Quality assessment of *Borassus aethiopum* Mart fruit pulp pectin precipitated with various solvents

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Received 28 February, 2020; Accepted 18 August, 2020

*Borassus aethiopum* Mart fruits are underutilized in Africa and most of them are left to rot in the field. These fruits have great potential as an alternative and commercially viable pectin source for the pectin industry. Physicochemical and rheological characteristics of *B. aethiopum* pectins are significantly affected by the extraction process which permits the isolation of tailored pectin of specific applications in the food industry. One important step in pectin recovery is its precipitation from the liquid extract and this can considerably impact the quality attributes of the final product. In this study the effects of precipitating solvents (isopropanol, acetone and 50/50 isopropanol-acetone) on the physicochemical and functionalities of pectin recently extracted from *B. aethiopum* fruit were investigated. Most of the results for the physicochemical characteristics were not statistically different however great variability was noticed in the functional and rheological properties of pectins precipitated with isopropanol (IPA), acetone (ACTN) and the 50/50 IPA-ACTN solvents. Isopropanol precipitated pectin exhibited statistically higher (p<0.05) emulsifying activity and a better gel sensorial property than the ACTN and IPA-ACTN precipitated pectins. Moreover, regardless of the precipitating solvent, high purity pectin with high viscosifying, emulsifying and gelling properties were obtained. Therefore, production of pectin from *B. aethiopum* fruit pulp must be governed by its intended use since extraction and precipitation processes isolate pectin samples with various functionalities.

Key words: *Borassus aethiopum* Mart, isopropanol, acetone, precipitating solvent, solvent mixture.

INTRODUCTION

The palm *Borassus* is a dioecious plant that belongs to the Arecaceae family. The species *Borassus aethiopum* Mart is mostly found in the savannah region of Côte d’Ivoire where it grows wild. A single tree of *B. aethiopum* can bear around 6 to 12 bunches of about 50 fruits per year. Ripe and mature fruits are large ovoid drupes with diameter between 15 to 20 cm and one fruit weighs about 1.5 kg. Because of its high water content and lack of effective conservation methods, more than 60% of the fruits are lost during the harvesting period (Ali et al., 2010).

Recent studies aiming at valorizing the fruits and
therefore reducing the post-harvest losses have been reported. The fresh fruit of *B. aethiopum* has 80% water and contains 42.6 mg/100 g, 12 mg/100 g and 33 mg/100 g of potassium, sodium and calcium respectively. The energy calorie is 72.1 Kcal/100 g and the pulp is rich in vitamin C (17.68 mg/100 g), vitamin A (445.8 µg/100 g) and carotenoid (5350 µg/100 g) (Oryema and Oryem-Origa, 2016). By using soft drying conditions, it is possible to produce flour with acceptable technological attributes and economically exploitable (Ali et al., 2010). Oven, solar or freeze-dried flours of *B. aethiopum* fruit pulp present good flowability and can potentially be used as ingredients in the manufacture of pasta, puddings, cakes, biscuits, breads, crackers, and doughnuts (Abe-Inge et al., 2018a). Moreover, the antioxidant properties as well as the mineral, anti-nutrient and phytochemical compositions of these flours were reported by Abe-Inge et al. (2018b). Juice with acceptable sensory attributes can be produced by enzymatic hydrolysis of *B. aethiopum* fresh fruit pulp puree at 35°C for 2 h (Koffi et al., 2010).

*B. aethiopum* fruit is described as a good source of pectin. Yield and galacturonic acid content of pectin extracted from the fruit pulp at various pH (natural pH of 5.2-5.5, 2.5, 7), temperature (70, 80 and 90°C) and time (30-120 min) were 47-149 g kg⁻¹ and 808-852 g kg⁻¹ respectively. This high content of galacturonic acid makes *B. aethiopum* pectin an excellent raw material for industrial production of pectin (Assoi et al., 2014). *B. aethiopum* pectin forms gel in presence of acid and sucrose despite its high degree of acetylation (~5%) (Assoi et al., 2016). The gels produced were very stable thus indicating strong intermolecular associations between *B. aethiopum* pectin molecules during network formation (Ibănescu et al., 2010). When compared to commercial pectin Fluka (76280, Sigma-Aldrich), gel with equivalent characteristics was noticed. Furthermore, *B. aethiopum* pectin exhibits good emulsifying properties probably because of its high protein (80 g kg⁻¹) and galacturonic acid content (Assoi et al., 2016).

