Instructions for Use

LACTOSE INTOLERANCE TEST

Reference 133001007

Version 2, March 2023

Medical device for In Vitro Diagnostic Use

CE IVD

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The manufacturer continues to improve its products and reserves the right to change the information at any time.

If the product is used in a manner not specified by the manufacturer, the device may be compromised. Refer to the product's warnings, precautions, measures to be taken and limitations for use.

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INTRODUCTION

The Doctor Vida Genetic Test - Lactose Intolerance Test is a CE-IVD test, for diagnosis of lactase persistence or primary lactose intolerance in the Caucasian population.

This kit is an in-vitro diagnostic assay that allows the qualitative detection of polymorphisms in a specific region in intron 13 of the MCM6 gene, namely -13910 C/T (rs4988235) by Loop-mediated isothermal amplification (LAMP) from a sample human biological (buccal swab or drop of blood). These tests allow the identification of the phenotype of each individual relative to the persistence or non-persistence of lactase (enzyme that degrades lactose) in adults and, in turn, determine the degree of tolerance to lactose. Lactose intolerant individuals have a CC genotype, individuals with lactase persistence can digest lactase (TT genotype) and heterozygous individuals (CT genotype) generally have several degrees of lactase persistence and thus several levels of lactose intolerance.

The LAMP technique is an isothermal nucleic acid amplification technique, where the target sequence is amplified at a constant temperature (around 65°C). This technique needs 3 pairs of specific primers that serve to identify 8 distinct regions of the target gene, thus increasing the specificity. The detection of each sample's genotype is done by analysing the melting curve after amplification.

INTENDED PURPOSE OF THE DEVICE

The Lactose Intolerance Test is suitable for detecting the -13910 C/T genetic variant in the MCM6 gene associated with lactase persistence among adults of the Caucasian population from a human biological sample, namely buccal swab. The result of the test helps in the diagnosis of patients with suspected primary lactose intolerance. Although typical symptoms are abdominal pain, diarrhea, nausea and meteorism after consuming products with lactose, lactose intolerance in adults is not considered a genetic disease, but rather an ancestral trait.

This test must be used in combination with the Dr Vida Pocket device (V6) for DNA molecular amplification and detection in 60-90 minutes. The system is suitable for near-patient-testing and should only be used by healthcare professionals. The user is responsible for the analysis of the sample, the compliance with applicable biosafety rules including the use of adequate personal protective equipment, the maintenance of the facilities and the treatment of waste resulting from the analysis.

TEST PRINCIPLES

Each Lactose Intolerance Kit assay is performed on a sample of buccal swab collected with a swab and placed in lysis buffer in the collection tube (Tube A) and is always performed by a healthcare professional following good practice procedures using the appropriate material included with this product and adequate personal protective equipment not included. After a 10-minute incubation at room temperature, approximately 10 µL of sample is added using component C (disposable dispenser) to the reaction tube (Tube B) following the instructions described in this document. The analysis is performed by isothermal amplification (Loop-mediated isothermal amplification, LAMP) which requires 6 types of primers designed to hybridize distinct regions around the polymorphism (primers FIP and F3 at the 3' end and primers BIP and B3 at the 5' end and the loop, LF and LB primers) and is based on the amplification at a constant temperature of the region of the MCM6 gene containing the -13910 C/T variant. Detection of the MCM6 gene variant is done with mutation-specific probes by fluorescence quenching detection.

