Journal of Applied Biology & Biotechnology Vol. 8(01), pp. 59-63, Jan-Feb, 2020
Available online at http://www.jabonline.in
DOI: 10.7324/JABB.2020.80110

Partial purification and characterization of lectin from serum of American cockroach, *Periplaneta americana*

Reymend Encily Martin*, Yogita Raosaheb Channe
Department of Zoology, Dr. Babasaheb Ambedkar Marathwada University, Aurangabad, Maharashtra, India

ARTICLE INFO

Article history:
Received on: July 25, 2019
Accepted on: October 01, 2019
Available online: January 10, 2020

Key words:
Purification, lectin, sugar specificity, pH, temperature, *Periplaneta americana*

ABSTRACT

The partial purification and characterization of haemagglutinin (lectin) were carried out from the hemolymph of the adult American cockroach, *Periplaneta americana*. The hemolymph was drawn from cockroach and lectin was purified by a single-step method, using ammonium sulfate $(NH_4)_2SO_4$ salt fractionation and gel filtration. Gel filtration showed two peaks. The Hemagglutination Activity (HA) was observed in the 20th fraction of the second peak. The purified lectin showed a molecular weight of 26.8kDa on Sodium Dodecyl Sulfate-Poly Acrylamide Gel Electrophoresis. The purified lectin showed an increase in HA at pH 7.5 and, subsequently, a sharp decline at pH 8. This indicates that HA was specific to a certain pH level. Similarly, an increase in HA was observed until 30°C, followed by a decline at 40°C. This indicates the heat labile nature of lectin. The HA showed a higher specificity to divalent Ca^{2+} and showed no specificity for Ba^{2+}. It also showed a higher inhibition for sugar D-galactose and a least inhibition for D-lactose. The HA to vertebrate blood group showed a highest activity to goat Red Blood Cells (RBCs). The study concludes that carbohydrate-binding-specific lectin is important for recognition of the cell surface carbohydrate of invading pathogens.

1. INTRODUCTION

Lectins have been purified from various insect species, such as fleshflies and cockroaches [1,2]. Some of them actively participate in self-defense by recognizing the polysaccharide chains on the surface of pathogens. Some insect lectins bind to microbial components such as lipopolysaccharide and activated prophenoloxidase (ProPO), and enhanced the cellular immune response by modulating hemocytes. The insect immune system activation mechanism was studied in *Drosophila melanogaster*, as a model organism [3,4]. *Drosophila* is estimated to have more than 30 C-type lectin genes [5,6]. However, very little is known about the participation of lectin in Drosophila immunity [7]. Undoubtedly, the presence of lectin in the hemolymph of insects appears to have an important function in insect immune defense system [8,9].

In invertebrates, including insects where they lack an antibody-based immunity, lectin probably plays a major role in nonself recognition, with several reports of endogenous serum lectin having opsonic activity, for invading pathogens [10,11]. Other than opsonization, lectin trigger ProPO activation pathway [12]. Arumugan, et al.[13] purified, characterized and identified humoral immune function on the induction of PO activity with the hemolymph lectin of *Periplaneta americana*. In this study, we examine in detail the hemolymph lectin from *P. americana* hemolymph, for its role in immune recognition and its properties.

2. MATERIALS AND METHODS

2.1. Insect Rearing

The cockroach, *P. americana* was collected from Godown's and reared in the laboratory in a dark container. They were reared in room temperature 27 ± 1°C and 12:12 Total Light (LL): Total Dark (DD) cycle. All experiments were conducted at this room temperature unless specified.

2.2. Preparation of Hemolymph Sera

The insect hemolymph was collected from 40 cockroaches of the same sizes. Before collecting hemolymph, the individual cockroaches were first anesthetized by keeping them at low temperature for 20 minutes. Then, they were bled by severing

*Corresponding Author
Reymend Encily Martin, Department of Zoology, Dr. Babasaheb Ambedkar Marathwada University, Aurangabad, Maharashtra, India.
E-mail: martinbamu@gmail.com

© 2020 Martin and Channe. This is an open access article distributed under the terms of the Creative Commons Attribution License -NonCommercial-ShareAlike Unported License (http://creativecommons.org/licenses/by-nc-sa/3.0/).

hind legs with fine scissors, and hemolymph was collected in a sterile centrifuge tube containing phenyl thiourea crystals. Phenyl thiourea crystals were used to inhibit phenoloxidase activity. After 10 minutes, the tubes were centrifuged at 10,000 rpm at 4°C for 10 minutes to remove hemocytes. The supernatant was carefully removed and used as a serum for HA assay against goat's Red Blood Cells (RBCs). The collected samples were tested immediately to prevent counterfeit negative results.