Pectin is used in the manufacture of jam, jellies, and marmalade; low sugar and calorie foods; bakery; meat packing; acidified milk drinks; and is also used as emulsifier and stabilizer (Voragen et al., 2009, 1995). Commercial production of pectin is typically derived from apple pomace and citrus peels. After extraction, pectin precipitation from the mother liquor is generally done with alcohol. However, at the laboratory level, various organic solvents as well as salts of polyvalent metals and physical techniques are used to evaluate the appropriate precipitation conditions of pectin from a specific plant material. Several studies aiming at exploring the impact of precipitating solvents on the yield and physicochemical indices of pectin are available in the literature. The work of Hunt (1918) reported comparable yield when apple pectin was precipitated with ammonium sulfate and alcohol. Saulnier and Thibault (1987) recovered pectin from the pulp of grape berries through cupric ion precipitation. Muminov (1997) used organic solvents and salts of polyvalent metals to determine the best precipitating solvents for pectin extracted from the valves of cotton bolls. Kravtchenko et al. (1992b) reported the recovery of more purified commercial apple and lemon pectins after precipitation with copper acetate. Alcohol and aluminum-chloride precipitation of galgul (Citrus pseudolimium) pectin affected the physicochemical characteristics and functional properties of the purified pectin (Attri and Maini, 1996). The quality, composition and physicochemical properties of yellow passion fruit ring pectin precipitated by dialysis or with alcohol and metal ion were also reported by Yapo (2009). The influence of pH on sugar beet pectin precipitation was mentioned by Guo et al. (2016a). Moreover, coexistence of five pectin fractions of different chemical and molecular characteristics have been identified when a stepwise ethanolic-precipitation was used during the recovery of sugar beet pectin from the water soluble extract (Guo et al., 2016b). Nagel et al. (2017) used selective precipitation steps to recover mango peel pectin from the liquid extract. Also, to establish an alternative method for pectin recovery from pomelo albedo residues, Vega et al. (2018) evaluated the effect of methanol, ethanol and 1-propanol on the quality of the precipitated pectin.

Therefore, since pectin was extracted for the first time from Palmyra palm (*B. aethiopum* Mart) (Assoi et al., 2016) the need to examine the impact of precipitating solvents on the quality attributes of that pectin must be determined to complement the work started on pectin extraction from this potential raw material. In this study pectin isolated at room temperature and under the natural pH of the fruit (5.2-5.5) was precipitated with isopropanol, acetone and the 50/50 mixture of the two solvents; and their effects on the physicochemical, functional, and rheological properties of *B. aethiopum* pectins were analyzed.

**MATERIALS AND METHODS**

**Sample preparation, *B. aethiopum* pectin extraction and recovery**

Mature, ripe fruits of Palmyra palm (*B. aethiopum* Mart), that weighed between 1.4 and 2 kg, were purchased at a local market in Dimbokro (Côte d’Ivoire). After cleaning, the fruit pulp was separated from the skin and kernel using a knife. An aliquot of 77 fruits was randomly selected which had a total weight of 100.4 kg and a yield of 39.10 kg of pulp was obtained. The pulp was cut into small pieces and dried in a Memmert laboratory oven (ULE 500, Memmert, Schwabach, Germany) at 60°C for 48 h (Agbo and Simard, 1992) and packed in plastic storage bags before being shipped to the Food Biotechnology Laboratory at Alabama A & M Huntsville, AL, USA where it was ground, alcohol washed and stored at room temperature until use. Pectin extraction was done as described by Assoi et al. (2016). Briefly, the alcohol insoluble solids (1 g) was dispersed in water (25 ml) and pectin was extracted using a shaking water bath (model 50, Thermo Fisher Scientific Inc. Waltham, MA, USA) maintained at room temperature. After
Proximate analysis and physicochemical characterization of *B. aethiopum* pectin

