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Anti-Diabetic Assessment of Endophytes Isolated From Neem

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***Abstract* – Endophyte of medicinal plants have sharp potential in *de-novo* synthesis of different bioactive metabolites. These bioactive compounds are used directly or indirectly as therapeutic agent in several ailments. Defect in insulin secretion, insulin action or both in human system named diabetes. Diabetes mellitus is causing serious health problem and nearly half of the adult world population is affected. Mycelia extract of endophytes isolated from neem were reported best inhibition ranging 15 to 38% α-amylase and sucrose. Haemoglobin enhancement is also reported. Isolated endophytes have fair activity against diabetes.**

***Keywords* – Medicinal Plants, Neem, Endophytes, Diabetes, Glucose level.**

I. INTRODUCTION

Medicinal plants are a major source of traditional remedy and also securing many basic requirements of health and livelihood. A large population of India realizes remedy of medicinal plant and they are almost dependent on medicinal plants. The world health organization reported that about 80% population of Asia and South Africa sub-continent use medicinal plants and their products as primary health care (Tapsell *et al.*, 2006; Lai and Roy, 2004; Sherman and Hash, 20011).Neem is a member of Meliacae family and also known as *Azadirachta india* A.juss. This is a tropical evergreen tree native to Indian sub-continent (Anonymous, 1985; Roxburgh, 1874). This is medium to large tree with up to 25 meter height with spreading branches. It started fruiting after 3-5 years but on full production it comes after 10 years (Kumar and Gupta, 2002). Neem is used in preparation of Ayurveda medicines since 4000 years back. In Sanskrit neem is called *arista* means *perfect, complete and imperishable.* Every part of this plant has medicinal potency which is extensively used in Ayurveda, Unani, Siddha, Naturopathy, Homeopathy and other medicines. (Pai *et al.*, 2004; Nathan *et al.*, 2005; Dasgupta *et al.*, 2004).An endosymbiosis of bacterium or fungus with plant parts of its life cycle without causing any disease. These interactions are not only mutualistic but communalistic also. Endophytes were first illustrated in the Darnel (Freeman, 1904) and these lead to worldwide to search novel endophytes from different sources to better understand and applicability of endophytes and their produced compounds compounds (Arnold *et al.*, 2003, 2005, 2007; Stone *et al.*, 2004; Schulz and Boyle, 2005; Rodriguez *et al.*, 2009). A report of Berdy, 2005 details more than 20, 000 bioactive metabolites are originated from different microbes. Fungi are the important organism which produces many novel metabolites which are used as drug (Kock *et al.*, 2001; Bode *et al.*, 2002; Donadio *et al.*, 2002; Chin *et al.*, 2006; Gunatilaka, 2006; Mitchell *et al.*, 2008; Stadler and Keller, 2008).Diabetes Mellitus is a metabolic disorder caused due to defect in insulin secretion, insulin action or both. Diabetes mellitus is causing serious health problem and due cause of some inheritance or changed lifestyle and food habit (Kumar and Clark, 2002). World health organization is reported that nearly 200 million people in world are diagnosed with diabetes. This is the sixth leading cause of disease related death. Diabetes may lead to many vascular complications and cause nephropathy, retinopathy and neuropathy (Beverley and Eschwege, 2003). WHO is estimated that diabetes mellitus increase from 4.0 percent in the year 1995 to 5.4 percent by the year 2025 and number of people with diabetes mellitus in the world will increases from 135 million to 300 million in the year 2025.A study for assessment of antidiabetic potency present in neem endophytes extract was taken in this study.

II. MATERIALS AND METHODS

The healthy leaves were surface sterilized by modified method of Strobel *et al.*, 1996. Grown endophytes were isolated and maintained on PDA slants. Isolated fungi were identified in order to morphological characteristics viz. colony growth, presence or absence of aerial mycelium, colony colour, presence of wrinkles and furrows, pigment production etc. in reference to Barnett, 1992 by lactophenol and other stains under microscope with 40X resolution. Qualitative and Quantitative characterization of Amylase, Protease, Cellulose and Lipase were performed. Collected compound were access for anti-diabetic activities. α-amylase inhibitory activity, Assay of sucrase inhibition activity and Glucose diffusion method were performed for *in-vitro* antidiabetic properties assessment. *In-vivo* anti-diabetic activities were also performed with animal model in respect to serum glucose level and Blood function test according to standardized laboratory method.