2.3. Isolation of Lectin: (Salting out of Serum Protein)

The lectin was isolated according to the procedure as described by Dorrah et al. [9].

2.4. Preparation of Erythrocyte Suspension and Assay of Hemagglutination Activity (HA)

Blood samples from goat and chicken were collected aseptically, each in an equal volume in heparin. Human blood (types A, B, and O) was collected in citrate/dextrose. The erythrocyte suspension and HA was determined by the procedure described in [14].

2.5. Effect of Divalent Cations

To study the effect of divalent cation for HA, the agglutination tests were performed as described by Richards et al. [15]. An aliquot of 100 µl of diluted serum was dialyzed for 24 hours at 4°C against 200 ml Tris Buffer Saline (TBS), pH 7, either without or with 20 mM Ca^{2+}, 20 mM Ba^{2+}, 20 mM Mg^{2+}, 20 mM Zn^{2+}, 20 mM Hg^{2+}, or 10 mM Ethylene Di-amine Tetra Acetic acid (EDTA) with 2 changes of buffer. HA assays were carried out by using goat RBCs washed in dialysis buffer.

2.6. Inhibition Assay

The binding of sugars to serum lectin was studied by competitively binding and measuring HA in the presence of goats RBCs. The method of inhibition followed in this study was as described by Ayyad et al. [14]. The sugars assessed in this study are monosaccharides (D-galactose, D-glucose, and D-fructose) and oligosaccharides (D-lactose). The minimum concentration of sugars for causing 50% inhibition (IC50) of HA was studied.

2.7. Effect of Temperature and pH

For the heat stability test, an aliquot of the protein sample was incubated in a heating block at different temperatures (10°C, 20°C, 30°C, and 40°C) for 30 minutes. The solution was cooled on ice immediately and then allowed to warm up to 27°C at room temperature, and thereafter the HA was assayed. The percentage of HA was calculated by dividing the activity of the sample with the activity of the control kept at temperature 27°C Room Temperature (RT) multiplied by 100.

For pH test, the lectin in buffer solutions with different pH values, i.e., 6–6.5 pH (0.2 M Citrate buffer) and 7–8 pH (0.2 M Tris-HCl buffer), was used. Equal volumes of protein sample and buffer solution were mixed and incubated at room temperature for 30 minutes. The solution was neutralized and the HA assay was performed. The hemagglutination titer of the protein sample was incubated in Phosphate Buffer Saline (PBS) (pH 7), which served as control. The percentage of HA was calculated by dividing the activity of the sample with the activity of the control of pH 7.5 multiplied by 100.

2.8. Determination of Protein Concentration

The total protein concentration was determined according to the method of Lowry et al. [16], using bovine serum albumin (BSA) dissolved in 0.15 M NaCl was used as a standard.

2.9. SDS-Poly Acrylamide Gel Electrophoresis

The SDS-PAGE of the isolated lectin was carried out according to the procedure described by Laemmil [17]. The electrophoresis was performed at a constant voltage of 200 V for 90 minutes. The molecular weight of the protein bands was determined using standard medium-range molecular markers: 97.4KDa-Phosphorylase-b; 66KDa-BSA; 43 kDa ovalbumin, 29kDa carbonic anhydrase, 20.1kDa soyabean trypsin inhibitor, 14.2kDa lysozyme.

