Exemption Request Form

Date of submission: _____

1. Name and contact details

A) Name and contact details of applicant:

Company:	Roche Diagnostics Ltd.	Tel.:	+41 41 792 3741
Name:	Nick Lewis, M.Sc.	E-Mail:	nick.lewis@roche.com
Function:	Manager, Regulatory Affairs	Address:	Forrenstrasse 2 6343 Rotkreuz Switzerland

B) Name and contact details of responsible person for this application (if different from above):

Company:	Roche Diagnostics Ltd..	Tel.:	+41 41 798 5452
Name:	Dr. Prisca Zammaretti	E-Mail:	prisca.zammaretti@roche.com
Function:	Head, Regulatory Affairs	Address:	Forrenstrasse 2 6343 Rotkreuz Switzerland

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2. Reason for application:

Please indicate where relevant:

☒ Request for new exemption in:

☐ Request for amendment of existing exemption in:

☐ Request for extension of existing exemption in:

☐ Request for extension of existing exemption in:

☐ Request for deletion of existing exemption in:

☐ Provision of information referring to an existing specific exemption in:

☐ Annex III ☒ **Annex IV**

No. of exemption in Annex III or IV where applicable: **N / A**

Proposed or existing wording: **Lead in solders used to construct and connect to Peltier thermal cyclers used for in-vitro diagnostic analysers that use polymerase chain reaction**

Duration where applicable: **Until 31 December 2020**

☐ Other:

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3. Summary of the exemption request / revocation request

This exemption request is to allow the use of lead in the solder used to attach Peltier elements in in-vitro diagnostics (IVD) analysis instruments that analyse samples from human blood and tissue samples for a variety of diseases. The analysis procedure requires multiple, very precise thermal cycles which are provided by the Peltier heating elements. This precise temperature control is critical to the analysis procedure; incorrect temperature during any phase of the procedure can have significant detrimental effects on the analysis, which can in turn result in risk to patient safety.

The Peltier thermal cyclers are connected electrically with solder, and research has shown that most Peltier elements that are bonded with lead-free solders do not provide the same required precision of temperature control for the thermal cycles. One version has been shown to give the required precision, but has proven to be unacceptably unreliable, failing far sooner than the tin/lead soldered version. As with temperature control, similar risks to patient safety exist for Peltiers that fail to demonstrate sufficient reliability; instrument unavailability as a result of a non-functioning Peltier increases the risk that a time-sensitive result could not be obtained in time for a physician to take potentially life-saving action.

There are several different IVD analysers on the market produced by several manufacturers. Roche does not know which solders its competitors use, Roche has also been able to design three instruments that use Peltier thermal cyclers which are soldered using lead-free solder. However, none of these other instruments are equivalent to the designs that are the subject of this exemption request; in many cases, these other instruments cannot perform the same range of tests (which often is the most important factor for a laboratory),or they are designed for much larger, high-throughput laboratories.

National health service laboratories in EU Member States, as well as smaller private laboratories, often have limited budgets and thus may not purchase a new, high throughput instrument if a lower throughput instrument is sufficient for their needs. Because there also are stringent regulations associated with the accreditation or licensure of these laboratories, extensive planning, validation, and often, submissions to regulatory bodies are necessary to implement use of new instruments. The costs and effort associated with these requirements can be extensive.

Finally, it is important to note that if the current instrument that uses leaded solder were not available after July 2016, these laboratories may be forced to continue to use old instruments for longer than planned. These instruments could become increasingly unreliable, which could also have a negative effect on patients' health.

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4. Technical description of the exemption request / revocation request

(A) Description of the concerned application:

1. To which EEE is the exemption request/information relevant? Name of applications or products:

   IVD analysers of the types COBAS TaqMan and COBAS TaqMan48 Instruments (these models are made by Roche)

   a. List of relevant categories: (mark more than one where applicable)

   ☐ 1 ☐ 7

   ☐ 2 ☒ **8**

   ☐ 3 ☐ 9

   ☐ 4 ☐ 10

   ☐ 5 ☐ 11

   ☐ 6

b. Please specify if application is in use in other categories to which the exemption request does not refer: **Not applicable.**

c. Please specify for equipment of category 8 and 9:

The requested exemption will be applied in

☐ monitoring and control instruments in industry

☒ **in-vitro diagnostics**

☐ other medical devices or other monitoring and control instruments than those in industry

2. Which of the six substances is in use in the application/product? (Indicate more than one where applicable)

☒ **Pb** ☐ Cd ☐ Hg ☐ Cr-VI ☐ PBB ☐ PBDE

3. Function of the substance: **Constituent of solder used to join materials within and to Peltier element.**

4. Content of substance in homogeneous material (%weight): **37% Lead solder.**

5. Amount of substance entering the EU market annually through application for which the exemption is requested:

- **CTM: 4 elements x 0.56g per element = 2.25g per Instrument.**
- **CTM48: 2 elements x 0.56g per element = 1.125g per Instrument.**

Information regarding the total amount entering the EU market annually is confidential.

Please supply information and calculations to support stated figure. **See above.**

6. Name of material/component: **Lead with tin in a solder alloy.**

7. Environmental Assessment: **Not applicable to this exemption request.**

LCA: ☐ Yes
☐ No

(B) In which material and/or component is the RoHS-regulated substance used, for which you request the exemption or its revocation? What is the function of this material or component?

The component at issue is the Peltier element that is used in the Thermal Cycler units in the COBAS TaqMan (CTM) and COBAS TaqMan 48 (CTM 48) Analyzers. The lead is present in the solder used to join together the various internal components of the Peltier element, which include cables (copper), ceramic, and copper elements affixed to the ceramic. The joining together of these components within the Peltier element is critical for the efficient transfer of electrical and thermal energy.

Peltier elements are used for either heating or cooling and have many applications. Peltier heaters are constructed from a series of p-n junctions made from bismuth telluride. When a voltage is applied across the p-n junction, heat is transferred from one end to the other giving a hot end and a cold end so that these devices can be used for either heating or cooling.

For IVD analysers like the COBAS TaqMan, where the element is used for heating, a heat sink is placed at the cold end as the heat energy source. Peltier elements are an ideal choice for this application as reversal of the applied voltage draws heat away from the heated end so that accurately controlled temperature cycling is possible by controlling the applied voltage.

The temperature control provided by the Peltier is fundamental to processes integral to successful PCR. Accurate temperature control is critical for primer and probe hybridization as well as for DNA polymerase activity, which includes extension rate, processivity, and fidelity.

(C) What are the particular characteristics and functions of the RoHS-regulated substance that require its use in this material or component?

The RoHS-regulated substance, lead, is a constituent of solder that is used to make electrical connections to the Peltier elements. Lead solders such as 63%Sn37%Pb are known to have very different physical properties to lead-free solders such as Sn0.7%Ag0.3%Cu. Research has been carried out that shows that the thermal fatigue properties are very different and the comparable performance depends on the size of the stress on the solder bond.

There is an additional complication with soldering to Peltier elements in that the bismuth telluride is a brittle semiconductor material that can fracture and fail under sufficient strain. Lead-free solders that can be cycled over the ambient to 100˚C temperature range are all harder and less ductile than SnPb solders. When stress is imposed due to dimensional changes caused by thermal expansion mismatch, the more ductile SnPb can distort more easily and so reduce stress levels imposed on the bismuth telluride compared to lead-free solders.

The frequent temperature changes impose thermal stresses on the Peltier heater and in particular to the solder bonds that are used to make electrical connections to each element. In a lead-free application, the thermal coefficient of expansion (TCE) of bismuth telluride would be different to the metal used for making electrical connections, and also to the solder. The resulting TCE of Bi_2Te_3 would be different for the two directions parallel to and perpendicular to the p-n junction at 2.1 and 1.4 x 10^{-5} K^{-1}, respectively[1]. The TCE of lead-free solders are typically 2.2 x 10^{-5} K^{-1}, so stresses would be induced because of the smaller perpendicular TCE of Bi_2Te_3 as well as due to the different TCE of connecting metal parts.