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III. RESULTS

Tab. 1. Morphological characterization of isolated fungi.

Endophyte (Morphological basis)	Colour of mycelia	Pigmentation	Spore arrangement
Cladosporiumsp.	Olive brown	Dark brown	Un-branched
Nigrosporaoryzae sp.	White	Black	Un-branched
Streptomyces sp.	White	Whitish green	Branched filamentous
Acremonium sp.	White	Not appeared	Single celled
Fusarium sp.	White	Whitish pink	Sickle shaped
Curvularia sp.	Blackish brown	Greenish brown	Branched septa

Tab. 2. Qualitative enzyme activities characterization of isolated fungi

Name of Endophyte (Morphological basis)	Amylase	Protease	Cellulase	Lipase
Cladosporiumsp.	-	+	+	+
Nigrosporaoryzae sp.	-	-	-	+
Streptomyces sp.	+	+	+	-
Acremonium sp.	+	-	+	+
Fusarium sp.	+	-	-	+
Curvularia sp.	-	+	-	-

Tab 3. Quantitative enzyme activities characterization of isolated fungi

Endophyte (Morphological basis)	Amylase (U/ml)	Protease (U/ml)	Cellulase (U/ml)	Lipase (U/ml)
Cladosporiumsp.	0.00	1.22	0.25	0.16
Nigrosporaoryze sp.	0.00	0.00	0.00	0.25
Streptomyces sp.	2.31	1.63	0.45	0.00
***Acremonium* sp.**	1.90	0.00	0.47	0.12
***Fusarium* sp.**	2.71	0.00	0.00	0.05
***Curvularia* sp.**	0.00	1.17	0.00	0.00

Tab 4. In-vitro antidiabetic activities viz. α-amylase (%) Sucrase (%).

Name of Endophyte (Morphological basis)	α-amylase (%)			Sucrase (%)		
	50 µg	100 µg	200 µg	50 µg	100 µg	200 µg
Cladosporiumsp.	06.0	13.0	22.4	08.7	14.5	27.2
Nigrosporaoryze sp.	05.5	09.6	14.0	07.6	11.5	18.4
Streptomyces sp.	09.8	17.4	29.3	07.2	15.2	21.5
Acremonium sp.	00.6	01.3	03.0	00.5	01.1	02.9
Fusarium sp.	00.0	00.0	00.3	00.0	00.2	00.5
Curvularia sp.	00.1	00.4	01.0	00.4	00.9	01.4

Tab 5. In-vitro glucose diffusion test.

Name of Endophyte (Morphological basis)	Glucose diffusion test		
	50 µg	100 µg	200 µg
Cladosporiumsp.	-	-	+
***Nigrosporaoryze* sp.**	-	-	+
***Streptomyces* sp.**	+	+	++
***Acremonium* sp.**	-	-	+
***Fusarium* sp.**	-	-	-
***Curvularia* sp.**	-	-	+

Tab 6. In-vivo Serum glucose levels determination of diabetic induced rats with treatments

Groups	Serum glucose level (fasting) mg/dL		
	0 Day	30 Days	60 Days
Control	104±5.6	105±6.2	102±5.9
Diabetic control	242±22.5	240±25.8	243±26.5
Diabetic with	240±20.9	106±7.2	102±8.9

Glibenclaimide(30mg/kg)				
Diabetic with mycelium extract				
Cladosporiu msp.	**250 mg/kg**	241±19.4	160±12.8	150±11.6
	500 mg/kg	239±21.3	128±10.2	120±10.5
***Nigrosporaoryze* sp.**	**250 mg/kg**	244±20.8	148±10.6	145±12.1
	500 mg/kg	235±19.8	121±09.2	117±11.6
***Streptomyces* sp.**	**250 mg/kg**	238±19.5	147±11.8	139±12.4
	500 mg/kg.	231±18.9	118±11.4	109±12.7
***Acremonium* sp.**	**250 mg/kg**	237±19.6	162±14.1	151±11.7
	500 mg/kg	239±20.2	122±11.9	110±11.7
***Fusarium* sp.**	**250 mg/kg**	240±18.9	139±15.8	134±12.6
	500 mg/kg	239±19.9	108±14.2	102±13.8
***Curvularia* sp.**	**250 mg/kg**	242±21.5	145±11.5	142±13.2
	500 mg/kg	240±22.8	112±11.2	103±11.9