3. RESULTS AND DISCUSSION

3.1. The Physico-Chemical Properties of the Isolated Lectins

In this study, a single-step purification procedure was used to purify lectin. The chromatographic step using column Sephadex G-75 was used to remove most of the impurities and the purification fold of 2.38 was obtained (Table 1). The gel filtration showed two peaks and the second peaks fraction 20 showed HA (Fig. 1). The obtained data in the present study revealed that HA showed a decrease at 40°C (Table 2). On the other hand, a high HA was observed at 30°C. At 10°C the HA showed a least activity. This observation indicates that lectin from cockroach are heat-labile in nature, and shows the highest activity at certain temperatures only. The

Table 1: Purification of lectin from serum of American cockroach, *P americana*.

Step	Volume (ml)	Total Protein (mg/ml)	Hemagglutinating Activity	Yield (%)	Purification (fold)
HLS	5.3	1023	0.25	100	1
Aspt	4.8	672	0.2	65.68	1.21
Dialysis	4.0	486	0.15	47.50	1.26
Gel Filtration (F-20)	3.0	366	0.0375	35.77	2.38

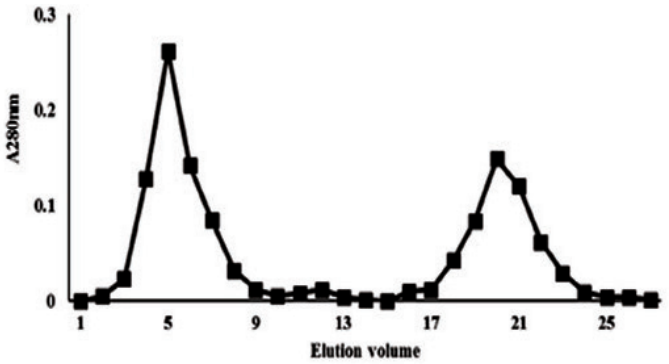

Figure 1: The profile of gel filtration on Sephadex G 75 showing two peaks. Only the second peaks fraction 20 showed hemagglutinating Activity.

heat instability is a characteristic of lectins of some other insects, e.g., the orthopteran, *Teleogryllus commodus* [18], *Melanoplus sanguinvpes,* [19] and the dipteran, *Glossina fuscipes* [20], in phasmid *Exatosoma tiaratatum* [15]. However, in contrast to this, in *Leptinotarsa decemlineata* [21], and in *Locusta migratoria* [22], the lectins were reported to be heat resistant. Similarly, we observed HA at different pH ranges of 6–8 pH with the highest activity at pH 7.5 (Table 3). Similar results were observed in *Pterockdiella capillacea* (pH 7.5) [23], and *P. americana* (pH 7.5) [13]. Ayaad [24] reported an HA at pH 6.2 for *Aedes caspius*.

3.2. Divalent Cation Requirement

The study revealed that isolated lectins are calcium-ion-dependent. The cations like Mg^{2+} or Zn^{2+} appear to have limited ability to replace Ca^{2+}; however, Ba^{2+} showed no effect on HA (Table 4). When EDTA was added, complete inhibition of HA was observed. Similar observations were reported on insects having C-type lectin, *T. commodus, M. sanguinipes, E. tiaratum*, and adult *Schistcerca gregari* [15,18,19,25]. The mechanism of carbohydrate binding of C-type lectin is suggested by Zelensky and Gready [26]. According to Zelensky and Gready [26], each of the C-type lectins possesses a carbohydrate recognition domain structure with conserved Ca^{2+} binding sites, and a second site [27]. The lectin of *S. gregaria* [25] is Ca^{2+} dependent. However, Yu and Ma [28] suggested that calcium is not required for lectin binding to the sugar, but is necessary to protect it from proteinase digestion. Several workers have reported that the C-type lectin possesses a common carbohydrate recognition domain [29,30] and a second domain [31] that may help in increasing their binding affinity to carbohydrate. The suggested mechanism may involve interactions with the conserved Ca^{2+} and carbohydrate, which may possibly result in the formation of hydrogen bonding with acid and amide side groups [32]. A similar mechanism is carried out in this study.

We observed that the HA of *P. americana* was strong in the presence of calcium, followed by zinc, magnesium, and mercury. The results showed that cockroach HA may be condensed by heavy metal ions, possibly due to protein denaturation. The inhibition of HA of *P. americana* by Ba^{2+} was important because it did not show any HA. The plausible reason for this may be that barium ions may be directly bound to the carbohydrate-binding sites, which may seriously affect its conformation modifications resulting in blocked active sites.