Pectin moisture was determined after drying in a Precision Telco laboratory oven (Winchester, VA, USA) according to the method of Kalapathy and Proctor (2001). Ash content was determined by incinerating pectin sample (1 g) in a muffle furnace (Lindberg Blue M, Ashville, NC, USA) at 660°C overnight. Pierce BCA (bicinchoninic acid) protein assay kit (Pierce Biotechnology, Rockford, IL, USA) was used for protein analysis. Galacturonic acid content (GalA) was estimated colorimetrically using m-phenyl phenol; degree of methylolation (DM) and degree of acetylation (DaC) were estimated by methanol released after saponification; individual neutral sugars (arabinose, rhamnose, xylose, mannose, galactose and glucose) were estimated from per-O-trimethylsilyl derivatives by gas chromatography–mass spectrometry. Molecular weight (Number average molecular weight, Mn; Weight average molecular weight, Mw) and polydispersity were estimated by laser light scattering (Dawn Heleos® II) and refractive index Optilab-T-Rex detectors (Wyatt Technology, Santa Barbara, CA) using a PL Aquagel-OH size exclusion columns (Ailgnet Technologies, Santa Clara, CA) as described by Corredig and Wicker (2001). Intrinsic viscosity of diluted pectin samples (10-50 g kg⁻¹) prepared in 0.1 mol/L sodium phosphate buffer (pH 7) was determined according to a modified protocol described by Constenla et al. (2002). Samples were analyzed at 25°C and constant shear stress of 0.1 Pa using a controlled stress dynamic rheometer Rheometric Scientific model SR-500 (Rheometrics, Piscataway, New Jersey, USA). A concentric cylinder geometry with a cup diameter of 32 mm; bob diameter of 29.5 mm, and bob length of 44.5 mm was used during the test. Zeta potential, which provides information on the surface charge of polymers in solution, was measured using a Particle Size Analyzer coupled with the BI-Zeta option (90 Plus, Brookhaven Instruments Corporation, Holtsville, NY, USA), and equipped with a 50 mV diode laser (90 angle) and a BI-9000 AT correlator. Refractive index and laser beam were set at 1.330 and 659.0 nm and the temperature at 25°C (Kim and Wicker, 2011). Before running the analysis, 3 ml of the aqueous dispersion of pectin (5 g kg⁻¹) containing 0.2 g kg⁻¹ of sodium azide was filtered through a 5.0 μm filter (Millipore, Bedford, MA) after pH adjustment to 4.

Functional and rheological properties of *B. aethiopum* pectin

Emulsifying activity was determined according to the modified method of Dalev and Simoneonova (1995). Aliquot (3 ml) of aqueous pectin solutions (5 g kg⁻¹) containing 0.2 g kg⁻¹ of sodium azide was homogenized for 3 min with 3 ml of vegetable oil. The oil-in-water emulsion was centrifuged at 600 × g for 5 min using Allegra X-22R centrifuge (Brea, CA, USA). The whole volume (Wv) of the emulsion and the volume of the emulsified layer (ELv) were used to calculate the emulsifying activity (EA) as follows:

$$EA (\%) = \frac{(ELv / Wv)}{100}$$

All rheological analyses were performed with a ARG-2000 controlled stress rheometer (TA instrument, New Castle, Delaware, USA) mounted with a truncated cone (diam. 40 mm; angle 2º; truncation 54 μm) and peltier geometry. Solvent trap was used to prevent drying of the sample during the test. Steady shear flow behavior test was performed to determine the apparent viscosity of 20 g kg⁻¹ *B. aethiopum* pectin solution prepared by dissolving pectin in distilled water at room temperature for 1 h by magnetic stirring. Data was collected over a shear rate range of 10⁴ to 10000 s⁻¹ at 25°C. Viscoelastic behavior of *B. aethiopum* aqueous solution was analyzed through small oscillatory deformation test. Stress sweep test conducted at 25°C and constant frequency of 1 Hz was performed to estimate the maximum deformation attainable by a sample in the linear viscoelastic region (LVR). Frequency sweep test was carried out in the frequency range of 0.1 to 10 Hz at constant stress (0.35 Pa) selected within the LVR. The mechanical spectra of the solution was obtained by reporting the elastic modulus (G') and viscous modulus (G'') as a function of frequency. *B. aethiopum* gels properties were determined on 1 g/100 g pectin gels prepared in a sealed vial. Pectin samples and 65 g/100 g of dried sucrose were dissolved in citrate buffer (pH 3). The mixture was heated for 30 min in boiling water under continuous stirring and kept overnight at room temperature for gel formation. Textural properties of *B. aethiopum* pectin gels were assessed through oscillatory measurements performed at 25°C and 0.35 Pa selected in the LVR. The magnitude of tan delta, elastic (G') modulus and viscous modulus (G'') were recorded and analyzed to describe the quality attributes of the gels (strength, softness).

