Pectin as a Modulator of Lipase Activity using an Olive Oil Substrate

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Introduction

Lipase is a pancreatic enzyme which catalyses the breakdown of fat in our diet [(1)]. In this study, olive oil was used as a natural substrate for lipase and orlistat, a known inhibitor of lipase, was used as a positive inhibitor.

Pectin is a polysaccharide, dietary fibre found in plant cell walls [(2)]. Three different pectin samples were provided by the University of York, extracted from potato peel. Each pectin sample was used at varied concentrations with lipase to see if pectin could increase or decrease the enzymatic activity of lipase. Consequently, altered lipase activity, due to pectin, could provide a new method of treatment in diseases, including obesity and pancreatic disorders.

Method – Olive Oil Assay

- Olive oil was passed through 8cm of Al_2O_3 to remove free fatty acids. A 10% solution was made using 8.0g of olive oil collected with 80ml of acetone. A 4ml 1% solution was then produced. 100ml of 0.05M Tris buffer was heated to 70℃ before adding the 1% olive oil solution and homogenizing for 10 minutes. A fresh substrate solution was required for each repeat [(3)].
- 1mg/ml of lipase was made in dH_20 and 60µl of colipase was added to the enzyme solution.
- Orlistat was used and made up to 5ml in substrate solution.
- Three pectin samples (120, 140 and 160) were made up to 5mg/ml in substrate solution. A serial dilution of each pectin sample in substrate solution was prepared in a 96 well plate.
- 10µl of lipase solution was added to the first three rows and 10µl of Tris buffer was added to the fourth and fifth rows of a second 96 well plate. 200µl was then transferred from the first well plate into the well plate containing lipase and Tris buffer.
- The assay was read at 405nm for 2 hours at 5 minute intervals.

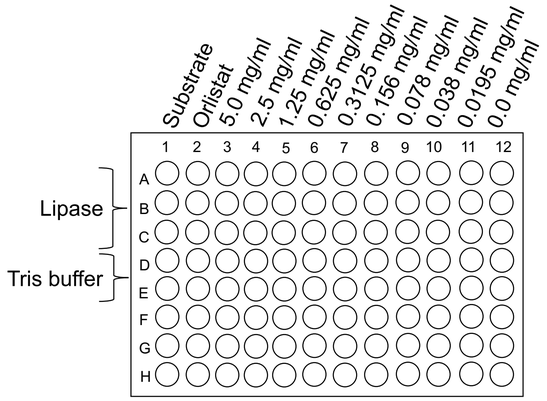

Figure 1. 96 well plate containing the reagents required for the olive oil assay.

Results

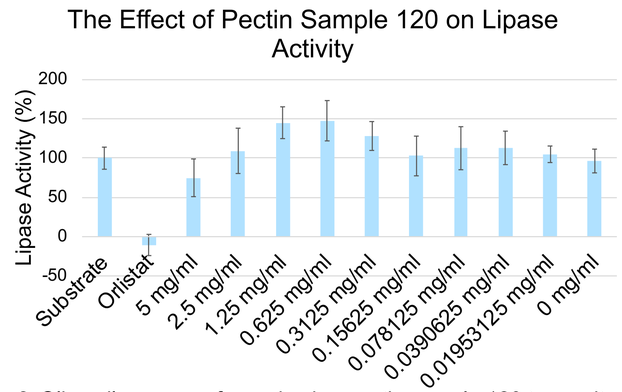

Figure 2. Olive oil assay performed using pectin sample 120 to monitor lipase activity. The data corresponds to the mean SD of four repeats.

