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Staphylococcus D vaccine kit, used in the treatment of periodontal disease, produced by the National Institute of Development In Microbiology and Immunology Cantacuzino - Romania

A new method of periodontitis treatment by modulating the immune response with SVD associated with polyvaccine (Polidin) and antibiotics together with conventional treatment.

Language: English

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Prof. Marian Vladimir Constantinescu, DDS, PhD, Assist. Prof. Mihaela Răescu, DDS, PhD,
Titu Maiorescu University, Faculty of Dentistry, Bucharest, Romania

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15th of the Balcan Stomatological Society Thesaloniki
Thesaloniki, Greece

Introduction

Staph vaccine was prepared by dr.Pasteur in 1892 and used for treating staphylococcus infection, but gradually abandoned since the appearance of the antibiotics. Since 1985 hypothesis of staphylococcus involvement in periodontal disease and its right treatment by staph vaccine, have been checked out.
Staphylococcus belongs to oral cavity flora, as saprophyte and opportunistic - pathogenic determined, even ubiquitous ancient bacteria that parasites all living organisms passing from human to animal and vice versa, with great adapting capacity and becoming immune to antibiotics by mutations (aerobic and discretionary anaerobic). Currently there is a pandemic of methicilline resistant staphylococcus.
Piogen infections with Staphylococcus appears most commonly in the medical practice (70-80% out of suppuration) and it is the agent in more than 90% of osteitis and osteomyelitis cases. Staphylococcus is high calcium consumer.
Bacteriological tests conducted by Cantacuzino Institute revealed a considerable proportion (45%) of staphylococcus in periodontal pockets, on a 200 subjects sample and 800 bacteriological identifications by selective breeding ground (Chapman). Administrating a new scheme of Staph vaccine has reduced periodontal inflammation.

Objectives

Modulating the Immune response with staphylococcal vaccine (SVD) in the periodontal disease. A new method of periodontitis treatment by modulating the immune response with SVD associated with polyvaccine (Polidin) and antibiotics together with conventional treatment staph vaccine produced by Romanian Cantacuzino Institute in 1927. First generation vaccine was originally used only for treating chronic recurrent staphylococcal infections. Since 1997, Cantacuzino Institute introduced staph vaccine in D classification.

Fig. 1

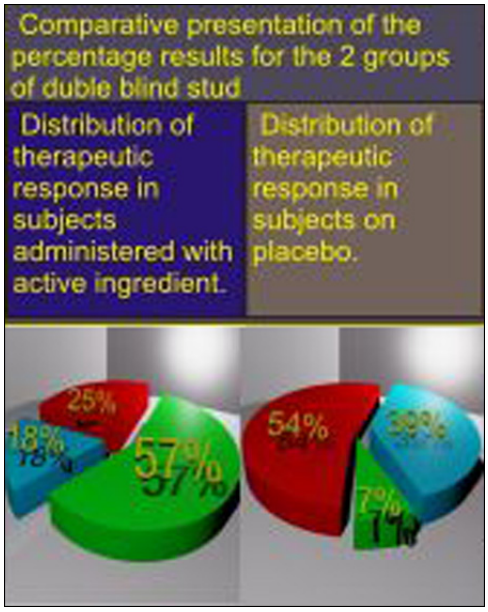


Fig. 2

Material and Methods

SVD kit consists of 4 staph vaccine boxes in different dilutions of one billion 1/10, 1/100, 1/1000, bacterial/ml with therapeutic indication for immunomodulater in periodontal disease.
Active ingredient - corpuscular suspension of fixed and inactivated bacteria in saline solution from a balanced 15 strains mix of Staphylococcus aureus as commonly circulated bacterial types.
Preparation procedures for staph vaccine are up to date according to recommendations of European Pharmacopoeia Guidelines and the European Agency for the Evaluation of Medicinal Products EMEA - Property Committees for Medicinal Products (CPMP) and also authorised by the National Medicine Agency. Control procedures for the reception of raw materials and materials control procedures in process, finished product inspection procedures.
Method of treatment - OSIM Romanian patent no. CL 111,022 / 1996 T. Georgescu-- Revendication Preparation of SVD in one billion decimal dilutions, 1/10, 1/100, 1/1000 corpora bacteria/ml - Field treatment - periodontal disease therapy. Changing management vaccine scheme as follows: - administrating 15 to 18 hypodermic inoculations, first 9 SVD daily inoculations of dilution, next 6 vaccine concentrate (1md/ml) inoculations hypodermic every 2 -3 days with one Polidin ampoule (Romanian polivaccine) and antibiotics (ampicilline + metronydazole) for VSD management scheme.