After amplification, the temperature drops to 40°C, allowing the probe to hybridize with the amplified fragment, which brings the fluorophore and quencher closer together, resulting in fluorescence *quenching* . During the analysis of the melting curve, the temperature gradually increases up to 70°C allowing the detection system included in the Doctor Vida Pocket device (Ref. 133001002) to detect the emitted fluorescence. As the probe is specific for the mutation (T), it does not hybridize perfectly with DNA fragments that do not contain the genetic variant (C) and therefore in these cases the emission of fluorescence occurs at lower temperatures than for DNA fragments that contain the genetic variant (T). The changes in fluorescence at different temperatures are detected by the Doctor Vida device allowing the distinction between the different nucleotides in the region of the -13910C/T. On the other hand, if there is no fluorescence emission or if it is residual, it means that the amount of biological material is below the detection limit of the technique or there are inhibitors in the sample causing an invalid result. The analysis of the sample in Dr Vida device, the results and report obtained are communicated to the user and the patient through the Dr Vida Pocket PCR (V3) app installed on the mobile phone.

QUALITY CONTROL

The quality control procedure is intended to monitor the performance of the kit reagents produced and the test assays themselves. The components of this product are manufactured in accordance with the lactose intolerance test manufacturing and quality control procedure, ISO 13485 and applicable regulatory requirements.

Each batch produced is properly tested along with Doctor Vida device and the Certificate of Analysis (*CoA*) for each batch is available upon request. For each batch, tests are carried out with negative controls (one with H_2O and another assay with only collection solution) that allow to verify if the kit reagents have any nucleic acid contamination and positive controls (2 assays with 2 samples with known genotype - aliquot of human cell line A549, 50 cells/µL, with TT genotype and the control LacI – Ctrl+, with CT genotype) essential to evaluate the efficiency of the procedure and also allows the verification of the quality of the reagents (integrity of the probes, primers and the enzyme activity).

SYMBOL TABLE

	Consult instructions for use		CE Mark
	Catalogue number		In vitro diagnostic medical device
	Lot number		Temperature limits
	Manufacturer		Contains sufficient for <n> tests
	Do not reuse		**Caution:** Follow the instructions in this manual; improper use may cause damage to the device or health
	Use by		
	Device for near-patient testing		The device is not intended for self-testing

WARNINGS, PRECAUTIONS, SAFETY MEASURES AND LIMITATIONS FOR USE

Warnings, precautions and safety measures

When performing a Lactose Intolerance test it is necessary to consider the instructions for use provided with the product, namely the warnings, precautions and safety measures of the test (see Table 1), as well as the Lactose Intolerance Safety Data Sheet sent with the product. Also consider the Doctor Vida device's precautions and safety measures (see the Doctor Vida and Software Instructions for Use), concerning:

- Fluid/Reagents handled near the device;
- Electrical power to the device;
- The operating environment of the device;
- Physical environment where you place the device

For operator safety and to ensure the reliability of the test, it is important that the following safety precautions are read and understood before performing the test.

- **Do not use any kit components if they are damaged or expired, or if the sample or reagent solution has potential contaminants (turbidity).** The use of unsuitable components may cause damage to health and/or compromise the reliability of the test. In this case, contact technical support.
- **Do not use components from different kits (lots) or components that are not included in the kit in the same assay.** The use of such components may compromise the reliability and veracity of the test result.
- **The handling of this product should be limited to healthcare professionals, taking into account the use of Good Laboratory Practices,** in order to prevent risks to the operator and to the integrity of the reagents.

- **Avoid reagent contamination and/or cross-contamination** when performing each assay:

  -Use appropriate personal protective equipment (gown, gloves, eye protection and mask) according to applicable regulatory guidelines;

  -Keep the cap open only on the tube you are using. Do not interchange or reuse tube caps.

  -Always change gloves and clean the device, required materials and surface before and after each test;

  -Perform amplification in an area isolated from the sample collection and reaction mixture (reagents + sample) preparation area;

  -Do not reopen reaction tubes after the assay to avoid releasing amplified DNA fragments (amplicons) in significant quantities;

  -Residue materials and reagents should be properly disposed of in the biological waste bag, according to applicable regulatory guidelines;

  -The facility should be **cleaned daily with DNA-free products, and ventilated (natural ventilation).**

- All users should be aware of the instruction manual for both the test and the Doctor Vida device. **Failure to follow the instructions may cause damage to the device and/or cause harm to health.**

