Research Article

Physico-Chemical Properties of Guna Seed Protein (GPR)

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ABSTRACT

Guna protein was charaterised for its physicochemical properties .The properties investigated included moisture content, melting point, viscosity, total dissolved solids(TDS).Guna seeds were powdered using mortar and pestle and subsequently extracted with phenol and extraction buffer and centrifuged for 30minutes,this was then left to stand overnight .The purified protein extract was separated at lower layer of the separation. Moisture content was determined by drying and weighing constantly in a desiccator to a constant weight, total dissolved was determined by silica gel method, melting was determined by means of a melting point apparatus and viscosity was evaluated using micro-syringe method. Comparative extraction procedure was also used with acetone at various concentrations. Results from moisture studies showed a value of 1.2%, however a melting point value after triplicate determination of 220^{o}C using Stunt SMB model. Determination of TDS showed a value of 2.09%.Protein was extracted using the solvents:acetone,ethanol,ammonium tetraoxosulphate(vi) salt and phenol. The isolation ability was in the order of acetone>phenol>ammonium tetraoxosulphate(iv)salt>DETERGENT>

Keywords: Physicochemical, melting point, TDS, viscosity, guna protein, moisture.

INTRODUCTION

Guna is the Hausa name for the drought tolerant crop that belongs to the family of plants cucumbitaceace.Verieties of the plant include citrillus lanatus[1] and citrillus colocynthis[2].These are cultivated mainly in arid and semi-arid zones of north eastern Nigeria.Guna (citrillus vulgaris) which is an edible specie of the cucumbitaceace family is now found growing in southern and even north eastern Nigeria[3].

Recently, polymers from renewable resources have been receiving increasing attention predominantly due to two reasons partly to environmental concerns and owing to the fact that our petroleum resources are finite. In addition to providing income to those involved in agriculture, moreso, plant proteins have been used extensive in plastic applications[4].Protein from plants consist of amino-acids which can be able to form linkages and consequently resulting in a spectrum of possibility in terms of functionalities and functional properties.

Legume seed proteins have been studied extensively in the last few decades due to their economic viability and nutritional importance[5].More so knowledge derived from plant protein is widely used in applied sciences, medical sciences and biochemistry[6]. These contributions in plant protein analysis were in studies in functional properties such as emulsion capacity, water and oil absorption capacities, and foaming ability and viscosity measurements. Many applications requiring the utilization of protein and its derivatives requires the function in the final product. These denote the performance of proteins either as a processing aid or as contributor in the attributes of the final product[19].

More so, the manufacture of true biocomposites demands that the matrix be made predominantly from renewable resources. Green composites with moderate

mechanical properties are excellent for such applications in which at the end of their life they can be composted or disposed without harming the environment. With several plant based resin, such plant based resin abundantly present all over the world, protein resin obtained from them can be used after characterization for fabricating green composites.

Recently, soy protein composites[7] in the forms of isolates containing up to 90%protein and concentrates of up to 70% protein have been used as resins for composites(8).As a result, biodegradable bio-based biopolymer products have attracted much attention in recent years(8-10)

This served as stimulus to characterize Guna protein resin from guna seed (citrillus vulgaris) which is sustainable based on the physicochemical studies .

MATERIALS AND METHODS

MATERIALS

Guna seeds were obtained from a farmland in Mubi Local government area of Adamawa state being one of the major cultivators of the plant. Chemicals of analytical grade were supplied by the British Drug House(BDH) and these included silica gel , ethanol, acetone, ammonium acetate, phenol, EDTA, sucrose, hydrogen tetraoxosulphate (iv) acid etc.

Equipment used includes melting point apparatus model SMS smart, centrifuge and other laboratory equipment.

METHODS

These were cleaned stored in clean polyethylene bags. This was then followed by processing the seed using mortar and pestle and extraction using the method described by Tanaka [11] using phenol and extraction buffer (0.1 Hcl P^H8.8, 5M EDTA, 30% SUCROSE. This was left over night for further re-extraction. The substance was centrifuged for 30minutes to achieved complete separation.5% of 0.1% ammonium acetate was aaddedin100% methanol to further precipitate proteins before further analysis.

The proteins obtained were characterized for their viscosity, total dissolved solids, moisture, meltingpoint etc. as follows;

VISCOSITY MEASUREMENTS

The methods described by[12] were adopted for the viscosity measurements.

The intrinsic viscosity average molecular weight respectively in water ethanol system (Mv) of the polymer was determined using the modified Staudinger equation for branched and cross linked polymers in equation below.

$= KM_v^a = 0.74 \times 10^{-3} M_v^{1.05}$

Where K and a are constant given as 0.7×10^{-3} and 1.

SOFTENING TEMPERATURE

The softening temperature of GPR was determined by melting point apparatus at a heating rate of 5^oC per minute over a temperature range of 40^oC – 150^oC in accordance with the methods[12].

TOTAL DISSOLVED SOLID CONTENT (TDS)

This was carried out according to the methods described by (14). Silica gel weighing about 25g was added to an evaporating dish, dried in an oven at 110^oC for 30 minutes, cooled in a desiccators and then weighed. About $7cm^3$ of Guna Protein Resin (GPR) Syrup (W_2.g) was added into a dish and reweighed. The dish was covered with aluminum foil and placed in an oven and heated to a constant weight W_1g. The temperature will be recorded.

