation de l'instrument approprié pour obtenir des instructions détaillées ou contacter Helena Biosciences Europe pour obtenir des notes d'application spécifiques à l'instrument.

INTERPRÉTATION DES RÉSULTATS

Le taux prévu de fibrinogène chez un adulte sain est de 150-350 mg/dL (1.5–3.5 g/L)[4,5].

LIMITES

Des taux d'héparine >0,6 unités/mL et de produits de dégradation fibrinolytique >100 mg/mL peuvent donner une quantification du fibrinogène erroné(e) faussement bas(se). Si les valeurs de l'échantillon patient se situent hors des valeurs de la courbe d'étalonnage, réaliser à nouveau l'analyse en utilisant la dilution appropriée pour les amener dans la plage de l'étalon.

CONTRÔLE QUALITÉ

Chaque laboratoire doit établir un programme de contrôle qualité. Les plasmas de contrôle, normaux et anormaux, doivent être testés avant chaque lot d'échantillons patients afin de s'assurer que l'instrument et l'opérateur offrent des performances satisfaisantes. Si les contrôles ne donnent pas les résultats prévus, les résultats de patient doivent être considérés comme non valables. Helena Biosciences Europe distribue les contrôles suivants à utiliser avec ce produit:

REF 5301	Specialty Assayed Control N
REF 5302	Specialty Assayed Control A
REF 5186	Routine Control N
REF 5187	Routine Control A
REF 5183	Routine Control SA

VALEURS DE RÉFÉRENCE

Les valeurs de référence peuvent varier d'un laboratoire à l'autre suivant les techniques et les systèmes utilisés. C'est pour cette raison qu'il appartient à chaque laboratoire de déterminer sa propre plage normale.

CARACTÉRISTIQUES DE PERFORMANCES

Helena Biosciences Europe ou ses représentants ont déterminé à titre indicatif les performances suivantes. Chaque laboratoire doit établir ses propres données de performance. Le dosage manuel du fibrinogène est conçu pour donner un étalonnage linéaire sur la plage 1,5 – 6,5 g/L.

Reproductibilité

Précision intra-série *Fibrinogène (g/L)*	*n*	*CV (%)*	Précision inter-séries *Fibrinogène (g/L)*	*n*	*CV (%)*
1,24	5	4,88	1,02	10	6,17
3,01	5	3,58	3,62	10	3,75

BIBLIOGRAPHIE

1. Clauss A (1957) Gerinnungsphysiologische Schnellmethode zur bestimmung des Fibrinogens. *Acta Haematol.* 17:237-246
2. Shaw TS (1977) Assays for Fibrinogen and its Derivatives CRC. *Crit. Rev. Clin. Lab. Sci.* 8:145-192.
3. Clinical and Laboratory Standards Institute (2008) Collection, Transport and Processing of Blood Specimens for Testing Plasma-Based Coagulation Assays and Molecular Haemostasis Assays; Approved Guideline, 5th edn. CLSI: H21-A5
4. Scully RE et al. (1980) Normal Reference Laboratory Values. *New England Journal Medicine*, 302(37):37-48.
5. Okuno T, Sakenko V (1972) Plasma fibrinogen determination by automated thrombin time. *American Journal of Medical Technology*, 38(6):196-201

Clauss Fibrinogen 100
Anleitung

de

VERWENDUNGSZWECK

Das Clauss Fibrinogen 100-Kit ist für klassenallmähliche Gerinnungstests vorgesehen.

Clauss[1] entwickelte zur quantitativen Bestimmung von Fibrinogen eine einfache Methode, bei der die Gerinnungszeit von verdünntem Plasma nach Zugabe von Thrombin gemessen und (>30 NIH Einheiten/mL). Diese Gerinnungszeit ist der Fibrinogenkonzentration direkt proportional. Das Fibrinogen kann durch Entzündung, Schwangerschaft oder Einnahme von oralen Verhütungsmitteln erhöht sein[2]. Vermindert wird es durch Lebererkrankung und Verbrauchskoagulopathie. Angeborene Mangelzustände sind insbesondere Afibrinogenämie (kein nachweisbares Fibrinogen), Hypofibrinogenämie (<1mg/mL) und Dysfibrinogenämie (abnormales Fibrinogenmolekül). Das Clauss Fibrinogen 100 dient der quantitativen Bestimmung von Fibrinogen im Humanplasma.

WARNHINWEISE UND VORSICHTSMASSNAHMEN

Die in diesem Kit enthaltenen Reagenzien sind ausschließlich für die Verwendung von in-vitro-Diagnosen vorgesehen – NICHT VERSCHLUCKEN. Tragen Sie beim Umgang mit sämtlichen Komponenten des Kits geeignete Schutzausrüstung. Beachten Sie gegebenenfalls die Verweise auf entsprechende Gefahren- und Vorsorgemaßnahmen in der Produktgefahrbestätigung. Entsorgen Sie die Komponenten gemäß den örtlichen Vorschriften.

Die Blutprodukte wurden untersucht und sind für folgende Sone ohne Befund (soweit nicht anderweitig auf der Verpackung oder den Ampullen angegeben):
Hepatitis-B-Antikörper (HbsAg)
HIV-Antikörper 1
HIV-Antikörper 2
HCV-Antikörper
Sie sind jedoch mit den gleichen Vorkehrungen zu behandeln wie Proben von menschlichen Patienten.

ZUSAMMENSETZUNG

Komponente	Inhalt	Beschreibung	Vorbereitung
Thrombin 100 NIH/mL	5 x 2 mL (REF 5376) 5 x 1 mL (REF 5376H)	Jedes Fläschchen enthält ca. 200 (REF 5376) / 100 (REF 5376H) Einheiten Rinder-Thrombin mit Stabilisatoren.	Jedes Fläschchen mit: 2 mL (REF 5376) 1 mL (REF 5376H) destilliertem Wasser oder Kaolinsuspension je nach manueller oder gerätespezifischer Methode rekonstituieren. Kontamination beim Pipettieren vermeiden. Leicht schwenken und 15 Minuten stehen lassen. Vor Gebrauch gut mischen. Nicht schütteln.
Fibrinogen Calibrator	2 x 1 mL (REF 5376) 1 x 1 mL (REF 5376H)	Jedes Fläschchen enthält 1 mL lyophilisiertes normales Humanplasma mit bekanntem Fibrinogen-Gehalt	Jedes Fläschchen mit genau 1 mL destilliertem Wasser rekonstituieren. Leicht schwenken und 10 minuten stehen lassen. Vor Gebrauch leicht mischen. Nicht schütteln.
Owren's Buffer	2 x 25 mL (REF 5376) 1 x 25 mL (REF 5376H)	Jede Flasche enthält 25 mL Puffer aus Barbital- und Natrium(chlor) mit Natriumazid als Konservierungsmittel	Der Puffer ist gebrauchsfertig verpackt.
Kaolin Suspension 0.5 g/L	2 x 5 mL (REF 5376) 1 x 5 mL (REF 5376H)	Enthält 5 mL Kaolinsuspension bei 0.5 g/L.	Unmittelbar vor Gebrauch kräftig mischen. Sicher stellen, dass keine Sedimentation zu sehen ist.

Jedes Kit enthält eine Gebrauchsanweisung.
Jedes Kit enthält chargenspezifischen Referenzwerten.

ERFORDERLICHE, ABER NICHT MITGELIEFERTE ARTIKEL

Die Helena Biosciences Europe Fibrinogen Test-Reagenzien können auf jedem Gerinnungsanalyzer verwendet werden, auf dem man Fibrinogen-Bestimmungen nach Clauss durchführen kann (nicht zum Gebrauch mit dem AC-K, C-1, C-2, C-4). Zur Bedienung siehe Bedienungsanleitung des Geräts und/oder Anwendungshinweise.

Die Reagenzien dieses Kits sind auch einzeln erhältlich:

REF 5374	Clauss Fibrinogen (Thrombin only)
REF 5378	Clauss Fibrinogen (Thrombin only)
REF 5379	Fibrinogen Calibrator
REF 5376	Owren's Buffer
REF 5376S	Kaolin Suspension

LAGERUNG, HALTBARKEIT UND STABILITÄT

Ungeöffnete Fläschchen sind unter den auf Verpackung oder Fläschchen angegebenen Lagerbedingungen bis zum aufgedruckten Verfallsdatum stabil.

Thrombin: 100 NIH/mL	Nach Rekonstitution ist das Reagenz 8 Stunden bei Raumtemperatur, 1 Woche bei 2 – 8°C stabil oder 1 monate bei -20°C stabil.
Fibrinogen Calibrator	Rekonstituiert ist das Reagenz bei einer Temperatur von 2 – 8°C für 4 Stunden stabil.
Owren's Buffer	Nach dem Öffnen bei 2 – 8°C lagern.
Kaolin Suspension	Nach dem Öffnen bei 2 – 8°C lagern.

PROBENENTNAHME UND VORBEREITUNG

Nur Plastik oder Silikonglas verwenden. Blut (9 Teile) sollte in 3,2% oder 3,8%-Natriumcitrat als Antikoagulans (1 Teil) entnommen werden. 15 Minuten bei 1500 g zentrifugieren und Plasma abpipettieren. Plasma bei 2 – 8 oder 18 – 24 °C lagern. Plasma sollte innerhalb von 4 Stunden verarbeitet oder bei gefroren bei -20°C für 2 Wochen oder -70°C für 6 Monat gelagert werden. Vor dem Testen schnell bei 37°C auftauen. Nicht länger als 5 Minuten bei 37°C belassen[3].

VORGEHENSWEISE

Manuelle Methode

Alle Reagenzien wie unter „Inhalt" beschrieben vorbereiten.

1. Erstellung der Standardkurve.
   a. Für jede neue Reagenzien-Charge, oder wenn sich die Normalwerte der Qualitätskontrolle ändern, muss eine neue Standardkurve erstellt werden. Folgende Verdünnungen mit Owren's Buffer herstellen (Gut durchschütteln mischen):

Röhrchen	Verdünnung	Fibrinogen-Kalibrator (mL)	Puffer (mL)
1	1 + 4	0.2	0.8
2	1 + 9	0.1	0.9
3	1 + 19	0.1	1.9
4	1 + 29	0.1	2.9
5	1 + 39	0.1	3.9

2. Vorbereitung der Patientenprobe:
   a. Eine 1+9 Verdünnung des Patienten- oder Kontrollplasmas mit Owren's Buffer herstellen.
   b. Ohne zu schütteln mischen.
3. Testdurchführung:
   a. Alle Tests im Doppelansatz durchführen.
   b. 0,2 mL Standard, Patienten- oder Kontrollverdünnung in ein Röhrchen pipettieren und für 2 Minuten bei 37°C inkubieren.
   c. 0,1 mL Thrombin (~15 – 30 IU) hinzufügen.
   d. Die Gerinnungszeit bis auf 0,1 Sekunden genau bestimmen.
   e. Den Mittelwert der Standardgerinnungszeit (Y-Achse) gegen Fibrinogen-Wert (X-Achse) auf Fibrinogen-Millimeterpapier auftragen. Es sollte eine gerade Linie erzeugt werden. Den Referenzwert des Fibrinogen-Kalibrators der 1+9 Verdünnung zuordnen, um eine direkte Interpolation von Patienten- und Kontrollwerten aus der Standardkurve zu ermöglichen.

Automatisierte Methoden

Siehe die Bedienungsanleitung des entsprechenden Geräts für genaue Anweisungen oder wenden Sie sich an Helena Biosciences Europe für spezielle anwendungstechnische Hinweise.

INTERPRETATION DER ERGEBNISSE

Fibrinogen-Normalwerte bei gesunden Erwachsenen sind 150-350 mg/dL (1,5-3,5 g/L)[4,5].

EINSCHRÄNKUNGEN

Heparinwerte >0,6 Einheiten/mL und Fibrinolyse-Abbauprodukte >100 mg/mL können falsch niedrige Fibrinogen-Mengen ergeben. Liegen bei Patientenproben die Werte außerhalb der Standardkurve, die Proben erneut mit geeigneter Probenverdünnung testen, um die Werte in den Bereich der Standardkurve zu bringen.

QUALITÄTSKONTROLLE

Jedes Labor muss für eine eigene Qualitätskontrolle sorgen. Vor jedem Testlauf mit Patientenproben müssen normale und pathologische Kontrollplasmen getestet werden, um eine zufrieden stellende Gerätefunktion und Bedienung zu gewährleisten. Liegen die Kontrollen außerhalb des Normbereichs, sind die Patientenergebnisse nicht zu verwenden. In Verbindung mit diesem Produkt bietet Helena Biosciences Europe die folgenden Kontrollen an:

REF 5301	Specialty Assayed Control N
REF 5302	Specialty Assayed Control A
REF 5186	Routine Control N
REF 5187	Routine Control A
REF 5183	Routine Control SA

REFERENZWERTE

Referenzwerte können je nach Technik und verwendetem System von Labor zu Labor unterschiedlich sein. Aus diesem Grund sollte jedes Labor seinen eigenen Normalbereich erstellen.

LEISTUNGSMERKMALE

Die folgenden Leistungseigenschaften wurden von Helena Biosciences Europe oder in deren Auftrag ermittelt. Jedes Labor muss seine eigenen Werte ermitteln.
Der manuelle Fibrinogentest ist so konzipiert, dass sich eine lineare Kalibration von 1,5 – 6,5 g/L ergibt.

Reproduzierbarkeit

Intra-assay-Präzision *Fibrinogen (g/L)*	*n*	*CV (%)*	Inter-assay-Präzision *Fibrinogen (g/L)*	*n*	*CV (%)*
1,24	5	4,88	1,02	10	6,17
3,01	5	3,58	3,62	10	3,75

LITERATURVERZEICHNIS

1. Clauss A (1957) Gerinnungsphysiologische Schnellmethode zur bestimmung des Fibrinogens. *Acta Haematol.* 17:237-246
2. Shaw TS (1977) Assays for Fibrinogen and its Derivatives CRC. *Crit. Rev. Clin. Lab. Sci.* 8:145-192.
3. Clinical and Laboratory Standards Institute (2008) Collection, Transport and Processing of Blood Specimens for Testing Plasma-Based Coagulation Assays and Molecular Haemostasis Assays; Approved Guideline, 5th edn. CLSI: H21-A5
4. Scully RE et al. (1980) Normal Reference Laboratory Values. *New England Journal Medicine*, 302(37):37-48.
5. Okuno T, Sakenko V (1972) Plasma fibrinogen determination by automated thrombin time. *American Journal of Medical Technology*, 38(6):196-201

Clauss Fibrinogen 100
Istruzioni per l'uso

it

SCOPO PREVISTO

Il kit Clauss Fibrinogen 100 è concepito per l'esecuzione di dosaggi di emostasi basati sulla presenza di coagulo.

Clauss[1] ha messo a punto un semplice metodo per la determinazione quantitativa del fibrinogeno misurando il tempo di coagulazione del plasma diluito in seguito all'aggiunta di Thrombin (≥30 NIH units/mL). Questo tempo di coagulazione è proporzionale alla concentrazione di fibrinogeno. I livelli di fibrinogeno possono aumentare in seguito ad infiammazione, gravidanza o impiego di contraccettivi orali[2]. Livelli ridotti di fibrinogeno sono riscontrabili in alcuni stati, come ad esempio le patologie epatiche e la CID. Tra le carenze congenite rientrano l'afibrinogenemia (assenza di fibrinogeno rilevabile), l'ipofibrinogenemia (< 1 mg/mL) e la disfibrinogenemia (molecola di fibrinogeno anomala). Il kit di Fibrinogeno Clauss 100 è stato formulato per la determinazione quantitativa del fibrinogeno in plasma umano.

AVVERTENZE E PRECAUZIONI

I reagenti contenuti in questo kit sono destinati esclusivamente alla diagnostica in vitro - NON INGERIRE. Indossare un'adeguata attrezzatura protettiva personale durante la manipolazione di tutti i componenti del kit. Per conoscere i relativi rischi e precauzioni e di pericolo, fare pertinente, fare riferimento alla dichiarazione di sicurezza del prodotto. Smaltire i componenti conformemente alle normative locali vigenti.

I prodotti ematici sono stati sottoposti a screening e risultati negativi (salvo diversa indicazione sulle confezioni del kit o sulla bolla) per la presenza di:

Antigene dell'epatite B (HBsAg)
Anticorpo HIV 1
Anticorpo HIV 2
Anticorpo HCV

Questi prodotti devono tuttavia essere maneggiati con le stesse misure precauzionali adottate per un campione paziente umano.

COMPOSIZIONE

Componente	Contiene	Descrizione	Preparazione
Thrombin: 100 NIH/mL	5 x 2 mL (REF 5376) 5 x 1 mL (REF 5376H)	Ogni flacone contiene approssimativamente 200 (REF 5376) / 100 (REF 5376H) unità di Thrombin bovina con stabilizzatori.	Ricostituire ogni flacone con: 2 mL (REF 5376) 1 mL (REF 5376H) di acqua distillata o Kaolin Suspension a seconda della metodologia specifica del manuale o dello strumento. Durante il ripallagon, prestare attenzione ad evitare la contaminazione. Agitare delicatamente e lasciare riposare per 15 minuti. Miscelare bene immediatamente prima dell'uso. Non agitare.
Fibrinogen Calibrator	2 x 1 mL (REF 5376) 1 x 1 mL (REF 5376H)	Ogni flacone contiene 1 mL di plasma umano normale liofilizzato dosato per i livelli di fibrinogeno.	Ricostituire ogni flacone con esattamente 1,0 mL di acqua distillata. Agitare delicatamente e lasciare riposare per 10 minuti. Miscelare delicatamente prima dell'uso. Non scuotere.
Owren's Buffer	2 x 25 mL (REF 5376) 1 x 25 mL (REF 5376H)	Ogni flacone contiene 25 mL di tampone contenente barbital e sodio cloruro con sodio azide come conservante.	Il tampone è pronto all'uso così come viene fornito.
Kaolin Suspension: 0.5 g/L	2 x 5 mL (REF 5376) 1 x 5 mL (REF 5376H)	Contiene 5 mL di sospensione di caolino a 0,5 g/L.	Miscelare energicamente immediatamente prima dell'uso. Assicurarsi che non vi siano sedimenti visibili.

Ogni kit contiene un foglietto per l'uso.

Ogni kit contiene un inserto recante i valori di riferimento specifici per il lotto.

MATERIALI NECESSARI MA NON IN DOTAZIONE

I reagenti di dosaggio del fibrinogeno Helena Biosciences Europe sono idonei all'uso in qualsiasi analizzatore di coagulazione in grado di eseguire i dosaggi del fibrinogeno di Clauss (da non utilizzare con ACL-1, C-1, C-2, C-4). Fare riferimento al manuale utente dello strumento ed alla nota applicativa per conoscere le istruzioni specifiche.

Reagenti contenuti in questo kit sono disponibili anche come materiale separato:

REF 5374	Clauss Fibrinogen (Thrombin only)
REF 5375	Clauss Fibrinogen (Thrombin only)
REF 5376	Fibrinogen Calibrator
REF 5375	Owren's Buffer
REF 53765	Kaolin Suspension

CONSERVAZIONE, VITA UTILE E STABILITÀ

I flaconi non aperti sono stabili fino alla data di scadenza indicata se conservati nelle condizioni riportate sull'etichetta o sull'esterno del kit.

Thrombin: 100 NIH/mL	Dopo la ricostituzione, il reagente è stabile per 8 ore a temperatura ambiente, 1 settimana a 2 – 8°C o per 1 mese a -20°C.
Fibrinogen Calibrator	Dopo la ricostituzione, il reagente è stabile per 4 ore a 2 – 8°C.
Owren's Buffer	Conservare a 2 – 8°C dopo l'apertura.
Kaolin Suspension	Conservare a 2 – 8°C dopo l'apertura.