Tab 7. In-vivo haemoglobin levels determination of diabetic induced rats with treatments

Groups		**Haemoglobin gm/dL**		
		0 Day	**30 Days**	**60 Days**
Control		15.80±0.85	15.20±0.80	15.40±0.86
Diabetic control		10.10±1.00	09.52±0.95	07.15±0.98
Diabetic with Glibenclaimide (30mg/kg)		10.25±0.95	12.60±0.88	13.20±0.92
Diabetic with mycelium extract				
Cladosporiumsp.	**250 mg/kg**	09.95±0.94	10.92±0.91	12.05±1.00
	500 mg/kg	10.05±0.98	11.56±0.94	13.00±1.10
***Nigrosporaoryze* sp.**	**250 mg/kg**	10.10±1.00	10.95±0.93	12.00±0.98
	500 mg/kg	10.16±0.85	11.02±0.85	12.90±0.99
***Streptomyces* sp.**	**250 mg/kg**	10.00±0.92	10.25±0.91	11.10±0.82
	500 mg/kg.	09.80±0.87	11.10±0.88	12.15±0.95
***Acremonium* sp.**	**250 mg/kg**	09.90±0.92	10.24±0.86	10.98±0.86
	500 mg/kg	09.80±1.00	11.30±0.91	12.65±0.89
***Fusarium* sp.**	**250 mg/kg**	10.10±0.92	10.53±0.92	11.10±0.88
	500 mg/kg	10.15±0.99	11.40±0.89	12.95±0.85
***Curvularia* sp.**	**250 mg/kg**	10.25±0.86	10.13±0.83	11.00±0.91
	500 mg/kg	10.22±0.90	11.65±0.91	13.00±0.92

Cladosporium sp., *Nigrosporaoryze* sp., *Streptomyces* sp., *Acremonium* sp., *Fusarium* sp. and *Curvularia* sp.endophytes were found in neemleaves. Morphology of endophytes was determined according to their colour of mycelia, pigmentation and spore arrangement as in *Cladosporiumsp*. Olive brown, Dark brown and Un-branched; *Nigrosporaoryzae* sp. White, Black and Un-branched; *Streptomyces* sp. White, Whitish green and Branched filamentous; white colour of mycelia, nonappearance of pigmentation and single celled spore arrangement was found in isolate *Acremonium* sp; white colour, whitish pink pigment and sickle shaped spore arrangement found in species of *Fusarium*; blackish brown colour of mycelia, greenish brown pigmentation and branched septa spore arrangement was found in isolate of *Curvularia* sp. Qualitative characters of isolated endophytes were shown as Amylase in *Streptomyces* sp., *Acremonium* sp. and*Fusarium* sp.; Protease in *Cladosporiumsp*., *Streptomyces* sp., and *Curvularia* sp.; Cellulase in *Cladosporiumsp.*, *Streptomyces* sp. and*Acremonium* sp. whereas Lipase in in *Cladosporiumsp*., *Nigrosporaoryze* sp, *Acremonium* sp. and *Fusarium* sp. Quantity of Amylase in *Streptomyces* sp.-2.31, *Acremonium* sp.-1.90 and *Fusarium* sp.-2.71; Protease in *Cladosporium*sp.-1.22, *Streptomyces* sp.-1.63, and *Curvularia* sp.-1.17; Cellulase in *Cladosporium*sp.-0.25, *Streptomyces* sp.-0.45 and *Acremonium* sp-0.47. whereas Lipase in in *Cladosporium*sp.-0.16, *Nigrosporaoryze* sp.-0.25, *Acremonium* sp.-0.12 and *Fusarium* sp.-0.05.α-amylase inhibitory activity for 50µg, 100 µg and 200 µg concentration of*Cladosporium* species mycelia was determined. 0.6%, 13.0% and 22.4 % amylase inhibition was shown in 50µg, 100 µg and 200 µg concentration of *Cladosporium* species mycelia respectively. Sucrase inhibitory activity for 50µg, 100 µg and 200 µg concentration of *Cladosporium* species mycelia was determined. 08.7%, 14.5% and 27.2 % sucrase inhibition was shown in 50µg, 100 µg and 200 µg concentration of *Cladosporium* species mycelia respectively. α-amylase inhibitory activity for 50µg, 100 µg and 200 µg concentration of*Nigrospora oryze* species mycelia was determined. 05.5%, 09.6% and 14.0 % amylase inhibition was shown in 50µg, 100 µg and 200 µg concentration of *Nigrospora oryze* species mycelia respectively. Sucrase inhibitory activity for 50µg, 100 µg and 200 µg concentration of *Nigrospora oryze* species mycelia was determined. 07.6%, 11.5% and 18.4 % sucrase inhibition was shown in 50µg, 100 µg and 200 µg

