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REVIEW ARTICLE

The contribution of Franz Schardinger to cyclodextrins: a tribute on the occasion of the centenary of his death

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Abstract

For nearly 130 years, cyclodextrins, cyclic oligosaccharides obtained by enzymatic degradation of starch, have continued to fascinate researchers and the industrial world. These substances are remarkable macrocyclic molecules with significant theoretical and practical impacts in chemistry, biology, biochemistry, health science and agriculture. Amongst the list of prestigious researchers who have contributed to the fundamental knowledge of cyclodextrins, Franz Schardinger has played an important role. Schardinger was the first to describe their particular properties and lay the foundations for their chemistry. Indeed, he is known as the "Founding Father" of cyclodextrins. The purpose of this historical mini-review, on the occasion of the centenary of his death, is to commemorate his contribution to cyclodextrin chemistry.

Graphic abstract

Keywords Schardinger · Dextrins · Cyclodextrins · Tribute

Introduction

Cyclodextrins, synthetic substances obtained from the enzymatic degradation of starch, belong to the family of cage molecules. The core of their structure is composed of a hydrophobic cavity that can encapsulate other molecules

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[1]. This is at the origin of many industrial applications, e.g. pharmaceuticals, cosmetics, hygiene and toiletries, medical, food, biotechnology, agrochemistry, catalysis, packaging, textile industry, and environmental domains. For nearly 130 years, cyclodextrins have continued to fascinate researchers and the industrial world from practical and academic point of view [2–11].

The history of cyclodextrins began in France in the late nineteenth century with the work of the pharmacist and chemist Antoine Villiers (1854–1932) on the action of enzymes on various carbohydrates. In 1891, Villiers, studying the action of butyric ferment *Bacillus amylobacter* on potato starch, was the first to observe the formation of unwanted crystals with particular properties, which he named "*cellulosines*", due to the similarities with cellulose [12–15]. Although Villiers discovered cyclodextrins, their recognition is mainly attributed to Franz Schardinger (1853–1920), an Austrian chemist and bacteriologist, who was the first Great Scientist to have marked the history of these oligosaccharides, laying the foundations for their chemistry. Indeed, since the 1940s, Schardinger has been recognized as the "Founding Father" of cyclodextrins [16].

Schardinger's obituary is rare. Two short obituary notices were published in 1920 in German in the journals *Zeitschrift für angewandte Chemie* (Fig. 1) and *Chemiker Zeitung* [17]. An English translation can be found in French's review published in 1957 [18]. Schardinger was Chief Inspector of the *Untersuchungsanstalt für Lebensmittel* in Vienna. He was a member of the famous German-Austrian School of chemistry and biochemistry working on carbohydrates, saccharides and other natural substances, including sugars, starch, cellulose and chitin, at the beginning of the twentieth century. Schardinger has worked on many academic and industrial topics, the main one being heat-resistant micro-organisms that could cause food poisoning [1].

At the beginning of the last century [19–27], Schardinger discovered that a type of microorganism extremely resistant to heat was capable of dissolving potato starch and forming crystalline by-products/products remarkably similar to the *cellulosines* reported by Villiers. Part of the product formed, about 25–30% of the starch, was crystalline, the rest being amorphous and gum-like. Schardinger has obtained two "interesting" products that he studied in detail and a byproduct to which he initially pays less attention. He observed that, when chloroform or ether were used as precipitants, the two products were precipitated from the fermenting liquid. The resulting "crystalline slush"/slime was dissolved in a "little" amount of water and boiled to remove precipitants. The first precipitate was a "mud" (later the term "heavy sludge" was used) of a fine amorphous powder that was difficult to filter, whilst by concentrating the filtrate "*ein schönes kristallisierte Substanz B*" (a beautiful crystalline product, i.e. β-dextrin) was obtained. However, the main product, "*kristallisierte Substanz A*" (α-dextrin) was obtained from the filtrate by precipitation with alcohol. Schardinger first named his *krystallisiertes dextrins* crystalline dextrin A and crystalline dextrin B, and later crystalline dextrin-α and crystalline dextrin-β, while the first precipitate was considered as a byproduct. The B form resembled Villiers' *cellulosine*. Later, another crystalline dextrin was obtained from the slime by crystallizing it from dilute alcohol, followed by purification. For eight years, between 1904 and 1911, Schardinger studied the two *krystallisiertes dextrins*/cyclodextrins in detail, from their isolation and purification to their composition and physical and chemical properties.

Schardinger was the first researcher to introduce the terms dextrin-α and dextrin-β (renamed later as Schardinger alpha-dextrin/α-cyclodextrin/cyclohexaamylose and Schardinger beta-dextrin/β-cyclodextrin/cycloheptaamylose, respectively), to isolate the strain of bacteria responsible for the degradation of starch into dextrins, to describe their fundamental properties, to hypothesize that the *krystallisiertes dextrins* were cyclic "polysaccharides", and also to suggest their ability to form complexes [19–27]. His major discovery was undoubtedly to have isolated the microorganism capable of synthesizing the enzyme that catalyzes the degradation of starch into cyclodextrins. Schardinger first found the new species in a retting pit and thought it was a retting bacteria. That's why he named it *Bacillus macerans*. In this species, Schardinger also discovered the first known microorganism capable of forming acetone.

The purpose of this historical mini-review, on the occasion of the centenary of his death, is to commemorate his contribution to the foundations of cyclodextrin chemistry, their preparation, isolation and characterization.


Wirtschaftlicher Teil] 33. Jahrgang 1920 Chemisch-wirtschaftliche Nachrichten 393

Zeitschrift für angewandte Chemie

Bd. II, S. 393—396 | Wirtschaftlicher Teil u. Vereinsnachrichten | 12. Oktober 1920

Regierungsrat Dr. Fr. Schardinger, Chemiker u. Bakteriologe, Oberinspektor i. R. der Untersuchungsanstalt f. Lebensmittel in Wien, der durch die Entdeckung des Linksmilchsäurebacillus, durch die Methylenblaureaktion zur Unterscheidung der ungekochten von gekochter Milch, durch Forschungen über Acetongärung und über krystallisierte Abbauprodukte der Stärke der Wissenschaft bedeutende Dienste geleistet hat, in Innsbruck im Alter von 67 Jahren.

Fig. 1 Brief obituary notice of Franz Schardinger published in the journal *Zeitschrift für angewandte Chemie* in 1920

Preparation and isolation of cyclodextrins

Schardinger's research

Franz Schardinger is the founder of several research fields in biochemistry and chemistry, mainly in bacteriology

and carbohydrate chemistry. He worked on many research themes, including fermentative production of levorotatory lactic acid or acetone [28], water contamination [29, 30], the reducing power of milk [31], the action of enzyme from milk on fermentation [31], food spoilage [32, 33], heat-resistant microorganisms that played a role in the deterioration of foods [32, 33], and chemistry and biochemistry of starches and their degradation [34].

