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Rapid Visco Analyser (RVA) as a Tool for Measuring Starch-Related Physiochemical Properties in Cereals: a Review

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Abstract

Starch, composed of amylose (AMY) and amylopectin (AP), is a common constituent of many agricultural grain crops and the main source of energy for both humans and domesticated animals. There are several physiochemical factors that determine the suitability of starch for a specific end use, which mainly entails the ratios of the AMY and AP, but also the granular and molecular structure thereof. This, in turn, determines its functional properties, i.e. swelling, gelatinisation, pasting and retrogradation. Different instruments, such as the Amylograph®, Falling Number® System, Ottawa Starch Viscometer and the Consistometer, in addition to the more recently developed Rapid Visco Analyser (RVA) are used to study the functional properties of starch. Due to its reliability, repeatability and versatility, the RVA is increasingly used to determine the physiochemical and, in particular, the pasting properties of cereal starches. This review gives an update on the current knowledge of starch-related physiochemical and functional properties of a selection of cereal (i.e. wheat, barley, rice and maize) starches, and the RVA as a measuring instrument thereof, including critical analysis and discussion.

Keywords RVA · Starch · Physiochemical properties · Gelatinisation · Pasting · Retrogradation

Introduction

Starch is a common component of cereal grains and the main form in which carbon is stored (Carciofi et al. 2012; Cuesta-Sejjo et al. 2013). After cellulose, starch is the most abundant and renewable polysaccharide in cereals (Jeon et al. 2010) and is therefore the foremost component of human food and animal feed (Copeland et al. 2009; Gilbert et al. 2013).

Molecular, crystalline and granular differences affect the physiochemical as well as functional properties of the different cereal starches (Li et al. 2017). These differences are closely linked to the ratio of amylose (AMY) to amylopectin (AP)

present in the starch granules (Juhász and Salgó 2008). AMY molecules are composed of mostly linear structures (glucose monomers with α -1,4 glucosidic bonds) with a few long branches. The AP molecules are highly branched with many short branches, making them considerably larger in terms of their molecular weight compared to the AMY molecules (Beckles and Thitisaksakul 2014; Li et al. 2017). The semi-crystalline growth rings in the starch granules comprised crystalline and amorphous lamellae. Regions of the formed AP double helices fall within the crystalline lamellae while the AP branch points lie in the alternating amorphous regions. The AMY molecules are also present in the amorphous lamellae. This results in a highly ordered structure in the starch granule which requires specific enzymes to initiate degradation.

Gelatinisation, a familiar and commonly used characteristic of starch, inflicts changes in granule swelling, birefringence and viscosity (Zaidul et al. 2007; Juhász and Salgó 2008). It results in irreversible changes in functional properties due to the breakdown of the intermolecular structure of the starch molecules (Batey 2007; Beckles and Thitisaksakul 2014). During gelatinisation, the starch granules absorb water and swell, melting the internal crystalline structures that lead to the granules breaking down (Batey 2007). Pasting follows gelatinisation and involves granular swelling and leaching of

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AMY molecules. During pasting, total disruption of the starch granules takes place at a temperature higher than that of gelatinisation. Pasting inflicts the development of a viscous gel and can be measured by observing changes in the viscosity based on rheological principles (Batey 2007; Zaidul et al. 2007). A further property of starch, i.e. retrogradation, involves the re-aligning and re-association of the AMY and AP chains in the gelatinised starch, again into an ordered crystalline structure (Batey 2007; Sandhu and Singh 2007; Beckles and Thitisaksakul 2014). Although both AMY and AP are capable of retrogradation, the AP component appears to be responsible for long-term changes in foods such as bread staling.

Different instrumental methods have been employed to study starch pasting properties, with the original method being the Consistometer (Merill 1925). Other instruments include the Amylograph®, Falling Number® System and Ottawa Starch Viscometer. However, the Rapid Visco Analyser (RVA), being more sensitive, repeatable and versatile, became the instrument of choice in recent years (Deffenbaugh and Walker 1989; Zhang and Hamaker 2005). The RVA mimics the cooking process of a cereal when a flour-water suspension is subjected to a heat-hold-cool-hold temperature cycle (Chen et al. 2008). During the heating stage, the gelatinisation temperature of AP is reached and subsequently the paste viscosity rises to a peak viscosity. The granules hydrate and swell causing AMY and small AP molecules to leach out (Chen et al. 2008). This is followed by the hot holding stage when a drop in viscosity is seen. This sudden decrease in viscosity has been associated with swelling and rigidity of starch granules as a result of variations in the AP, AMY, lipid and protein fractions content and/or molecular structure (Chen et al. 2008). The viscosity rises again during the cooling phase as the soluble AMY retrogrades, forming a gel containing gelatinised starch granules (Chen et al. 2008).

The starch viscosity or pasting property of a given cereal enables the improvement thereof, and aid with the selection or screening of new breeding lines in breeding programmes (Zhou and Mendham 2005; Cozzolino et al. 2012; Cozzolino 2016; Batey 2007; Glennie-Holmes 1995a). The information provided by RVA viscograms (or curves) makes it highly suitable to study the genetic traits of cereal grains (Chen et al. 2008). Although originally being developed to identify pre-harvest sprouting in wheat (Ross et al. 1987), applications have extended to pre-harvest sprouting also in barley (Bason et al. 1993; Batey et al. 1997) and studying the microstructure of starch pastes to elaborate on the relationship between chemical composition and pasting properties (Singh et al. 2003b). The RVA has been shown to be useful for a wide range of applications (Almeida-Dominguez et al. 1997a). This review aims to present the current status of knowledge on the RVA as a tool to measure the physicochemical and functional properties of starch in a selection of cereals,

emphasising pasting properties and interpretation of results, with critical analysis and discussion.

Starch Biology

Starch is synthesised in many organs within plants and stored in two forms, i.e. storage and transient starch. Storage starch is used for long-term storage in amyloplasts and is one of the main components in cereal grains and tubers, while transient starch is used for short-term storage in chloroplasts of vegetative organs such as leaves (Mutisya et al. 2015; Tetlow et al. 2004; Regina et al. 2010; Cuesta-Seijo et al. 2013; Sonnewald and Kossmann 2013). Starch granules are found in different sizes, shapes, crystalline type and composition (Table 1) depending on their botanical origin (Cuesta-Seijo et al. 2013; He et al. 2013). Granule size ranges from 1 to 100 µm in diameter depending on the starch type (He et al. 2012). Starch is composed of glucose polymers in the form of semi-crystalline granules with an internal lamellar structure made up of AMY and AP. The primary structure of these fractions is similar. Both AMY and AP are poly-glucans with the glucose moieties linked by α -1,4-glucosidic linkages with branch points composed of α -1,6 linkages. These two fractions, however, differ in chain length and the degree of branching, both of which influence the physicochemical properties of the starch (Slattery et al. 2000; Tester et al. 2004a; Hannah and James 2008).

AMY comprises linear chains of glucose (D-glucan chain) molecules joined by α -1,4 glucosidic linkages (Fig. 1a) with less than 0.1% α -1,6 branch points (Kossmann and Lloyd 2000; Regina et al. 2010). The molecular weight of AMY is ca. 10^5 – 10^6 Da (Tester et al. 2004a; Copeland et al. 2009; Thitisaksakul et al. 2012) and contains ca. 9–20 branch points resulting in ca. 3–11 chains per molecule. Each chain contains 100–10,000 glucose residues resulting in an approximate molecular weight of between 30,000 and 110,000 Da (Tester et al. 2004b; Keeling and Myers 2010; Simsek et al. 2013).