concentration of *Nigrospora oryze* species mycelia respectively. α-amylase inhibitory activity for 50µg, 100 µg and 200 µg concentration of *Streptomyces* species mycelia was determined. 09.8%, 17.4% and 29.3 % amylase inhibition was shown in 50µg, 100 µg and 200 µg concentration of *Streptomyces* species mycelia respectively. Sucrase inhibitory activity for 50µg, 100 µg and 200 µg concentration of *Streptomyces* species mycelia was determined. 07.2%, 15.2% and 21.5 % sucrase inhibition was shown in 50µg, 100 µg and 200 µg concentration of *Streptomyces* species mycelia respectively. α-amylase inhibitory activity for 50µg, 100 µg and 200 µg concentration of *Acremonium* species mycelia was determined. 0.6%, 01.3% and 03.0% amylase inhibition was shown in 50µg, 100 µg and 200 µg concentration of *Acremonium* species mycelia respectively. Sucrase inhibitory activity for 50µg, 100 µg and 200 µg concentration of *Acremonium* species mycelia was determined. 00.5%, 01.1% and 02.9% sucrase inhibition was shown in 50µg, 100 µg and 200 µg concentration of *Acremonium* species mycelia respectively. α-amylase inhibitory activity for 50µg, 100 µg and 200 µg concentration of *Fusarium* species mycelia was determined. 00.0%, 00.0% and 00.3% amylase inhibition was shown in 50µg, 100 µg and 200 µg concentration of *Fusarium* species mycelia respectively. Sucrase inhibitory activity for 50µg, 100 µg and 200 µg concentration of *Fusarium* species mycelia was determined. 00.0%, 00.2% and 00.5% sucrase inhibition was shown in 50µg, 100 µg and 200 µg concentration of *Fusarium* species mycelia respectively. α-amylase inhibitory activity for 50µg, 100 µg and 200 µg concentration of *Curvularia* species mycelia was determined. 00.1%, 00.4% and 01.0% amylase inhibition was shown in 50µg, 100 µg and 200 µg concentration of *Curvularia* species mycelia respectively. Sucrase inhibitory activity for 50µg, 100 µg and 200 µg concentration of *Curvularia* species mycelia was determined. 00.4%, 00.9% and 01.4% sucrase inhibition was shown in 50µg, 100 µg and 200 µg concentration of *Curvularia* species mycelia respectively. Positive Glucose diffusion test was reported at 200 µg in *Cladosporium*sp. and *Nigrosporaoryze* sp.; at 50 µg, 100 µg and 200 µg *Streptomyces* sp. gives positive, fairly positive and strong positive results. *Acremonium* sp. and *Curvularia* sp. shown mild positive result in 200 µg but *Fusarium* sp. never showed any activity. Serum glucose (fasting) levels in Control, diabetic control, diabetic treated with glibenclaimide and treated with different mycelia extracts of isolated fungi with 250mg/kg and 500mg/kg concentration were reported in table 6. Levels of serum glucose (mg/dL) in control at 0^{th}, 30^{th} and 60^{th} days were 104±5.6, 105±6.2 and 102±5.9 respectively; diabetic control 242±22.5, 240±25.8 and 243±26.5 at 0^{th}, 30^{th} and 60^{th} day samples respectively; diabetic with glibenclaimide treated samples 240±20.9, 106±7.2 and 102±8.9 at 0^{th}, 30^{th} and 60^{th} day respectively; diabetic treated with 250mg/kg *Cladosporium*sp. mycelia extract 241±19.4, 160±12.8 and 150±11.6 at 0^{th}, 30^{th} and 60^{th} day correspondingly; diabetic treated with 500mg/kg *Cladosporium*sp. mycelia extract 239±21.3, 128±10.2 and 120±10.5 at 0^{th}, 30^{th} and 60^{th} day respectively; diabetic treated with 