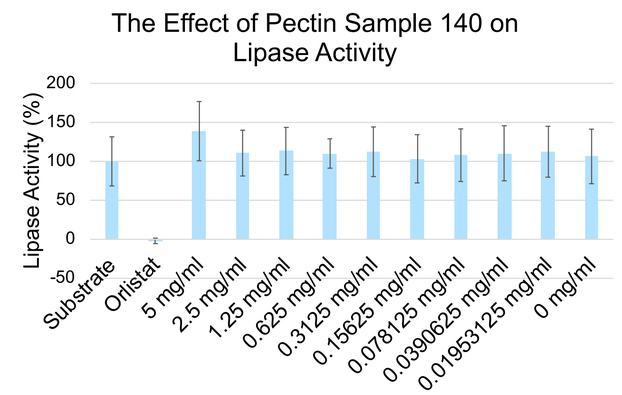

Figure 3. Olive oil assay performed using pectin sample 140 to monitor lipase activity. The data corresponds to the mean SD of four repeats.

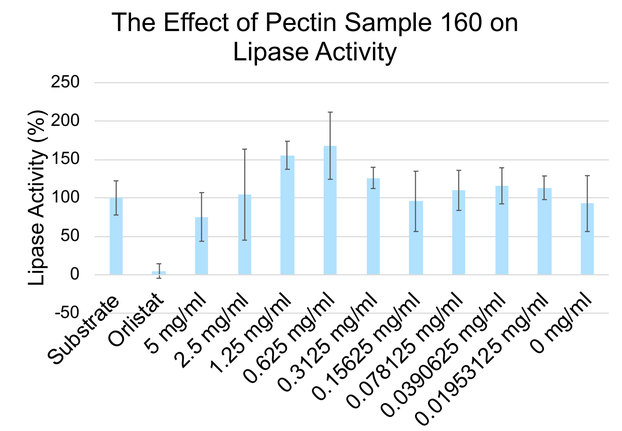

Figure 4. Olive oil assay performed using pectin sample 160 to monitor lipase activity. The data corresponds to the mean SD of four repeats.

Discussion

Pectin sample 120 caused both an increase and decrease in lipase activity. The highest activity of lipase was observed at 0.625 mg/ml at 148%, whereas a decrease of lipase activity to 75% was observed at 5mg/ml.

On the other hand, pectin sample 140 increased lipase activity at all concentrations used. The highest activity of lipase was established at 5 mg/ml of pectin at 138% and the smallest increase of lipase activity was seen at 0.156 mg/ml at 103%.

Similarly to pectin sample 120, pectin sample 160 caused both an increase and decrease in lipase activity. An increase in activity was observed at all concentrations excluding both 5mg/ml (75%) and 0.156 mg/ml (95%) which decreased enzymatic activity. The highest increase of lipase activity was observed at 0.625 mg/ml (168%).

As expected, orlistat caused a significant decrease or absence of lipase activity in the assay.

Conclusion

- Pectin can increase and decrease lipase activity.
- Pectin has the capacity to cause a more significant increase than decrease in lipase activity.
- Pectin is more likely to be used in pancreatic disorders where lipase function is impaired, compared to in obesity where lipase activity would be required to be subsided to reduce the breakdown of fat and therefore absorption of fat into the body.
- Further research would look into the effect of pectin on other digestive enzymes, including pepsin and amylase.

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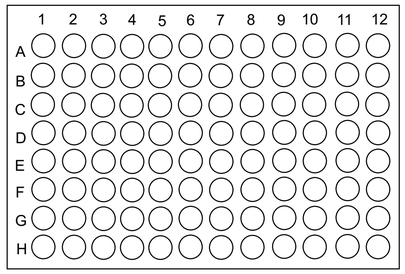
References

1. MedlinePlus. Lipase Tests. 2018. [cited 5/8/19]; Available from: https://medlineplus.gov/lab-tests/lipase-tests/
2. ScienceDirect. Pectin. 2003. [cited 5/8/19]; Available from: https://www.sciencedirect.com/topics/neuroscience/pectin
3. Wilcox MD, Brownlee IA, Richardson JC, Dettmar PW, Pearson JP. The modulation of pancreatic lipase activity by alginates. Food Chemistry 2014; 146: 479-484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