RACCOLTA E PREPARAZIONE DEI CAMPIONI

Nel corso dell'intera procedura è necessario utilizzare plastica o vetro siliconizzato. Il sangue (9 parti) deve essere raccolto in sodio citrato al 3,2% o al 3,8% come anticoagulante (1 parte). Separare il plasma in seguito a centrifugazione a 1500 x g per 15 minuti. Il plasma può essere conservato a 2 – 8°C o 18 – 24°C. I test devono essere completati entro 4 ore dalla raccolta dei campioni; in alternativa, il plasma può essere conservato congelato a -20°C per 2 settimane o a -70°C per 6 mesi. Decongelare rapidamente a 37°C prima di eseguire i test. Non conservare a 37°C per oltre 5 minuti[3].

PROCEDURA

Metodo Manuale

Preparare tutti i reagenti secondo le istruzioni riportate nel paragrafo "Composizione".

1. Preparazione della curva standard
   a. Quando i lotti di lotto dei reagenti cambiano o se i valori previsti per i test del controllo qualità variano, è necessario utilizzare una nuova curva standard. Preparare le seguenti diluizioni nel Owren's Buffer (Mezclar sin agitar):

Provetta	Dilución	Calibrador per fibrinogeno (mL)	Tampone [mL]
1	1 + 4	0.2	0.8
2	1 + 9	0.1	0.9
3	1 + 19	0.1	1.9
4	1 + 29	0.1	2.9
5	1 + 39	0.1	3.9

2. Preparazione dei campioni dei pazienti
   a. Preparare 1+9 diluizioni di plasma del paziente o di plasma di controllo nel Owren's Buffer.
   b. Miscelare senza scuotere.
3. Esecuzione del test.
   a. Eseguire tutti i test per 2 volte.
   b. Pipettare 0,2 mL di diluizione standard del paziente o di controllo in una provetta di reazione ed incubare a 37°C per 2 minuti.
   c. Aggiungere 0,1 mL di Thrombin (~15 – 30°C).
   d. Determinare il tempo di coagulazione con un'approssimazione a 0,1 secondi.
   e. Tracciare i tempi di coagulazione standard media (sull'asse Y) rispetto ai livelli di fibrinogeno (sull'asse X) sulla carta/algoritmo del fibrinogeno. Si dovrà ottenere una linea retta. Assegnare il valore di riferimento del Fibrinogen Calibrator alla diluizione 1+9. È poi possibile l'interpolazione diretta dei valori del paziente e di controllo dalla curva standard.

Metodo Automatico

Fare riferimento al manuale utente dello strumento appropriato per istruzioni dettagliate oppure contattare Helena Biosciences Europe per le note applicative specifiche dello strumento.

INTERPRETAZIONE DEI RISULTATI

Nei soggetti adulti sani i valori previsti per il fibrinogeno sono di 150-350 mg/dL (1,5-3,5 g/L)[4,5].

LIMITAZIONI

I livelli di plasma >6,0 unità/mL e i prodotti fibrinolitici di degradazione >100 mg/mL possono causare una quantificazione del fibrinogeno falsamente bassa. Se i valori superiscono dei valori della curva standard per i campioni dei pazienti, risalire a dosaggio utilizzando una diluizione appropriata per portare i valori all'interno del range standard.

CONTROLLO QUALITÀ

Ogni laboratorio deve definire un programma di controllo qualità. I plasmi di controllo normali e anormali devono essere testati prima di ogni lotto di campioni di pazienti, per garantire un livello prestazionale soddisfacente sia per quanto riguarda la sfumerità che per l'operatore. Qualora i controlli non funzionassero come previsto, i risultati relativi ai pazienti non sono stati correttamente non validi. Helena Biosciences Europe mette a disposizione i seguenti controlli utilizzabili con questo prodotto:

REF 5301	Specialty Assayed Control N
REF 5302	Specialty Assayed Control A
REF 5186	Routine Control N
REF 5187	Routine Control A
REF 5183	Routine Control SA

VALORI DI RIFERIMENTO

I valori di riferimento possono variare da un laboratorio all'altro in funzione delle tecniche e dei sistemi in uso. Per tale motivo ciascun laboratorio dovrà elaborare un proprio range normale.

CARATTERISTICHE PRESTAZIONALI

Le caratteristiche prestazionali sotto riportate sono state determinate da Helena Biosciences Europe e dai propri rappresentanti a titolo di linee guida. Ciascun laboratorio dovrà pertanto elaborare i propri dati prestazionali. Il dosaggio fibrinogeno manuale è concepito per offrire una calibrazione lineare da 1,5-6,5 g/L.

Riproducibilità

Precisione intra-dosaggio			Precisione tra i dosaggi		
Fibrinogeno (g/L)	*n*	*CV (%)*	*Fibrinogeno (g/L)*	*n*	*CV (%)*
1.24	5	4.88	1.02	10	6.17
3.01	5	3.56	3.62	10	3.75

BIBLIOGRAFIA

1. Clauss A (1957) Gerinnungsphysiologische Schnellmethode zur bestimmung des Fibrinogens. *Acta Haematol.* 17:237-246
2. Shaw TS (1977) Assays for Fibrinogen and its Derivatives CRC, *Crit. Rev. Clin. Lab. Sci.* 8:145-192.
3. Clinical and Laboratory Standards Institute (2008) Collection, Transport and Processing of Blood Specimens for Testing Plasma-Based Coagulation Assays and Molecular Haemostasis Assays' Approved Guideline, 5th edn. CLSI: H21-A5.
4. Scully RE *et al.* (1960) Normal Reference Laboratory Values. *New England Journal Medicine.* 302(37):37-48.
5. Okuno T, Sekenka V (1972) Plasma fibrinogen determination by automated thrombin time. *American Journal of Medical Technology.* 38(6):196-201

Clauss Fibrinogen 100
Instrucciones de uso

es

USO PREVISTO

El uso previsto del kit Clauss Fibrinogen 100 es realizar ensayos de hemostasia basados en la coagulación.

Clauss[1] desarrolló un método simple para la determinación cuantitativa de fibrinógeno midiendo el tiempo de coagulación del plasma diluido después de la adición de Thrombin (≥30 unidades NIH/mL). Este tiempo de coagulación es proporcional a la concentración de fibrinógeno. Los niveles de fibrinógeno pueden aumentar como consecuencia de inflamación, embarazo o uso de anticonceptivos orales[2]. Pueden encontrarse niveles disminuidos en determinados estados como la hepatopatía y la CID. Entre las deficiencias congénitas están la afibrinogenemia (sin fibrinógeno detectable), la hipofibrinogenemia (< 1 mg/mL) y la disfibrinogenemia (molécula de fibrinógeno anómala). El kit de Fibrinógeno de Clauss 100 está prevista para la determinación cuantitativa del fibrinógeno en el plasma humano.

ADVERTENCIAS Y PRECAUCIONES

Los reactivos que contiene este kit son sólo para uso de diagnóstico in vitro- NO INGERIR. Lleve el equipo de protección personal adecuado cuando utilice todos los componentes del kit. Consulte la declaración de seguridad del producto para saber más sobre las indicaciones adecuadas de advertencia y riesgo. Deseche los componentes de conformidad con las normativas locales.

La sangre se ha sometido a pruebas que han resultado negativas (a menos que se indique lo contrario en la caja del kit o en el vial) de la presencia de:

Antígeno de la hepatitis B (HBsAg)
Anticuerpos del VIH 1
Anticuerpos del VIH 2
Anticuerpos del VHC

Sin embargo, deben manipularse con las mismas precauciones que una muestra de un paciente.

COMPOSICIÓN

Componente	Contiene	Descripción	Preparación
Thrombin: 100 NIH/mL	5 x 2 mL (REF 5376) 5 x 1 mL (REF 5376H)	Cada vial contiene aproximadamente 200 (REF 5376) / 100 (REF 5376H) unidades de trombina bovina con estabilizadores.	Reconstituya cada vial con: 2 mL (REF 5376) 1 mL (REF 5376H) de agua purificada o suspensión de Kaolin dependiendo de la metodología específica manual o de cada instrumento. Tener cuidado al pipetear para evitar la contaminación. Agitar suavemente y permitir que repose durante 15 minutos. Mezclar bien inmediatamente antes de usar. No agitar.
Fibrinogen Calibrator	2 x 1 mL (REF 5376) 1 x 1 mL (REF 5376H)	Cada vial contiene 1 mL de plasma humano normal liofilizado ajustado en cuanto a niveles de fibrinógeno.	Reconstituir cada vial con exactamente 1,0 mL de agua destilada. Agitar suavemente y permitir que repose durante 10 minutos. Mezclar suavemente antes de su uso. No agitar.
Owren's Buffer	2 x 25 mL (REF 5376) 1 x 25 mL (REF 5376H)	Cada frasco contiene 25 mL de tampón con barbital y cloruro sódico con azida sódica como conservante.	El tampón viene envasado listo para usar.
Kaolin Suspension: 0.5 g/L	2 x 5 mL (REF 5376) 1 x 5 mL (REF 5376H)	Contiene 5 mL de suspensión de kaolín a 0,5 g/L.	Mezclar vigorosamente inmediatamente antes de usar. Comprobar que no hay sedimentación visible.

Cada kit contiene instrucciones de uso.

Cada kit contiene valores de referencia específicos insertados del lote.

ARTÍCULOS NECESARIOS NO SUMINISTRADOS

Los reactivos de la valoración del fibrinógeno de Helena Biosciences Europe son adecuados para usarse en cualquier analizador de coagulación capaz de realizar valoraciones del fibrinógeno de Clauss (no debe usarse con el ACL-1, C-1, C-2, C-4). Consúltese el Manual del Operador del instrumento y/o la nota de aplicación para instrucciones adecuadas.

Los reactivos contenidos en este kit están también disponibles como artículos separados:

REF 5374	Clauss Fibrinogen (Thrombin only)
REF 5378	Clauss Fibrinogen (Thrombin only)
REF 5376	Fibrinogen Calibrator
REF 5375	Owren's Buffer
REF 53765	Kaolin Suspension

ALMACENAMIENTO, CADUCIDAD Y ESTABILIDAD

Los viales no abiertos son estables hasta la fecha de caducidad indicada cuando se conservan en las condiciones indicadas en el vial o en la etiqueta del kit.

Thrombin: 100 NIH/mL	El reactivo reconstituido permanece estable durante 8 horas a temperatura ambiente, 1 semana a 2 –8°C o 1 meses a -20°C.
Fibrinogen Calibrator	Una vez reconstituido, el reactivo permanece estable durante 4 horas a 2 – 8°C.
Owren's Buffer	Conservar a 2 – 8°C una vez abierto.
Kaolin Suspension	Conservar a 2 – 8°C una vez abierto.

RECOGIDA Y PREPARACIÓN DE LAS MUESTRAS

Debe usarse siempre plástico o vidrio siliconizado. Debe recogerse sangre (9 partes) en el anticoagulante citrato sodico al 3,2% o al 3,8% (1 parte). Separar el plasma después de la centrifugación a 1500 x g durante 15 minutos. El plasma debe conservarse a 2 – 8°C o 18 –24°C. Las pruebas deberían terminarse en 4 horas desde la recogida de las muestras o el plasma puede conservarse congelado a -20°C durante 2 semanas o -70°C durante 6 mes. Descongelar rápidamente a 37°C antes de realizar la prueba. No conservar a 37°C durante más de 5 minutos[3].

PROCEDIMIENTO

Método Manual

Preparar todos los reactivos como se indica en "composición".

1. Preparación de la curva estándar:
   a. Debe usarse una nueva curva estándar cuando los números de lote de los reactivos cambien o si los valores esperados en las pruebas de control de calidad cambian. Preparar las siguientes diluciones en Owren's Buffer (Mezclar sin agitar):

Tubo	Dilución	Calibrador del fibrinógeno (mL)	Tampón [mL]
1	1 + 4	0.2	0.8
2	1 + 9	0.1	0.9
3	1 + 19	0.1	1.9
4	1 + 29	0.1	2.9
5	1 + 39	0.1	3.9

2. Preparación de la muestra del paciente:
   a. Preparar 1 + 9 diluciones del plasma del paciente o el plasma control en Owren's Buffer.
   b. Mezclar sin agitar.
3. Realizacion de las pruebas:
   a. Realizar todas las pruebas por duplicado.
   b. Pipetear 0,2 mL de solución estándar, del paciente o control en un tubo de reacción e incubar a 37°C durante 2 minutos.
   c. Añadir 0,1 mL de Thrombin (~15 – 30°C).
   d. Determinar el tiempo de coagulación en los 0,1 segundos más próximos.
   e. Dibujar el tiempo de coagulación estándar (eje Y) frente al nivel del fibrinógeno (eje X) en el papel de gráfico del fibrinógeno. Debe obtenerse una línea recta. Asignar el valor de referencia del Fibrinogen Calibrator a la dilución 1 + 9 para permitir la interpolación directa de los valores del paciente y control a partir de la curva estándar.

Método Automatizado

Consulte el manual del usuario del instrumento adecuado para instrucciones detalladas o póngase en contacto con Helena Biosciences Europe para notas de aplicación específicas del instrumento.

INTERPRETACIÓN DE LOS RESULTADOS

Los valores esperados para el fibrinógeno en adultos sanos son de 150-350 mg/dL (1,5-3,5 g/L)[4,5].

LIMITACIONES

Los niveles de heparina >0,6 unidades/mL y los productos de degradación fibrinolíticos >100 mg/mL pueden producir una cuantificación falsamente baja del fibrinógeno. Si los valores caen fuera de los valores de la curva estándar para las muestras de pacientes, vuelva a realizar la valoración usando una dilución adecuada para llevar los valores al intervalo estándar.

CONTROL DE CALIDAD

Cada laboratorio debe establecer un programa de control de calidad. Los controles normales y anormales deben analizarse antes de cada lote de muestras de paciente, para asegurar un funcionamiento adecuado del instrumento y el operador. Si los controles no se realizan como se esperaba, los resultados del paciente deben considerarse inválidos. Helena Biosciences Europe suministra los siguientes controles disponibles para usar con este producto:

REF 5301	Specialty Assay Control N
REF 5302	Specialty Assay Control A
REF 5186	Routine Control N
REF 5187	Routine Control A
REF 5183	Routine Control SA

VALORES DE REFERENCIA

Los valores de referencia pueden variar entre los laboratorios dependiendo de las técnicas y sistemas usados. Por esta razón, cada laboratorio debe establecer su propio intervalo normal.

CARACTERÍSTICAS FUNCIONALES

Helena Biosciences European sus representantes han determinado las siguientes características de rendimiento como directrices. Cada laboratorio debe establecer sus propios datos de rendimiento. La valoración del fibrinógeno manual está diseñada para dar una calibración lineal de 1,5 a 6,5 g/L.

Reproducibilidad

Precisión intra-ensayo			Precisión inter-ensayo		
Fibrinógeno (g/L)	*n*	*CV (%)*	*Fibrinógeno (g/L)*	*n*	*CV (%)*
1.24	5	4.88	1.02	10	6.17
3.01	5	3.56	3.62	10	3.75

BIBLIOGRAFÍA

1. Clauss A (1957) Gerinnungsphysiologische Schnellmethode zur bestimmung des Fibrinogens. *Acta Haematol.* 17:237-246
2. Shaw TS (1977) Assays for Fibrinogen and its Derivatives CRC, *Crit. Rev. Clin. Lab. Sci.* 8:145-192.
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4. Scully RE *et al.* (1960) Normal Reference Laboratory Values. *New England Journal Medicine.* 302(37):37-48.
5. Okuno T, Sekenka V (1972) Plasma fibrinogen determination by automated thrombin time. *American Journal of Medical Technology.* 38(6):196-201

Тест-система "Определение фибриногена методом Клусса 100"
инструкция

ru

НАЗНАЧЕНИЕ

Комплект "Определение фибриногена методом Клусса 100" предназначен для проведения анализов гемостаза на основе коагуляционного сгустка.

Метод Клусса[1] – простой метод количественного определения фибриногена, основанный на измерении времени образования сгустка. Метод проводится в разведенной пробе после добавления тромбинового реагента (≥ 30 NIH/мл). Время образования сгустка пропорционально концентрации фибриногена. Уровень фибриногена может увеличиваться при беременности, воспалении или приеме пероральных контрацептивов[2]. Низкие значения могут наблюдаться при заболеваниях печени и ДВС. Изменение содержания фибриногена в плазме показано диагностика врожденных дефектных протеинов, включая афибриногенемию (полное отсутствие фибриногена), гипофибриногенемию (концентрация фибриногена менее 1 г/л) и дисфибриногенемию (структурные нарушения в молекуле фибриногена). Тест-система "Определение фибриногена методом Клусса 100" предназначена для проведения количественного определения фибриногена по Клусса в плазме крови человека на полуавтомате.

ПРЕДУПРЕЖДЕНИЯ И МЕРЫ ПРЕДОСТОРОЖНОСТИ

Содержащиеся в данном наборе реагенты предназначены только для in vitro диагностики- НЕ ПРОГЛАТЫВАТЬ. Внутри. При работе со всеми компонентами набора используйте соответствующие средства индивидуальной защиты. В случае необходимости см. свидетельство о безопасности изделия для ознакомления с соответствующими опасными воздействиями и связанными с ними предосторожности. Удаление компонентов в строгом соответствии с местными правилами.

Препараты крови были подвергнуты скринингу и получены отрицательный результат (если не указано в конкретно упаковке комплект или на пробирке) на указанные ниже:

Антиген к гепатиту В (HbsAg)
Антитела к ВИЧ 1
Антитела к ВИЧ 2
Антитела к вирусу гепатита С (HCV)

Тем не менее с ними следует обращаться, соблюдая те же меры предосторожности, что и при обращении с образцом, полученным от человека.

СОСТАВ

Компоненты	Состав набора	Описание	Приготовление реагентов
Тромбиновый Реагент: 100 NIH/мл	5 x 2 мл (Кат. № 5376) 5 x 1 мл (Кат. № 5376H)	содержит 200 единиц (Кат. № 5376) / 100 единиц (Кат. № 5376H) бычьего тромбина со стабилизаторами.	Внесите во флакон: 2,0 мл (Кат. № 5376) или 1,0 мл (Кат. № 5376H) дистиллированной триэтаноламиновой буферной или каолиновой суспензии (0,5 г/л) в зависимости от протокола и конкретного прибора. Осторожно пипетировать и не допускать попадания посторонних веществ. Аккуратно перемешать и оставить при комнатной температуре в течение 15 минут. Перед использованием еще раз перемешать. Не встряхивать.
Калибратор Фибриногена	2 x 1 мл (Кат. № 5376) 1 x 1 мл (Кат. № 5376H)	Содержит 1,0 мл лиофилизированной нормальной плазмы человека, отрегулированной по уровню фибриногена.	Внесите во флакон 1,0 мл дистиллированной или деионизированной воды. Осторожно перемешать и оставить при комнатной температуре в течение 10 минут. Перед использованием еще раз перемешать. Не встряхивать.
Буфер Оуренса	2 x 25 мл (Кат. № 5376) 1 x 25 мл (Кат. № 5376H)	Флакон содержит 25,0 мл готового к использованию буфера, содержащего натрия хлорид и натрия азид в качестве стабилизаторов.	Дополнительных разведений не требуется.
Каолиновая суспензия 0,5 г/л	2 x 5 мл (Кат. № 5376) 1 x 5 мл (Кат. № 5376H)	Содержит 5 мл каолиновой суспензии с концентрацией 0,5 г/л.	Перед использованием перемешать. Не встряхивать. Убедитесь в отсутствии видимого осаждения и материала.

