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Control of the Maillard reaction in food systems

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The Maillard reaction is of primary importance to the food manufacturer, since it is frequently responsible for the aromas and colours that develop during heating or storage of food products. Our ability to control the reaction is still very limited, although recent studies have indicated how it may be manipulated, particularly with regard to aroma development. The introduction of new processing and cooking techniques, such as microwave heating, presents a constant challenge to the manufacturer to find ways of manipulating the reaction. The chemistry of the reaction and the factors affecting it are reviewed.

The Maillard reaction is a type of non-enzymic browning of fundamental interest to food chemists and food processors, because it occurs when virtually all foods are heated[1]. It also occurs during the storage of certain commodities at, or below, room temperature. Non-enzymic browning reactions usually involve the reaction of carbonyl groups with free amino groups, but similar reactions (caramelization) occur in the absence of amino compounds.

As far as the manufacturer is concerned, the most significant and obvious effects of the Maillard reaction are the development of colour and flavour. Most of the effects of the reaction, such as the golden brown colours or caramel aromas that develop on heating, are desirable. Nevertheless, the Maillard reaction is sometimes undesirable (e.g. when dehydrated foods darken and develop off-flavours on storage or during the sterilization of milk). The Maillard reaction may also result in a reduction in nutritional value, the development of antioxidant properties, and the formation of potentially toxic compounds[2]. It also has physiological ramifications. All aspects of the Maillard reaction have been reported in the proceedings of four symposia[3–6].

Chemistry of the reaction

An outline of the Maillard reaction is given in Fig. 1, which illustrates that it is a complex network of reactions. In summary, it involves condensation of the carbonyl group of a reducing sugar (aldose) with a free amino group of a protein or an amino acid to give an *N*-substituted glycosylamine (step A), which rearranges to form the Amadori rearrangement product (ARP) (step B). The ARP dehydrates to form furfurals and reductones (step C) and fission (dicarbonyl) products (step D). These compounds, and aldehydes formed by Strecker degradation of amino acids (step E), may react either in the absence of amino compounds, to give aldols and high molecular weight, nitrogen-free polymers (step F), or in the presence of amino compounds, to give brown nitrogenous polymers and copolymers, called melanoidins (step G). Step H in Fig. 1 illustrates a direct route to fission products from *N*-substituted glycosylamines, without the formation of an ARP (Refs 8 and 9). Some steps of the Maillard reaction are well defined (steps A–E, and many of the reactions leading to the formation of volatile aroma components, not shown in Fig. 1). The reactions leading to the development of non-volatile colour compounds (mainly outlined by steps F and G) remain obscure.

The food manufacturer's interest in the Maillard reaction concerns how it can aid in the production of food of superior quality. In order to achieve this, the factors that affect the reaction must be understood, the most important determinants being temperature, time, composition of the system, water activity (a_w), pH and sulphur dioxide. In fact, the Maillard reaction represents thousands of individual reactions, and each is influenced in different ways by the factors listed above[10]. The following is a summary of the current state of knowledge, and illustrates how aroma and colour formed during the reaction may be controlled.

Temperature and time

An increase in temperature increases the rate of Maillard browning. The reaction has a relatively high temperature coefficient (Q_{10} values range from 2 to 8)[11]. This property is useful when the objective is to prevent the reaction completely (e.g. in food that is to be stored). However, finer control, leading to manipulation of the reaction, is also of value to the food processor.

Early studies of model systems showed that an increase in temperature and/or time of heating resulted in increased colour development. However, very little information is available about the nature of the components responsible for colour[12], although it has been shown that, for melanoidins formed from the reaction of glucose or xylose with glycine, the carbon-to-nitrogen ratio, the degree of unsaturation and the chemical aromaticity increase with temperature and time[13,14].

