Nutrition and Foods Safety Agency of the Centre for Disease Prevention and Control, People's Republic of China

Testing Report

Sample processing number: 200405018

Name of Sample:	Eleotin
Sample received from:	Botali International Enterprise Co., Ltd Tianjin Port Free Trade Zone
Testing type:	As per requested
Publication date:	June 23, 2005

Nutrition and Foods Safety Agency of the Centre for Disease Prevention and Control, People's Republic of China

Xi Yuan Hospital of China Academy of Traditional Chinese Medicine

Testing Report

Name of Sample:	Eleotin
Sample received from:	Botali International Enterprise Co., Ltd Tianjin Port Free Trade Zone
Manufacturer:	Eastwood Bio-Medical Research Inc.
Sample Number:	20040204
Expiration Date:	24 months from manufacturing date **condition: store under normal room temperature**
Sample Description:	Type: capsule Colour: contents- light brown powder
Amount of Sample Received:	Eleotin capsules and placebo capsules, at 0.3g/capsule x 30 capsules/bottle x 360 bottles each
Sample Received on:	Jan 04th, 2005
Testing Criteria:	Sample's functions as a supplement in decreasing blood glucose level (human-testing)
Testing Reference:	Natural Health Products Testing and Evaluation Guidelines- 2003

Testing Results:

One hundred subjects with high blood glucose levels were selected and randomly divided into two groups: treatment group and control group, each with fifty subjects. During the testing period, subjects took their original blood glucose controlling medications on regular schedules and doses; Eleotin doses were added to the treatment group while placebo doses were added to the control group. Intra-group (differences before and after treatment within the same group) and inter-group (differences between treatment group and control group) comparisons were made and observations were recorded. One month later, results showed significant

differences in the empty-stomach (fasting) blood glucose levels of the treatment group before and after testing. The percentage decrease in the blood glucose level of empty-stomach treatment group subjects was 11.2%; when this percentage decrease was compared with that of the control group, the differences reach statistical significance. The 2-hour post meal blood glucose levels of the treatment group were significantly different before and after treatment started, with a 20.9 % decrease in the blood glucose levels post-meal; when this percentage decrease was compared with that of the control group, differences reached statistical significance. According to the standards set by Natural Health Products Testing and Evaluation Guidelines-2003, the testing of whether Eleotin is effective in supplementing the decrease of blood glucose level in a human experiment returned positive results. (end of page)

Eleotin's functions as a supplement in decreasing blood glucose level: Human-Testing Report

1 Materials and Methods

1.1 Sample: Eleotin capsules and placebo capsules, provided by Botali International Enterprise Co., Ltd, Tianjin Port Free Trade Zone; manufactured by Eastwood Bio-Medical Research Inc.

1.2 Selection of Subjects:

Selection Criteria: Adults, Type II Diabetics, with stable conditions controlled either by diet modifications or by taking oral medications (no modifications were made to these patients' medications and doses), with empty-stomach blood glucose level ≥7.8 mmol/L (140mg/dl) and 2-hour post-meal blood glucose level ≥11.1 mmol/L (200mg/dl); subjects with high blood glucose levels of 7.8 mmol/L ≥ empty stomach blood glucose level ≥ 6.7 mmol/L (120mg/dl) and 11.1 mmol/L (200mg/dl) ≥ 2-hour post-meal blood glucose level ≥ 7.8 mmol/L were also eligible.

1.3 Ineligible Subjects:

1.3.1 Type I diabetics.

1.3.2 Persons ages 18 and under or 65 and over; pregnant or nursing women.

1.3.3 Persons with heart, liver, and/or kidney diseases, or other severe illnesses, mental illnesses.

1.3.4 Persons who cannot adapt to diet modifications thus influencing testing outcomes.

1.3.5 Persons not meeting the selection criteria, not taking treatments on time, and/or with insufficient data thus influencing testing outcomes.

