STRUCTURAL CARBOHYDRATE CHEMISTRY

E. G. V. PERCIVAL

Revised by

ELIZABETH PERCIVAL

Second Edition

STRUCTURAL CARBOHYDRATE CHEMISTRY

BY

E. G. V. PERCIVAL

D.Sc., Ph.D., F.R.I.C.

LATE READER IN CHEMISTRY, UNIVERSITY OF EDINBURGH

REVISED BY

ELIZABETH PERCIVAL

D.Sc., Ph.D., F.R.S.E.

LECTURER IN CHEMISTRY, UNIVERSITY OF EDINBURGH

FOREWORD BY

E. L. HIRST

C.B.E., F.R.S.

PROFESSOR OF ORGANIC CHEMISTRY
UNIVERSITY OF EDINBURGH

LONDON
J. GARNET MILLER LTD
13 Tottenham Street W. 1

SECOND EDITION, 1962
PRINTED IN GREAT BRITAIN BY
ROBERT MACLEHOSE AND CO. LTD., GLASGOW

FIRST PUBLISHED BY FREDERICK MULLER, LTD., IN 1950
REPRINT PUBLISHED BY J. GARNET MILLER LTD.,
IN 1953

FOREWORD

In view of the importance of the structural chemistry of the carbohydrates as a necessary background in many fields of study it is surprising that so few textbooks have been published which aim at serving the needs of students of chemistry and biochemistry and which would at the same time be of general use to research workers in both industrial and academic laboratories. After the publication of Sir Norman Haworth's *The Constitution of Sugars* in 1928 there was a long gap and when this came to be recognised one of the first and most successful of the new publications was the late E. G. V. Percival's *Structural Carbohydrate Chemistry* which appeared in 1950. This filled admirably the need for a short but comprehensive and authoritative guide to carbohydrate chemistry as it had developed during the previous twenty-two years. The ten years since 1950 have seen astonishing changes in our knowledge of the structural chemistry of natural products and the rate of progress has been by no means least in the carbohydrate field. This is clearly an appropriate time for a new edition of *Structural Carbohydrate Chemistry*, incorporating the manifold changes and additions to knowledge made during the past decade, and new readers as well as those who have so greatly appreciated the earlier volume will welcome the second edition which has been prepared by Dr. Elizabeth Percival. A major difficulty which has to be faced by the author of such a book is how to select the fundamental and important aspects from the immense mass of published material, but in her position as an acknowledged authority in this field Dr. Percival has been able to make a wise assessment of the salient points. The new book is necessarily somewhat longer than the first edition, little of which could be discarded as unnecessary or outmoded, but the reader will find in it a concise, reasoned and critical survey of structural carbohydrate chemistry as it stands in 1961. In welcoming the advent of the new edition I have every reason to believe that it will fulfil its purpose as admirably and as completely as did the previous volume.

E. L. Hirst

PREFACE TO THE SECOND EDITION

IN this revised edition an attempt has been made, while retaining the same method of approach, to bring all the subject matter up to date. At the same time it has been necessary to keep the book within a reasonable format.

Only minor changes have been made in the first four chapters. These include, among other things, the inclusion of Hudson's Isorotation rules and a new section on sugar conformation, the revision of the structure of sugar phenylosazones including their conversion to formazan derivatives and isotriazoles, and a considerable increase has been made in the section on amino sugars.

A number of new disaccharides and their more recent syntheses have been added and a completely new chapter on oligosaccharides written. This latter is divided into three sections: Those oligosaccharides which contain a molecule of sucrose linked to other sugar units, the human milk oligosaccharides including lactaminic acid, and the Schardinger dextrins. The sections on nitrogen glycosides, and on the inositols, together with all the chapters on polysaccharides, have been re-written and re-grouped. A description of more recently investigated polysaccharides, such as the blood group substances, has been added. Although the manuscript was completed in 1959, owing to delays in publication it has been found possible to include new material from original work published during 1960.

My grateful thanks are due to Professor E. L. Hirst, C.B.E., F.R.S., who has read the whole of the manuscript. I am indebted to my colleagues in the Department of Chemistry, and to Dr. D. J. Bell for helpful discussions, and particularly to Dr. G. O. Aspinall and Dr. D. J. Manners for helpful criticism on the chapters on cellulose, and on starch and glycogen respectively, and I am pleased to acknowledge the help I have had from Dr. J. C. P. Schwarz in the preparation of the sections on Conformation and on the Inositols. Finally I wish to express my gratitude to Dr. Eric Dewar for his generous and willing help in the arduous task of proof correction.