In the 1890s, Schardinger began studying drinking water for the presence of *Bacterium coli* (now *Escherichia coli*) and, in 1892, he proposed a water analysis test for its detection [28]. Over time, the test was extended first to dairy products, then to food products [35], and food processors began to use indicator testing as a means of determining the adequacy of a process. Schardinger also discovered the reducing power of milk. He had originally found that fresh cow's milk reduced methylene blue in the presence of formaldehyde but not in its absence and not if the milk had previously been boiled. Indeed, Schardinger observed that milk contained a heat-labile factor that catalyzed the rapid destruction of methylene blue when an aldehyde was added as substrate. In 1902, Schardinger isolated *Bacillus lactis* aerogenes from unclean drinking water and studied at length its action on milk and its slime production. He noted its property of making milk slimy. Later, Schardinger confirmed that the bovine mill contained an enzyme capable of oxidizing aldehydes to acids and hypothesized that the reducing action of milk on the discoloration of methylene blue was not due to bacteria but to a reducing enzyme [32, 33]. This was demonstrated 10 years later with Bach's works. This enzyme, involved in the oxidation of this and other aldehydes [36], was then commonly referred to "Schardinger enzyme" for many years to pay tribute to his work. The same year, Schardinger reported the presence of galactan polysaccharides in bacterial slime. In 1905, he discovered that acetone can be produced by biological means (fermentation) [37]. In *Bacillus macerans*, Schardinger discovered the first known microorganism capable of forming acetone and the production of acetone by fermentation was commercially exploited. On the basis of his work, later, other species were also been studied.

Like many authors, Schardinger also aimed to improve the understanding of dextrins, substances discovered in 1821 [37]. At that time, dextrins had a fascination for (carbohydrate) chemists and biochemists in that they had markedly different properties from those of other saccharides and carbohydrates. At the beginning of the last century, there was a confusion about their preparation, nomenclature, and composition. There were also doubts and debates about whether dextrins were true components of starch. For Schardinger, dextrins were first (partial) degradation products [38] of starch obtained by heating and later intermediate decomposition products [39], while Villiers considered these novel substances as isomers of starch [12–15]. For Pringsheim, they were as the intermediate products ("scission products") formed in the course of the hydrolysis of starch to maltose and glucose by diastatic enzymes [1]. Moreover, at that time, the structure of dextrins had not yet been established (it will be determined by Freudenberg in the mid-1930s), nor that of starch, chitin or cellulose. In addition, it was not clearly established that starch was a macromolecule composed of several residues of "glucose".

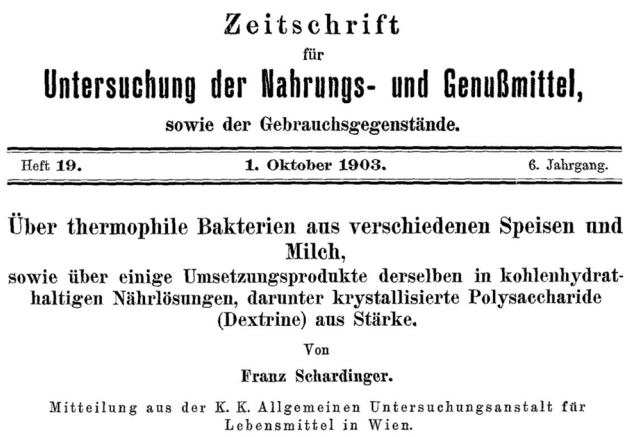
At the time, the ability of certain bacteria and fungi to produce starch hydrolyzing enzymes was well known, to such an extent that these processes were used for various commercial purposes where diastatic enzymes were required. Thanks to the work of Villiers, however, it was known that some bacteria were able to break down starches with the formation of acids, alcohols, and acetone. In this, the action of bacteria upon starch was different from that of the diastatic enzymes which produced 100 per cent maltose, and acid hydrolysis of starch, which resulted in the formation of glucose. Schardinger will clearly demonstrate this [40].

The Schardinger dextrins

In 1903, Schardinger, in the course of his investigations on micro-organisms that can lead to food poisoning, observed that a type of microorganism extremely resistant to heat could dissolve starch to form crystalline by-products, called "*krystallisiertes dextrins*" [34]. He published his results in the journal *Zeitschrrift für Untersuchuing von Nahrungs- und Genussmittel* (Fig. 2).

Using an iodine microscopic test, Schardinger distinguished two types of "*krystallisiertes dextrins*", referred as "*krystallisierte Substanz A*" and "*krystallisierte Substanz B*", as most of their properties were similar to the already know partial degradation products of starch, i.e. the dextrins [35]. "*Krystallisiertes dextrins*" were first considered as the degradation byproducts of starch through heating [35]. Schardinger isolated larger quantities of crystalline dextrin A than crystalline dextrin B, the latter being more surprising. In this seminal publication, Schardinger also indicated that the B form resembled Villiers' "*cellulosine*". Indeed, the chemical behavior and the physical constants given by Schardinger for his substance agree very well with those of the dextrin previously described by Villiers [12–15]. Schardinger found that it was possible to isolate pure fractions with a maximum yield of 30% crystalline dextrins from starch, the main form obtained being always dextrin A.

In 1904, Schardinger isolated a heat-resistant organism with considerable starch-fermenting power [36]. To monitor the progression of enzyme action on starch, Schardinger proposed a simple microscopic iodine test based on the observation that the "*krystallisiertes dextrins*" formed characteristic crystalline compounds with iodine [36]. This



Zeitschrift
für
Untersuchung der Nahrungs- und Genußmittel,
sowie der Gebrauchsgegenstände.

Heft 19. 1. Oktober 1903. 6. Jahrgang.

Über thermophile Bakterien aus verschiedenen Speisen und Milch,
sowie über einige Umsetzungsprodukte derselben in kohlenhydrathaltigen Nährlösungen, darunter krystallisierte Polysaccharide (Dextrine) aus Stärke.

Von

Franz Schardinger.

Mitteilung aus der K. K. Allgemeinen Untersuchungsanstalt für Lebensmittel in Wien.

Fig. 2 Extract of the first page of the article published in the journal *Zeitschrrift für Untersuchuing von Nahrungs- und Genussmittel* by Schardinger on dextrins in 1903

test was inspired by the work of Colin and de Claubry, who were the first to mention in 1814 the formation of a violet to blue–black substance produced by the action of iodine on starch [41]. This product was soluble in water and gave an intensely blue solution which was rapidly decolorized by boiling, as reported by von Stromeyer a year later [42]. As dextrins had a particular structure, it was conceivable that their reaction with iodine was similar to that of starch with iodine.

One year later, Schardinger confirmed the simplicity and usefulness of the iodine test to characterize his "*krystallisiertes dextrins*". During the course of the action of the enzyme upon starch, there was no significant change in reducing power or optical rotation. The dextrins, considered as the intermediate decomposition products of starch, were then designated by the term "*krystallisiertes polysaccharides*", i.e. crystallized/crystalline polysaccharides [37].

