



**LiliF**

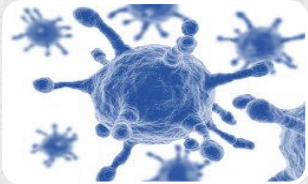
# Detection of Honey Bee Pathogens

Real-time PCR Detection System



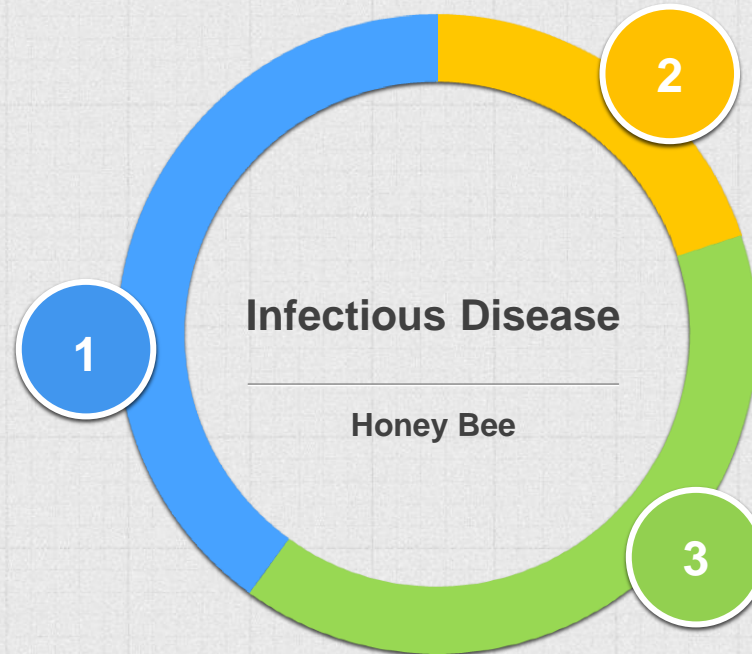
# Infectious Disease of Honey Bee

❖ The widespread decline of the global bee population in recent years, a result of unexplained bee colony collapses and declines, highlights the potential involvement of a variety of fungal, bacterial and viral infections.



## Viral disease

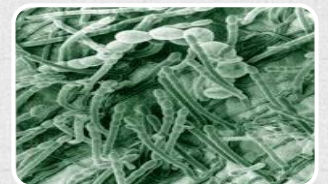
- Virus infects larvae of the honey bee, resulting in failure to pupate and death. Larvae with sacbrood fail to pupate, and ecdysial fluid, rich in SBV, accumulates beneath their unshed skin, forming the sac for which the condition is named.



## Bacterial disease



- Foulbrood (FB), is the most serious bacterial disease of honey bee brood and is caused by the bacterium. The disease is transferred and initiated only by the spore stage of the bacterium. The reason this disease is so serious is that the spores can remain viable and last indefinitely on beekeeping equipment.



## Fungal disease

- Brood is a fungal disease of honey bee brood that infects the gut of the larvae. It is caused by a spore-forming fungus named *Ascosphaera apis* that is consumed along with larval food. Although it can affect workers, drones, or queens it most often occurs in workers and drones.



# Viral, bacterial and fungal honey bee pathogens covered by the Multi-Path

Viral agents	Bacterial agents	Fungal agents
ABPV, Acute bee paralysis virus	AFB, Paenibacillus larvae (causative agent of American foulbrood)	CB, Ascosphaera apis (causative agent of chalkbrood)
BQCV, Black queen cell virus	EFB, Melissococcus plutonius (primary agent of European foulbrood)	Nosema apis (causative agent of nosema disease)
CBPV, Chronic bee paralysis virus	Paenibacillus alvei (associated or secondary agent of European foulbrood)	Nosema ceranae
DWV, Deformed wing virus	Enterococcus faecalis (associated or secondary agent of European foulbrood)	
IAPV, Israeli acute paralysis virus		
KBV, Kashmir bee virus		
SBV, Sacbrood virus		