Statistical analysis

Data were analyzed for comparison of the means using SAS 9.2. Differences in the means were obtained using the Tukey test at α = 0.05, values are expressed as means ± standard deviation of the means.

RESULTS AND DISCUSSION

Effect of precipitating solvents on yield and physicochemical characteristics of *B. aethiopum* pectins

In this study, water soluble pectin was extracted from *B. aethiopum* fruit pulp for 30 min at room temperature and natural pH (5.2-5.5) of the fruit. To improve the yield and purity of *B. aethiopum* pectin, isopropanol (IPA), acetone (ACTN), and the 50/50 mixture of the pure solvents were used to recover pectin from the liquid extract. At industrial level, pectin precipitation from the mother liquor is generally done using organic solvents such as IPA or ethanol (May, 1990). The use of ACTN as pectin precipitant was mostly reported in the literature (Suhaila and Zahariah, 1995; Muminov, 1997; Aina et al., 2012). Good yield, color and gelling characteristics were reported for pectins extracted from various tropical agrowastes when acetone was used as precipitating solvent (Suhaila and Zahariah, 1995). Therefore, IPA and ACTN were selected to assess their effects on the physicochemical and functional properties of *B. aethiopum* pectin. In addition, mixture of the pure solvents (50/50) for pectin precipitation was also investigated.
Table 1. Yields, proximate composition and physicochemical characteristics of *B. aethiopum* pectin precipitated by the 3 solvents.

| Parameter                  | Precipitating solvents |
|----------------------------|------------------------|
|                            | Isopropanol | IPA-ACTN (50/50) | Acetone |
| Yield (g kg⁻¹)             | 102.3 ± 0.04 | 96.2 ± 0.03 | 105.8 ± 0.11 |
| Moisture (g kg⁻¹)          | 76.1 ± 0.02 | 64.3 ± 0.28 | 67.2 ± 0.31 |
| Ash (g kg⁻¹)               | 35.0 ± 0.03 | 35.3 ± 0.01 | 41.9 ± 0.01 |
| Protein (g kg⁻¹)           | 87.4 ± 0.11 | 87.2 ± 0.27 | 90.5 ± 0.05 |
| GalA (g kg⁻¹)              | 845.2 ± 2.17 | 889.1 ± 3.16 | 909.0 ± 1.62 |
| DM (%)                     | 71.92 ± 2.61 | 71.35 ± 2.46 | 68.31 ± 1.56 |
| Dac (%)                    | 5.25 ± 0.54 | 4.14 ± 0.18 | 5.07 ± 0.64 |

**Molecular weight**

**Firstly eluted pectin molecules**

| Parameter                  | Isopropanol | IPA-ACTN (50/50) | Acetone |
|----------------------------|-------------|------------------|---------|
| Polydispersity             | 1.48        | 1.24             | 1.41    |
| Mn × 10⁵ (g mol⁻¹)         | 2.02ᵃ       | 1.61ᵇ            | 2.15ᵃ   |
| Mw × 10⁵ (g mol⁻¹)         | 2.92ᵃ       | 1.97ᵇ            | 3.00ᵇ   |

**Secondly eluted pectin molecules**

| Parameter                  | Isopropanol | IPA-ACTN (50/50) | Acetone |
|----------------------------|-------------|------------------|---------|
| Polydispersity             | 1.0         | 1.03             | 1.0     |
| Mn × 10⁵ (g mol⁻¹)         | 1.44ᵃ       | 1.07ᵇ            | 1.51ᵇ   |
| Mw × 10⁵ (g mol⁻¹)         | 1.46ᵃ       | 1.11ᵇ            | 1.52ᵇ   |
| Intrinsic viscosity (ml g⁻¹)| 291.7ᵇ     | 316.56ᵃ          | 295.67ᵇ |
| Zeta potential (mV)        | -25 ± 0.12  | -22 ± 0.95       | -24 ± 0.83 |

GalA: Galacturonic acid; DM: Degree of methylation; Dac: Degree of acetylation; Mn: Number average molecular weight; Mw: Weight average molecular weight; IPA-ACTN: Isopropanol- Acetone. Values are mean±SD;ᵃᵇMeans in a row with the same superscript are not significantly different (p > 0.05).