AP also consists of linear chains of glucose (D-glucan chain) linked by α -1,4 glucosidic bonds, but contains, in contrast to AMY, a high degree of branched points, ca. 5% α -1,6 linkages (Fig. 1b) (Kossmann and Lloyd 2000; Tester et al. 2004b; Regina et al. 2010). These linkages are organised as such that there are distinct regions comprising a high level of branching (amorphous region), alternating with regions devoid of branches. With a molecular weight of 10^7 – 10^9 Da (Tester et al. 2004a, 2004b), AP is thus larger and has a more organised structure compared to AMY. AMY, however, has a longer average chain length than AP (Tester et al. 2004a, 2004b) due to the lower frequency of branch points. AP aggregates into clusters where the chains within the cluster form double helices resulting in a crystalline region (James et al. 2003; Sonnewald and Kossmann 2013).

Table 1 Shape, composition and properties of wheat, barley, rice and maize starch granules (adjusted from Belitz et al. 1999)

Source	Shape	Diameter (μm)	Crystallinity (%)	Amylose (%)	Amylose polymerisation degree	Amylopectin iodine binding constant ^a	Amylopectin polymerisation degree
Wheat	Lenticular Polyhedral	2–38	36	22–28	2100	0.21	19–20
Barley	Lenticular	2–5	–	22–29	1850	–	26
Rice	Polyhedral	3–8	38	14–32	–	0.59	–
Maize	Polyhedral	5–25	–	28	940	0.91	25–26

^a mg iodine/100 mg starch

AP typically makes up the major portion of cereal grain starch varying from ca. 60 to 85% in normal starch (Keeling and Myers 2010) and up to 100% in waxy starch types. This makes starch, and more specifically AP, the most abundant single component within cereal grains which is considerably more than proteins and other grain components considered important in cereal grain quality.

Starch Biochemistry

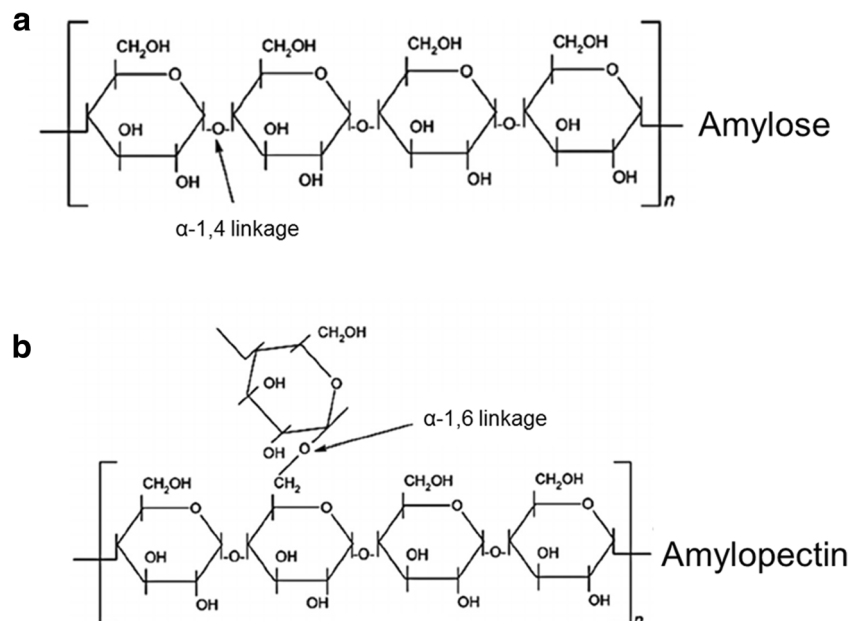
ADP-glucose pyrophosphorylase (AGPase) is responsible for the first step of starch metabolism, i.e. the production of ADP-glucose. ADP-glucose is the substrate for starch synthase in the production of AMY and AP (Smith et al. 1997; Tetlow 2011; Tetlow and Emes 2017). Starch synthase comprises of two classes, soluble starch synthases (SSI, SSII, SSIII) and insoluble granule-bound starch synthases (GBSSI and GBSSII). These enzymes catalyse starch synthesis by transferring the glucosyl moiety from ADP-glucose to the non-reducing end of the α -1,4 linkage to form AP and AMY

(Ball and Morell 2003; Li et al. 2003; Morell et al. 2003; Tetlow and Emes 2017).

Starch-branching enzymes (SBEI and SBEII) catalyse the hydrolysis of α -1,4 glucosidic linkages within the polymer and transfers the hydrolysed chain to form α -1,6 glucosidic linkages (Slattery et al. 2000; Thitisaksakul et al. 2012; Tetlow and Emes 2017). Starch-debranching enzymes (ISA1, ISA2 and PUL) determine AP structure by regulating the branching and maintenance of AP crystallinity (Morell et al. 2003). This process occurs in special organelles known as plastids and the biosynthesis can occur in either amyloplasts or chloroplasts (Smith et al. 1997; Carciofi et al. 2012). The modes of enzymatic actions that affect the rate in which these enzymes function are not yet defined (Wu et al. 2013).

Natural variation in cereal types, usually from a wild source, can result in allelic variation in one or more of the starch synthesis genes (Tikapunya et al. 2017; Burton et al. 2002; Campbell et al. 2016). The most common is a mutation or knockout of GBSS, which can result in cereal starch comprising 100% AP, called waxy-type starch (Pedersen et al. 2005). Where there is allelic variation in GBSS, a hetero-

Fig. 1 Glucose moieties linked by **a** α -1,4 to form amylose and **b** by α -1,4 glucosidic linkages with branch points composed of α -1,6 linkages to form amylopectin



waxy is formed, which could be between 90 and 95% AP. Variation in SSI and SSII genes can result in high AMY expression, but usually not more than 40% (Swanston et al. 2001). Variation is, however, not limited to the SS genes. Allelic variation in either SBE or SDE will result in changes to the structure of AP, through the degree of branching (Campbell et al. 2016; Regina et al. 2004). The effects of these changes on starch properties can be measured through pasting analysis, e.g. using the RVA (Campbell et al. 2016).

The lowest level of organisation found in starch granules is alternating crystalline (hard) and semi-crystalline (soft) shells (Fig. 2a), only several hundred nanometers (nm) in thickness (Gallant et al. 1997). The hard shells consist of larger blocklets which are ca. 50–500 nm in size while, the soft shells are smaller and range between 20 and 50 μm . At the higher level of structure organisation, the blocklet structure shows amorphous radial channels connected by a central cavity with the exterior of starch granule (Kossmann and Lloyd 2000; Gallant et al. 1997). At the highest level of structure, one blocklet is observed containing numerous amorphous crystalline lamellae besides the crystalline structure of the starch polymers (Gallant et al. 1997) (Fig. 2b). The crystalline structure is exclusively associated with the AP component while the amorphous regions mainly represent AMY (Singh et al. 2006) (Fig. 2b) (Table 1).

