

Irena Nazarov
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August 18, 2020

Re: MICROBIOLOGICAL TEST OF SEEDS

Official sample No.46881-2020/2 Certificate import no. 99030-2020

Sample Details:

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Sample:	Pepper seeds Capsicum Annum	Lab ID:	4782	
Variety:	TIRZA	Seeds origin:	PERU	
Lot No.	6017380	Name of official Sampler:	Irena Nazarov	
Weight of sample:	120gr	Seed owner:	Efal Chemicals	
Weight of shipment:		Date sampled: 22.07.2020	Date Received: 22.07.2020	

Tested for:	Results:
Pseudomonas corrugata - 2 replicates of 5000 seeds each	Negative
Clavibacter michiganensis subsp. michiganensis - 2 replicates of 5000 seeds each	Negative
Erwinia carotovora - 2 replicates of 5000 seeds each	Negative
Xanthomonas vesicatoria- 2 replicates of 5000 seeds each	Negative
Tobamoviruses (TMV ToMV, PMMoV, ToBRFV) - 12 replicates of 250 seeds each, by bioassay method.	Negative
Tomato black ring nepovirus - 1 replicate of 250 seeds	Negative
Tomato ringspot nepovirus -1 replicate of 250 seeds	Negative
Pospiviroids (CEVd, CLVd, MPVd, PCFVd, PSTVd, TASVd, TCDVd and TPMVd) -6 replicates of 500 seeds each, by the PCR method.	Negative
Phytophthora capsici - 400 seeds	Negative

Tested for:	Results: Positive*		
Tested for.	CSP	CT28	Menzel
Tomato Brown Rugose Fruit Virus (ToBRFV) - 12 replicates of 250 seeds each then unifying each 4 replicates to make 3 final replicates, by the PCR method.	Ct ₁ =33.10 Ct ₂ =29.20 Ct ₃ =27.80	Ct ₁ =35.90 Ct ₂ =36.50 Ct ₃ =36.80	Ct ₁ =34.10 Ct ₂ =31.50 Ct ₃ =30.40
Negative control	Ct=37.90	Ct=N/A	Ct=38.30
Positive control	Ct=29.50	Ct=30.10	Ct=29.90

Lab ID 4782:

The tests were performed according to:

S.O.P 54: "Testing Tomato and Pepper seeds for Pt, Pc, Xv, Cmm, Er".

S.O.P 56: "Detection of infectious Tobamoviruses in tomato and pepper plants and seeds"

(Tomato: TMV ToMV, ToBRFV) (Pepper: TMV ToMV, PMMoV, ToBRFV)

S.O.P 80: "Detection of Viroid from the Pospiviroid group including Potato Spindle Tuber Viroid (PSTVd) in plants and seeds by RT-PCR or qPCR method".

"REFERENCE PROTOCOL NAKTUINBOUW:Real-time RT-PCR (RT TaqMan PCR) for pospiviroids (CEVd, CLVd, MPVd, PCFVd, PSTVd, TASVd, TCDVd and TPMVd) on seeds of tomato (Solanum lycopersicum)" by Naktuinbouw, 2014

S.O.P 83: "Detection of Fungi in seeds for: Pythium spp., Phytophthora spp., Fusarium spp., Rhizoctonia spp., Verticillium spp., Lasiodiplodia spp., Cylindrocladium spp., Myrothecium spp., Didymella spp., Phoma spp., Colletotrichum spp., Alternaria spp., Stemphylium* spp., Pestalotia spp., Rhizopus spp., Penicillium spp., Aspergillus spp., Diplodia spp., Helminthosporium spp., Drechslera spp., Bipolaris spp., Septoria spp., Cladosporium *spp., Macrophomina sp., Phacidiopycnis spp., Neofabraea spp. Urocystis cepulae, Sclerotium cepivorum, Sclerotinia spp., Botryosphaeria spp., Botrytis spp., Ascochyta spp."

S.O.P 103: "Detection of viruses in Solanacea seeds".

S.O.P 126:" Detection of bacteria, viruses, viroids by the RT-PCR or qPCR method".

Comments:

- *According to regulation requirements as described at ISHI-Veg (March 2020) and at S.O.P 56. RNA fragments of ToBRFV could be found. A $Ct \le 33.50$ considered as positive $Ct=32\pm1.5$ (protocol attached).
- 1. <u>Transportation:</u> The sample was received in the laboratory without any means of protection from temperature changes.
- 2. Treatment: Untreated seeds.
- 3. ISRAC is not responsible for the testing results conducted by the laboratory and the laboratory's accreditation is not considered as an approval of ISRAC or different party related to the assessed.
- 4. This document must be referred to entirely and copying of any part of it to other documents is forbidden
- 5. The test performed is under accreditation to ISO/IEC 17025 standard.
- 6. The data in this certificate reflect results that are valid for analyzed samples only as received in the lab.

Final results are the PPIS representative responsibility. End of certificate.

Sincerely yours,

Head, Microlab Laboratories (99) LTD.