Board 1 – Precautions and safety measures for the Lactose Intolerance Test Kit

Limitations of use

- Failure to follow proper sample transport, storage, collection, processing, and analysis procedures may invalidate or compromise the test result due to cross-contamination.
- The possible existence of additional rare mutations that may generate false genotyping results. Therefore, the manufacturer should evaluate possible genetic alterations that compromise the result and report them as a limitation, if applicable.
- If the sample amount in the assay is less than the limit of detection (LoD=5 cells per reaction), the result may be invalid and you will have to repeat the sample collection and have to use a new test kit.
- The presence of certain substances in the patient's blood, derived from the patient's state of health, may interfere with the test result. List of previously described substances that have interfered with test performance.

Substances	Concentration of substances in human whole blood
Bilirubin	5 mg/dl whole blood
Cholesterol	250 mg/dl whole blood
Potassium EDTA	10 mg/ml whole blood
Triglycerides	500 mg/dl whole blood

Table 7 – List of substances interfering with the test.

BEFORE STARTING THE EXPERIMENT

Materials Provided

The Lactose Intolerance Test Kit consists of the test package and the accessories provided to perform the test. Each component is single use.

Componentes do Kit	Descrição	Quantidade	Condições de armazenamento
Test packing	Tube A (Collection tube)	200µL collection solution	-25°C a -15°C
	Tube B (Assay tube)	90µL reaction solution	
	Component C (Disposable dispenser)	1 unit	
Accessories	Component D (Disposable swab)	1 unit	4°C a 25°C
	Biological waste bag	1 unit	Ambient temperature

Table 1 – Components and storage conditions of the Lactose Intolerance Test kit

Materials required for testing, but not provided

-Dr Vida device (including power supply and micro-USB-B cable) - available under reference no. 133001002
-Dr Vida Pocket PCR app (V3) – available on Google play and iOS store
-Cell phone with Bluetooth – to install and use the Dr Vida Pocket PCR app.
-Internet (wireless) – required for the transfer of data to the server.

To perform a Lactose Intolerance test it is necessary to ensure that the Doctor Vida Pocket PCR app is installed on your mobile phone and that the Doctor Vida device is plugged in and connected to the mains. For further details please refer to the Doctor Vida device and software Instructions for use.

Product transport

For the transport of the Lactose Intolerance Kit to be carried out accordingly, the test package is properly packed in styrofoam boxes containing dry ice or ice sheets, to guarantee the temperature conditions of the components (-25ºC to -15ºC). The Test accessories are packaged and transported at room temperature.

Product storage

The components of the Lactose Intolerance Test Kit must be stored in accordance with the temperature printed on the labels of each component, as described in table 1 - Components and storage conditions of the Lactose Intolerance Test kit until expiration date indicated on the label.

Product handling

When handling the Lactose Intolerance Test kit, keep in mind that:

- For each test, use only 1 test package, which is for single use only.
- Place the collection tube (Tube A) and the assay tube (Tube B) contained in the package on a support and allow it to defrost at room temperature;
- After completely defrosting, the tubes are ready to start the test.

⚠ **Avoid freeze-thaw cycles as this will decrease product efficiency**.

⚠ **Minimise light exposure in the assay tube (Tube B) as this will decrease the efficiency of the product.**

Product stability

This product maintains the stability of the reagents and the performance of the kit in accordance with the expiry date imposed on the packaging label, provided that the conditions of transport, storage and handling are met.

Classificação dos reagentes do Kit do Teste

The classification of the substances and/or mixtures of substances present in the Lactose Intolerance Test Kit has been carried out in accordance with Regulation (EC) No 1272/2008 and is available for consultation in the Safety Data Sheet of the test kit concerned.