November 1959 ELIZABETH PERCIVAL

PREFACE TO THE FIRST EDITION

THIS book is intended to be of service to students reading for an Honours degree in Chemistry, and to serve as a text-book in the later stages of preparation for ordinary and combined degrees, and for examinations in biochemistry and pharmaceutical chemistry. It is hoped also that research workers in both the academic and industrial fields will find some useful information within its pages.

When the original manuscript was completed early in 1948, the only text-book in English on the subject was *The Constitution of the Sugars*, and the advances made since the publication of Sir Norman Haworth's classic in 1928 appeared to be a sufficient justification for an attempt to bring the subject up to date. Owing to delays in publication it has been found possible to include new material from original work up to the early part of 1949.

I wish to acknowledge the debt I owe to my collaborators in research, and especially to those in whose laboratories I have had the good fortune to work, Professor W. Wardlaw, (the late) Professor H. Hibbert, Professor Sir Norman Haworth, Professor Kendall and Professor E. L. Hirst.

In the preparation of this book I have had the invaluable assistance of my wife, especially in the preparation of the formulae, the index and the manuscript, as well as in the reading of proofs and I wish to place on record my appreciation and thanks.

Finally I wish to thank the publishers for their assistance at every stage, and especially Dr. J. G. F. Miller who, indeed, persuaded me to write this book.

September 1949 E. G. V. P.

CONTENTS

CHAPTER I

INTRODUCTION. THE GENERAL PROPERTIES, CONFIGURATIONS AND INTERRELATIONSHIPS OF THE MONOSACCHARIDES

Introduction

It is generally conceded that the carbohydrates, as a group, are compounds of the highest importance and interest, viewed from both the practical and the theoretical aspects. The process of photosynthesis by means of which all life on this planet is sustained, is concerned with the manufacture of carbohydrates in the plant from carbon dioxide and water, although we do not yet know the precise way in which this apparently facile transformation is brought about. Carbohydrates therefore represent a great storehouse of energy, either as foodstuffs for men and animals, or after transformations in the geological past, as coal and peat. Apart from these considerations and the special importance of carbohydrates in plant and animal biochemistry, the carbohydrates are of intrinsic interest as chemical individuals, and by their study chemistry, and especially stereochemistry, has been greatly enriched.

The term carbohydrate originated in the belief that naturally occurring compounds of this class for example glucose, $C_6H_{12}O_6$, sucrose $C_{12}H_{22}O_{11}$, starch $(C_6H_{10}O_5)_n$, could be represented formally as hydrates of carbon, that is $C_x(H_2O)_y$. This definition is too rigid, however, since the important deoxy sugars, for example rhamnose, $C_6H_{12}O_5$, the uronic acids and such compounds as ascorbic acid, would thereby be excluded and acetic acid and phloroglucinol would qualify for inclusion. Nevertheless the term carbohydrate remains in general use to describe those polyhydroxy compounds which reduce Fehling's solution either before or after hydrolysis with mineral acids.

The main divisions it is customary to make within the carbohydrate group are the *Monosaccharides* or simple sugars like glucose, xylose, fructose, the *Disaccharides* such as maltose, lactose, and sucrose, formed, as the name implies, by the union of two monosaccharide units, the *Trisaccharides* of which raffinose is an example, and the *Polysaccharides*, a group which includes starch, glycogen and cellulose, in each of which a large number of monosaccharide units are combined. The term *Polyuronide* denotes a polysaccharide containing uronic acid residues, and the term *Oligosaccharide* is used, especially on the Continent, to denote compound sugars containing between two and six simple sugar units, that is ranging from di- to hexa-saccharides.

It is necessary to subdivide the monosaccharide groups in such a way that the number of carbon atoms in the molecule is indicated in the name. Thus a hexose contains six, a pentose five, and a heptose seven carbon atoms in the molecule, and by using the prefix aldo-, or keto-, a distinction is made between those sugars which have a potential aldehydic group at one end of the chain, and those in which a keto group is fundamental to the structure. Thus glucose is described as an aldohexose and fructose a ketohexose. Finally, as will appear in the sequel, since sugars, whether simple or combined, normally exist in cyclic forms, and the type of ring may vary according to the circumstances of formation, it is frequently necessary to denote the precise ring system by a suffix, for example pyranose or furanose, to indicate a six- or a five-membered ring respectively, and so we have such expressions as glucopyranose, and xylofuranoside to define even more precisely the nature of the compound under consideration.