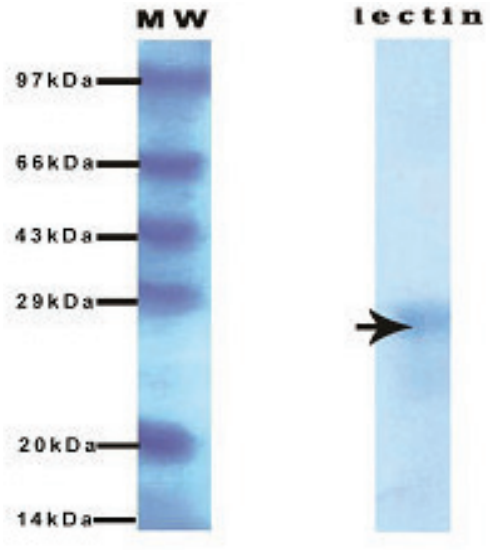

Figure 2: Molecular weight of lectin calculated by SDS–PAGE. Lane 1 shows Standard molecular weight markers, and lane 2 shows the purified lectin molecular weight.

Table 2: The HA of cockroach serum to different temperature.

Temperature (°C)	HA	%HA
10	0.025 ± 0.00	68.3
20	0.143 ± 0.0037	39.07
30	0.366 ± 0.0023	100
40	0.122 ± 0.0038	33.33

Values are $n = 3$, $\bar{x} \pm$ SD

Table 3: The HA of cockroach serum to different pH.

pH	HA
6.0	0.027 ± 0.0046
6.5	0.030 ± 0.0046
7.0	0.211 ± 0.025
7.5	1.001 ± 0.023
8.0	0.1 ± 0.00

Values are $n = 3$, $\bar{x} \pm$ SD

Table 4: The HA of cockroach serum to different divalent cations.

Metal ion (20 mM)	HA	%HA
Ba^{2+}	ND	
Ca^{2+}	0.211 ± 0.0025	100
Zn^{2+}	0.1 ± 0.00	47.39
Mg^{2+}	0.122 ± 0.003	57.81
Hg^{2+}	0.03 ± 0.0046	14.21
EDTA	0	0

Values are $n = 3$, $\bar{x} \pm$ SD, ND = Not detected.

Table 5: The HA of cockroach sera to different blood groups.

	HA
Goat RBCs	0.366 ± 0.023
Chicken TBCs	0.233 ± 0.0023
Human erythrocytes	
A	0.1 ± 00
B	0.083 ± 0.00
O	0.025 ± 0.00

Values are $n = 3$, $\bar{x} \pm$ SD.

Measured in a sample pool of 30 insects for each RBCs types. Standard assay conditions using TBS/Ca^{2+}-Mg^{2+}.

high specificity of serum lectin to goat RBCs when compared to that of chicken and human.

In this study, the erythrocytes from human, goat, and chicken showed a non-specific HA with cockroach lectin. The lectin agglutinated all these erythrocytes with varying degrees of HA titers. The serum agglutinating activity is rather not highly specific since all the erythrocytes tested showed agglutination and it might be common as naturally occurring agglutinins are specific toward a wide range of vertebrate erythrocytes. The presence of lectin sugar on the surface of erythrocytes may result in an interaction with the lectin resulting in agglutination. The lectin showed a significantly higher HA for goat compared to chicken erythrocytes. The general specificity is influenced by the limited number of contacts with carbohydrates and the depth of the sugar-binding sites [30]. In addition, any modification or substitutions to a binding site can influence the binding specificity [33]. The observed difference in lectin activity with different blood groups may be due to the difference in carbohydrate lectin binding interactions, which can be attributed to differences in carbohydrates present in the cell surface of the different blood groups. The carbohydrates on the erythrocyte cell surface are differently distributed in various animal groups [34]. In the presence of lectin, sugar on the surface of erythrocytes results in an interaction with the lectin, leading to agglutination, or the carbohydrate unit structure and positions may be more specific with higher affinity for binding the lectin. It is possible that a similar mechanism may be responsible for increased goat erythrocyte agglutination. Moreover, lectin characteristics, such as multi-valency, may determine cross-linking interactions in binding recognition spatial distribution of multivalency among lectin structures, which, in turn, produce a higher level of specificity [35,36]. A high HA observed for goat erythrocytes might be due to the occurrence of specific carbohydrate moiety on the surface of goat erythrocytes other than RBCs types [13]. Unfortunately, several erythrocyte types were not examined for the HA activity in the previous reports [37,38].

