3207-0010

chemagic™ DNA CS200 kit

Instructions for use. Reagents for 960 extractions.

Manufacturer:
Wallac Oy
Mustionkatu 6, FI-20750 Turku, Finland
Phone: +358 2 2678 111

FOR *IN VITRO* DIAGNOSTIC USE

CE

revvity

SYMBOLS

	*In vitro* diagnostic medical device
	Batch code
	Packing number
	Catalog number
	Use by
	Temperature limitation
	Store in the dark
	Contains sufficient for <n> tests
	Consult instructions for use
	Manufacturer
	GHS02
	GHS08

GHS07

GHS05

This way up

Recyclable

Fragile, handle with care

Keep dry

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chemagic™ DNA CS200 kit

INTENDED PURPOSE

The chemagic™ DNA CS200 kit is intended for the extraction and purification of DNA from human whole blood or plasma for analysis by PCR based *in vitro* diagnostic test systems. The disorder and determination of analytes are dependent on the PCR based downstream assay. The product is intended to be used with an automated workflow by trained laboratory personnel.

SUMMARY AND PRINCIPLE

The chemagic DNA CS200 kit is based on a magnetic bead technology platform proprietary to Revvity chemagen Technologie GmbH. Cells or other source of DNA present in whole blood or plasma samples are lysed during the extraction process. The released nucleic acids bind to small magnetizable particles which are then magnetically separated from the sample material. During subsequent steps contaminants are removed and the purified nucleic acids are transferred into an elution buffer. The automated sample processing is performed using the chemagic 360-D instrument (2024-0010) with chemagic 360 96 Rod Head Set (CMG-370) or chemagic Prime™ Jr-D (2029-0010).

KIT CONTENTS

The kit contains reagents sufficient to perform 960 extractions.

The expiry date of the unopened kit is stated on the outer label. Do not use any component beyond the expiry date. Store at +2 to +25 °C.

Once opened, the kit components have a limited stability. The stability after opening is stated for each component separately in the reagent listing below. Note: Recap the bottles tightly immediately after use to prevent evaporation.

The bottles may discolor during storage. The discoloration of the bottles has no effect on the functionality of the assay.

The kit contains following items:

Component	Quantity
Magnetic Beads B	1 bottle, 150 mL
Lysis Buffer P	1 bottle, 480 mL
Binding Buffer P	2 bottles, 550 mL
Lysis Buffer B	1 bottle, 480 mL
Binding Buffer B	2 bottles, 550 mL
Wash Buffer BB	1 bottle, 700 mL
Wash Buffer BA	1 bottle, 700 mL
Wash Buffer E	1 bottle, 700 mL
Wash Buffer H	1 bottle, 700 mL
Elution Buffer	1 bottle, 240 mL
Proteinase K	5 bottles (lyophilized)
Poly(A)RNA	10 tubes (dried)
Poly(A)RNA buffer	10 tubes, 0.5 mL
Disposable Tips (96 ea)	10 x 96 ea

Reagents

Component	Stability and storage
Magnetic Beads B	+2 to +25 °C until expiry date stated on the bottle label. Once opened, stable for 60 days at +2 to +25 °C.
Suspension of particles containing nanoparticular iron oxide encapsulated in a matrix of polyvinyl alcohol. Magnetic Beads (28.0 ± 0.5 mg/mL) bind the DNA during the extraction process.	
Lysis Buffer P  DANGER	+2 to +25 °C until expiry date stated on the bottle label. Store in the dark. Once opened, stable for 60 days at +2 to +25 °C.
Ready-for-use aqueous buffer solution containing guanidinium thiocyanate (50–75%). Lysis Buffer is used to lyse the cells or other DNA source present in the sample in order to get the DNA in solution.	

Lysis Buffer P contains guanidinium thiocyanate:
H302+H312+H332 Harmful if swallowed, in contact with skin or if inhaled.
H314 Causes severe skin burns and eye damage.
H412 Harmful to aquatic life with long lasting effects.
P260 Do not breathe dusts or mists.
P303+P361+P353 IF ON SKIN (or hair): Take off immediately all contaminated clothing. Rinse skin with water [or shower].
P305+P351+P338 IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.
P310 Immediately call a POISON CENTER/doctor.
P362+P364 Take off contaminated clothing and wash it before reuse.
P405 Store locked up.
P501 Dispose of contents/container in accordance with local regulations.
EUH032 Contact with acids liberates very toxic gas.

Binding Buffer P

+2 to +25 °C until expiry date stated on the bottle label. Once opened, stable for 60 days at +2 to +25 °C.

DANGER

Ready-for-use Tris-HCl-buffered (pH 5.0–5.9) solution with sodium perchlorate (25–50%), acetic acid (1–2.5%) and ethanol (25–50%). Binding Buffer is used to create the appropriate conditions to get the DNA bound to the Magnetic Beads.

Binding Buffer P contains sodium perchlorate and ethanol:
H225 Highly flammable liquid and vapor.
H302 Harmful if swallowed.
H319 Causes serious eye irritation.
H373 May cause damage to organs through prolonged or repeated exposure.
P210 Keep away from heat, hot surfaces, sparks, open flames and other ignition sources. No smoking.
P241 Use explosion-proof [electrical/ventilating/lighting] equipment.
P303+P361+P353 IF ON SKIN (or hair): Take off immediately all contaminated clothing. Rinse skin with water [or shower].
P305+P351+P338 IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.
P403+P235 Store in a well-ventilated place. Keep cool.
P501 Dispose of contents/container in accordance with local regulations.

