Name (section 101)
20 October 2003
Lab 4

Isolation and isomerization of lycopene from tomato paste

Abstract

Pigment was extracted from tomato paste using a 50:50 hexane-acetone solution and washed with sodium chloride and potassium carbonate. Lycopene was separated from this mixture through column chromatography and the use of hexane and acetone. Spectral analysis of the lycopene revealed that 58% (class results ranged from 28%-63%) of the lycopene was all-trans.

Introduction

This experiment was conducted primarily as an introduction to the technique of column chromatography, though the technique of liquid-solid extraction was also emphasized, and the theory behind ultraviolet-visible spectrometry was touched upon. These techniques were utilized in order to determine the percentage of all-trans lycopene present in various kinds of tomato paste. The working hypothesis for this experiment was that a lower percentage of all-trans lycopene present reflected poorly upon the quality of the tomato paste, in that exposure to heat and light will isomerize all-trans lycopene into a completely or partially cis-compound.

Column chromatography allows for the separation of several components in solution by taking advantage of the varying solubilities and polarities of the dissolved components. In this

case, alumina was used as the stationary phase in the column, and hexane was used as the mobile phase to saturate the column and move the components through. Two pigments, carotene and lycopene, were present in tomato paste in sufficient quantities to observe in the column. Carotene, with its lesser affinity for alumina, moved through the column first, while lycopene followed shortly after, aided by the addition of acetone.

The technique of liquid-solid extraction varies little from liquid-liquid extraction, with the exception that one of the materials is a solid. In this experiment the solid was tomato paste, which can almost be considered more as a very viscous liquid, and the liquid was a 50:50 hexane-acetone solution.

While the ultraviolet-visible spectral analysis was not performed by the students but rather by the professor, the theory behind it is based upon the fact that stereoisomers will absorb ultraviolet light maximally at different wavelength because of their varying structures. By determining these absorption maxima, one can determine the percentage of all-trans lycopene present.

Material and Methods

The experiment was followed as dictated by the procedure laid out in the distributed lab manual[1] (Lehman 78-86) and the distributed lab supplement[2] with the following adjustment: the carotene could not be observed in the column, so it was not collected separately, and all the recovered pigment was labeled as lycopene.

Results

The lycopene from a sample of tomato paste (1.45 g) was extracted and isolated. An ultraviolet-visible spectral analysis was performed, revealing three distinct absorption peaks (see attached graphs and Table 1). The percent of all-trans lycopene was determined by dividing the height of the third peak by that of the second (equation 1).

(1) $$\frac{0.90\text{ cm}}{1.55\text{ cm}} = 58\%$$

Discussion

Overall, the percentage of all-trans lycopene present in the sampled tomato pastes seemed rather low. The results ranged from 28% to 63%, with the upper limit at first glance appearing to be a reasonable measure of a quality product. But when one considers the fact that tomatoes in their natural state contain only all-trans lycopene (Lehman 78), these results indicate that minimally, 37% of the all-trans lycopene has isomerized to its cis form. This does not reflect well upon the quality of the product, and certainly does not show a high grade of quality for those pastes with only 28% all-trans lycopene.

It is possible that the quality of the products was not reflected accurately by this experiment. That is, perhaps the very isolation of the lycopene created conditions conducive to isomerization. This could very well have been the case, especially when you consider the one instance in which one brand was analyzed by two different lab sections. In section 103, Country's Delight was determined to have 33% all-trans lycopene, while in section 105, the

same brand was determined to have 63% all-trans lycopene. While it is possible that the cans differed in composition, it seems more likely that the lycopene was mishandled somehow to give such a low reading. Treatment of the paste with hexane, acetone, sodium chloride, and potassium carbonate may have induced isomerization, and undue care in handling the lycopene (exposure to too much heat and light) would have accomplished the same thing.

While the results did not reveal a high degree of freshness or quality in the tomato pastes tested, it did permit an introduction to the technique of column chromatography, and allow for the refinement of previously learned extraction techniques (solid-liquid in this case).

References/Literature cited

1. Lehman, John W. *Multiscale Operational Organic Chemistry*; Prentice Hall: Upper Saddle River, 2002; Part 1, 78-86.

2. Dintzner, M. R. http://condor.depaul.edu/~mdintzne/171-03*Lab4sup.pdf

Figures/Tables/Data

Table 1: Spectral analysis of lycopene (see attached graphs)

Lab section	Brand	height of third peak (cm)	height of second peak (cm)	third/second (% all-trans)
101	Dominick's	0.90	1.55	58%
102	Contadina	0.85	1.39	61%
103	Country's Delight	0.61	1.85	33%
104	Whole Foods 365	0.58	1.10	53%
105	Country's Delight	0.82	1.10	63%
106	Hunts	0.55	1.95	28%

Questions

Lehman, Questions 3-5, p 85-86

3. a) *structures hand-drawn in*

b) 13-cis-lycopene is considerably more stable because, unlike in the other two isomers, there is no steric strain between the functional methyl groups.

4. a) If the tomato paste had been opened for several days, it would have likely been oxidized and isomerized from the exposure to the atmosphere and other chemical agents in the refrigerator.

b) There would not be a large amount of measurable isomerization if the second UV-VIS reading was taken immediately after the iodine was added because the iodine would not have had time to interact with the lycopene.

c) The acid in the alumina would likely have isomerized the all-trans lycopene to other cis stereoisomers.

5. a) Most hydrocarbons are not colored because they reflect the majority of the light that they encounter, whereas lycopene and β-carotene absorb certain frequencies of light, giving them a colored appearance.

b) Adding excess iodine will saturate some of the double bonds, breaking up the conjugation and reducing absorption in the visible region (loss of color).