ORIGINAL RESEARCH

Role of Parenteral Amino Acid in Patients of Diabetic Foot Ulcer

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Abstract
Aim: To analyse the role of amino acids among subjects with type 2 diabetes having foot ulcer.
Material and method: The present study was conducted in the Department of Surgery, Rohilkhand Medical College and Hospital, Bareilly from February 2020 to March 2021. The study comprised of 60 patients, aged 18 years and above, suffering from diabetic foot ulcer. Patients were divided into two groups i.e. group A and B. Patients of group A continued to take diabetic diet as they were taking before while patients of group B were given IV amino acid (200ml) on alternate day till 15th day in addition to the diet they were taking before. Ulcers of all patients were properly cleaned and dressed under aseptic condition with normal saline and povidone iodine everyday, wound debridement was done as required. Ulcers were assessed on day 0, 5, 10 and 15.
Results: Mean cholesterol increased in group A while it decreased in group B at different intervals with statistically insignificant difference as $p>0.05$. At the end of study, normal healing was found among 26.67% and 66.67% of the subjects in group A and B respectively. Major haematoma was reported only in group A and that among 10% of subjects. Hence healing was observed better among group B subjects.
Conclusion: It can be concluded from the results that among subjects with diabetic foot ulcers, amino acid administration leads to better healing and satisfaction among the patients. Therefore, amino acids supplement should be given to diabetic foot ulcer subjects.
Keywords: Diabetes, Ulcer, Amino acid, Healing

Introduction: Diabetes is a chronic disease that stems from pancreatic β cell dysfunction, impairing normal production of insulin which leads to high and also fluctuating, blood glucose levels.[1,2] Diabetes mellitus is a group of metabolic diseases associated with long term effects on many organs which results in micro and macro vascular complications[3,4]. A combination of these latter two conditions with improper footwear leads to loss of sensitivity in the feet of diabetic patients, which facilitates instalment of chronically-infected lesions, the so-called diabetic foot ulcers (DFU), ultimately leading to limb amputations[1]. DFU is a non-healing or poorly healing, partial or full thickness wound below the ankle, in an individual with diabetes[5].

Diabetic foot ulcer is one of the most common complications of diabetes which is often difficult to heal.[2] Wound healing process consists of a series of interactions between different cell types, cytokine mediator and the extracellular matrix. This process is successful if there is adequate blood supply and nutrients supply to the site of damaged tissue[3,4].

Management of DFU should always start with prevention, through policies for foot inspection and use of custom therapeutic footwear to prevent ulceration, as DFU are difficult to heal once installed. Nutrition therapy plays a vital role in the management of diabetes and complications related to it. Poor nutrition before or during the healing process may delay wound healing and impair wound strength. Protein is one of the most important macronutrient as it is essential for woundhealing.[6,7] It is required for all stages in healing process including fibroblast proliferation, collagen synthesis, angiogenesis and immune function[8].

Amino acids are the structural units (monomers) that make up proteins. Specifically, a protein is made up of one or more linear chains of amino acids, each of which is called a polypeptide. Amino acids can, under appropriate conditions, enhance insulin secretion from primary islet cells and βcell lines.[9] In vivo, L-glutamine and L-alanine are quantitatively the most abundant amino acids in the blood and extracellular fluids followed closely by the branched chain amino acids. However, unlike glucose, individual amino acids do not provoke insulin secretion in vitro when added at physiological concentrations.[9,10]

In periods of fasting or starvation, amino acid release from skeletal muscle (primarily L-glutamine and L-alanine)may modulate glucagon release from pancreatic αcells,which subsequently may influence insulin secretion from βcells. Dietary amino acids may also stimulate incretin release, e.g., GLP-1, from intestinal L-cells and therefore stimulate insulin secretion via indirect mechanisms.[11]

European Pressure Ulcer Advisory Panel and Agency for Health Care Policy and Research have both recommended increasing intake of protein for wound healing process. Inadequate protein intake may result in delay in wound healing process. Although the effect of inadequate protein intake has been observed in non-diabetic people with wounds, there is very scarce data regarding the role of parenteral amino acid in patients of diabetic foot. Hence the present study was conducted to analyse the role of amino acids among subjects with type 2 diabetes having foot ulcer.

