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Studies on preparation and functional properties of carboxymethyl starch from sorghum

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Abstract

Attempts were made to explore the potential of hybrid sorghum (Variety CSH-9) to prepare starch by wet milling process. Starch yield from sorghum found to be 72.5 percent. The starch so prepared was further utilized for the production of carboxymethyl starch (CMS) derivative. The reaction conditions during chemical modification of sorghum starch such as reaction time and monochloroacetic (MCA) acid concentration were optimized. Reaction time of 90 min. and 15 ml of 40 percent MCA acid (w/v) resulted in the maximum DS of CMS. The functional properties of CMS like swelling power, solubility and rheological properties were studied. The solubility and swelling power of sorghum CMS were maximum at 70°C. Apparent viscosity of sorghum CMS yielded 1172 Cps at 90°C using shear rate 4.51 per second. The viscosity of CMS was found to be less as compared to native starch.

Introduction

Sorghum (*Sorghum bicolor* L. Moench) is one of the major cereal crops in India after rice and wheat. Sorghum is used both as food and feed due to its carbohydrate content and starch is the principal component in it. Sorghum is rich source of starch ranging from 62.6 to 73.3% and posses better physicochemical pasting characteristics and amylolytic susceptibility (Wankhede *et al*., 1990; Gopalan *et al*., 1994). Intensive research work carried out on the isolation of starch from sorghum grains revealed that sorghum is the best source of starch (Watson, 1967; Akingbola *et al*., 1981; Carcea, 1992) and has the potential to replace other sources for starch manufacture. Starch is principal component in many fabricated or engineered food products. Native starches have their own limitation in functional attributes while modified starches are tailor made for specific function (Light, 1990). Native starches may be subjected to modification procedures to provide the desired properties for specific applications. Various types of modification procedures have been developed including physical, enzymatic and chemical modifications (Hofrieter, 1986; Sajilata, 2005; Kavlani *et al*., 2012). Carboxymethylation of polysaccharide (Chemical modification method) is a vital and versatile transformation since it provides access to water soluble polymers and intermediate with valuable functional attributes and most widely used food colloids. Carboxymethyl starch (CMS) is a starch derivative in which the –OH groups of the starch molecule, are partially substituted by ether group (-O-CH_2COOH). It exhibits varying degree of viscosity depending on its degree of substitution (DS). Food starches typically have DS range of 0.01 to 0.5 (Stojanovic *et al*., 2000). CMS has wide applicability due to high ranges of viscosity and stability (Sloan *et al*., 1962; Dong-fang *et al*., 2005). This article describes the preparation of CMS from sorghum variety CSH 9, its some functional properties to explore sorghum as cheap and economical source of starch, its derivative source for food, pharmaceutical and textile industries in tropical region.

Materials and Methods

Sorghum (*Sorghum bicolor* L. Moench) variety CSH 9 grains were procured from Sorghum Research Station, Marathwada Agricultural University, Parbhani, India. Chemicals used in the investigation are of analytical grade. Proximate composition of sorghum grains (moisture, protein, crude fat, crude fiber and ash content) was determined as given in AOAC 1990. Total carbohydrate estimation was essentially performed by the method suggested by Dubois *et al.* (1956) as modified by Wankhede *et al*. (1976). Starch content was estimated as per the

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method McCredy (1950). Isolation and purification of starch from sorghum grains were carried out by the method of Wankhede *et al.* (1979). Iodine affinity of starch was estimated by the method suggested by Schoch (1964).

Preparation of carboxymethyl starch (CMS)

Preparation of carboxymethyl starch was carried out by the method suggested by Khalil *et al.* (1990) with some modifications. 5 g of sorghum starch was dispersed in aqueous solution (iso-propanol : water - 80:20) and required pH was adjusted with 2N NaOH solution. Specific volume of MCA acid (40% w/v) was added to the suspension and incubated at room temperature (30°C) for specified time with intermittent stirring. Carboxymethylation was performed under nitrogen atmosphere to avoid degradation of polymer which takes place via β-alkoxy-carboxyl mechanism and found to result that the main alkaline degradation product such as β-D saccharinic acid (Whistler and Bemiller, 1958). During the experiment, attempts were made to standardize two parameters i.e. reaction time (30, 60, 90 and 120 minutes) and volume of 40% MCA acid concentration (5 ml, 10 ml, 15 ml and 20 ml) for the production of CMS from native sorghum starch.