Lysis Buffer B

WARNING

+2 to +25 °C until expiry date stated on the bottle label. Avoid direct sunlight. Once opened, stable for 60 days at +2 to +25 °C.

Ready-for-use aqueous buffer solution (pH 6.9–7.4) containing guanidinium chloride (15–25 %) and detergent. Lysis Buffer is used to lyse the blood cells in order to get the DNA in solution.

Lysis Buffer B contains guanidinium chloride:
H315 Causes skin irritation.
H319 Causes serious eye irritation.
P264 Wash thoroughly after handling.
P280 Wear protective gloves / eye protection / face protection.
P305+P351+P338 IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.
P332+P313 If skin irritation occurs: Get medical advice/attention.
P362+P364 Take off contaminated clothing and wash it before reuse.
P337+P313 If eye irritation persists: Get medical advice/attention.

Binding Buffer B

DANGER

+2 to +25 °C until expiry date stated on the bottle label. Once opened, stable for 60 days at +2 to +25 °C.

Ready-for-use Tris-HCl-buffered (pH 5.0–5.9) solution with sodium perchlorate (15–25 %) and ethanol (25–50 %). Binding Buffer is used to create the appropriate conditions to get the DNA bound to the Magnetic Beads.

Binding Buffer B contains sodium perchlorate and ethanol:
H225 Highly flammable liquid and vapor.
H319 Causes serious eye irritation.
H373 May cause damage to organs through prolonged or repeated exposure.
P210 Keep away from heat, hot surfaces, sparks, open flames and other ignition sources. No smoking.
P241 Use explosion-proof [electrical/ventilating/lighting] equipment.
P303+P361+P353 IF ON SKIN (or hair): Take off immediately all contaminated clothing. Rinse skin with water [or shower].
P305+P351+P338 IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.
P403+P235 Store in a well-ventilated place. Keep cool.
P501 Dispose of contents/container in accordance with local regulations.

Wash Buffer BB

+2 to +25 °C until expiry date stated on the bottle label. Once opened, stable for 60 days at +2 to +25 °C.

DANGER

Ready-for-use Tris-HCl-buffered (pH 5.0–5.6) solution with sodium perchlorate (15–25%) and ethanol (25–50%). Used for removing non-DNA contaminants during washing step.

Wash Buffer BB contains sodium perchlorate and ethanol:
H225 Highly flammable liquid and vapor.
H319 Causes serious eye irritation.
H373 May cause damage to organs through prolonged or repeated exposure.
P210 Keep away from heat, hot surfaces, sparks, open flames and other ignition sources. No smoking.
P241 Use explosion-proof [electrical/ventilating/lighting] equipment.
P303+P361+P353 IF ON SKIN (or hair): Take off immediately all contaminated clothing. Rinse skin with water [or shower].
P305+P351+P338 IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.
P403+P235 Store in a well-ventilated place. Keep cool.
P501 Dispose of contents/container in accordance with local regulations.

Wash Buffer BA

+2 to +25 °C until expiry date stated on the bottle label. Once opened, stable for 60 days at +2 to +25 °C.

DANGER

Ready-for-use Tris-HCl-buffered (pH 5.0–5.6) solution with sodium perchlorate (15–25%) and ethanol (25–50%). Used for removing non-DNA contaminants during washing step.

Wash Buffer BA contains sodium perchlorate and ethanol:
H225 Highly flammable liquid and vapor.
H319 Causes serious eye irritation.
H373 May cause damage to organs through prolonged or repeated exposure.
P210 Keep away from heat, hot surfaces, sparks, open flames and other ignition sources. No smoking.
P241 Use explosion-proof [electrical/ventilating/lighting] equipment.
P303+P361+P353 IF ON SKIN (or hair): Take off immediately all contaminated clothing. Rinse skin with water [or shower].
P305+P351+P338 IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.
P403+P235 Store in a well-ventilated place. Keep cool.
P501 Dispose of contents/container in accordance with local regulations.

Wash Buffer E  DANGER	+2 to +25 °C until expiry date stated on the bottle label. Once opened, stable for 60 days at +2 to +25 °C.

Ready-for-use solution contains ethanol 50–75%. Used for removing last traces of non-DNA contaminants during washing step.

Wash Buffer E contains ethanol:
H225 Highly flammable liquid and vapor.
P210 Keep away from heat, hot surfaces, sparks, open flames and other ignition sources. No smoking.
P241 Use explosion-proof [electrical/ventilating/lighting] equipment.
P280 Wear protective gloves/protective clothing/eye protection/face protection.
P303+P361+P353 IF ON SKIN (or hair): Take off immediately all contaminated clothing. Rinse skin with water [or shower].
P403+P235 Store in a well-ventilated place. Keep cool.
P501 Dispose of contents/container in accordance with local regulations.

Wash Buffer H	+2 to +25 °C until expiry date stated on the bottle label. Once opened, stable for 60 days at +2 to +25 °C.