Каждый набор содержит инструкции по применению.

Паспорт с результатами испытаний.

НЕОБХОДИМЫЕ КОМПОНЕНТЫ, НЕ ВКЛЮЧЕННЫЕ В КОМПЛЕКТ ПОСТАВКИ

Тест-система "Определение фибриногена методом Клусса 100" может использоваться на любом анализаторе свертываемости, способном определять фибриноген по Клусса. (Не рекомендуется использовать на Helena C-1/2/4/ACL-6). Адаптации реагентов и различные пробирки доступны по запросу у производителя дистрибьюторов в компании Хелена.

Реагенты, входящие в набор могут быть заказаны отдельно:

Кат. № 5374	Тест-система "Фибриноген по Клауссу (только тромбиновый реагент)"
Кат. № 5378	Тест-система "Фибриноген по Клауссу (только тромбиновый реагент)"
Кат. № 5376	Калибратор фибриногена
Кат. № 5375	Тестовый реагент "Буфер Оуренса"
Кат. № 53765	Тестовый реагент "Фибриноген по Клауссу, суспензия каолиновая"

ХРАНЕНИЕ, СРОК ГОДНОСТИ И УСТОЙЧИВОСТЬ

Неоткрытые флаконы хранятся до истечения срока годности в условиях, указанных на упаковке или этикетке.

Тромбиновый: 100 МЕ/мл	После растворения, тромбиновый реагент стабилен в течение 7 дней при 2 – 8°C и 8 часов на борту анализатора при 15 – 30°C, и 1 месяц при -20°C.
Калибратор Фибриногена	После растворения, калибратор стабилен в течение 4 часов при 2 – 8°C.
Буфер Оуренса	Вскрытый флакон с буфером следует хранить при 2 – 8°C.
Каолиновая суспензия	Вскрытый флакон с буфером следует хранить при 2 – 8°C.

ОТБОР И ПОДГОТОВКА ОБРАЗЦОВ

Для работы следует использовать только пластиковую или силиконированную стеклянную посуду. Кровь забирается в пробирку с цитратным антикоагулянтом (3,2% или 3,8% раствор натрия) в соотношении 9 + 1. После центрифугирования при 1500 g в течение 15 минут осторожно отделяют плазму. Другие параметры должны применяться лабораторией), полученную плазму необходимо отделить от форменных элементов крови. Плазму следует хранить при температуре 2 – 8°C или 18 – 24°C. Тестирование должно быть проведено в течение 4 часов после забора образцов, либо плазму можно однократно заморозить и хранить при температуре -20°C в течение 2 недель или при -70°C в течение 6 месяцев. Перед проведением исследования плазму следует быстро разморозить при 37°C. Не держать плазму при 37°C более 5 минут[3].

ПРОЦЕДУРА

Ручной Метод

1. Подготовьте все реактивы, см. выше раздел Приготовление реагентов.
2. Новая калибровка должна использоваться при изменении номера серии реагентов или если ожидаемые значения контроля выходят за пределы референсных значений. Разведите калибратор буфером Оуренса как указано в таблице:

Пробирки	Разведение	Калибратор Фибриногена (мл)	Буфер (мл)
1	1 + 4	0,2	0,8
2	1 + 9	0,1	0,9
3	1 + 19	0,1	1,9
4	1 + 29	0,1	2,9
5	1 + 39	0,1	3,9

3. Подготовка образца
   a. Смешайте 1 часть плазмы пациента или контроля с 9 частями буфера Оуренса
   b. Аккуратно перемешайте. Не встряхивайте!
4. Выполнение теста в дубле
   a. Отберите пипеткой 0,2 мл стандарта, пробы пациента или контроля в пробирки для реакции и инкубируйте в течение 2 минут при 37°C.
   b. Добавьте 0,1 мл тромбинового реагента (~15 – 30 °C).
   c. Определите время свертывания с точностью 0,1 секунды.
   d. На миллиметровой бумаге (отложений в набор реагентов) отложите по оси Y значения времени свертывания стандартов, а по оси X значения концентрации фибриногена. Должна быть получена прямая линия. Определите значение калибратора фибриногена – разведение 1+9, интерполируйте получение значения с целью контроля стандартной кривой.

Автоматизированный Метод

Для получения более подробной информации обратитесь к инструкции по эксплуатации для полуавтоматического коагулометр.

ИНТЕРПРЕТАЦИЯ РЕЗУЛЬТАТОВ

Средний уровень фибриногена в популяции практически здоровых людей находится между 150-350 мг/дл (1,5-3,5 г/л)[4,5].

ОГРАНИЧЕНИЯ

На низко точные результаты концентрации фибриногена, полученные на коагулометре, может влиять наличие в концентрации >0,6 Ед/мл и ПДФ в концентрации >100 мг/мл. Если значения образцов пациентов не входят в график стандартной кривой, то требуется дальнейшее разведение образца, чтобы данные попадали в линейный диапазон от 1,0 до 6,5 г/л.

КОНТРОЛЬ КАЧЕСТВА

Каждая лаборатория должна установить программу контроля качества. Перед применением каждой партии образцов пациентов необходимо протестировать с нормальными и аномальными контрольными образцами и удовлетворительной работе оборудования и оператора. Если контрольные измерения не совпадают с ожидаемыми значениями, то измеренные данные пациентов следует считать недействительными. Helena Biosciences рекомендует следующие контрольные материалы:

Кат. № 5301	Контроль качества специальные тесты, норма
Кат. № 5302	Контроль качества специальные тесты, патология
Кат. № 5186	Контроль качества, норма
Кат. № 5187	Контроль качества, умеренно нарушенная патология
Кат. № 5183	Контроль качества, высокая патология

НОРМАЛЬНЫЕ ПОКАЗАТЕЛИ

Референсные значения могут варьировать между лабораториями в зависимости от используемых методов и компонентов. По этой причине каждая лаборатория должна устанавливать свои собственные значения.

ЭКСПЛУАТАЦИОННЫЕ ХАРАКТЕРИСТИКИ

Компания Хелена, как ее дистрибьюторы определили следующие ориентировочные аналитические характеристики. Каждая лаборатория должна определять свои собственные аналитические характеристики. При использовании ручного метода было определено следующее соотношение величин (CV) и воспроизводимости в диапазоне от 1,0 до 6,5 г/л (без дополнительных разведений плазмы).

Воспроизводимость

в пределах аналитической серии			между разными аналитическими сериями		
Фибриноген (г/л)	*n*	*CV (%)*	*Фибриноген (г/л)*	*n*	*CV (%)*
1,24	5	4,88	1,02	10	6,17
3,01	5	3,56	3,62	10	3,75

ЛИТЕРАТУРА

1. Clauss A (1957) Gerinnungsphysiologische Schnellmethode zur bestimmung des Fibrinogens. *Acta Haematol.* 17:237-246
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5. Okuno T, Sekenka V (1972) Plasma fibrinogen determination by automated thrombin time. *American Journal of Medical Technology.* 38(6):196-201

Declaration of Conformity

HL-7-0135DC DOI 2015/07 (7)

In Application of the Council Directive 98/79/EC on the approximation of the laws of the Member States relating to *In Vitro* Diagnostic Medical Devices & CE marking.

Declaration of conformance to applicable sections of Annex I - Essential Requirements and Annex III (EC Declaration of Conformity) imposed by sections 2 to 5. The below listed products are not classified under Annex II Lists A or B. Access to the appropriate technical files will be made available to the appropriate body in the event this is required.

Product Code	Description	GMDN Classification Code
5183	Routine Control SA	30590

I, the undersigned declare that the devices registered against the above GMDN Classification Code conforms to the said Directives.

Full Name: M.J. Stephenson Title: Managing Director

Signed: Date: 28 Jul 2015

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Declaration of Conformity

HL-7-0137DC DOI 2015/07 (7)

In Application of the Council Directive 98/79/EC on the approximation of the laws of the Member States relating to *In Vitro* Diagnostic Medical Devices & CE marking.

Declaration of conformance to applicable sections of Annex I - Essential Requirements and Annex III (EC Declaration of Conformity) imposed by sections 2 to 5. The below listed products are not classified under Annex II Lists A or B. Access to the appropriate technical files will be made available to the appropriate body in the event this is required.

Product Code	Description	GMDN Classification Code
5186	Routine Control N	30590

I, the undersigned declare that the devices registered against the above GMDN Classification Code conforms to the said Directives.

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Declaration of Conformity

HL-7-0138DC DOI 2015/07 (7)

In Application of the Council Directive 98/79/EC on the approximation of the laws of the Member States relating to *In Vitro* Diagnostic Medical Devices & CE marking.

Declaration of conformance to applicable sections of Annex I - Essential Requirements and Annex III (EC Declaration of Conformity) imposed by sections 2 to 5. The below listed products are not classified under Annex II Lists A or B. Access to the appropriate technical files will be made available to the appropriate body in the event this is required.

Product Code	Description	GMDN Classification Code
5187	Routine Control A	30590

I, the undersigned declare that the devices registered against the above GMDN Classification Code conforms to the said Directives.

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Coagulation Control Plasmas

REF 5186 Routine Control N
REF 5187 Routine Control A
REF 5183 Routine Control SA
REF 5482 Routine Coagulation Control Set

CE IVD

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HL-2-0482P 2016/01 (16)

Coagulation Control Plasmas
Instructions for use

en

INTENDED PURPOSE

The Coagulation Control Plasmas kit is intended for use as a quality control material.

Routine Control N, Routine Control A and Routine Control SA are for use as normal, moderately prolonged and markedly prolonged controls for PT and aPTT assays. They are also assayed for Fibrinogen, TCT and ATIII, and are prepared from normal human plasma.

WARNINGS AND PRECAUTIONS

The reagents contained in this kit are for *in vitro* diagnostic use only – DO NOT INGEST. Wear appropriate personal protective equipment when handling all kit components. Refer to the product safety declaration for the link to appropriate hazard and precautionary statements where applicable. Dispose of components in accordance with local regulations.

Blood products have been screened and found negative (unless otherwise stated on the kit box or vial) for the presence of:
Hepatitis B Antigen (HbsAg)
HIV 1 antibody HIV 2 antibody
HCV antibody
However they should be handled with the same precautions as a human patient sample.

COMPOSITION

REF	Component	Content	Description
5186	Routine Control - N	10 x 1 mL	Prepared from pooled normal plasma.
5187	Routine Control - A	10 x 1 mL	Prepared from adsorbed human plasma.
5183	Routine Control - SA	10 x 1 mL	Prepared from adsorbed human plasma.
5482	Routine Coagulation Control Set:		
	Routine Control - N	4 x 1 mL	
	Routine Control - A	3 x 1 mL	
	Routine Control - SA	3 x 1 mL	

Each kit contains instructions for use.
Each kit contains lot specific reference values insert.

Each vial contains 1 mL of buffered, lyophilised human plasma.
Preparation: Reconstitute each vial of the appropriate control with 1 mL of distilled or deionised water. Swirl gently. Allow to stand for 10 minutes for complete dissolution and mix well before use.

ITEMS REQUIRED BUT NOT PROVIDED

Coagulation Control Plasmas may be used when performing tests on any mechanical or photo-optical coagulation instrument in conjunction with all suitable, commercial reagents.

STORAGE, SHELF-LIFE AND STABILITY

Unopened vials are stable until the given expiry date when stored under conditions indicated on the vial or kit label. The reconstituted controls are stable for 8 hours when kept at 2 –8°C or 4 weeks at -20°C when flash frozen. Keep covered.

SAMPLE COLLECTION AND PREPARATION

Not applicable.

PROCEDURE

Each control should be treated in the same manner as the unknown specimen in accordance with the instructions outlined in each particular test protocol.

INTERPRETATION OF RESULTS

Routine Control N should give values within the laboratory normal range for PT, aPTT and fibrinogen assays. Routine Control A and Routine Control SA have been standardised to give prolonged and markedly prolonged PT and aPTT times respectively. Lot and instrument specific expected values are provided with each pack of controls.

LIMITATIONS

The results obtained with Coagulation Control Plasmas depend on several factors strongly associated with instrumentation, types of reagents, deficient substrates and laboratory to laboratory variations[1,2,3]. Each laboratory should establish an expected range for the particular instrument-reagent system.

QUALITY CONTROL

Each laboratory should establish a quality control program. Normal and abnormal control plasmas should be tested prior to each batch of patient samples, to ensure satisfactory instrument and operator performance. If controls do not perform as expected, patient results should be considered invalid.

REFERENCE VALUES

Reference values can vary between laboratories depending on the techniques and systems in use. For this reason each laboratory should establish its own reference ranges.

PERFORMANCE CHARACTERISTICS

The following performance characteristics have been determined by Helena Biosciences Europe or their representatives using an opto-mechanical coagulation instrument. Each laboratory should establish its own performance data.

Reproducibility

		Intra-assay precision	
Sample	*n*	*aPTT CV (%)*	*PT CV (%)*
Routine Control N	5	2.83	1.01
Routine Control A	5	2.76	1.71
Routine Control SA	5	1.72	1.03

BIBLIOGRAPHY

1. Kirkwood TBL *et al.* (1977) Identification of Sources of Variation in Factor VIII Assay, *British Journal of Haematology*, 37:559-568.
2. Goldenfarb MD (1971) Reproducibility in Coagulation Assays, *AJCP*, 55:561-564.
3. Palkuti HA and Longberry JR (1973) A Precision Study of Coagulation Factor Assay Techniques, *AJCP*, 59:231-235.

Plasmas de contrôle de coagulation
Fiche technique

fr

UTILISATION

Le kit Coagulation Control Plasmas est destiné à être utilisé comme produit de contrôle qualité.

Les contrôles Routine Control N, Routine Control A et Routine Control SA servent de témoins normal, modérément prolongé et nettement prolongé dans les déterminations du TP et du TCA. Le fibrinogène, la TCT et l'ATIII ont été dosés et ils sont préparés à partir de plasma humain normal.

AVERTISSEMENTS ET PRÉCAUTIONS

Les réactifs du kit sont à usage diagnostique *in vitro* uniquement – NE PAS INGERER. Porter un équipement de protection individuelle approprié lors de la manipulation de tous les composants du kit. Consulter la fiche de données de sécurité du produit pour obtenir le lien vers les phrases de risque et les conseils de prudence le cas échéant. Éliminer les composants conformément aux églementations locales.

Un dépistage des produits sanguins a été réalisé et a donné un résultat négatif (sauf indication contraire sur la boîte du kit ou sur le flacon) quant à la présence de :
Antigène de l'hépatite B (AgHBs)
Anticorps anti-VIH 1
Anticorps anti-VIH 2
Anticorps anti-VHC
Cependant, ils doivent être manipulés avec les mêmes précautions que celles prises pour les échantillons patients humains.

COMPOSITION

REF	Composant	Contient	Description
5186	Routine Control - N	10 x 1 mL	Préparé à partir d'un pool de plasma normal.
5187	Routine Control - A	10 x 1 mL	Préparés à partir de plasma humain adsorbé.
5183	Routine Control - SA	10 x 1 mL	Préparés à partir de plasma humain adsorbé.
5482	Routine Coagulation Control Set:		
	Routine Control - N	4 x 1 mL	
	Routine Control - A	3 x 1 mL	
	Routine Control - SA	3 x 1 mL	

Chaque kit contient une fiche technique.
Chaque kit contient valeurs de référence spécifiques du lot.

Chaque flacon contient 1 mL de plasma humain tamponné lyophilisé.
Préparation: Reconstituer chaque flacon du contrôle approprié avec 1 mL d'eau distillée ou désionisée. Agiter doucement. Attendre 10 minutes jusqu'à dissolution totale et bien mélanger avant d'utiliser.

MATÉRIEL NÉCESSAIRE NON FOURNI

Le Coagulation Control Plasmas peut être utilisé dans les analyses réalisées sur des instruments de coagulation mécanique ou photo-optique avec les réactifs appropriés vendus dans le commerce.

CONSERVATION, DURÉE DE VIE UTILE ET STABILITÉ

Les flacons non ouverts sont stables jusqu'à la date de péremption indiquée s'ils sont conservés dans les conditions indiquées sur l'étiquette du kit ou du flacon. Une fois reconstitués, les contrôles sont stables 8 heures entre 2 –8°C ou 4 semaines à -20 °C en cas de congélation instantanée. Couvrir le produit.

PRÉLÈVEMENT ET PRÉPARATION DES ÉCHANTILLONS

Non applicable.

PROCÉDURE

Chaque contrôle doit être traité de la même manière que l'échantillon à analyser en observant les instructions de chaque protocole spécifique.

INTERPRÉTATION DES RÉSULTATS

Le Routine Control N doit donner des valeurs se situant dans la plage normale du laboratoire pour le TP, le TCA et le fibrinogène. Le Routine Control A et le Routine Control SA ont été standardisés pour donner des temps TP et TCA prolongés et très prolongés respectivement. Les valeurs prévues spécifiques du lot et de l'instrument sont fournies avec chaque kit de contrôles.

LIMITES

Les résultats obtenus avec le Coagulation Control Plasmas dépendent de plusieurs facteurs largement corrélés avec l'instrument, les types de réactifs, les substrats carencés et les variations inter-laboratoires[1,2,3]. Le laboratoire doit déterminer une plage prévue pour chaque système instrument-réactif.

CONTRÔLE QUALITÉ

Chaque laboratoire doit établir un programme de contrôle qualité. Les plasmas de contrôle, normaux et anormaux, doivent être testés avant chaque lot d'échantillons patients afin de s'assurer que l'instrument et l'opérateur offrent des performances satisfaisantes. Si les contrôles ne donnent pas les résultats prévus, les résultats du patient doivent être considérés comme non valables.

VALEURS DE RÉFÉRENCE

Les valeurs de référence peuvent varier d'un laboratoire à l'autre suivant les techniques et les systèmes utilisés. C'est pour cette raison qu'il appartient à chaque laboratoire de déterminer ses propres plages de référence.

CARACTÉRISTIQUES DE PERFORMANCES

Helena Biosciences Europe ou ses mandataires ont déterminé les caractéristiques de performance suivantes en utilisant un instrument de coagulation opto-mécanique. Chaque laboratoire doit établir ses propres données de performance.

Reproductibilité

		Précision intra-série	
Échantillon	*n*	*TCA CV (%)*	*TP CV (%)*
Routine Control N	5	2.83	1.01
Routine Control A	5	2.76	1.71
Routine Control SA	5	1.72	1.03

BIBLIOGRAPHIE

1. Kirkwood TBL *et al.* (1977) Identification of Sources of Variation in Factor VIII Assay, *British Journal of Haematology*, 37:559-568.
2. Goldenfarb MD (1971) Reproducibility in Coagulation Assays, *AJCP*, 55:561-564.
3. Palkuti HA and Longberry JR (1973) A Precision Study of Coagulation Factor Assay Techniques, *AJCP*, 59:231-235.

Kontrollplasma für die Gerinnung
Anleitung

de

VERWENDUNGSZWECK

Das Coagulation Control Plasmas-Kit ist für die Qualitätskontrolle vorgesehen.

Routine Control N, Routine Control A und Routine Control SA sind als normale, mäßig verzögerte und stark verzögerte Kontrollen für PT und aPTT Tests geeignet. Sie sind auch auf Fibrinogen, TZ und AT-III getestet und werden aus normalem Humanplasma hergestellt.