One manufacturer of Peltier elements (Ferrotec) has published a detailed description of reliability issues around lead free solders[2]. This states that a “Mean Time Between Failures” (MTBF) of 200,000 hours is possible, but that the MTBF is “significantly worse” in applications involving thermal cycling. Ferrotec explains that four thermal cycling parameters affect the reliability of the Peltier element; 1) the total number of cycles, 2) the temperature range, 3) the upper temperature, and 4) the rate of temperature change. Parameters 2, 3 and 4 are fixed for PCR analysis and cannot be changed, and the number of cycles needs to be as many as is possible to maximize equipment lifetime.

In the example given by Ferrotec, the first failure occurred after approximately 30,000 cycles (MTBF was 68,000 cycles which is the mean failure rate for a series of tests). The results of the reliability analysis performed by Roche indicate that in some cases the elements will achieve less than half, and in one case less than one-third of the specified number of cycles, which supports the Ferrotec results. The lead-free alternatives also demonstrated markedly variable performance, with a wide range of cycle counts at element failure.

Refer to Section 6B for detail on the reliability testing performed on the alternative (lead-free) Peltier elements.

References:

1. “Thermal Expansion Coefficients of Bi2Se3 and Sb2Te3 Crystals from 10 K to 270 K”, X.Chen, et.al. Downloaded from http://arxiv.org/pdf/1112.1608.pdf
2. https://thermal.ferrotec.com/technology/thermoelectric/thermalRef10

5. Information on Possible preparation for reuse or recycling of waste from EEE and on provisions for appropriate treatment of waste

1) Please indicate if a closed loop system exist for EEE waste of application exists and provide information of its characteristics (method of collection to ensure closed loop, method of treatment, etc.)

A closed loop system exists for recycling the Pelier elements in compliance with all applicable EU and Swiss regulations.

2) Please indicate where relevant:

☐ Article is collected and sent without dismantling for recycling

☐ Article is collected and completely refurbished for reuse

☒ Article is collected and dismantled:

☐ The following parts are refurbished for use as spare parts: _____

☒ The following parts are subsequently recycled: **Peltier element**

☐ Article cannot be recycled and is therefore:

☐ Sent for energy return

☐ Landfilled

3) Please provide information concerning the amount (weight) of RoHS substance present in EEE waste accumulates per annum:

This information is deemed proprietary by Roche.

☐ In articles which are refurbished _____

☐ In articles which are recycled _____

☐ In articles which are sent for energy return _____

☐ In articles which are landfilled _____

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6. Analysis of possible alternative substances

(A) Please provide information if possible alternative applications or alternatives for use of RoHS substances in application exist. Please elaborate analysis on a life-cycle basis, including where available information about independent research, peer-review studies, development activities undertaken

Roche has initiated a comprehensive process to identify a suitable alternative Peltier element that complies with RoHS requirements. This effort is part of a larger, multi-step process that is generally required to implement significant changes to an IVD instrument and subsequently introduce the changed instrument into the market:

- Step One – Performance Feasibility: Verification that the alternative elements meet temperature performance specifications
- Step Two – Reliability/Endurance Testing: Assessment of the long-term reliability of the alternative elements
- Step Three – Assay Verification: After identification of a suitable alternative element, a verification campaign must be undertaken to confirm there is no impact to the performance of the assays run on instruments using the alternative elements
- Step Four – Customer Validation: Most IVD customers must perform extensive validation in their laboratories to assess the performance of the changed instrument

The following sections describe the steps Roche has undertaken to date in order to identify a suitable alternative Peltier element.

Performance Feasibility of Alternative Peltier Elements for the CTM/CTM48 Instruments

I. Background:

The Peltier element currently deployed in the thermal cyclers on the COBAS TaqMan (CTM) and TaqMan 48 (CTM48) Analyzers is not RoHS compliant. The feasibility of several alternative RoHS-compliant elements was evaluated.

II. Study Design:

The alternative Peltiers were evaluated using a series of different PCR profiles called the Combined Test Profile to evaluate two key performance parameters: ***temperature*** (achievement of specific temperature points within a defined specification band) and the ***ramp*** (the rate at which the temperature increases or decreases).

Determination of Temperature and Ramp Specifications

The Combined Test Profile was used to acquire data from 100 current Thermal Cyclers (containing the current non-RoHS compliant Peltiers) in order to define 74 temperature specification data points and 6 ramp specification data points which reflect the temperature band and the ramp performance for the current Thermal Cyclers.

With the temperature and ramp specifications set, the Combined Test Profile was then used to acquire data from Thermal Cyclers containing the alternative Peltiers, in order to assess whether their temperature & ramp performance were within the temperature and ramp specifications which were derived from the current Thermal Cyclers.

Physical Measurement Points Within the Thermal Cycler

The Thermal Cyclers include a “cup holder” with 24 wells, which correspond to the number of wells contained in the PCR amplification plate used with the CTM and CTM48 instruments; this cup holder is depicted in Figure 1.

Figure 2 summarizes the collection and analysis of the temperature and ramp data.

Figure 1: Well Positions / Measurement Points in the CTM Thermal Cycler Cup Holder

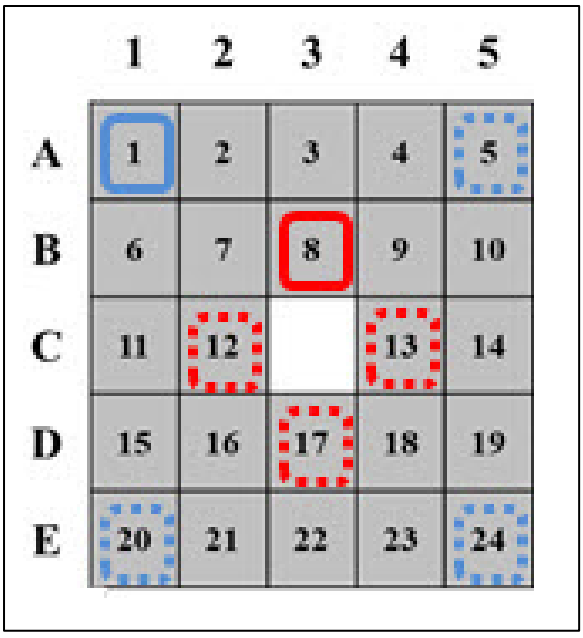
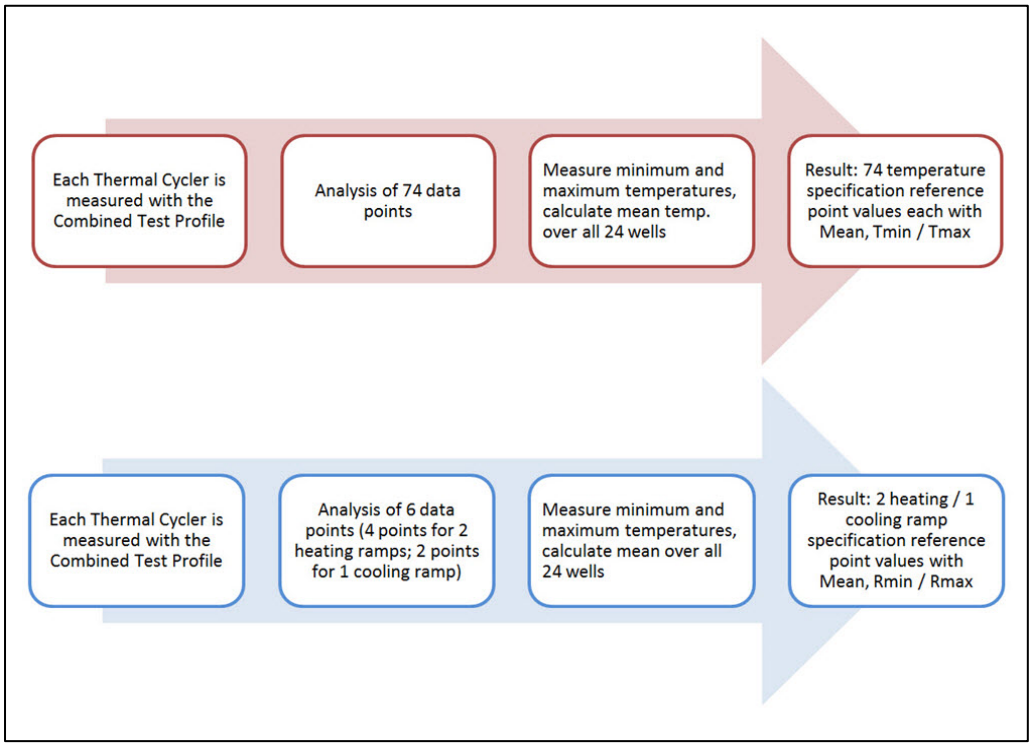


Figure 2: Collection and Analysis of Thermal Cycler Temperature and Ramp Data



III. Data Analysis and Acceptance Criteria:

A test case was considered to have passed if the actual outcome met the expected outcome. If the expected outcome was not achieved or other anomalies were observed, these findings were documented in the test report.