Foods stored at room temperature may develop off-flavours due to the Maillard reaction. On heating at intermediate temperatures (e.g. 100°C), caramel or cooked flavours are produced, while at higher temperatures (e.g. 150°C), the aroma is 'toasted' or 'roasted'[10] (Fig. 2). It is clear that the aroma profile varies with the temperature and time of heating. At any given temperature–time combination, a unique aroma, which

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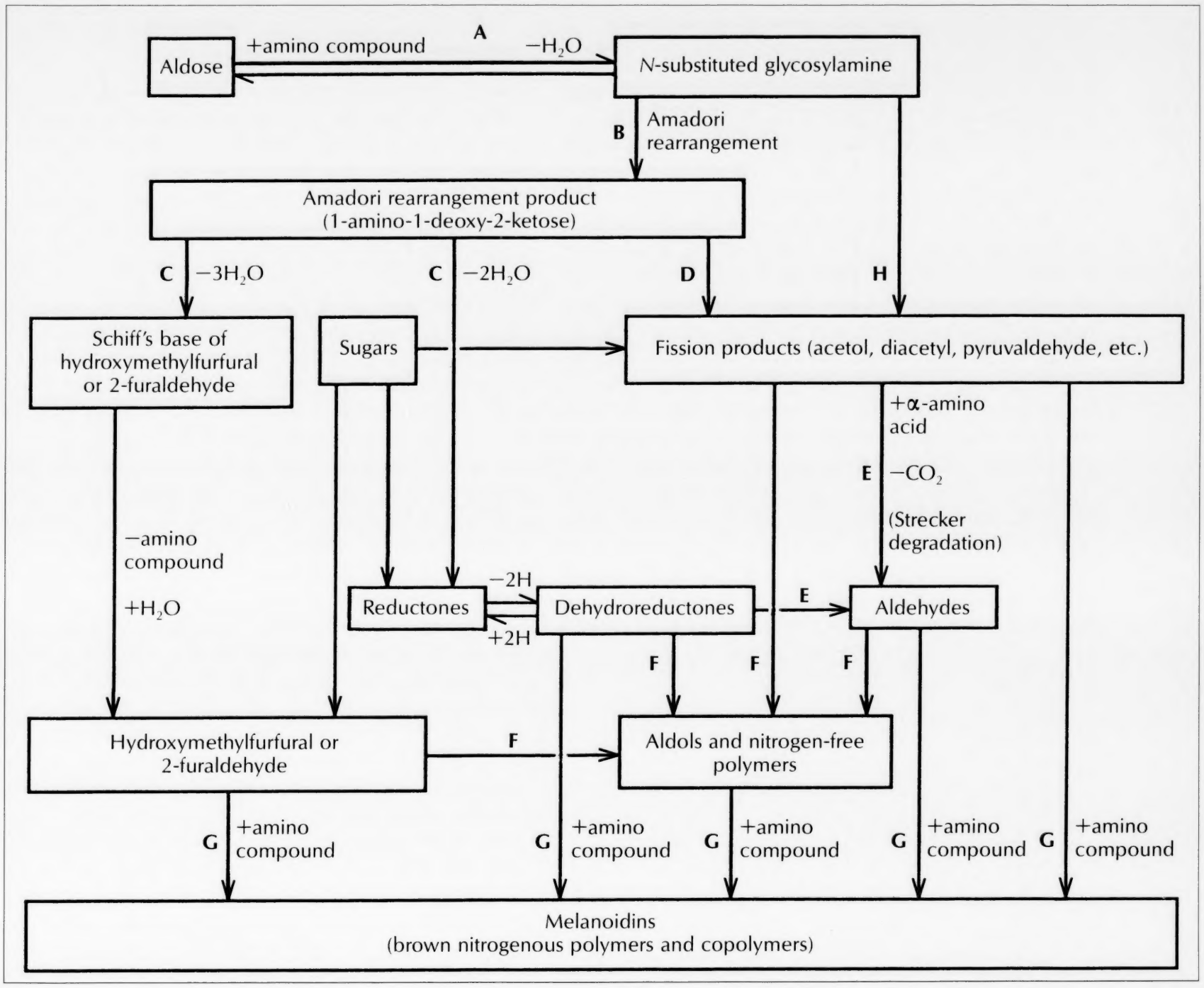

Fig. 1

An outline of the Maillard reaction. (Adapted from Refs 7 and 8.)

is not likely to be reproduced at any other combination of heating conditions, is produced[10]. This was confirmed by a study of a model meat flavour system consisting of cystine and 2,5-dimethyl-4-hydroxy-2,3-dihydrofuran-3-one (DMHF). Shu and Ho[15] showed that, of all the parameters investigated, temperature had the greatest effect on both the yield of total aroma components (the higher the temperature, the higher the yield) and the final aroma profile of the system. Heating in a water : glycerol (75 : 25) mixture at 160°C gave the most desirable aroma, described as 'pot-roasted, meaty and clean'; the major reaction products were trithiolanes, thiophenones and 2,4-hexanedione. Temperatures of 100°C or 200°C resulted in different aroma descriptions, due to the formation of different predominant reaction products. Heating in pure water at 160°C resulted in optimum aroma after 30 minutes; more burnt and biting flavours developed after longer heating times.