Pectin purity is mostly based on GalA content and lack of ash. In this study, statistical analysis showed no significant difference (p>0.05) in the yield (96-106 g kg⁻¹), moisture (64 -76 g kg⁻¹), ash (35-42 g kg⁻¹) and protein (87-91 g kg⁻¹) of the *B. aethiopum* pectin precipitated with the different solvents (Table 1). It is noted that despite the cold extraction process *B. aethiopum* pectin contained high level of protein and this could suggest the presence of elevated amount of protein into the ripe and mature fruit. This protein may exist either in free form or bound to the arabinogalactan moiety (Oosterveld et al., 2002) probably liberated during the ripening process (Sirisomboon et al., 2000, Steele et al., 1997). Lower ash content in pectin sample is desired (Ceylan et al., 2017) because of its ability to negatively affect gel formation. Ash maximum limit to produce good quality gel is 100 g kg⁻¹ (Huda, 2016). El-Nawawi and Fadia (1988) reported a decrease in the jelly grade of Egyptian orange peel pectin due to an increased amount of ash in the pectin. Ash content of *B. aethiopum* pectins was below the maximum limit regardless of the solvents used.

GalA content of *B. aethiopum* ranged from 845 g kg⁻¹ (IPA) to 909 g kg⁻¹ (ACTN). Acetone and the 50/50 solvent mixture provided pectin with significantly (p<0.05) higher GalA content as compared to the IPA solvent (Table 1). High GalA is known to improve the functional properties of the biopolymer. GalA content of *B. aethiopum* pectin was higher than the maximum limit (65%) defined for commercial pectin (May, 1990). Pectin with high purity was extracted from *B. aethiopum* fruit pulp as indicated by the high GalA content and low ash.

*B. aethiopum* fruit pulp pectins were highly methylated (HM) and acetylated, and values ranged from 68.31% (acetone) to 71.92% (isopropanol) and from 4.14% (50/50 IPA-ACTN) to 5.25% (IPA) respectively. The degree of methylation (DM) and the degree of acetylation (Dac) were not significantly affected (p>0.05) by the precipitating methods (Table 1). Based on their DM, ACTN pectin can be classified as medium rapid set pectin and both IPA and 50/50 IPA-ACTN pectins as rapid set pectins (Thibault and Ralet, 2003).

Neutral sugar (NS) content of precipitated *B. aethiopum* pectin is presented in Figure 1. NS contributes to the functional (gelling and rheological) properties of pectin in the food system (Hwang et al., 1993). Acetone precipitated *B. aethiopum* pectin was richer (7.95 g/100 g) in neutral sugars than isopropanol precipitated (5.18 g/100 g) pectin. The lowest neutral sugar content (3.8 g/100 g) was observed for *B. aethiopum* pectin precipitated with the 50/50 mixed solvent. These data showed a partial purification of *B. aethiopum* pectin by the pure solvents as compared to the 50/50 mixed solvent (Kravtchenko et al., 1992b). Acetone solvent probably allowed the co-precipitation of higher amount of free neutral polysaccharides; such as xyloglucans, arabinans, arabinogalactans, and mannans as compared...
to the 70% isopropanol solvent. A co-precipitation of salts, free proteins and sugars has also been reported (Brigand et al., 1990; Garna et al., 2007). The efficiency of isopropanol solvent at reducing co-precipitation of impurities in B. aethiopum pectin was about 1.3 times improved when isopropanol was mixed with acetone rather than water. Consequently, the mixture of acetone and isopropanol solvent, in the ratio of 50:50, which helped to obtain B. aethiopum pectin with low amount of impurities, could be better choice.

A bimodal distribution of the molar mass was observed, regardless of the precipitating solvent, thus revealing two distinct populations of the B. aethiopum pectin polymers. First eluted pectin population was composed of high molecular weight polymer chains \( (1.97 \times 10^5 - 3.0 \times 10^5 \text{ g mol}^{-1}) \) while the second eluted population contained polymers chains of much lower molecular weight \( (1.11 \times 10^5 - 1.52 \times 10^5 \text{ g mol}^{-1}) \) (Table 1). Molecular weights of pectin precipitated by the pure solvents were statistically higher \( (p<0.05) \) than the molar mass of the pectin precipitated with the solvent mixture.