The following criteria were used to compare each alternative Peltier with the temperature and ramp specification reference point values. The tolerance specification (0.XX°C; this information is deemed proprietary by Roche) was derived from the uncertainty of the calibration process performed on the temperature measuring device; specifically, twice the uncertainty as documented on the calibration certificate for the device, in alignment with NIST Technical Note 1297 "Guidelines for Evaluating and Expressing the Uncertainty of NIST Measurement Results".

<u>Evaluating maximum temperature:</u>

A = green if: $T_{i,\,Max\,(alternative\,Peltier)} - T_{i,\,Max\,(current\,Peltier)} \leq 0$

A = yellow if: $T_{i,\,Max\,(alternative\,Peltier)} - T_{i,\,Max\,(current\,Peltier)} \leq C$

A = red if: $T_{i,\,Max\,(alternative\,Peltier)} - T_{i,\,Max\,(current\,Peltier)} > 0.XX°C$

<u>Evaluating minimum temperature:</u>

B = green if: $T_{i,\,Min\,(alternative\,Peltier)} - T_{i,\,Min\,(current\,Peltier)} \geq 0$

B = yellow if: $T_{i,\,Min\,(alternative\,Peltier)} - T_{i,\,Min\,(current\,Peltier)} \geq -0.XX°C$

B = red if: $T_{i,\,Min\,(alternative\,Peltier)} - T_{i,\,Min\,(current\,Peltier)} < -0.XX°C$

<u>Evaluating the Mean maximum of heating and cooling ramp:</u>

C = green if: $R_{Max\,(alternative\,Peltier)} - R_{Max\,(current\,Peltier)} \leq 0$

C = yellow if: $R_{Max\,(alternative\,Peltier)} - R_{Max\,(current\,Peltier)} \leq 0.XX°C/s$

C = red if: $R_{Max\,(alternative\,Peltier)} - R_{Max\,(current\,Peltier)} > 0.XX°C/s$

<u>Evaluating the Mean minimum of heating and cooling ramp:</u>

D = green if: $R_{Min\,(alternative\,Peltier)} - R_{Min\,(current\,Peltier)} \geq 0$

D = yellow if: $R_{Min\,(alternative\,Peltier)} - R_{Min\,(current\,Peltier)} \geq -0.XX°C/s$

D = red if: $R_{Min\,(alternative\,Peltier)} - R_{Min\,(current\,Peltier)} < -0.XX°C/s$

IV. Study Results:

Of the alternative Peltier elements evaluated, only one met the acceptance criteria. It demonstrated good performance, with all temperature and ramp reference points in the required temperature and ramp tolerance values.

(B) Please provide information and data to establish reliability of possible substitutes of application and of RoHS materials in application

<u>Endurance Testing of Alternative Peltier Elements for the CTM/CTM48 Instruments</u>

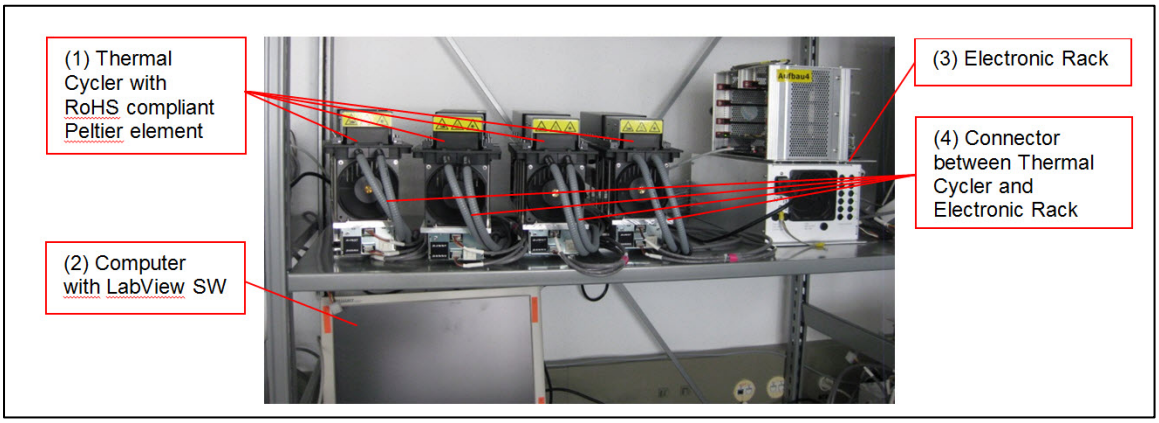
I. Background:

The endurance testing study was conducted on eight examples of the single alternative Peltier element to pass the temperature performance feasibility study. The study was designed to repeatedly simulate the PCR process to determine how many heating and cooling cycles the alternative Peltier elements could withstand before they failed.

II. Study Design:

A testing rig was setup as shown below in Figure 3, consisting of an array of CTM Thermal Cyclers (1) assembled with the alternative Peltier element; a computer workstation (2) running LabView Endurance Software to manage and track the heating and cooling cycles; a COBAS TaqMan Electronics Rack (3), and the connectors between the Thermal Cyclers and the Electronics Rack (4).

Figure 3: Peltier Element Endurance Testing Setup



III. Acceptance Criteria:

All eight of the Peltier elements must meet the specification for acceptable reliability.

IV. Results:

The results of the endurance testing are summarized below.

Table 1: Endurance Testing Results for Alternative Peltier Element

Peltier #	% Specification Achieved
Element 1	79%
Element 2	87%
Element 3	49%
Element 4	99%
Element 5	>100%
Element 6	>100%
Element 7	33%
Element 8	62%

V. Summary and Conclusion:

The endurance study conducted with the RoHS compliant Peltier element failed because 6 of the 8 Peltier elements tested did not meet the reliability specification. Moreover, the data show that the element reliability varied significantly from one another, further supporting the conclusion that this Peltier element is not a suitable replacement for the existing element in the CTM Thermal Cycler.

7. Proposed actions to develop possible substitutes

(A) Please provide information if actions have been taken to develop further possible alternatives for the application or alternatives for RoHS substances in the application.

Roche has other PCR analysis platforms available for sale in the EU market (the cobas 4800 System and the cobas 6800/8800 Systems), but they are not suitable alternatives to the CAP/CTM platform. Refer to Section 9 of this application for greater detail on the cobas 4800, 6800, and 8800 Systems and rationale as to why they are not suitable alternatives.

Roche will continue research into possible use of alternative RoHS-compliant Peltier elements with the CAP/CTM Platform.

(B) Please elaborate what stages are necessary for establishment of possible substitute and respective timeframe needed for completion of such stages.

The remaining technical issue is the reliability of the lead-free soldered Peltier, and the exemption is needed until this can be resolved. At present, the cause of failure has not been identified and so it is not possible to estimate how long it will take to resolve this issue.

8. Justification according to Article 5(1)(a):

(A) Links to REACH: (substance + substitute)

1) Do any of the following provisions apply to the application described under (A) and (C)? **NO. Lead in solder is not encompassed by the REACH regulation.**

☐ Authorisation

☐ SVHC

☐ Candidate list

☐ Proposal inclusion Annex XIV

☐ Annex XIV

☐ Restriction

☐ Annex XVII

☐ Registration of intrusions

☐ Registration

2) Provide REACH-relevant information received through the supply chain.

Name of document: _____

(B) Elimination/substitution:

1) Can the substance named under 0(A)1 be eliminated?