3.4. Sugar specificity and inhibitions to HA

The inhibition of HA by sugar is shown in Table 6. The carbohydrate specificity of agglutinin is detected in the serum of cockroach and is called as the HA inhibition assay [39,40]. The HA of these lectins was preferentially inhibited by D-galactose, followed by D-lactose, D-glucose, and D-fructose. These results suggest that the carbohydrate-binding and carbohydrate recognition domains prefer α linked D-galactosides rather than free D-galactose. The affinity of the cockroach sera lectins toward α-linked galactosides is reported in *L. migratoria* [22] and in adult of *S. gregaria* [25]. The β-D-galactosides, as lectin-specific ligands, are found in insects, coleopteran *Allomyrins dichotoma* [41]. Barondes [42] also reported β-galactosides, as the lectin specificity in vertebrates,

Table 6: The HA of cockroach serum to different Sugars.

Sugar specificity	Concentration for 50% inhibition of lectin activity	% inhibition
D-Galctose	0.122 ± 0.0038	100
D-Glucose	0.061 ± 0.0034	50
D-Fructose	0.052 ±. 0041	42.62
D-Lactose	0.066 ± 0.0028	54.09

Inhibition was assayed at concentration of 25 mM sugars.

and in orthopterans like *T. commodus* [18] and the dipteran *P. duboscaqi* [40] also possess lectins with amino sugars binding affinity. In many species of insects, especially lepidopteran, hemolymph lectins show an affinity for galactose and lactose [43] or to glucosides [21,44]. Some insects also show a binding affinity to mannose, e.g., the dictyopteran, *Blaberus discoidalis* [45] and lepidopteran, *Heliothis virescans* pupae [46]. Sugars such as galactose and lactose have been reported as inhibitors of the agglutinating activity of hemolymph of *P Americana* [37,38]. Volf et al. [40] and McGreal et al. [47] reported that sugars binding specificity is significant for the recognition of carbohydrate moiety on pathogen surface, which are essential and responsible for the ability of the insect as a vector in pathogen transmission. A similar property has been reported in lectins of other insect, *B. discoidalis* [45], *Phlebotomus duboscqu* [40], and *A. caspius* [24].

3.5. SDS-PAGE

SDS-PAGE showed a single band of molecular weight of 26.8 KDa. This molecular weight was near to 30 KDa lectin of *P. americana* [38], and 20KDa lectin of *Saracophaga peregrina* [48].

4. CONCLUSION

In conclusion, we say that the lectin purified from cockroach has the highest HA at temperature 30°C and at pH 7.5. The cockroach serum lectin contains a C-type lectin and shows inhibition to D-galactose sugar. Furthermore, the insect's C-type lectin appears to play an important role in pathogen recognition and cellular interactions. This ability is achieved due to binding with the carbohydrate component of the surface molecular patterns. The SDS-PAGE of lectin showed a molecular weight of 26.8kDa. The isolated lectin was highly specific to goat RBCs than for those of other vertebrates. Accordingly, we can say that the carbohydrate-binding specificity by lectin is necessary for the recognition of the cell surface carbohydrate including those of the invading pathogens.