Ready-for-use ultra-filtered water solution. Used for removing possible residuals of ethanol.

Elution Buffer	+2 to +25 °C until expiry date stated on the bottle label. Once opened, stable for 60 days at +2 to +25 °C.

Ready-for-use 10 mM Tris-HCl-buffered (pH 7.8–8.4) solution.

Proteinase K  DANGER	+2 to +25 °C until expiry date stated on the bottle label. Once reconstituted, stable for 28 days at +2 to +8 °C.

The Proteinase K (Proteinase 50–90%) is reconstituted by adding 2.5 mL of molecular grade water. Proteinase K is added to enhance the efficiency of the lysis step.

Proteinase K contains Proteinase, Tritirachium album serine:
H315 Causes skin irritation.
H319 Causes serious eye irritation.
H334 May cause allergy or asthma symptoms or breathing difficulties if inhaled.
H335 May cause respiratory irritation.
P261 Avoid breathing dust/fume/gas/mist/vapors/spray.
P280 Wear protective gloves / eye protection / face protection.
P284 [In case of inadequate ventilation] wear respiratory protection.
P305+P351+P338 IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.
P405 Store locked up.
P501 Dispose of contents/container in accordance with local regulations.

Poly(A)RNA	+2 to +25 °C until expiry date stated on the tube label. Once reconstituted, stable for 30 days at +2 to +8 °C.

Poly(A)RNA is reconstituted by adding 440 µL of Poly(A)RNA Buffer. The Poly(A)RNA functions as a DNA carrier to enhance the efficiency of the extraction process.

Poly(A)RNA buffer	+2 to +25 °C until expiry date stated on the tube label.

DANGER

Ready-for-use aqueous buffer solution containing guanidinium thiocyanate (25–50%). Poly(A)RNA Buffer is used for reconstitution of Poly(A)RNA.

Poly(A)RNA buffer contains guanidinium thiocyanate:
H302+H332 Harmful if swallowed or if inhaled.
H314 Causes severe skin burns and eye damage.
H412 Harmful to aquatic life with long lasting effects.
P260 Do not breathe dusts or mists.
P303+P361+P353 IF ON SKIN (or hair): Take off immediately all contaminated clothing. Rinse skin with water [or shower].
P305+P351+P338 IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.
P310 Immediately call a POISON CENTER/doctor.
P405 Store locked up.
P501 Dispose of contents/container in accordance with local regulations.
EUH032 Contact with acids liberates very toxic gas.

Disposable Tips (96 ea)	+2 to +25 °C until expiry date stated on the label.

MATERIALS REQUIRED BUT NOT SUPPLIED WITH THE KIT

The chemagic DNA CS200 kit requires the following items, which are available from Wallac Oy or Revvity, Inc. and its distributors:

- chemagic 360-D (prod. no. 2024-0010) with chemagic 360 96 Rod Head Set (prod. no. CMG-370) or chemagic Prime Jr-D (prod. no. 2029-0010)
- consumables for chemagic DNA Extraction (low well plates, deep well plates, prod. no. 4148-0010)

Additional required items:

- pipettes and pipette tips with aerosol barriers
- molecular grade water

Additional optional items:

- chemagic Stand 96 (prod. no. CMG-301)

SPECIMEN COLLECTION AND HANDLING

Human plasma samples (200 µL) should be used, either fresh or stored typically for up to five days at +2 to +8 °C or stored frozen at -20 to -80 °C. Frozen samples must not be thawed more than once. The recommended sample stabilizers are EDTA or citrate. The use of heparin stabilized plasma samples can cause inhibition problems in PCR and is therefore not recommended.

Human whole blood samples (200 µL), fresh or stored typically for a maximum of one week at +2 to +8 °C should be used. The recommended blood stabilizers are EDTA or citrate. The use of heparin stabilized blood samples can cause inhibition problems in PCR and is therefore not recommended. The white blood cell count in the whole blood sample decreases during storage.

The white blood cell count in the whole blood sample decreases during storage. Storing the samples may cause a poor yield of the DNA extraction.

The extraction efficiency using other types of sample material has not been determined.

Specimen stability of plasma

The influence of storage time and temperature was studied[1] using several confirmed Cytomegalovirus (CMV) negative and positive plasma specimens collected to collection tubes containing either EDTA or citrate as an anticoagulant. Samples were stored at -20 °C. Extracted DNA was analysed with CE IVD registered downstream for diagnosis of CMV in singlicate and was categorized as CMV-positive and CMV-negative. No clinically significant deviation was observed on results compared to comparator extraction method.

[1] Study performed at Turku University Hospital, Turku, Finland.

Specimen stability of whole blood

The influence of storage time and temperature was studied[2] using several whole blood specimens collected from healthy donors to collection tubes containing either EDTA or citrate as an anticoagulant. Samples were stored at +2 to +8 °C up to 7 days. Extracted DNA samples were replicated in CE IVD-registered PCR assay and samples were tested by a commercially available downstream assay kit for detecting Fragile X syndrome using recommended reference samples by the National Institute for Biological Standards and Control (NIBSC). No clinically significant deviation was observed on results.

Influence of interfering substances

The effect of substances contained in human whole blood or plasma possibly interfering with the DNA extraction was tested[3] in both whole blood and plasma samples. The tested substances and their concentrations are presented in the table below. Based on the results, it was concluded that the tested substances do not interfere with DNA extraction.