Continuing his investigations on the structure of starch, Schardinger hypothesized in 1907 that "*krystallisiertes polysaccharides*" were true components of the starch "molecule". Repeating the experiments on the action *Bacillus macerans* on a starch suspension (by varying the source of the starch), he again succeeded in isolating three fractions of his fermentation mixture: a soluble fraction called dextrin alpha (dextrin A), a slightly soluble fraction called dextrin beta (dextrin B), and an insoluble residue called "*Schlamm*" [38].

One year later, Schardinger introduced a distinction between a "*crystallized amylose*" for dextrin A and a "*crystallized amylodextrin*" for dextrin B, because, for him, there was an analogy between his dextrins and amylose and amylodextrin, especially with respect to their iodine color-reactions [39]. In 1909, he confirmed that the cultivation of *Aerobacillus macerans* upon starch solutions produced a mixture of water-soluble dextrins, from which two distinct, non-reducing, crystalline compounds (*crystallized amylose* and *crystallized amylodextrin*) might be readily isolated. These dextrins were closely related to starch components [39]. Finally, two years later, Schardinger considered that these names were inappropriate, and thus decided to rename it "*krystallisierte dextrin*-α" and "*krystallisierte dextrin*-β" [18].

All the Schardinger's results on dextrins were summarized in a book on polysaccharides published in 1931 by Pringsheim [19] and later discussed by French [18] in the first comprehensive review entitled The Schardinger Dextrins.

Bacillus macerans

Between 1902 and 1903, Schardinger observed that dextrin B/*cellulosine* was systematically formed in starch-based media containing putrefying microorganisms [34], regardless of the starch source used. He then tried to isolate the strain of bacteria responsible for the degradation of starch into crystallized dextrins. In 1903, Schardinger isolated a thermophilic microorganism capable of synthesizing the enzyme that catalyzed the reaction, twelve years after the first publication of Villiers. Schardinger called it *strain II* [35]. This heat-resistant organism had considerable starch-fermenting power. Schardinger observed that, in starch subculture, *strain II* decomposed the starch, giving an alcohol-insoluble "soluble starch" together with "*krystallisierte dextrin*" A (fine hexagonal plates) and "*krystallisierte dextrin*" B (stout prismatic crystals). However, he also observed that with time, the activity of the *strain II* microorganism decreased. Indeed, Schardinger was unsuccessful in maintaining a culture of *strain II* which had the characteristic starch-degrading activity.

Several months later, Schardinger found a similar action in cultures of another isolate from starch. He isolated a new microorganism, considered as "an accidental contaminant", which he first called *Rottebacillus I* owing to its action on potato starch, *i.e.* it produced acetone and ethyl alcohol by fermentation of carbohydrate media. The name *Rottebacillus I* was used to express the fact the microorganism was able to form both acetone and ethyl alcohol.

In 1904, Schardinger published a second seminal publication in the journal *Wiener Klinische Wochenschrift* where he used the Latin term *Bacillus macerans* to name his microbe, i.e. *macerare*, to rot (Fig. 3) [36]. His first interest in this macerans organism had been as "*ein aceton bildender Rottebacillus*", and that is why he had recorded bacteriological studies on the pure culture: its occurrence, morphology, nutrition, and acetone production [37].

Subsequently, Schardinger turned to the study of the unique crystalline dextrins formed by this organism from

starchy media. By inoculating *Bacillus macerans* with a starch, he again found that the starch was decomposed. By evaporating and extracting the residue with alcohol, Schardinger obtained in the alcoholic extract crystalline dextrins of the same nature (elemental composition, specific rotation, properties, complexes) as those he had previously described. Pursuing the experiments, he showed that the crystalline dextrins formed from wheat, rice, potato, and arrowroot starches were always the same. In particular, all products formed had the same characteristics with iodine reaction and were halfway between maltose and starch. This was discussed in 1957 by French [18].

Schardinger then hypothesized that the two "*krystallisiertes polysaccharides*" were the intermediate crystallized decomposition products of starch, i.e. amylose and amylodextrin, or closely related to these true components [39]. They could be of considerable theoretical interest for the study of the starch "molecule", and to extent that they formed crystalline iodine addition products, they could be used to shed some light on the starch-iodine reaction. Schardinger highlighted the analogy between his A and B dextrins and amylose and amylodextrin, that he proposed the names "*krystallisiertes amylose*" and "*krystallisiertes amylodextrin*".

Finally, in 1911, Schardinger confirmed that *Bacillus macerans* was able to give the same crystalline dextrins as before, and these products were true components of starch. The same year, Schardinger developed a procedure for the isolation of two different crystalline dextrins (dextrin-α and dextrin-β), and characterized each by specific rotation, elemental analysis, and their unique crystalline iodine complexes [40]. It is important to note that at the time, it was still doubtful that dextrins were products of the synthetic metabolism of the organism. It was only with the work of the group of Hudson [43–47] in the late 1930s that this last hypothesis was abandoned, and the first, proposed by Schardinger, was demonstrated.

The fact that the cultures of *B. macerans* contained an enzyme that converted gelatinized starch into a highly rotating mixture of non-reducing dextrins, without production of maltose, glucose or other reducing sugars (Schardinger's crystalline dextrin-α and dextrin-β were then starch components), was demonstrated by Freudenberg [48–50] in the late 1930s and by French [51–53] in the 1940s.

Following Schardinger's early work on the action of *Bacillus macerans* on starches and Villiers with *Bacillus amylobacter* on potato starch, little interest has been shown for *Bacillus macerans* in its ability to produce dextrins in the years following their work. It was only in the 1940s that researchers became interest in this bacillus again for the production of cyclodextrins/cycloamyloses [54–61]. However, since the 1920s, many researchers have studied the organism as a fermentation source of ethyl alcohol and acetone [62–66].

Bacillus macerans, ein Aceton bildender Rottebacillus.

[Mitteilung aus der k. k. Untersuchungsanstalt für Lebensmittel in Wien.]

Von **Franz Schardinger.**

Ueber die durch den genannten Mikroben bewirkte Acetongärung wurde bereits in No. 8 der Wiener klinischen Wochenschrift, Jahrg. 1904 berichtet. Die Ergebnisse fortgesetzter Studien über diese Gärung und die Verrottung pflanzlicher Gebilde sollen im nachfolgenden einem weiteren Leserkreise unterbreitet werden.