Statistical analysis

Analysis of variance (ANOVA) was used to study the effect of reaction time and MCA concentration on DS of sorghum CMS (Panse and Sukhatme, 1985). Daniel's XL toolbox (Version 4) was used for analyzing variations (analysis of variance – ANOVA) and for Tukey's comparison of means of samples treated with same 40% MCA acid volume, but for different reaction time. Differences between mean values with probability $p < 0.05$ were recognized as statistically significant differences.

Sorghum carboxymethyl starch properties

Chemcial properties of CMS like moisture, ash, protein, crude fat (AOAC, 1990), iodine affinity (Schoch, 1964) were estimated. Degree of substitution (DS) of CMS was determined according to the procedure of Green (1963).

Swelling and solubility behaviour of CMS was determined by the method of Leach *et al.* (1959) from temperature of 40 to 80°C. The starch samples were accurately weighed in centrifuge tubes and appropriate amount of distilled water was added to tubes. The slurry was heated at different temperatures for constant time intervals (10 minute) followed by centrifugation at 4000 rpm for 10 minute. The supernatant and residues were analysed separately for solubility and swelling characteristics as adopted by Wankhede *et al.* (1977). The following formulas are used for the calculations.

$$\% \text{ solubility} = \frac{\text{Weight of starch (g)} \times 100}{\text{Weight of sample (g)}}$$

$$\% \text{ swelling power} = \frac{\text{Weight of sedimented paste} \times 100}{\text{Weight of sample} \times (100 - \% \text{ solubility})}$$

Haake's rotoviscometer (RV-20 model, Haake, Germany) was used for the determination of viscosity. Observations were recorded over different deformation speed ranging from 4.51 per second to 451 per second (total 10 numbers of determinations for shear rate) at various temperature (60 - 90°C) by using spindle MV-II sensors.

Results and Discussion

Proximate composition of sorghum grains

Sorghum genotype namely CSH 9 was analyzed quantitatively for their chemical composition. The results reported in Table 1, revealed that the starch content was found to be 72.5 percent in CSH 9. However, protein and fat content in the grains were low. The protein content of CSH 9 was observed as 9.45 percent and fat content was observed as 3.6 percent.

Chemical composition of starch

Isolation and purification of starch from sorghum variety CSH 9 was achieved by wet processing method (Wankhede *et al.*, 1979) by keeping the optimum conditions (i.e. soaking time, temperature and pH). Chemical composition of sorghum starch is presented in Table 2. Yield of sorghum starch found to be 72.5 percent. After exhaustive purification, the protein content of sorghum starch could be reduced to 1.05 percent. The results also revealed that total carbohydrate content was 95.6 percent in sorghum starch. Iodine affinity and bulk density of sorghum starch was found to be 3.9 and 0.58 g/ml respectively. These results are in good conformity with the results reported by Wankhede and Umadevi (1982).

Standardization of carboxymethyl starch production from native sorghum starch

Attempts were made to standardize parameters (i.e. reaction time and volume of solvent) for the production of carboxymethyl starch. The results pertaining to the effect of concentration of monochloroacetic acid and reaction time on the DS of CMS from genotype CSH 9 are presented in Table 3. DS was found to be in the range of 0.37 to 0.45.