Interfering substance	Concentration	Interference
Bilirubin conjugated	332 µg/mL	No
Bilirubin unconjugated	200 µg/mL	No
Triglycerides	30 mg/mL	No
Human Serum Albumin	30 mg/mL	No

WARNINGS AND PRECAUTIONS

The product is intended to be used by trained laboratory personnel.

To minimize irregularities in diagnostic results, the product is intended to be used with an internal control as well as positive and negative controls throughout the process of sample preparation, and sample amplification and detection according to the downstream assay used.

Handle all patient specimens as potentially infectious. Nevertheless all recommended precautions for the handling of blood derivatives should be observed. Please refer to the U.S. Department of Health and Human Services publication "Biosafety in Microbiological and Biomedical Laboratories" or any other local or national regulation.

Lysis Buffer P contains guanidinium thiocyanate and is harmful if swallowed, in contact with skin or if inhaled, causes severe skin burns and eye damage and is harmful to aquatic life with long lasting effects. **Binding Buffer P contains sodium perchlorate and ethanol** and is highly flammable liquid and vapor and is harmful if swallowed, causes serious eye irritation and may cause damage to organs through prolonged or repeated exposure. **Poly(A)RNA buffer contains guanidinium thiocyanate** and is harmful if swallowed or if inhaled, causes severe skin burns and eye damage and is harmful to aquatic life with long lasting effects. **Wash Buffer BB, Wash Buffer BA and Binding Buffer B contain sodium**

[2] Study performed at Wallac Oy, Turku, Finland.
[3] Study performed at Revvity chemagen Technologie GmbH, Baesweiler, Germany.

perchlorate and ethanol and are highly flammable liquids and vapors, cause serious eye irritation and may cause damage to organs through prolonged or repeated exposure. **Lysis Buffer B contains guanidinium chloride** and causes skin irritation and serious eye irritation. **Wash Buffer E contains ethanol** and is highly flammable liquid and vapor. **Proteinase K contains Proteinase, Tritirachium album serine** and causes skin irritation and serious eye irritation, may cause allergy or asthma symptoms or breathing difficulties if inhaled and may cause respiratory irritation. See specific precautions from the section “KIT CONTENTS”.

To avoid injuries when working with the kit components, always wear safety glasses, disposable gloves, and protective clothing. For detailed information, please refer to the corresponding safety data sheets (SDS).

Follow local regulations for handling of ethanolic solutions.

Disposal of all waste should be in accordance with local regulations.

For a patient/user/third party in the European Union and in countries with an identical regulatory regime (IVDR; EU 2017/746/EU); if, during the use of this device or as a result of its use, a serious incident has occurred, please report it to the manufacturer and to your national authority. The contact information for the manufacturer of this device to report a serious incident is stated on the cover page of these instructions for use.

PROCEDURE

Extraction protocol using the chemagic 360-D instrument

The duration of the automated extraction protocol is approximately 75 minutes.

The protocol is suitable for processing up to 96 samples in parallel (see protocol steps below). For detailed instructions on the use of the chemagic 360-D instrument, please refer to the chemagic 360-D User manual.

Samples and reagents must be brought to room temperature (+19 to +25 °C) before use.

NOTE: Recap the bottles tightly immediately after use or keep the bottles connected tightly to the chemagic 360-D instrument. Binding Buffer P, Binding Buffer B, Wash Buffer BB, Wash Buffer BA and Wash Buffer E contain ethanol. If ethanol evaporates, the optimal yield or detection sensitivity cannot be guaranteed.

Processing steps in detail

<u>Preparing steps</u>

1. Check all kit components for integrity. In case of damage, contact your supplier.

2. For plasma samples: Reconstitute the Proteinase K and Poly(A)RNA components
    a. Proteinase K: Add 2.5 mL molecular grade water to Proteinase K bottle and mix gently until dissolved.

b. Poly(A)RNA: Add 440 µL of Poly(A)RNA Buffer to the Poly(A)RNA tube and mix thoroughly until dissolved.

3. For plasma samples: If Lysis Buffer P contains precipitate (formed during transfer or storage), the solution should be heated to 50–60 °C and thoroughly mixed until the solution is clear. The clarity of the Lysis Buffer P should be visually confirmed always before use (visual inspection should be done followed by mixing and by opening the bottle cap).

4. Connect the reagent bottles to chemagic 360-D instrument as follows:
   Manifold 1: No bottle connected
   Manifold 2: No bottle connected
   Manifold 3: Wash Buffer BB
   Manifold 4: Wash Buffer BA
   Manifold 5: Wash Buffer E
   Manifold 6: Wash Buffer H

5. Fill and prime the chemagic 360-D tubing with reagents by choosing protocol **'prime manifolds H96 all 360 V150116'**. Press [Insert IDs] button, follow the instructions given in the software and start priming by pressing [OK] button. If functions enabling the ID data input are deactivated, start priming directly by pressing [Start] button. Priming needs to be done when reagent bottles are connected to the chemagic 360-D instrument for the first time or when the instrument's tubing is not already filled with the above mentioned reagents.