250mg/kg *Nigrosporaoryze* sp. mycelia extract 244±20.8, 148±10.6 and 145±12.1 at 0^{th}, 30^{th} and 60^{th} day correspondingly ; diabetic treated with 500mg/kg. *Nigrosporaoryze* sp.mycelia extract 235±19.8, 121±09.2 and 117±11.6 at 0^{th}, 30^{th} and 60^{th} day respectively; diabetic treated with 250mg/kg. *Streptomyces* sp. mycelia extract 238±19.5, 147±11.8 and 139±12.4 at 0^{th}, 30^{th} and 60^{th} day correspondingly; diabetic treated with 500mg/kg *Streptomyces* sp.mycelia extract 231±18.9, 118±11.4 and 109±12.7 at 0^{th}, 30^{th} and 60^{th} day respectively; diabetic treated with 250mg/kg *Acremonium* sp. mycelia extract 237±19.6, 162±14.1 and 151±11.7 at 0^{th}, 30^{th} and 60^{th} day correspondingly; diabetic treated with 500mg/kg *Acremonium* sp. mycelia extract 239±20.2, 122±11.9 and 110±11.7 at 0^{th}, 30^{th} and 60^{th} day respectively; diabetic treated with 250mg/kg *Fusarium* sp. mycelia extract 240±18.9, 139±15.8 and 134±12.6 at 0^{th}, 30^{th} and 60^{th} day correspondingly; diabetic treated with 500mg/kg *Fusarium* sp. mycelia extract 239±19.9, 108±14.2 and 102±13.8 at 0^{th}, 30^{th} and 60^{th} day respectively; diabetic treated with 250mg/kg *Curvularia* sp. mycelia extract 242±21.5, 145±11.5 and 142±13.2 at 0^{th}, 30^{th} and 60^{th} day correspondingly; diabetic treated with 500mg/kg *Curvularia* sp. mycelia extract 240±22.8, 112±11.2 and 103±11.9 at 0^{th}, 30^{th} and 60^{th} day respectively.Haemoglobin level in Control, diabetic control, diabetic treated with glibenclaimide and treated with different mycelia extracts of isolated fungi with 250mg/kg and 500mg/kg concentration were reported in table 7. Levels of haemoglobin (gm/dL) in control at 0^{th}, 30^{th} and 60^{th} days were 15.80±0.85, 15.20±0.80 and 15.40±0.86 respectively; diabetic control 10.10±1.00, 09.52±0.95 and 07.15±0.98 at 0^{th}, 30^{th} and 60^{th} day samples respectively; diabetic with glibenclaimide treated samples 10.25±0.95, 12.60±0.88 and 13.20±0.92 at 0^{th}, 30^{th} and 60^{th} day respectively; diabetic treated with 250mg/kg *Cladosporiumsp.* mycelia extract 09.95±0.94, 10.92±0.91 and 12.05±1.00 at 0^{th}, 30^{th} and 60^{th} day correspondingly; diabetic treated with 500mg/kg *Cladosporiumsp.* mycelia extract 10.05±0.98, 11.56±0.94 and 13.00±1.10 at 0^{th}, 30^{th} and 60^{th} day respectively; diabetic treated with 250mg/kg *Nigrosporaoryze* sp. mycelia extract 10.10±1.00, 10.95±0.93 and 12.00±0.98 at 0^{th}, 30^{th} and 60^{th} day correspondingly ; diabetic treated with 500mg/kg *Nigrosporaoryze* sp. mycelia extract 10.16±0.85, 11.02±0.85 and 12.90±0.99 at 0^{th}, 30^{th} and 60^{th} day respectively; diabetic treated with 250mg/kg *Streptomyces* sp. mycelia extract 10.00±0.92, 10.25±0.91 and 11.10±0.82 at 0^{th}, 30^{th} and 60^{th} day correspondingly; diabetic treated with 500mg/kg *Streptomyces* sp. mycelia extract 09.80±0.87, 11.10±0.88 and 12.15±0.95 at 0^{th}, 30^{th} and 60^{th} day respectively; diabetic treated with 250mg/kg *Acremonium* sp. mycelia extract 09.90±0.92, 10.24±0.86 and 10.98±0.86 at 0^{th}, 30^{th} and 60^{th} day correspondingly; diabetic treated with 500mg/kg *Acremonium* sp. mycelia extract 09.80±1.00, 11.30±0.91 and 12.65±0.89 at 0^{th}, 30^{th} and 60^{th} day respectively; diabetic treated with 250mg/kg *Fusarium* sp.mycelia extract 10.10±0.92, 10.53±0.92 and