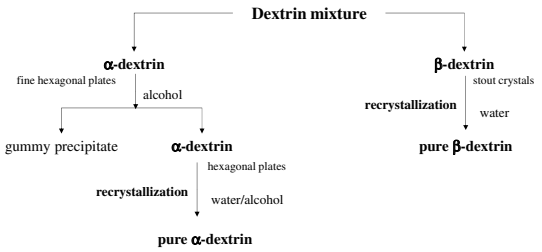
Fig. 3 Abstract of the article published in the journal *Wiener Klinische Wochenschrift* by Schardinger on *Bacillus macerans* in 1904

Isolation of the crystalline dextrins

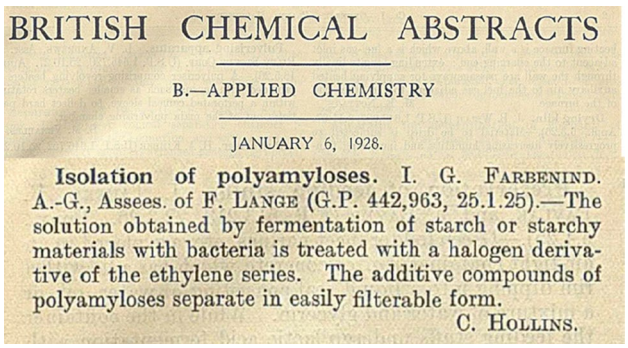
Schardinger was not convinced by Villiers' conclusions on the reaction yields obtained for the two *cellulosines*/dextrins. In his early experiments [36], for its dextrin alpha and dextrin beta, the yields obtained were ten times higher than those of Villiers. To explain this "*dieses interessante Ergebnis*" (interesting result), Schardinger suggested that, in the conditions of sterilization described by Villiers, the bacillus used was "*wahrscheinlich unrein*" (probably not pure).

Between 1905 and 1911, Schardinger studied the preparation, fractionation/separation, and purification of the *cellulosines/krystallisierte dextrins*, with the objective of preparing pure products with high yields. In 1905, Schardinger pointed out the difficulty of isolating the two dextrins A and B, the problems of selective precipitation and the different solubilities [37]. Two years later, he described a first protocol for the preparation of dextrins with high purity. The *krystallisierte α-dextrin* and *krystallisierte β-dextrin* were synthesized from several sources of starch, e.g. potatoes, rice and wheat, and bacteria. About 25–30% of the starch was converted to crystalline dextrins depending on these parameters [38]. However, yields were always 10 times higher than those reported by Villiers [13, 14].

The method of separation proposed by Schardinger was based on the ease of crystallization of the *krystallisierte β-dextrin* from water and its low solubility, about 1.5% at room temperature, followed by precipitation of the *krystallisierte α-dextrin* from the mother liquor by the addition of alcohol. During four years, this method was studied and modified [39]. In 1911, Schardinger published the first fractionation and purification protocol for the production of α-dextrin and β-dextrin with high purity (Scheme 1) but yields remained low [40]. This protocol has been modified by Lange [67] in 1925 (Fig. 4) who introduced trichloroethylene as a precipitating agent for the crystalline dextrins. However, yields were yet low. It was not until 1935 and Freudenberg's work that the first protocol were developed to obtain almost pure dextrins with high yields [68]. This procedure was based not only on solubility differences of the



Scheme 1 The first fractionation and purification protocol proposed by Schardinger in 1911 for the production of dextrins with high purity



BRITISH CHEMICAL ABSTRACTS

B.—APPLIED CHEMISTRY

JANUARY 6, 1928.

Isolation of polyamyloses. I. G. FARBENIND. A.-G., Assees. of F. LANGE (G.P. 442,963, 25.1.25).—The solution obtained by fermentation of starch or starchy materials with bacteria is treated with a halogen derivative of the ethylene series. The additive compounds of polyamyloses separate in easily filterable form.

C. HOLLINS.

Fig. 4 Extract of the British Chemical Abstracts announcing the first patent, entitled "*Verfahren zur gewinnung von polyamylosen*", on polyamyloses/dextrins filed by Fritz Lange in 1925

dextrins themselves, as proposed by Schardinger, but also on the differences in solubilities and rates of crystallization of their acetates. In the protocols proposed by Lange and Freudenberg, the distinction between the two products has always been made by their ability to form complexes of different colors with iodine, as Schardinger already suggested.

Nomenclature

As early as 1903, Schardinger suggested that "*krystallisiertes dextrins*" (dextrin alpha and dextrin beta) was a better name than "*cellulosines*". Later, he used the term "*krystallisiertes polysaccharides*" and finally, in 1911, he changed the names to "*krystallisierte dextrin-α*" and "*krystallisierte dextrin-β*".

A major controversy then developed over the nomenclature of dextrins [1]. For instance, Pringsheim first suggested the name of "amyloses" for this new group of sugars, of which the dextrin-α and dextrin-β of Schardinger were a diamylose and a triamylose, respectively. Later, Pringsheim preferred to call them "*krystallisiertes polyamylosen*", crystalline polyamyloses, considering that "these substances were the basic units of the starch molecule". Pringsheim introduced two series: the α-series of dextrins obtained from the amylose fraction of starch (four distinct substances) and the β-series from amylopectin fraction degradation (two distinct products, hexaamylose). For him, the "*Schlamm*" substance consisted of either three or four diamylose substances. However, at that time, Karrer disagreed with the nomenclature proposed by Pringsheim. Karrer regarded maltose as the fundamental unit of the whole of the starch molecule ("starch is a polymeric maltose anhydride"). The confusion that results from the terminology of the different compounds lasted for more than 20 years [1].

In the 1930s, the German chemist Karl Johann Freudenberg (1886–1983) renamed more simply the two *krystallisiertes dextrins* into α-dextrin and β-dextrin, and this terminology is finally adopted. Freudenberg also proposed to call them "Schardinger dextrins" in recognition of the fact that Schardinger had first described their preparation, isolation and properties with reliable details and first identified *Bacillus macerans*, the strain responsible for their formation [48–50]. This recognition was probably first suggested in 1935 [1].

For many years, cyclodextrins were called "Schardinger dextrins" in his honor, almost up to the 1970s. Schardinger dextrins were subsequently named cycloamyloses [69] by the American chemist Dexter French (1918–1981) in 1942, cycloglucanes [70–72] by Freudenberg in 1943, and finally cyclodextrins [73] in 1949 by the German chemist and biochemist Friedrich Cramer (1923–2003), a pupil of Freudenberg.

The foundations of cyclodextrin chemistry

Chemical properties

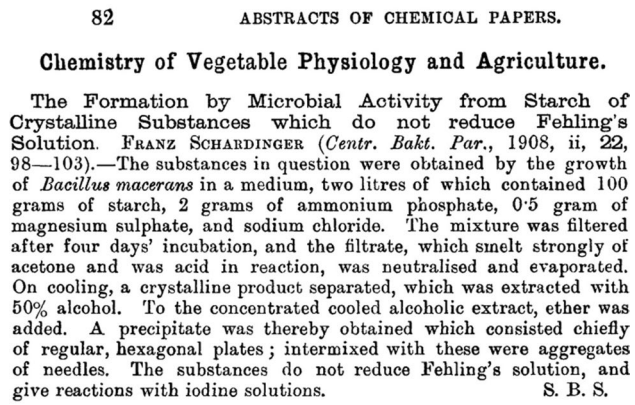
Between 1904 and 1911, Schardinger made several important observations from a chemical point view. In 1904, he observed that *cellulosines*/dextrins were often formed in starch-based media containing putrefying microorganisms [36]. The formation of the two "*krystallisiertes dextrins*" depended on the type of bacteria digesting starch. The products were well crystallized from water and aqueous alcohols, and quite resistant to acid hydrolysis, in accordance with Villiers' observations. They lose alcohol of crystallization without any important change in their physical properties. Like Villiers, Schardinger observed that, in air, the crystals became opaque. The crystals also showed high optical activity, much higher than those of certain dextrins. Schardinger stated that, by using the characteristic reaction that starch derivatives showed with iodine, it was easy to distinguish the two dextrins. Iodine colored dextrins that had high optical activity, and the intensity of coloration decreased with optical activity.