Analytical performance of the Lactose Intolerance test

The analytical validation of the Lactose Intolerance test was performed in combination with the Doctor Vida® device and the Dr Vida app. To analytically validate the test, several analyses were performed:

a. Intra-batch reproducibility

Intra-batch reproducibility was assessed on 2 sample types in triplicate within the same batch. The data collected shows that the replicate results corresponded to 100% of the expected qualitative results, thus an excellent reproducibility of results within the same batch (see table 2).

# Batch	Sample	% Correct results (Expected Result/Replicates)	Total % Correct Results
22AT002	NTC (Harvest Buffer)	**Expectable Result**: Invalid 100% (3/3)	**100%** (6/6)
	POS (Cells A549)	**Expected result**: TT 100% (3/3)	

Table 2 – Analysis of the Intra-lot Reproducibility

b. Inter-batch reproducibility

Inter-lot reproducibility was evaluated on the same 2 sample types in triplicate with 3 different lots. The data collected indicate that in the 3 batches tested, all results matched 100% of the expected qualitative results, which means that there is little variability from batch to batch (see table 3).

Sample	# Batch	% Correct results (Expected Result/Replicates)	Total % Correct Results
NTC (Harvest Buffer)	22AT002 22AT003 22AT004	**Expectable Result**: Invalid 100% (3/3)	**100%** (6/6)
POS (Cells A549)	22AT002 22AT003 22AT004	**Expected Result**: TT 100% (3/3)	

Table 3 – Inter-batch reproducibility analysis

c. Analytical Sensitivity (Limit of Detection)

To determine the limit of detection (LoD) of the test, triplicate assays were performed using different concentrations of a sample with known concentration and known genotype. The data collected denote that for a detection rate > 95% the lowest concentration level is 5 cells per reaction (i.e. 100 cells/mL), thus defining the analytical sensitivity of the test. (See Table 4).

	Sample level (No. of cells A549/rection)	% Correct results (Detected/Replicated)
	1	33% (1/3)
LoD→	5	100% (3/3)
	10	100% (3/3)
	50	100% (3/3)
	75	100% (3/3)
	100	100% (3/3)

Table 4 – Analytical sensitivity of the test (LoD)

d. Stability of the test in storage

In order to analyse the storage stability of the test and thus determine its shelf life, assays were performed in duplicate and with the same lot, stored at -20ºC with different time periods (0, 32, 60 and 120 days). The results collected demonstrate that the tests performed after 120 days of storage at -20ºC are still fully functional. Therefore, the validity period of the test was set to 4 months under the conditions defined on the packaging and in this document (See table 5).

Sample	Storage time (days)	% Correct results (Expected Result/Replicates)	Total % Correct Results
NTC (Harvest Buffer)	0	**Expectable Result**: Invalid 100% (2/2)	**100%** (16/16)
	32	**Expectable Result**: Invalid 100% (2/2)	
	60	**Expectable Result**: Invalid 100% (2/2)	
	120	**Expectable Result**: Invalid 100% (2/2)	
POS (Cells A549)	0	**Expected Result**: TT 100% (2/2)	
	32	**Expected result**: TT 100% (2/2)	
	60	**Expected result**: TT 100% (2/2)	
	120	**Expected result**: TT 100% (2/2)	

Table 5 – Test storage stability analysis

Clinical performance of the Lactose Intolerance test

The clinical performance of the Lactose Intolerance Test - isothermal method was performed in comparison with the laboratory reference laboratory method - Sanger method sequencing (using DNA purified from FTA cards). The clinical validation study was performed following a non-probability sampling with a total of 63 mouth exudate samples from volunteers from Portugal (N=63).

All data were analysed using the 3x3 confusion matrix method, after exclusion of invalid results. General statistics and statistics by genotype classes were calculated using the statistical computing software R (R Foundation, version 4.2.2).

For clinical validation study, the results obtained by the portable isothermal laboratory method showed an overall accuracy of 98.41% (with 95% confidence interval between 91.47% and 99.96%) compared to the reference method. From a total of 63 assays performed only 1 had an invalid result (1.59%) and another had a genotyping not concordant with reference method - *Sanger* Sequencing.