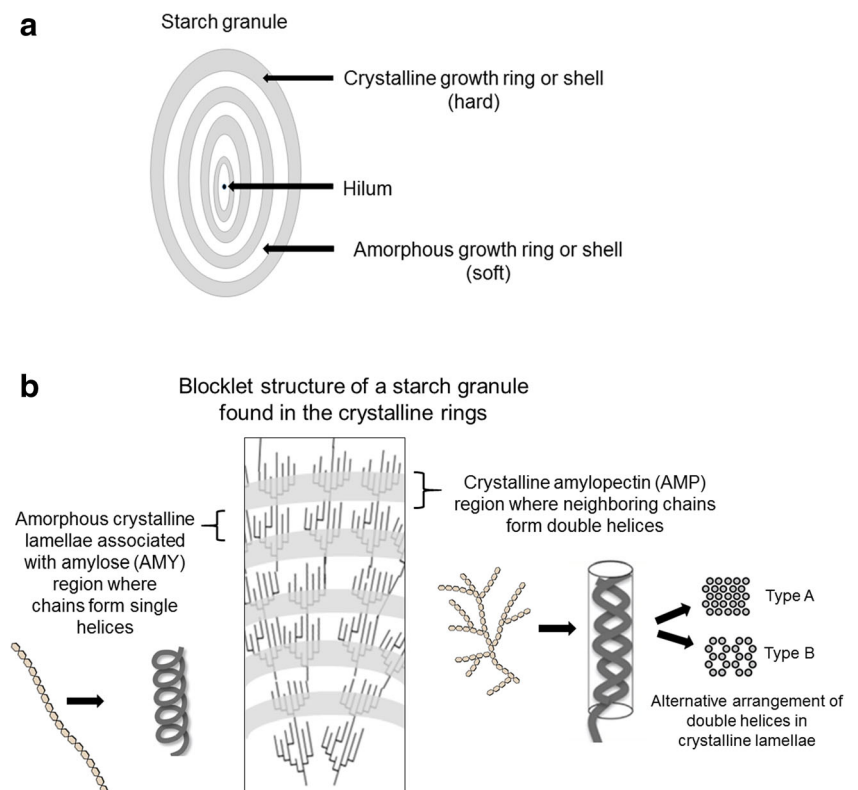
Starch differ structurally with respect to granules, crystallinity and molecule complexes (Li et al. 2017) (Table 2). In

wheat starch, the granular structures found are either the larger A-type or the smaller B-type granules, with similar size-related differences found amongst other types of cereals as well. Further, the surface morphology of the granules also differ and thereby contribute towards the structural differences. Also, surface pores and internal channels are present in varying sizes, intensities and distributions. The functionality of respective starch granules implies that the larger pores and channels allow easier access of water, chemicals and enzymes to the granule interior, and thereby enhance the starch gelatinisation.

The chemical composition of starch granules differs due to the varied AMY and AP contents (inherently determined by species), which are related to the crystalline structure thereof (Li et al. 2017). This also correlates with the size of the granules, i.e. the larger A-type granules. High-amylose maize starch granules, when exposed to water, slowly swell due to the large amounts of AMY packed between the small amounts of crystalline AP (Batey 2007); thus, the level of crystallinity, and thereby the higher amount of longer chain fractions (AMY), plays an important role in the retrogradation as functional property of starch (Li et al. 2017).

Besides the AMY and AP ratio differences, the varied amylose-lipid complexes and the protein and phosphorus contents also lead to differences in gelatinisation, pasting and swelling of starch, in conjunction with varying granule sizes (Li et al. 2017). Soft maize with loosely packed starch granules has

Fig. 2 **a** Schematic view of the structure of a starch granule, with alternating amorphous and crystalline regions, constituting the growth rings; and **b** represents a blocklet structure of a starch granule, found in the crystalline regions and consisting of amorphous lamellae representing amylose, as well as the crystalline structures, associated with amylopectin



reduced protein-to-starch bounds and can therefore hydrate and swell more rapidly in the presence of heat opposed to tightly packed granules of hard maize (Almeida-Dominguez et al. 1997b). For milled rice, the absence of surrounding protein matrix makes hydrated and swollen starch granules more fragile, aiding disruption with a smaller ability to develop viscosity during heating (Almeida-Dominguez et al. 1997b).

Functional Properties of Starch and its Measurement

The application of starch and starch-based products is determined by its physicochemical or rheological properties. The main functional properties of starch include pasting and viscosity of the starch paste. Rheological methods play an important role in measuring functional properties of starch-based materials (Panozzo and McCormick 1993; Higley et al. 2003). Pasting properties were originally measured by means of the Consistometer; however, this instrument was not economically viable and required highly skilled operators (Merill 1925). Subsequent instruments included the Ottawa Starch Viscometer (Deffenbaug and Walker 1989) and the Brabender Visco-Amylograph which became available during the 1930s. The Brabender Visco-Amylograph was modified into the Amylograph® (Deffenbaug and Walker 1989).

More recently, the Rapid Visco Analyser (RVA) was developed by the Bread Research Institute of Australia, in collaboration with the Wheat Research Unit of CSIRO Division of Plant Industry (Goode et al. 2005). The RVA was initially invented as a tool to measure the extent of sprout damage, first in wheat and later in barley (Ross et al. 1987; Wrigley 1994; Deffenbaug and Walker 1989). The first RVA did not include a temperature-controlling computer; instead, the temperature was increased manually using a heater and monitored with a thermometer (Hazelton and Walker 1996). Recognising this

problem, the RVA was improved by incorporating computer technology which resulted in enhanced reproducibility, versatility and more rapid measurements. In addition, development of the paddle has ensured homogeneity of the sample and optimal heat transfer (Hazelton and Walker 1996). The RVA has several advantages over the Amylograph, i.e. small sample size (4 g compared to 65 g); ability to set a temperature profile; and to report to a computer (Deffenbaug and Walker 1989; Zhou and Mendham 2005; Crosbie and Ross 2007). In addition, the Amylograph analysis time is approximately 2 h due to the cooling rate of 1.5 °C/min. The RVA result obtained was shown to be similar to that obtained with the Falling Number apparatus, when used to determine the extent of sprout damage (Glennie-Holmes 1995a). The RVA has thus been recommended for its speed as well as its strong mixing action, compared to the Ottawa Starch Viscometer and Amylograph (Zhou et al. 1998). Other significant differences between the RVA and the latter two instruments include rotation speed, spindle geometries, heat transfer and sample temperature-measuring systems (Mariotti et al. 2005).

The RVA is easy to operate and versatile in terms of test procedures using different temperature/time profiles. The standard profile takes 13 min to complete. The parameters measured by the RVA are similar to the other viscometers (Deffenbaug and Walker 1989; Ryu et al. 1993).

In addition to the mentioned viscometers, the dynamic Bohlin CS-50 rheometer (Goode et al. 2005) has also been used to study the rheological properties of starch (Singh et al. 2003a). This system has a unique star-shaped paddle rotor and considers temperature/time, grit loads, adjunct and enzyme levels (Goode et al. 2005). Although the rheometer is very useful, the RVA is favoured due to the following functionalities: (1) readouts are given in standard viscosity units (mPa·s), (2) it is less sensitive to localised disturbances, (3) it is user friendly and (4) it is considerably cheaper to purchase (Goode et al. 2005) (Table 3).

Table 2 Structural differences in cereal starches and the effect thereof on the functional, as well as physiochemical properties

Starch structure	Differences	Physiochemical and functional properties	Reason	Effect
Granular structure and surface morphology	Size of granules and amount and position of surface pores and internal channels	Gelatinisation	Pores and channels allow easier access of water, chemicals and enzymes to the granule interior	Larger pores and channels enhance digestibility and gelatinisation
Crystallinity	Varied AMY and AP contents: higher AMY content granules have a higher crystallinity (waxy starches)	Retrogradation		
Molecular structure	AMY: AP AMY-lipid complexes Protein content Phosphorus content	Pasting Swelling Gelatinisation	Loosely packed starch granules have reduced protein-to-starch bounds opposed to tightly packed granules	Hydrate and swell more rapidly in the presence of heat