Temperature also affects the development of aroma during extrusion cooking. Low levels of pyrazines were formed during the processing of potato flakes when the extruder temperature was less than 130°C, but increased by about tenfold when the temperature was increased to 160°C (Ref. 16). Pyrazine yields also increased over the same temperature range during the processing of malt, but decreased at 190°C, probably due to evaporation or binding to macromolecules[17].

Composition of the system

Pentose sugars (e.g. ribose) react more readily than hexoses (e.g. glucose) which, in turn, are more reactive than disaccharides (e.g. lactose). In addition, individual sugars within each of these groups exhibit different reaction rates; in particular, aldoses, such as glucose, behave differently to ketoses, such as fructose[18]. Of all the amino acids, lysine results in the most colour in the Maillard reaction, due to its ε-amino group; cysteine results in the least colour[19]. Therefore, foods containing proteins that are rich in lysine residues (e.g. milk proteins) are likely to brown readily. The sugar-to-amino-compound ratio also influences the amount of colour produced; for example, a glucose–glycine model system containing 65% water and stored at 65°C showed a

rapid increase in colour formation as the glucose-to-glycine ratio was decreased (e.g. from 10 : 1 or 2 : 1 to 1 : 1 or 1 : 5)[20]. In foods, attempts to prevent the reaction should be directed towards removing one of the reactants: the amino compounds (for carbohydrate-rich foods) or the reducing sugars (for protein-rich foods). For example, in jam making, cooking times of only 15–20 minutes limit the inversion of sucrose to glucose and fructose in the acidic medium[21]; when making Cheddar cheese for processed cheese manufacture, starter cultures should adequately ferment the reducing sugars in the milk, in order to prevent brown colour formation in the processed product[22]. An alternative to reducing the levels of one of the reactants is to process the carbohydrate and protein ingredients separately. This approach has improved the quality of some products (e.g. cream-style soups and dairy products)[23].

The aroma produced on heating is greatly influenced by the reactant amino compound[24]. Many amino acids lead to the development of caramel aromas in heated aqueous systems, but others lead to very specific aromas. The sulphur amino acids, notably cysteine, result in meaty aromas, whereas proline and hydroxyproline result in bread and cracker aromas[10]. Although the sugar has much less effect on the final aroma, the choice of sugar can strongly influence the total amount of flavour produced[25].