☐ Yes. Consequences? _____

☒ **No.** Justification? **See Section 6.**

2) Can the substance named under 0(A)1 be substituted?

☐ Yes.

☐ Design changes: _____

☐ Other materials: _____

☐ Other substance: _____

☒ **No.**

Justification? **See Section 6.**

3) Give details on the reliability of substitutes (technical data + information): **Not applicable; see Section 6.**

4) Describe environmental assessment of substance from 0(A)1 and possible substitutes with regard to

1. Environmental impacts: **Not applicable.**
2. Health impacts: **Patients health would be adversely affected, as described above, by unexpected early failure of analysers or if analysers provide inaccurate results.**
3. Consumer safety impacts: **Not applicable.**

ð Do impacts of substitution outweigh benefits thereof? **Not applicable.**

Please provide third-party verified assessment on this: **Not applicable.**

(C) Availability of substitutes:

1) Describe supply sources for substitutes: **Not applicable; no suitable substitutes are available.**

2) Have you encountered problems with the availability? Describe: **Not applicable.**

3) Do you consider the price of the substitute to be a problem for the availability?

☐ Yes ☒ **No, but none that are suitable have yet been developed**

4) What conditions need to be fulfilled to ensure the availability? **Not applicable.**

(D) Socio-economic impact of substitution: **Not applicable; no suitable substitutes are available.**

ð What kind of economic effects do you consider related to substitution?

☐ Increase in direct production costs

☐ Increase in fixed costs

☐ Increase in overhead

☐ Possible social impacts within the EU

☐ Possible social impacts external to the EU

☐ Other: _____

ð Provide sufficient evidence (third-party verified) to support your statement:

9. Other relevant information

Please provide additional relevant information to further establish the necessity of your request:

Medical and Scientific Justification in Support of Exemption

I. Background

Roche's blood screening and virology products using the COBAS TaqMan Analyzer (CTM) detect viral DNA or RNA from numerous viral targets to screen blood and blood products for infectious agents and to diagnose and to manage patients' infection. The underlying technology in these products includes reverse transcription (RT) and polymerase chain reaction (PCR) with real-time fluorescence detection (see Exhibit 1 for greater detail on PCR). This analysis technique is used to diagnose patients for diseases, and to monitor if a patient is responding adequately to his/her treatment; it furthermore is used for screening blood before it can be used for transfusions, and for screening organs before they are transplanted (more details below).

A timely, correct result depends on the thermal cyclers in the CTM functioning properly; any malfunction or failure of the thermal cyclers can lead to incorrect or delayed results, which, in turn, can have devastating medical consequences.

II. Criticality of Temperature Control during PCR

PCR depends on variation of temperature in a controlled and stable manner. Not only is precise temperature control critical to this technology, but a high level of reliability is required as numerous repetitive temperature cycles occur during use of the instruments. When used in a blood screening or diagnostic laboratory, these instruments are expected to achieve the correct temperature for each process step with high accuracy and precision.

Two critical design specifications for the Peltier are precise temperature control and reliability.

Temperature Control

The temperature control provided by the Peltier is fundamental to processes integral to successful PCR. Incorrect temperature during any phase of the reaction can have significant detrimental effects. For example, temperature directly affects primer and probe hybridization (binding of the primers and probes onto the viral nucleic acid), DNA polymerase activity (which impacts the quality of the PCR reaction and resulting amplified nucleic acid), and various processes related to fluorescence and detection. An example of a secondary effect of temperature control is change in pH, which together with temperature also affects primer and probe hybridization and DNA polymerase activity, as well as other critical processes in PCR.

A description of temperature-dependent processes critical to PCR is presented below in Table 2, together with the potential effects of Peltier malfunction on each process, the assay result, and most importantly, patient safety. Greater detail on the PCR process, and definitions for the terms used below, can be found in Exhibit 1 to this application.

Table 2: Temperature-dependent Processes During PCR and Effects of Peltier Malfunction

Temperature-Dependent Process	Potential Effect on PCR	Potential Effect on Assay Result	Effect on Patient Safety
Primer and probe hybridization	Decreased: · Sensitivity · Inclusivity	· False negative result · Misquantification · Invalid result	· Infection of transfusion or organ recipient · Failure to notify or defer donor · Improper diagnosis or treatment · Delay to treatment

Temperature-Dependent Process	Potential Effect on PCR	Potential Effect on Assay Result	Effect on Patient Safety
	Decreased: · Specificity	· False positive result · Misquantification · Invalid result	· Infection of transfusion or organ recipient · Incorrect donor notification and deferral · Improper diagnosis or treatment · Delay to treatment
Extension rate, processivity, and fidelity of DNA polymerase	Decreased: · Sensitivity · Inclusivity	· False negative result · Misquantification · Invalid result	· Infection of transfusion or organ recipient · Failure to notify or defer donor · Improper diagnosis or treatment · Delay to treatment
	Decreased: · Specificity	· False positive result · Misquantification · Invalid result	· Infection of transfusion or organ recipient · Incorrect donor notification and deferral · Improper diagnosis or treatment · Delay to treatment
Fluorescence signal	· Change in signal-to-noise ratio · Uncompensated error in signal versus hard-coded cutoffs	· False negative result · False positive result · Misquantification · Invalid result	· Infection of transfusion or organ recipient · Incorrect donor notification and deferral · Improper diagnosis or treatment · Delay to treatment
Probe quenching	· Change in signal-to-noise ratio · Uncompensated error in signal versus hard-coded cutoffs	· False negative result · False positive result · Misquantification · Invalid result	· Infection of transfusion or organ recipient · Incorrect donor notification and deferral · Improper diagnosis or treatment · Delay to treatment
Structure of target and amplification product (amplicon)	Decreased: · Sensitivity · Inclusivity	· False negative result · Misquantification · Invalid result	· Infection of transfusion or organ recipient · Failure to notify or defer donor · Improper diagnosis or treatment · Delay to treatment

Temperature-Dependent Process	Potential Effect on PCR	Potential Effect on Assay Result	Effect on Patient Safety
	Decreased: · Specificity	· False positive result · Misquantification · Invalid result	· Infection of transfusion or organ recipient · Incorrect donor notification and deferral · Improper diagnosis or treatment · Delay to treatment
pH	· Primer and probe hybridization · DNA polymerase rate, processivity, and fidelity · Buffering of divalent metal ion concentrations	· False negative result · False positive result · Misquantification · Invalid result	· Infection of transfusion or organ recipient · Failure to notify or defer donor · Improper diagnosis or treatment · Delay to treatment

Reliability

Similar risks to patient safety exist for Peltiers that fail to demonstrate sufficient reliability. If the Peltier element is not functioning, then it is not possible to obtain test results. Instrument unavailability as a result of a non-functioning Peltier increases the risk that a time-sensitive result could not be obtained in time for a physician to take potentially life-saving action.[1-4]

For example, before an organ is transplanted, blood screening tests are used to test the blood of an organ donor to make sure that the organ (e.g., heart, lungs, or liver) to be transplanted is not infected with HIV, HCV, HBV, and West Nile virus. Transplanting an infected organ or tissue into an immunocompromised person (as transplant recipients are) could lead to the recipient's serious illness or death.[5,6] Organ transplantation is time sensitive; physicians have only a few hours after an organ becomes available to transplant that organ or the organ will not be viable for transplant.[4] Heart or lungs, for instance, must be transplanted within 6 hours of removal from the donor.[4] The result of screening tests for infectious disease must be complete before the transplant can occur. If those results are not available in a timely fashion, the organ may not be transplanted, and the intended recipient may face adverse health consequences, including death, if the recipient cannot receive a transplant of the organ intended to save that person's life. The failure of a Peltier element during blood screening in this setting could result in the loss of the organ for transplant and result in dire consequences for the intended recipient.