In 1905, Schardinger confirmed that a simple microscopic iodine test showed that the "*krystallisiertes dextrins*" formed

characteristic crystalline compounds with iodine [37]. In control tests made upon digests of starch with other amylases (*Aerobacillus polymyxa*), from which the main products were reducing substances, the crystalline iodine solids were not formed. In addition, Schardinger observed that the microscopic appearances were entirely different from those observed in the case of the *macerans* digests. The iodine test appeared to be specific for the products formed.

The same year, Schardinger was also the first to observe that different starchy substrates differed in their behavior with *Bacillus macerans*, especially in the yields obtained [37]. He indicated for the first time, the fact that the two forms of dextrins are able to form complexes of different colors with iodine [38]. He again pointed out their lack of reducing power and hydrolysis to reducing sugar. In 1908, Schardinger reported the formation by microbial activity from starch of crystalline substances, which did not reduce Fehling's solution (Fig. 5). One year later, he confirmed that the *krystallisierte dextrin-α* and *krystallisierte dextrin-β* were not only non-reducing to copper reagents but also non-fermentable by yeast [39]. The two dextrins were however soluble in alcohol and they could be converted into ethers under the action of acid chlorides.

In 1911, Schardinger demonstrated that, regardless of the starch source, the main product, dextrin-α, was characterized by a relatively high solubility in water and by the formation of crystallized greenish needles on the addition of iodine. The second product, dextrin-β, was much less soluble in water and it formed reddish brown crystals on the addition of iodine [40]. However, the solubility of dextrin-β raised with temperature. These characteristics were confirmed by Pringsheim [74–79] in 1924 and by Freudenberg [68] in 1935, but only demonstrated by French [69] in 1942, when these authors attempted to determine the structure of the dextrins. French also demonstrated that the crystalline



82 ABSTRACTS OF CHEMICAL PAPERS.

Chemistry of Vegetable Physiology and Agriculture.

The Formation by Microbial Activity from Starch of Crystalline Substances which do not reduce Fehling's Solution. Franz Schardinger (*Centr. Bakt. Par.*, 1908, ii, 22, 98—103).—The substances in question were obtained by the growth of *Bacillus macerans* in a medium, two litres of which contained 100 grams of starch, 2 grams of ammonium phosphate, 0·5 gram of magnesium sulphate, and sodium chloride. The mixture was filtered after four days' incubation, and the filtrate, which smelt strongly of acetone and was acid in reaction, was neutralised and evaporated. On cooling, a crystalline product separated, which was extracted with 50% alcohol. To the concentrated cooled alcoholic extract, ether was added. A precipitate was thereby obtained which consisted chiefly of regular, hexagonal plates; intermixed with these were aggregates of needles. The substances do not reduce Fehling's solution, and give reactions with iodine solutions. S. B. S.

Fig. 5 Extract of the American Chemical Abstracts announcing innovative Schardinger's results on dextrins in 1908 and published a year later in the journal *Centralblatt für Bakteriologie Parasitenkunde und Infektionskrankheiten*

complexes with iodine-iodide resembled in many respects the starch-iodine complexes.

It is important to note that the chemistry and the techniques originally developed by Schardinger have been extended and perfected by Freudenberg [48–50] and French [51–53] in the 1930s and 1940s respectively, to whom much of our present knowledge of the Schardinger dextrins/cycloamyloses/cyclodextrins can be attributed.

First concepts in terms of chemical structure and inclusion properties

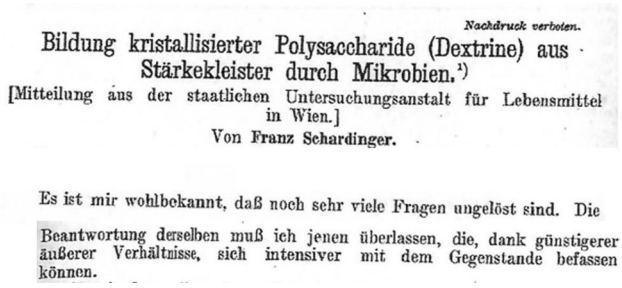
In 1903, Schardinger was able to isolate only small amounts of dextrin A, but this amount was much larger than that of dextrin B which he identified as Villiers' *cellulosine* and tried to determine its composition. Schardinger proposed for his *krystallisierte dextrin* an empirical formulae $[(C_6H_{10}O_5)_2 + 3H_2O]$, similar to that of Villiers. However, Schardinger did not attempt their molecular-weight determinations. This was only determined in 1942 by French [69] using X-ray data.

When purified by fractional precipitation, the crystals presented different optical rotation properties and were difficult to hydrolyze any further. By varying the solvent or the conditions of crystallization, it was also found possible to obtain different types of crystal forms (patterns) but these were not investigated by Schardinger. It was not until 1933 that Ulmann's group examined the 10 different forms in which alpha dextrin crystallized using X-ray powder diffraction [80].

Schardinger also hypothesized that the *krystallisiertes dextrins* were "cyclic polysaccharides" but did not propose a structure for dextrins. It will take another twenty-years before the cyclic nature of Schardinger's dextrins was clearly identified; first hypothesized in 1935 [68] and then demonstrated in 1948 [81, 82].

A distinctive character of the dextrins which facilitated their separation from crude starch digests was their ability to from crystalline insoluble complexes with many liquids. In 1911, Schardinger reported the behavior of the dextrin-α and dextrin-β in the presence of alcohols, chloroform, ether, and iodine solution. He used the complexes with these solvents as a means of selective precipitation of dextrins [40]. This was also the first indication of the ability of dextrins to form "inclusion" complexes [1]. This remarkable property was not put forward until the 1950s with Cramer's work [74–79]. The observations on complexes reported by Schardinger were later used by other researchers, particularly when they looked for other precipitants for the isolation of dextrins or when they collected data on their solubility in water and solvents.

Schardinger decided to stop his research into dextrins in 1911 [40], and as a conclusion he wrote: "I realize that still



Nachdruck verboten.

Bildung kristallisierter Polysaccharide (Dextrine) aus Stärkekleister durch Mikrobien.[1])

[Mitteilung aus der staatlichen Untersuchungsanstalt für Lebensmittel in Wien.]