Table 1. Proximate Composition of hybrid sorghum CSH 9

Parameters	Results (%)
Moisture	8.80 ±0.30
Ash	1.26 ±0.15
Protein	9.45-- ±0.68
Starch	72.50 ±0.94
Soluble sugars	3.10 ±0.26
Crude fiber	2.70 ±0.17
Crude fat	3.60 ±0.43

Results are presented as mean +SD, n = 3

Table 2. Physio-chemical composition of sorghum CSH 9 starch

Parameters	Results
Moisture (%) on wet basis	9.20 ±0.30
Ash (%)	0.26 ±0.04
Protein (%)	1.05 ±0.13
Crude fat (%)	0.83 ±0.10
Total carbohydrate (%) on dry basis	95.6 ±0.72
Iodine affinity	3.90 ±0.17
Bulk density (g/ml)	0.580 ±0.017

Results are presented as mean +SD, n = 3

Table 3. Effect of reaction time and monochloroacetic acid concentration on DS of CMS

Reaction Time	Degree of substitution (DS)			
	Volume of 40% MCA			
	5 ml	10 ml	15 ml	20 ml
30 min	0.37 ±0.01^{a}	0.39 ±0.01^{a}	0.39 ±0.01^{a}	0.39 ±0.01^{a}
60 min	0.40 ±0.01^{b}	0.40 ±0.01^{ab}	0.42 ±0.01^{ab}	0.43 ±0.02^{ab}
90 min	0.40 ±0.02^{ab}	0.42 ±0.02^{ab}	0.45 ±0.01^{b}	0.45 ±0.01^{b}
120 min	0.41 ±0.01^{b}	0.43 ±0.01^{b}	0.43 ±0.02^{b}	0.43 ±0.02^{ab}

Results are presented as mean +SD, n = 3
a-b, different superscripts within the same column indicate significant differences ($p < 0.05$) Unless and otherwise mentioned reaction is carried out at temperature of 30°C and pH of 10.5.

These results are in line with the findings reported by Jingwu *et al.* (1993) and Bhattacharya *et al.* (1995) for corn CMS starch in aqueous isopropanol reaction medium. The values of DS increased almost linearly with increasing monochloroacetic acid concentration in the reaction mixture. However, after certain extent no significant change in DS observed as function of MCA concentration at specific reaction time. Moreover, no significant changes in DS value were observed at or after 90 min reaction time at specific MCA acid concentration. The selectivity for carboxymethylation appears to be rather independent of time. These observations are in line with results reported by Kooijman *et al.* (2003) and Tijsen *et al.* (2001). The highest DS value can be obtained at 15 ml of 40% MCA acid concentration and 90 min reaction time. The same was further studied for its proximate and functional properties.

Properties of sorghum CMS starch

Proximate composition of CMS starch (DS 0.45, reaction time 90 minute, 15 ml of 40% MCA acid treated) is presented in Table 4. Percent moisture, protein, crude fat and total carbohydrate are found to be 8.40, 0.63, 0.85 and 97.2 respectively. The iodine affinity of CMS decreased as compared to native starch. The results are in good agreement with results reported by Bemiller (1993).

Table 4. Chemical Composition of CMS starch from sorghum

Parameters	Results
Moisture (%)	8.40 ±0.26
Protein (%)	0.63 ±0.07
Fat (%)	0.85 ±0.04
Total carbohydrate on dry basis	97.2 ±0.53
Ash (%)	0.21 ±0.03
Iodine Affinity	1.09 ±0.01
Bulk density (g/ml)	0.569 ±0.009

