final report

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Evaluation of novel DNA-based test kits

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Abstract

SwissDecode DNAFoil PCR kit is an on-site DNA test kit that can detect pork contamination in beef without a requirement for expensive PCR instrumentation and laboratory settings. This study was conducted to test the DNAFoil technology and assess the suitability of the test in controlled conditions. A total of five (5) DNAFoil tests were conducted on beef, pork and beef mixed with varied percentages of pork. The DNAFoil kit is self contained, easy to use and was able to successfully detect the pork contamination at low levels. However, a false positive was identified when beef was tested in the same environment where pork was handled previously. Although all care was taken to clean and decontaminate the working area and procedures to prevent cross contamination were followed, a false positive was observed. The number of kits tested is too small to draw conclusions on robustness of the kits and further testing on a larger sample size in different testing conditions is highly recommended.

Executive summary

SwissDecode DNAFoil PCR kit is an on-site DNA test kit that can detect pork contamination in beef without a requirement for expensive PCR instrumentation and laboratory settings. The test takes 30 minutes and does not require prior experience or specific molecular biology skills, making it suitable for field staff checking supply chain integrity in the meat industry. This study was conducted under the DNAFoil Early Access Program (DEAP) that allows priority access to test this technology and assess the suitability of this test. A total of fiver (5) DNAFoil tests were conducted on beef, pork and beef mixed with pork at both 1% and 0.1%. The DNAFoil kit was first evaluated on negative (100% beef) and positive (100% pork) controls and appropriate lines were observed on the test strip as directed by the kit manufacturer. The kit was then evaluated on beef with pork at both 1% and 0.1% and it was able to successfully detect the pork at these typical levels of contamination. Finally, the kit was evaluated for robustness and 100% beef samples were again tested in an environment where there was pork testing had previously been conducted. The kit detected a positive result on the 100% beef sample. Although all care was taken to clean and decontaminate the working area and procedures to prevent cross contamination were followed, a false positive was observed. A possible cause for this false positive could be the storage of the meat samples in the same fridge and/or the kit is extremely sensitive to pork.

The findings of this study demonstrate that the kit is self contained and has all that is required for performing the test. It was simple to use and detected pork contamination at relatively low levels. However, the number of samples (kits) tested is too small to draw any conclusions on robustness and further testing on a larger sample size in different testing conditions is highly recommended.

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1 Background

DNAFoil is claimed to be the world's first portable, completely self-administered, on-site DNA test that does not require expensive PCR equipment or laboratory settings to confirm detection of pork contamination in meat in as little as 30 minutes.

DNAFoil Early Access Program allows priority access to this technology and allows hand-on experience in handling these kits. The program also allows the user to evaluate whether the kits are fit for purpose.

2 Project objectives

The objective of the project was to evaluate novel DNA kits, in this first example, test if the DNAFoil kit detects pork contamination in beef at lower levels under controlled conditions.

3 Methodology

The DNAFoil kit is a self contained kit and comes with preparation barrel, reaction mixture tube and test strips. The homogenised meat is extracted using the preparation barrel in the kit and a drop of the extract is used in the reaction mix tube. The tube is then incubated in the kit water bath for 30mins and a test strip is dipped for detection. A single line on the strip indicates negative material and successful reaction. Development of double lines on the strip indicates positive material.

Beef and pork were sourced from the super market and were sub-sampled using fresh scalpel, disposable plates and knives. A sample size of at least 1000mg was taken from the meat chunks and finely chopped and homogenised using scalpel and knives. The homogenised sample was loaded into the preparation barrel in appropriate quantity as described in the kit operating procedures. The sample preparation and digestion work flow is shown in Figure 1.

Prior to testing each sample (kit), the work bench was decontaminated using 50 % bleach and thoroughly cleaned. The work bench was appropriately covered with a disposable bench coat before each test was performed. The DNAFoil kits were first tested on the negative control (100% beef). Homogenised beef was used as the starting material and processed according to the kit operating procedures. A small drop of the final extract from beef was then used in a reaction mix. After 30 minutes incubation in the kit water bath, the test strip was introduced into the reaction mix for the lines to develop. A single line on the strip indicated negative material and successful reaction. Development of double lines on the strip would indicate positive material. After the negative control test was performed the work area was cleaned and decontaminated. A new kit was opened and positive material (100% Pork) was tested.

The kit was then evaluated on beef with 1% and 0.1% pork. For 1 % pork in beef, 990 mg of beef was weighed on a balance and 10 mg pork was added. The meats were then mixed thoroughly and homogenised using disposable scalpels, plates and knives. Similarly for 0.1% pork in beef, 9990 mg of beef was weighed on a balance and 10 mg pork was added, homogenised and used for detection.

After the 4 kits were evaluated for the mixed meats, another negative control test was conducted using beef to check if the handling of pork in the same area affects the results for the negative samples. Following similar work flow, the work bench for this test was thoroughly cleaned and decontaminated using bleach. A section of beef was homogenise and processed using a fresh kit and the extracted material was used in the reaction mix.