SBV



DWV



BQCV



ABPV



ABPV



CVPB



IAPV

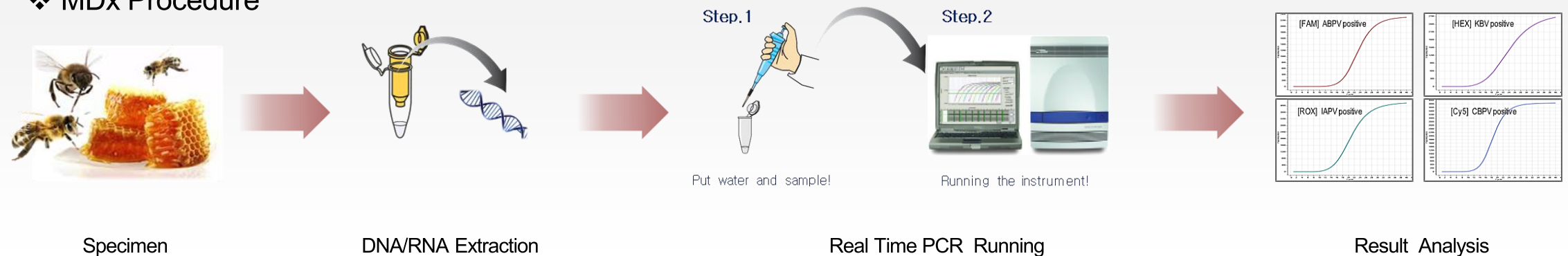
\* ANSES, Dec 2010 , Journal No. 4, Research for reference, J. Carletto



# Process of Molecular Diagnosis

- ✓ The PCR detection kits has advantages over other methods for early detection and monitoring of disease due to its high sensitivity and specificity.

## ❖ MDx Procedure



**Extraction Kits**

Patho Gene-spin™ DNA/RNA Extraction Kit0



**Detection Kits**

LiliF™ ABPV/KBV/IAPV/CBPV Real-time RT-PCR Kit  
LiliF™ SBV/KSBV/DWV/BQCV Real-time RT-PCR Kit

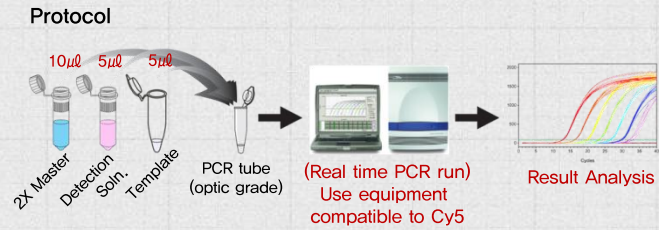


# Viral Disease Detection Kits

## LiliF™ SBV/KSBV/DWV/BQCV Real-time RT-PCR Kit

### Description

- The kit contains a specific primer and probe set for a highly conserved region based on current sequence alignments of all type of SBV, KSBV, DWV and BQCV, allowing the RNA detection. It can determine the infecting all serotype and accurately and sensitivity detect multiple detection genes at one time using the real-time RT-PCR (quantitative) method, and take only 2 hours for detection.



### KIT Contents

Product	Amount	Cat. No.
LiliF™ SBV/KSBV/DWV/BQCV Real-time RT-PCR Kit	50 Test/Kit	IPC12023
Contents		Amount
2X qRT-PCR Master Mix Solution		520 µl
SBV/KSBV/DWV/BQCV Detection Solution		260 µl
Positive control (External PC)		75 µl
DNase/RNase Free Water		1 ml
Related Product	Amount	Cat. No.
Patho Gene-spin™ DNA/RNA Extraction Kit	50 Col./Kit	17154

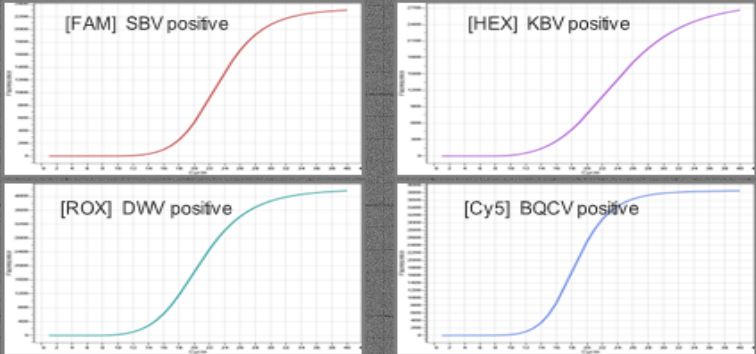


### Step of the PCR reaction & Fluorescent Channel

Step	Cycle	Temp	Time	Channel setting	
Reverse Transcription Step	1	45 °C	30 min.	SBV	FAM
		95 °C	10 min.	KSBV	HEX
PCR Step	40	95 °C	15 sec.	DWV	ROX
		62 °C	1 min.	BQCV	Cy5



### Example Results



- ✓ This product contains a positive control in the product. Therefore, in order for the user to judge whether the performance of this product is working properly, please check whether the positive result and the negative reference solution react with each other and whether the result is normal