Transfusion of blood or blood products could be similarly, adversely affected if a Peltier failed and screening results were delayed. Not all units of blood or blood products are acceptable for transfusion into all patients. In particular, persons who depend on blood transfusions, such as persons undergoing chemotherapy or with hematologic disorders, often develop antibodies that make finding compatible units of blood or blood products quite difficult.[7-11] In some instances, one particular donor may be the only compatible source of blood or platelets for a particular critically-ill patient. If a unit of blood or platelets is collected from such a single compatible source for a bleeding, anemic, or thrombocytopenic patient, then that patient is vulnerable to a delay in screening results. If a Peltier fails and delays a screening result, that rare unit may not be available when the critically-ill person waiting for it needs it. The intended recipient could face serious adverse health consequences or die because the blood or platelet because of Peltier failure.

Peltier failure could create unacceptable medical risk in the virology diagnostic laboratory, as well. A false negative result carries a severe hazard to infants who undergo NAT testing for HIV diagnosis. Infants at risk for HIV infection would undergo repeat testing because of the difficulty of diagnosing HIV in this population.[1] Infants typically undergo repeat testing after birth, at age 14-21 days, at 1-2 months, and at 4-6 months, so, while a false negative result could be detected at subsequent time points, a delay in treatment of several weeks to months could

occur. Guidelines recommend that infants under 12 months of age diagnosed with HIV initiate combination antiretroviral treatment upon diagnosis. Children under 12 months of age experience rapid immunological and clinical deterioration including death, which makes critical the timeliness of accurate results. Among infants within the lowest CD4 count percentile category, the death rate is 5% per month.

In adults, a patient with a false negative test could be lost to follow up, which would permit irreversible immune system damage and severe or fatal infectious complications to occur.[2,3] Delays in diagnosis could lead to delay in treatment of acute infections would allow the size and diversity of the viral reservoir to expand, compromising a patient's chance of achieving a functional cure. Further, a risk to public health exists in that the patient could unknowingly transmit HIV to their sexual or drug use partners.

Underquantification of viral DNA on an HIV viral load test could result in an HIV-infected patient with a low CD4+ cell counts and failing antiretroviral treatment progressing to AIDS-defining illness(es) or even death because of a falsely underquantified HIV viral load measurement.[2,3] The risk of HIV drug resistance would also be elevated.

III. Clinical Advantages of the CAP/CTM Platform vs. Alternative PCR Platforms

The COBAS AmpliPrep / COBAS TaqMan (CAP/CTM) PCR analysis platform, which is marketed as the cobas s 201 System for use with donor screening applications, provides clinicians with certain advantages due to the unique combination of instrumentation features, performance, and the portfolio of assays that can be run on the platform, when compared to alternative PCR platforms that have been CE marked by their manufacturers under the In Vitro Diagnostics Directive (IVDD) 98/79/EC and are available on the EU market.

It is important to note that Roche does not know which solders are used by the manufacturers of these alternative PCR analyzer platforms; some or all may contain lead or other substances that exceed RoHS limits.

Table 3 below summarizes several differentiating features which are key to PCR analysis, and presents a comparison of these features on the CAP/CTM versus alternative PCR analysis platforms.

Table 3: Alternatives to Roche CAP/CTM / Roche cobas s 201 PCR Analysis Platforms in the EU Market

Feature / Parameter	**Roche CAP/CTM (cobas s 201)**	**Abbott m2000[1]**	**Hologic Panther[2]**	**Siemens kPCR[3]**	**Beckman Veris[4]**	**Cepheid[5]**	**Qiagen[6]**	**Roche cobas 4800**	**Roche cobas 6800/8800**
Sample Throughput (8 hours)	144 (CAP/CTM) 72 (cobas s 201)	96	275	96	150 for DNA 100 for RNA	Will depend on the size of the module	*	Varies by assay	384 / 960
Interleaving (Processing of multiple assays simultaneously)	Yes (up to 3)	Yes (HIV & HCV only)	Mixed batching of up to 4 assays	No	Each sample is processed individually. Up to 20 assays can be run at one given time.	Each sample is processed individually. Number of assays depends on the size of the module.	Yes (up to 2)	Mixed batching of up to 3 assays within the microbiology portfolio only (MRSA, C. diff & HSV 1/2)	Mixed batching of up to 3 assays
Automated transfer between sample preparation and amplification / detection processes	Yes (via Docking Station; CTM 96 only)	No	Yes – Sample preparation and amplification / detection processes integrated in single instrument	No	Yes – Sample preparation and amplification / detection processes integrated in single instrument	Yes – Sample preparation and amplification / detection processes integrated in single instrument	No	No	Yes – Sample preparation and amplification / detection processes integrated in single instrument
Hands-on Time (minutes)	10 (96 tests)	65 (96 tests)	37	35 (48 tests)	15 (48 tests)	2 (per test)	*	30 (96 tests)	30 (384 tests)
Daily Maintenance (minutes)	30	10	14	*	30	0	*	< 10 minutes	0
Ready-to-Use Reagents	Yes	No	No	No	Yes	Yes	Yes	Yes, but they need to be manually transferred to reservoirs	Yes
Contamination control	Yes (UNG)	Yes (UNG; manual addition)	Yes (Bleach)	Yes (UNG)	Yes (UNG)	No	No	Yes (UNG)	Yes (UNG)
Assay calibration required by user	No	Yes	Yes	Yes	Yes	No	No	No	No
Reagent Storage	Refrigerator	Freezer	Freezer	Refrigerator	Refrigerator	Refrigerator	Freezer	Refrigerator	Refrigerator
Required Floor Plan	1 single room	2 separated areas are recommended	1 single room (integrated system)	1 single room	1 single room (integrated system)	1 single room (integrated system)	1 single room	1 single room	1 single room
IVD Assays (Quantitative)	· HIV · HBV · HCV · CMV	· HIV · HBV · HCV · CMV · EBV · BKV	· HIV	· HIV · HBV · HCV	· HBV · CMV · HIV · HCV	· HIV · HCV	· HIV · HBV · HCV	N / A	· HIV · HBV · HCV · CMV

Feature / Parameter	Roche CAP/CTM (cobas s 201)	Abbott m2000[1]	Hologic Panther[2]	Siemens kPCR[3]	Beckman Veris[4]	Cepheid[5]	Qiagen[6]	Roche cobas 4800	Roche cobas 6800/8800
IVD Assays (Qualitative)	· HIV · HCV · HLA*B5701 · Mtb†	· HIV · HCV Genotyping · CT/NG · HPV · C. diff · HSV · Mtb/MAI · Influenza A/B · RSV	· HPV · CT/NG · Progensa PCA3	· HCV Genotyping · kPCR PLX (CMV, EBV, HSV, VZV, HHV-6, BKV, JCV, Adenovirus, Parvovirus B19) · CT/NG	N / A	· HIV · Mtb · Flu/RSV · HPV · CT/NG · Group B Strep · Trichomonas · MRSA/SA · C.diff · VRE · Norovirus · Enterovirus · Carba-R	· CMV · BKV · EBV · VZV · HSV · CT/NG · C.diff · MRSA · VanR · Group B Strep · EGFR (plasma)	· CT/NG · HPV · C. diff · MRSA · HSV 1/2	N / A
Donor Screening Assays	· MPX (Qualitative HIV/HBV/HCV) · DPX (Quantitative Parvo B19, Qualitative HAV) · WNV (Qualitative)	N / A	· Ultrio Elite (Qualitative HIV/HBV/HCV) · WNV · Parvo B19 · HAV · HEV	N / A	N / A	N / A	N / A	N / A	· MPX (Qualitative HIV/HBV/HCV) · DPX (Quantitative Parvo B19, Qualitative HAV) · WNV (Qualitative) · HEV

[1] Abbott Global website; MaxCycle Brochure; m2000 rt Brochure; Abbott RealTime HIV-1, HCV & HBV Package Inserts; CAP-Today; Argent Blood Screening Analysis 2008

[2] Novartis Global Website; Procleix® Panther® System Brochure; Argent Blood Screening Analysis 2008; CAP-Today

[3] Siemens Website

[4] Beckman European Customer Presentation; GenomeWeb

[5] Cepheid Website; Argent Microbiology Analysis 2013; GeneXpert Brochure

[6] Qiagen website

[*] Information not available

[*] Information not available

[†] COBAS TaqMan 48 only – Manual sample preparation

When looking individually at key features such as throughput, the level of automation between sample preparation and amplification/detection, and the ability to run multiple tests in parallel, the CAP/CTM platform in many cases provides an outright technical advantage over alternative solutions; when taken together in combination, there is no single alternative PCR analysis platform that offers the flexibility and technical advantages afforded by the CAP/CTM platform.