Table 3 Advantages of the Rapid Visco Analyser

Advantage	Reference
Small sample size requirement (<i>ca.</i> 4 g)	(Deffenbaugh and Walker 1989; Zhou and Mendham 2005; Crosbie and Ross 2007; Zhou et al. 2007)
Standard profile completes in 13 min or less	(Deffenbaugh and Walker 1989)
Durability and ease of operating the instrument	(Deffenbaugh and Walker 1989; Crosbie and Ross 2007)
Versatility of test procedure and direct demonstration of starch application in foods	Deffenbaugh and Walker 1989; Hazelton and Walker 1996)
Parameters measured by RVA are similar to the other viscometers, i.e. Amylograph and Falling Number System.	(Deffenbaugh and Walker 1989; Ryu et al. 1993; Glennie-Holmes, 1995a)
Relatively cheap to purchase	(Goode et al. 2005; Zhou et al. 2007)

The RVA as Rheological Method of Analysis

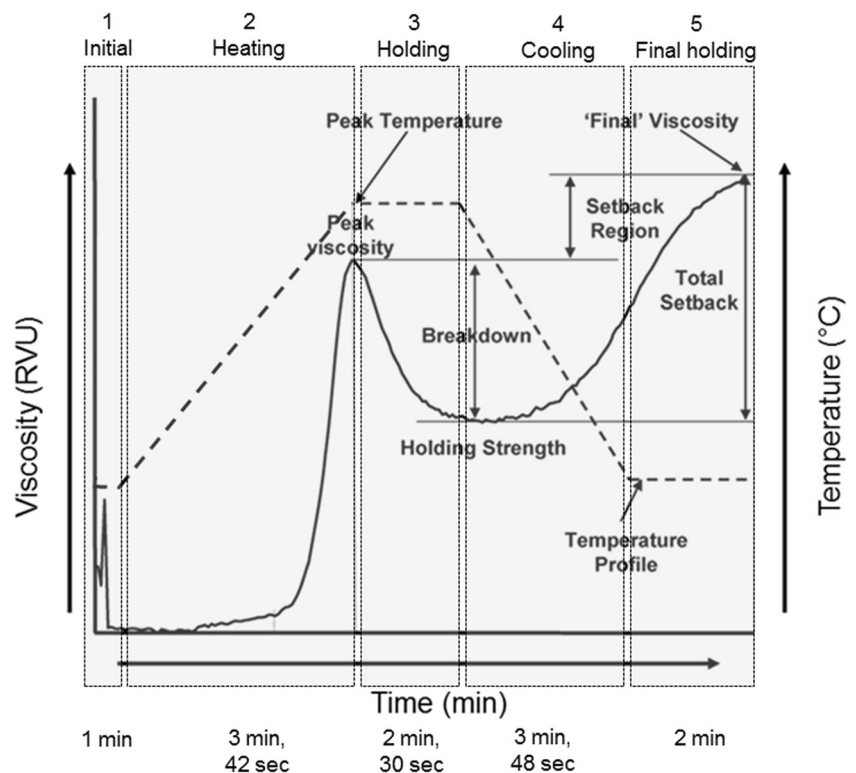
By definition, the RVA is a heating and cooling viscometer that measures the viscosity of a sample over a given period of time while it is stirred (Gamel et al. 2012). During an RVA test, the process of pasting is observed in a starch/flour and water slurry subjected to stirring and heat, i.e. the formation of a gel as the starch granules swell and totally dislocate following gelatinisation (Batey 2007). The test is divided into 5 stages: (1) the addition of water to the starch/flour sample; (2) heating; (3) holding at a maximum temperature; (4) cooling; and (5) a final holding stage. Consequently, the observed RVA profile reflects the complex interactions of the starch and water, affected by temperature and time. The

standard RVA profile (RVA STD 1) comprises the following: initial temperature set at 50 °C; holding time of 1 min at 50 °C; heating to 95 °C over 3 min 42 s; holding at 95 °C for 2 min 30 s; cooling to 50 °C over 3 min 48 s; and lastly, holding at 50 °C for 2 min (Fig. 3).

Initial Stage

The first or *initial stage* involves the combination of sample and solvent in a canister, which is placed into the RVA at temperatures above ambient, followed by mixing (Batey 2007). During this stage, hydration is taking place where water moves into the interior of the starch and also binds to other components such as protein. At temperatures below 50 °C,

Fig. 3 A typical RVA pasting profile of a cereal (using the standard profile), indicating the main parameters measured during analysis



swelling of starch granules is minimal, but at higher temperatures aggressive swelling is found and breakdown of granules can also start to occur.

Heating Stage

The *heating stage* simulates the cooking cycle. Pasting, displayed as a sudden increase in viscosity, occurs during this stage (Batey 2007; Sandhu et al. 2007). Gelatinisation occurs when starch is heated in the presence of water, resulting in an expansion due to hydration. The starch granules rupture resulting in the breakdown of hydrogen bonds (Tester et al. 2006; Lii et al. 1996; Srichuwong et al. 2005). When the gelatinised granules are continually heated, AMY leaches out of the granules resulting in a paste. The paste consists of a molecular dispersion of dissolved starch molecules and discontinuous phase of swollen granules and granule fragments (BeMiller 2011). Increased viscosity is thus observed during the heating process. The temperature, when the viscosity of the starch begins to rise, is recorded as the *pasting temperature* (PT) (Liang and King 2003). Since the viscosity only begins to increase once the starch granules are completely gelatinised, the PT is usually higher than the gelatinisation temperature. *Pasting* thus takes place immediately after gelatinisation, and it refers to changes in viscosity before, during and after the gelatinisation process (Zeng et al. 1997). The PT thus provides an indication of the minimum temperature required to cook a flour (Sandhu et al. 2007). The increase in viscosity with increase in temperature may be attributed to the removal of water from the exuded AMY by the granules as they swell.

The highest viscosity reached during heating or pasting is recorded as the *peak viscosity* (PV). This is reached at the end of the heating stage when the high number of swollen starch granules results in pasting (Thomas and Atwell 1999). Pasting is the combined effect of swelling and rate of disruption of the granules (Batey 2007). Peak viscosity is also an indication of the water-holding capacity of the starch or mixture thereof (Cozzolino 2016) and is often correlated with other quality properties of the sample. The *time to peak viscosity* (TTPV) is used as an indication of the time taken by a sample to reach peak viscosity (Zhou et al. 2007; Zhou and Mendham 2005). *Peak temperature* is related to the rate at which the granules reach maximum swelling (Batey 2007).

Holding Stage

During the *holding stage* (third stage) at maximum temperature, a decrease of viscosity is found and the curve starts to flatten (Batey 2007). This decrease is due to the melting of the crystalline regions of the starch granule and thereby allowing the rapid movement of water into the granule. The available water is thus reduced and collisions between the granules are

seen. As pasting is a temperature-related phenomenon, the heating rate is also an important factor. Faster heating causes more rapid swelling of the granules and thereby enables more granules to reach their maximum before *breakdown* (BD) or decrease in viscosity starts. The breakdown viscosity is measured by the difference between the peak viscosity (stage 2 measurement) and the *holding strength* (HS), which is the lowest viscosity reached during the holding stage. The rate and extent of swelling and *breakdown* depend on the type and amount of starch, the temperature gradient, shear force and composition of the mixture (Copeland et al. 2009; Schirmer et al. 2013). Differences in pasting properties amongst different genotypes are attributed to granule rigidity, extent of AMY leaching from the granule, phosphate and lipid content, AMY content and starch granule crystallinity (BeMiller and Whistler 2009).

Cooling Stage (Setback)

The holding stage is followed by the *cooling stage*, also known as the *setback* (SB). During the cooling stage, the viscosity increases again as the starch granules are cooled and retrogradation occurs (Batey 2007). During this stage, the AP and AMY chains realign themselves to form a more crystalline structure. The increase in viscosity during the cooling period indicates the tendency of the AMY present in the hot paste to re-associate with decrease in temperature (Kaur et al. 2007). Lower setback viscosities indicated a lower tendency of the starch granules to retrograde (Sandhu et al. 2007). The *setback viscosity* is determined by the difference between the peak viscosity (stage 2 measurement) and the final viscosity (stage 5 measurement) (Zhou et al. 2007; Zhou and Mendham 2005) (Fig. 3).