# Viral Disease Detection Kits

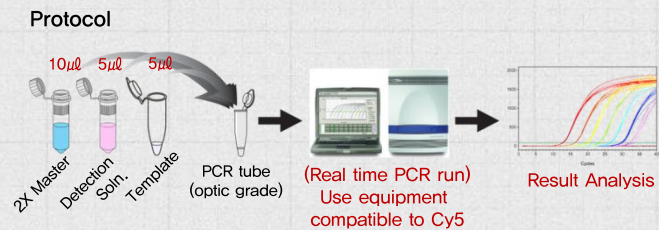
LiliF™

ABPV/KBV/IAPV/CBPV

Real-time RT-PCR Kit

## Description

- The kit contains a specific primer and probe set for a highly conserved region based on current sequence alignments of all type of ABPV, KBV, IAPV and CBPV, allowing the RNA detection. It can determine the infecting all serotype and accurately and sensitivity detect multiple detection genes at one time using the real-time RT-PCR (quantitative) method, and take only 2 hours for detection.



## KIT Contents

Product	Amount	Cat. No.
LiliF™ ABPV/KBV/IAPV/CBPV Real-time RT-PCR Kit	50 Test/Kit	IPC12024
Contents		Amount
2X qRT-PCR Master Mix Solution		520 µl
ABPV/KBV/IAPV/CBPV Detection Solution		260 µl
Positive control (External PC)		75 µl
DNase/RNase Free Water		1 ml
Related Product	Amount	Cat. No.
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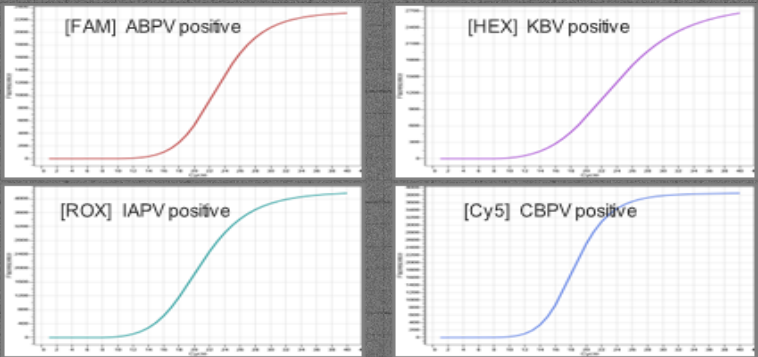


## Step of the PCR reaction & Fluorescent Channel

Step	Cycle	Temp	Time	Channel setting	
Reverse Transcription Step	1	45 °C	30 min.	ABPV	FAM
		95 °C	10 min.	KBV	HEX
PCR Step	40	95 °C	15 sec.	IAPV	ROX
		62 °C	1 min.	CBPV	Cy5



## Example Results



- ✓ This product contains a positive control in the product. Therefore, in order for the user to judge whether the performance of this product is working properly, please check whether the positive result and the negative reference solution react with each other and whether the result is normal



# Viral Disease Detection Kits

## LiliF™ SBV RT-PCR Kit

- **Sacbrood virus(SBV) Detection kit** is able to detect sacbrood virus directly on the basis of a genetic database. So this enables to diagnose it very fast and accurately with reliability. It can amplify only the specific gene using the RT-PCR method, and take only 2~3 hours for detection.

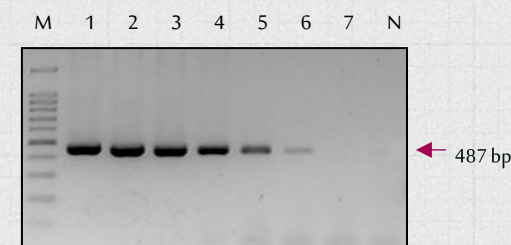


## LiliF™ CSBV RT-PCR Kit

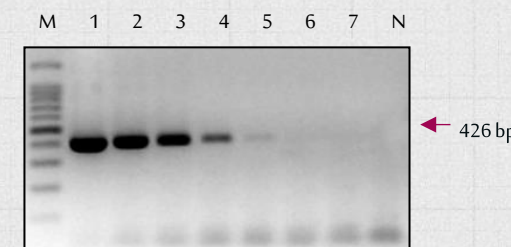
- **Chinese Sacbrood virus(CSBV) Detection kit** is able to detect Chinese sacbrood virus directly on the basis of a genetic database. So this enables to diagnose it very fast and accurately with reliability. It can amplify only the specific gene using the RT-PCR method, and take only 2~3 hours for detection.



