Short Paper

Lipase Activity in Scallop Hepatopancreas

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The high content of free fatty acids in the lipids of scallop hepatopancreas suggests lipase activity in the tissue,^{1,2)} but virtually nothing is known about it. We found that acetone-ether powders from scallop hepatopancreas are capable of hydrolyzing the ester bonds of the *sn*-1(3)-position of long-chain triacylglycerol molecules.

Acetone-ether powders were prepared as described by Bier³⁾ from the hepatopancreas of living scallops *Patinopecten yessoensis* and used without further purification. For the assay, the method of Luddy *et al.*⁴⁾ was modified. The acetone-ether powder (50 mg) was added to a solution containing olive oil triacylglycerols (5 mg), 22% CaCl₂ (0.2 ml), 0.05% sodium chorate (0.5 ml), and 0.32 M universal buffer (Britton-Robinson, pH 7.0 2 ml). The digestion was carried out in a screw-cap vial by stirring at 700 rpm for 20 h at 20°C. To avoid acyl migration under the acidic and alkaline conditions of *sn*-1,2(2,3)-diacylglycerols released during enzymatic hydrolysis, the solution was kept at pH 7.0. The reaction products were extracted with ether without acidification and separated by thin-layer chromatography (TLC) on silica gel G (0.5 mm thickness) impregnated with 5% boric acid.⁵⁾ Spots were

detected by exposing to iodine vapor and identified by comparing their *R_f* values with those of authentic standards. Porcine pancreatic and *Candida cylindracea* lipases (Sigma) were also used as standards of 1,3-specific and non-specific lipases, respectively.

The acetone-ether powders had an activity level of 0.3 units/mg solid. One unit will hydrolyze 1.0 nanoequivalent of fatty acid from a triacylglycerol in 1 h under the above conditions. Figure 1 shows the TLC chromatogram of the hydrolysates obtained from olive oil triacylglycerols by lipase preparations of scallop hepatopancreas, porcine pancreas, and *C. cylindracea*. Unhydrolyzed triacylglycerols (*R_f* 0.61), free fatty acids (*R_f* 0.40), diacylglycerols (*R_f* 0.21 and 0.14), and monoacylglycerols (*R_f* 0.01) were clearly resolved on borate TLC using *n*-hexane-ether (60:40, v/v) as the developing solvent. As expected, *C. cylindracea* lipase showed two clearly resolved diacylglycerol spots, which represent *sn*-1,3- and *sn*-1,2(2,3)-isomers; porcine pancreatic lipase showed only the *sn*-1,2(2,3)-isomer spot. The scallop lipase preparation also showed only one diacylglycerol spot, which had the same *R_f* value as that of the *sn*-1,2(2,3)-diacylglycerols obtained from the triacylglycerols by porcine pancreatic and *C. cylindracea* lipases. These results clearly show that the lipase preparation from scallop hepatopancreas is highly specific for the *sn*-1(3)-position of long-chain triacylglycerol molecules. Patton and Quinn⁶⁾ reported that the digestive lipase of the surf clam *Spisula solidissima* was capable of hydrolyzing the oleate or palmitate in the *sn*-1(3)-position of triacylglycerol molecules. Except for their study, there are no reports on the positional specificity of bivalve lipase, although 1,3-specific lipase has been found in fish.⁷⁾ The hepatopancreas of scallops is inedible and is an untouched marine resource. The present study suggests that the hepatopancreas would be a potential source of 1,3-specific lipases.

References

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Fig. 1. TLC separation of the hydrolysates of olive oil triacylglycerols by lipases of scallop hepatopancreas (A), porcine pancreas (B), and *Candida cylindracea* (C).

TG, triacylglycerols; FFA, free fatty acids; DG, diacylglycerols; MG, monoacylglycerols. 15, 100, and 350 units of A, B, and C were used for the reactions, respectively.