Materials and Method: The present study was conducted in the Department of Surgery, Rohilkhand Medical College and Hospital, Bareilly from February 2020 to March 2021. The study comprised of 60 patients, aged 18 years and above, suffering from diabetic foot ulcer. Patients were enrolled in the study after obtaining written informed consent and approval from Institutional Ethical Committee.Physicians and laboratory technicians who worked with the study participants,the participants themselves, and researchers involvedin the study were also blinded to whether the patients wereassigned to treatment group or control group.

Group A 30- Patient continued taking same diet as they were taking before.

Group B 30- Patients Amino acids (200ml) was given intravenously on alternate days for 15 days in addition to the diet they were taking previously.

Method of Data Collection- Patients were recruited into this study according to the following inclusion and exclusion criteria:

Inclusion criteria: All adult patients suffering from type 2 diabetes mellitus and having diabetic foot ulcer of any size and severity admitted in surgery wards of Medical College.

Exclusion criteria:Children and pregnant women, patients with acute illnesses, type 1 diabetes mellitus, complications like nephropathy, liver diseases etc, lower extremity wounds of other than diabetic origin, those who received hyperbaricoxygen therapy, had collagen-

based topicalwound therapy or recent (within 3 month) skin grafting were excluded from the study.

The data was collected by a preformed structured interviewer-administered questionnaire that was pretested with modifications made prior to its use in the study. The patients were interviewed about the demographic, socioeconomic status, medical history and previous history of taking any medications and supplements.

Patients of group A continued to take diabetic diet as they were taking before while patients of group B were given IV amino acid (200ml) on alternate day till 15th day in addition to the diet they were taking before.

Demographic data of both groups were recorded i.e. age, gender, weight, height, body mass index, smoker/non-smoker, duration of diabetes mellitus, systolic blood pressure and diastolic blood pressure.

Investigations viz. complete blood count (CBC), serum creatinine, Total Protein, S.Albumin, SGOT, SGPT, ALP, Serum Triglycerides, HDL, LDL Cholesterol, HBA1c and Random Blood Sugar was done.

Procedure:

- All patients with diabetic foot were admitted.
- All baseline investigations were done.
- X-ray of affected foot in antero-posterior and oblique view was taken to rule out bony involvement.
- Pus-culture and sensitivity test were for all patients.
- Ulcers of all patients were properly cleaned and dressed under aseptic condition with normal saline and povidone iodine everyday, wound debridement was done as required.
- Dressings were changed daily.
- Antibiotics were given according to culture and sensitivity report.
- Ulcers were assessed on day 0, 5, 10 and 15.

Patients were assessed on the basis of blood investigations results, size of ulcer, wound healing and pain. Hb, TLC, DLC, S.Creatinine, HBA1c, Total Protein,wound healing were documented at intervals of 5 days on 5, 10 and 15th day. Signs of inflammation were assessed along with edema.

Wound healing was assessed on the basis of:

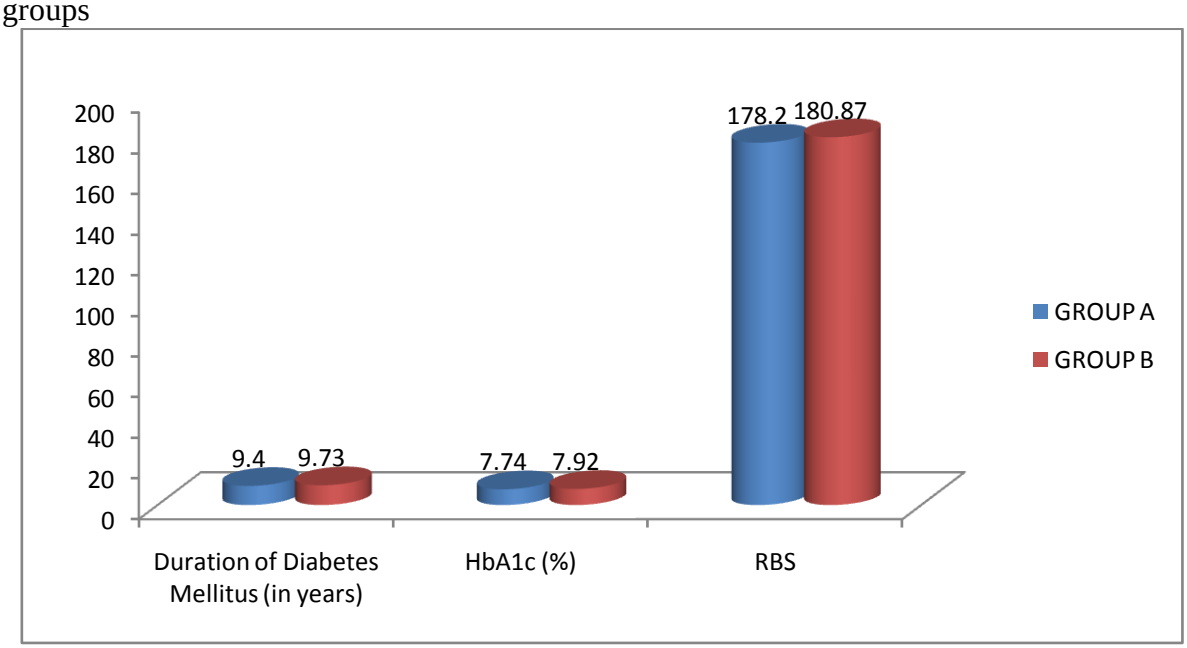
a. Southampton Scoring System
b. Asepsis Wound Scoring System