Polydispersity (PD) provides information on the uniformity of the polymer chain lengths, and it is calculated as the ratio of Mw and Mn. For PD closers to 1 all polymers chains approach uniform chain length. When PD falls between 1.01 and 1.2, low variation in chain length is observed. Higher variability in chain length is reported when PD > 1.4 (Whitfield et al., 2019). PD of B. aethiopum pectin varied from 1.24 (IPA-ACTN) to 1.48 (IPA). Pectin precipitated with the 50/50 solvent mixture was less polydisperse than the others, and it also displayed the lowest Mw probably due to the removal of the covalently unlinked substances to the pectin molecules. These impurities may have contributed, to a certain extent, to the relatively high molar mass of pectin precipitated with acetone and isopropanol (Berth, 1988; Kravtchenko et al., 1992a). This result was in a close agreement with observations previously reported for the neutral sugars, intrinsic viscosity and molar mass analysis. Comparable results were also reported by Kravtchenko et al. (1992b) when attributes of the aqueous copper acetate purified commercial pectin was compared to that of the alcoholic purified one.

Intrinsic viscosity, which specifies the hydrodynamic volume of the individual polymer molecule in solution, is reported by using the Solomon-Ciuta equation (Pamies et al., 2008) because conditions for a linear extrapolation to zero polymer concentration could not be met (Evageliou et al., 2005). The 50/50 IPA-ACTN precipitated B. aethiopum pectin \( (\text{Mw of } 1.97 \times 10^5 \text{ g mol}^{-1}) \) exhibited the highest intrinsic viscosity \( (316.56 \text{ ml g}^{-1}) \) as compared to isopropanol \( (291.7 \text{ ml g}^{-1}, \text{Mw of } 2.92 \times 10^5 \text{ g mol}^{-1}) \) and

![Figure 1. Individual neutral sugar (g/100 g) of B. aethiopum pectin extracted at room temperature and pH natural pH of the fruit and precipitated with Isopropanol, Acetone or 50/50 isopropanol/acetone. Symbols: Arabinose ( ), Rhamnose ( ), Xylose ( ), Mannose ( ), Galactose ( ), Glucose ( ).](image-url)
acetone (295.67 ml g⁻¹, Mw of 3.00 × 10⁵ g mol⁻¹) precipitated ones (Table 1). Since the intrinsic viscosity of polymer solutions is mostly related to the volume of the polymers in solution (Round et al., 2010); it could be inferred that the lower intrinsic viscosity exhibited by the higher Mw pectin samples may be due to the presence of compact polymers in the solution (Koliandris et al., 2010).

Zeta potential, which illustrates the magnitude of charges at a specific pH on the surface of a particle in colloidal dispersion, derives mostly from the chemical ionization of functional groups in solution (Alkan et al., 2005). All aqueous solutions of B. aethiopum pectin exhibited negative zeta potential values at pH 4. IPA (-25 mV) and ACTN (-24 mV) precipitated pectins exhibited more negatively charged surface (p<0.05) than the 50/50 IPA-ACTN precipitated pectin (-22 mV). At pH of 4, carboxyl groups are dissociated and pectin molecules carry a negative surface charge. The density of charge depends both on the DM of the pectin and the pH used (Schmidt et al., 2016). DM of B. aethiopum precipitated pectins were not statistically different but IPA and ACTN precipitated pectin exhibited statistically higher negative surface charge than the 50/50 IPA-ACTN precipitated pectin (Table 1). This difference could be related to a different distribution of the negatively charged carboxyl groups within the pectin molecule (Gawkowska et al., 2018). Indeed, a blockwise charge distribution generates a more negatively surfaced charge than a randomly distributed charged carboxyl groups (Lutz et al., 2009). A high zeta potential is a sign of greater stability of the aqueous dispersion of pectin (Pacheco et al., 2019). Therefore, more stable aqueous dispersions will be obtained with the IPA and ACTN precipitated pectins than the 50/50 IPA-ACTN precipitated one.