Results are presented as mean +SD, n = 3

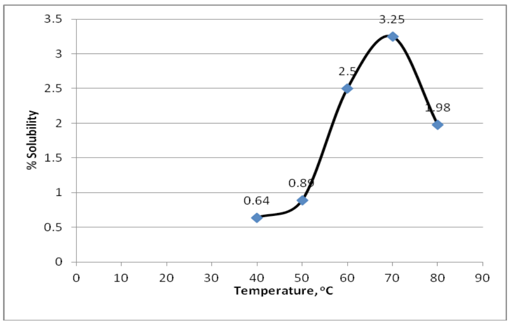

Figure 1. Effect of temperature on % solubility of sorghum CMS

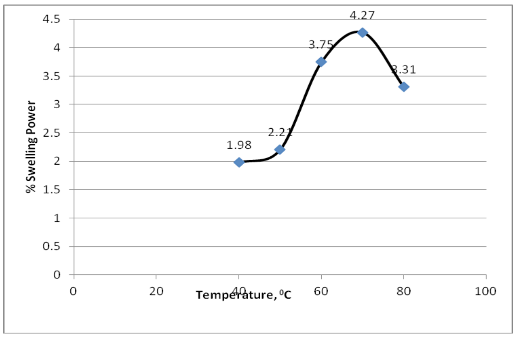

Figure 2. Effect of temperature on % swelling power of sorghum CMS

Swelling and solubility behaviour of CMS from sorghum

To study the nature of associative bonding forces within the granules the swelling and solubility behaviour in an aqueous system has been investigated. The results are presented in Fig. 1 and Fig. 2. CMS from sorghum exhibited moderate swelling in comparative higher solubility. Similarly, it is clear from the data that CMS solubility was increased as the temperature increased from 40 to 70°C and thereafter it declined. Maximum swelling and solubility was observed at 70°C. CMS solubility in water is a complex phenomenon. It mainly depends

Table 5. Effect of temperature on flow behaviour of 10% sorghum starch solution at different shear rates

Temperature (°C)	Apparent viscosity (Cps)				
	Shear rate (per second)				
	4.51	34.9	94.7	270	451
60	-	1.56	2.78	5.67	13.5
70	78.3	61.2	48.7	33.6	21.6
80	1204.7	808.3	701.8	443.1	152.7
90	2349.1	1543	810.2	690.3	427.6
After cooling at 30	4103.6	2751.2	1604.2	1303.1	853.7

Table 6. Effect of temperature on flow behaviour of 10% sorghum CMS starch solution at different shear rates

Temperature (°C)	Apparent viscosity (Cps)				
	Shear rate (per second)				
	4.51	34.9	94.7	270	451
60	76.5	4.2	2.8	2.1	1.9
70	390.4	65.6	5.9	3.8	1.9
80	750.9	290.2	160.7	80.1	85.7
90	1172	205.2	105.6	98.4	59.8
After cooling at 30	2183.4	434.5	267.3	190.8	110.4

on temperature. Observations are in good conformity with results reported by Stojanovic *et al.* (2009). In general, higher substituted CMS exhibits higher cold water solubility (Tatongjai and Lumdubwong, 2010). Moreover, the amylose content in a starch source is important as Volkert *et al.* (2004) reported lower solubility of waxy maize starch-based CMS than potato or corn-based CMS.

Rheological properties of sorghum CMS

Rheological properties are considered as important parameters to decide the performance of the said modified starch in particular industry. Knowledge of flow characteristics is important because of their effect on the finished product attributes like mouthfeel, texture and other properties. In the present study, viscosity of 10 percent sorghum starch and sorghum CMS solution over wide range of shear rates at various temperature (60 - 90°C) were recorded by using Haake's rotoviscometer (Model RV-20) and results are reported in Table 5 and 6 respectively. Results revealed that viscosity increases considerably when the temperature of starch and CMS solution increases from 60-900C. Apparent viscosity of sorghum native starch and sorghum CMS yielded 2349.1 Cps and 1172 Cps respectively at 90°C using shear rate 4.51 per second. However, it has been observed that viscosity was drastically reduced to several folds as compared to native starch. Thus, the results indicated that the incorporation of carboxymethyl functional group on the polymer yielded reduction in viscosity. It indicates that thermal stability is improved after carboxymethylation. Results are well in conformity with the results reported by Lu-feng *et al.* (2010) for kudzu root CMS.

Conclusion

From the above discussion, it can be concluded that sorghum genotype CSH 9 can be used for the production of starch. The resultant starch can be modified for the preparation of CMS of desired DS value using acid hydrolysis. In context of standardization of CMS production, Sorghum native starch treated with 15 ml of 40 percent MCA acid for 90 minutes yielded 0.45 DS CMS. The results obtained in this study will be valuable input for the development of kinetic models for the carboxymethylation process of sorghum starch. Total carbohydrate content of prepared CMS was found to be 97.2%. Maximum swelling and solubility of sorghum CMS were observed at 70°C. CMS exhibited moderate swelling in comparison to higher solubility. Rheological study of sorghum native and modified starch i.e. CMS at different temperature and shear rates characterizes application in food industry. The viscosity of sorghum CMS was observed to be significantly low as compared to that of native sorghum starch. Increased in the temperature results in increased in the viscosity with maximum values after cooling. This rheological property of prepared sorghum CMS can be explored as food additive not only to facilitate processing but also for improving food product attributes as of other modified starches do.