Von Franz Schardinger.

Es ist mir wohlbekannt, daß noch sehr viele Fragen ungelöst sind. Die Beantwortung derselben muß ich jenen überlassen, die, dank günstigerer äußerer Verhältnisse, sich intensiver mit dem Gegenstande befassen können.

Fig. 6 Extract of the last paper published in the journal *Centralblatt für Bakteriologie Parasitenkunde und Infektionskrankheiten* in 1911 by Schardinger

very many questions remain unsolved; the answer to these I must leave to another, who, owing to more favorable external conditions, can deal with the subject more intensively (Fig. 6)."

In the 24 years following Schardinger's final paper [40], little real progress has been made in investigations on crystalline dextrins, their structure and method of preparation, although the German chemist and biochemist Hans Pringsheim (1876–1940) [83–85] and the Swiss chemist Paul Karrer (1889–1971) [86–89] are recognized as the two first researchers to have published extensively on dextrins [1]. Indeed, their works were repetitive, marred by frequently contradictory results and by even hot debate between the two groups [90]. A major controversy then developed over the structure of dextrins.

As stated by Freudenberg [68] and later by French [18], all researchers at the time used impure products and relied too much on cryoscopic measurements of molecular weights. As Schardinger dextrins had a relatively high molecular weight and were very difficult to separate from low molecular weight impurities, the use of cryoscopic techniques led to many abnormal results and bitter disputes between groups, particularly those of Pringsheim and Karrer. As a result, data in the literature between 1911 and 1935 are now generally considered to be of little importance. The next important step in the investigation of Schardinger dextrins did not begin until 1935 [1], when concerted efforts began on the problems associated with their separation, purification and determination of their structure.

From an academic point of view, research on cyclodextrins developed increasingly exponentially from the 1940s onwards, with the work of Freudenberg, French, Cramer and József Szejtli (Hungarian chemist, 1933–2004) in the 1940s, 1950s, 1960s and 1970s, respectively. From an industrial point of view, cyclodextrins only really took off in the 1980s with the first applications in the chromatography, pharmaceutical and food industries [1]. Actually, applications are found in practically all sectors of industry [91].

Conclusion

Although, in retrospect, the discovery of cyclodextrins should be attributed to Villiers, Schardinger was the first to describe their preparation, isolation, and properties in reliable detail. Franz Schardinger is recognized as the "Founding Father" of cyclodextrins. In this mini-review, on the occasion of the centenary of his death, we commemorated his contribution to the cyclodextrin chemistry by highlighting his many results. Schardinger laid the foundation for the chemistry of cyclodextrin. His major discovery was undoubtedly to have isolated the microorganism capable of synthesizing the enzyme that catalyzes the degradation of starch into cyclodextrins, identified a few years later as cyclodextrin glucosyltransferase. In his honor, *cellulosines/krystallisiertes dextrins* were called "Schardinger dextrins" for many years, almost until the 1970s.

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Compliance with ethical standards

Conflict of interest The author declares no competing financial interest.

References

1. Crini, G.: Review: a history of cyclodextrins. Chem. Rev. **114**, 10940–10975 (2014)
2. Saenger, W.: Cyclodextrin inclusion compounds in research and industry. Angew. Chem. Int. Ed. **19**, 344–362 (1980)
3. Smolková-Keulemansová, E., Krysl, S.: Inclusion compounds in chromatography. J. Chromatogr. **184**, 347–361 (1980)
4. Hinze, W.L.: Applications of cyclodextrins in chromatographic separations and purification methods. Sep. Purif. Methods **10**, 159–237 (1981)
5. Szejtli, J.: Industrial applications of cyclodextrins. In: Atwood, J.L., Davies, J.E.D., McNicol, D.D. (eds.) Inclusion Compounds, vol. 3, pp. 331–390. Academic Press, London (1984)
6. Sybilska, D., Smolková-Keulemansová, E.: Application of inclusion compounds in chromatography. In: Atwood, J.L., Davies, J.E.D., McNicol, D.D. (eds.) Inclusion Compounds, vol. 3, pp. 173–243. Academic Press, London (1984)
7. Bender, H.: Production, characterization and applications of cyclodextrins. In: Liss, A.R. (ed.) Advances in Biotechnological Processes, Chap 6, pp. 31–71. Wiley, New York (1986)
8. Li, S., Purdy, W.C.: Cyclodextrins and their applications in analytical chemistry. Chem. Rev. **92**, 1457–1470 (1992)