All the findings and observations were coded and entered in Excel master sheet.

Statistical analysis: Data so collected was tabulated in an excel sheet, under the guidance of statistician. The means and standard deviations of the measurements per group were used for statistical analysis (SPSS 22.00 for windows; SPSS inc, Chicago, USA). For each assessment point, data were statistically analyzed using one way ANOVA. Difference between two groups was determined using student t-test as well as chi square test and the level of significance was set at $p < 0.05$.

Results:In both the groups, male was more as compared to females. In group A and B, 60% and 46.67% of the subjects were having age between 51-60 years. Minimum subjects were in the age group of 31-40 years followed by >60 years. Graph 1 shows the comparison of mean duration of diabetes mellitus (in years), HbA1c (%) and RBS among the study groups. Mean duration of diabetes mellitus (in years), HbA1c (%), RBS was 9.40, 7.74, 178.20 and 9.73, 7.92, 180.87 in group A and B respectively.

Graph 1: Mean duration of diabetes mellitus (in years), HbA1c (%) and RBS among the study groups



Mean cholesterol increased in group A while it decreased in group B at different intervals with statistically insignificant difference as p>0.05. Mean HDL at baseline was 35.17 and 35.97 in group A and B respectively with statistically insignificant difference. Mean HDL increased in both the groups i.e. 36.02 in group A and 36.48 in group B at different intervals, though statistically insignificant as p>0.05. Mean LDL decreased in both the groups i.e. 102.33 in group A and 95.16 in group B at different intervals, though statistically insignificant as p>0.05.

Table 1: Comparison of lipid profile among the study groups at different intervals

Groups		**CHOLESTEROL**				**Anova Test**	**p value**
		Baseline	**5th Day**	**10th Day**	**15th Day**		
Group A	Mean	171.67	178.33	183.07	183.14	2.82	0.10
	SD	36.33	38.39	33.41	32.89		
Group B	Mean	165.20	166.27	162.11	160.76	2.32	0.27
	SD	34.81	35.79	33.08	37.71		
t test		0.49	1.37	2.13	2.27		
p value		0.48	0.29	0.13	0.10		
		HDL					
Group A	Mean	35.17	36.07	36.32	36.02	1.07	0.51
	SD	12.99	8.70	8.88	9.09		
Group B	Mean	35.97	36.23	36.53	36.48	0.89	0.64
	SD	13.08	9.25	9.20	8.91		
t test		0.69	0.52	0.81	0.40		
p value		0.41	0.48	0.36	0.62		
		LDL					

Group A	Mean	102.40	104.40	100.32	101.33	1.43	0.37
	SD	18.81	16.53	16.94	15.71		
Group B	Mean	99.82	96.27	96.18	95.16	1.68	0.38
	SD	17.01	16.13	18.59	15.51		
t test		0.91	0.30	1.38	2.31		
p value		0.32	0.58	0.24	0.12		

Mean Creatinine (mg/dl) at baseline was 1.46 and 1.51 in group A and B respectively with statistically insignificant difference. Mean Creatinine (mg/dl) decreased in both the groups i.e. 1.07 in group A and 1.09 in group A and B at different intervals with statistically insignificant difference as $p>0.05$. Mean uric acid (mg/dl) decreased in both the groups i.e. 4.68 in group A and 4.83 in group A and B at different intervals with statistically insignificant difference as $p>0.05$ (table 2).