The portfolio of CAP/CTM and cobas s 201 assays also differentiates the Roche platform from the alternatives. None of the alternative platforms provide as extensive a menu of assays, combined with the ability to perform both IVD and donor screening assays.

Finally, CAP/CTM reagent technology highlights the following differences versus the alternatives:

- CAP/CTM and cobas s 201 assay reagents can be stored in the refrigerator and do not require additional preparation such as reconstitution or centrifugation. Several of the alternative platforms require the reagents to be reconstituted, frozen, or both.
- CAP/CTM and cobas s 201 assay reagents include UNG to prevent cross contamination, which is critical to the integrity of PCR; assays on some alternative platforms require manual addition of UNG, the use of bleach, or do not provide methods to prevent cross contamination.
- CAP/CTM and cobas s 201 assays do not require the user to perform any calibration; while not a unique feature, the alternative platforms that also do not require assay calibration are only intended for IVD testing and not donor screening.

It is important to note that Roche has developed a next-generation PCR analysis platform, the **cobas**® 6800/8800 Systems, which are CE marked under the IVD Directive 98/79/CE and were launched for sale in the EU market in September 2014. The platform consists of two separate instruments, the **cobas**® 6800 System and the **cobas**® 8800, both of which offer fully-integrated sample preparation, PCR amplification, and target detection in a single instrument. Both the cobas 6800 and 8800 Systems are fully RoHS-compliant.

Roche also offers for sale in the EU another IVD CE-marked PCR analysis platform, the **cobas**® 4800 System, which provides sample preparation and PCR amplification / target detection in two separate instruments. The cobas 4800 System also is RoHS-compliant. However, as can be seen in Table 15, neither of these platforms are suitable alternatives to the CAP/CTM platform.

- While the cobas 6800/8800 Systems offer a similar assay portfolio as CAP/CTM, they are much larger instruments that are intended for use in significantly higher-throughput laboratories. Laboratories that currently use the CAP/CTM would not use this system.
- As for the cobas 4800 System, while similar in functionality and throughput to the CAP/CTM platform, the assay portfolio is completely different; there are no quantitative IVD assays, in particular for HIV/HBV/HCV, currently offered for use on the cobas 4800 System, nor are there any donor screening assays.

In summary, the unique combination of features offered by the CAP/CTM and cobas s 201 PCR analysis platforms provide optimum diagnostic performance in lower-throughput IVD (diagnostic / viral load monitoring) and donor screening applications and there are no alternatives on the market that provide the same combination of functions, analyses and performance.

IV. Conclusion

The alternative Peltier elements evaluated by Roche do not meet design specifications for temperature control or reliability, or both.

- Incorrect temperatures applied during PCR could have significant detrimental effects. For qualitative tests, this could lead to invalid, false negative, or false positive results; for quantitative tests, this could additionally cause underquantification or overquantification. Such errors in patient results can lead to significant disability, permanent harm, or even death in some cases, due to incorrect or delayed diagnoses and/or improper clinical management.[1-4]
- Unexpected instrument downtime as a result of poor reliability of the Peltier element increases the risk that a time-sensitive result cannot be obtained in time for a physician to take potentially life-saving action. [1-4]

Furthermore, analysis of alternative PCR analyzers highlights the unique combination of features which provide optimum diagnostic performance in diagnostic / viral load monitoring and donor screening applications.

In conclusion, the CTM Analyzers with their existing Peltiers must remain available because a suitable replacement Peltier for the CTM is not available, and because alternative PCR analyzers do not provide equivalent medical value. Any restriction on the availability of these instruments will thus introduce unacceptable levels of medical risk.

IV. References

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10. Information that should be regarded as proprietary

Please state clearly whether any of the above information should be regarded to as proprietary information. If so, please provide verifiable justification:

Roche has provided this redacted copy of the Exemption Request Form for distribution to the public, in which information deemed proprietary by Roche has been removed.

ROCHE CTM EXEMPTION REQUEST – EXHIBIT 1

1. Overview of the COBAS AmpliPrep / COBAS TaqMan (CAP/CTM) Platform

1.1. COBAS TaqMan Analyzer

The COBAS TaqMan (CTM) Analyzer is a flexible, automated bench top batch analyzer. It performs real-time (kinetic) PCR amplification and detection using fluorescent multicolor nucleic acid probes. It includes four thermal cyclers.

Figure 1: COBAS TaqMan Analyzer

1.2. COBAS TaqMan 48 Analyzer

The COBAS TaqMan 48 (CTM 48) analyzer also is an automated bench-top analyzer for performing real-time PCR amplification and detection using fluorescent multicolor nucleic acid probes. It is a smaller instrument than the CTM, and only contains two thermal cyclers versus four in the CTM.

Figure 2: COBAS TaqMan 48 Analyzer

1.3. COBAS AmpliPrep Instrument

The COBAS AmpliPrep (CAP) Instrument automates sample preparation (sample pipetting, incubation and washing) for qualitative or quantitative nucleic acid testing using PCR technology. Serum and/or plasma specimens and method-defined control samples may be used, depending upon the pathogen that is being analyzed and the kit being used. The CAP can be docked to a CTM analyzer via a Docking Station for to support an automated workflow between sample preparation on the CAP, and amplification and detection on the CTM.

Figure 3: COBAS AmpliPrep Instrument

Figure 4: COBAS AmpliPrep Instrument Docked with COBAS TaqMan Analyzer

1.4. COBAS TaqMan Thermal Cyclers

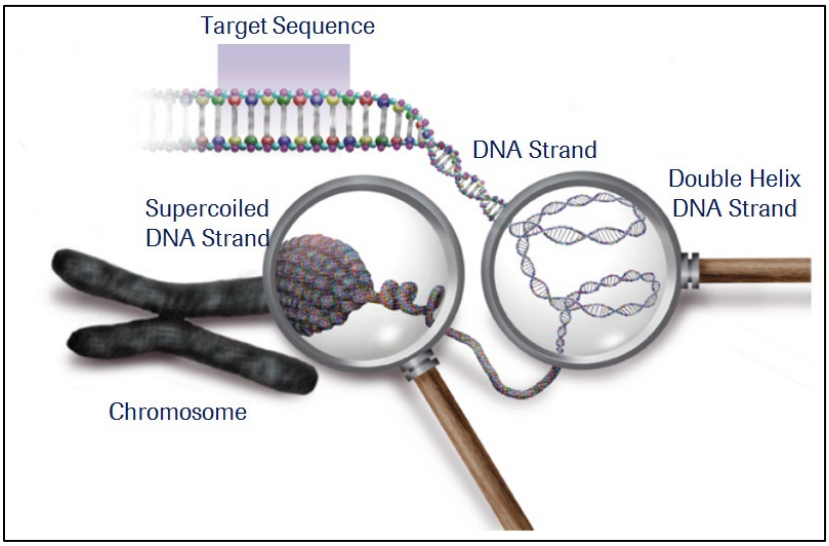
The CTM contains four, and the CTM48 two, simultaneous and centrally controlled thermal cyclers necessary for the PCR amplification steps to take place. The thermal cycler also collects fluorescent data via fiber optic filaments into the photometer. The design of the thermal cycler incorporates the following components:

- A single power stage
- Two double temperature transducers
- Heat sink with fan
- Cover heater lid with an automatic closing mechanism
- Peltier heating/cooling device