4 Results

When 100% beef was used as the starting material, a single line was observed indicating negative for pork and a successful reaction (Figure 2 A). As expected a double line was observed for the positive control 100% pork (Figure 2 B). Similarly, the 1% and 0.1% Pork in Beef samples resulted in double lines in the test strips (Figure 2 C&D), i.e. positive for pork.

After all the tests were performed with either beef or beef mixed with pork, the 5th and final kit was used to test the negative control (beef) again. Although all care was taken to clean and decontaminate the working area and all procedures to prevent cross contamination were followed, the final testing with 100% beef turned out to be positive for pork (Figure 3).

5 Discussion

The results of this study demonstrate that the kit is self contained and has all that a user requires for performing the test. It was simple to use and detected pork contamination at relatively low levels. The testing required little previous experience and test results were available in 30mins. However, a positive result for the 100% beef sample in the final test indicates a false positive result for what was undoubtedly a negative sample. One of the possible reasons for this false positive detection may be the storage of the beef and pork samples in the same fridge or the kits are extremely sensitive to pork and detect pork once it is present in the testing environment. However, the sample size for this trial was too small (5 tests) to draw conclusions on the robustness of the kits. Further testing on a large sample size in different testing conditions is highly recommended to investigate the false positive result and establish robustness.

6 Conclusions/recommendations

1. DNAFoil kits are easy to use and are self contained. The user does not require any additional equipment or specialised skills to perform testing. The test results are developed in 30 mins and can be easily interpreted.
2. The sample size of the tests performed in this round of testing is too small to draw conclusions on the robustness of the kits and further testing is recommended.