In the clinical validation the statistical analysis per class of genotics was also performed. The result of the relative sensitivity per class is 96.67%, 100% and 100%, for the genotypes CC, CT

and TT respectively. The result of relative specificity by class is 100%, 97.44% and 100%, for the genotypes CC, CT and TT respectively. In the same study the positive predictive value was also calculated for each of the genotypes, CC (100%), CT (96%) and TT (100%) and negative predictive value for each of the genotypes, CC (97.06%), CT (100%) and TT (100%). Parameters such as prevalence, detection rate, detection prevalence as well as balanced accuracy were also calculated for each genotype (see table 6).

Buccal swab samples

Number of invalid assays: 1/63 (1.59%)

		Reference method – Sanger sequencing			
		-13910 CC	-13910 CT	-13910 TT	**Total**
Isothermal lab-on-phone - Lactose Intolerance	-13910 CC	28	0	0	**28**
	-13910 CT	1	24	0	**25**
	-13910 TT	0	0	9	**9**
	Total	**29**	**24**	**9**	**62**

Overall statistics

Accuracy: 98.41%
95% CI: (91.47% - 99.96%)
No Information Rate: 47.62%
P-Value [Acc > NIR]: < 2.2e-16
Kappa: 0.9739

Class statistics by Genotype:	-13910 CC	-13910 CT	-13910 TT
Sensitivity	96.67%	100%	100%
Specificity	100%	97.44%	100%
Pos Pred Value	100%	96.00%	100%
Neg Pred Value	97.06%	100%	100%
Prevalence	47.62	38.10%	14.29%
Detection Rate	46.03	38.10%	14.29%
Detection Prevalence	46.03	39.68%	14.29%
Balanced Accuracy	98.33%	98.72%	100%

Table 6 – Statistical analysis of the clinical performance of the lactose intolerance test - portable isothermal laboratory method vs. reference method - Sanger sequencing.

Preparation of the site and all equipment used in the test

Before starting the sampling and the analysis of the test itself, you should prepare all the place where the test is carried out and also all the equipment and material inherent to the analysis of the test.

- The place should have an ambient temperature between 15ºC to 30ºC and humidity levels between 20 to 80%;
- Keep the place clean and ventilated, daily natural ventilation;
- Have well defined spaces (with a minimum acceptable distance) for the sample collection, transferring it to the assay tube (test B) and analysing the assay;
- Clean and disinfect work surfaces with 10% bleach and 70% ethanol to mitigate the risk of contamination;
- Wipe the outside of the Dr Vida Pocket devices with paper moistened with 70% ethanol (do not spray) and leave it to air dry;
- Remove the lid from the Dr Vida Pocket and clean it with paper dampened in 70% ethanol and leave it to air dry;

  Note: Be careful not to get Ethanol into the hole where the test tube is placed

- Keep the lid open until a new test;
- Put on some new gloves at the beginning of each analysis.

For further information please read the Installation topic in the Doctor Vida and Software instructions for use (https://www.stabvida.com/drvida/INSTRUCTIONS-PT or QR code).

a. Connecting the Dr Vida device

Plug Dr Vida device into the mains or connect to a portable charger, output: 5VDC, 2A. The device may take a few minutes to stabilise the temperature.

b. Installing the Dr Vida Pocket PCR app

Install and configure the Dr Vida Pocket PCR app on your mobile phone (see Doctor Vida device instructions for use).

c. Select the test to be performed and Pair the Dr Vida device

On the main screen of the app, select the Doctor Vida genetic test you want to perform - "Lactose Intolerance". Then click "Select device" and connect the device you want manually or scan the QRcode of the device you want to select. The device is ready to perform the test.

d. Registering the information of Doctor Vida's genetic test reagents

The information of the genetic test reagents in question is recorded, either by manually entering the test reference and lot number or by scanning the QRcode included in the kit.

e. Fill in the sample data

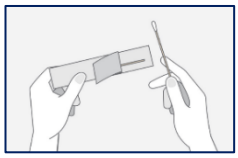
Click on "read sample ID" to be able to fill in all the data relative to the sample to be tested, such as the ID itself and the type of sample in question, "Mouth Exudate". Also, you have the option to select the "Test with certificate at the end" and for that you should go to "More details" fill in or verify (in case the filling in was done by reading the QRcode of the sample) the patient's data.