Final Holding Stage

The last stage entails a *final holding* period where the temperature stays constant while the viscosity continues to increase until a plateau is reached (Batey 2007). This is referred to as the *final viscosity* (FV) or the viscosity at the end of the pasting cycle (Zhou et al. 2007; Zhou and Mendham 2005). With continual heating, after peak viscosity has been reached, the viscosity of the starch paste reduces quickly due to disintegration of the granules and leaching out of solubilised starch polymers from the swollen granules into the solution. The polymers can also align themselves in the direction of shear, which further reduces the viscosity (BeMiller 2007).

Often, the time or temperature of any stage is altered for the same material in order to determine a specific feature or functionality of the given starch, thereby changing the RVA parameters of peak viscosity, holding strength and final viscosity as examples (Batey 2007). Importantly, the pasting properties of starch are affected

by starch granule size, AMY and lipid or protein contents and AP structure (Ao and Jane 2007). The effects that these or other substances may have can be altered by the addition of components such as silver nitrate. Due to the digestion of the starch during the RVA test, α -amylase causes a reduction in the viscosity, whereas wheat gluten proteins can increase or decrease the viscosity of the starch. The addition of lipids only slightly reduced the peak viscosity, whereas the addition of detergents caused large increases in the peak viscosity (Batey 2007).

Application of the Rapid Visco Analyser in Cereal Analysis

The RVA was developed to rapidly estimate sprout damage in wheat (Ross et al. 1987; Walker et al. 1988). This application was soon extended to include the detection of sprout damage in barley at grain receival points (Bason et al. 1993; Batey et al. 1997). Nowadays, applications include variations of the basic test in order to determine end-use starch quality parameters and specifically starch pasting properties. Examples of such investigations include the prediction of noodle texture quality in wheat (Crosbie et al. 1999; He et al. 2004; He et al. 2006); bread-making potential of wheat flour (Leon et al. 2010); functionality of starch and protein during processing of whole grain products (Ragaee and Abdel-Aal 2006); barley and malting quality (Glennie-Holmes 1995a; Zhou and Mendham 2005; Izydorczyk 2008); the evaluation of maize hardness (Almeida-Dominguez et al. 1997b); and the prediction of rice cooking quality (Champagne et al. 1999). The RVA has been an integral part of the selection processes in breeding programmes of all the major cereals (Batey and Curtin 2000; Ketthaisong et al. 2014).

Wheat

Wheat cultivars from the Chinese winter wheat regions account for 90% of all wheat produced in China. Characterising the starch properties of the wheat flour can facilitate improved noodle quality through genetic advances (He et al. 2006). Using RVA parameters, i.e. peak viscosity, holding strength, breakdown, final viscosity, setback, peak time and pasting temperature in correlation with the AMY content ideal crossing parents could be identified.

Using the RVA, it was shown that the preferred texture of Japanese noodles (udon and ramen) was associated with high peak viscosity (Crosbie et al. 1999). Indian waxy wheat varieties were assessed to establish the relationship between starch pasting properties and noodle quality (Ram and Sharma 2013). Small reduction in

AMY content of partial waxy wheats showed significant improvement in the eating quality of noodles, indicated by higher peak viscosities that relate to a soft elastic noodle texture (Ram and Sharma 2013). The decreased paste gel rigidity contributed to a softer texture.

Earlier studies consistently reported breakdown as the useful indicator of noodle texture. The inactivation of the α -amylase, which considerably affected the other RVA parameters, was however not considered (Crosbie et al. 1999). Batey et al. (1997) optimised the RVA test conditions for the prediction of Asian noodle quality, using Australian wheat, and confirmed that α -amylase altered the pasting viscosity of flour as well as that of whole meal. An investigation into the inhibition of α -amylase activity before and during viscosity measurements using acids, chelating agents and heavy-metal salts, showed silver nitrate to be the most effective inhibitor (Batey et al. 1997).

The physicochemical and functional properties of wheat starch also contribute to bread-making quality (Leon et al. 2010). Peak, breakdown, setback and final viscosity were identified as clear RVA indicators of bread-making quality (Leon et al. 2010). These RVA parameters as used in bread-making quality studies are elaborated in Table 4 and wheat applications in Table 5.

Purna et al. (2015) compared the pasting properties of normal wheat flour, measured using the RVA, with that of waxy wheat flour. Normal wheat endosperm consists of ca. 25% AMY and 75% AP, whereas the endosperm of waxy wheat comprised essentially only AP (thus AMY free). The waxy wheat flours were shown to have lower pasting temperatures and final viscosities although their peak viscosities were more than 2.5 times higher than that of normal wheat flours. In order to assess the role of protein in the pasting properties of these flours, they were treated with protease and the RVA analysis performed in a silver nitrate solution. The lack of granular rigidity of the waxy wheat starches, when heated in excess water, resulted in lower pasting temperatures and final viscosities. The higher peak viscosities suggested waxy flours to be more susceptible to amyolytic degradation. Therefore, the factors that directed the differences in pasting properties were the protein matrix that affected the swelling of starch granules, as well as the susceptibility to α -amylase activity (Purna et al. 2015).

An increasing interest in high AMY flours (mainly used for pasta production) led to the manipulation of the starch content of durum wheat (Sestili et al. 2010). The AMY content of durum wheat can be markedly increased through the silencing of specific genes. RVA analysis of high-amylose lines showed an overall decrease in viscosity, with significant variations in mean trough, breakdown, setback and final viscosity, when compared to the control wheat samples. The decrease in viscosity indicates a lower level of retrogradation.

Table 4 Rapid Visco Analyser bread-making investigations (Batey 2007; Leon et al. 2010)

RVA parameter	Definition	Property	Effect on bread-making
Peak viscosity	The viscosity that is reached due to an increase of temperature after reaching the pasting temperature	The point where equilibrium is reached between starch granule swelling and rupture; when AMY is leaching out into the solution; related to the water uptake capacity of the starch; lower peak viscosity relates to a lower degree of swelling of the starch granules and also a lower starch content	Associated with final product quality; predictors of bread firming behaviour during storage
Breakdown viscosity	Takes place during the holding period at a constant high temperature, when the swollen granules rip, or rupture thereof occurs	A breakdown in viscosity is caused by rupture of swollen granules; lower breakdown viscosities relate to a decrease in the rate of rupturing of starch granules	Associated with the cooking or hot stability or ease of cooking of the starch; reveals different heating stabilities of starch granules
Setback viscosity	Last phase of the pasting curve, also known as the cooling stage; where the viscosity increases again as granules are cooled and retrogradation is also taking place	Corresponds with to the gelling process of the starch; AMY chains recrystallise and form a gel structure	Low setback viscosities indicate low rates of starch retrogradation
Final viscosity	Follows the final holding period where the temperature stays constant while the viscosity continues to climb until a plateau is reached		Indicates the ability of a flour to form a viscous paste after cooking and cooling

Barley and malt

Initial RVA analysis of barley comprised the determination of the extent of damage due to sprouting in barley or wheat (Bason et al. 1993) by indirectly measuring the α -amylase in the barley meal (Izydorczyk 2008). Applications have since expanded into studying relationships between barley flour pasting properties and potential malting quality (Cozzolino et al. 2012; Cozzolino et al. 2014; Cozzolino 2016) (Table 6). Glennie-Holmes (1995a) optimised sample preparation and analysis conditions for routine testing of barley and malt using RVA (Glennie-Holmes 1995a). RVA peak viscosity, peak area and final viscosity as determined from the viscograms were related to malt extract values (Glennie-Holmes 1995b; Glennie-Holmes 1995c; Cozzolino et al. 2012).