### Sacbrood virus(SBV) Detection kit



### Chinese Sacbrood virus(CSBV) Detection kit



Pathogen	PCR type	CAT. No.	PCR type
Virus	RT-PCR	IPC12001	LiliF™ SBV RT-PCR Kit
	RT-PCR	IPC12002	LiliF™ CSBV RT-PCR Kit
	RT-PCR	IPC12012	LiliF™ KSBV RT-PCR Kit



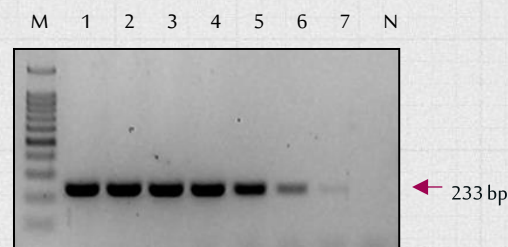
# Bacterial Disease Detection Kits

## LiliF™ AFB PCR Kit

- **American foulbrood (AFB) Detection kit** is able to detect *Paenibacillus* larvae directly on the basis of a genetic database. So this enables to diagnose it very fast and accurately with reliability. It can amplify only the specific gene using the PCR method.



American foulbrood (AFB) Detection kit

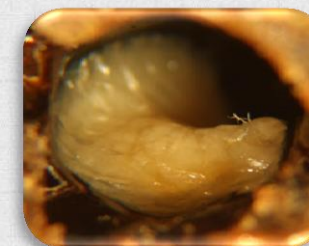
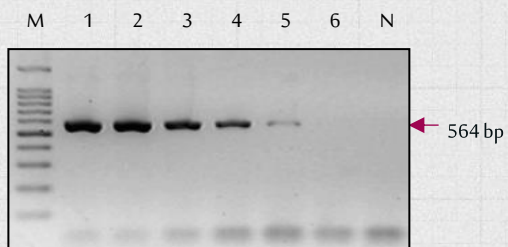


## LiliF™ EFB PCR Kit

- **European foulbrood (EFB) Detection kit** is able to detect bacteria, *Melissococcus plutonius* directly on the basis of a genetic database. So this enables to diagnose it very fast and accurately with reliability. It can amplify only the specific gene using the PCR method.



European foulbrood (EFB) Detection kit



Pathogen	PCR type	CAT. No.	PCR type
Virus	RT-PCR	IPC11087	LiliF™ AFB PCR Kit
	RT-PCR	IPC11084	LiliF™ EFB PCR Kit



# Fungal Disease Detection Kits

## LiliF™ CB PCR Kit



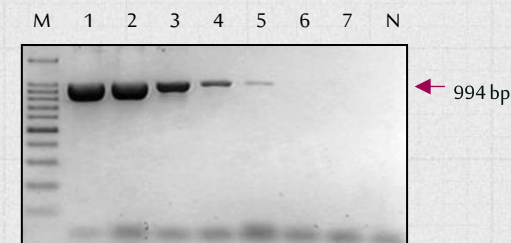
- **Chalkbrood(CB) Detection kit** is able to detect *Ascosphaera apis* directly on the basis of a genetic database. So this enables to diagnose it very fast and accurately with reliability. It can amplify only the specific gene using the PCR method.

## LiliF™ SB PCR Kit

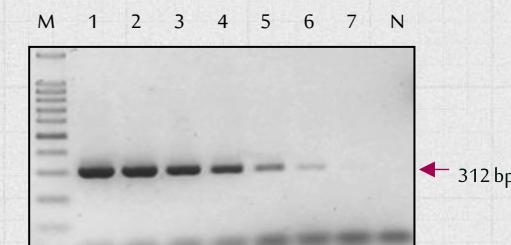


- **Stonebrood(SB) Detection kit** is able to detect *Aspergillus fumigatus*, *Aspergillus flavus*, and *Aspergillus niger* directly on the basis of a genetic database. So this enables to diagnose it very fast and accurately with reliability. It can amplify only the specific gene using the PCR method.

### Chalkbrood(CB) Detection kit



### Stonebrood(SB) Detection kit



Pathogen	PCR type	CAT. No.	PCR type
Virus	RT-PCR	IPC11088	LiliF™ CB PCR Kit
	RT-PCR	IPC11089	LiliF™ SB PCR Kit



# Diagnosis and Prescription of Honey Bee

## Summary

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### **iNtRON Products**

Honey Bees play a vital role in the environment by pollinating both wild flowers and many agricultural crops as they forage for nectar and pollen, in addition to producing honey and beeswax.



### **Customized Service**

Less set up time / Less errors / Less contamination