Collecting the sample - buccal swab

Sample collection should be performed by healthcare professionals in accordance with national regulatory guidelines, following the instructions in this Instruction Manual and the diagrams provided in the Dr Vida Pocket PCR app.

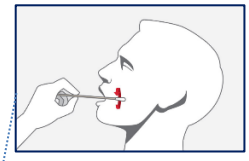
Avoid eating, drinking or smoking 30 min before collection

General instructions:

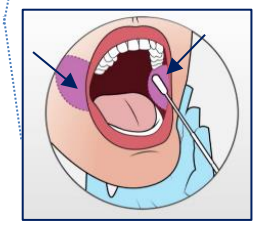


a) Remove the swab supplied from its sterile packaging, taking care to avoid contact with any object or surface.

⚠ Hold the swab by the plastic end and keep your fingers away from the cotton area.

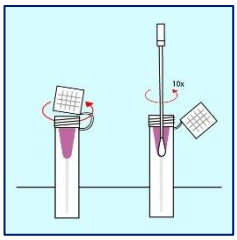


b) Insert the swab into your mouth and rub the inside of both cheeks vigorously in circular motions for at least 30 seconds.



⚠ The aim is to collect a swab from the inner wall of the cheek(s). An insufficient sample may require a further collection.

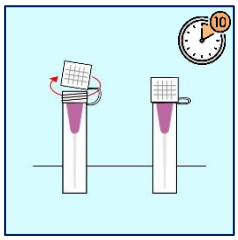
c) Remove the swab carefully so it does not touch the teeth, lips or other surface.



d) Unscrew the cap of the sample collection tube (tube A) containing the pink liquid, place the swab inside and rotate it repeatedly about 10 times.

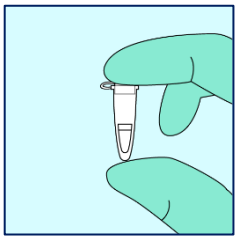
e) Remove the swab to ensure maximum sample transfer and dispose of it in the biohazard bag.

f) Close tube A with its screw cap and leave it to incubate at room temperature for 10 minutes.



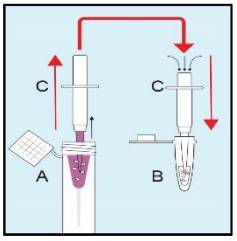
- If **you want to** analyse the sample, follow the procedure described in "Analysis of the sample".
- If you do **not wish to** analyse the sample immediately, store it at 2-8°C for up to 24 hours.

Sample handling and preparation and analysis



g) Remove tube B and thaw it with your fingers until the liquid is completely clear.

h) After the 10 minutes of sample incubation, unscrew the cap of tube A, and collect 10 µL of sample using the supplied sample dispenser.



⚠ Make sure to leave the sample dispenser plunger out of the tube to prevent it from getting wet.

⚠ If the sample volume does not rise to the mark on component C, remove and replace the dispenser tip several times so that the pressure differential makes it easier to raise the total sample volume.

i) Dispense the collected sample into tube B by placing the tip of component C into the tube and plugging the hole at the top, then lightly depressing the plunger.

⚠ After dispensing, a pink intermediate layer should be visible inside tube B

⚠ Close tube A again and dispose of it in the biological waste bag provided.

j) Close tube B tightly and tap the tube on the bench 5 times to mix the sample and reagents well.

k) Check that the information that was previously entered is correct in the 'Settings' menu and edit if necessary.

l) Click on start assay and wait for the device to verify that the requirements to start are met.

m) When prompted, insert the B tube into Doctor Vida, unscrew the lid, place the tube and screw the lid back on.

n) The test will start automatically and will take approximately 90 minutes.