2. Principles of Polymerase Chain Reaction (PCR)

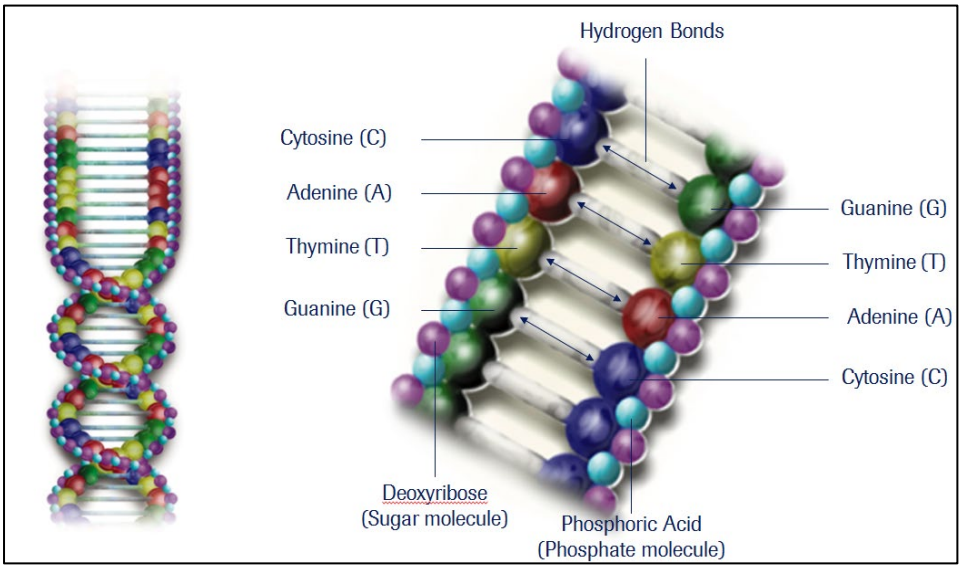
PCR amplifies a targeted sequence of nucleic acid (DNA or RNA). Such targeted sequence is found in the genetic material of the viruses or pathogens (e.g. HIV, HBV) that are to be detected in blood samples during the analysis. The targeted sequence is thus a specific sequence of genetic material of viruses which allows their detection and identification. Through the use of customized nucleic acid primer and fluorescent-labeled probe sets, the COBAS TaqMan methodology can be applied to all viral, bacterial and parasitic pathogens. Figure 5 below depicts with increasing granularity the structure of a chromosome, the double helix of DNA, an individual strand of DNA, and finally, a target sequence of nucleotides.

Figure 5: Chromosome and DNA



The DNA is made of two strands of nucleotides which are bound together by hydrogen bonds, as depicted in Figure 6:

Figure 6: DNA and Nucleotides



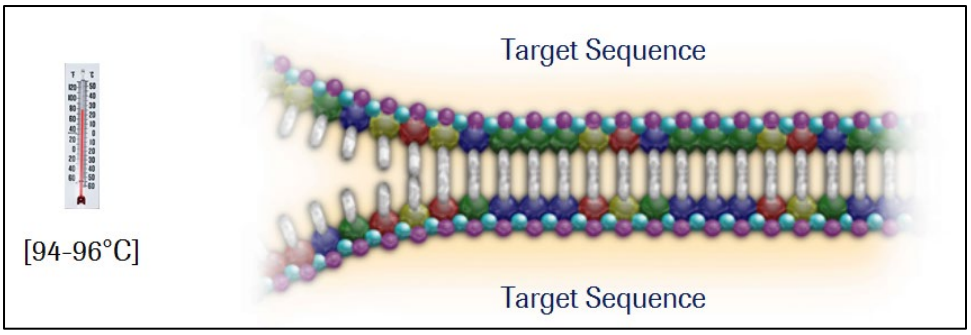
PCR involves multiple rounds, or cycles, of copying a specific sequence of nucleotides so that small amounts of genetic material from viruses or pathogens which may be present in a patient sample can be multiplied exponentially; this is called amplification, and the resulting sequence of nucleotides is called the amplicon.

During the process of amplification, fluorescent particles, or dyes, are attached to each amplicon. These particles can then be detected via excitation with specific wavelengths of light. The amount of fluorescence detected after a certain number of amplification cycles will correspond to the amount of genetic material originally present in the sample.

PCR can be explained in four main processes: denaturation, annealing, synthesis, and amplification.

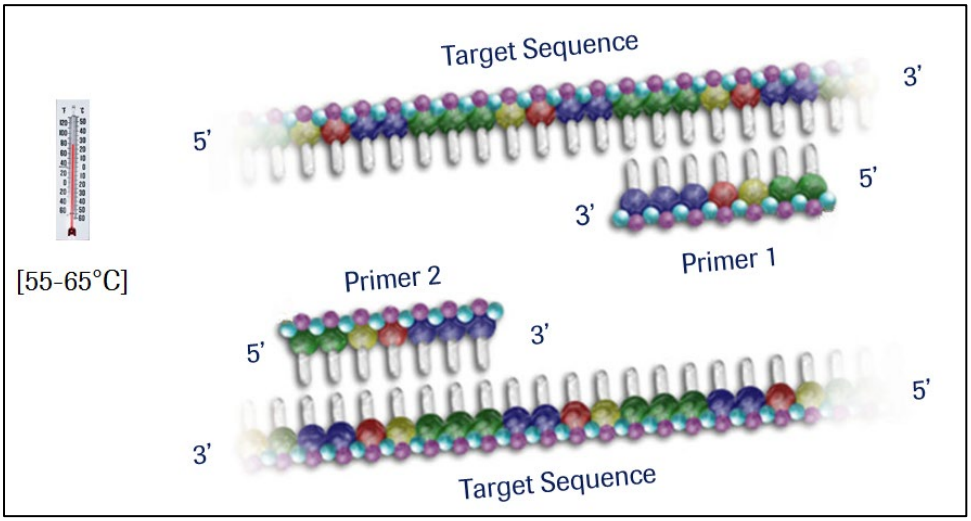
PCR Step 1 – Denaturation: As depicted in Figure 7, the first step of PCR consists of breaking the hydrogen bonds that bind complementary nucleotide strands with heat in order to separate the two DNA strands.

Figure 7: Denaturation



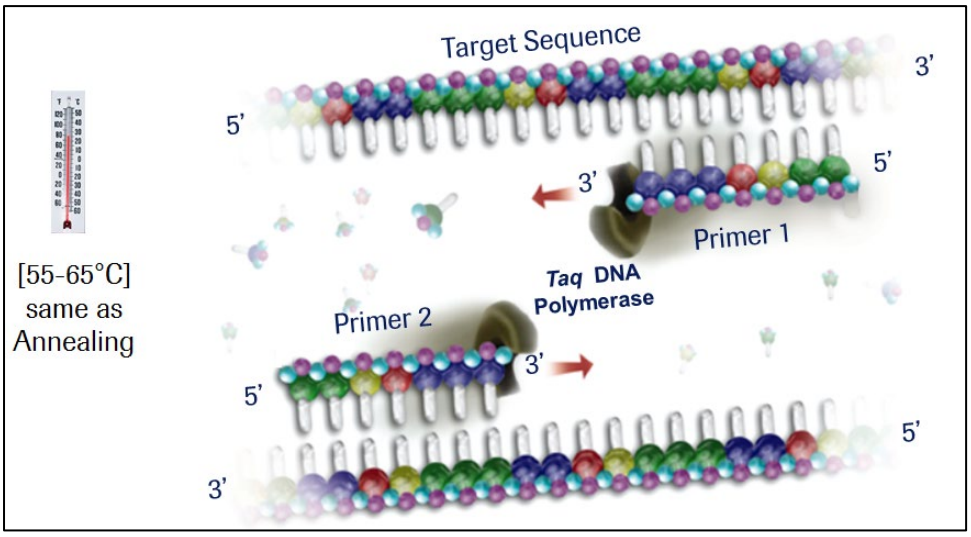
PCR Step 2 – Annealing: During this step, primers are annealed (attached in the double-stranded form) at both ends of the target sequence. This process happens at a temperature of 55°C to 65°C.