Zhou and Mendham (2005) showed RVA measurements to significantly correlate with malting quality. Using 0.1 M silver nitrate in the water slurry, the pasting temperature showed a very close relationship with malting quality. Silver nitrate is considered the most effective inhibitor of α -amylase and thus malt enzymes. The viscogram thus resembles the rheological profile of barley and displays the intrinsic viscosity of starch (Fox et al. 2014). Furthermore, the incorporation of the grain protein content in the regression made the prediction of malt extract (implicating quality) even more accurate (Zhou and Mendham 2005).

Barley malt extract is a key quality indicator (Zhou et al. 2008). Micromalting requires a relatively large amount of grain, usually not available in the early stages of a breeding

programme. The RVA can thus effectively be used when only a small amount of grain is available for quality assessment (Fox et al. 2014).

Pre-germination (pre-harvest sprouting) is the premature sprouting of grain while still in the ear as a consequence of prolonged spells of wet weather before the mature grain is harvested (Izydorczyk 2008). Using the RVA to detect and measure the degree of pre-germination in barley, Izydorczyk (2008) suggested the results to be interpreted as follows: high final viscosity values (≥ 120 RVU (units) or ≥ 1440 cP) indicated no pre-germination (due to low amylase activity), whereas low final viscosity values (< 50 RVU or < 600 cP) indicated moderate to high pre-germination (due to high amylase activity). Edney et al. (2013) confirmed that the RVA can successfully be used to indicate pre-harvest sprouting in barley.

Goode et al. (2005) studied the use of the RVA to assess the impact of barley adjunct additions during mashing. They considered the RVA rheological profile of mash, which is a mixture of malt (and/or other ingredients) and water, heated to convert starches into fermentable sugars for use in brewing. Different ratios of malt:barley adjunct showed major viscosity changes during starch gelatinisation, furthermore revealing minor viscosity changes due to proteolytic actions (Goode et al. 2005). In this instance, peak viscosity (PV) correlated with the level of barley adjunct and was found to be a useful measurement of barley quality (Goode et al. 2005).

Waxy barley is used for specialised foods in Japan and Korea (Yanagisawa et al. 2006). Comparing the AMY contents of 17 waxy barley cultivars with their respective RVA

Table 5 Rapid Visco Analyser wheat applications

Sample set	Grinding: sieve size	RVA slurry	RVA test profile	Interesting finds	Reference
Chinese winter wheat (<i>n</i> = 260)	0.5 mm	Wheat flour (3.5 g) + dH ₂ O (25 mL) or 0.1 M silver nitrate solution (2.5 g)	Starting temp = 65 °C; held for 2 min; raised to 95 °C; held for 6 min; decreased to 50 °C; held for 5 min; heating/decrease rate = 15 °C/min; Stirring speed = 960 rpm for 10 s and 160 rpm for the remainder of the test period	AMY content correlated with RVA parameters (peak viscosity, breakdown, final viscosity, setback and pasting temp.), except with peak time in terms of Chinese noodle texture and quality	(He et al. 2006)
Japanese wheat (<i>n</i> = 20)	0.5 mm	Wheat flour (3.5 g) + 0.1 M silver nitrate solution (25 g)	Starting temp = 65 °C; held for 2 min; raised to 95 °C; held for 6 min; decreased to 50 °C; held for 5 min; heating/decrease rate = 15 °C/min; Stirring speed = 960 rpm for 10 s and 160 rpm for the remainder of the test period	Peak viscosity was the most relevant RVA parameter to indicate preferred Japanese noodle texture	(Crosbie et al. 1999)
Indian wheat (<i>n</i> = 270)	0.5 mm	3.5 g flour +25 mL dH ₂ O	Stirring speed = 960 rpm for 10 s and 160 rpm for the remainder of the test period Starting temp = 50 °C; raised to 95 °C; held for 150 s; decreased to 50 °C; heating/cooling rate = 1 °C/5 s; Stirring speed = 960 rpm for 10 s and 160 rpm for the remainder of the test period	Peak viscosities relate to preferred soft elastic texture in noodles	(Ram and Sharma 2013)
7 transgenic wheat lines derived from the same genetic background	Not available	ICC standard method 162		Peak, final and breakdown viscosity were the indicators that showed differing pasting properties of bread wheat flour	(Leon et al. 2010)
6 hard waxy, 1 hard partial waxy and 1 hard normal wheat	Not available	Wheat flour + dH ₂ O or 1 mM AgNO ₃ or protease +1 mM AgNO ₃ = 28 g	Starting temp = 50 °C; held for 1 min; raised to 95 °C over 3 min; held for 3 min; decreased to 50 °C over 4 min; held for 2 min; Stirring speed = 960 rpm for 10 s and 160 rpm for the remainder of the test period	Waxy wheat flours had lower pasting temperatures and final viscosities although their peak viscosities were more than 2.5 times higher than that of normal wheat flours	(Purna et al. 2015)
Durum wheat	Not available	Wheat flour (3.87 g) + 25.13 ml 0.2% AgNO ₃	Starting temp = 50 °C; held for 1 min; raised to 95 °C; held for 4 min; decreased to 50 °C and held for 4 min; Stirring speed = 960 rpm for 10 s and 160 rpm for the remainder of the test period (heating and cooling rates not available)	In high-amylose lines, a decrease in viscosity were observed due to lower retrogradation	(Sestili et al. 2010)