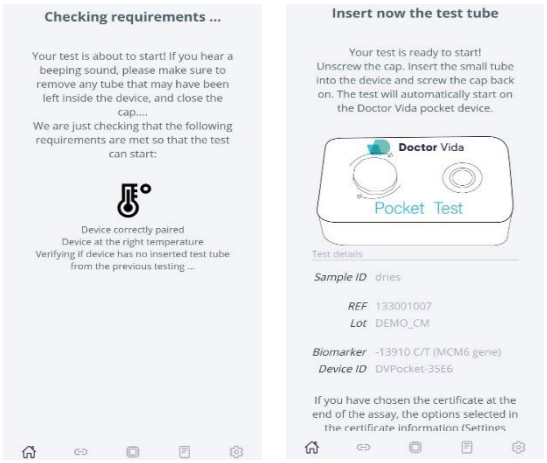


Figure 1 – Screenshots of the app – Start the assay

⚠ Do not touch or move the Dr Vida device while the sample is being processed as this may compromise the reliability of the test.

⚠ While the test is in progress, do not disconnect the Dr Vida device from the mains.

Note: If you lose internet or *Bluetooth* connection while the assay is in progress, you will not lose the assay. Just reopen the app and select the 'Pair available devices' screen to reconnect the device(s) that are in progress.

Consult the Result obtained

a) Once the test is complete, Dr Vida device transfers and stores the data in real time to the Doctor Vida® API server via the Dr Vida Pocket PCR app.

Note: It may take about 2 minutes to transfer the data until you get the result.

b) After data transfer, click on the 'Results' button in the app to be able to view the test result and/or consult the test result certificate in your email if you have selected and registered that option.

c) The test result tells us the nucleotide(s) present in the -13910 C/T polymorphism region (rs4988235), based on the temperature at which the peak(s) is (are) observed.

d) The lactose intolerance test assay may have three possible results (see Table 8). If no peaks are observed the result is considered invalid, therefore the test must be repeated, including a re-collection.

Interpretation of results				
Chart				
Description and Chart Interpretation	1 Peak at 48-54ºC - Indicates presence of Nucleotide C.	1 Peak at 48-54ºC and 1 Peak at 56-64ºC - It indicates the presence of Nucleotide C and T, simultaneously.	1 Peak at 56-64ºC - Indicates presence of T nucleotide.	0 Peaks - indicates test failure. Repeat.
Genotype	**CC**	**CT**	**TT**	**Invalid**
Phenotype	Primary lactose intolerance - Lactase deficiency in adults.	Different degrees of primary lactose intolerance.	Lactose tolerance - Lactase persistence in adults	--

Table 8 – Several possibilities of Lactose Intolerance test results.

Cleaning of the site and equipment and waste disposal

At the end of each test, ensure that the different rooms are cleaned and ventilated as well as all the equipment used in the test, including the Doctor Vida device and ensure that the disposal of the tubes A and B and all the inherent material is carried out according to the rules for the treatment of biological waste and in compliance with regulatory guidelines.

- After the analysis, open the lid of the Doctor Vida device, remove the tube B and dispose it in the biological waste;
- Also dispose of tube A as well as the dispenser in the biological waste;
- Wipe the outside of the Dr Vida device and the lid with paper moistened with 70% ethanol (do not spray) and leave to air dry;

Note: Be careful not to get Ethanol into the hole where tube B is placed.

- Clean and disinfect work surfaces with 10% bleach and 70% ethanol to mitigate the risk of contamination;
- Keep the place clean and ventilated, daily natural ventilation;
- Keep the device lid open until a new test;
- Dispose of paper and gloves used in cleaning the space and equipment to bio-waste.

