

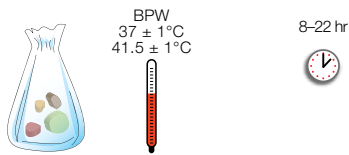


iQ-Check STEC VirX, 3578139

Easy I and Easy II Deep Well Protocols

Using the Thermoshaker

Quick Guide



A. Raw beef, MicroTally swab, fresh spinach (Easy II)

- Enrich the sample in prewarmed Buffered Peptone Water (BPW) (375 g in 1,125 ml or 1 swab in 200 ml) for 8–22 hr at $41.5 \pm 1^\circ\text{C}$

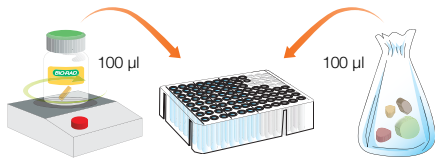
B. Raw milk products (Easy I)

- Enrich the sample in prewarmed BPW + STEC Supplement (25 g in 225 ml) for 16–20 hr at $37 \pm 1^\circ\text{C}$

C. Cannabis flower (Easy I)

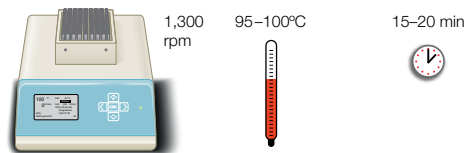
- Enrich the sample in BPW (10 g in 90 ml) for 20–22 hr at $37 \pm 1^\circ\text{C}$

Check with your Bio-Rad representative for specific validations.

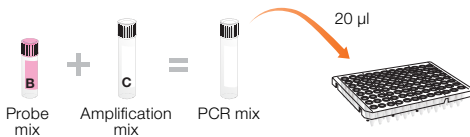


Be sure the lysis reagent is constantly stirring to keep it in suspension.

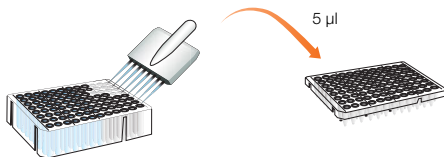
- Add 100 µl of the lysis reagent (Easy I: reagent A or Easy II: reagents A + F) to each well of a deep well plate
- Transfer 100 µl of enriched sample to the deep well plate
Avoid shaking the stomacher bag to prevent collecting large fragments of food debris.
- Seal the deep well plate using the prepierced sealing film



- Incubate at $95\text{--}100^\circ\text{C}$ for 15–20 min at 1,300 rpm in the thermoshaker
- Cool the deep well plate



- Prepare the PCR mix (see PCR Mix Calculation Guide on back)
- Distribute 20 µl/well in a PCR microplate



- Add 5 µl each of positive and negative controls to each well
- Transfer 5 µl/well of extracted DNA from the deep well plate to the microplate
Do not vortex before collecting the sample.
Check there are no bubbles.
- Seal the PCR microplate



- Launch CFX Manager IDE Software
- Create the plate setup
- Start the amplification by clicking **Run**

* Steps automated by iQ-Check Prep System.

For detailed instructions, review the kit user guide.

PCR Mix Calculation Guide

To find the correct volumes to use when preparing the PCR mix, add the total number of samples and controls to be analyzed and find the corresponding volumes of reagent B and reagent C in the table.

Total Number of Samples and Controls	Probes Reagent B, µl	Amplification Mix Reagent C, µl	Total Number of Samples and Controls	Probes Reagent B, µl	Amplification Mix Reagent C, µl	Total Number of Samples and Controls	Probes Reagent B, µl	Amplification Mix Reagent C, µl
1	5	15	33	178	535	65	351	1,050
2	11	33	34	184	550	66	356	1,070
3	16	48	35	189	565	67	362	1,090
4	22	66	36	194	580	68	367	1,100
5	27	81	37	200	600	69	373	1,120
6	32	96	38	205	615	70	378	1,130
7	38	115	39	211	635	71	383	1,150
8	43	130	40	216	650	72	389	1,170
9	49	147	41	221	665	73	394	1,180
10	54	160	42	227	680	74	400	1,200
11	59	177	43	232	696	75	405	1,210
12	65	195	44	238	715	76	410	1,230
13	70	210	45	243	730	77	416	1,250
14	76	230	46	248	745	78	421	1,260
15	81	245	47	254	760	79	427	1,280
16	86	260	48	259	777	80	432	1,300
17	92	275	49	265	795	81	437	1,310
18	97	290	50	270	810	82	443	1,330
19	103	310	51	275	825	83	448	1,340
20	108	325	52	281	845	84	454	1,360
21	113	340	53	286	860	85	459	1,380
22	119	357	54	292	880	86	464	1,390
23	124	370	55	297	890	87	470	1,410
24	130	390	56	302	905	88	475	1,420
25	135	405	57	308	925	89	481	1,445
26	140	420	58	313	940	90	486	1,460
27	146	440	59	319	960	91	491	1,470
28	151	450	60	324	970	92	497	1,490
29	157	470	61	329	990	93	502	1,510
30	162	485	62	335	1,000	94	508	1,520
31	167	500	63	340	1,020	95	513	1,540
32	173	520	64	346	1,040	96	518	1,550

Visit bio-rad.com/foodscience for more information.

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