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References

Akingbola, J. O., Rooney, L. W., Placious, L. G. and Sweet, V. E. 1981. Thermal properties of sorghum starches. In: Proceedings of the International Symposium on Sorghum Grain quality. p. 251-261. Patancheru, Hyderabad, India: ICRISAT

AOAC. 1990. Association of official analytical chemists. Edn 12. Association of official analytical chemists : Wahington D.C.

Bemiller, J. N. 1993. Dextrins. In Macrac, R., Robinson, R. K. and Salder, M. J. (Ed). Encyclopedia of food science and nutrition. p. 4381-4384. New York: Academic Press

Bhattacharya, D., Singhal, R. S. and Kulkarni, P. R. 1995. A comparative account of conditions for the synthesis of sodium carboxymethyl starch from corn and amaranth starch. Carbohydrate Polymers 27 (4): 247-253.

Carcea, M., Cubadda, R. and Acquistucci, R. 1992. Physicochemical and rheological characterization of sorghum starch. Journal of Food Science 57 (4): 1024-1028.

Dong-fang, Z., Ben-zhi, J., Shu-fen, Z. and Jin-zong, Y. 2005. Progress in the synthesis and application of green chemicals, carboxymethyl starch sodium. Proceeding of the 3rd International Conference on Functional Molecules. p. 25-30. Dalian, China: Dalian University of Technology

Dubois, M., Gilles, K. B., Hamilton, G. K., Rebers, P. A. and Smith, F.1956. Colorimetric method for the determination of sugars and related substances. Analytical Chemistry 28 (3): 350-356.

Gopalan, C., Ramasastry, B. V. and Balasubramaniam, S. C. 1994. Tables of food composition. Nutritive value of Indian foods p. 47-58. Hyderabad, India: National Institute of Nutrition Press

Green, J. H. 1963. O-carboxymethyl cellulose. In Whistler, R.L. (Ed). Methods in Carbohydrate Chemistry vol.3. p. 322-327. New York: Academic Press.

Hofrieter, B. T. 1986. Modified starches: Properties and uses. In Wurzburg, O. B. (Ed). Miscellaneous modifications. Boca Raton, FL: CRC Press.

Jingwu, Z., Dongali, L., Yuquan, S., Xiaohong, Z. and Dahua, W. 1993. The study on the effect of alkalization on the carboxymethylation of starch. Journal of Tianjin University. 1: 124-131.

Kavlani, N., Sharma, V. and Singh, L. 2012. Various techniques for the modification of starch and the applications of its derivatives. International Research Journal of Pharmacy 3 (5): 25-31.

Khalil, M. I., Hashem, A. and Hebeish, A. 1990. Carboxymethylation of maize starch. Starch/Starke 42 (2): 60-63.

Kooijman, L. M., Ganzeveld, K. J., Manurung, R. M. and Heeres, H. J. 2003. Experimental studies on the carboxymethylation of arrowroot starch in isopropanol-water media. Starch/Starke 55 (11): 495-503.

Leach, H. W., McCowen, L. D. and Schoch, T. J. 1959. Structure of starch granule: swelling and solubility patterns of various starches. Cereal Chemistry 36: 534.

Light, J. M. 1990. Modified food starches: why, what, where and how. Cereal Food World 35 (11): 1081-1091.

Lu-Feng, W., Si-Yi, P., Hao, H., Wen-Hua M. and Xiao-Yun X. 2010. Synthesis and properties of carboxymethyl kudzu root starch. Carbohydrate Polymers 80 (1): 174-179.

McCready, R. 1970. Starch and Dextrin. In Joslyn, M. (Ed). Methods in food analysis. p. 541-563. New York: Academic Press.

McCready, R., Guggolz, J., Silviera, V. and Owens, H. 1950. Determination of starch and amylose in vegetables, application to peas. Analytical Chemistry 22 (9): 1156-1158.

Panse, V. G. and Sukhatme, P. V. 1985. Statistical methods for agricultural workers. 2nd Ed. Indian council of agricultural research: New Delhi, India.