General Properties of the Monosaccharides

If we take the hexoses as typical examples we find that the simple sugars are generally colourless crystalline solids with a sweet taste. This latter property is evidently dependent on the constitution, for glucose is much sweeter than mannose and the ketohexose, fructose, is the sweetest of all the sugars; this explains the sweetness of honey which is a mixture of glucose and fructose. All the hexoses are readily soluble in water to give optically active solutions which readily reduce Fehling's solution and ammoniacal silver nitrate, char or caramelise very readily on heating or in the presence of concentrated sulphuric acid, and suffer extensive decomposition on heating with caustic alkalis.

Emil Fischer* (1852-1919) applied his great gifts to a study of the monosaccharides and showed how they were interrelated. Although in some points of detail modifications have taken place since

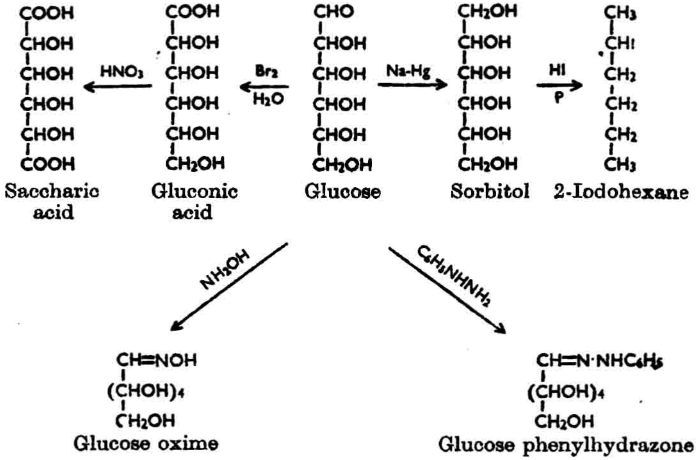
---

* *Untersuchungen über Kohlenhydrate und Fermente*, Berlin, 1909.

his time, Fischer laid the foundations of the present edifice of carbohydrate chemistry. Since Fischer's work on the sugars commenced (1884) with a study of the properties of glucose it is appropriate to make that sugar our starting point.

Glucose as a Pentahydroxy Aldehyde

That glucose can be represented as a pentahydroxy aldehyde is shown by the following facts. Reduction with sodium amalgam in faintly acid solution gives the hexahydric alcohol sorbitol.* This gives a hexaacetate, and, on vigorous reduction yields 2-iodohexane, clearly showing that no branchings occur in the chain. Oxidation of glucose with bromine water gives gluconic acid and more vigorous oxidation with nitric acid yields saccharic acid,† a tetrahydroxy dibasic acid. The fact, that on reduction glucose gives an alcohol and on mild oxidation a carboxylic acid without the loss of a carbon atom, is strong presumptive evidence that it contains an aldehydic group. This view is strengthened by the fact that glucose forms an oxime and a phenylhydrazone. Confirmatory evidence that an aldehydic group is present on the terminal position in glucose is furnished by the highly important reaction, discovered by H. Kiliani in 1886‡ wherein glucose unites with hydrocyanic acid to form a cyanhydrin (or more exactly two cyanhydrins), which on hydrolysis followed by reduction yield *n*-heptylic acid.

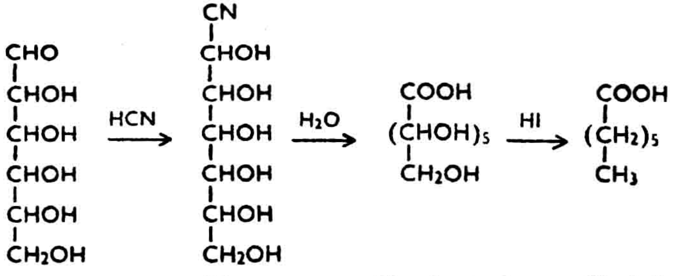


---

* Now generally known as D-glucitol to indicate relationship to parent sugar.

† Now generally known as D-glucaric acid.