9. Wenz, G.: Cyclodextrins as building blocks for supramolecular structures and functional units. Angew. Chem. Int. Ed. **33**, 803–822 (1994)
10. Hedges, A.R.: Industrial applications of cyclodextrins. Chem. Rev. **98**, 2035–2044 (1998)
11. Armspach, D., Gattuso, G., Koeniger, R., Stoddart, J.F.: Cyclodextrins. In: Hecht, S.M. (ed.) Bioorganic Chemistry: Carbohydrates, pp. 458–488. Oxford University Press Inc., New York (1999)
12. Villiers, A.: Sur la transformation de la fécule en dextrine par le ferment butyrique. Compte Rendus des Séances de l'Académie des Sciences **CXII**, 435–437 (1891)
13. Villiers, A.: Sur la transformation de la fécule en dextrine par le ferment butyrique. Bull. Soc. Chim. Paris **45**, 468–470 (1891)
14. Villiers, A.: Sur la fermentation de la fécule en dextrine par le ferment butyrique. Compte Rendus des Séances de l'Académie des Sciences **CXII**, 536–538 (1891)
15. Villiers, A.: Sur la fermentation de la fécule en dextrine par le ferment butyrique. Bull. Soc. Chim. Paris **46**, 470–472 (1891)
16. Szejtli, J.: Cyclodextrins and their inclusion complexes. Akadémiai Kiadó, Budapest (1982)
17. Brief obituary notices of Franz Schardinger are published in a) WirtschaftlicherTeil u. Vereinsnachrichten. Personal und Hoschshulnachrichten. Z. Angew. Chem **33**, 393–396 (1920) and b) Vom Tage. Regierungsrat Dr. Franz Schardinger. Chem. Ztg. **44**, 744 (1920). The author is indebted to Professor Benito Casu for providing these reprints.
18. French, D.: The Schardinger dextrins. In: Wolfrom, M.L. (ed.) Advances in Carbohydrate Chemistry, vol. 12, pp. 189–260. Academic Press Inc., New York (1957)
19. Pringsheim, H.: Dextrine: charakteristik, gewinnung und eigenschaften. In: Die Polysaccharide, Chapter VII. Berlin: Verlag von Julius Springer, pp. 248–274 (1931).
20. Samec, M., Blinc, M.: Die neueren ergebnisse der stärkeforschung. Kolloid Beih. **49**, 211–223 (1939)
21. Thoma, J.A., Stewart, L.: Cycloamyloses. In: Whistler, R.L., Paschall, E.F. (eds.) Starch, Chemistry and Technology, Volume 1: Fundamental Aspects, Chapter IX, pp. 209–249. Academic Press, New York (1965)
22. Caesar, G.V.: The Schardinger dextrins. In: Radley, J.A. (ed.) Starch and Its Derivatives, EC4, Fourth edition, Chapter X, pp. 290–305. Chapman and Hall Ltd., London (1968)
23. Kainuma, K.: Starch oligosaccharides: linear, branched, and cyclic. In: Whistler, R.L., BeMiller, J.N., Paschall, E.F. (eds.) Starch, Chemistry and Technology, 2nd edition, Chap V, pp. 125–152. Academic Press Ltd., London (1984)
24. Clarke, R.J., Coates, J.H., Lincoln, S.F.: Inclusion complexes of the cyclomalto-oligosaccharides (cyclodextrins). Adv. Carbohydr. Chem. Biochem. **46**, 205–249 (1988)
25. Robyt, J.F.: Cyclodextrins. In: Essentials of Carbohydrate of Chemistry, Chapter 8. New York: Springer, pp. 245–261 (1998).
26. Szejtli, J.: Introduction and general overview of cyclodextrin chemistry. Chem. Rev. **98**, 1743–1753 (1998)
27. Loftsson, T., Duchêne, D.: Cyclodextrins and their pharmaceutical applications: historical perspectives. Int. J. Pharm. **329**, 1–11 (2007)
28. Schardinger, F.: Über eine neue optisch active modification der milchsäure, durch bacterielle spaltung des rohrzuckers erhalten. Monatshefte für Chemie **11**, 545–559 (1890)
29. Schardinger, F.: Über das vorkommen gährung erregender spaltpilze im trinkwasser und ihre bedeuting für die hygienische beurtheilung desselben. Wien. Klin. Wschr. **5**, 403–405 (1892)
30. Schardinger, F.: Über das vorkommen gährung erregender spaltpilze im trinkwasser und ihre bedeuting für die hygienische beurtheilung desselben. Wien. Klin. Wschr. **5**, 421–423 (1892)
31. Schardinger, F.: Ueber das vrhalten der kuhmilch gegen methylenblau und seine verwendung zur unterscheidung von ungekochter und gekochter milch. Z. Unters. Nahr. Genussm. **5**, 1113–1121 (1902)
32. Schardinger, F.: Über die gärprodukte eines schleimbildenden Bacillus in rohrzuckerlösungen und die zusammemsetzung eines aus dem schleime isolierten. Centralbl. Bakteriol. Parasitenkd. Infektionskrankh. Hyg. Abt. II **8**, 144–147 (1902)
33. Schardinger, F.: Über die gärprodukte eines schleimbildenden Bacillus in rohrzuckerlösungen und die zusammemsetzung eines aus dem schleime isolierten. Centralbl. Bakteriol. Parasitenkd. Infektionskrankh. Hyg. Abt. II **8**, 175–180 (1902)
34. Schardinger, F.: Über thermophile bakterien aus verschiedenen speisen und milch, sowie über einige umsetzungsprodukte derselben in kohlenhydrathaltigen nährlösungen, darunter krystallisierte polysaccharide (dextrine) aus Stärke. Z. Unters. Nahr. Genussm. **6**, 865–880 (1903)
35. Schardinger, F.: Ueber die zulässigkeit des warmhaltens von zum genuβ bestimmten nahrungsmitteln mittelst wärme speichernder apparate, sog. thermophore. Wien. Klin. Wochenschr. **16**, 468–474 (1903)
36. Schardinger, F.: Mitteilung aus der staatlichen untersuchungsanstalt für lebensmittel in Wien Azetorgärung. Wien. Klin. Wochenschr. **17**, 207–209 (1904)
37. Schardinger, F.: *Bacillus macerans*, ein aceton bildender Rottebacillus. Centralbl. Bakteriol. Parasitenkd. Infektionskrankh. Hyg. Abt. II **14**, 772–781 (1905)
38. Schardinger, F.: Zur biochemie des *Bacillus macerans*. Centralbl. Bakteriol. Parasitenkd. Infektionskrankh. Hyg. Abt. II **19**, 161–163 (1907)
39. Schardinger, F.: Ueber die bildung kristallisierter fehlingsche lösung nicht reduzierender körper (polysaccharide) aus stärke durch mikrobielle tätigkeit. Centralbl. Bakteriol. Parasitenkd. Infektionskrankh. Hyg. Abt. II, **22**, 98–103 (1908–1909)
40. Schardinger, F.: Bildung kristallisierter polysaccharide (dextrine) aus särkekleister durch mikrobien. Centralbl. Bakteriol. Parasitenkd. Infektionskrankh. Hyg. Abt. II **29**, 188–197 (1911)
41. von den Colin, H.H., de Claubry, G.: Ueber die verbindungen der jodine mit den pflanzen- und den therischen körpern. Ann. Physik. **48**, 297–304 (1814)
42. von Stromeyer: Ein sehr empfindliches reagens für jodine, aufgefunden in der stärke (amidon). Ann. Physik. **49**, 146–153 (1815)
43. Tilden, E.B., Hudson, C.S.: Conversion of starch to crystalline dextrins by the action of a new type of amylase separated from cultures of *Aerobacillus macerans*. J. Am. Chem. Soc. **61**, 2900–2902 (1939)
44. Tilden, B., Hudson, C.S.: Preparation and properties of the amylases produced by *Bacillus macerans* and *Bacillus polymyxa*. J. Bacteriol. **43**, 527–544 (1942)
45. McClenahan, W.S., Tilden, E.B., Hudson, C.S.: A study of the products obtained from starch by the action of the amylase of *Bacillus macerans*. J. Am. Chem. Soc. **64**, 2139–2144 (1942)
46. Tilden, E.B., Adams, M., Hudson, C.S.: Purification of the amylase of *Bacillus macerans*. J. Am. Chem. Soc. **64**, 1432–1433 (1942)
47. Wilson, E.J., Schoch, T.J., Hudson, C.S.: The action of Macerans amylase on the fractions from Starch. J. Am. Chem. Soc. **65**, 1380–1383 (1943)
48. Freudenberg, K.: Beiträge zur cemie der stärke und anderer polysaccharide. Angew. Chemie **39**, 675–677 (1934)
49. Freudenberg, K., Rapp, W.: Zur kenntnis der stärke und der Schardinger-dextrine. Ber. Dtsch. Chem. Ges. **69**, 2041–2045 (1936)
50. Freudenberg, K., Meyer-Delius, M.: Über die Schardinger-dextrine aus stärke. Ber. Dtsch. Chem. Ges. **71**, 1596–1600 (1938)
51. French, D., Rundle, R.E.: The molecular weights of the Schardinger alpha and beta dextrins. J. Am. Chem. Soc. **64**, 1651–1653 (1942)

