

bodies to produce derivatives of triphenyl methane. Danckwortt<sup>6</sup> condensed *p*-nitrobenzaldehyde with phenolic bodies. Since these condensations present possibilities for the preparation of dyes they are now being investigated.

The above tabulated list of compounds like many other similar condensation products are affected to a considerable extent by the action of light. It was noted that the halogenated derivatives were particularly susceptible to light.

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### NOTES.

**The Enzymes of the Abdominal Adipose Tissue of the Common Turkey, *Meleagris Gallopavo*.**—While the constants of turkey fat have been studied by several investigators, the enzymes, which occur in the adipose tissue (crude fat), have not been studied. Such a study, made upon samples of known history, is reported in this paper. The turkeys were hens, grown and killed in central Tennessee. They were marketed during the Christmas season. In trade parlance, they were dry picked, air chilled, well bled, undrawn (uneviscerated) birds. After slaughter, they were kept in a mechanically refrigerated chill room or cooler at a temperature of approximately 0° for 3 days, then were shipped to this city by express, two days being required for the journey. Upon arrival at Philadelphia, they were again placed in a mechanically refrigerated chill room, in which they were kept at a temperature of approximately 0° for from 3 to 7 days; they were then removed for study.

An aqueous extract of the abdominal adipose tissue (crude gizzard fat) was prepared as in the studies of chicken fat by Pennington and Hepburn.<sup>1</sup> The technic in the test for lipase, esterase, catalase, reductases, oxidase, and peroxidase was that used in the studies cited with the following modifications. Tributyrin was used as a substrate for lipase, and ethyl butyrate for esterase. In testing for catalase, 20 cc. of the aqueous extract was taken; and the volume of the evolved oxygen was recorded without correction. As reagents for oxidizing enzymes, use was made of  $\alpha$ -naphthol, trikresol, and phenolphthalin; when phenolphthalin was the substrate, 0.1 *N* sodium hydroxide solution was added after incubation until the phenolphthalein present showed its maximum red color.

The proper control experiments were always made on the boiled aqueous extract in order to allow for all changes in the substrate not due to enzyme action. Trikresol was used as a bactericide at a concentration of 0.2% in the tests for lipase, esterase, simple reductase, and protease.

<sup>6</sup> Danckwortt, *Ber.*, **42**, 4163 (1909).

<sup>1</sup> Pennington and Hepburn, *THIS JOURNAL*, **34**, 210 (1912); *Bur. Chemistry, Circ.*, **75**, 1 (1911); **103**, 6 (1912).

The results of the tests for *lipase*, *esterase*, *catalase*, *simple reductase*, and *aldehyde reductase* have been collected in the table; a plus + indicates the presence, a zero (0) the absence of reductase.

TABLE I.

Occurrence of Certain Enzymes in the Abdominal Adipose Tissue of the Common Turkey.

Expt. Number.	Lipase.	Esterase.	Catalase.		Reductase.	
			Cc. O <sub>2</sub> evolved.		Simple.	Aldehyde.
	Cc. 0.1 N butyric acid. Liberated in 72 hours.		30 min.	Time. 24 hours.		
Hen.						
1.....	2.20	....	12.7	39.0	+	....
2.....	5.35	1.25	10.8	....	+	0
3.....	4.90	0.35	....	7.9	+	0
4.....	6.40	0.85	....	22.5	+	Trace
5.....	6.80	0.80	....	31.4	+	Trace
6.....	4.70	0.40	....	2.0	0	0

**Oxidases** acting upon trikresol and upon  $\alpha$ -naphthol were not found. All but one of the samples contained an oxidase acting upon phenolphthalin. One sample was also tested for cresol tyrosinase,<sup>2</sup> the oxidase which produces colored compounds by its action on a mixture of *p*-cresol and amino acids or secondary protein derivatives; the substrates used were trikresol plus asparagin and trikresol plus Witte peptone; both tests were entirely negative.

**Peroxidase** acting upon  $\alpha$ -naphthol occurred in one sample, and peroxidase acting upon phenolphthalin was found in another sample. Peroxidase acting upon trikresol was not detected.

**Protease.** Two of the samples were tested for the presence of a protease. The results indicated that such an enzyme, active in the presence of hydrochloric acid, did not occur in the adipose tissue.

A 20cc. portion of the aqueous extract of one sample was permitted to act upon a flock of carmine fibrin in the presence of 0.2% hydrochloric acid; the fibrin was not attacked after incubation for 24 hours at a temperature of 37.5°.

A 2.5cc. portion of the aqueous extract of another sample was mixed with 2 cc. of a 2% solution of castor bean globulin in 5% sodium chloride solution; and 0.5 cc. of 0.1 N hydrochloric acid was then added. The precipitated protean, derived from the globulin, was not attacked during incubation for 7 days at a temperature of 37.5°.

### Summary.

Catalase, lipase (acting on tributyrin), and esterase (acting on ethyl butyrate) were always found in the adipose tissue. Simple reductase

<sup>2</sup> *Arch. sci. phys. nat.*, [4] 33, 70 (1912).

and oxidase acting upon phenolphthalin were usually present. Tests for oxidases, which act on  $\alpha$ -naphthol and on trikresol, and for protease gave negative results. Aldehyde reductase and peroxidases were found in several of the samples.

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**Friedel and Crafts' Reaction. The Preparation of 2-*para*-Toluy-Benzoic Acid.**—In order to determine whether the results obtained by Rubidge and Qua<sup>1</sup> in the study of the reaction between phthalic anhydride, benzene and aluminum chloride are of general application the reaction between phthalic anhydride, toluene and aluminum chloride has been studied. In the following table the yields are calculated on the phthalic anhydride used.

No.	Toluene. G.	Phthalic anhyd. G.	AlCl <sub>3</sub> G.	Time. Hrs.	Toluybenz. acid. %.	Ditoly- phthalide. %.
1	20	5	9	12	89	5.4
2	20	5	9	12	92.5	3.9
3	43	5	9	12	90	5
4	20	5	9	12	95	1.9
5	20	5	10	10	97	trace
6	20	5	4.5	12	19	34
7	20	5+5	9	12+12	19	27
8	20	5+5 cc. acet. anh.	9	12-12	none	52
9	20	5+3.2 cc. acet. anh.	9	12-12	none	45

Nos. 1 and 2 show that using the proportions of the reagents indicated the yield of toluyl-benzoic acid is about 90%, and that some ditolyl-phthalide is formed. In No. 3 the increased amount of toluene did not materially affect the yield. In No. 4 the aluminum chloride was finely powdered and in No. 5 the slightly larger amount of aluminum chloride prevented the formation of any ditolyl-phthalide. In No. 6 with half the usual amount of aluminum chloride the yield of acid is greatly reduced and ditolyl-phthalide increased. In No. 7 the usual proportions of the reagents were used, but after 12 hours' boiling additional phthalic anhydride was added and boiling continued for 12 hours. The results show that the additional phthalic anhydride reacted with the intermediate compound to give ditolyl-phthalide. Nos. 8 and 9, with acetic anhydride used instead of the additional phthalic anhydride after the first 12 hours' boiling, gave no toluyl-benzoic acid and a better yield of ditolyl-phthalide.

These results agree completely with those obtained by Rubidge and Qua.

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<sup>1</sup> Rubidge and Qua, THIS JOURNAL, 36, 732 (1914).