Figure 8: Annealing



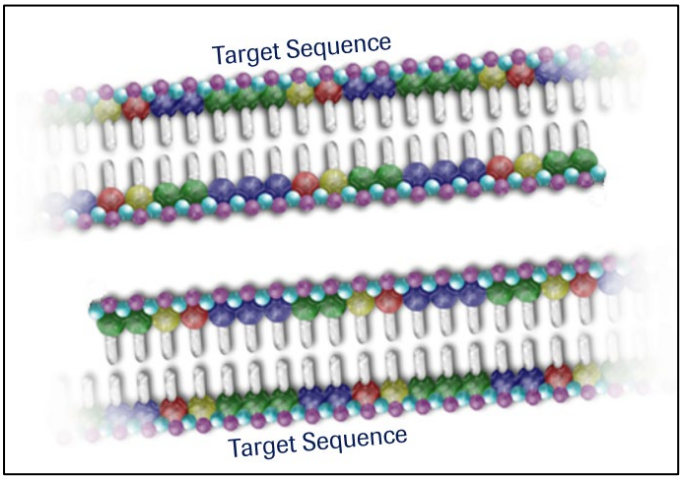
PCR Step 3 – DNA Synthesis: At the same temperature as the annealing, the Taq DNA polymerase, an enzyme already included in the solution (Master Mix), acts as a catalyzer and synthesizes the missing DNA strand with the complementary nucleotides. Figure 9 illustrates this process.

Figure 9: DNA Synthesis



As depicted in Figure 10, at the end of this process, two copies of the target sequence are created.

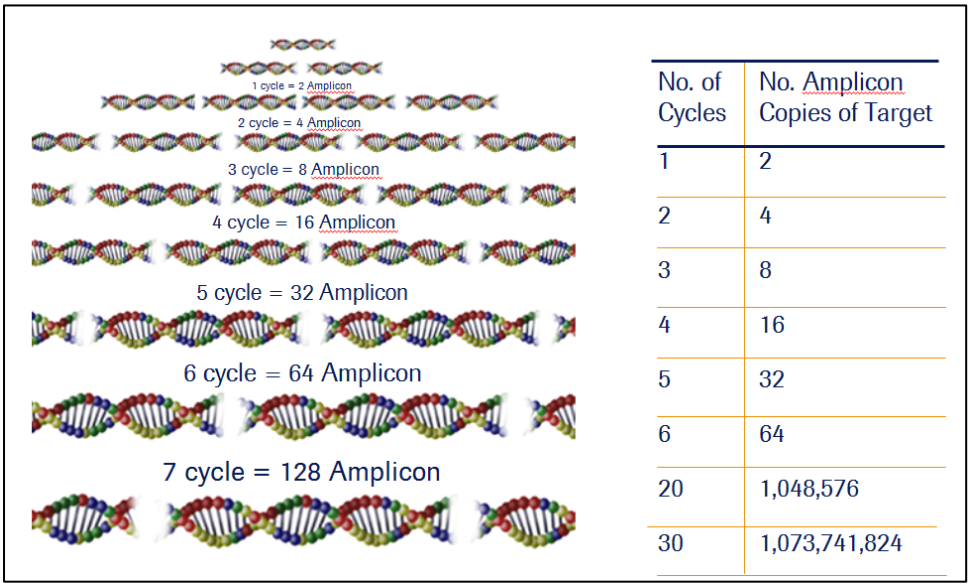
Figure 10: Two Copies of the Target Sequence Created



PCR Step 4 – Target Amplification: Repeating sequentially these steps doubles the number of target sequences during each cycle. This creates an exponential increase of target strands, which is called target amplification. After 30 cycles, more than a billion copies of the target sequence have been created. This amplification dramatically increases the detectability of the target sequence in the sample. As noted above, fluorescent agents are incorporated on target sequence strands. The total fluorescence signal thus increases with the total number of available target sequence strands.

This concept is illustrated in Figure 11.

Figure 11: Target Amplification



No. of Cycles	No. Amplicon Copies of Target
1	2
2	4
3	8
4	16
5	32
6	64
20	1,048,576
30	1,073,741,824

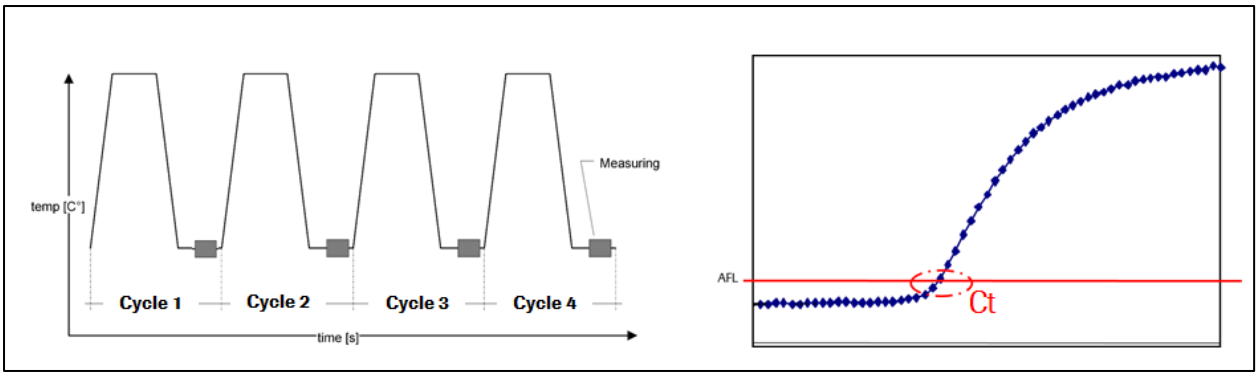
The COBAS TaqMan technology also employs an Internal Control/Quantitation Standard (IC/QS), consisting of non-infectious nucleic acid that is incorporated into each sample at a known copy number. The IC/QS is carried through sample preparation, amplification, and 5' nuclease detection along with the target nucleic acid. The IC/QS corrects for any instrument, chemistry, or sample variances by relating the fluorescence from the target probe to the fluorescence from the IC/QS probe. The IC/QS is also used in quantitative tests to determine the copy number of the unknown target nucleic acid.

3. The COBAS TaqMan Analyzer Detection Process

During PCR, fluorescence data is collected at every cycle. The detection process is dependent on the Critical Threshold value (Ct), which is defined as the cycle number (measured in fractions of cycles) at which point the sample fluorescence level exceeds the Assigned Fluorescence Level (AFL).

The AFL indicates the beginning of an exponential growth phase. The Ct is often referred to as the “Elbow” or crossing point (Cp). Figure 12 illustrates the detection process; specifically, the measurement points and the CT value.

Figure 12: Detection Process



There are three phases of the fluorescence curve:

- Beginning of PCR: Limited amount of template, also limited possibilities for target and primer and polymerase to assemble
- Exponential (and therefore quantifiable) phase of PCR
- End of PCR: Inhibition by accumulation of pyrophosphate; primer and nucleotides are exhausted

4. Glossary

Greater detail is provided below for the terms used in Section 9 of the Application to describe the temperature-dependent processes which are critical to PCR,

Term	Definition
Extension Rate	The number of nucleotides per second which a DNA polymerase adds to the 3’ end of an extending DNA primer.
Fidelity	A measure of how accurately the correct complimentary deoxyribonucleotide is bound to an extending DNA primer, or, the ability to bind to the 3’ terminal end of a DNA primer in order to extend by adding nucleotides.
Fluorescence Signal	The optically detectable accumulation of fluorescent dye, which signals the presence of the target of interest.
pH	pH is the negative log of the hydrogen ion concentration in aqueous solution. pH can impact many PCR processes. The PCR buffer pH changes dynamically with temperature.
Primer and Probe Hybridization	The binding of a specific DNA fragment to its complimentary target, which occurs during every cycle of PCR, and is required for initiation of target replication (amplification) and detection of each replicated copy. Because the concentration of primers and probes in PCR is fixed, hybridization is driven primarily by temperature.
Probe Quenching	Each intact probe molecule is quenched through energy transfer of a fluorescent reporter (dye) to a quencher. The extent of quenching (and therefore baseline signal) is affected by temperature.
Processivity	The number of nucleotides which a DNA polymerase enzyme adds to the 3’ end of an extending DNA primer during the “on” time of the “on/off” binding equilibrium of enzyme to substrate. The equilibrium, and therefore processivity, are impacted by temperature.
Structure of Target and Amplicon	Nucleic acid targets and the amplified copies of these targets are folded in three dimensions (secondary structure) based on their primary sequence and the temperature at which they are held. Incorrect reaction temperatures can cause changes to secondary structure which prevent efficient amplification during PCR.