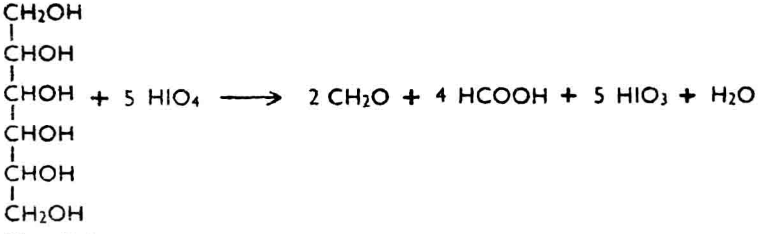
‡ *Ber.*, 1888, **21**, 915; 1886, **19**, 3029.



$$\begin{array}{c}CHO\\|\\CHOH\\|\\CHOH\\|\\CHOH\\|\\CHOH\\|\\CH_2OH\end{array} \xrightarrow{HCN} \begin{array}{c}CN\\|\\CHOH\\|\\CHOH\\|\\CHOH\\|\\CHOH\\|\\CHOH\\|\\CH_2OH\end{array} \xrightarrow{H_2O} \begin{array}{c}COOH\\|\\(CHOH)_5\\|\\CH_2OH\end{array} \xrightarrow{HI} \begin{array}{c}COOH\\|\\(CH_2)_5\\|\\CH_3\end{array}$$

Glucose cyanhydrin Glucoheptonic acid *n*-Heptylic acid

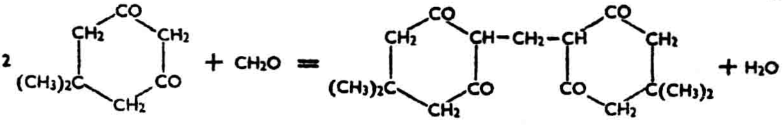
Perhaps the most direct confirmation of the above structure is furnished by the reaction, discovered by Malaprade*, and destined to have a very powerful influence on modern research in carbohydrate chemistry.† Malaprade showed that periodic acid (HIO_4) smoothly oxidised α-glycols with the production of formic acid from a secondary alcohol group and formaldehyde from a primary alcohol residue. Thus the hexahydric alcohol, mannitol, consumed five molecules of periodic acid, readily estimated by titration, with the production of two molecules of formaldehyde and four molecules of formic acid.



$$\begin{array}{c}CH_2OH\\|\\CHOH\\|\\CHOH\\|\\CHOH\\|\\CHOH\\|\\CH_2OH\end{array} + 5\ HIO_4 \longrightarrow 2\ CH_2O + 4\ HCOOH + 5\ HIO_3 + H_2O$$

Mannitol

The formaldehyde is readily estimated as the crystalline condensation product with " dimedone " (5,5-dimethyl *cyclo*hexane 1,3-dione),



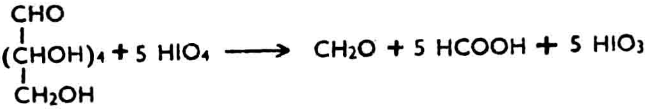
---

* *Bull. soc. chim.*, 1928, **43**, 683; 1934, (5), 1, 833.

† See Bobbitt, *Advances in Carbohydrate Chem.*, 1956, **11**, 1.

and the formic acid by titration. Colorimetric methods for the assay of μg quantities of formaldehyde have been developed[*],

When this facile reaction is applied to glucose the products[†] set



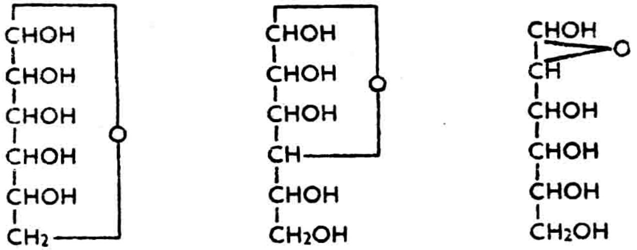
$$\begin{array}{l} CHO \\ | \\ (CHOH)_4 \\ | \\ CH_2OH \end{array} + 5\,HIO_4 \longrightarrow CH_2O + 5\,HCOOH + 5\,HIO_3$$

out in the above equation are liberated.

The oxidation of sugars with lead tetraacetate, although not so convenient to apply, follows a somewhat similar course to that using periodic acid; lead tetraacetate, however, oxidises *cis* α-glycol groups preferentially,[‡] whereas periodic acid does not distinguish between *cis* and *trans* α-glycols. It is considered that the formation of a cyclic intermediate between the α-glycol groups and the oxidant is generally,[§] although not always, necessary before cleavage can take place.