### Effect of precipitating solvent on functional and rheological properties of B. aethiopum pectins

Emulsifiying activity (EA) of the B. aethiopum pectin ranged from 52.1% (acetone) to 62.1% (isopropanol). B. aethiopum pectin with statistically (p<0.05) higher EA was precipitated by the isopropanol solvent. Precipitation with acetone and the 50/50 IPA-ACTN solvent led to the isolation of B. aethiopum pectin with reduced EA (Table 2). All of the pectin samples, at concentration of 5 g kg⁻¹, exhibited high emulsifying activities (EA> 50%) probably due to their high protein content (<90 g kg⁻¹). B. aethiopum pectins can be considered as true emulsifiers as they were able to adsorb and form at the surface of the newly formed oil droplets a protecting layer which prevents them from coalescing with neighboring oil droplets (Ngouémazong et al., 2015).

Precipitating solvents significantly affected (p<0.05) the rheological properties of the B. aethiopum pectins. Apparent viscosity of B. aethiopum pectin solutions decreased with increasing shear rates (Figure 2), and this is an indicative of a shear thinning flow behavior. Similar flow behavior was reported by Hwang and Kokini (1992) for commercial apple pectin prepared by precipitation with Cu(II) followed by 1.0% acid alcohol washing or EDTA treatment. Thinning of a polymer solution during shear is the result of the disruption of network and alignment of the polymers in the direction of the flow (Dangi and Yadav, 2020; Pacheco et al., 2019). For all pectin samples, the Newtonian plateau (NP) was not observed probably because the concentration used was high enough to shift the NP toward much lower values than the lowest shear rate available. The same trend was reported by Razavi et al. (2011) for sage seed gum. At very low shear rate, apparent viscosity of acetone precipitated pectin solution was statistically higher (4.71 Pa.s) than that of IPA (3.57 Pa.s) and 50/50 IPA-ACTN (3.08 Pa.s) precipitated pectins. This higher viscosity, at similar concentration (20 g Kg⁻¹), suggests the presence of higher intermolecular interactions between ACTN precipitated pectin chains at near-zero shear rates (Chen et al., 2020). When the shear rate was increased, a rapid decrease of the apparent viscosity was observed for the acetone precipitated pectin solution. This might reflect a much weaker association of the acetone precipitated polymers in solution (Sato et al., 2004). The overlapping of the graphs obtained with isopropanol and 50/50 IPA-ACTN precipitated pectin may reveal the formation of

| Parameter | Precipitating Solvents | Isopropanol | IPA-ACTN (50/50) | Acetone |
|-----------|-------------------------|-------------|-----------------|---------|
| Emulsifying activity (%) | 62.1±0.59 | 56.3±0.05 | 52.1±2.95 |
| Apparent viscosity (Pa.s) of 20 g/100 kg⁻¹ pectin solution at near zero shear rates | 3.57b | 3.08b | 4.71b |
| Elastic modulus (Pa) of 1 g/100 g gel (obtained at 1 Hz) | 1634a | 1497b | 1611a |
| Tan delta of 1 g/100 g gel | 0.18-0.23 | 0.2-0.25 | 0.18-0.23 |
| Sensory property (G’/G”) of 1 g/100 g gel | 5-6 | 4-5 | 4-5 |

IPA / ACTN: Isopropanol/Acetone; Pa: Pascal; G**: Elastic modulus; G”: viscous modulus; Hz: herz. Values are mean±SD; a,b,cMeans in a row with the same superscript are not significantly different (p> 0.05).
network structures of comparable characteristics (Assoi et al., 2016).

Results of the oscillatory tests revealed that viscoelastic properties of *B. aethiopum* pectin solutions were affected by the precipitating solvents. Stress sweep results (Figure 3) exposed viscoelastic liquid-like characteristics (G’ > G”) for *B. aethiopum* pectin solutions (20 g kg⁻¹). Moreover, at very low shear stress (0.01 - 1 Pa), acetone precipitated pectin solution exhibited higher elastic modulus (G’) than IPA and 50/50 IPA-ACTN precipitated pectin solutions. Since network strength is measured by the magnitude of G’ (Ström et al., 2014), it could be inferred that sufficient entanglements were formed between acetonate precipitated pectin polymers chains thus leading to the formation of a continuous network (Tam et al., 1999). At shear stress beyond 1 Pa (critical stress), G’ of acetone precipitated pectin decreased faster than that of the others, thus revealing weak associations of the pectins polymers. Mechanical spectra displayed in Figure 4 is showing changes in elastic moduli (G’) and viscous moduli (G”) with frequency variation. All pectin solutions exhibited an increase in the magnitude of G’ and G” with increasing frequency which is a characteristic of fluid-like behavior typical of random-coil entanglement networks. Moreover, G’ and G” tend to converge at higher frequency thus indicating network formation between polymers.