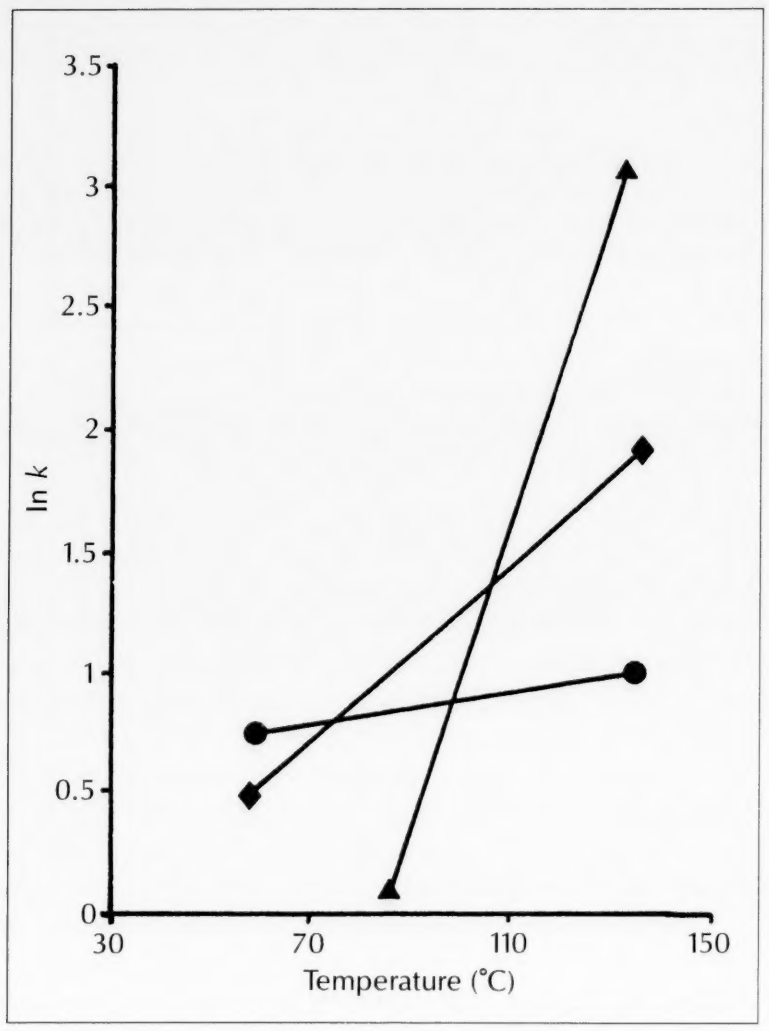

Fig. 2

Hypothetical semi-log plots showing the effects of temperature on rate constants (k, ppm/h) for the formation of: 5-hydroxymethyl-2-furaldehyde (circles), which is associated with off-flavours in foods on storage; 2-hydroxy-1-methylcyclopent-1-en-3-one (diamonds), which possesses a caramel aroma; and 2,3-dimethylpyrazine (triangles), which is associated with toasted aromas. (Reprinted with permission from Ref. 10.)

Water activity

Figure 1 shows that water is produced during the Maillard reaction. Thus, as a consequence of the law of mass action, the reaction occurs less readily in foods with a high a_w value. In addition, the reactants are diluted at high a_w values while, at low a_w, the mobility of reactants is limited, despite their presence at increased concentrations[26]. In practice, the Maillard reaction occurs most rapidly at intermediate a_w values (0.5–0.8)[21], and a_w is of most significance to the reaction in dried and intermediate-moisture foods (IMFs), which have a_w values in this range[11].

However, a_w values for maximum browning are affected by other components of the system; humectants, such as glycerol, can lower the a_w value for maximum browning (e.g. from 0.5–0.7 to below the a_w of many IMFs)[11]. If sugars (e.g. glucose) are used as the humectants in IMFs, serious browning can result. Liquid humectants, such as glycerol and propylene glycol, are useful alternatives.

The food processor is most likely to be concerned about the effect of a_w on the Maillard reaction when concentrating or drying foods, and steps must be taken to ensure that the food passes through the critical a_w values as rapidly as possible, without remaining at elevated temperatures longer than necessary[21]. The water content that results in the maximum reaction rate depends on temperature; for example, on heating freeze-dried carrots at 60°C or 75°C, the maximum rate of formation of ARPs occurred at a moisture content of about 5% ($a_w \approx 0.3$); on heating at 90°C, the reaction rate increased with decreasing water content over the range 25–3%, showing no apparent maximum[27]. In contrast, a study of nonfat dried milk powder, heated at 95°C for 30 minutes to 3 hours, showed that maximum formation of pyrazine and 2-methylpyrazine occurred at an a_w value of 0.75 (Ref. 28).

Heating cystine–DMHF systems containing 0–100% water showed that the sample containing 75% water resulted in the most balanced meaty aroma[15], reflecting high yields of trithiolanes and thiophenones (classes of compound correlated with desirable meat aroma[15]). The moisture content of potato flakes affects aroma formation during extrusion cooking, and the formation of pyrazines has been shown to increase twentyfold on decreasing the moisture content from 48% to 25% (Ref. 16).