Table 2: Comparison of Creatinine (mg/dl) and Uric acid (mg/dl) among the study groups at different intervals

Groups		**Creatinine (mg/dl)**				**Anova Test**	**p value**
		Baseline	**5th Day**	**10th Day**	**15th Day**		
Group A	Mean	1.46	1.24	1.16	1.07	1.39	0.32
	SD	1.09	1.24	1.13	1.38		
Group B	Mean	1.51	1.22	1.13	1.09	1.04	0.47
	SD	1.42	1.33	1.21	1.44		
t test		0.26	0.12	0.14	0.21		
p value		0.61	0.73	0.71	0.65		
		Uric acid (mg/dl)					
Group A	Mean	5.58	5.27	4.86	4.68	1.93	0.26
	SD	2.96	2.57	2.90	3.07		
Group B	Mean	6.03	5.50	5.10	4.83	1.67	0.34
	SD	2.53	2.64	2.84	2.91		
t test		1.16	0.34	0.71	0.19		
p value		0.29	0.56	0.32	0.66		

At the end of study, normal healing was found among 26.67% and 66.67% of the subjects in group A and B respectively. Major haematoma was reported only in group A and that among 10% of subjects. Hence healing was observed better among group B subjects. When group A and B was compared statistically w.r.t Southampton Scoring System, it was found to be statistically significant at all the intervals as $p<0.05$ (table 3).

Table 3: Southampton Scoring System among the study groups at 15th day

Category	**Group A (Control)**		**Group B (With Amino Acids)**		**Chi Square**	**p value**
	N	**%**	**N**	**%**		
Normal healing	8	26.67	20	66.67	12.16	0.007*
Minor Complication	9	30	7	23.33		

Wound Infection	10	33.33	3	10		
Major Haematoma	3	10	0	0		
Total	30	100	30	100		

*: statistically significant

At the end of study, satisfactory healing was found among 23.33% and 66.67% of the subjects in group A and B respectively. Severe wound infection was reported only in group A i.e. among 10% of subjects. When group A and B was compared statistically according to Asepsis Wound Scoring System, it was found to be statistically significant at all the intervals as $p<0.05$ (table 4).

Table 4: Asepsis Wound Scoring System among the study groups at 15th day

Category	**Group A (Control)**		**Group B (With Amino Acids)**		**Chi Square**	**p value**
	N	**%**	**N**	**%**		
Satisfactory healing	7	23.33	20	66.67	12.27	0.006*
Disturbance of Healing	10	33.33	7	23.33		
Minor Wound Infection	10	33.33	3	10		
Severe Wound Infection	3	10	0	0		
Total	30	100	30	100		

*: statistically significant

Discussion: In the treatment of diabetes and problems linked to it, diet therapy plays a crucial role. Poor nutrition can slow down wound healing and impair wound strength before or during the healing process. As it is crucial for wound healing, protein is one of the most essential macronutrients. It is essential for all phases of the healing process, including fibroblast proliferation, synthesis of collagen, angiogenesis, and immune function[1].

Inadequate protein intake can contribute to a delay in the process of wound healing. While the impact of insufficient protein consumption in non-diabetic people with injuries has been observed, there is relatively little knowledge on the role of parenteral amino acids in patients with diabetic foot ulcers. In order to examine the function of amino acids among subjects with type 2 diabetes with foot ulcer, the present research was therefore carried out.

In both the groups, males were more in number as compared to females in this study. Maris S. Joneset al[12] in their study revealed similar male dominance. Similarly Bhaktavatsalam M et al[13] found that more males were affected than females. In a study by Nida Sajid et al[14] a total of 542 subjects were included in the study, out of which 365 (67.2%) were males and 178 (32.8%) were females. SambashivaRao. G et al[15] in their study reported that male preponderance was due to their more exposure to injuries during their occupational and recreational activities.