1.4 Testing Procedure: 100 subjects were selected and randomly assigned into two groups. Subjects took their original blood glucose controlling medications on regular schedules and doses. Eleotin doses were added to the treatment group, while placebo

doses (capsules with similar appearance, colours, and packaging) were added to the control group both at 2 capsules each time and 3 times daily. Subjects were asked to consistently monitor their food intake and consume accordingly. Results were compared within the groups and between the groups.

1.5 Instrument and Testing Reagent: Sysmex F-820 haematology analyzer (UK), Midiron urine analysis instrument (Germany), Bayer Technicon RA-1000 analyzer (United States), biochemical testing reagents/ papers, all provided by Biosino Bio-Technology & Science Inc.

2 Parameter / Index of Observation:

All indexes were recorded before and after testing. Empty-stomach blood glucose levels were measured on the 15^{th} day.

2.1 Observation of Effects

2.1.1 Observation of Symptoms

Carefully examined subjects' medical history, diet, level of physical fitness, and major clinical symptoms: increased appetite, polydipsia (increased thirst, and consequent increased fluid intake), polyuria (frequent urination), and irreducible fatigue. According to the level of severity of the symptoms (severe:3, less severe:2, non severe:1), statistical analysis was performed before and after testing. Observed the percentage improvement of symptoms (an improvement of 2 points means significant, 1 point means effective).

2.1.2 Observed and recorded empty-stomach blood glucose levels and 2-hour post meal blood glucose levels.

2.1.3 Tested subjects for urine sugar content as well as for urine ketones.

2.1.4 Tested subjects' serum lipid: total cholesterol (TC) and triglyceride (TG).

2.2 Subjects observed for treatment safety:

2.2.1 Subjects received routine blood, urine, and stool check ups:
Red blood cells (erythrocyte) count, haemoglobin (Hb or HGB), white blood cells (leukocyte) count, routine urine check up, routine stool check up.

2.2.2 Testing of biochemical indexes:
Blood serum ALB (Albumin), TP (total protein); heart, liver, and kidney functions— AST (Aspartate aminotransferase) (GOT), ALT (Alanine aminotransferase) (GPT), urea (BUN), and Cre.

2.2.3 Abdominal ultrasound, electrocardiogram, chest X-ray (tests done before treatment)

3 Statistical Analysis and Results Determination:

3.1 Statistical Analysis: Paired t-test was performed on values obtained within the group, regular t-test was performed to compare the mean values of the two groups, the equality (homogeneity) of variance test was also performed to compare the mean values obtained from both groups, appropriate $\log(Y_i)$ transformation was performed on means with non-normal distributions and/or inequality of variance to stabilize the variance. After normal equality of variance was achieved, t-test was performed on the transformed data; when normal equality of variance could not be achieved even with the transformed data, t-test

or Wilconxon's rank sum test was performed; data with equality of variance but too high a critical value (i.e. CV>50%) were analyzed with Wilconxon's rank sum test.
3.2 Results Determination: The difference in fasting blood glucose level before and after testing of the treatment group was statistically significant; in addition, an average of ≥ 10% decrease was observed in the blood glucose level of treatment subjects. The blood glucose level and the percentage decrease in the blood glucose level of the treatment group after testing were significantly different when compared with those of the control group. With the results stated above, it can be concluded that the testing of whether Eleotin decreases fasting blood glucose level returned positive results.

The difference in the 2-hour post meal blood glucose level before and after testing of the treatment group was statistically significant; in addition, an average of ≥ 10% decrease was observed in the blood glucose level of treatment subjects. The blood glucose level and the percentage decrease in the blood glucose level of the treatment group after testing were significantly different when compared with those of the control group. With the results stated above, it can be concluded that the testing of whether Eleotin decreases the 2-hour post meal blood glucose level returned positive results.