Table 6 Rapid Visco Analyser barley and/or malt quality applications

Sample set	Grinding: sieve size	RVA slurry	RVA test profile	Objective of study and/or interesting finds	Reference
Barley variety with average malting ability: <i>Bandulla</i>	1 mm	Ground barley (4 g) + liquid (24 mL) or malt (7 g) + liquid (21 mL) Slurries vortexed for 10 s	Stirring speed = 960 rpm for 10 s and 160 rpm for the remainder of the test period or High-speed stirring at 960 rpm for the duration of the test (the rest of the profile was not mentioned)	Effect of changes in sample weight, liquid volume, fineness of grind, initial and final temperatures, speed of mixing and delay before commencing was investigated; also, the effect of chemicals used for inhibition of enzymes was determined.	(Glennie-Holmes 1995a)
Varieties or breeding lines ($n = 60$)	1 mm	Ground barley meal (4 g) + dH ₂ O (25 g) or 0.1 M silver nitrate solution (25 g)	Starting temp = 50 °C; held for 1 min; raised to 95 °C in 3.7 min; held for 2.5 min; cooled to 50 °C in 3.8 min; held for 2 min; Stirring speed = 960 rpm for 10 s and 160 rpm for the remainder of the test period	Investigated different methods of measuring pasting temp: 11.5 RVU in 0.2 min, were used in the profile and not 2 RVU in 0.1 min Also, incorporated the grain protein content by multiple regression which improved the extract prediction	(Zhou and Mendham 2005)
Barley variety: <i>Optic</i> ($n = 1$)	0.2 mm	Ground malted and unmalted (in different ratios) barley (7.84 g) + dH ₂ O (20.16 g)	Starting temp = 50 °C; held for 30 min; raised to 62 °C; held for 40 min; raised to 78 °C; held for 5 min; heating rate = 1 °C/min. Stirring speed = 960 rpm for 10 s and 160 rpm for the remainder of the test period	The temperature profile simulated an upward infusion-mashing programme as is typically used during a brewery mashing programme	(Goode et al. 2005)
Waxy barley ($n = 17$)	Not available	Starch extracts: Starch (2.5 g, 13.5% wb) + 25 mL dH ₂ O	Starting temp = 35 °C, raised to 95 °C at 3 °C/min and held at 95 °C for 5 min, then cooled to 35 °C at 3 °C/min	Temperature at peak viscosity, hot paste viscosity, final viscosity and setback correlated significantly with AMY content	(Yanagisawa et al. 2006)
Barley varieties ($n = 10$)	1 mm	Similar to Zhou and Mendham (2005)	Similar to Zhou and Mendham (2005)	Pasting temperature was highly significant for the selection of parents for improving malt extract. Selection can therefore be made in the early segregating generations	(Zhou et al. 2008)
Barley varieties	0.8 mm	Ground barley (± 4 g, depending on moisture %) + dH ₂ O (25 ml)	Test temp set at 95 ± 1 °C Stirring speed = 960 rpm for 10 s and 160 rpm for the remainder of the test period (2 min 50 s)	RVA indirectly estimates the amount of α -amylase in barley since the level thereof in sound grain is very low compared to its level in germinating grain.	(Lzydorezyk 2008)
Sib lines of Canadian barley ($n = 475$)	0.8 mm	Method of Lzydorezyk (2008)	Method of Lzydorezyk (2008)	RVA results identified possible parents of future genotype combinations for PHS resistance as well as good malting potential	(Edney et al. 2013)
Malt samples from different varieties	1 mm	Malt-water ratio was modified to 1:1.5 in this study.	Stirring speed = 960 rpm for 10 s and 160 rpm for the remainder of the test period (14 min 50 s) Starting temp = 50 °C, raised to 90 °C over 3 min 42 s; held at 90 °C for 5 min, then cooled to 50 °C over 5 min Method 1: Kilned malt method Stirring speed = 960 rpm for 10 s and 160 rpm for the remainder of the test period (14 min 50 s) Starting temp = 55 °C, raised to 95 °C over 3 min 42 s; held at 95 °C for 5 min, then cooled to 55 °C over 5 min Method 2: Commercial malt method maintaining a constant temperature of 65 °C during the RVA run	'Kilned malt' method (Newport Scientific, 1997) Showed the potential of RVA to discriminate between poor and good fermentable barley malts	(Fox et al. 2014)
Commercial malt ($n = 4$) and rice ($n = 2$)	Not available	Not available	Method 1: Kilned malt method Stirring speed = 960 rpm for 10 s and 160 rpm for the remainder of the test period (14 min 50 s) Starting temp = 55 °C, raised to 95 °C over 3 min 42 s; held at 95 °C for 5 min, then cooled to 55 °C over 5 min Method 2: Commercial malt method maintaining a constant temperature of 65 °C during the RVA run	Time of gelatinisation onset and time to peak viscosity were positively correlated to AMY chain length distribution; time of gelatinisation onset and time to peak viscosity had a negative correlation to the amount of fermentable sugars produced in a high-temperature infusion mash	(Fox et al. 2018)

profiles, significant correlations between setback ($r = 0.61$), final viscosity ($r = 0.64$), temperature at peak viscosity ($r = 0.72$) and hot paste stability (time maintained > 80% of peak viscosity) ($r = 0.79$) were observed.

Fox et al. (2014) used barley malt samples with different degrees of fermentability to investigate the effect of various RVA operating conditions on the viscograms. The variants included instrument models, time-temperature profiles, enzyme activity, heating rates and degree of starch damage. Fox et al. (2014) also showed that the RVA could deliver a fast and reliable indication of a malt's performance when simulating the brewery mashing process. The RVA was also used to measure gelatinisation temperature of malts and rice adjuncts when conducting mashing experiments to relate AMY structure to the development of fermentable sugars (Fox et al. 2018). A number of RVA parameters such as the time of gelatinisation onset and time to peak viscosity were positively correlated to AMY chain length distribution which in turn had a negative correlation with the amount of fermentable sugars produced in a high-temperature infusion mash.

Rice

The texture of cooked rice is affected by AMY content, gelatinisation temperature and gel consistency and to a lesser extent protein content (Champagne et al. 1999; Boa et al. 2006). Sensory evaluations and sensory textural attributes of cooked rice are not always clearly defined. Using RVA as an objective method of analysis, setback was shown to correlate the strongest with stickiness (Champagne et al. 1999). Cooking quality was shown to correlate best with peak viscosity, breakdown and setback (Allahgholipour et al. 2006) (Table 7). The study included 167 included waxy (no AMY present), as well as low, intermediate and high AMY content rice varieties. Good cooking quality rice had low AMY content, associated with high peak viscosity and breakdown values, and low setback values. In the study of Boa et al. (2006), a large selection ($n = 499$) of non-waxy rice varieties and lines were analysed to correlate physiochemical properties with the textural properties of the cooked rice. AMY content significantly correlated with almost all the pasting viscosity properties, except for peak viscosity that was found to be greatly influenced by environment.

Inconsistency in the reporting of correlations of AMY content to RVA pasting properties was reported by Chen et al. (2008). The portions of the pasting viscosity variation that explained the AMY content were identified as being the peak viscosity, as well as the breakdown and setback values (Chen et al. 2008). The viscograms of rice flours with different AMY content were compared and

clear distinction was found between three groups differing in AMY content: AMY > 12%, AMY < 5% and AMY < 2% (Wang et al. 2010).

To elaborate on genotype-by-environmental interactions, Tong et al. (2014) applied the RVA to different rice cultivars. AMY content, cool paste viscosity, breakdown and setback values were mainly affected by genotype, while peak viscosity and hot paste viscosity were mainly affected by environmental (Tong et al. 2014), similar to the findings of Boa et al. (2006). More recently, Tikapunya et al. (2017) showed that wild rice varieties from Australia have lower RVA pasting properties compared to commercial rice.

Shafie et al. (2016) classified whole grain rice samples based on high, medium and low RVA stability ratio and final viscosity that could be used as indicators for selection of whole grain rice on the basis of functional properties.

Characterising the physiochemical properties of starch in commercially and traditionally cultivated rice varieties using the RVA, polished rice flour was shown to have a higher range of peak, hold, breakdown, final and setback viscosity than brown rice flour (Rithesh et al. 2018). This indicated the major influence of rice polishing on the pasting curve.

Furthermore, the RVA has been used to study the effect of nitrogen fertiliser at heading stage on the quality of rice under elevated temperature during filling stage (Dou et al. 2017). Significant effects of temperature elevation during filling stage were observed on setback and pasting temperature, while the use of nitrogen fertiliser significantly affected setback and consistency. Effects of earthworm on quality of rice grown in rice-oilseed rape rotation field were investigated using the RVA (Yin et al. 2018). No alteration in any of the pasting properties has been found.