Results

VSD in periodontal therapy has been demonstrated by:

V.1. Longitudinal clinical study on 1000 subjects: improvement and cure of 70-90% on any periodontitis form as a result of joint action of immunotherapy, antibiotics and hygiene. In a significant proportion relapse occurred after 6 months and therefore 6 months, 1 year and 2 years-revaccination are recommended.

V.2. Double-blind clinical study on 150 subjects administered SVD without other antibiotics and hygiene measures - see charts V2.

Lot test- evolution: Lot martor - evolution: (vaccin) - 57% -favorable (placebo) -7% -favorable -18% -stationare -54% -stationare -25% -agravation -39% -agravation

V.3. Identify the total flora of periodontal pockets, aerobic and anaerobic, on 15 subjects and total 120 matches - see table V.3. The presence of golden staphylococcus in periodontal pockets before immunotherapy with SVD and its disappearance after treatment but remained non-pathogenic Staphylococcus in some subjects. (Tabel V.3)

V.4. Biochemical analysis - the use of enzymatic markers in evaluating the inflammatory process evolution after treatment with SVD: HDL, LDL, PAF-AH, C-reactive protein ROS and inflammation-reducing confirmed - (VIASAN research program no.333/2004).

V.5. Clinical cases (case 1, 2, 3, 4, 5).

V.6. Morpho-pathological exam applied on 30 subjects with biopsy before and after immunotherapy: see results - case 6, 7, 8, 9.