6. Make sure that the samples are homogenous at the time of pipetting the samples on a plate by mixing gently.

Protocol steps

Magnetic Beads B (low well plate on plate position 2 in chemagic 360-D instrument) is resuspended by mixing thoroughly and pipetted manually (150 µL / well) to each corresponding sample well in use.

Elution Buffer (deep well plate on plate position 8 in chemagic 360-D instrument) is pipetted manually (100 µL / well) to each corresponding sample well in use.

The sample preparation steps are done manually. The reagents used in sample preparation are dependent on the sample type (whole blood/plasma). Upon completion of sample preparation steps (refer to sample preparation step table), the sample plate is placed to the chemagic 360-D instrument and the automated DNA extraction process is initiated.

NOTE: The automated extraction run must be started immediately after adding the Binding Buffer P and/or Binding Buffer B to the lysed sample wells. Hesitation may result in low yield and purity.

Please refer to the table below for more detailed information on plate position and protocol step details.

Automated DNA Extraction run in chemagic 360-D instrument

Position on tracking system*	Material in position	Protocol step in detail
		Select the protocol **'check manifolds H96 all 360 V150116'** to flush the tubing prior to starting the automated extraction run. Press [Insert IDs] button, follow the instructions given in the software and start flushing by pressing [OK] button. If functions enabling the ID data input are deactivated, start flushing directly by pressing [OK] and [Start] buttons.
		When using the functions enabling the ID data input, select the protocol **'---chemagic CS200 IVD prefilling V141203.che'** and press [Insert IDs] button. Follow the instructions given in the software to fill in the required data. Load the plates to tracking system positions 1-8. After all plates are in place, press [OK] button.
1	Rack with Disposable Tips	Use Disposable Tips according to the positions of the samples. Note: Tips need to be present in rack in full rows.
2	Low well plate 150 µL Magnetic Beads B	Pipette thoroughly resuspended Magnetic Beads B to each sample well in use. Place the plate to rack position 2.
Prepare the samples according to the procedures described in separate tables. The samples should be prepared after all the other preparing steps are ready and the plates placed on tracking system positions 1-2 and 4-8.		
3	Sample plate (Deep well plate)	Place the plate with prepared samples to rack position 3 and check all plates for accurate positioning and fitting. Close the front door and **start the DNA extraction process immediately**.
4	Deep well plate	Place empty plate to rack position 4. Wash Buffer BB is dispensed to the plate automatically.
5	Deep well plate	Place empty plate to rack position 5. Wash Buffer BA is dispensed to the plate automatically.
6	Deep well plate	Place empty plate to rack position 6. Wash Buffer E is dispensed to the plate automatically.
7	Deep well plate	Place empty plate to rack position 7. Wash Buffer H is dispensed to the plate automatically.
8	Deep well plate 100 µL Elution Buffer	Place the prefilled Elution Buffer plate to rack position 8.
		When using the functions enabling the ID data input, the extraction run is started by pressing [Start] button at the end of the dialogue. If the functions enabling the ID data input are deactivated, load the plates to tracking system positions 1-8. After all plates are in place, select the protocol **'---chemagic CS200 IVD prefilling V141203.che'**, mark the Columns in use on the plate map in the dialogue and start the extraction run directly by pressing [Start] button.

* Numbers in tracking system refer to the positioning of the plate in chemagic 360-D instrument

Sample preparation, Whole Blood samples

Material	**Protocol step in detail**
Deep well plate 200 µL blood (sample) 450 µL Lysis Buffer B	 Dispense up to 96 wells of the Sample Plate with 200 µL whole blood. Add Lysis Buffer B to the wells containing sample and incubate the plate for 10 minutes.
1050 µL Binding Buffer B	Add the Binding Buffer B to each lysed whole blood sample well. Place the sample plate to rack position 3 and **start the run immediately**.

Sample preparation, Plasma samples

Material	**Protocol step in detail**
Deep well plate 4 µL reconstituted Poly(A)RNA 10 µL reconstituted Proteinase K 200 µL plasma (sample) 450 µL Lysis Buffer P	 Add the reconstituted Poly(A)RNA and Proteinase K to the sample wells. Dispense up to 96 wells of the Sample Plate with 200 µL plasma. Add Lysis Buffer P to the wells containing sample and incubate the samples in 50–60 °C for 10 minutes. The Proteinase K activity will decrease after incubation longer than 10 minutes in Lysis Buffer P. Ensure that all samples are mixed with Poly(A)RNA/Proteinase K/Lysis Buffer P during incubation.
1050 µL Binding Buffer P	Add the Binding Buffer P to each lysed plasma sample well. Place the sample plate to rack position 3 and **start the run immediately**.

After the isolation procedure has finished, collect the DNA eluates and use the [Turn Table] button to unload the tracking system. Each click on the [Turn Table] moves the tracking system (table) clockwise by one position. Never move the tracking system (table) manually. Note: Do not turn the x-axis by hand, because it may cause damage to the equipment. All movements have to be performed with the [Turn Table] function.

Extraction protocol using the chemagic Prime Jr-D instrument

The duration of the automated extraction protocol is approximately 3 hours 10 minutes.

The protocol is suitable for processing up to 48 samples per run and provides automated sample processing. For detailed instructions on the use of the chemagic Prime Jr-D instrument, please refer to the chemagic Prime Jr-D Instrument manual.