In group A and B, 60% and 46.67% respectively of the subjects were of the age between 51-60 years. Minimum subjects were in the age group of 31-40 years followed by >60 years in our study. Mean age in present study is 56 ± 11.68yrs In a study by NidaSajid et

al,[14] mean age of the subject was 54.61±10.51 years. Pemayun TG et al[16], found that the mean age in their study was 54.3±8.6 years. Similarly Bhaktavatsalam M et al[13] found that majority of the patients were found in the age group of 51-60 years (35.5%) followed by 41-50 years (23.5%).

Mean duration of diabetes mellitus, HbA1c (%), RBS was 9.40 years, 7.74, 178.20 and 9.73 years, 7.92, 180.87 in group A and B respectively. Hence prolonged duration of diabetes mellitus is a significant risk factor for diabetic foot ulcers. Al-Rubeaan K et al[17] in their study revealed that the prevalence of complications of foot was associated with duration of diabetes.

Mean S.HDL at baseline were 35.17mg/dl and 35.97mg/dl in group A and B respectively with statistically insignificant difference. Mean HDL increased in both the groups i.e. 36.02 in group A and 36.48 in group B at different intervals, though statistically insignificant as $p>0.05$. In a study by NidaSajidet al[14], mean HDL at baseline was 28.95±10.63.

Mean S.Creatinine (mg/dl) at baseline was 1.46mg/dl and 1.51mg/dl in group A and B respectively with statistically insignificant difference. Mean Creatinine (mg/dl) decreased in both the groups i.e. 1.07 in group A and 1.09 in group A and B at different intervals with statistically significant difference as $p<0.05$ in our study. In a study by NidaSajidet al[14], mean serum creatinine baseline was1.13±0.29 (mg/dl).

At the end of study, normal healing was observed among 26.67% and 66.67% of the subjects in group A and B respectively. Major haematoma was reported only in group A(in 10% of subjects). Hence better healing was observed among group B subjects. When group A and B were compared statistically w.r.t Southampton Scoring System, it was found to be statistically significant at all the intervals ($p<0.05$) in our study. David and Graham study reported about the relationship between wound healing and nutritional state among surgical subjects, they observed that the wound healing processwas prolonged among malnourished subjects[18]. In a study by Gray D et al observed a mixture of three amino acid arginine,glutamine and β-hydroxy-β-methyl-butyrate had reduced cost of antibiotic and improvement occuring wound healing process[19]. One of the study conducted by Lee et al[20] in Maryland malnourished nursing home between the subjects of pressure ulcer showed that high protein diet improves the healing of pressure ulcers in malnourished nursing home subjects. Randomized control trial showed the benefit of protein hydrolysate supplement in the residents who have pressure ulcers. Another study by Dickhaut et al[21] observed the importance of protein in wound healing after amputation. Subjects consuming inappropriate diet will increase time of wound healing so, nutritional states of subjects with diabetic foot ulcer have a significant role in recovery of wound. A study also reported that every third diabetic foot ulcer subjects were malnourished.

At the end of this study, satisfactory healing was found among 23.33% and 66.67% of the subjects in group A and B respectively. Severe wound infection was reported only in group A (among 10% of subjects). When group A and B were compared statistically according to Asepsis Wound Scoring System, it was found to be statistically significant at all the intervals as $p<0.05$. Maris S. Jones et al[12] in their study revealed that the amino acid supplement used in their study has a benefit in promoting the healing of diabetic foot ulcers. In a retrospective review of 11 diabetic patients with foot ulcers who were given supplementation with arginine, glutamine, and HMB, degeneration was not identified in any wounds assessed with standardized scoring systems for wound depth and wound appearance, but on the contrary, statistically significant improvements in scores for wound depth and wound appearance were identified. This implies a decrease in diabetic wound catabolism leading to diminished wound degeneration[22].

The limitation of the present study is its small sample size and short follow up. So the author recommends further longitudinal studies with maximum sample size.

Conclusion: In the clinical setting, healing failure is common in diabeticpatients and leads to chronic pain, infections, gangrene andamputation. Also, diabetic foot ulceration is the most common problem among diabetic subjects. Malnutrition is involved in delayed wound healing among this population. In our study, we found that among subjects with diabetic foot ulcers,amino acid administration leads to better healing and satisfaction among the patients. Therefore,amino acids supplement should be given to diabetic foot ulcer subjects.

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