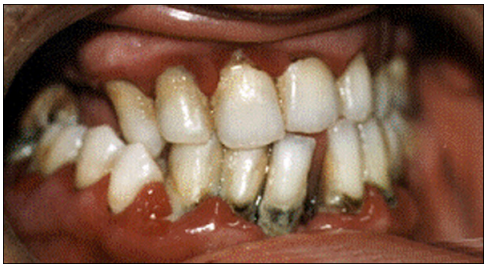


Fig. 3: Results 1 before

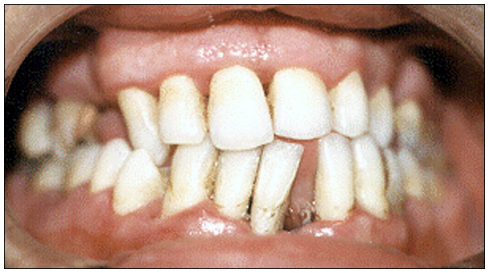


Fig. 4: Results 1 after

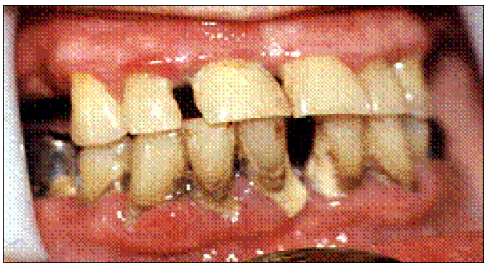


Fig. 5: Results 2 before

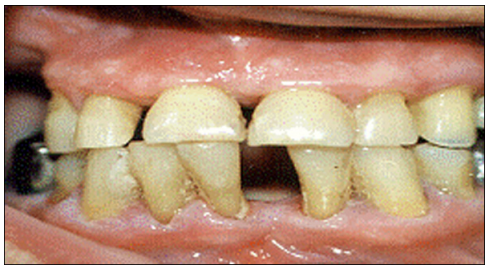


Fig. 6: Results 2 after

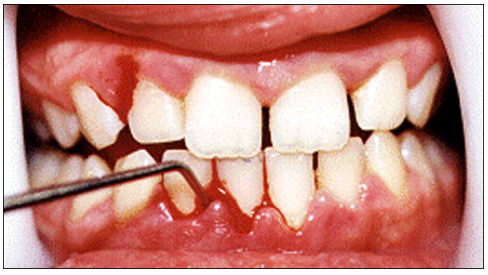


Fig. 7: Results 3 before

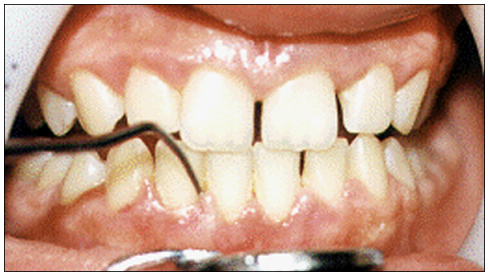


Fig. 8: Results 3 after

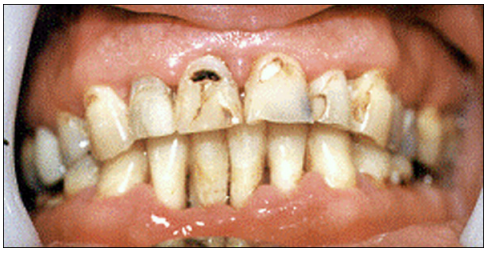


Fig. 9: Results 4 before

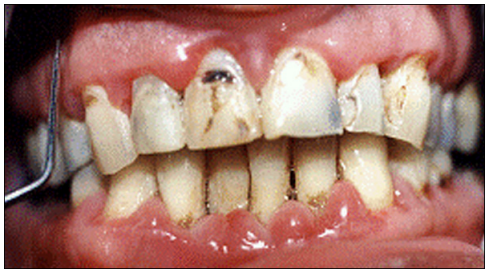
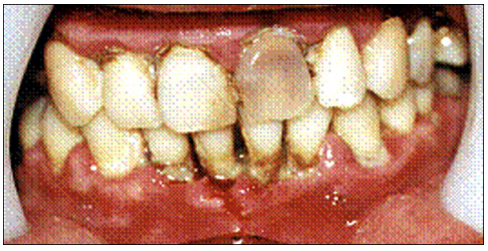


Fig. 10: Results 4 after



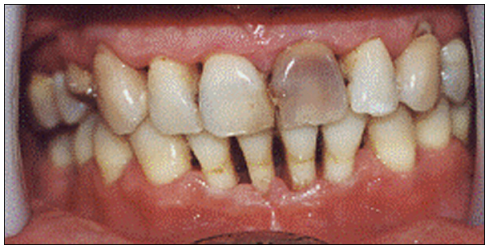


Fig. 11: Results 7 before Fig. 12:

V.5. Signs of clinical efficacy assessment of immunotherapy with SVD

BEFORE TREATMENT

- purple color, glossy gum
- massive edema of gum
- periodontal pockets
- spontaneous or induced gum bleeding
- pockets suppuration
- soft, friable gum
- fetid halena
- occlusal instability
- excessive concern for periodontal suffering

AFTER TREATMENT

- pink, healthy gum
- reduction of inflammatory edema
- reduced depth of periodontal pockets
- reducing spontaneous or induced gums bleeding, up to disappearance
- reducing suppurations up to disappearance
- scaring marginal epithelium with gum, modified consistency, firm and normal texture gum
- feeling of occlusal stability
- reduced mobility
- no more halena

1.
Before: Dg C.C 32 years. - Periodontitis - purple coloration and clinical signs of gum hypertrophy and bleeding, plaque, tartar, bags up to 8 mm, up to grade 3 mobility, suppuration blocking the reverse gear 13 to 44, 11 - ulceration of gingival margins.
After: SVD immunotherapy and local treatment - reducing inflammation, no more gum bleeding or suppurations, normal color, texture and consistency of gum, reduced pockets mobility and size, regeneration of the gingival margins.

2.
Before: G.L Dg 79 years. Periodontitis - clinical signs - massive gingival swelling, bleeding, suppuration, plaque and tartar, periodontal pockets up to 10mm, tooth mobility to grade 3, horizontally and vertically dental migrations, occluzo-articular imbalance by edentation and loss of mastication units.
After: immunotherapy with VSD - reducing inflammation, bleeding and suppurations, reduced size and no mobility of periodontal pockets, normal color, texture and consistency of gums.

3.
Before: Dg S.A.G 12 years - juvenile periodontitis: clinical signs and papillae hypertrophy, bleeding, reduced bacterial plaque, deep pockets 7 -8 mm.
After: immunotherapy with VSD and local treatment - reduced or no more inflammation, normal texture of gum. Reduced deep of pockets.