The Maillard reaction is of considerable importance when dried and concentrated foods are to be stored, particularly if the moisture content has not been adequately controlled. Problems arise due to the development of yellow colours and off-odours. An example of the

development of undesirable colour may be taken from the results of a study of tomato powders with a_w values of 0.33 or 0.43, stored at 23°C (Ref. 29). The shelf life of the powder at the lower a_w was approximately double that of the powder at the higher a_w. However, preheating the powder at 40°C before storage reduced the shelf life by two-thirds at both moisture levels, due to the formation of ARPs during preheating.

pH

Low pH values (≤7) favour the formation of furfurals from ARPs, while the routes to reductones and fission products are preferred at a high pH (>7). However, in practice, the effect of pH is not this clear cut, and reactions take place by all three pathways, with the pH of the system influencing the ratio of products formed. This has been exemplified by Nursten[26], who showed that several low molecular weight coloured products possess moieties derived from both the furfural and reductone routes. However, the browning potential of the route to furfurals is less than that of the route to reductones and, consequently, the rate of colour formation can be reduced by decreasing the pH (Ref. 26).

The pH also influences aroma; for example, optimum meat flavour is produced in the pH region 4–5.5. This is supported by the cystine–DMHF study[15]. The most desirable meaty and pot-roasted aroma was obtained at pH 4.7, but pH had a less dramatic effect on aroma than did temperature, time or water content. On the other hand, it is well known that pyrazine formation increases greatly with pH, especially above pH 5 (Ref. 28).

Microwave heating

The use of microwave ovens has increased dramatically over the past few decades[30]. However, microwave cooking does not favour the Maillard reaction, and this is one of the problems facing the food industry at present[31]. In order to produce acceptable microwavable food products, a multifaceted approach is required, based on the control of several of the factors discussed above. Conventionally cooked food is subjected to relatively high temperatures for a relatively long time, and the surface of the food dries out to give a crust with a low a_w, favouring the Maillard reaction. In contrast, during microwave cooking, there is a lack of hot, dry air surrounding the product, and the food is usually cooked for a much shorter time[31]. In practice, the surface of many baked and roasted products remains moist during microwave heating, and the development of the expected flavours and colours does not occur, resulting in a product that is perceived as undercooked. Many of the aroma compounds that are formed may be lost by steam distillation.

Several steps can be taken to overcome the lack of browning during microwave cooking. Metallized films, which are used in microwave susceptor packaging, can reach temperatures of up to 260°C. They promote surface drying and high temperatures in the food during microwave cooking by conventional contact heating, resulting in surface browning of the product[30,31]. Unfortunately, flavour development throughout the food is not improved[31]. This may be addressed by adding precursors, such as ARPs, prior to heating[31], but losses of aroma compounds by steam distillation or binding to the food matrix are not prevented. Other ways of improving the flavour and colour of microwave-heated foods include treating the surface of the food with mixtures of reducing sugars and amino compounds before heating, coating the food with a moisture barrier that maintains a low a_w at the food surface, and combining microwave heating with conventional or infrared heating[30].

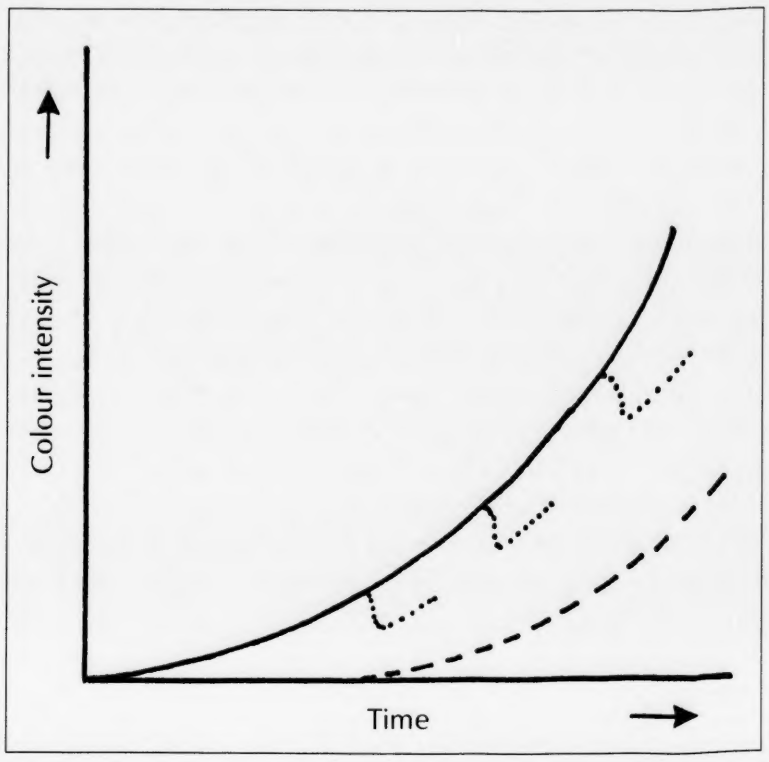

Fig. 3
Effect of added SO_2 on colour development in glucose–glycine solutions at 50°C: unbroken line, no SO_2; dashed line, SO_2 added before incubation; dotted line, SO_2 added during incubation. (Reprinted with permission from Ref. 32.)