Objections to the Representation of Glucose as a Free Aldehyde

Although the above evidence that glucose contains an aldehyde group may seem convincing, there are important facts which are in disagreement with this conclusion. As B. Tollens[‖] pointed out, as far back as 1883, although glucose vigorously reduces Fehling's solution to cuprous oxide, and ammoniacal silver nitrate to metallic



Early cyclic formulae for glucose

---

[*] Mitchell and Percival, *J.C.S.*, 1954, 1423; Hough, Powell and Woods, *J.C.S.*, 1956, 4799.

[†] Fleury and Lange, *J. Pharm. Chim.*, 1933, **17**, 409.

[‡] Hockett, Nickerson and Reeder, *J. Amer. Chem. Soc.*, 1944, **66**, 472; Abraham, *ibid.*, 1950, **72**, 4050.

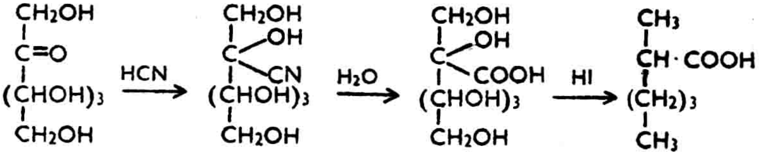
[§] Criegee, Kraft and Rank, *Annalen*, 1933, **507**, 159; Buist and Bunton, *J.C.S.*, 1954, 1406.

[‖] *Ber.*, 1883, **16**, 921.

silver, the reducing power is far in excess of that which could be accounted for by a single aldehydic residue. Furthermore it may be stated that fructose, which as will be seen shortly is apparently a pentahydroxy ketone, reduces equally vigorously. The evidence in favour of an aldehydic structure based on these experiments is therefore suspect, and the suspicion is deepened by the fact that the colour is not restored to Schiff's reagent unless a specially sensitive reagent* is used. To account for these facts Tollens suggested 1,4- and 1,6-oxide ring formulae; it should be added that a 1,2-oxide ring had been suggested as early as 1870 by A. Colley.† It will be seen later that this matter of ring formation must be taken into account, but since sugar chemistry made remarkable progress at the end of the nineteenth century on the basis of the open-chain formulae and because certain stereochemical complications are necessarily introduced on ring closure, we shall continue for the present to consider only the acyclic formulae. There is in fact no real inconsistency in this for although the amount of the acyclic form may be very small, a proportion is always present, in aqueous solution, in equilibrium with the ring forms.

Fructose as a Pentahydroxy Ketone

Kiliani,‡ by methods similar to those already outlined for glucose, converted fructose into 2-methyl *n*-caproic acid, indicating that fructose is a 2-ketohexose.



Fructose

Fructose cyanhydrin

2-Methyl *n*-caproic acid

Sugar Osazones

Emil Fischer's introduction§ of phenylhydrazine as a reagent in sugar chemistry is one of the milestones in the development of the

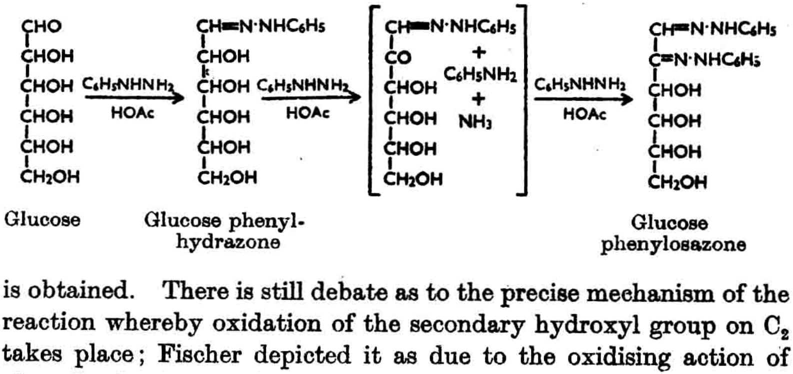
---

* Tobie, *Ind. Eng. Chem.* (Anal.), 1942, **14**, 405.
† *Compt. rend.*, 1870, **70**, 403.
‡ *Ber.*, 1885, **18**, 3066.
§ *Ber.*, 1884, **17**, 579.

subject. Sugars are notoriously difficult to crystallise and their melting points are frequently indefinite; in the phenylosazones, however, Fischer found a means of identifying the sugars by the isolation of bright yellow products usually of characteristic crystalline form. It may perhaps be mentioned in passing that although compared with the parent sugars the osazones are superior, most modern workers would agree that these compounds, since they are difficult to purify, are only used as a last resort for purposes of identification, and the osotriazoles described by C. S. Hudson obtained by oxidising the osazones with copper sulphate* are to be preferred.