Samples and reagents must be brought to room temperature (+19 to +25 °C) before use.

PROCEDURAL NOTES

1. A thorough understanding of this package insert and the chemagic 360-D instrument manual is necessary for successful use of the chemagic DNA CS200 kit.

2. Do not use kit reagents after the expiry date printed on the kit label. Once opened, the reagents can be used for the time period stated in the reagent listing of this kit insert.

3. Any deviation from the protocol may affect the results.

4. The reagents are automatically dispensed in whole rows and therefore the tip covers (included in the kit) should be used also in whole rows on each rod in contact with any reagent solution. It should also be noted that if partial plates are run, the solutions may not be sufficient for 960 extractions.

5. Opening the chemagic 360-D instrument door while the automated extraction run is on-going, will terminate the run and the samples in process may be lost.

6. The cleaning and maintenance of the system is described in detail in the chemagic 360-D User Manual.
   - The system cleaning is performed once per week: Clean the chemagic Dispenser. Select the protocol ’**regular cleaning procedure 96 dispenser 360 V150116.che’** and press the [Insert IDs] or the [Start] button if the enhanced functions are deactivated. Follow the instructions as given in the software.
   - Prior to the next use of the chemagic Dispenser perform the appropriate priming protocol.
   - The cleaning of the chemagic Dispenser with 70 % ethanol is recommended once per month. Simply use the **’intensive cleaning procedure H96 dispenser 360 V150116.che’** instead of the regular one for this purpose.
   - If the chemagic Dispenser will not be used for longer period of time it is mandatory to perform the "regular cleaning procedure" to maintain the performance of the instrument when bringing it back into service.

7. The amount of DNA from whole blood sample obtained can be quantified using an independent method, e.g. UV measurement.

LIMITATIONS OF THE PROCEDURE

In some cases traces of Magnetic Beads B may be left in the eluate. Though such particles will usually not interfere with PCR or most downstream applications, an additional separation step either using a magnetic separator (chemagic Stand 96, provided with the chemagic 360 96 Rod Head Set prod. no. CMG-370) or centrifugation is recommended, in order to separate any traces of particles. For UV measurement of DNA eluates from whole blood samples, traces of Magnetic Beads may cause a higher background and a separation step should be done prior to quantitation.

Extracted DNA should be used immediately after extraction in the desired *in vitro* diagnostic test.

The kit is not intended to be used for extraction and purification of human genomic or human cfDNA from plasma sample, or human cfDNA from whole blood sample.

The DNA yield strongly depends on blood characteristics, e.g. low leukocyte count results in a decreased DNA yield.

PERFORMANCE CHARACTERISTICS

Blood samples

The performance of the 3207-0010 chemagic DNA CS200 kit and 2024-0010 chemagic 360-D instrument using whole blood samples was established by conducting DNA extractions specimens from healthy donors. The mean yield for each specimen was calculated and plotted against the white blood cell count. Table 1 shows the descriptive statistics of the specimen means and the DNA yields are presented in figures 1 and 2 using both EDTA and citrate sample tubes. Note: The DNA yield is also effected by the sample dilution caused by the used sample tube (the volume of preservative is different in EDTA and citrate sample tubes).

The results on the purity of the extracted DNA are presented in figure 3.

Table 1. The descriptive statistics of the specimen results

Variable	N	Median	Mean	Minimum	Maximum
Citrate Yield (µg/200 µL sample)	41	4.6	4.6	3.0	6.6
EDTA Yield (µg/200 µL sample)	41	5.1	5.2	2.8	7.9