REFERENCES

1. Notori, S. Insect lectins and innate immunity. Adv Exp Med Biol 2001;484:223–8.
2. Franc NC, White K. Innate recognition systems in insect immunity and development. New approaches in Drosphila. Microbes infect 2000;2:243–50.
3. Hoffman JA. The immune response of *Drosophila.* Nature 2003;426:33–8.
4. Naitza S, Ligoxygakis P. Antimicrobial defenses in *Drosophila* the story so far. Mol Immunol 2004;40:887–96.
5. Dodd RB, Drickamer K. Lectin like proteins in model organisms: implecation a for evolution of carbohyrates—binding activity. Glycobiology 2001;11:71–9
6. Tanji T, Kobayashi A, Natori S. Activation of the *Sarcophaga* lectin gene promoter in transgenic *Drosophila*. Arch Insect Biochem Physiol 2002;50:131–8.
7. Pace KE, Lebestky T, Hummel T, Amoux P, Kwan K, Baum LG. Characterization of novel *Drosophila melanogaster* galectin: expression in developing immune, neural and muscle tissues. J Biol Chem 2002;277:13091–8.
8. Vasta GR, Ahmed H, Taumi S, Odom, EW, Saito K. Biological roles of lectins in innate immunity: molecular and structural basis of diversity in self /non self recognition. In: Lambris JD (ed.). Current topics in innate immunity, Springer, New York, NY, pp 389–406, 2007.