WARNHINWEISE UND VORSICHTSMASSNAHMEN

Die in diesem Kit enthaltenen Reagenzien sind ausschließlich für die Verwendung von *in-vitro*-Diagnosen vorgesehen. NICHT VERSCHLUCKEN. Tragen Sie beim Umgang mit sämtlichen Komponenten des Kits geeignete Schutzausrüstung. Beachten Sie gegebenenfalls die Verweise auf entsprechende Gefahren- und Vorsichtserklärungen in der Produktsicherheitserklärung. Entsorgen Sie die Komponenten gemäß den örtlichen Vorschriften.

Die Blutprodukte wurden untersucht und sind für folgende Gene ohne Befund (soweit nicht anderweitig auf der Verpackung oder den Ampullen angegeben):
Hepatitis-B-Antikörper (HbsAg)
HIV-Antikörper 1
HIV-Antikörper 2
HCV-Antikörper
Sie sind jedoch mit den gleichen Vorkehrungen zu behandeln wie Proben von menschlichen Patienten.

ZUSAMMENSETZUNG

REF	Komponente	Inhalt	Beschreibung
5186	Routine Control - N	10 x 1 mL	Aus gepooltem Humanplasma hergestellt.
5187	Routine Control - A	10 x 1 mL	Adsorbiertem Humanplasma hergestellt.
5183	Routine Control - SA	10 x 1 mL	Adsorbiertem Humanplasma hergestellt.
5482	Routine Coagulation Control Set:		
	Routine Control - N	4 x 1 mL	
	Routine Control - A	3 x 1 mL	
	Routine Control - SA	3 x 1 mL	

Jedes Kit enthält eine Gebrauchsanweisung.
Jedes Kit enthält chargenspezifischen Referenzwerten.

Jedes Fläschchen enthält 1 mL gepuffertes, lyophilisiertes Humanplasma.
Vorbereitung: Jedes Fläschchen Kontrollplasma mit 1 mL destilliertem oder entionisiertem Wasser rekonstituieren. Leicht schwenken. Zum vollständigen Auflösen 10 Minuten stehen lassen und vor Gebrauch gut mischen.

ERFORDERLICHE, ABER NICHT MITGELIEFERTE ARTIKEL

Coagulation Control Plasmas kann in Verbindung mit allen entsprechenden kommerziellen Reagenzien bei der Durchführung von Tests an mechanischen oder lichtoptischen Koagulometern verwendet werden.

LAGERUNG, HALTBARKEIT UND STABILITÄT

Ungeöffnete Fläschchen sind unter den auf Verpackung oder Fläschchen angegebenen Lagerbedingungen bis zum aufgedruckten Verfalldatum stabil. Rekonstituierte Kontrollen sind bei 2 –8°C 4 Wochen stabil bzw. 4 Wochen bei -20 °C, wenn schockgefroren. Verschlossen aufbewahren.

PROBENENTNAHME UND VORBEREITUNG

Entfällt.

VORGEHENSWEISE

Jede Kontrolle sollte gemäß den Anleitungen der einzelnen Testprotokolle wie unbekannte Probe behandelt werden.

INTERPRETATION DER ERGEBNISSE

Routine Control N sollte für PT, aPTT und Fibrinogen Tests Werte im Normalbereich ergeben. Routine Control A und Routine Control SA wurden standardisiert, um verlängerte bzw. stark verlängerte PT und aPTT Zeiten zu ergeben. Chargen und Geräte spezifische Normalwerte sind in jeder Packung mit Kontrollen enthalten.

EINSCHRÄNKUNGEN

Die mit Coagulation Control Plasmas erzielten Resultate hängen von mehreren Faktoren ab, die stark mit dem Gerät, den verwendeten Reagenzien, mangelnden Substraten und Unterschieden zwischen den Labors in Verbindung stehen[1,2,3]. Jedes Labor sollte daher für jedes Geräte-Reagenzien-System einen eigenen Normalwertebereich erstellen.

QUALITÄTSKONTROLLE

Jedes Labor muss für eine eigene Qualitätskontrolle sorgen. Normale und pathologische Kontrollplasmen müssen vor jeder Testreihe mit Patientenproben getestet werden, um eine zufrieden stellende Geräteleistung und Bedienung zu gewährleisten. Liegen die Kontrollen außerhalb des Normbereichs, sind die Patientenergebnisse nicht zu verwenden.

REFERENZWERTE

Referenzwerte können je nach Technik und verwendetem System von Labor zu Labor unterschiedlich sein. Aus diesem Grund sollte jedes Labor seine eigenen Referenzwertbereiche ermitteln.

LEISTUNGSMERKMALE

Die folgenden Leistungseigenschaften wurden von Helena Biosciences Europe oder in ihrem Auftrag mit einem optomechanischen Gerinnungsgerät ermittelt. Jede Labor muss seine eigenen Werte ermitteln.

Reproduzierbarkeit

		Intra-assay-Präzision	
Probe	*n*	*aPTT CV (%)*	*PT CV (%)*
Routine Control N	5	2.83	1.01
Routine Control A	5	2.76	1.71
Routine Control SA	5	1.72	1.03

LITERATURVERZEICHNIS

1. Kirkwood TBL *et al.* (1977) Identification of Sources of Variation in Factor VIII Assay. *British Journal of Haematology*. **37**:559-568.
2. Goldenfarb MD (1971) Reproducibility in Coagulation Assays. *AJCP*. **55**:561-564.
3. Palkuti HA and Longberry JR (1973) A Precision Study of Coagulation Factor Assay Techniques. *AJCP*. **59**:231-235.

Plasmi di controllo della coagulazione
Istruzioni per l'uso

it

SCOPO PREVISTO

Il kit Coagulation Control Plasmas è concepito per l'uso come materiale di controllo qualità.

Routine Control N, Routine Control A e Routine Control SA sono destinati ad essere utilizzati come controlli normali, moderatamente prolungati e marcatamente prolungati per i dosaggi di PT e aPTT. Essi vengono dosati anche per fibrinogeno, TCT e ATIII e sono preparati con plasma umano normale.

AVVERTENZE E PRECAUZIONI

I reagenti contenuti in questo kit sono destinati esclusivamente alla diagnostica *in vitro* - NON INGERIRE. Indossare un'adeguata attrezzatura protettiva personale durante la manipolazione di tutti i componenti del kit. Per conoscere i relativi simboli precauzionali e di pericolo, laddove pertinente, fare riferimento alla dichiarazione di sicurezza del prodotto. Smaltire i componenti conformemente alle normative locali vigenti.

I prodotti ematici sono stati sottoposti a screening e trovati negativi (salvo diversa indicazione sulla confezione del kit o sulla fiala) per la presenza di:
Antigene dell'epatite B (HbsAg)
Anticorpo HIV 1
Anticorpo HIV 2
Anticorpo HCV
Questi prodotti devono tuttavia essere manipolati con le stesse misure precauzionali adottate per un campione paziente umano.

COMPOSIZIONE

REF	Componente	Contiene	Descrizione
5186	Routine Control - N	10 x 1 mL	Preparato con un pool di plasma normale.
5187	Routine Control - A	10 x 1 mL	Preparati con plasma umano adsorbito.
5183	Routine Control - SA	10 x 1 mL	Preparati con plasma umano adsorbito.
5482	Routine Coagulation Control Set:		
	Routine Control - N	4 x 1 mL	
	Routine Control - A	3 x 1 mL	
	Routine Control - SA	3 x 1 mL	

Ogni kit contiene un'istruzioni per l'uso.
Ogni kit contiene un inserto recante i valori di riferimento specifici per il lotto.

Ogni flacone contiene 1 mL di plasma umano tamponato liofilizzato.
Preparazione: Ricostituire ogni flacone di controllo appropriato con 1 mL di acqua distillata o deionizzata. Agitare delicatamente. Attendere 10 minuti per consentire al prodotto di sciogliersi completamente e miscelare bene prima dell'uso.

MATERIALI NECESSARI, MA NON IN DOTAZIONE

Il Coagulation Control Plasmas può essere utilizzato durante l'esecuzione di test su qualsiasi strumento di coagulazione meccanico o foto-ottico in combinazione con tutti i reagenti idonei disponibili in commercio.

CONSERVAZIONE, VITA UTILE E STABILITÀ

Ogni riferimento a nomi e dati utilizzati negli esempi è casuale, se non specificato diversamente. I controlli ricostituiti sono stabili per 8 ore se conservati a °2 — °8°C oppure 4 settimane a -20°C se congelato molto velocemente. Mantenere i prodotti coperti.

RACCOLTA E PREPARAZIONE DEI CAMPIONI

Non applicabile.

PROCEDURA

Ogni controllo deve essere trattato seguendo la stessa procedura adottata per il campione non noto, conformemente alle istruzioni riportate in ciascun protocollo di test specifico.

INTERPRETAZIONE DEI RISULTATI

Routine Control N deve fornire valori compresi nel range normale di laboratorio per i dosaggi di PT, aPTT e fibrinogeno. Routine Control A e Routine Control SA sono stati standardizzati per fornire, rispettivamente, tempi di PT e aPTT prolungati e marcatamente prolungati. I valori previsti specifici per il lotto e lo strumento vengono forniti con ciascuna confezione di controlli.

LIMITAZIONI

I risultati ottenuti con il Coagulation Control Plasmas dipendono da innumerevoli fattori, strettamente legati alla strumentazione, ai tipi di reagenti, a substrati carenti e alle variazioni dovute ai singoli laboratori[1,2,3]. Ogni laboratorio dovrà definire un range di previsione per il sistema strumento-reagente specificamente utilizzato.

CONTROLLO QUALITÀ

Ogni laboratorio deve definire un programma di controllo qualità. I plasmi di controllo normali e anormali devono essere testati prima di ogni lotto di campioni di pazienti, per garantire un livello prestazionale soddisfacente sia per quanto riguarda lo strumento che per l'operatore. Qualora i controlli non funzionassero come previsto, i risultati relativi ai pazienti dovranno essere considerati non validi.

VALORI DI RIFERIMENTO

Per la sicurezza del paziente, è necessario che il sistema sia monitorato continuamente da un operatore qualificato. Per tale motivo ciascun laboratorio dovrà elaborare i propri range di riferimento.

CARATTERISTICHE PRESTAZIONALI

Le seguenti caratteristiche prestazionali sono state determinate da Helena Biosciences Europe o dai propri rappresentanti con l'utilizzo di uno strumento di coagulazione opto-meccanico. Ciascun laboratorio dovrà pertanto elaborare i propri dati prestazionali.

Riproducibilità

		Precisione intra-dosaggio	
Campione	*n*	*aPTT CV (%)*	*PT CV (%)*
Routine Control N	5	2.83	1.01
Routine Control A	5	2.76	1.71
Routine Control SA	5	1.72	1.03

BIBLIOGRAFIA

1. Kirkwood TBL *et al.* (1977) Identification of Sources of Variation in Factor VIII Assay. *British Journal of Haematology*. **37**:559-568.
2. Goldenfarb MD (1971) Reproducibility in Coagulation Assays. *AJCP*. **55**:561-564.
3. Palkuti HA and Longberry JR (1973) A Precision Study of Coagulation Factor Assay Techniques. *AJCP*. **59**:231-235.

Plasmas de control de la coagulación
Instrucciones de uso

es

USO PREVISTO

El uso previsto del kit Coagulation Control Plasmas es como material de control de calidad.

Routine Control N, Routine Control A y Routine Control SA se usan como controles normal, moderadamente prolongado y notablemente prolongado para las valoraciones de TP y TTPa. También se valoran para fibrinógeno, TCT y ATIII y se elaboran a partir de plasma humano normal.

ADVERTENCIAS Y PRECAUCIONES

Los reactivos que contiene este kit son sólo para uso de diagnóstico *in vitro*; NO INGERIR. Lleve el equipo de protección personal adecuado cuando utilice todos los componentes del kit. Consulte la declaración de seguridad del producto para saber más sobre las indicaciones adecuadas de advertencia y riesgo. Desechar los componentes de conformidad con las normativas locales.

La sangre se ha sometido a pruebas que han resultado negativas (a menos que se indique lo contrario en la caja del kit o en el vial) de la presencia de:
Antígeno de la hepatitis B (HbsAg)
Anticuerpos del VIH 1
Anticuerpos del VIH 2
Anticuerpos del VHC
Sin embargo, deben manipularse con las mismas precauciones que una muestra de un paciente.

COMPOSICIÓN

REF	Componente	Contiene	Descripción
5186	Routine Control - N	10 x 1 mL	Elabora a partir de plasma normal de reserva.
5187	Routine Control - A	10 x 1 mL	Elaboran a partir de plasma humano adsorbido.
5183	Routine Control - SA	10 x 1 mL	Elaboran a partir de plasma humano adsorbido.
5482	Routine Coagulation Control Set:		
	Routine Control - N	4 x 1 mL	
	Routine Control - A	3 x 1 mL	
	Routine Control - SA	3 x 1 mL	

Cada kit contiene instrucciones de uso.
Cada kit contiene valores de referencia específicos insertados del lote.

Cada vial contiene 1 mL de plasma humano tamponado, liofilizado.
Preparación: Reconstituya cada vial del control adecuado con 1 mL de agua destilada o desionizada. Agite suavemente. Deje que repose durante 10 minutos para que la disolución sea completa y mezcle bien antes de su uso.

ARTÍCULOS NECESARIOS NO SUMINISTRADOS

El Coagulation Control Plasmas puede usarse cuando se realizan pruebas con cualquier instrumento de coagulación mecánica o foto-óptica junto con todos los reactivos adecuados, comerciales.

ALMACENAMIENTO, CADUCIDAD Y ESTABILIDAD

Los viales no abiertos son estables hasta la fecha de caducidad indicada cuando se conservan en las condiciones indicadas en el vial o en la etiqueta del kit. Los controles reconstituidos son estables durante 8 horas cuando se conservan a °2 — °8°C o durante 4 semanas a -20°C cuando se conserva con congelación instantánea. Manténgase cubierto.

RECOGIDA Y PREPARACIÓN DE LAS MUESTRAS

No aplicable.

PROCEDIMIENTO

Cada control debe tratarse de la misma forma que la muestra desconocida, de acuerdo con las instrucciones indicadas en cada protocolo de prueba concreto.

INTERPRETACIÓN DE LOS RESULTADOS

Routine Control N debe dar valores dentro del intervalo normal de laboratorio para TP, TTPa y valoraciones de fibrinógeno. El Routine Control A y el Routine Control SA han sido estandarizados para dar tiempos de TP y TTPa prolongados y notablemente prolongados, respectivamente. Se aportan los valores esperados específicos de lote y de instrumento con cada paquete de controles.

LIMITACIONES

Los resultados obtenidos con Coagulation Control Plasmas dependen de varios factores fuertemente asociados a la instrumentación, los tipos de reactivos, sustratos deficientes y variaciones entre laboratorios[1,2,3]. Cada laboratorio debe establecer un intervalo esperado para el sistema instrumento-reactivo concreto.

CONTROL DE CALIDAD

Cada laboratorio debe establecer un programa de control de calidad. Los plasmas de control normales y anormales deben estudiarse antes de cada lote de muestras del paciente, para asegurar un funcionamiento adecuado del instrumento y el operador. Si los controles no se realizan como se esperaba, los resultados del paciente deben considerarse inválidos.

VALORES DE REFERENCIA

Los valores de referencia pueden variar entre los laboratorios dependiendo de las técnicas y sistemas usados. Por esta razón, cada laboratorio debe establecer sus propios intervalos de referencia.

CARACTERÍSTICAS FUNCIONALES

Las siguientes características de rendimiento han sido determinadas por Helena Biosciences Europe o sus representantes usando un instrumento de coagulación opto-mecánico. Cada laboratorio debe establecer sus propios datos de rendimiento.

Riproducibilità

		Precisión intra-ensayo	
Muestra	*n*	*TTPa CV (%)*	*TP CV (%)*
Routine Control N	5	2.83	1.01
Routine Control A	5	2.76	1.71
Routine Control SA	5	1.72	1.03

BIBLIOGRAFÍA

1. Kirkwood TBL *et al.* (1977) Identification of Sources of Variation in Factor VIII Assay. *British Journal of Haematology*. **37**:559-568.
2. Goldenfarb MD (1971) Reproducibility in Coagulation Assays. *AJCP*. **55**:561-564.
3. Palkuti HA and Longberry JR (1973) A Precision Study of Coagulation Factor Assay Techniques. *AJCP*. **59**:231-235.

КОНТРОЛЬНЫЕ ПЛАЗМЫ
ИНСТРУКЦИЯ

ru

НАЗНАЧЕНИЕ

Комплект Coagulation Control Plasmas предназначен для использования в качестве материала для контроля качества.

Контрольные плазмы «Контроль качества, норма», «Контроль качества, высокая патология», «Контроль качества, умеренно выраженная патология» – контроли качества с нормальным, умеренно увеличенным и значительно увеличенным активированным парциальным тромбопластиновым временем (АПТВ/АЧТВ) и протромбиновым временем (ПВ). Их также можно использовать для определения количества фибриногена, тромбинового времени (ТВ) и антитромбина III (АТ-III). Контроли приготовлены из человеческой плазмы практически здоровых людей.

ПРЕДУПРЕЖДЕНИЯ И МЕРЫ ПРЕДОСТОРОЖНОСТИ

Содержащиеся в данном наборе реагенты предназначены только для *in vitro* диагностики– НЕ ПРИНИМАТЬ ВНУТРЬ! При работе со всеми компонентами набора использовать соответствующие средства индивидуальной защиты. В случае необходимости см. свидетельство о безопасности изделия для ознакомления с соответствующими описаниями опасного воздействия и сведениями о мерах предосторожности. Удаление компонентов в отходы производите в соответствии с местными правилами.

Препараты крови были подвергнуты скринингу и показали отрицательный результат (если на коробке, в которую упакован компонент или на пробирке не указано иное) на:
Антиген к гепатиту В (HbsAg)
Антитела к ВИЧ 1
Антитела к ВИЧ 2
Антител к вирусу гепатита С (HCV)
Тем не менее с ними следует обращаться, соблюдая те же меры предосторожности, что и при обращении с образцом, полученным от человека.

СОСТАВ

Кат. №	Компоненты	Состав набора	Описание
5186	Контроль качества, норма	10 x 1 мл	приготовлен из пула нормальной плазмы.
5187	Контроль качества, умеренно выраженная патология	10 x 1 мл	приготовлен из адсорбированной плазмы.
5183	Контроль качества, высокая патология	10 x 1 мл	приготовлен из адсорбированной плазмы.
5482	Контроль коагуляции, уровень П1:		
	Контроль качества, норма	4 x 1 мл	
	Контроль качества, умеренно выраженная патология	3 x 1 мл	
	Контроль качества, высокая патология	3 x 1 мл	

Каждый набор содержит инструкцию по применению.
Паспорт с референсными значениями (допустимыми/контрольных пределов).

Каждый флакон содержит 1,0 мл лиофилизированной плазмы человека.
Подготовка: Разведите соответствующую контрольную плазму добавлением во флакон 1,0 мл дистиллированной или деионизированной воды. Осторожно перемешайте и оставьте при комнатной температуре в течение 10 минут. Перед использованием флакон с разведенной плазмой переворачивайте (без встряхивания).

НЕОБХОДИМЫЕ КОМПОНЕНТЫ, НЕ ВКЛЮЧЕННЫЕ В КОМПЛЕКТ ПОСТАВКИ

Контрольные плазмы могут быть использованы с анализаторами, имеющими различный метод детекции (механический, оптический и т.д.) в сочетании с реагентами различных производителей.