Sulphur dioxide

Discussion of ways of controlling the Maillard reaction has, so far, centred on modifying the process conditions or the product composition. An important method of control of the reaction involves using an inhibitor of the reaction, sulphur dioxide, which is normally added as a solution of sodium bisulphite or sodium metabisulphite. Sulphur dioxide also inhibits enzymic browning and the growth of microorganisms. It may affect the early stages of the Maillard reaction, combine with active intermediates, and bleach the final colour[32]. When added to a glucose–glycine system before any browning has occurred, the time for browning to become apparent is extended (Fig. 3). During this period of inhibition, some of the sulphite becomes irreversibly bound to the food, due to combination with reducing sugars and intermediates of the

Maillard reaction (carbonyls and dicarbonyls). In the early stages of the reaction, the rate of irreversible binding of sulphite is proportional to the amount of the free additive in solution[33]. However, sulphite only delays colour formation. When it is added to a model system that has already started to brown, there is an almost immediate reduction in colour (Fig. 3), but the melanoidins are only partially bleached and the colour is not reduced to the level observed when sulphite is added at the beginning of the browning reaction. This indicates that other mechanisms, apart from bleaching, operate when sulphite is added at the beginning of the reaction[32]. The chemistry of the interactions of sulphite with melanoidins still remains to be explored[33], but it is interesting to note that the colour formed in sulphite-treated systems is less red and more yellow than in untreated systems[32].

Conclusion

The Maillard reaction is extremely complex and is influenced by several factors. Much is known about the way in which individual factors affect the reaction in model systems, particularly with regard to aroma development. However, there are few good data describing the kinetics of the Maillard reactions; more studies of the kinetics of such reactions are needed[10]. Investigations aimed at correlating sensory and instrumental data are also required[10]. Little is known about the chemistry of the formation of the colour compounds, and more work is required in this area in order to understand the reactions that we wish to manipulate. Studies on model systems are invaluable; it is also important to study systems that more closely resemble food, and to understand how simultaneous optimization of different factors may assist in controlling the reaction.