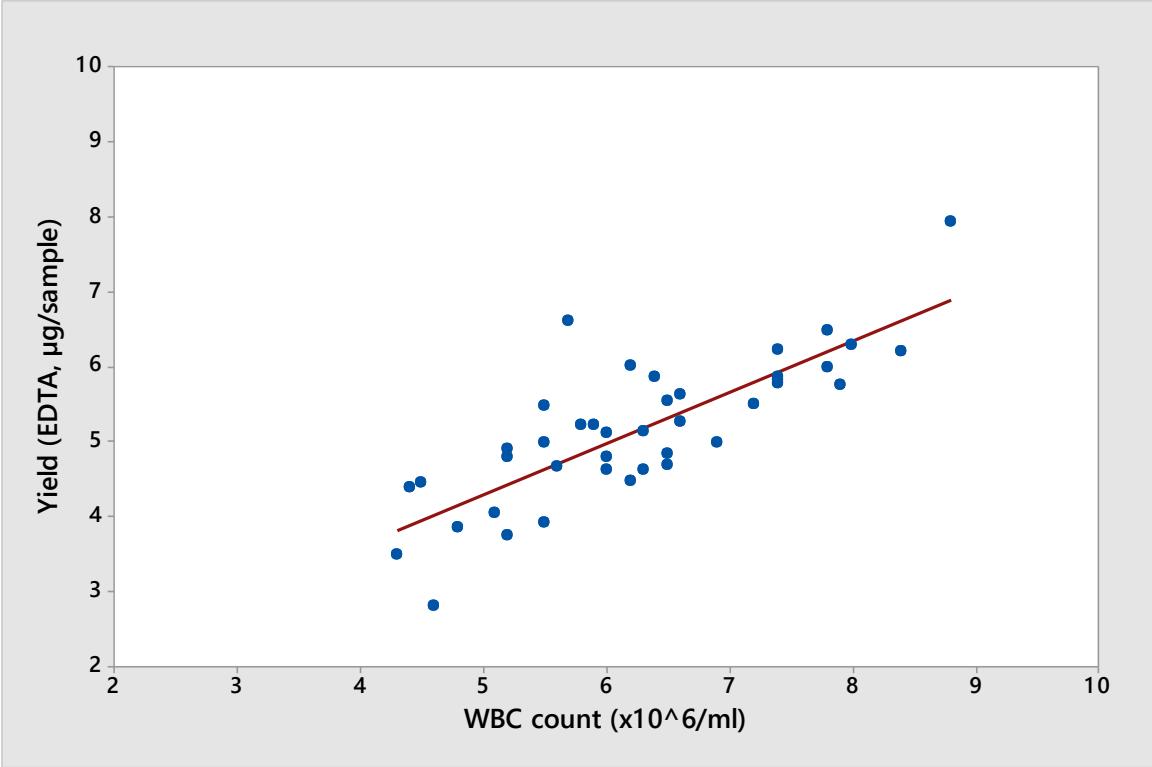

Figure 1. The DNA yield (EDTA tubes, 41 specimens) from 200 µL sample volume. White blood cell counts of healthy donors were determined and were in the range 4.3 – 8.8 x 10e6 cells/mL.

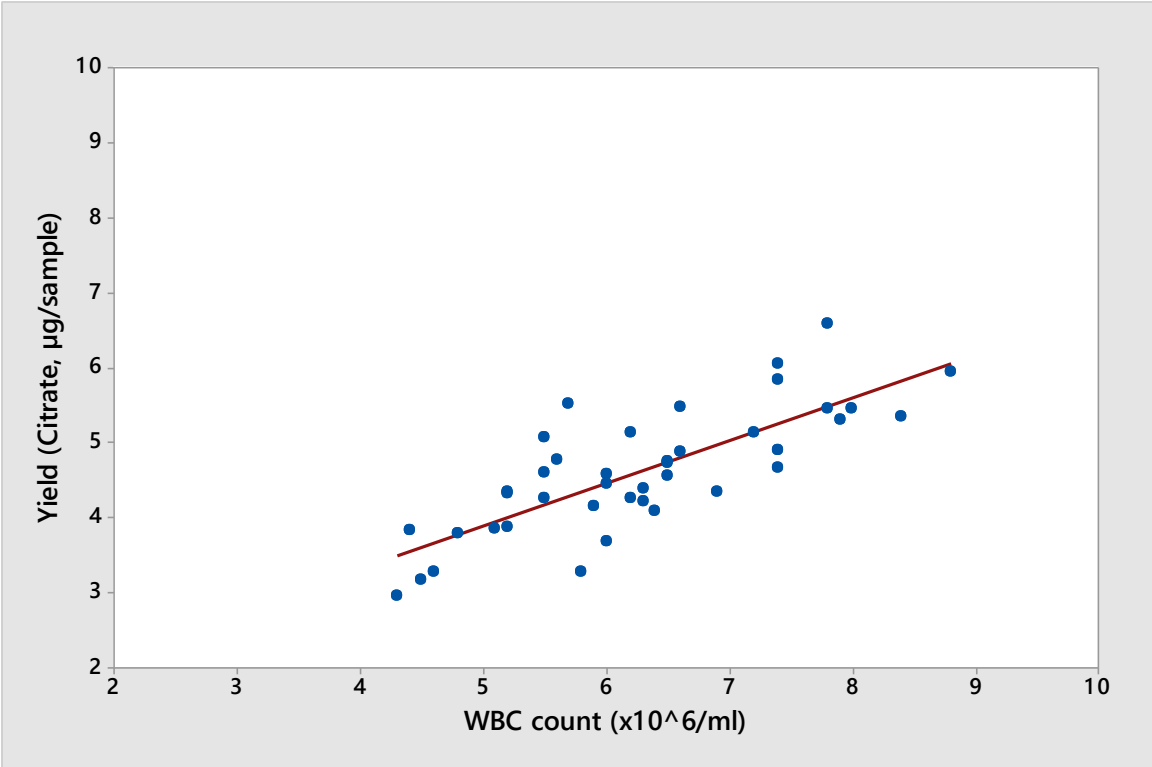

Figure 2. The DNA yield (citrate tubes, 41 specimens) from 200 µL sample volume. White blood cell counts of healthy donors were determined and were in the range 4.3 – 8.8 x 10e6 cells/mL.