9. Dorrah AM, Ayaad TH, Mohamad A, Bassal TTM. Isolation and characterization of multiple lectins from serum of the desert locust, *Schistocerca gregaria* (Orthopera: Acridiade). J Orthroptera Res 2009;18(1):103–12.
10. FujitaT, Matsushita M, EndoY. The lectin complement pathways its role in innate immunity and evolution. Immunol Rev 2004;198:185–202.
11. Denis M, ThayappanK, Ramaswamy MS, Arumugan M. Lectin in innate immunity of crustacea. Austin Biol 2016;1:1001–7.
12. Shi XZ, Yu XQ. The extended loop of the C terminal carbohydrate recognition domains of *Manduca sexta* immulectin-2 is important for ligand binding and functions. Amino Acids 2012;42:2383–91.
13. Arumugan MG, Sreramulu B, Paulchamy R, Thangevel S, SundaramJ. Purifcation and functional characterization of lectin with phenol oxidase activity from hemolymph of cockroach *P ameriana*. Arch Insect Biochem Physiol 2017;95:21–39.
14. Ayaad TH, Dorrah MA, Mohamed AA, Bassal TTM. Specificity and developmental changes in hemagglutinating activity of serum of the desert locust Schistocerca gregaria (Orthoptera; Acrididae). J Orthoptera Res 2009;18(1):51–6.
15. Richards EH, Ratcliffe NA, Renwrantz I. Isolation and characterization of a serum lectin from the giant stick insect, *Exatosoma tiaratatum*. Insect Biochem 1988;18:691–700.
16. Lowry OH, Rosebrough NL, Forr Al, Randell RJ. Protein measurement with Folin Phenol reagent. J Biol Chem 1951;192:265–75.
17. Laemmli UK. Cleavage of structural protein during the assembly of head of bacteriophage T4. Nature 1970;227:680–5.
18. Hapner KD, Jermyn MA. Hemagglutinin activity in the hemolymph of *Teleogryllus comodus.* Insect Biochem 1981;11:287–95.
19. Stebbins MR, Hapner KD. Preparation and properties of hemagglutinin from hemolymph of Acrididae (Grasshopper). Insect Bichem 1985;15:451–62.
20. Ingram GA, Molyneux DH. Lectins in the hemolymph of *Glossina fuscipes fuscipes*: isolation, partial characterization, selected physico-chemical properties and carbohydrate binding specificities. Insect Biochem 1990;20:13–27.
21. Minnick ME, Rupp RA, Spence KD. A bacteria induced lectin which triggered hemocyte coagulation in *Manduca sexta*. Biochem Biophysical Res Commun 1986;137:729–36.
22. Drif L, Brehelin M. Purification and characterization of an agglutinin from hemolymph of *Locusta migratoria* (orthropotera). Insect Bioche Mol Biol 1994;24:223–8.
23. Oliveira JTA, Melo VMM, CamaraMFL, Vasconcelos IM, Beltramini LM, Machado OLT, et al. Purification and characterization of lectin from *B. mori*. Phytochem 2002;61:301002E.
24. Ayaad TH. Isolation and partial characterization of Ca^{2+} independent lectin from *Aedes caspius* (Diptera: Culicidae). Efflatounia 2008;8: 17–28.
25. Ayaad TH. Isolation, characterization, and N–terminal amino acid sequence of lectin from plasma of *Schisticerca gregaria*. Efflatounia 2004;4:9–22.
26. Zelensky AN, Gready JE. The C-type lectin-like domain superfamily. FEBSJ 2005;272:6179–217.
27. Drickamer K. C-type lectin-like domain. Curr Opin Structural Biol 1999;9:585–90.
28. YuXO, Maa Y. Calcium is not required for immulectin-2, but protects the protein from proteinase digestion. Insect Biochem and Mol Biol 2006;36:505–16.
29. Weis WI, Kahn R, Fourme R, DrickamerK, Henddrickson WA. Structure of the calcium dependent lectin domain from rat manose binding protein determined by MAD phasing. Science 1991;254:1608–15.
30. Drickamer, K. Multiplicity of lectin—carbohydrate interactions. Nat Struct Biol 1995;2:437–9.
31. Yu XQ, Kanost MR. Immulection-2, a lipopolysaccharide specific lectin from insect, *Manduca sexta*, is induced in response to gram negative bacteria. J Biol Chem 2000; 275:37373–81.
32. WeisWI, Drickamer K.Structural basis of lectin carbohydrate recognition. Ann Rev. Biochem 1996;65: 441–473.
33. DrickamerK, Taylor E. Glycan arrays for functional glycomics. Genome Biol 2002;3:10341–4.
34. Ynalvez RA, Carmen GC, Ynalvez MA. Isolation, partial purification and characterization of texas live oak lectin. Adv Biosci and Biotechnol 2015;6:470–84.
35. Loris R. Principles of structures of animal and plant lectins. Biochim Biophys Acta 2002;1572:198–208.
36. Sacchettini JC, Baum LG, Brewer CF. Multivalent protein carbohydrate interactions. A new paradigm for supermolecular assembly and signal transduction. Biochem 2001;40:3009–15.
37. Jomori T, Kubo T, Natori S. Purification and characterization of lipopolysaccharide binding protein from hemolymph of american cockroach *Periplaneta americana*. Eur J Biochem 1990;190:201–6.
38. Kubo T, Natori S. Purification and some properties of a lectin from the hemolymph of *Periplaneta americana* (American cockroach). Eur J Biochem, 1987;16:875.
39. Sivaprasanth P, Nalini M. Lactose specific hemagglutinins with *in vitro* opsonic effects from the hemolymph of *Ariadne merione* (Cramer). Int J Sci Res 2014;3:2013–48.
40. Volf P, Skarupova, Man P. Characterization of the lectin from female *Phlebotomus duboscqui* sand flies. Eur J Biochem 2002;269:6294–301.
41. Umetsu K, KosakaS, SuzukiT. Isolation and characterization of a lectin from the beetle *Allomyrins dichotoma*. J Biochem 1984;95:239–46.
42. Barondes SH. Soluble lectins a new class of extracellular proteins. Science 1984;223:1259–64.
43. Pendland JC, BouciasDG. Characteristics of a galactose binding hemagglutinin (lectin) from hemolymph of *Spodoptera exigua* larvae. Dev Comp Immunol 1986;10:477–87.
44. Qu XM, Zhang CE, Komano II, Natori S. Purification of a lectin from the hemolymph of chinese oak silkmoth (*Antheraea pernyl*) pupae. J Biochem 1987;101:545–51.
45. Chen C, Ratcliffe NA, Rowley AF. Detection, isolation and characterisation of multiple lectins from the hemolymph of the cockroach, *Blaberus discoidalis*. Biochem J 1993;294:181–90.
46. Ourth DD, Narra MB, Chung KT. Isolation of mannose binding C-type lectin from *Heliothis virescens* pupae. Biochem and Biophysical Res Comm 2005;335:1085–9.
47. McGreal EP, Martinez PI, Gordon S. Divergent roles for C-type lectins expressed by cells of the innate immune system. Mol Immunol 2004;41:1109–21.
48. Fujita Y, KurataS, Homm K, Natori S. A novel lectin from *Sarcophaga* its purification, characterization, and cDNA cloning. J Biol Chem 1998;273:9667–72.

How to cite this article:
Martin RE, Channe YR. Partial purification and characterization of lectin from serum of American cockroach, *Periplaneta americana*. J Appl Biol Biotech 2020;8(01):59–63. DOI: 10.7324/JABB.2020.80110