4 Results:
4.1 General Information:
Total number of subjects: 100.
Treatment group: 17 male and 33 female subjects, ages 36-65, with an average age of 55.3, and an average diabetes history of 4.8 years.
Control group: 18 male and 32 female subjects, ages 41-65, with an average age of 57.0, and an average diabetes history of 5.4 years.
4.2 A comparison of the two groups before testing:

Table 1. Comparison of Conditions before testing (X±SD)

Groups	Number of subjects	Age	Sex		History of diabetes (years)	Blood glucose level	
			M	F		After fasting	2 hour post meal
Treatment	50	55.3 ± 7.4	17	33	4.8 ± 4.0	9.1 ± 1.8	13.2 ± 2.5
Control	50	57.0 ± 7.3	18	22	5.4 ± 3.9	8.5 ± 1.9	12.8 ± 2.9

Table 2. Blood glucose controlling medications taken by subjects

Groups	Number of subjects	Sulfonylurea group	Biguanide group	Sulfonylurea + biguanide	Not on medication
Treatment	50	17	13	16	4
Control	50	18	12	17	3

Table 1 and 2 show that the conditions of the subjects in both groups were similar before testing, it would therefore be beneficial to compare their results after the test.

4.3 Effects in decreasing blood glucose level:

4.3.1 Comparison of after fasting and post meal blood glucose levels:

Table 3. Comparison of after fasting and 2-hour post meal blood glucose levels of both groups (mmol/L, X±SD)

Groups	After fasting blood glucose level			2-hour fasting blood glucose level		
	Before treatment	Post treatment	% Decrease	Before treatment	Post treatment	% Decrease
Treatment	9.1±1.8	8.0±1.4**	11.2±9.6##	13.2±2.5	11.5±2.0**#	20.9±17.8##
Control	8.5±1.9	8.4±1.9	1.2±4.5	12.8±2.9	12.7±2.8	2.0±7.6

Intra-group comparison **p<0.01 Inter-group comparison #p<0.01, ##p<0.01

One month later, results showed significant differences in the empty-stomach (fasting) blood glucose levels of the treatment group before and after testing. The percentage

decrease in the blood glucose level of empty-stomach treatment group subjects was 11.2%; when this percentage decrease was compared with that of the control group, the differences reach statistical significance. The 2-hour post meal blood glucose levels of the treatment group were significantly different before and after treatment started, with a 20.9 % decrease in the blood glucose levels post-meal; when this percentage decrease was compared with that of the control group, differences reached statistical significance.

4.3.2 Comparison of changes in after fasting blood glucose level, urine sugar content, and urine ketones:

Table 4. Comparison of after fasting blood glucose level before, during, and after treatment and changes in urine sugar content and urine ketones (X±SD)

Types	Subjects	Before treatment	After treatment	Difference
Blood glucose level (mmol/L)	50	9.1±1.8	8.0±1.4**	-1.1±1.1**
	(50)	(9.2±2.3)	(9.4±2.5)	(+0.2±1.6)#
Urine sugar content	50	0.4±0.8	0.2±0.6*	0.2±0.5
	(50)	(0.57±1.02)	(0.5±1.0)	(-0.0±0.1)
Urine keytone body	50	-	-	
	(50)	(-)	(-)	

( )Control group intra-group comparison *p<0.05, **p<0.01 inter-group comparison #p<0.05

4.4 Comparison of effects in decreasing the blood glucose level:

Table 6. Blood glucose level and effectiveness of treatment

Groups		Effective	Ineffective	Total % effectiveness
Treatment	Number of subjects	32	18	32
	% effectiveness	64.0	36.0	64.0#
Control	Number of subjects	4	46	4
	% effectiveness	8.0	92.0	8.0

The differences between the two groups reach statistical significance, #$p<0.05$

4.5 Improvement of symptoms:

Table 7. Improvement of major clinical symptoms

Symptoms	Number of subjects	Minor improvement	Effective	Ineffective	% Improvement
Increased appetite + increased hunger	16	1	8	7	56.3
	(17)	(0)	(5)	(12)	(29.4))
Polydipsia	20	1	9	10	50.0
	(17)	(1)	(6)	(10)	(41.2)
Polyuria	16	1	8	7	56.3
	(15)	(0)	(6)	(9)	(40.0)
Irreducible fatigue	21	4	7	10	52.4
	(22)	(0)	(9)	(13)	(40.9)