ХРАНЕНИЕ, СРОК ГОДНОСТИ И УСТОЙЧИВОСТЬ

Невскрытые флаконы с лиофилизированной плазмой хранятся до истечения срока годности в условиях, указанных на этикетке. Разведенная контрольная плазма в закрытом флаконе может храниться при °2 —°8°C или 4 недель при -20°C в случае мгновенной заморозки, в течение 8 часов.

ОТБОР И ПОДГОТОВКА ОБРАЗЦОВ

Не относится.

ПРОЦЕДУРА

Каждый образец контрольной плазмы должен анализироваться так же, как образец плазмы больного, в соответствии с инструкциями к используемому реагенту и прибору.

ИНТЕРПРЕТАЦИЯ РЕЗУЛЬТАТОВ

«Контроль качества, норма», должен показывать значения в пределах нормального диапазона используемого в лаборатории для ПВ, АПТВ и определения концентрации фибриногена. «Контроль качества, умеренно выраженная патология» и «Контроль качества, высокая патология», были стандартизированы, таким образом, чтобы показывать соответственно умеренно или выраженное увеличение АПТВ и ПВ. Номер партии и допустимые/контрольные пределы для различных коагулометров поставляются с каждым набором контролей.

ОГРАНИЧЕНИЯ

Референсные значения могут варьировать между лабораториями в зависимости от используемых реагентов, методов, коагулометров и других факторов[1,2,3]. По этой причине каждая лаборатория должна установить свои собственные нормативные параметры метода.

КОНТРОЛЬ КАЧЕСТВА

Каждая лаборатория должна установить программу контроля качества. Перед измерением каждой партии образцов пациентов необходимо протестировать нормальную и патологическую плазму, чтобы удостовериться в удовлетворительной работе оборудования и оператора. Если контрольные измерения не совпадают с ожидаемыми значениями, то измеренные данные пациентов следует считать недостоверными.

НОРМАЛЬНЫЕ ПОКАЗАТЕЛИ

Референсные значения могут варьировать между лабораториями в зависимости от используемых методов и коагулометров. По этой причине каждая лаборатория должна установить свои собственные значения.

ЭКСПЛУАТАЦИОННЫЕ ХАРАКТЕРИСТИКИ

Компания Хелена или её дистрибьюторы определили следующие ориентировочные аналитические характеристики. Каждая лаборатория должна определить свои собственные аналитические характеристики.
При использовании оптико-механического коагулометра и реагентов Хелена были определены следующие коэффициенты вариации (CV):

Воспроизводимость

		в пределах аналитической серии	
Контрольная плазма	*Кол-во образцов*	*CV (%) АПТВ*	*CV (%) ПВ*
Контроль качества, норма	5	2.83	1.01
Контроль качества, умеренно выраженная патология	5	2.76	1.71
Контроль качества, высокая патология	5	1.72	1.03

ЛИТЕРАТУРА

1. Kirkwood TBL *et al.* (1977) Identification of Sources of Variation in Factor VIII Assay. *British Journal of Haematology*. **37**:559-568.
2. Goldenfarb MD (1971) Reproducibility in Coagulation Assays. *AJCP*. **55**:561-564.
3. Palkuti HA and Longberry JR (1973) A Precision Study of Coagulation Factor Assay Techniques. *AJCP*. **59**:231-235.

Medi-Test URYXXON® Stick 10

de | In-vitro-Diagnostikum

Teststreifen zum Schnellnachweis von Blut, Urobilinogen, Bilirubin, Protein, Nitrit, Keton, Glucose, pH-Wert, Dichte und Leukozyten im Urin, zur reflektionsphotometrischen Auswertung mit URYXXON® 300, 500 und URYXXON® Relax

Anwendung
Suchtest zur Erkennung von Diabetes, Stoffwechselanomalien, Leberschäden, Verschlusskrankheiten, hämolytischen Erkrankungen sowie Erkrankungen im Bereich der Nieren und Harnwege.

Anwendung nur durch Fachpersonal.

Gebrauchsanleitung
Frischen, unzentrifugierten Harn verwenden und die Harnprobe gut durchmischen. Teststreifen ca. 1 Sekunde in Harn eintauchen. Seitliche Kante am Gefäßrand abstreifen, um überschüssigen Harn zu entfernen. Teststreifen parallel Bedienungsanleitung in das Gerät einlegen. Die Teilfelder werden reflektionsphotometrisch ausgewertet und die Resultate auf dem Bedienfeldmonitor angezeigt. Die Werte entsprechen denen des visuellen Vergleich tabellarischen Konzentrationsbereichen. Bei visueller Bewertung Reaktionsstreifen nach 30–60 Sekunden (Leukozytenfeld nach 60–120 Sekunden) mit der Farbskala vergleichen. Farbveränderungen, die nach mehr als 2 Minuten auftreten, sind ohne Bedeutung. Der Harn sollte nicht länger als 2 Stunden gestanden haben.
Bei der Auswertung sollte beachtet werden, daß nicht in allen Fällen eine völlige Übereinstimmung der URYXXON® 300, 500 und der URYXXON® Relax-Resultate mit den Ergebnissen der visuellen Auswertung gegeben sein muß, da das menschliche Auge Farben anders bewerten kann als das URYXXON® 300, 500 und das URYXXON® Relax-Auswertgerät. Für visuelle Auswertung vorgesehene Medi-Test Harnteststreifen können mit URYXXON® 300, 500 und URYXXON® Relax nicht ausgewertet werden.

Prinzip
Blut: Der Nachweis beruht auf der pseudoperoxidatischen Aktivität des Hämoglobins bzw. Myoglobins, die die Oxidation eines Farbindikators durch ein organisches Hydroperoxid zu einem blaugrünen Farbstoff katalysieren.
Urobilinogen: Das Testfeld enthält ein stabiles Diazoniumsalz, das mit Urobilinogen einen rötlichen Azofarbstoff bildet.
Bilirubin: Durch Kupplung des Bilirubins mit einem Diazoniumsalz im sauren Milieu entsteht ein roter Azofarbstoff.
Protein: Der Test basiert auf dem Prinzip des „Eiweißfehlers" von Indikatoren, d. h. bei einem konstant gepufferten pH-Wert erfolgt der Farbumschlag in Gegenwart von Albumin von gelb nach grünblau. Andere Proteine reagieren mit geringerer Empfindlichkeit.
Nitrit: Mit diesem Test werden nitritbildende Mikroorganismen nachgewiesen, die Nitrat zu Nitrit reduzieren können. Dem Test liegt die Griess'sche Reaktion zugrunde. Das Testpapier enthält ein Amin und eine Kupplungskomponente. Durch Diazotierung mit anschließender Kupplung entsteht ein rot gefärbter Azofarbstoff.
Keton: Der Test beruht auf dem Prinzip der Legal'schen Probe. Acetessigsäure und Aceton reagieren mit Nitroprussidnatrium in alkalischem Medium zu einem violetten Farbkomplex.

Bewertung – Fehlerquellen
Komponentenprüfung: Das Komponentenfeld ist frei von Chemikalien und dient ausschließlich der reflektionsmessischen Auswertung der Testfelder.
Blut: Der Test erfasst Werte ab 5 Erythrozyten/µl. Harn, die einen Konzentration von ca. 0,015 mg Hämoglobin bzw. Myoglobin/dl. Harn entsprechen. Akute Entzündungen werden durch punktformige Verfärbungen des Testfeldes angezeigt. Die Farbvergleichsfelder entsprechen

0 (negativ), ca. 5–10, ca. 50, ca. 250 Ery/µl bzw.
einer Hämoglobinmenge aus ca. 10, ca. 50, ca. 250 Ery/µl.

Normale Konzentrationen von Ascorbinsäure (< 40 mg/dl.) beeinflussen das Testergebnis nicht. Falsch positive Reaktionen können durch Reste peroxidhaltiger oder anderer Reinigungsmittel hervorgerufen werden.
Urobilinogen: Je nach Ergebnis des Urins basiert von Konzentrationen von 0,5 bis 1 mg Urobilinogen/dl. Harn nachweisen. Die normale Ausscheidungsrate liegt bei 1 mg/dl. Werte darüber sind pathologisch. Ein deutlich pathologisches völliges Fehlen von Urobilinogen im Harn liegt sich mit Teststreifen nicht nachweisen. Die Farbfelder sind folgenden Urobilinogenkonzentrationen zugeordnet:

norm. (normal), 2, 4, 8, 12 mg/dl, bzw.
norm. (normal), 35, 70, 140, 280 µmol/l.

Der Nachweis wird durch höhere Konzentrationen an Formaldehyd gehemmt. Längeres Stehen des Harns am Licht kann infolge Oxidation zu erniedrigten oder falsch negativen Werten führen. Zu hohe oder falsch positive Resultate können durch im Harn ausgeschiedene Farbstoffe oder Medikamente verursacht werden. Größere Mengen Bilirubin färben das Testfeld grün.
Bilirubin: Werte ab 0,5 mg/dl. Harn werden angezeigt. Die Farbfelder sind folgenden Bilirubinkonzentrationen zugeordnet:

0 (negativ), 1(+), 2(++), 4(+++) mg/dl, bzw
0 (negativ), 17(+), 35(++), 70(+++) µmol/l.

Einige Harnbestandteile können eine Gelbfärbung des Testfeldes verursachen. Der Nachweis wird durch höhere Konzentrationen an Ascorbinsäure nicht mehr gehemmt. Längeres Stehen des Harns am Licht kann infolge Oxidation zu erniedrigten oder falsch negativen Werten führen. Ausgeschiedene Farbstoffe und Medikamente mit roter Eigenfarbe können ein positives Resultat vortäuschen.
Protein: Der Test erfasst Werte ab 10 mg Protein/dl. Harn. Die Farbfelder sind folgenden Albuminkonzentrationen zugeordnet:

negativ, 30, 100, 500 mg/dl, bzw.
negativ, 0,3, 1,0, 5,0 g/l.

Falsch positive Befunde können bei stark alkalischem Harn (pH > 9), nach Infusionen mit Polyvinylpyrrolidon (Blutersatzmittel), bei der Behandlung mit chinolinhaltigen Präparaten und durch Reste von Desinfektionsmitteln im Uringlas auftreten. Farbstoffe aus Arzneimitteln (z. B. Methylenblau) oder der Fleischprobe der roten Rüben können die Proteinfärbung überdecken.
Nitrit: Der Nachweis erfasst Werte ab 0,05 mg Nitrit/l. Harn, dass Reagenz bedingt einen bakteriellen Harnwegsinfekt. Die Farbentwicklung hängt zwar von der Nitritkonzentration ab, erlaubt aber keine Aussage über den Infektionsgrad. Ein negatives Resultat kann einen Harnwegsinfekt nicht ausschließen.
Falsch negative Resultate können durch hohe Ascorbinsäurekonzentrationen, bei der Antibiotika-Therapie und bei zu niedrigem Nitratgehalt im Harn infolge nitratarmer Kost bzw. starker Verdünnung (Diurese) auftreten. Auch können Keime ohne die Fähigkeit der Nitrit-Bildung vorliegen. Eine falsch positive Reaktionsfarbe kann durch im Harn ausgeschiedene Farbstoffe verursacht werden.
Keton: Acetessigsäure reagiert mit dem Testfeld empfindlicher als Aceton. Werte ab 5 mg/dl. Acetessigsäure bzw. 50 mg/dl. Aceton werden angezeigt. Die Farbfelder sind folgenden Acetessigsäurekonzentrationen zugeordnet:

0 (negativ), 25(+), 100(++), 300(+++) mg/dl, bzw.
0 (negativ), 2,5(+), 10(++), 30(+++) mmol/l.

Phenylketone führen in höherer Konzentration, ergeben aber eine abweichende Färbung. β-Hydroxybuttersäure wird nicht erfasst. Phthaleinverbindungen ertragen sich dem Testfeld falsche Farbtöne.
Glucose: Pathologische Glucosekonzentrationen werden durch einen Umschlag von grün nach blaugrün angezeigt. Geben bis schwach grüne Testfelder sind als negativ (bzw. normal) zu bewerten. Die Farbfelder entsprechen folgenen Glucosekonzentrationen:

neg. (gelb), neg. bzw. normal (gelbgrün), 5%, 10%, 20%, ≥ 1000 mg/dl, bzw.
neg. (gelb), neg. bzw. normal (gelbgrün), 2,8, 5,5, 27,8, ≥ 55,5 mmol/l.

Die Eibung durch Ascorbinsäure (Vitamin C) wurde weitgehend festgelegt. Hemmwirkung zeigt Gentisinsäure. Falsch positive Reaktionen können durch Reste peroxidhaltiger oder anderer Reinigungsmittel hervorgerufen werden.
pH: Bei Gesunden liegt der pH-Wert des Morgenharns meist zwischen pH 5 und 6. Die Farbfelder erlaubt eine deutliche Unterscheidung des pH-Wertes zwischen pH 5 und pH 9.
Dichte: Der Test erlaubt die Bestimmung der Harndichte zwischen 1.000 und 1.030. Der Normalwert für Erwachsene liegt bei normaler Nahrungsaufnahme und Flüssigkeitszufuhr etwa zwischen 1.015 und 1.025.
Da mit dem Teststreifen ermittelte Dichte kann gegenüber anderen Methoden leicht unterschiedliche Werte anzeigen, da z. B. eine Erhöhung der Dichte durch Glucosekonzentrationen > 1000 mg/dl (≥ 56 mmol/l.) nicht erfasst wird. Zu hohe Resultate ergeben sich bei erhöhter Proteinausscheidung. Alkalische Harne mit hohem Gehalt an Pufferbestandteilen lassen zu niedrige Werte erwarten.
Leukozyten: Der Test erfasst Werte ab ca. 10 Leukozyten/µl. Harn. Verfärbungen, die nicht mehr dem negativen Vergleichsfeld zuzuordnen sind, und schwache violette Verfärbungen nach 120 Sekunden müssen positiv bewertet werden. Die Farbvergleichsfelder entsprechen folgenden Leukozytenkonzentrationen:

negativ (normal), 25, 75, 500 Leukozyten/µl.

Eine abgeschwächte Reaktion ist bei Eiweißausscheidungen über 500 mg/dl. und einer Glucosekonzentration über 2 g/dl. sowie bei der Einnahme von Präparaten mit Cephalexin bzw. Gentamycin zu erwarten. Bakterien, Trichomonaden und Erythrozyten reagieren nicht mit diesem Test. Formaldehyd (als Konservierungsmittel) kann zu einer falsch positiven Reaktion führen. Sondere als Konservierungsstoff vermindert die Empfindlichkeit der Reaktion. Ausscheidungen von Eiweiß, Nitritbildnern oder anderen stark gefärbten Verbindungen können die Reaktionsfarbe überdecken. Bei Proteinen mit weiblichen Patienten kann durch vaginalen Ausfluss eine falsch positive Reaktion vorgetäuscht werden.

Reagierende Substanzen
(Menge bzw. Aktivität*/l nach der Imprägnierung)

Blut:		Filtrat:		pH:	
Tetramethylbenzidin	31 µg	Salicylsäure	35 µg	Methylrot	3 mg
Cumolhydroperoxid	315 µg	Gerüstsubstanz	37 µg	Bromthymolblau	10 mg
Urobilinogen:		**Keton:**		**Dichte:**	
Diazoniumsalz	75 µg	Natriumnitroprussid-Natrium	183 µg	Bromthymolblau	42 mg
Bilirubin:		Glycinat		Copolymer	1008 µg
Diazoniumsalz	29 µg	Glucoseoxidase	7 U	**Leukozyten:**	
Protein:		Peroxidase	1 U	Carbonsäureester	16 mg
Tetrabromphenolblau	10 µg	o-Tolidin	85 µg	Diazoniumsalz	14 µg

Qualitätskontrolle bei Anwendung durch Fachpersonal
Eine Überprüfung der Testfelder sollte mit positiven und negativen Kontrolllösungen erfolgen. Die positiven und negativen Kontrollen sollten einmal im Tag, nach Öffnen einer neuen Dose, bei Einsatz einer neuen Testreifenchargе und nach jeweils 30 Tagen zur Prüfung der Lagerbedingungen durchgeführt werden. Jedes Labor sollte seine eigenen Zielwerte für ordnungs Leistungsmäßigkeit festlegen und Testverfahren und Abläufe überprüfen, wenn diese Standards nicht erreicht werden.

Hinweise
Grundsätzlich können einzelne Teststreifenresultate und im Zusammenhang mit anderen ärztlichen Befunden eine definitive Diagnose und eine gezielte Therapie ermöglichen.
Die Auswertung von Medikamenten oder deren Metaboliten auf den Test ist nicht in allen Fällen bekannt. Im Zweifelsfall wird deshalb empfohlen, den Test vor Abnahme der Medikation zu wiederholen.
Zur Harnsammlung nur gut gespülte, saubere Gefäße verwenden. Übliche Harnkonservierungsmittel stören den Test nicht.
Stets nur die notwendige Anzahl an Testfeldern entnehmen. Packung nach der Entnahme sofort wieder fest verschließen. Restkassette nicht benutzen! Testfelder vor Sonnenlicht und Feuchtigkeit schützen. Dose kühl und trocken aufbewahren (Lagertemperatur nicht über + 30 °C). Bei sachgemäßer Lagerung sind die Teststreifen bis zum aufgedruckten Verfalldatum haltbar.

Der Stapfen der Teststreifendose enthält ein ungiftiges Trockenmittel. Sollte es einmal verschluckt werden, reichlich Wasser nachtrinken.
Symptomlosen Vergiftungen finden Sie am Ende der Packungsbeilage.
Entsorgung: Entsorgen Sie die benutzten Testblechen unter Beachtung der geltenden Sicherheitsbestimmungen.
Hersteldosis: Packungen mit 100 und 125 Testreifen
Datum der Überarbeitung: 08/2014

Verwendbar bis / Use by / Fecha de caducidad / A utiliser avant
LOT Chargennr. / Batch identification / Código de lote / Numéro de lot
IVD In-vitro-Diagnostikum / In vitro diagnostic product / Diagnóstico in vitro / Diagnostic in vitro
Dieses Testsystem entspricht der Richtlinie 98/79/EG vom 27. 10. 1998 (IVD-Richtlinie) / These test strips conform to the directive 98/79/EC dated 27.10.1998 (IVD-directive) / Las tiras reactivas corresponden a la norma 98/79/EG del 27.10.1998 (IVD-norma) / Les bandelettes correspondent à la directive 98/79/CE du 27.10.1998 (IVD-directive)
CE

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en | In vitro diagnostic medium

Medi-Test URYXXON® Stick 10

Test strips for rapid determination of blood, urobilinogen, bilirubin, protein, nitrite, ketones, glucose, pH-value, density and leukocytes in urine, the reflectrophotometrical evaluation with URYXXON® 300, 500 and URYXXON® Relax

Use
Screening test for detection of diabetes, metabolic abnormalities, liver diseases, biliary and hepatic obstructions, hemolytic diseases and diseases of kidney and urinary tract.
Only for use by qualified personnel.

Instructions for use
Use fresh and uncentrifuged urine. Shake urine sample well before use. Dip the test strip for approximately 1 second into the urine. Draw it across the rim of the container to remove excess urine. Place the test strips into the instrument according to instructions for use in the manual. The test pads are reflectrophotometrically evaluated and the results are printed out. The results obtained with the URYXXON® 300, 500 and URYXXON® Relax correspond to the concentration ranges indicated on the color chart for visual evaluation. For visual evaluation, please compare color changes with the color chart after 30–60 seconds (leukocyte test field after 60–120 seconds). Color changes that take place after more than 2 minutes are of no significance. When tested the urine should not be older than 2 hours.
Due to the fact, that the human eye evaluates color changes somewhat differently than a URYXXON® 300, 500 and URYXXON® Relax reflectrometer, there can also be differences between these two evaluations. Our standard Medi-Test urine dipsticks for visual evaluations cannot be used for instrumental evaluations with our URYXXON® 300, 500 and URYXXON® Relax reflectometers.