11.10±0.88 at 0th, 30th and 60th day correspondingly; diabetic treated with 500mg/kg *Fusarium* sp.mycelia extract 10.15±0.99, 11.40±0.89 and 12.95±0.85 at 0th, 30th and 60th day respectively; diabetic treated with 250mg/kg *Curvularia* sp.mycelia extract 10.25±0.86, 10.13±0.83 and 11.00±0.91 at 0th, 30th and 60th day correspondingly; diabetic treated with 500mg/kg *Curvularia* sp.mycelia extract 10.22±0.90, 11.65±0.91 and 13.00±0.92 at 0th, 30th and 60th day respectively.

IV. DISCUSSION

Morphological characters of isolated fungi were olive brown colour, dark brown pigment and un-branched spore arrangement and shows positive enzyme activities for protease, cellulose and lipase with 1.22, 0.25 and 0.16 U/ml production. Isolated endophytic fungi with these characters were earlier reported as species of *Cladosporium* (Ogorek *et al.,* 2012; Braun *et al.,* 2003). isolated fungi were white colour of mycelia, black pigmentation and unbranched spore arrangement and shows positive enzyme activities for lipase only with 0.25 U/ml production. Similar result was reported by Abbas and Mohammad in 2014 and confirms the isolated endophytic fungi with these characters were a species of *Nigrosporaoryze*. Morphological characters of isolated fungi were white colour of mycelia, whitish green pigmentation and branched filamentous spore arrangement and shows positive enzyme activities for amylase, protease and cellulose with 2.31, 1.63 and 0.45 U/ml production. Isolated endophytic fungi with these characters were earlier reported as species of *Streptomyces* (Tadai *et al*., 2006).Morphological characters of isolated fungi were white colour of mycelia, non-appearance of pigmentation and single celled spore arrangement and shows positive enzyme activities for amylase, cellulose and lipase with 1.90, 0.47 and 0.12 U/ml productions. Similar result was reported by Lim *et al.,* 2002; Matsumura *et al.,* 1980 and confirms the isolated endophytic fungi with these characters were a species of *Acremonium.* Endophytic isolate was reported white colour, whitish pink pigment and sickle shaped spore arrangement morphological characters and shown enzymatic activities amylase and lipase only with 2.71 and 0.05. Joshi *et al.,* 2013; Minuto *et al.,* 1995a reported isolate species having similar characters was a member of *Fusarium*. Morphological characters of isolated fungi were blackish brown colour of mycelia, greenish brown pigmentation and branched septa spore arrangement and shows positive enzyme activities for protease only with 1.17 U/ml production. Isolated endophytic fungi with these characters were earlier reported as species of *Curvularia* (Jain, 1962; Pereira *et al.,* 1998).Mycelia extract of *Streptomyces*, *Cladosporium* and *Nigrospora oryze* species isolates were reported best inhibition ranging 15 to 38% α-amylase and sucrose. Isolates of *Acremonium*, *Curvularia* and *Fusarium* species were showed low potency to inhibit α-amylase and sucrose. Similarly, *Streptomyces*, *Cladosporium* and *Nigrospora oryze* species chronically gives best results as compare to isolates of *Acremonium*, *Curvularia* and *Fusarium* species in glucose diffusion test. Ushasri and Anusha worked on *in-vitro* anti-diabetic properties in 2015 and reported that the endophytic extracts have good potency to α-amylase and sucrose inhibition with up to 23.7% inhibition. Wan *et al.,* (2013) also reported that swartia extract resulted best result for reducing diabetes. Blood functions tests (% haemoglobin and White blood cells count) and Body weight studies were performed at 0th, 30th and 60th days for *in-vivo* anti-diabetic activities determinations. Yuan and associates (2014) worked on effect of *Actinida* extract in diabetic induced mice and reported that alpha-glucosidase inhibitory activity test, ethanol extract of roots showed the best inhibitory activity (74.2%, 6 mg/ml). Ushasri and Anusha (2015) previously reported the effect of endophytic extract on allexon induced diabetic mice that showed highest glycosidase inhibition in treated mice. In this study serum glucose level was decrease from 240gm/dL to 102gm/dL in 60 days administration.

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