Sajilata, M. G. and Singhal, R. S. 2005. Speciality starches for snack foods. Carbohydrate Polymers 59 (2): 131-151.

Sangseethong, K., Ketsilp, S. and Sriroth, K. 2005. The role of reaction parameters on the preparation and properties of carboxymethyl cassava starch. Starch/Starke 57: 84–93.

Schoch, T. J. 1964a. Iodometric determination of amylose, potentiometric titration: standard method. In Whistler, R.L. and Wolfrom, M.L. (Eds). Methods in carbohydrate chemistry: Satrch. Vol.4. p.157. New York and London: Academic Press.

Schoch, T. J. 1964b.Solubility and swelling power of starches. In Whistler, R.L. and Wolfrom, M.L. (Eds). Methods in carbohydrate chemistry: Starch/Starke. Vol.4. p.106-109. New York and London: Academic Press

Sloan, J. W., Mehltretter, C.I. and Senti, R.F. 1962. Carboxymethyl high amylose starch. Journal of Chemical Engineering Data 7 (1): 156-158.

Stojanovic, Z., Jeremic, K. and Jovanovic, S. 2000. Synthesis of carboxymethyl starch. Starch/Starke 52 (11): 413-419.

Stojanovic, Z ., Jeremic, K., Jovanovic, S., Nierling, W. and Lechner, M. D. 2009. Light scattering and viscosity investigation of dilute aqueous solutions of carboxymethyl starch. Starch/Starke 61: 199–205.

Tatongjai, J. and Lumdubwong, N. 2010. Physicochemical properties and textile utilization of low- and moderate-substituted carboxymethyl rice starches with various amylose content.Carbohydrate Polymer 81 (2): 377–384.

Tijsen, C. J., Kolk, H. J., Stamhuis, E. J. and Beenackers, A.A.C.M. 2001a. An experimental study on the carboxymethylation of granular potato starch in non-aqueous media. Carbohydrate Polymers 45 (3): 413-419.

Tijsen, C. J., Voncken, R. M. and Beenackers, A.A.C.M. 2001b. Design of a continuous process for the production of highly substituted granular carboxymethyl starch. Chemical Engineering Science 56 (2): 411-418.

Volkert, B., Loth, F., Lazik,W. and Engelhardt, J. 2004. Highly substituted carboxymethyl starch. Starch/Starke 56: 307–314.

Wankhede, D. B. and Tharanathan, R. N.1976. Sesame (*Sesamum indicum*) carbohydrates. Journal of Agricultural Food Chemistry 21: 655-659.

Wankhede, D.B. and Umadevi. 1982. Preparation and some of the physiochemical properties of pyrodextrins from ragi, wheat and rice starches. Starch/Starke 34: 162-165.

Wankhede, D. B., Rathi, S. S., Gunjal, B. B., Patil, H. B., Walde, S. G., Rodge, A. B. and Sawte, A. R. 1990. Studies on isolation and characterization of starch from pearl millet (*Pennisetum americanum leeke*) grains. Carbohydrate Polymers 13 (1): 17-28.

Wankhede, D. B., Saroja, R. and Raghvendrarao, M. R. 1977. New starches: Preparation and properties of starch and its fractions of two varieties of groundnuts (*Arachis hypogea*) Starch/Starke 29: 223-228.

Wankhede, D. B., Shehnaz, A. and Raghvendra, M. R. 1979. Preparation and physiochemical properties of starches and their functions from finger millet (*Eleusine coracana*) and foxtail millet (*Steria italica*). Starch/Starke 31 (5): 153-159.

Watson, S. A. 1967. Manufacture of corn and milo starch. In Whistler, R.L. and Paschall, E.F. (Eds). Starch Chemistry and Technology. vol. II. p. 1-51. New York and London: Academic Press.

Whistler, R. L. and Bemiller, J. N. 1958. Alkaline degradation of polysaccharides. In Wolfrom, M.I. and Tipson, R.S. (Eds). Advances in Carbohydrate Chemistry. Vol. 13. p. 289-329. New York: Academic Press.