Maize

Maize can be separated into normal, waxy and high-amylose based on the AMY and AP content ratio (Sandhu and Singh 2007). Consumers prefer sticky and tender maize when consumed as cooked green ears. Stickiness and tenderness are determined by the physiochemical properties of starch in the kernels (Ketthaisong et al. 2014). The improvement of waxy maize quality is one of the major aims in breeding programmes and pasting viscosity traits are used as quality criteria (Table 8). Similarly, maize kernel hardness is an important quality property determining milling potential. The RVA has been used with apparent viscosity profiles related to hardness measurements (Almeida-Dominguez et al. 1997a). The selection of particular RVA testing conditions improved the sensitivity to differences. Using 18% solids (opposed to 15%) at either 2.4 or 10 °C/min heating rate enabled differentiation of hard and soft maize most effectively. Increased solids cause higher peak viscosities as more starch granules are available for swelling to increase viscosity. When

Table 7 Rapid Visco Analyser rice applications

Sample set	Grinding: sieve size	RVA slurry	RVA test profile	Interesting finds	Reference
Non-waxy rice (<i>n</i> = 76)	Not available	Not available	AACC Approved Method 61-02; Stirring speed = 960 rpm for 10 s and 160 rpm for the remainder of the test period Starting temp = 50 °C held for 1 min; raised to 95 °C over 3.8 min; held at 95 °C for 2.5 min, then cooled to 50 °C in 3.8 min; held for 1.4 min	Setback correlated best with sensory textural attributes such as stickiness when describing good cooking texture	(Champagne et al. 1999)
Waxy, low AMY and high AMY (<i>n</i> = 167)	Not available	4-g rice flour + 25 mL dH ₂ O	Stirring speed = 960 rpm for 10 s and 160 rpm for the remainder of the test period Starting temp = 50 °C held for 1 min; raised to 93 °C over 5 min; held at 93 °C for 6 min, then cooled to 50 °C over 4 min; held for 3 min	High peak viscosity and high breakdown values, together with low setback values, describe good cooking rice quality	(Allahgholipour et al. 2006)
Non-waxy (AMY present) rice landraces and cultivars (<i>n</i> = 499)	100-mesh sieve	3-g rice flour + 25 g dH ₂ O	AACC Approved Method 61-02	AMY content correlated significantly with all the pasting properties except peak viscosity	(Bos et al. 2006)
Non-glutinous accessions (<i>n</i> = 146)	Not available	3-g rice flour + 25 g dH ₂ O	AACC Approved Method 61-02	AMY content related with peak viscosity, as well as the breakdown and setback values	(Chen et al. 2008)
Rice accessions (<i>n</i> = 20)	Not available	3-g rice flour + 25 g dH ₂ O	AACC Approved Method 61-02	Paste viscosity, breakdown and setback values were mainly affected by genotypic variance, while peak viscosity and hot paste viscosity were mainly affected by environmental variance	(Tong et al. 2014)
Wild rice from Australia (<i>n</i> = 3), <i>O. sativa</i> (<i>n</i> = 5), unpolished rice (<i>n</i> = 2)	Not available	3-g rice flour + 25 g dH ₂ O	AACC Approved Method 61-02	Setback values suggested that intermediate AP chains resulted in less recrystallisation after gelatinisation	(Tikapunya et al. 2017)

Table 8 RVA related studies on maize for the determination of physicochemical properties in relation to eating and textural properties

Sample set	Grinding: sieve size	RVA slurries	RVA test profile	Interesting finds	Reference
Commercial maize ($n = 6$)	1-mm and 2-mm screens	15 and 18% solids	Stirring speed = 960 rpm for 10 s and 160 rpm for the remainder of the test period Starting temp = 50 °C held for 2 min; raised to 95 °C over 19 min; held at 95 °C for 4 min (2.4 °C/min) and Starting temp = 50 °C, held for 2 min; raised to 95 °C over 4.5 min; held at 95 °C for 4.5 min (10 °C/min); then cooled to 50 °C over 4 min; held for 10 min	Finely ground samples had higher viscosity slopes and more distinct peaks that developed both earlier and at lower temperatures than the coarser samples; soft maizes were more affected by grinding conditions than hard maize; reduced peak viscosities were found when greater heating rates were used	(Almeida-Dominguez et al. 1997a)
Indian maize ($n = 9$)	Milled using sieve no. 72 (British Sieve Standards)	10% w/w, 28 g total	Starting temp = 50 °C held for 1 min; raised to 95 °C (6 °C/min); held at 95 °C for 5 min; then cooled to 50 °C (6 °C/min); held for 2 min	Peak viscosity correlated negatively with protein content	(Sandhu et al. 2007)

increased heating rates were applied, more energy was provided to gelatinise the starch, which also accelerated the development of viscosity, and thus higher peak viscosities were observed (Almeida-Dominguez et al. 1997a).

A study undertaken by Sandhu et al. (2007) describes the physicochemical properties of flours from different maize varieties grown in India, as well as the relations to chapati-making. Again, the grains with higher hardness had tightly packed starch granules and consequently developed lower viscosity levels.

Maize genotypes with suitable properties (hardness, density and structural composition) are required for the nixtamalisation and tortilla-making processes. In a recent study, the RVA was demonstrated as a suitable rapid method to screen maize varieties for these characteristics. A range of values of each RVA parameter was obtained that allows predicting a high yield of masa and a good tortilla quality (Vázquez-Carrillo and Santiago-Ramos D 2019).

Multivariate data analysis

A further improvement on and enhancement of RVA measurements is the use of multivariate data analysis (MDA) in combination with the RVA curves or viscograms. This is in contrast to only considering the selected values, typically used when interpreting RVA results. This enables pattern recognition as well as the development of quantitative prediction and qualitative classification models.

Principal component analysis (PCA) and partial least squares (PLS) regression were used in combination with RVA curves of barley flour and malt from two different

localities (Cozzolino et al. 2012). Using MDA resulted in the extraction of more useful information to describe the starch pasting properties of the samples. Separation between samples from the two different localities could be seen in the PCA score plots. Pre-processing of the RVA profiles included baseline transformation using first or second Savitzky-Golay derivative (20-point smoothing) resulted in improved prediction of hot water extract (%) with a correlation coefficient of 0.8 and a standard error of cross-validation of 0.58%.

Using locally weighted partial least squares (LW-PLS) regression, successful calibration models were developed from RVA viscograms that predicted maize hardness irrespective of the RVA profile (hard, soft and standard) used (Guelpa et al. 2015). The models predicted the conventional hardness results with accuracies comparable to those of the laboratory errors.

The difference in the composition of rice types is reflected in their pasting properties. Zhu et al. (2018) applied multivariate data analysis to obtain more information from the RVA viscograms of 152 rice samples. PCA and partial least squares discriminate analysis (PLS-DA) enabled classification of two rice subspecies groups. A classification accuracy of 100% was obtained when the model was tested on 60 unknown samples.

First derivatives have been used with success as pre-processing technique to enhance the information generated by the RVA profile (Juhász and Salgó 2008). The viscograms were transformed with first derivative (Norris derivative method, including smoothing) which enabled better understanding of the physical/chemical interactions in starch systems. Near-infrared spectroscopy spectra and RVA curves both provide information physicochemical changes in samples. These methods were used in association to study the germination process in wheat (Juhász et al. 2005).

Conclusions

The RVA is a useful instrument increasingly being used for the determination of the viscous properties of cereals. Many studies aim to optimise the conditions of analysis to provide profiles to be used for measurement of different commodities or samples. This paper reviewed the effect of operating conditions on the RVA viscosity parameters. It also illustrated the versatility of the RVA enabling different temperature profiles for a particular product or application. Unique methods can thus be established for each product or sample. This review elaborates on the principles of operation and key parameters determined during RVA analysis. Maximising the use of the RVA as an analytical technique requires establishing the relationships between starch (referring predominantly to the AMY and AP contents) and its pasting properties with end-use qualities of a product or commodity. Furthermore, advantages of the RVA opposed to other rheological methods are presented and this review provides an overview of RVA applications with respect to wheat, barley, rice and maize. The discriminating ability of RVA tests in association with multivariate data analysis is acknowledged illustrating the usefulness of the RVA not only for routine analysis and quality control tests, but also for research.

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Compliance with Ethical Standards

Conflict of Interest Sandra Balet declares that she has no conflict of interest. Anina Guelpa declares that she has no conflict of interest. Glen Fox declares that he has no conflict of interest. Marena Manley declares that she has no conflict of interest.

Ethical Approval This article does not contain any studies with human participants or animals performed by any of the authors.

Informed Consent Not applicable

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