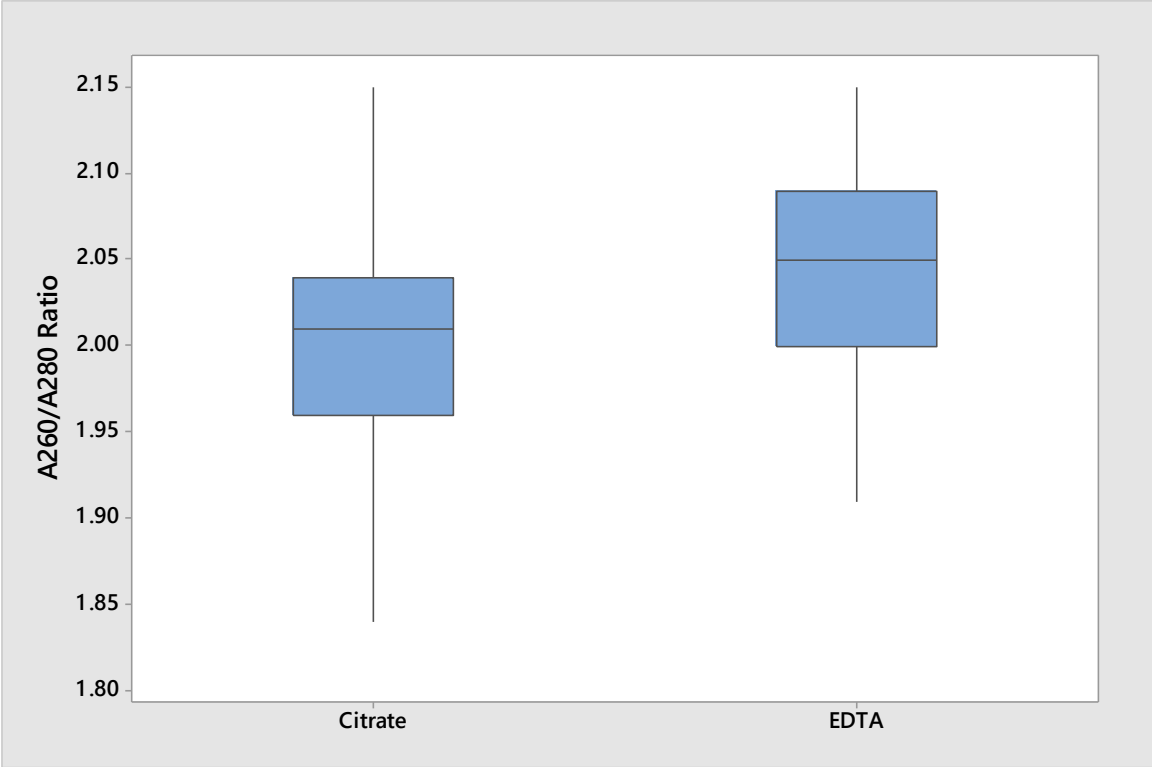

Figure 3. The purity of the extracted DNA (Absorbance ratio A260/A280) of 41 Citrate specimens and 41 EDTA specimens.

Plasma samples

The performance evaluation study of 3207-0010 chemagic DNA CS200 kit and 2024-0010 chemagic 360-D instrument using plasma samples was conducted in FINAS accredited testing laboratory (EN ISO/IEC 17025) using CE IVD registered downstream application for diagnosis of Cytomegalovirus (CMV). The results of positive patient samples with CMV copy numbers are presented in table 2.

Table 2. The results of CMV positive patient samples

	Investigational Device (chemagic DNA extraction platform)		Comparator Device (CE IVD registered DNA extraction platform)	
Sample ID	CMV kit result (copies / mL)	Evaluation of Detection	CMV kit result (copies / mL)	Evaluation of Detection
001	2400	Positive	650	Positive
003	700	Positive	2100	Positive
005	2000	Positive	1500	Positive
007	600	Positive	550	Positive
009	650	Positive	450	Positive
011	200	Positive	100	Positive
013	1100	Positive	300	Positive

015	24000	Positive	14000	Positive
017*	**50**	**Positive**	**NA**	**Negative**
019	16000	Positive	9900	Positive
021	6.8x10E6	Positive	4.5x10E6	Positive
025	8600	Positive	3800	Positive
029	NA	Negative	NA	Negative
031	1200	Positive	250	Positive
033	1000	Positive	800	Positive
035*	**NA**	**Negative**	**100**	**Positive**
037	2000	Positive	2300	Positive
039	400	Positive	100	Positive
041	250	Positive	150	Positive
043	84000	Positive	67000	Positive
047	1100	Positive	1000	Positive
049	27000	Positive	15000	Positive
051	1300	Positive	1100	Positive
053	9500	Positive	13000	Positive
057	5000	Positive	2300	Positive
059	230000	Positive	130000	Positive
061	1200	Positive	1400	Positive
065	1600	Positive	2700	Positive
067	16000	Positive	11000	Positive
069	5700	Positive	4300	Positive
071	8400	Positive	4100	Positive
073	83000	Positive	70000	Positive
075	4200	Positive	5900	Positive
077	950	Positive	1400	Positive
079	800	Positive	400	Positive
081	2000	Positive	600	Positive
082	2200	Positive	1100	Positive
083	750	Positive	600	Positive
084	1200	Positive	350	Positive
085	500	Positive	300	Positive

* The virus copy numbers of samples 017 and 035 are below the detection limit of the downstream assay.

WARRANTY

The performance data presented here were obtained using the assay procedure indicated. Any change or modification of the procedure not recommended by the manufacturer may affect the results, in which event Wallac Oy and its affiliates disclaim all warranties expressed, implied or statutory including the implied warranty of merchantability and fitness for use.

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No content changes between the current and previous version. Company name and logo updated.

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