Principle
Blood: The detection is based on the pseudoperoxidative activity of hemoglobin and myoglobin, which catalyze the oxidation of an indicator by an organic hydroperoxide producing a green color.
Urobilinogen: The test paper contains a stable diazonium salt forming a reddish azo compound with urobilinogen.
Bilirubin: A red azo compound is obtained in the presence of acid by coupling of bilirubin with a diazonium salt.
Protein: The test is based on the "protein error" principle of indicators. The test zone is buffered to a constant pH value and changes color from yellow to green/blue in the presence of albumin. Other proteins are indicated with less sensitivity.
Nitrite: Microorganisms, which are able to reduce nitrate to nitrite, are indicated indirectly by this test. The principle of Griess reagent is the basis of this test. The test paper contains an amine and a coupling component. A red colored azo compound is formed by diazotization and subsequent coupling.
Ketones: The test is based on the principle of Legal's test. Acetoacetic acid and acetone form with sodium nitroprusside in alkaline medium a violet colored complex.
Glucose: The detection is based on the glucoseoxidase-peroxidase-chromogen reaction. Apart from glucose, no other compound in urine is known to give a positive reaction.
pH: The test paper contains indicators which clearly change color between pH 5 and pH 9 (from orange to green to turquoise).
Density: The test determines the concentration of ions in urine and shows a good correlation to the refractometrical method. The color of the test strip changes from deep blue in urine with low ionic concentration through green to yellow in urines with high ionic concentrations.
Leukocytes: The test is based on the esterase activity of granulocytes. This enzyme splits carboxylic acid ester. The alcohol constituent released reacts with a diazo salt producing a violet color.

Evaluation – Sources of Error
Compensation area: The compensation test pad is free of any chemicals and is solely used as reference for the reflectometric wash action of the other test pads.
Blood: The minimum sensitivity of the test strip is 5 erythrocytes/µl, which corresponding to approx. 0.015 mg hemoglobin/dl. urine. Intact erythrocytes are indicated by flecky discolorations of the test field. The color fields correspond to the following values:

0 (negative), ca. 5–10, ca. 50, ca. 250 Ery/µl. resp.
hemoglobin concentration out of ca. 10, ca. 50, ca. 250 Ery/µl.

Normal concentrations of ascorbic acid (< 40 mg/dl.) do not influence the test results. Falsely positive reactions can be produced by a residue of peroxide containing cleansing agents.
Urobilinogen: In dependence upon the urine color 0.5 to 1 mg urobilinogen/dl. urine are indicated. 1 mg/dl. is considered to be the normal excretion rate. Higher values are pathological. A complete absence of urobilinogen in the urine, which is physiologically nonlogical, cannot be indicated by the strips. The color fields correspond to the following urobilinogen concentrations:

norm. (normal), 2, 4, 8, 12 mg/dl. or
norm. (normal), 35, 70, 140, 280 µmol/l.

The test will be inhibited by higher concentrations of formaldehyde. Exposure of the urine to light for a longer period of time may lead to lowered or falsely negative results. Too high or falsely positive results can be caused by the presence of drugs/salts or therapeutic dyes in the urine. Larger amounts of bilirubin produce a yellow colorization.
Bilirubin: The minimum sensitivity of the test strip is 0.5 to 0.75 mg bilirubin/dl. urine. The color fields correspond to the following values:

0 (negative), 1(+), 2(++), 4(+++) mg/dL or
0 (negative), 17(+), 35(++), 70(+++) µmol/l.

Some urine contents can produce a yellow colorization of the test strip. Ascorbic acid and nitrite in higher concentrations inhibit the test. Exposure of the urine to light for a longer period of time may lead to lowered or falsely negative results. Too high or falsely positive results can be caused by the presence of drugs/salts or therapeutic dyes in the urine.
Protein: The minimum sensitivity of the test strip is 10 mg protein/dl. urine. The color fields correspond to the following ranges of albumin concentrations:

negative, 30, 100, 300 mg/dL or
negative, 0.3, 1.0, 3.0 g/L

Falsely positive results are possible in alkaline urine samples (pH > 9), after infusions with polyvinylpyrrolidone (blood substitute), after intake of medicaments containing quinine and also by disinfectant residues in the urine sampling vessel. The protein coloration may be masked by the presence of medicaid dyes (e.g. methylene blue) or beetroot pigments.
Nitrite: The test detects concentrations from 0.05 mg nitrite/dl. urine. Every pink color indicates a bacterial infection of the urinary tract. The color intensity depends only on the nitrite concentration, but does not provide information about the extent of the infection. A negative result does not preclude an infection of the urinary tract, if bacteria which can not produce nitrite are present. Falsely negative results can be produced by high doses of ascorbic acid, by antibiotics therapy and by very low nitrate concentrations as come as the result of low nitrate diet or strong dilution (diuresis). Falsely positive results can be caused by the presence of diagnostic or therapeutic dyes in the urine.
Ketones: The test is more sensitive to acetoacetic acid than to acetone. Values of 5 mg/dl. acetoacetic acid or 50 mg/dL acetone are indicated. The color fields correspond to the following acetoacetic acid values:

0 (negative), 25(+), 100(++), 300(+++) mg/dL or
0 (negative), 2.5(+), 10(++), 30(+++) mmol/l.

Phenylketones in higher concentrations interfere with the test, and will produce variable colors.
β-Hydroxybutyric acid is not detected. Phthalein compounds interfere by producing a red coloration.
Glucose: Pathological glucose concentrations are indicated by a color change from green to bluish green. Yellow or greenish test fields should be considered negative or normal. The color fields correspond to the following ranges of glucose concentrations:

neg. (yellow), neg. or normal (greenish), 50, 150, 500, > 1000 mg/dL or
neg. (yellow), neg. or normal (greenish), 2.8, 8.3, 27.8, > 55.5 mmol/l.

The influence of ascorbic acid (vitamin C) has been largely eliminated. An inhibitory effect is produced by gentisic acid. Falsely positive reactions can be produced by a residue of peroxide containing cleansing agents.
pH: The pH value of fresh urine of healthy people varies between pH 5 and pH 6. The color scale gives a clear distinction of pH value between pH 5 and pH 9.
Density: The test permits the determination of urine density between 1.000 and 1.030. Urines from adults with normal fluids and normal food intake will have a density of 1.015 – 1.025.
The chemical nature of the test strip may cause slightly different results from those obtained with other methods when specified amounts of certain urine constituents are present, e.g. the increase of urine density in dependence on glucose concentrations of > 1000 mg/dL (≥ 55 mmol/L) cannot be demonstrated by the strips. Elevated density readings may be obtained in the presence of moderate quantities of protein. Highly buffered alkaline urines may cause low readings.
Leukocytes: The test responds to values starting from approx. 10 leukocytes/µl. urine. Changes in color that can not be assigned to the negative reference field and faint violet colors after 120 seconds must be evaluated as positive. The color reference fields correspond to the following leukocyte concentrations:

negative (normal), 25, 75, 500 leukocytes/µl.

A weakened reaction can be expected in the case of proteinuria of over 500 mg/dl. and a glucose concentration of over 2 g/dl, as well as in the case of patients taking preparations containing cephalexin and gentamycin. Bacteria, trichomonads and erythrocytes do not react with this test. Formaldehyde (as a preservative) can result in a false positive reaction. Boric acid used as preservative decreases the sensitivity of the reaction. Excretion of proteins, nitrite-forming or other strongly-colored compounds may disguise the color of the reaction. Tests with female patients have shown that vaginal discharges can cause a false positive reaction.

Reacting substances
(Quantity resp. activity/cm² at time of impregnation)

Blood:		Nitrite:		pH:	
tetramethylbenzidine	31 µg	sulfanilic acid	35 µg	nitrazine yellow	3 mg
cumene hydroperoxide	315 µg	granular substance	37 µg	bromthymolblue	10 mg
Urobilinogen:		**Ketones:**		**Density:**	
diazonium salt	75 µg	sodium nitroprusside	183 µg	bromthymolblue	42 mg
Bilirubin:		**Glucose:**		copolymer	1008 µg
diazonium salt	29 µg	glucose oxidase	7 U	**Leukocytes:**	
Protein:		peroxidase	1 U	carbonic acid ester	16 mg
tetrabromphenolblue	10 µg	o-tolidine	85 µg	diazonium salt	14 µg

Quality Control in professional use
The performance of the test strips should be confirmed by use of positive and negative control solutions. Positive and negative controls should be analyzed once a day, when a new bottle of strips is opened, whenever a new lot of strips is started, and every 30 days to check storage conditions. Each laboratory should establish its own goals for adequate standards of performance, and should question handling and testing procedures if these standards are not met.

Directions
In any case, in order to establish a final diagnosis and prescribe an appropriate therapy, the results obtained with test strips should be verified with other medical results.
The effect of medicaments or their metabolic products on the test is not known in all cases. In case of doubt it is recommended not to take the medicaments and then repeat the test.
Only use well washed and clean vessels for urine collection. The presence of usual urine preservatives will not affect the test results.
Remove only as many test strips as are required and reseal the container immediately after use. Do not touch the test pads. Avoid exposing the strips to sunlight and moisture. Store the container below + 30 °C in a dry place. The test strips are stable, when stored properly up to the date of expiry indicated.
The glass contains a non-poisonous and harmless desiccant. In case this desiccant is swallowed accidentally, then drink plenty of water.
Explanations of symbols can be found in the package insert.
Disposals: Please dispose all used test strips in accordance with your local laws and regulations.
Package units: Tubes of 100 and 125 test strips
Date of change: 08/2014

Manufacturer / Manufacturer / Fabricante / Fabricant
REF Article number / Item number / Número de referencia / Référence produit
Consult instructions for use / Please read the instructions for use! / Observe les instructions de use / Respecter les instructions d'utilisation
Temperature storage range / Permitted storage temperature range / Limites de temperatura / Limites de température
Do not reuse / Do not reuse / Ne pas réutiliser / Ne pas réutiliser
CE

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Medi-Test URYXXON® Stick 10

es | In-vitro-Diagnostikum

Tiras reactivas para la determinación rápida de sangre, urobilinógeno, bilirrubina, proteínas, nitritos, cetonas, glucosa, valor pH, densidad y leucocitos en orina, para la evaluación reflecto-fotométrica con URYXXON® 300, 500 y URYXXON® Relax

Uso

Prueba de selección (screening) para la detección de diabetes, anormalidades metabólicas, enfermedades del hígado, obstrucciones biliares y hepáticas, enfermedades hemolíticas y enfermedades en la región renal y vías urinarias.

Utilizar solo bajo control médico.

Instrucciones de manejo

Emplear orina fresca no centrifugada y mezclar bien la prueba de orina. Sumerja la tira reactiva durante aproximadamente 1 segundo en orina. Separe, apoyando en el borde del contenedor para eliminar el exceso de orina. Colocar las tiras de comprobación en el aparato según las instrucciones de servicio. Los campos de comprobación se evalúan de un modo reflecto-fotométrico y los resultados se muestran en el formulario de diagnóstico. Los valores se corresponden a ciertas zonas de concentración de modo semejante a la comparación visual. En el caso de concentración visual se comparan los colores reactivos tras un tiempo de 30 o 60 segundos (campo de prueba de leucocitos después de 60–120 segundos) con la escala cromática. Los cambios de color que tienen lugar pasados 2 minutos no tienen significado. La orina no debe tener más de 2 horas, cuando se analice.

En la evaluación debe cuidarse que en todos los casos no coincidarán perfectamente por necesidad los resultados del URYXXON® 300, 500 y URYXXON® Relax con los resultados procedentes de la evaluación visual, ya que el ojo humano evalúa los colores de un modo diferente al aparato de evaluación URYXXON® 300, 500 y URYXXON® Relax. Las tiras de comprobación de la orina previstas para la evaluación **no pueden** evaluarse con el URYXXON® 300, 500 y URYXXON® Relax.

Principio

Sangre: La detección se basa en la actividad pseudoperoxidativa de la hemoglobina y mioglobina, que catalizan la oxidación de un indicador por un hidroperoxido orgánico produciendo un color verde.

Urobilinógeno: El papel de análisis contiene una sal de diazonio estable que produce un compuesto azo rojizo con urobilinógeno.

Bilirrubina: Se obtiene un compuesto azo roja en presencia de un ácido por el acoplo de la bilirrubina a una sal de diazonio.

Proteínas: La prueba se basa en el principio de los indicadores de "error proteico". La zona de reacción está tamponada a un pH ácido y cambia de color del amarillo al azul grisáceo en presencia de albúmina. Se indican otras proteínas con menor sensibilidad.

Nitritos: Los microorganismos causantes de reducir el nitrato a nitrito quedan indirectamente indicados por esta prueba. El reactivo del principio de Griess es la base de la prueba. El papel reactivo contiene una amina y un componente acoplante. Se obtiene un azocompuesto coloreado en rojo por la diazotización y acople subsiguiente.

Cetonas: La prueba se basa en el principio de Legal. El ácido acetoacético y la acetona forman con nitroprusiato sódico en medio alcalino un complejo coloreado violeta.

Glucosa: La detección se basa en la reacción enzimográfica glucosa-oxidasa-peroxidasa. A excepción de la glucosa en orina otra comúnmente conocida de la orina, da reacción positiva.

pH: El papel reactivo contiene indicadores que claramente cambian de color entre pH 5 y pH 9 (del naranja al verde brillante).

Densidad: El test determina la concentración de iones en la orina y logra buenas correlaciones con el método refractométrico. El color vira de azul verdoso, por verde, a amarillo, en la medida que aumenta la concentración de iones.

Leucocitos: La prueba se basa en la actividad esterasa de los granulocitos. Dicha enzima divide un éster del ácido carbónico. El componente alcohólico que se libera reacciona con una sal de diazonio produciendo una coloración violeta.

Evaluación – Fuentes de error

Zona de compensación: El campo de compensación se encuentra libre de agentes químicos y se emplea exclusivamente para la evaluación reflectométrica de los campos de pruebas.

Sangre: La mínima sensibilidad de la tira es de 5 eritrocitos por µL de orina, correspondiendo aproximadamente a 0,015 mg de hemoglobina o mioglobina/dL de orina. De hecho, los eritrocitos vienen indicados por unas pautas de decoloración del campo de análisis. Las pautas de colores corresponden a las siguientes valores:

0 (negativo), ca. 5–10, ca. 50, ca. 250 Ery/µL o bien una concentración de hemoglobina de ca. 10, ca. 50, ca. 250 Ery/µL respectivamente.

Las concentraciones normales de ácido ascórbico (< 40 mg/dL) no afectan a los resultados de las pruebas. Pueden producirse también reacciones falsamente positivas por residuos porosos contenidos en agentes limpiadores.

Urobilinógeno: Dependiendo del color de la orina, la adición de 0,5 a 1 mg/dL de urobilinógeno, 1 mg/dL se considerado como una excreción normal. Valores más altos son patológicos. Una ausencia total de urobilinógeno en orina, que es asimismo patológica, no puede demostrarse con las tiras. El campo de colores corresponde a las concentraciones de urobilinógeno siguientes:

norm. (normal), 2, 4, 8, 12 mg/dL o
norm. (normal), 35, 70, 140, 200 µmol/L.

La prueba será inhibida por una alta concentración de formaldehído. La exposición de la orina a la luz durante un largo tiempo puede dar valores bajos o falsos negativos. Resultados muy altos o falsos positivos pueden ser causados por la presencia de fases diagnósticas o terapéuticas en la orina. Grandes cantidades de fármacos producen una coloración amarilla.

Bilirrubina: La sensibilidad mínima de las tiras reactivas es de 0,5 a 0,75 mg de bilirrubina/dL de orina. La gama de colores corresponde a los valores siguientes:

0 (negativo), 1(+), 2(++), 4(+++) mg/dL o
0 (negativo), 17(+), 35(++), 70(+++) µmol/L.

Algunos componentes de la orina pueden producir una coloración amarillo de la tira reactiva. El ácido ascórbico y los nitritos en concentraciones elevadas inhiben la prueba. La exposición de la orina a la luz durante un largo período de tiempo puede dar resultados bajos o falsos negativos. Resultados demasiado altos o falsos positivos pueden ser causados por la presencia de fases diagnósticas o terapéuticas en la orina.

Proteínas: La mínima sensibilidad de la tira reactiva es 10 mg de proteínas/L de orina. Los valores corresponden a las concentraciones de albúmina siguientes:

negativo, 30, 100, 500 mg/dL o
negativo, 0,3, 1,0, 5,0 g/L.

Resultados falsamente positivos son posibles en muestras de orina alcalinas (pH > 8), después de infusiones con polimetilmidona (sustituto de la sangre), después de repetidas administraciones conteniendo quinina y también por residuos desinfectantes en los contenedores de orina. La coloración de las proteínas puede enmascararse por la presencia de tintas médicas (p. ej. azul de metileno) o pigmentos de restos de comidas.

Nitritos: La prueba detecta concentraciones desde 0,05 mg de nitrato/dL de orina. Toda pista roja indica una infección bacteriana de las vías urinarias. La densidad del color depende tan sólo de la concentración de nitrito, pero no proporciona información acerca de la magnitud de la infección. Un resultado negativo no excluye una infección de las vías urinarias, si existen bacterias que no producen nitritos. Pueden producirse resultados falsamente negativos por alta dosis de ácido ascórbico, por terapias con antibióticos y por muy bajas concentraciones de nitratos en la orina como resultados de dietas con bajo contenido en nitratos o fuerte diuresis (diuresis). Resultados falsamente positivos pueden ser motivados por la presencia de tintas diagnósticas o terapéuticas en la orina.

Cetonas: El ácido acetoacético reacciona con mayor sensibilidad que la acetona. Se detectan valores de ácido acetoacético de 5 mg/dL ó 500 mg/dL de acetona. El campo de colores corresponde a los valores de ácido acetoacético siguientes:

0 (negativo), 25(+), 100(++), 300(+++) mg/dL o
0 (negativo), 2,5(+), 10(++), 30 (+++) mmol/L.

Altas concentraciones de fenilcetonas interfieren la prueba y producen colores vacilantes. El ácido hidroxibutírico no se detecta. Los compuestos thiólicos interfieren produciendo una coloración roja.

Glucosa: Las concentraciones patológicas de glucosa vienen indicadas por un cambio de color que va desde el verde hasta el verde azulado. Las pruebas que den color amarillo o verdoso deben considerarse como normales o negativas. El campo de variación del color corresponde a los siguientes rangos de concentración de glucosa:

neg. (amarillo), neg. o normal (verdoso), 50, 150, 500, o 1000 mg/dL
neg. (amarillo), neg. o normal (verdoso), 2,8, 8,3, 27,6, o 55,5 mmol/L.

El colorido por ácido ascórbico se puede eliminar ampliamente. Además se produce un efecto inhibidor por el ácido gentísico. Pueden producirse también reacciones falsamente positivas por un residuo de peróxido contenido en agentes limpiadores.

pH: El valor de pH de la orina fresca de la mayor parte de la población varía entre pH 5 y pH 8. La escala de colores da una clara distinción del valor de pH entre pH 5 y pH 9.