( ) Control group

Percentage improvements of major clinical symptoms of treatment group are higher than that of the control group

4.6 Statistical values of clinical symptoms

Table 8. Statistical values of clinical symptoms (X±SD)

Groups	Number of subjects	Before treatment	During treatment	After treatment
Treatment	50	7.0±4.2	6.1±3.8**	5.4±3.6**
Control	50	7.6±4.5	7.2±4.3**	6.9±4.2**

Intra-group comparison **p<0.01

Both the treatment group and the control group showed improvements in clinical symptoms.

4.7 Serum lipid changes:

Table 9. Comparisons of serum lipid changes before and after treatment (X±SD)

Types	Treatment group (n=50)		Control group (n=50)	
	Before treatment	After treatment	Before treatment	After treatment
TC (mmol/L)	6.1±1.2	5.9±1.1	6.4±1.0	6.4±1.1
TG (mmol/L)	1.7±1.3	1.6±1.1	1.7±0.9	1.8±0.9
HDL-C(mmol/L)	1.1±0.3	1.2±0.3	1.1±0.3	1.1±0.3

Changes in serum lipid before and after treatment with both groups are not statistically significant.

4.8 Monitoring of changes in blood, urine, and stool:

Table 10. Comparison of changes in blood, urine, and stool before and after treatment for monitoring the safety of treatments (X±SD)

Types	Treatment group (n=50)		Control group (n=50)	
	Before treatment	After treatment	Before treatment	After treatment
TP (g/L)	75.0±4.2	74.8±4.2	76.3±3.8	76.3±3.5
ALB (g/L)	42.8±2.3	43.7±2.6	42.4±2.7	42.3±2.6
ALT (u/L)	17.8±8.3	15.2±7.5	19.3±8.9	18.3±7.1
AST (u/L)	14.9±5.4	16.9±4.7	13.0±4.3	12.9±3.8
UREA(mmol/L)	5.3±1.5	5.2±1.6	5.7±1.0	5.6±1.0
Cre (umol/L)	103.3±11.9	102.3±13.6	106.9±9.8	106.1±10.2
HGB (g/L)	145.3±13.8	144.6±13.8	145.5±13.8	146.5±12.9
RBC(x1012/L)	4.7±0.4	4.8±0.4	4.7±0.4	4.8±0.4
WBC(x109/L)	7.0±1.6	6.9±1.5	7.0±1.5	6.8±1.4
Urine (except for urine sugar content)	Normal	Normal	Normal	Normal
Stool	Normal	Normal	Normal	Normal

All indicators (except for urine sugar content) are within normal range for both groups before and after treatment.

4.9 Testing results of abdominal ultrasound, electrocardiogram, and chest X-rays: all within normal range.

5 Conclusion:

5.1 Results showed significant differences in the empty-stomach (fasting) blood glucose levels of the treatment group before and after testing. The percentage decrease in the blood glucose level of empty-stomach treatment group subjects was 11.2%; when this percentage decrease was compared with that of the control group, the differences reached statistical significance. The 2-hour post meal blood glucose levels of the treatment group were significantly different before and after treatment started, with a 20.9 % decrease in the blood glucose levels post-meal; when this percentage decrease was compared with that of the control group, differences reached statistical significance. According to the standards set by Natural Health Products Testing and Evaluation Guidelines- 2003, the testing of whether Eleotin is effective in supplementing the decrease of blood glucose level in a human experiment returned positive results.

5.2 After Eleotin treatment, haemoglobin (Hb or HGB), red blood cells (erythrocyte) count, haemoglobin (Hb or HGB), white blood cells (leukocyte) count, blood serum ALB (Albumin) and TP (total protein), AST (GOT), ALT (GPT), urea (BUN), Cre, urine and stool levels were all within normal range, thus it can be concluded that Eleotin has no negative side effects on the subjects' health.

5.3 Allergic reactions and undesirable effects were not detected during the duration of treatment.