52. Rundle, R.E., French, D.: The configuration of starch and the starch-iodine complex. II. Optical properties of crystalline starch fractions. J. Am. Chem. Soc. **65**, 558–561 (1943)
53. Norberg, E., French, D.: Studies on the Schardinger dextrins. III. Redistribution reactions of Macerans amylase. J. Am. Chem. Soc. **72**, 1202–1205 (1950)
54. Myrbäck, K.: Beta-dextrin and starch. I. Announcement. Biochem. Z. **297**, 160–171 (1938)
55. Blinc, M.: Gewinnung des enzyms von *Bacillus macerans* und seine wirkungsweise. Arch. Mikrobiol. **12**, 183–191 (1941)
56. Blinc, M.: Experiments to concentrate the Macerans enzyme. Kolloid Z. **101**, 126–128 (1942)
57. Samec, M.: Über die hydrolyse von abbauprodukten der stärke durch den *Bacillusmacerans* und Sein Enzym. Ber. Dtsch. Chem. Ges. **75**, 1758–1760 (1942)
58. Kerr, R.W.: Action of Macerans enzyme on a component of corn starch. J. Am. Chem. Soc. **64**, 3044–3045 (1942)
59. Kerr, R.W.: On the significance of the degradation of starch by Macerans amylase. J. Am. Chem. Soc. **65**, 188–193 (1943)
60. Akiya, S., Watanabe, T.: A crystalline decomposition product of starch by a bacillus. 3. A new strain of *Bacillus macerans*. J. Pharm. Soc. Japan **70**, 572–576 (1950)
61. Hale, W.S., Rawlins, L.C.: Amylase of *Bacillus macerans*. Cereal Chem. **28**, 49–58 (1951)
62. Northrop, J.H., Ashe, L.H., Senior, J.K.: Biochemistry of *Bacillus acetoethylicum* with reference to the formation of acetone. J. Biol. Chem. **39**, 1–21 (1919)
63. Peterson, W.H., Fred, E.B., Verhulst, J.H.: A fermentation process for the production of acetone, alcohol, and volatile acids from corncobs. Ind. Eng. Chem. **13**, 757–759 (1921)
64. Speakman, H.B.: The biochemistry of acetone formation from sugars by *Bacillus acetoethylicum*. J. Biol. Chem. **64**, 41–52 (1925)
65. Bakonyi, S.: The theory of acetone-ethyl alcoholic fermentation. Biochem. Z. **169**, 125–128 (1926)
66. Zacharov, J.P.: Some morphological and physiological characteristics of *Bacillus macerans*. Centralbl. Bakteriol. Abt. **II**(80), 205–218 (1930)
67. Lange, F.: Verfahren zur gewinnung von polyamylosen. German Patent, Patentschrift n° 442963, I.G. Farbenindustrie Akt.-Ges. Frankfurt (1925)
68. Freudenberg, K., Jacobi, R.: Über Schardingers dextrin aus stärke. Justus Liebigs Ann. Chem. **518**, 102–108 (1935)
69. French, D.: An investigation of the configuration of starch and its crystalline degradation products. Dissertation. Iowa State University. University Microfilms, Inc., Ann Arbor, MI, UMI number: DP12704, 100 p (1942)
70. Freudenberg, K.: Beiträge zur chemie der kohlenhydrate. Ber. Dtsch. Chem. Ges. **76**, A71–A96 (1943)
71. Freudenberg, K.: Hydrolysis and optical rotation of cellulose, starch, and cycloglucans. J. Polym. Sci. **23**, 791–799 (1957)
72. Freudenberg, K.: Beiträge zur chemie der stärke und der cycloglucane (Schardinger-Dextrine). Angew. Chemie **69**, 419–422 (1957)
73. Cramer, F.: Die cyclodextrine aus stärke. Dissertation, Heidelberg, (1949)
74. Cramer, F.: Einschlussverbindungen von cyclodextrinen und die jod-raktion der starke. Angew. Chemie **63**, 487–487 (1951)
75. Cramer, F.: Einschluβverbindungen. Angew. Chemie **64**, 437–447 (1952)
76. Dietrich, H.V., Cramer, F.: Über einschlussverbindungen. VII. Mitteilung, zür struktur der jodketten in kanal-einschlussverbindungen. Chem. Ber. **87**, 806–817 (1954)
77. Cramer, F., Steinle, D.: Die wirkungsweise der amylase aus *Bacillus macerans*. Ann. Chem. Justus Liebig **595**, 81–100 (1955)
78. Cramer, F., Henglein, F.M.: Einschlussverbindungen der cyclodextrine mit gasen. Angew. Chem. Int. Ed. **68**, 649–649 (1956)
79. Cramer, F., Hettler, H.: Inclusion compounds of cyclodextrins. Naturwissenschaften **54**, 625–632 (1967)
80. Hess, K., Trogus, M., Ulmann, M.: Information on the modifications of alpha-dextrin by F Schardinger. Z. Phys. Chem. Abt. B **21**, 1–6 (1933)
81. Freudenberg, K., Cramer, F.: Die constitution der Schardingerdextrine dextrine-alpha, dextrin-beta and dextrin-gamma. Z. Naturforsch. B **3**, 464–464 (1948)
82. Freudenberg, K., Cramer, F.: Über die Schardinger-dextrine aus stärke. Chem. Ber. Recl. **83**, 296–304 (1950)
83. Pringsheim, H.: Über die konstitution der stärke, des glykogens und der flechtenstärke (Beiträge zur chemie der stärke, XII). Ber. Dtsch. Chem. Ges. **57**, 1581–1598 (1924)
84. Pringsheim, H., Beiser, A.: A complement of amylase and the marginal dextrin. Third Memorandum. Biochem. Z. **148**, 336–343 (1924)
85. Pringsheim, H., Leibowitz, J.: Über die constitution der polyamylosen (Beiträge zur Chemie der stärke, IX). Ber. Dtsch. Chem. Ges. **57**, 884–887 (1924)
86. Karrer, P.: Untersuchungen über polymere kohlenhydrate. Angew. Chem. **35**, 85–90 (1922)
87. Karrer, P.: Polymere kohlenhydrate. Akademische Verlagsgesellschaft, Leipzig (1925)
88. Karrer, P., Nägeli, C.: Polysaccharides. II. On the constitution of diarrylose. Helv. Chim. Acta **4**, 169–172 (1921)
89. Karrer, P., Staub, M., Wälti, A.: Polysaccharides XIII. On the understanding of Inulin and the alkali hydroxide bonding of anhydrous sugar. Helv. Chim. Acta **5**, 129–139 (1922)
90. Szejtli, J.: Cyclodextrin Technology, p. 450. Kluwer Academic Publishers, Dordrecht (1988)
91. Crini, G., Fourmentin, S., Fenyvesi, E., Torri, G., Fourmentin, M., Morin-Crini, N.: Cyclodextrins, from molecules to applications. Environ. Chem. Lett. **16**, 1361–1375 (2018)