7 Appendix

Figure 1 –Work Flow

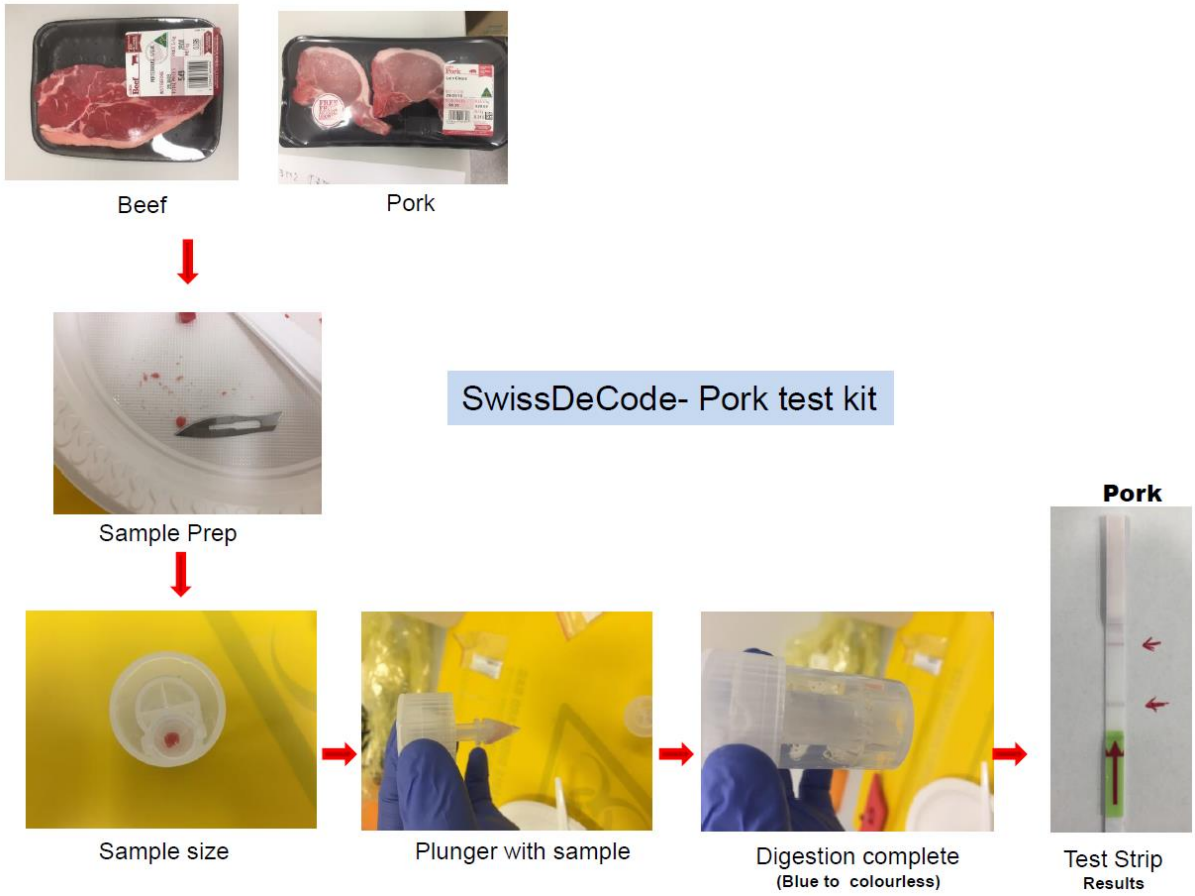


Figure 2

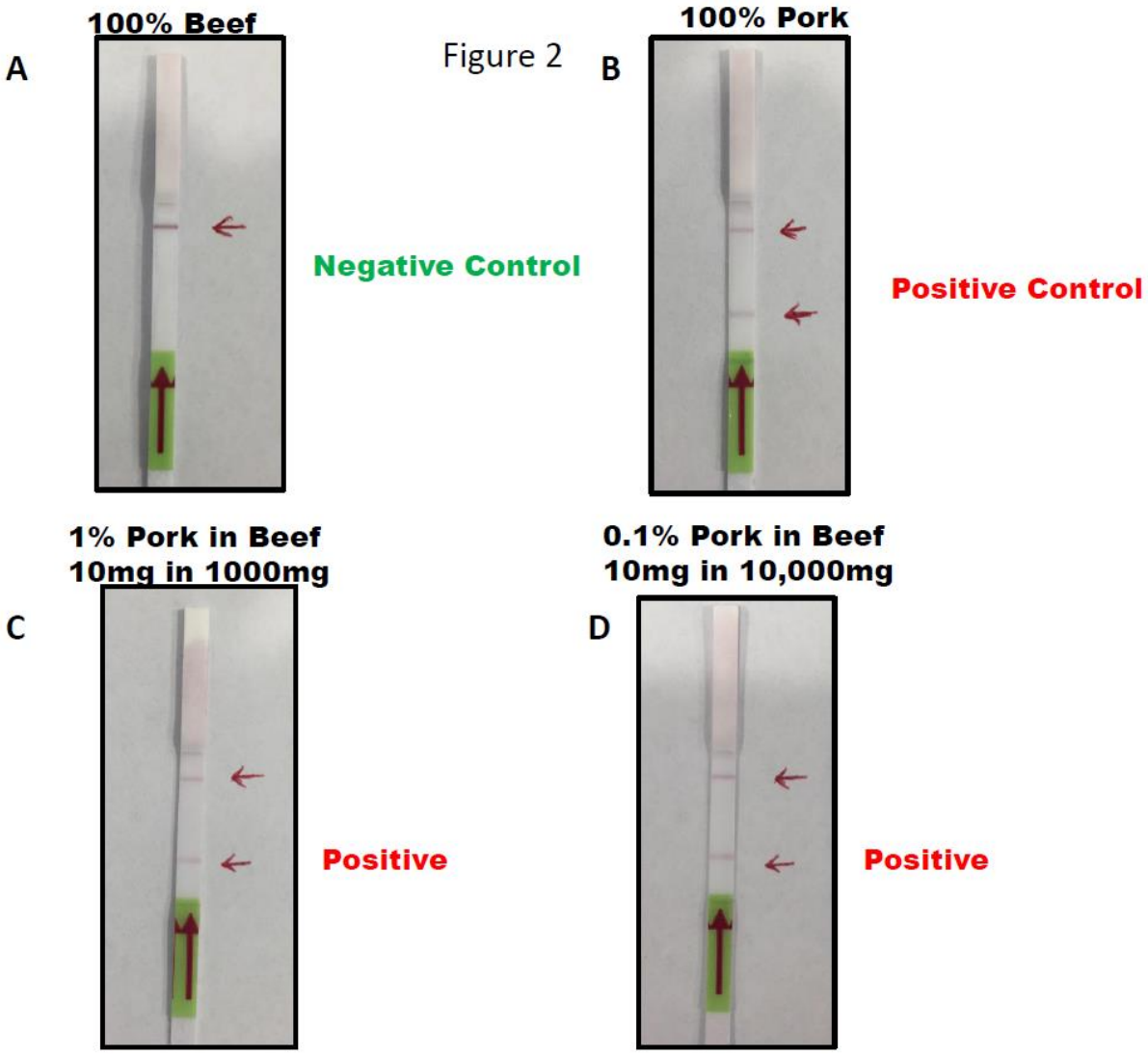


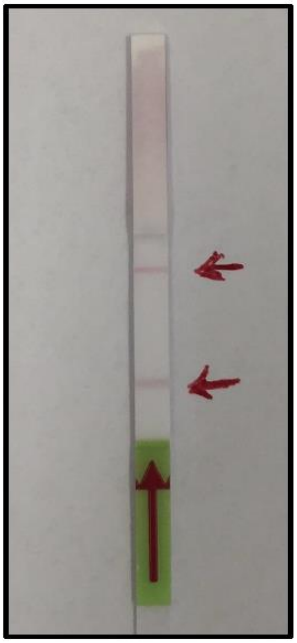
Figure 2
A
100% Beef
Negative Control
B
100% Pork
Positive Control
C
1% Pork in Beef
10mg in 1000mg
Positive
D
0.1% Pork in Beef
10mg in 10,000mg
Positive

Figure 3

Figure 3

Testing to check if Positive samples (PORK) in the environment impacts Negative Sample (Beef)

Beef-after-Pork testing



Positive for Pork