The percentage weight by weight of the resin solids was calculated using the relation in equation

$$\% \text{ TDS} = \frac{W_2 g}{W_1 g} \times 100$$

MOISTURE CONTENT

Thermal properties of the GPR were determined by the methods of (13). This included a known weight of resin samples

introduced into a desiccator containing a saturated solution of sodium chloride. The differences between the wet and dry samples were recorded as moisture intake by the resin samples. The amount of water absorbed was calculated as follows;

$$M_{t(\%)} = (W_t - W_o) / W_o \times 100\%$$

Where W_t and W_o are the weights of the samples before and after immersion in water respectively.

RESULTS AND DISCUSSIONS

RESULTS

Table 1 **PHYSICOCHEMICAL PROPERTIES**

Properties	Melting point	TDS	Moisture Content
	220^{O}C	2.09%	1.2%

Table 11: Extraction Abilities of various solvents

Solvents	Acetone	Ammonium sulphate	Detergent	Ethanol	Phenol	
Ability	+++	+		+	++	++

Key:

+++ highest + relative low ++ relatively low

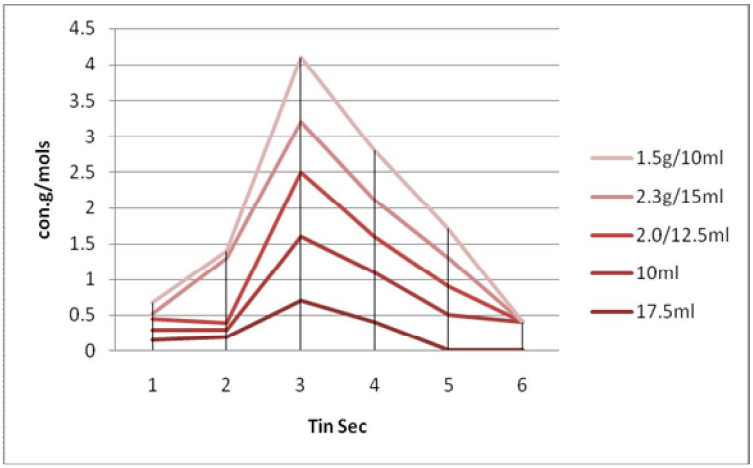

Fig. 1: change in the viscosities of protein resin at various concentrations with time

DISCUSSION

Result of moisture studies showed that guna protein has a moisture content of 1.2% which falls within acceptable codex for plant materials of this category, this is shown in Table. However the melting point of protein so obtained after crystallization was found to be within the range of 220-280^{o}C .This high temperature recorded may be attributed to the fact that proteins exist as ions i.e. positively charged, negatively charged or as zwitterions (14).The stability of protein is key and useful in the determination or prediction of applications and uses of the end products of the materials synthesized in combination with protein. The melting temperature of the crystalline protein appeared high suggesting a broad range of applications with respect to temperature sensitivity. The solvents scrutinized for use in protein extraction included; acetone, phenol, detergent, ethanol and ammonium tetraoxosulphate (vi) salts .Proteins can be solubilized from plant membrane by organic solvents, salts or detergents as partitioning by proteins is a function of factors such as ionic composition, concentration targeted, molecular weight, and stability of proteins15-17. In the present investigation, the greatest concentration of proteins was seen in the case of the acetone extract. This may not be unconnected to the fact that acetone is a non-polar solvent. It has been established that acetone is more efficient in the extraction of protenous substances compared to ethanol [18].

The relative extraction abilities of solvents namely: acetone, phenol, ammonium tetraoxosulphate (iv) salts, ethanol are shown in Table11. From the result, acetone has the highest was seen in the case of acetone. This may not be unconnected to the fact that for the extraction of protein a number of factors are taken into consideration among which nature of solvent, volume of solvent used, molecular weight of the protein under study and a host of other factors[17].Hence ,no single buffer is appropriate for use as a universal for the extraction of all targeted proteins[20].Therefore, the solvent that showed the most or maximum extraction for proteins was selected and was used in investigations and further analysis. The total dissolved solutes was 2.09%, this is shown in Table. This is a percentage representing various silica gel determinations of solute content over a period of time. Proteins in legumes may be characterized by accessing their physical and chemical properties. These properties are reflected in physicochemical properties which in turn are a function of inherent primary composition and sequence of amino-acids. In the present investigation, the percentage total dissolved indicated a good texture of the protein concentrate .This is in line with studies of[22]. The relative viscosities of the various concentrations of protein obtained at 35^{o}C are shown in Fig1. The viscosities of the various concentration under the present study tend to reduce as the concentration is increased, likewise the time of flow decreases with increase in concentration of the guna protein .These are all attributable to the increase in molecular weight as the concentration is increased. These points were earlier buttressed in studies carried out by[21].

CONCLUSION

Acetone showed the best protein pre - concentration at2.5g in 17.5ml and therefore selected as the most suitable solvent in this study. The viscosity of protein was considered most desirable at concentration(2.5g in 17.5ml).The total dissolved solid indicated a good texture and therefore useful in application were enhanced performance in considered. The high melting temperature 782recorded suggest its utility were high medium to high temperature in food packaging. In addition the low moisture content observed reiterates its application in packaging were improved shelf life is considered.

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