

Determination of Pancreatic Elastase 1 on epMotion 5070 or 5075

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Abstract

Here we describe a rapid, simple and semi- or fully automated Pancreatic Elastase 1 Determination on epMotion 5070 or 5075. In the procedure all washing, dispensing and aspirating steps are performed automatically. The automation of this ELISA kit greatly facilitates the test procedure and delivers reliable results.

Introduction

Human pancreatic elastase 1 (E1) remains undegraded during intestinal transit. Therefore, its concentration in feces reflects exocrine pancreatic function. The reliable and cost-effective ScheBo® Pancreatic Elastase 1 Stool Test is the new gold standard for noninvasive pancreatic function testing. In contrast to other laboratory parameters for the diagnosis of pancreatic disease (fecal chymotrypsin activity for the diagnosis of exocrine pancreatic insufficiency), the determination of pancreatic elastase 1 has the following advantages:

- E1 is absolutely pancreas-specific.
- Since E1 is stable during intestinal transit, the fecal elastase 1 concentration reflects the secretory capacity of pancreas.
- E1 determination correlates with both the gold standard invasive secretin-pancreozymin test and the secretin-caerulein test.
- Intra-individual variation of fecal E1 concentration is low.
- Enzyme substitution therapy has no influence on the determination of E1. The monoclonal antibodies used in the test do not cross-react with elastases of animal origin, which are contained in enzyme substitution preparations.

The ScheBo Pancreatic Elastase E1 is a test kit based on monoclonal antibodies. Eppendorf integrated this test into the workstation epMotion 5070 and also epMotion 5075.

Basic principle of the assay:

The ELISA plate is coated with a monoclonal antibody which only recognizes human pancreatic elastase 1 (E1). E1 from samples and standards binds to the antibody and is immobilized on the plate. A complex of monoclonal anti-Elastase 1-Biotin and Peroxidase (POD)-Streptavidin binds to E1 during the next incubation. The peroxidase oxidizes ABTS(2,2'-Azino-bis-(3-ethylbenzothiazolin-6-sulfonic acid) diammonium salt), which turns dark green. Finally, the concentration of oxidized ABTS is determined photometrically.

Materials and methods

Eppendorf epMotion 5070 or epMotion 5075 Reservoir holder Height Adapter (55 mm) Lid for MTP (optionally) Gripper and Gripper Holder Deepwell plates (optionally) ScheBo Pancreatic Elastase 1 Refer to the ScheBo Pancreatic Elastase 1 manual for complete instructions on preparing working solutions.

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Fig. 1: Screenshot from the epMotion editor showing the setup of the epMotion 5070 for the protocol Elastase 1



Fig. 2: Screenshot from the epMotion editor showing the setup of the epMotion 5075 for the protocol Elastase 1

On the epMotion 5075, incubation steps that needed to be carried out in the dark could automatically be processed on the worktable as the workstation operates with a special lid (Lid_1, position C4). On the epMotion 5070, this step was done manually.

Product use, limitations and safety information

Please read the ScheBo Pancreatic Elastase 1 manual before performing the method for the first time. More detailed instructions about the setup of the epMotion are available from Eppendorf upon request.

Quantification of results

After the end of the epMotion-program, the optical density has been measured at 405 nm with a microtiter plate reader. The evaluation is carried out by ELISA-software.

Results				
Standards				
Blank	0.108			
	Absor- bance1	Absor- bance2	Average	Concentration (µg E1/g stool)
STD1	0.179	0.197	0.188	15.0
STD2	0.486	0.524	0.505	50.0
STD3	1.214	1.263	1.239	150.0
STD4	2.321	2.29	2.306	500.0

Table 1: Absorbance of the Elastase Standards



Fig. 3: Standard Curve 15 to 500 µg/g

Control samples

Control	epMotion (μgE1/g stool)	Manually (µgE1/g stool)	Control value (µg E1/g stool)
1	195	195	200 (170 - 230)
2	201	209	200 (170 - 230)

Table 2: Control samples processed manually or by epMotion

Samples

Sample	epMotion (µgE1/g stool)	Manually (µgE1/g stool)
1	229	297
2	14	83
3	409	490
4	58	73
5	77	91
6	31	49
7	160	157
8	360	328

Table 3: Samples processed manually or by epMotion

Reference concentrations

for pancreatic elastase 1 in stool: normal:

- 200 to >500 µg E1 / g stool
- moderate to mild exocrine pancreatic insufficiency:
- 100 200 µg E1 / g stool
- severe exocrine pancreatic insufficiency:
- < 100 µg E1 / g stool

Conclusion

The Schebo Pancreatic Elastase 1 integrated into the workstation epMotion 5070 or 5075 offers a complete system for automated ELISA tests. The procedure is very easy to perform and user-friendly. The results using the epMotion correlate well with the manual results and allow a reliable determination of Pancreatic Elastase 1 in stool. As an additional benefit, a significant amount of time can be saved when working with the epMotion.

References

Eppendorf

Instruction Manuals for the epMotion 5070 and 5075 ScheBo

Instruction Manual Pancreatic Elastase 1

Ordering information			
Eppendorf			
Article	Description	Order no. international	Order no. North America
epMotion 5070	basic device includes control panel, software, optical sensor	5070 000.000 (200 – 240 V)	960000005 (100 – 130V)
epMotion 5075	basic device includes control panel, software,optical sensor	5075 000.008 (230V)	960020006 (120V)
Gripper	for transporting plates on the deck and for automatic opera- tion of the vacuum chamber	5282 000.018	960002270
Gripper holder	position plate for the gripper	5075 759.004	960002211
Reservoir rack	for use with 30 ml and 100 ml reagent reservoires	5075 754.002	960002148
Height adapter 55 mm	adapter for plates or racks	5075 752.000	960002113
Dispensing tool TM 1000-8	dispensing tool for the volume range from 40-1000 μl	5280 000.258	960001061
Deepwell plates (optional)	for processing, transport and storage of liquids		

ScheBo

Article	Supplier	Order no.
ScheBo Pancreatic Elastase 1	ScheBo Biotech	07



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