Variability in the properties of the gel (1 g/100 g) prepared with the precipitated *B. aethiopum* pectins was also noticed (Table 2 and Figure 5). All the precipitated pectins formed a gel in presence of 65 g/100 g of sucrose at pH of 3. Under Stress sweep test, the gel elastic modulus (G”) varied from 1497 Pa (50/50 IPA-ACTN) to 1634 Pa (IPA). Pectin precipitated with the pure solvents exhibited greater G’ than the 50/50 IPA-ACTN precipitated pectin, thus revealing the formation of denser network with more interconnected polymers chains. Gelling and thickening aptitude of pectin is generally assessed by its ability to form interconnected network in solution and the occurrence of these entanglements depends greatly on several intrinsic and extrinsic factors (Dangi and Yadav, 2020). The differences observed in the gel properties could be attributed to the low Mw and neutral sugar content of the pectin precipitated by the mixed solvents (Abid et al., 2017). Therefore it could be assumed that compounds not precipitated by the 50/50 solvent mixture may have contributed to the increase in the elastic modulus of the gel prepared with *B. aethiopum* pectin precipitated with each of the pure solvents.

For all *B. aethiopum* pectin gels, the frequency sweep results displayed the characteristic spectrum of gel-like structure (G’ > G”) (Figure 5) with tan delta around 0.2 (Table 2). Over the entire available frequency range, a minimal dependency to frequency was observed for G’ and G”, thus proving the formation of stable three-dimensional networks (Abid, 2017; Rasidek et al., 2018). IPA precipitated pectin formed stronger gel than the acetone and 50/50 IPA-ACTN pectins as indicated by its higher G’. HM pectin gel is stabilized by intermolecular hydrogen bounds and hydrophobic interactions between the methyl ester of the carboxyl groups (Chan et al., 2017). Since the Degree of Methylation of all precipitated
Figure 3. Stress weep graph of *B. aethiopum* pectin extracted at pH natural and room extracted at pH natural and room temperature and precipitated with isopropanol (●), acetone (■), and 50/50 isopropanol/acetone mixed solvent (○). Measurements were done at 25°C and 1 Hz. Filled symbols represent the elastic modulus (G') and the open symbols represent the viscous modulus (G''). Pa: Pascal.

Figure 4. Dynamic mechanical spectra of 20 g kg⁻¹ of *B. aethiopum* pectin solution extracted at pH natural and room temperature and precipitated with isopropanol (●), acetone (■), and 50/50 isopropanol/acetone mixed solvent (○) done at 25°C using a constant stress of 0.35 Pa selected in the LVR. Filled symbols represent the elastic modulus (G') and the open symbols represent the viscous modulus (G''). Pa: Pascal; Hz: hertz.
pectins showed a difference that was not statistically significant (Table 1) it could be assumed that hydrogen bonds were solely responsible for gel formation and stabilization. Hydrogen bounds are weak and easily broken but a large number of them can confer great stability to a network (Chan et al., 2017). Sensorial properties of gels are usually described by the ratio of the elastic modulus to the viscous modulus (G' G") (Iglesias and Lozano, 2004). Table 2 shows the ratio (between 4 and 6) obtained for B. aethiopum pectins precipitated with the different solvents. These small values demonstrated that gels with soft texture were produced with B. aethiopum pectins samples (Iglesias and Lozano, 2004).

**Conclusions**

Palmyra palm (*Borassus aethiopum* Mart.) fruits offer an inexpensive raw material to extract pectin. In this study polar solvents (isopropanol and acetone) and their mixture (50/50) were used to precipitate pectin extracted at room temperature and natural pH (5.2-5.5) of the fruit. Results showed that precipitating solvents affected the physicochemical and functional properties of *B. aethiopum* pectins. Additionally, regardless of precipitating solvent, Palmyra palm pectin showed high viscosifying, emulsifying and gelling properties. Therefore, the choice of a specific extraction condition (Assoi et al., 2016) and precipitating method for the isolation of pectin from *B. aethiopum* fruit pulp should depend on the intended use of that pectin.

**CONFLICT OF INTERESTS**

The authors have not declared any conflict of interests.

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