References

1 Ames, J.M. (1988) *Chem. Ind. (London)*, 558–561
2 Nursten, H.E. (1980–1981) *Food Chem.* 6, 263–277
3 Eriksson, C. (1981) in *Maillard Reactions in Food (Progress in Food and Nutrition Science, 5)* (Eriksson, C., ed.), pp. 1–6, Pergamon Press
4 Waller, G.R. and Feather, M.S., eds (1983) *The Maillard Reaction in Foods and Nutrition* (ACS Symposium Series, 215), American Chemical Society
5 Fujimaki, M., Namiki, M. and Kato, H., eds (1986) *Amino–Carbonyl Reactions in Food and Biological Systems (Developments in Food Science, 13)*, Elsevier
6 Finot, P.A., Aeschbacher, H.U., Hurrell, R.F. and Liardon, R., eds (1990) *The Maillard Reaction in Food Processing, Human Nutrition and Physiology*, Birkhäuser Verlag
7 Hodge, J.E. (1953) *J. Agric. Food Chem.* 1, 928–943
8 Nursten, H.E. (1990) in *The Maillard Reaction in Food Processing, Human Nutrition and Physiology* (Finot, P.A., Aeschbacher, H.U., Hurrell, R.F. and Liardon, R., eds), pp. 145–153, Birkhäuser Verlag
9 Namiki, M. (1988) *Adv. Food Res.* 32, 115–184
10 Reineccius, G.A. (1990) in *The Maillard Reaction in Food Processing, Human Nutrition and Physiology* (Finot, P.A., Aeschbacher, H.U., Hurrell, R.F. and Liardon, R., eds), pp. 157–170, Birkhäuser Verlag
11 Labuza, T.P. and Saltmarch, M. (1981) in *Water Activity: Influences on Food Quality* (Rockland, L.B. and Stewart, G.F., eds), pp. 605–650, Academic Press
12 Ames, J.M. and Nursten, H.E. (1989) in *Trends in Food Science* (Lien, W.S. and Foo, C.W., eds), pp. 8–14, Singapore Institute of Food Science and Technology
13 Feather, M.S. and Huang, R.D. (1986) in *Amino–Carbonyl Reactions in Food and Biological Systems* (Fujimaki, M., Namiki, M. and Kato, H., eds), pp. 183–192, Elsevier
14 Benzing-Purdie, L.M. and Ratcliffe, C.I. (1986) in *Amino–Carbonyl Reactions in Food and Biological Systems* (Fujimaki, M., Namiki, M. and Kato, H., eds), pp. 193–205, Elsevier
15 Shu, C-K. and Ho, C-T. (1989) in *Thermal Generation of Aromas* (ACS Symposium Series, 409) (Parliment, T.H., McGorrin, R.J. and Ho, C-T., eds), pp. 229–241, American Chemical Society
16 Maga, J.A. and Sizer, C.E. (1979) *Lebensm. Wiss. Technol.* 12, 15–16
17 Fors, S.M. and Eriksson, C. (1986) in *Amino–Carbonyl Reactions in Food and Biological Systems* (Fujimaki, M., Namiki, M. and Kato, H., eds), pp. 257–261, Elsevier
18 Spark, A.A. (1969) *J. Sci. Food Agric.* 20, 308–316
19 Ames, J.M. (1986) *Chem. Ind. (London)*, 362–363
20 Wolfrom, M.L., Kashimura, N. and Horton, D. (1974) *J. Agric. Food Chem.* 22, 796–800
21 Baltes, W. (1982) *Food Chem.* 9, 59–73
22 Bley, M.E., Johnson, M.E. and Olson, N.F. (1985) *J. Dairy Sci.* 68, 555–561
23 Assinder, I., Supran, M.K. and Thompson, G.A.K. (1987) US Patent 4 640 840
24 Lane, M.J. and Nursten, H.E. (1983) in *The Maillard Reaction in Foods and Nutrition* (ACS Symposium Series, 215) (Waller, G.R. and Feather, M.S., eds), pp. 141–158, American Chemical Society
25 Shibamoto, T. (1983) in *Instrumental Analysis of Foods (Recent Progress, Vol. 1)* (Charalambous, G. and Inglett, G., eds), pp. 229–278, Academic Press
26 Nursten, H.E. (1986) in *Concentration and Drying of Foods* (MacCarthy, D., ed.), pp. 53–68, Elsevier
27 Eichner, K., Laible, R. and Wolf, W. (1985) in *Properties of Water in Foods* (Simatos, D. and Multon, J.L., eds), pp. 191–210, Martinus Nijhoff
28 Leahy, M.M. and Reineccius, G.A. (1989) in *Flavor Chemistry: Trends and Developments* (ACS Symposium Series, 388) (Teranishi, R., Buttery, R.G. and Shahidi, F., eds), pp. 76–91, American Chemical Society
29 Eichner, K. and Ciner-Doruk, M. (1981) in *Water Activity: Influences on Food Quality* (Rockland, L.B. and Stewart, G.F., eds), pp. 567–603, Academic Press
30 Lingnert, H. (1990) in *The Maillard Reaction in Food Processing, Human Nutrition and Physiology* (Finot, P.A., Aeschbacher, H.U., Hurrell, R.F. and Liardon, R., eds), pp. 171–185, Birkhäuser Verlag
31 Reineccius, G.A. and Whorton, C. (1990) in *The Maillard Reaction in Food Processing, Human Nutrition and Physiology* (Finot, P.A., Aeschbacher, H.U., Hurrell, R.F. and Liardon, R., eds), pp. 197–208, Birkhäuser Verlag
32 McWeeny, D.J. (1981) in *Maillard Reactions in Food (Progress in Food and Nutrition Science, 5)* (Eriksson, C., ed.), pp. 395–404, Pergamon Press
33 Wedzicha, B.L. (1984) *Chemistry of Sulphur Dioxide in Foods*, Elsevier

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