Densidad: El test permite la determinación de la densidad en la orina, en el rango de 1.000 a 1.030. El valor normal de adultos que obtuvieren una dieta y un consumo de líquido regular, se sitúa entre 1.015 y 1.025. Los valores de densidad que se obtengan a través de la tira reactiva pueden verse ligeramente bajos a los casos métodos, p.ej. no se evidencian valores elevados que resulten de concentraciones de glucosa superiores a 1000 mg/dL (≈ 56 mmol/L). Así también, cuando haya elevada secreción de proteínas. En cambio, orina alcalina altamente temperada es causa frecuente de valores falsamente bajos.

Leucocitos: La prueba detecta valores a partir de aprox. 10 leucocitos/µL de orina. Las coloraciones que no son claramente en el campo de comparación negativo y mostrando débiles coloraciones violeta después de 120 segundos deben evaluarse como positivas. Los campos de comparación normales corresponden a las siguientes concentraciones de leucocitos:

negativo (normal), 25, 75, 500 leucocitos/µL.

Otras sustancias que reaccionan con el enzimo tales como altamente por encima de 500 mg/dL y una concentración de glucosa superior a 2 g/dL así como en caso de tomar preparados con cefalexina o gentamicina. Las bacterias, filamentos y eritrocitos no reaccionan con dicha prueba. El formaldehído como medio de conservación puede originar una reacción positiva falsa. Usado como conservante, el ácido bórico aminora la velocidad de la reacción. Excesivos de bilirubina, fenilcetonina o otros componentes fuertemente coloreados pueden enmascarar el color de la reacción. En las pruebas realizadas a mujeres, el flujo vaginal puede producir una reacción positiva falsa.

Sustancias reaccionantes
(Cantidad o actividad/cm² cuajados de la impregnación)

Sangre:		Nitrito:		pH:	
Tetrametilbenzidina:	31 µg	Ácido sulfanílico:	11 µg	Rojo de metilo:	3 µg
Hidroperóxido orgánico:	3 mg	Derivado de quinolina:	17 µg	Azul de bromotimol:	10 µg
Urobilinógeno:		**Cetonas:**		**Densidad:**	
Sal de diazonio:	75 µg	Nitroprusiato de sodio:	450 µg	Azul de bromotimol:	60 µg
Bilirrubina:		**Glucosa:**		Copolímero:	1946 µg
Sal de diazonio:	29 µg	Glucosa-oxidasa:	7 U	**Leucocitos:**	
Proteínas:		Peroxidasa:	1 U	Éster del ácido carbónico:	46 µg
Azul de tetrabromofenol:	10 µg	o-Tolidina:	44 µg	Sal de diazonio:	11 µg

Control de calidad para el empleo por personal cualificado

Para verificar el buen funcionamiento de las tiras reactivas se recomienda el uso de soluciones de control positivas y negativas. Los controles negativos y positivos deberán realizarse una vez al día, cada vez que se abra un nuevo envase, cuando se use un lote nuevo de tiras, así como cada 30 días para comprobar que las condiciones de almacenamiento del producto son adecuadas. Cada laboratorio debe establecer valores de referencia individuales según estándares de rendimiento adecuados para sale, y verificar los métodos de ensayo a estos estándares no con cumplidos.

Directrices

En todo caso, a fin de establecer un diagnóstico definitivo y prescribir la terapia adecuada, los resultados obtenidos por medio de tiras reactivas deben verificarse con otras técnicas médico-diagnósticas.

El efecto de los medicamentos y sus productos metabólicos sobre la prueba no se conocido en todos los casos. En caso de duda se recomienda no tomar los medicamentos y luego repetir la prueba.

Utilizar solamente contenedores limpios y limpios para recoger la orina. La presencia de contaminadores líquidos de orina no afectará los resultados

Sacar tan sólo las tiras reactivas que se precisen y tapar el contenedor inmediatamente después. No tocar el papel de prueba. Evitar exponer las tiras a la luz solar y a la humedad. Conservar el contenedor por debajo de 30 °C en un sitio seco. Las tiras reactivas son estables, cuando se conservan cuidadosamente hasta la fecha de caducidad indicada.

El agente desecante contenido en el tapón no es tóxico ni peligroso. En caso de ingestión accidental, beber agua en abundancia.

La explicación de los símbolos se encuentra al final de las instrucciones

Desechar las tiras usadas de acuerdo con la legislación en vigor.

Presentación: Tubo con 100 y 125 tiras

Fecha de Modificación: 08/2014

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fr | In-vitro-Diagnostikum

Medi-Test URYXXON® Stick 10

Bandelettes pour la détermination rapide du sang, de l'urobilinogène, de la bilirubine, des protéines, de nitrite, des corps cétoniques, du glucose, du pH, de la densité et des leucocytes dans l'urine, détermination avec le réflectomètre-photomètre URYXXON® 300, 500 et URYXXON® Relax

Usage

Test servant au diagnostic du diabète, d'anomalies du métabolisme, de maladies du foie, d'obstruction biliaire et hépatique, de maladies du sang ainsi que de maladies au niveau des reins et des voies urinaires.

Utilisation réservée au personnel compétent.

Mode d'emploi

Utiliser de l'urine fraîche bien mélangée mais non centrifugée. Immerger la bandelette brièvement (1 seconde) dans l'urine. Égoutter bandelette en passant le bouchon contre le rebord du récipient. Passer la languette test dans l'appareil et suivre les recommandations du mode d'emploi. Le réflectomètre lit les champs du test et imprime le résultat des mesures. Les valeurs sont comparables aux différents champs de mesure du comparateur couleur visuel selon la concentration. En détermination visuelle, attendre un temps de réaction de 30–60 secondes (champ de test de leucocytes après 60–120 secondes) sur la languette avant de la comparer avec le colorimètre sur l'emballage. Après plus de 2 minutes, les variations de couleur n'ont aucune signification diagnostique. Ne pas utiliser pour l'analyse des urines recueillies depuis plus 2 heures.

Il est à noter que toutes les mesures données par le réflectomètre URYXXON® 300, 500 et URYXXON® Relax ne correspondront pas obligatoirement avec la lecture visuelle des couleurs sur le champ du comparateur de l'emballage des languettes. En effet, l'œil peuvre intégrer différemment la mesure par colorimétrie. Les bandelettes urinaires Medi-Test destinées uniquement à la détermination visuelle, **ne pourront pas** être utilisées avec URYXXON® 300, 500 et URYXXON® Relax.

Principe

Sang : La mise en évidence repose sur l'action catalytique de l'hémoglobine ou de la myoglobine entraînant l'oxydation d'un indicateur vers une coloration bleu-verte par l'intermédiaire de l'hydroperoxyde organique.

Urobilinogène : La zone réactive contient un sel de diazonium stable qui forme avec l'urobilinogène un composé azoïque rougeâtre avec l'urobilinogène.

Bilirubine : En milieu acide, la copulation de la bilirubine avec un sel de diazonium provoque un composé azoïque rouge.

Protéines : Le test est basé sur le principe d'erreur protéique des indicateurs de pH. La zone réactive, indicateur colore tamponne à pH acide, est jaune en présence des protéines. A ce même pH, en présence de protéines, elle prend une teinte verte. Ce test est particulièrement sensible à l'albumine (limite de détection: 10 mg d'albumine/dL d'urine).

Nitrite : Indirectement, ce test met en évidence des micro-organismes qui peuvent réduire les nitrates en nitrites. La base de ce test est le principe de la réaction Griess. Le papier indicateur contient un amine et un facteur de copulation. Une diazotisation suivie d'une copulation entraîne un composé azoïque de couleur rouge.

Corps cétoniques : Le test repose sur le principe de la réaction de Legal. La réaction de l'acide acéto-acétique et de l'acétone avec le sodium nitroprusside en milieu alcalin forme un complexe de couleur violette.

Glucose : Il est mis en évidence par la méthode spécifique glucose-oxydase-peroxydase. Le test n'est pas influencé par la présence de corps cétoniques.

pH : La zone réactive contient 2 indicateurs colorés qui changent de couleur pour des valeurs de pH comprises entre 5 et 9 (l'orange à vert).

Densité : Ce test évalue la concentration d'ions présents dans l'urine. La corrélation est bonne entre les résultats obtenus avec ce test et la réfractométrie. En fonction de l'augmentation de la concentration d'ions, la coloration du test vire bleu-vert au vert, puis au jaune.

Leucocytes : Le test repose sur l'activité au niveau estérases des granulocytes. Cet enzyme clive un ester d'acide carboxylique. Les composants d'alcool ainsi dégagés réagissent avec un sel de diazonium par rapport à un colorant violet.

Evaluations et sources d'erreurs

Zone de compensation : Le champ de compensation est exempt de produits chimiques et il sert exclusivement à la mesure des champs du test par le réflectomètre.

Sang : La limite de détection de la bandelette est de 5 erythrocytes/µL d'urine correspondant à approximativement 0,015 mg d'hémoglobine ou de myoglobine/dL d'urine. Des colorations en forme de petits points dans la zone réactive, indiquent la présence d'érythrocytes intacts. Correspondances des zones de coloration :

0 (négatif), ca. 5–10, ca. 50, ca. 250 éry/µL respectivement
une concentration d'hémoglobine de ca. 10, ca. 50, ca. 250 éry/µL.

Des concentrations normales d'acide ascorbique (< 40 mg/dL) n'ont pas d'influence sur le résultat du test. Des résultats faussement positifs peuvent être dus à des restes de détergents contenant des résidus peroxydes ou autres.

Urobilinogène : Selon la coloration propre de l'urine, l'on peut mettre en évidence 0,5 à 1 mg d'urobilinogène/dL d'urine. Le taux d'excrétion normal est d'environ 1 mg/dL. Des valeurs supérieures sont pathologiques. Une absence complète et pathologique d'urobilinogène dans l'urine, ne peut pas être détectée par les bandelettes. Les zones de coloration correspondent aux concentrations d'urobilinogène suivantes :

norm. (normal), 2, 4, 8, 12 mg/dL ou
norm. (normal), 35, 70, 140, 200 µmol/L.

Le test est inhibé par des concentrations élevées de formaldéhyde. L'exposition prolongée au soleil de l'urine, peut donner, suite à une oxydation, des résultats trop faibles ou faussement négatifs. Des résultats trop élevés ou faussement positifs peuvent être dus à des colorants ou des médicaments présents dans l'urine. Des quantités importantes de bilirubine conduisent à une coloration jaune de la zone réactive.

Bilirubine : La sensibilité du test est limitée à 0,5–0,75 mg de bilirubine/dL d'urine. Les zones de coloration sont en fonction de la concentration en bilirubine selon les valeurs suivantes :

0 (négatif), 1(+), 2(++), 4(+++) mg/dL ou
0 (négatif), 17(+), 35(++), 70(+++) µmol/L.

Certains composants de l'urine peuvent provoquer une coloration jaune. Une très grande concentration d'acide ascorbique ou de nitrite peut conduire à des résultats trop faibles. L'exposition de l'urine à la lumière solaire peut entraîner – par suite d'une orientation – des résultats trop faibles ou faussement négatifs. Les médicaments ou colorants qui colorent l'urine en rouge, peuvent entraîner des résultats faussement positifs.

Protéines : La sensibilité détectable de ce test est de 10 mg protéine/dL d'urine. Les zones de coloration sont en fonction de la concentration en albumine selon les valeurs suivantes :

négatif, 30, 100, 500 mg/dL ou
négatif, 0,3, 1,0, 5,0 g/L.

Des résultats faussement positifs sont possibles dans des urines à valeur pH élevée (pH > 9) à la suite de perfusions de polyvinylpyrrolidone (succédanée du plasma sanguin) lors de traitement à la quinine ou en cas de présence de restes de substances antiseptiques à groupement ammonium quaternaire dans le récipient de recueil de l'urine. Des colorants ou provenance de médicaments (bleu de méthylène) ou de colorant des bactéries rouges peuvent influencer la coloration.

Nitrite : Par ce test, des valeurs de 0,05 mg de nitrite/dL d'urine sont détectables. Une coloration rose même faible indique l'existence d'une bactériurie significative. L'intensité de la coloration est en fonction de la concentration en nitrite mais ne permet cependant pas un diagnostic quant au degré de l'infection. Un résultat négatif n'exclut pas l'existence d'une bactériurie.

L'absorption de grandes quantités d'acide ascorbique, d'antibiotiques ou le cas de faible concentration de nitrate dans l'urine – par suite d'une alimentation pauvre en nitrates – ou la diurèse peuvent conduire à un résultat faussement négatif. Certains germes n'ont pas la possibilité de réduire le nitrate en nitrite. Une coloration faussement positive peut être due à des colorants dans l'urine.

Corps cétoniques : La sensibilité de l'acide acéto-acétique est plus prononcée que celle de l'acétone. Des valeurs de 5 mg d'acide acétique/dL d'urine ou 50 mg d'acétone/dL d'urine sont détectées. Les zones de coloration sont en fonction de la concentration d'acide acéto-acétique selon les valeurs suivantes :

0 (négatif), 25(+), 100(++), 300(+++) mg/dL ou
0 (négatif), 2,5(+), 10(++), 30(+++) mmol/L.

Les phénylcétonures en concentrations importantes donnent des interférences et conduisent à une coloration différente. Les composés phtaléines conduisent à des teintes rouges.

Glucose : Des concentrations pathologiques en glucose provoquent un virage du vert au verdâtre de la zone de coloration. Le test est être considéré comme négatif normal dans le cas d'un virage au jaune ou vert foncé de la zone de coloration. Les zones de coloration sont en fonction de la concentration du glucose suivant les valeurs ci-dessous :

négatif (jaune), négatif ou normal (jaune-vert), 50, 150, 500, ou 1000 mg/dL ou
négatif (jaune), négatif ou normal (jaune-vert), 2,8, 8,3, 27,8, ou 55,5 mmol/L.

L'influence de l'acide ascorbique (vitamine C) a été éliminé très largement. L'acide gentisique est cause d'effets inhibiteurs. Des résultats faussement positifs peuvent être dus à des restes de substances antiseptiques des oxydantes dans le récipient de recueil de l'urine.

pH : Dans l'urine fraîche de sujets sains, la valeur pH est de pH 5 à pH 8. L'échelle de coloration permet la lecture nette de la valeur pH entre pH 5 et pH 9.

Densité : Ce test permet de déterminer la densité de l'urine de 1.000 à 1.030. Pour un individu dont l'alimentation saine et liquide est normale, la normalité se situe entre 1.015 et 1.025.

Une légère différence dans les résultats peut être remarquée en comparaison avec d'autres méthodes. Par exemple la concentration de glucose à 1000 mg/dL (≈ 56 mmol/L) dans l'urine, fait augmenter sa densité, mais le test ne la relève pas. En présence de protéines, des densités trop faibles sont indicables par ce test. Dans des urines fortement alcalines, la valeur de la densité relevée ne sera souvent pas représentative.

Leucocytes : Le test saisit des valeurs à partir d'environ 10 leucocytes/µL d'urine. Les décolorations qui ne doivent plus être détectées au champ comparatif négatif ainsi que les décolorations légèrement violettes intervenant après 120 secondes doivent être interprétées sous forme positive. Les champs de comparaison de couleurs correspondent aux concentrations de leucocytes suivantes

négatif (normal), 25, 75, 500 leucocytes/µl.

Une réaction infaillible est attendue lors d'éliminations de protéines supérieures à 500 mg/dL et d'une concentration de glucose supérieure à 2 g/dL, ainsi que lors de l'absorption de préparations contenant de la céphalexine ou de la pentamycine. Les bactéries, les incomplexes et les érythrocytes ne réagissent pas à ce test. Le formaldéhyde (en tant qu'agent de conservation) peut entraîner une fausse réaction positive. Utilisé en tant que conservateur, l'acide borique diminue la sensibilité de la réaction. Les médicaments de laboratoire, phtaléinesulfone et autres combinaisons fortement colorées peuvent masquer la couleur de la réaction. Lors de prélèvements sur patients de sexe féminin, une fausse réaction positive peut s'avérer temporaire suite à des pertes vaginales.

Substances réactionnelles
(Quantité ou activité/cm² après l'imprégnation)

Sang :		Nitrite :		pH :	
Tétraméthylbenzidine :	31 µg	Acide sulfanilique :	11 µg	Rouge de méthyle :	3 µg
Cumènehydropéroxyde :	3 mg	Dérivé de quinoline :	17 µg	Bleu de bromothymol :	10 µg
Urobilinogène :		**Corps cétoniques :**		**Densité :**	
Sel de diazonium :	75 µg	Nitroprussiate de sodium :	450 µg	Bleu de bromothymol :	60 µg
Bilirubine :		**Glucose :**		Copolymère :	1946 µg
Sel de diazonium :	29 µg	Glucose oxydase :	7 U	**Leucocytes :**	
Protéines :		Peroxydase :	1 U	Ester d'acide carbonique :	46 µg
Bleu de tétrabromophénol :	10 µg	o-Tolidine :	44 µg	Sel de diazonium :	11 µg

Contrôle de qualité en cas d'utilisation par un personnel qualifié

Pour s'assurer du bon fonctionnement des bandelettes tests, il est recommandé d'utiliser des solutions de contrôle positives et négatives. Les contrôles positifs et négatifs devraient être réalisés une fois par jour, à l'ouverture d'un nouveau flacon, lors de l'utilisation d'un nouveau lot de bandelettes tests et tous les 30 jours pour vérifier les conditions de stockage. Chaque laboratoire devrait établir ses propres valeurs cibles pour des standards de performance adéquats et vérifier les méthodes de test à ces standards ne sont pas atteints.

Remarques

Nos bandelettes tests sont à associer à d'autres techniques médicales pour établir un diagnostic définitif, et prescrire une thérapie. L'influence des médicaments ou de leurs métabolites sur les tests n'est pas toujours connue. En cas de doute, il est conseillé de renéter les tests après arrêt de toute médication.

Recueillir l'urine dans des récipients bien lavés et rincés. Les conservateurs usuels de l'urine ne gênent pas les tests. Ne retirer que le nombre nécessaire de bandelettes de la boîte. Refermer aussitôt immédiatement. Ne pas toucher les zones de coloration. Ne pas exposer les bandelettes à la lumière solaire ni à l'humidité. Conserver la boîte dans un endroit frais et sec (ne pas dépasser 30 °C). Les bandelettes se conservent dans leur emballage d'origine jusqu'à la date de conservation indiquée sur la conditionnement.

La dessiccateur dans le bouchon n'est pas toxique. En cas d'ingestion accidentelle, boire abondamment de l'eau.

Légende : voir mode d'emploi joint

Destruction : détruire les bandelettes usagées selon les règles locales en vigueur

Contenu : boîte de 100 et 125 bandelettes tests

Date d'actualisation : 09/2014

CE

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EC Certificate

TÜVRheinland®

Directive 98/79/EC Annex IV, excluding Sections 4 and 6
Full Quality Assurance System
In Vitro Diagnostic Medical Devices

Registration No.: HL 60119814 0001

Report No.: 21265422 001

Manufacturer: Macherey-Nagel GmbH & Co. KG
Neumann-Neander-Str. 6-8
52355 Düren
Deutschland

Products: Products for self-testing

(see attachment for products and sites included)

Replaces Certificate, Registration No.: HL 60076687 0001

Expiry Date: 2022-05-28

The Notified Body hereby declares that the requirements of Annex IV, excluding section 4 and 6 of the directive 98/79/EC have been met for the listed products. The above named manufacturer has established and applies a quality assurance system, which is subject to periodic surveillance, defined by Annex IV, section 5 of the aforementioned directive. For placing on the market of List A devices covered by this certificate an EC design-examination certificate according to Annex IV, section 4 and a verification of manufactured products according to section 6 is required.