Note: Do not leave Dr Vida device switched on if you are not using it.

TROUBLESHOOTING

Problem observed	Possible Solution
- Failed after starting the test. Example: Power failure at the device during analysis.	- Turn off Dr Vida device and close the Dr Vida Pocket PCR app; - Reopen the Dr Vida Pocket PCR app; - Discard the old tube B and restart the process using a new tube B.
- Failure of the app to achieve a result. Example: The app appears to have the test blocked, and cannot obtain the result.	- Close the Dr Vida Pocket PCR app; - Check internet connection and Bluetooth; - Open the app and select the genetic test 'Lactose intolerance' again; - Click the 'Select device' button and connect the device where the test is taking place; - Select in the main menu the 3rd button 'Running assays', to be able to consult the test in progress; - If you still get an error, unpair the device and pair it again. It is important to check that the same device is not connected to more than one mobile phone; - If you can start a new test with this device, it means that either a power failure occurred or the device automatically restarted. In this case the test becomes invalid. Discard the tube B that was in use. - Switch the device off and on again and start the process using a new tube B. - If the problem persists, please contact technical support.

Problem observed	Possible Solution
-The device does not appear in the list of devices Example: The connection between the device and the mobile phone has failed	- Check that the device is plugged in, that there is no power failure (for example the power cable is damaged). If this is the case, you can use the mobile phone cable; - Check that Bluetooth is on, if in the Dr Vida Pocket PCR app you have given location permission; - Refresh the screen by sliding your finger downwards.
- The phone is out of space to store the data.	- If you run out of space before starting the assay you should free up space on your phone or use another phone. - If you run out of space during the assay, the assay will continue, however the results will not be available. In this case, do not disconnect your Doctor Vida device as the data will be stored on it. Free up some space on your phone and the connection will be restored. **Important note:** If you disconnect the device, all data will be lost and you will have to repeat the test with a new tube B.
-Test result invalid Example: If there is no peak in the melting curve	- A new test must be performed, repeat the sample collection and also the sample analysis **Note:** This type of result may occur when there is some inhibitor present in the sample or lack of biological material.

BIBLIOGRAPHY

- Enattah NS *et al.* 2002. Identification of a variant associated with adult-type hypolactasia, Nature Genetics, vol.30
- Rasinpera *et al*, 2004**.** A genetic test which can be used to diagnose adult-type hypolactasia in children, Gut; 53: 1571-1576

WARRANTY INFORMATION

The product is covered by a warranty period for the period of validity indicated on the product. This guarantee is to protect you against the costs associated with problems arising from manufacturing defects. The guarantee period begins on the date of receipt of the product at the desired location. For support during the guarantee period, please contact the manufacturer.

FORMATION

These operating instructions describe the correct use and operation of the device. Operators of the device must familiarize themselves with the applicable sections in the document before conducting tests to ensure safe and efficient use of the device. Ensure that you follow the training requirements in accordance with applicable regulatory guidelines. If you need more information about training in the use of this product, contact the manufacturer.

MANUFACTURER INFORMATION

Name: STAB VIDA- Investigação e Serviços em Ciências Biológicas, Lda.

Address: Madan Parque, Rua dos Inventores, Sala 2.18, 2825-182 Caparica, Portugal.

Website: https://www.stabvida.com/drvida

Technical assistance

In case of any problems please contact us by email drvida@stabvida.com or telephone 00351927151763.

Monday to Friday from 10h00 to 19h00 (GMT)

According to EU regulation 2017/746, any serious incident occurring in relation to the device must be reported to the manufacturer and the competent authority of the EU Member State where the user and/or patient is established.

REVISION HISTORY

Review		Revised points	
NO.	Date (dd/mm/yyyy)	NO.	Date (dd/mm/yyyy)
1	25/05/2022	-	**Issuance of the document**
2	31/03/2023	All	**Adaptation of the document in accordance with the requirements of EU Regulation 2017/746 and relevant harmonized standards.**