Fischer's great success with the osazones, however, was in their use as intermediaries in transformation reactions and their employment in his stereochemical proofs.

Taking glucose as the starting material, the first product of the reaction of phenylhydrazine acetate is glucose phenylhydrazone; this is then oxidised to a 2-keto derivative from which, by condensation with a further molecule of phenylhydrazine, glucose phenylosazone is obtained. There is still debate as to the precise mechanism of the reaction whereby oxidation of the secondary hydroxyl group on C_2 takes place; Fischer depicted it as due to the oxidising action of phenylhydrazine itself (which is converted to aniline and ammonia), but it is well known that the free base is normally a reducing agent. Kenner and Knight† have brought forward evidence that the phenylhydrazonium ion $[C_6H_5NHNH_3]^+$ is the effective oxidising agent, but other explanations have been advanced;‡ Weygand§ considers



Glucose

Glucose phenylhydrazone

Glucose phenylosazone

---

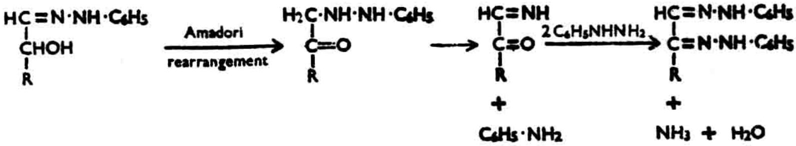
* Hann and Hudson, *J. Amer. Chem. Soc.*, 1944, **66**, 735; Hudson, *J. Org. Chem.*, 1944, **9**, 470; *J. Amer. Chem. Soc.*, 1947, **69**, 1461; 1948, **70**, 2288.

† *Ber.*, 1936, **69**, 341.

‡ See *Advances in Carbohydrate Chemistry*, 1948, **3**, 23, Academic Press, New York.

§ Weygand *et al.*, *Ber.*, 1940, **73**, 1284; 1949, **82**, 438; 1958, **91**, 1576.

that the phenylhydrazone is formed first and that this rearranges according to the following shortened scheme:

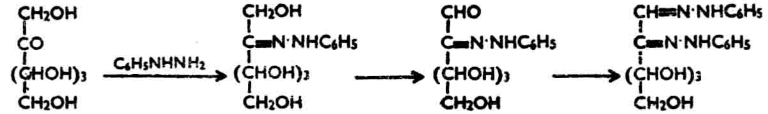


Since it has been pointed out[*] that β-alkyl substituted phenylhydrazines are readily oxidised in air to hydrazones:

$$RCH_2NHNHC_6H_5 \xrightarrow{air} R.CH{=}N.NHC_6H_5$$

an oxidising agent is no longer required; all that is necessary is a hydrogen acceptor such as the hydrazonium cation, which is then reductively cleaved to aniline and ammonia. On these grounds a mechanism similar to that formulated above but involving the hydrazonium ion seems inherently probable.

Ketoses also form phenylosazones, in fact fructose and phenylhydrazine react much more rapidly than glucose and the reagent, although the *same* osazone is produced. In this case it is the primary alcoholic group on C_1 which is oxidised to give an aldehydohydrazone.



Fructose

Glucose
phenylosazone

The fact that glucose and fructose give the same osazone is important since it proves that, apart from the arrangements on carbon atoms 1 and 2, glucose and fructose must have identical structures. The osazone reaction is of course not confined to the use of phenylhydrazine itself; many substituted osazones, for example *p*-bromophenylosazones, tolylosazones, etc., have been prepared for identification purposes. C. Neuberg[†] introduced α-methylphenylhydrazine [$(C_6H_5N(CH_3).NH_2$] as a reagent for detecting ketoses, since it

---

[*] Barry and Mitchell, *Nature*, 1955, **175**, 220.
[†] *Ber.*, 1904, **37**, 4616.