Effective Date: 2017-05-29

Date: 2017-05-29

Notified Body

Dipl.-Ing. Sven Hoffmann

TÜV Rheinland LGA Products GmbH - Tillystraße 2 - 90431 Nürnberg

TÜV Rheinland LGA Products GmbH is a Notified Body according to Directive 98/79/EC concerning in vitro diagnostic medical devices with the identification number 0197.

TÜVRheinland®

Doc. 1/1, Rev. 0

TÜV Rheinland
LGA Products GmbH
Tillystraße 2, 90431 Nürnberg

Attachment to
Certificate
Registration No.: HL 60119814 0001
Report No.: 21265422 001

Manufacturer: Macherey-Nagel GmbH & Co. KG
Neumann-Neander-Str. 6-8
52355 Düren
Deutschland

Products for self-testing:
- Single and multi-parameter disposable test strips for urine analysis
- indicator test strips and papers for measurement of pH in urine

Additional site for warehousing and logistics:

Bahnstr. 120
52355 Düren, Germany

Date: 2017-05-29

Notified Body

Dipl.-Ing. Sven Hoffmann

Medi-Test Control

en

Negative and positive control solutions for the Medi-Test urine test strips

Application:
Medi-Test Control are control solutions to verify the correct functioning of the Medi-Test Urine Test Strips and the URYXXON® Urine Analysis Systems. The measured values are compared with the target values shown in the table below.
Only qualified personnel must be allowed to carry out the application.

Reagents:
Each pack contains:
- 1 test tube with 15 mL reagent solution Medi-Test Control N
- 1 test tube with 15 mL reagent solution Medi-Test Control P

These are ready-to-use control solutions of purely chemical composition. The solution N simulates a urine sample with a value in the negative or normal range. The solution P will produce a positive color reaction with the Medi-Test Urine Test Strips for the parameters blood, urobilinogen, bilirubin, protein, nitrite, ketones, glucose and leukocytes so that values in the pathological range are indicated.

Safety precautions:
Reagent solutions Medi-Test Control contain Chloromethylisothiazolinone 0.0015–0.06% CAS 26172-55-4. WARNING H317 May cause an allergic skin reaction. P280 Wear protective gloves / eye protection. P302+352 IF ON SKIN: wash with plenty of water. P333+313 If skin irritation or rash occurs: get medical advice / attention. For further information, please ask for safety data sheet *(see www.mn-net.com/MSDS)*.

WARNING

Storage and Shelf Life:
When not in use, it is recommended that the reagent solutions Medi-Test Control N and P are stored in a dark place at 2–8 °C. Do not freeze the solutions under any circumstances.
If correctly stored, the reagent solutions may be used right up until the use-by-date printed on the packaging. After the first use, each reagent solution may be used for up to three months or for dipping the test strip up to 20 times, whichever occurs first.
Precipitations in the reagent solutions have no effect on the measurement result. If the solutions show other contamination, then they must no longer be used. The solutions may be disposed of with plenty of running water via the sewerage of the local wastewater treatment plant.

Instructions for use:
Take the test tubes with the solutions Medi-Test Control P and N out of the refrigerator, and allow them to warm up to room temperature, shake well so that the solutions can mix homogeneously. Avoid frothing.
Open the lids of the test tubes. Do not pour the solutions into another vessel. Perform measurements directly in the test tube. Where required, take out a test strip from a test strip container. After taking out a strip, immediately close the container again. Do not touch the test fields with your fingers.
Dip the test strip with all the test fields into the respective reagent solution for approx. 1 second.
After removing the test strip from the solution, briefly pat the edge along one side on an absorbent piece of material (e.g. kitchen roll) to dry it off. Insert the test strip into the device according to the instructions of the reflection photometers URYXXON® 300/500 and URYXXON® Relax. The test fields are analysed, and then the results are printed out.
When visually assessing the reaction colors of the test fields, compare the test fields with the color scale after about 30–60 seconds (leukocytes test field after 60–120 seconds). The most opportune reading time is after 30 seconds. Any color changes that occur after more than 2 minutes are of no significance.
Any visual assessment of the urine test strips must be made in daylight. Direct sunlight, however, must be avoided.
The urobilinogen test field may display a slightly more orange-red coloring in comparison to the color scale.
When not in use, and after completing the control procedures, replace the lids on the test tubes of the reagent solutions, and store them at 2–8 °C.

Note:
Please also note the instructions for the URYXXON® reflection photometers.
Do not ingest the solutions! Avoid any contact with skin or eyes! Store reagent solutions in a safe place inaccessible to children!
Rev. 10/2018

Explanation of symbols

CE	Statement of Conformity (Product corresponds to the In-Vitro Diagnostic Medical Devices Directive 98/79/EC of the European Union)
	Please read instructions for use!
	Permitted storage temperature range
	Use by
LOT	Batch identification
REF	Item number
	Manufacturer
IVD	In vitro diagnostics product
CONTROL	Control
CONTROL –	Control, negative
CONTROL +	Control, positive

Expected values:
The ranges identified in the table were determined using several different batches of the Medi-Test Combi 10® SGL and the URYXXON® Stick 10, and were ascertained using various different URYXXON® devices. Each laboratory should use the results provided only as a reference and establish own parameters of precision.

Analytes	Medi-Test Combi 10® SGL visually		URYXXON® Stick 10 with URYXXON® 300 / URYXXON® 500 / URYXXON® Relax	
	Control N	Control P	Control N	Control P
Blood	Negative	10–250 Ery/µL	Negative	10–250 Ery/µL
Urobilinogen	Normal	2–12 mg/dL[1] (35–200 µmol/L)	Normal	2–12 mg/dL[1] (35–200 µmol/L)
Bilirubin	Negative	1–4 mg/dL (1+–3+)	Negative	1–6 mg/dL (1+–3+)
Protein	Negative	100–500 mg/dL	Negative	25–500 mg/dL
Nitrite	Negative	Positive	Negative	Positive
Ketones	Negative	25–300 mg/dL (1+–3+) (2.5–30 mmol/L)	Negative	5–300 mg/dL (1+–3+) (0.5–30 mmol/L)
Glucose	Negative–Normal	500–≥1000 mg/dL (27.8–55.5 mmol/L)	Negative–Normal	50–1000 mg/dL (2.8–60 mmol/L)
pH value	5–7	7–9	5–7	7–9
Specific gravity (density)	1.010–1.030	1.005–1.025	1.010–1.030	1.005–1.025
Leukocytes	Negative	25–500 Leu/µL	Negative	15–500 Leu/µL

[1] The urobilinogen test field displays an orange-red coloring compared to the color scale

REF **93038** CONTROL IVD

A056642 / 108x

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CE

ФЕДЕРАЛЬНАЯ СЛУЖБА ПО НАДЗОРУ В СФЕРЕ ЗДРАВООХРАНЕНИЯ
(РОСЗДРАВНАДЗОР)

РЕГИСТРАЦИОННОЕ УДОСТОВЕРЕНИЕ НА МЕДИЦИНСКОЕ ИЗДЕЛИЕ

от 11 января 2017 года № ФСР 2010/08997

На медицинское изделие
Набор контрольных растворов белков мочи "БМ-контроль"
по ТУ 9398-269-52208224-2010

Настоящее регистрационное удостоверение выдано
Общество с ограниченной ответственностью "Медлакор С.-П."
(ООО "Медлакор С.-П."), Россия,
194100, Санкт Петербург, ул. А. Матросова, д. 4, корп. 2, Лит. П, офис 212

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Место производства медицинского изделия
ООО "Медлакор С.-П.", Россия, 194100, Санкт-Петербург, ул. А. Матросова, д. 4, корп. 2, Лит. П

Номер регистрационного досье **№ РД-14955/64156 от 20.12.2016**

Вид медицинского изделия **206630**

Класс потенциального риска применения медицинского изделия 1

Код Общероссийского классификатора продукции для медицинского изделия **93 9816**

Настоящее регистрационное удостоверение имеет приложение на 1 листе

приказом Росздравнадзора от 11 января 2017 года № 80
допущено к обращению на территории Российской Федерации.

Заместитель руководителя Федеральной службы
по надзору в сфере здравоохранения **Д.Ю. Павлюков**

0024833

ФЕДЕРАЛЬНАЯ СЛУЖБА ПО НАДЗОРУ В СФЕРЕ ЗДРАВООХРАНЕНИЯ
(РОСЗДРАВНАДЗОР)

ПРИЛОЖЕНИЕ К РЕГИСТРАЦИОННОМУ УДОСТОВЕРЕНИЮ НА МЕДИЦИНСКОЕ ИЗДЕЛИЕ

от 11 января 2017 года № ФСР 2010/08997

Лист 1

На медицинское изделие
Набор контрольных растворов белков мочи "БМ-контроль"
по ТУ 9398-269-52208224-2010:
- комплект 1 «БМ-контроль-ССК»;
- комплект 2 «БМ-контроль-ССК + глюкоза и pH»;
- комплект 3 «БМ-контроль-ССК с калибратором»;
- комплект 4 «БМ-контроль-ССК + глюкоза и pH с калибратором»;
- комплект 5 «БМ-контроль-ПГК»;
- комплект 6 «БМ-контроль-ПГК + глюкоза и pH» .

Заместитель руководителя Федеральной службы
по надзору в сфере здравоохранения **Д.Ю. Павлюков**

0026953

ИНСТРУКЦИЯ
ПО ПРИМЕНЕНИЮ НАБОРА КОНТРОЛЬНЫХ РАСТВОРОВ БЕЛКОВ МОЧИ + ГЛЮКОЗА и pH
«БМ-контроль-ССК + глюкоза и pH »

Назначение

Набор «БМ-контроль-ССК + глюкоза и pH» предназначен для контроля правильности и воспроизводимости результатов определения в моче :

белков – по их реакции с сульфосалициловой кислотой; с использованием диагностических тест- полосок;

глюкозы – ферментативным методом (глюкозооксидазным); качественным по реакции Бенедикта; с помощью диагностических тест – полосок;

pH – с помощью диагностических тест - полосок.

Набор предназначен только для диагностики in vitro.

Характеристика набора

«БМ-контроль-ССК + глюкоза и pH» представляет собой стабилизированные растворы, содержащие альбумин и глюкозу.

Контрольный материал готов к применению.

Состав набора

Набор «БМ-контроль-ССК + глюкоза и pH» содержит 8 флаконов по 10,0мл контрольных растворов альбумина и глюкозы, по 4 флакона двух уровней концентраций.

В паспорте набора указываются средние значения концентрации белка и глюкозы с контрольными пределами (X±2S).

Условия хранения и эксплуатации

Набор «БМ-контроль-ССК + глюкоза и pH» хранится при температуре (2 – 8) °С в темном месте.

Срок годности 9 месяцев.

В распечатанных и закрытых пробкой флаконах контрольный раствор белков мочи хранится при температуре (2 – 8) °С не более 14 дней.

. Меры предосторожности

При работе с набором необходимо соблюдать общие правила техники безопасности и производственной санитарии в клинико- диагностической лаборатории.

Аналитические характеристики

Диапазоны концентраций:

Белок	(0,2 - 0,4) г/л;
Глюкоза	(1,5 – 6,5) ммоль/л

Коэффициенты вариации:

Белок	не более 10%;
Глюкоза	неболее 5%.

Оборудование

Фотометр, кюветы с толщиной слоя 5 мм

Биохимический анализатор

Применение контрольных растворов

Контрольные растворы применяют в тех же условиях и с теми же реагентами, что и анализируемые образцы мочи.

Перед использованием флаконы с контрольными растворами выдерживают при комнатной температуре в течение 15 мин, затем перемешивают вручную путем переворачивания флакона 5-6 раз.

Определение концентраций компонентов проводят в соответствии с инструкциями к наборам реагентов или по методикам, утверждённым конкретным медицинским учреждением.

ИНСТРУКЦИЯ
ПО ПРИМЕНЕНИЮ НАБОРА КОНТРОЛЬНЫХ РАСТВОРОВ БЕЛКОВ МОЧИ
«БМ-контроль-ССК + глюкоза и pH с калибратором»

Назначение

Набор «БМ-контроль-ССК + глюкоза и pH с калибратором» предназначен для контроля правильности и воспроизводимости результатов определения в моче :

белков – по их реакции с сульфосалициловой кислотой; с использованием диагностических тест- полосок;

глюкозы – ферментативным методом (глюкозооксидазным); качественным по реакции Бенедикта; с помощью диагностических тест – полосок;

pH – с помощью диагностических тест - полосок.

Набор предназначен только для диагностики in vitro.

Характеристика набора

«БМ-контроль-ССК + глюкоза и pH с калибратором» представляет собой стабилизированные растворы, содержащие альбумин и глюкозу.

Контрольный материал готов к применению.

Состав набора

Набор «БМ-контроль-ССК + глюкоза и pH с калибратором» содержит 8 флаконов по 10,0 мл:

- 4 флакона калибратора с концентрациями белка 0,1; 0,2; 0,4 и 0,8 г/л;
- 4 флакона контрольных растворов альбумина и глюкозы в двух концентрациях, по 2 флакона каждой концентрации (уровень 1 и уровень 2).

В паспорте набора указываются средние значения концентрации белка и глюкозы с контрольными пределами (X±2S).

Условия хранения и эксплуатации

Набор «БМ-контроль-ССК + глюкоза и pH с калибратором» хранится при температуре (2 – 8) °С в темном месте.

Срок годности 9 месяцев.

В распечатанных и закрытых пробкой флаконах контрольный раствор хранится при температуре (2 – 8) °С не более 14 дней.

Меры предосторожности

При работе с набором необходимо соблюдать общие правила техники безопасности и производственной санитарии в клинико- диагностической лаборатории.

Аналитические характеристики

Диапазоны концентраций:

Белок	(0,1 - 0,8) г/л;
Глюкоза	(1,5 – 6,5) ммоль/л

Коэффициенты вариации:

белок	не более 10%;
глюкоза	не более 5%.

Оборудование

Фотометр, кюветы с толщиной слоя 5 мм

Биохимический анализатор

Применение контрольных растворов

Контрольные растворы применяют в тех же условиях и с теми же реагентами, что и анализируемые образцы мочи.

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Определение концентраций компонентов проводят в соответствии с инструкциями к наборам реагентов или по методикам, утверждённым конкретным медицинским учреждением.

Для построения калибровочного графика используют 4 раствора калибратора в разных концентрациях. Анализ каждого раствора повторяют в 5 параллельных пробах, для каждой концентрации рассчитывают среднее арифметическое оптической плотности. Используя полученные значения оптической плотности и паспортные значения концентрации калибратора, на логарифмической бумаге строят график. Началу координат соответствует концентрация белка 0,01 г/л.

Клиническая биохимия
Агат
105173, Москва, ул. Западная, д. 2, стр. 1, ООО «Агат-Мед».
Тел.: (495) 777-41-92.
Факс: (495) 741-25-19.
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БЕЛОК В МОЧЕ АГАТ

ИНСТРУКЦИЯ
по применению набора реактивов для определения белка в моче с сульфосалициловой кислотой

НАЗНАЧЕНИЕ

Диагностический набор предназначен для количественного определения содержания белка в моче по помутнению, образовавшемуся при добавлении сульфосалициловой кислоты.

Для клинико-диагностических и биохимических лабораторий.

Набор рассчитан на 660 определений при расходе 3,0 мл раствора сульфофосалициловой кислоты на один анализ.

ПРИНЦИП МЕТОДА

Интенсивность помутнения при коагуляции белка сульфосалициловой кислотой, измеренная по оптической плотности при 620 нм, пропорциональна его концентрации.

Калибровка осуществляется по раствору человеческого сывороточного альбумина.

СОСТАВ НАБОРА

1. **5-сульфосалициловая кислота**, дигидрат, 30 г – 2 упаковки;
2. **Калибровочный раствор альбумина 1000 мг/л**, 10 мл – 1 флакон.

ОБОРУДОВАНИЕ И РЕАГЕНТЫ

Спектрофотометр или фотоэлектроколориметр.

АНАЛИЗИРУЕМЫЕ ОБРАЗЦЫ

Моча профильтрованная.

ПОДГОТОВКА РЕАГЕНТОВ ДЛЯ АНАЛИЗА

Раствор сульфосалициловой кислоты. Содержимое одной упаковки (30 г) с сульфосалициловой кислотой количественно переносят в мерную колбу вместимостью 1000 мл, растворяют в дистиллированной воде и доводят объем до метки.

Раствор стабилен.

ПРОВЕДЕНИЕ АНАЛИЗА

В пробирки вносят реактивы по следующей схеме:

Отмерить, мл	Контрольная (холостая) проба	Опытная проба
Образец, профильтрованная моча	1,0	1,0
Раствор сульфосалициловой кислоты	-	3,0
Рствор натрия хлористого, 9 г/л	3,0	-

Содержимое пробирок тщательно перемешивают и выдерживают при температуре +18–22° С в течение 10 минут. Определяют оптическую плотность опытной пробы при длине волны 620 нм (590–650 нм, оранжевый или красный светофильтр) против холостой пробы в кювете с толщиной слоя 10 или 5 мм.

При стоянии образцов более 20 минут возможно уменьшение значений оптической плотности за счет оседания части преципитата. Непосредственно перед измерением пробирку с опытной пробой тщательно встряхнуть. Расчет проводят по калибровочному графику.

Построение калибровочного графика

Для построения калибровочного графика из калибровочного раствора альбумина и 9 г/л раствора натрия хлористого готовят следующие разведения:

№ пробирки	Калибровочный раствор альбумина, мл	9 г/л раствор NaCl, мл	Концентрация белка	
			мг/л	г/л
1	0,25	4,75	50	0,05
2	0,50	4,50	100	0,10
3	1,00	4,00	200	0,20
4	2,50	2,50	500	0,50
5	5,00	-	1000	1,00

Полученные разведения обрабатывают так же, как и образец.

Примечания: Линейная зависимость сохраняется до концентрации белка 1 г/л. При более высоких концентрациях пробу следует развести в 2–3 раза, результат умножить на разведение.

Результаты, получаемые данным методом чувствительны к изменениям температуры. Рекомендуется производить измерения при температуре +18–22° С.

Ложноположительные результаты могут быть получены при наличии в моче контрастных веществ, содержащих органический йод. Поэтому тест нельзя использовать у лиц, принимающих препараты йода. Ложноположительный тест может быть также обусловлен приемом сульфаниламидных препаратов, больших доз пенициллина и при высоких концентрациях в моче мочевой кислоты.

УСЛОВИЯ ХРАНЕНИЯ И ЭКСПЛУАТАЦИИ

Набор следует хранить в упаковке предприятия-изготовителя при температуре +2–8° С в течение всего срока годности.

Срок годности набора – 2 года.

Литература: Лабораторные методы исследования в клинике. Под редакцией проф. В.В. Меньшикова, М., 1987, с. 49.

По вопросам, касающимся приобретения наборов и их качества, просим обращаться по адресу: 105173, г. Москва, ул. Западная, д. 2, стр. 1, ООО «Агат–Мед». Телефон для справок: (495) 777-41-92.

Инструкция составлена: к.б.н. И.В. Смирновым – зав. лабораторией ГНЦ РАМН, В.В. Гладуном – главным технологом ООО «Агат-Мед».

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This Certificate of Conformity is issued to

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Address: 6, Glavnaya st., Moscow, 105173, Russia
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Date of issue: 26.01.2018 Period of validity: 26.01.2021

This certificate certifies that:

Medical devices. Quality management system. System requirements for regulatory purposes in relation to the works in accordance with Annex 1 to this certificate

(the attachment is an integral part of the certificate)

CORRESPONDS TO THE REQUIREMENTS OF GOST ISO 13485-2011 (EN ISO 13485:2003)

Manager of "Expert" authority **V.V. Koptsev** **O.V. Gundareva**

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