Hyaluronic and Synthetic Aminoacid Treatment of the post extraction tooth socket healing in subjects with diabetes mellitus type 2

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Introduction. Patients with Type 2 Diabetes Mellitus (TDM2), following dental extraction, experience more complications and a longer period of healing of the post-extraction socket.
One possible aid to promote tissue healing is Hyaluronic acid (HA). This study uses two versions of a medical device in a gel formulation composed of HA and amino acids: Aminogam 4 Gel (AG4: Glycine, L-proline, L-Leucine, L-Lysine) and Aminogam 6 Gel (AG6:Glycine, L-Proline, L-Leucine, L-Lysine, L-Valine, L-Alanine).

Purpose of the study. To evaluate the clinical and biological effects of HA therapy in the healing of post-extraction sockets in patients with TDM2. Primary end point is to evaluate the rate of healing of the post-extraction alveolus, while the secondary is to evaluate the quality of healing. A comparison between the efficacy of AG4 versus AG6 has been performed. The null hypothesis was that HA can significantly improve the post extractive healing of diabetic patients compared with no treatment.

Materials and methods. The study was designed as a single center randomized controlled trial. Patients requiring extraction of not impacted teeth were visited at the C.I.R. (Interdepartmental Research Center) of Dental School, Section of Oral surgery, Department of Surgical Sciences, University of Turin from September 2022 to July 2023. Patients were randomly assigned through a computer-generated random sequence of numbers to the test for one of the 4 groups:
T4 group (treated with AG4) included: post-operative application of AG4 3 times per day (8 hours distance between each application) for 7 days after oral hygiene and without swallowing, eating or drinking for one hour after the application, as follows: “wash your hands thoroughly before each application, apply a layer of gel on the injured mucosa, massage with a finger in order to facilitate spreading of the product over the treated area and compressing the product with gauze”.
C4 group (untreated) included: no treatment.
T6 group (treated with AG6) included: post-operative application of AG6 3 times per day (8 hours distance between each application) for 7 days after oral hygiene and without swallowing, eating or drinking for one hour after the application, as follows: “wash your hands thoroughly before each application, apply a layer of gel on the injured mucosa, massage with a finger in order to facilitate spreading of the product over the treated area and compressing the product with gauze”.
C6 group (untreated) included: no treatment.
All the surgeries were performed by the same experienced clinicians specialized in oral surgery who were blinded to the group allocation of the sites. All the pre- and post-operative assessments were performed by two calibrated and trained operators who were blinded to the T and C group allocation.
Inclusion criteria: age≥18 years, TDM2 for at least one year, adequate cognitive level, at least one systemic complication from TDM2, voluntary participation, need for dental extraction, informed consent, and willingness to complete the study. Randomization was random. After nonsurgical dental extraction, maximum mesio-distal (MD), vestibulo-oral (VO) socket

diameters and maximum probing depth (P) were measured. Follow-ups include 3-, 7-, 14-, and 21-day follow-ups where alveolus and healing index are measured using a version of Masse's HI.
At day 3, cytokines were analyzed by performing a healing tissue sampling and using the MACSPlex Cytokine 12 kit (Miltenyi Biotec), which quantitatively assesses 12 human cytokines: GM-CSF, IFN-α, IFN-γ, IL-2, IL-4, IL-5, IL-6, IL-9, IL-10, IL-12p70, IL -17A and TNF-α.

Results In total, 112 patients (n = 112), with a mean age of 69.44 ± 10.08 years, met the inclusion criteria and were enrolled in the study.
Based on the results, no statistically significant differences ($p > 0.05$) were highlighted between the T4 and C4 groups for any of the considered baseline variables. Therefore, it is possible to conclude that both the group T4 and C4 were similar, indicating an unbiased randomization and absence of covariates. Pre-operative data showed differences between T6 and C6 for: age, weight, BMI, PSR, high systemic risk; there was no difference for: sample size height. Analyzing the sex between groups, there are no statistically significant differences as shown in the table. The evaluation of BMI showed a higher value in the C6 group (mean 33.11 ± 9.12), with ANOVA Between Groups p-value = 0.019 (table 12).
T4 group showed significantly ($p < 0.05$) better Healing index values at D7 ($p = 0.01$) and D14 ($p = 0.02$), while no statistically significant difference was highlighted at D3 ($p = 0.08$) and D21 ($p =1$). In regard to the % of sockets that presented with optimal healing (healing index = 4), a statistically significant difference ($p < 0.05$) was highlighted at D14 ($p = 0.004$), with sockets treated with HA showing the better result. Values recorded in regard to OV diameters showed a statistically significant difference ($p = 0.03$) in favor of the T4 group at D3, while no statistically significant difference ($p > 0.05$) was recorded between the 2 groups at the other follow-up times. Regarding the MD diameters, a statistically significant difference was highlighted at D3 ($p = 0.03$) in favor of sites treated with HA (T group), while no statistically significant difference ($p > 0.05$) was recorded between the 2 groups at the other follow-up times. In regard to SD, a statistically significant difference was recorded at D14 ($p = 0.04$) in favor of the T group, while no statistically significant difference ($p > 0.05$) was recorded between the 2 groups at the other follow-up times. Sockets treated with HA (T4 vs C4 group), showed significantly ($p < 0.05$) better socket closure value at D 3 ($p = 0.04$), D7 ($p = 0.001$), and D14 ($p = 0.001$) compared to the C group.
In the postoperative evaluation between T6 and C6 groups. Statistically significant improved healing was observed at D7 ($p<0.001$) and D14 ($p<0.001$) in favor of the T6 group. In fact the mean healing index value of the T6 group was 4.0±0.2 versus 6.0±1.4 for the C6 group . At follow-ups at D14, healing is always in favor of the T6 group (4.0±0.1) compared with the C6 group (5.0±0.9). Sockets treated with AG6 (T6 group) showed significantly ($p < 0.05$) be[er Healing index values at D7 ($p = 0.01$) and D14 ($p = 0.02$), while no significant difference was highlighted at D3 ($p = 0.08$) and D21 ($p =1$).
Sockets treated with AG6 had optimal healing (healing index = 4) at day 7, C4 and C6 at day 21. To analyze the difference between T4 and T6 groups it has been conducted a Univariate Analysis (ANOVA) - (T6 vs T4) analysis of the median compared with the mean.
Analysis of the 12 cytokines showed a statistically significant difference for GM-CSF, IL-10 and TNF-α.

Discussion The aim of the present split-mouth randomized control trial was to investigate whether the employment of HA gel can provide benefits in the post-extraction tooth socket healing in subjects with DM type 2. This type of systemic patients often suffers from delayed wound healing and unfavorable changes in the tridimensional remodeling of the socket.
In both comparison of T4 vs C4 and T6 vs C6 a statistically better result was highlighted for the HA group at D7 and D14, while no statistically significant difference was found at D3 and D21. 7 and 14 days i.e., the days when healing in DM2 patients is slowest.
A higher amount of GM-CSF, IL-10 and TNF-α in the T6 group at 3 days might be a positive finding since these cytokines stimulate acute phase reaction, macrophage recruitment and tissue homeostasis
Conclusions HA has been shown to be coadjuvant in the healing of post-extraction sockets of DM2 patients. Therefore, its use can be considered to reduce the use of additional drugs (antibiotics, anti-inflammatories) in patients with severe systemic disease, polyorganic systemic complications and on polypharmacological therapy.