4.
Before: Dg M.M 35 years. Periodontiti - clinical signs: purple coloration and hypertrophy of gingival papillae, plaque, tartar, pockets up to 10 mm, mobility up to grade 3.
After: immunotherapy with VSD and local treatment - reducing inflammation, no more bleeding or suppurations, cvasinormal texture and color of gum. Reduced pockets depth.

5.
Before: HS - 32 years Dg. ulcero necrotic periodontitis, clinical signs - gum ulcerations, inflammation, bleeding, plaque, tartar, pockets up to 8- 10 mm, mobility up to grade 3 , gingival retraction.
After: SVD immunotherapy and local treatment - reducing inflammation, healing ulcerations, no more bleeding or suppurations. Reduced pockets depth and mobility

Nr. Crt.	BEFORE TREATMENT	AFTER TREATMENT
1	Porphyromonas asaccharolytica Peptostreptococcus anaerobius	Staphylococcus epidermidis
2	Bacteroides fragilis	*No bacteria developed*
3	Staphylococcus warneri	Staphylococcus warneri
4	Staphylococcus anreus	*No bacteria developed*
5	Fusobacterium nucleatum Staphylococcus epidermidis	Fusobacterium nucleatum
6	*No bacteria developed*	*No bacteria developed*
7	Staphylococcus chromogenes	*No bacteria developed*
8	Staphylococcus epidermidis Veillonella parvula	– Veillonella parvula
9	Staphylococcus epidermidis Bacteroides fragilis	Staphylococcus epidermidis Bacteroides fragilis
10	Staphylococcus capitis	*No bacteria developed*

11	Bifidobacterium sp. Staphylococcus anreus	Bifidobacterium sp.
12	Bifidobacterium sp. Streptococcus viridans	Bifidobacterium sp. Streptococcus viridans
13	Streptococcus viridans	Streptococcus viridans
14	Bifidobacterium sp. Staphylococcus epidermidis	Bifidobacterium sp. Staphylococcus epidermidis
15	Fusobacterium mortiferum Staphylococcus epidermidis	Fusobacterium mortiferum Streptococcus viridans

Tab. 1

V.6. Morpho-pathological exam applied on 30 subjects with biopsy before and after immunotherapy: see results (case 6, 7, 8, 9).

BEFORE TREATMENT
Gum:
• on its surface: necrotic cellular debris, microbial colonies, purulent exudate leukocyte.
• in the epithelium: different degrees of atrophy of epithelial cells, ulcerations.
• in the chorion: Chronic granulomatous inflammatory infiltrates with vessels of neoformations, colonies microbial and fungal.

AFTER TREATMENT
Gum:
• epithelium is regenerated, with benign cellular proliferation and para-keratosis and hiper-keratosis, mato papilo in chorion proliferation among groups of connective fibers papilomatoze, absence of microbial colonies.

In the chorion: numerous reticulin fiber bundles accompanying dense collagen fibers, decreased frequency of chronic inflammatory infiltration, no more granulomatous tissue, no more microbial or fungal colonies.

6.
Before treatment: neoformation vessels massive leukocyte infiltration, granulation tissue in the chorion, Col. HE x 100
After treatment: many papilla profiling, digitiforme with epithelial integrity; normal chorion joint-vascular axis with a few lymphocytes, which usually occur in a normal chorion. Basal cell proliferation and hyperkeratosis-Col. HE x 200

7.
Before treatment: - gingival epithelium atrophic with ulceration, and detritus necrotic. In the chorion granulation tissue with inflammatory infiltrates and new vessels .Col. HE x 64
After treatment: the epithelium cells displaying normal spinous layer with dezmozomi (intercellular bridges) in the surface layer is observed paraketoza process. Epithelial cell regeneration and chorion - Col. HE x 400

8.
Before treatment: - Microbial colony before and micelles in deeper layers, cellular alterations chorion, Col. HE x 64
After treatment: detail, complete restoration of spindle collagen interdental papilla. Col Gomory x 200

9.
Before treatment: - Extract of gum, stain Gomory- depolymerization massive network of collagen
After treatment: complete restoration of connective tissue with collagen proliferation explains the consistency of the gum farm after treatment, network of collagen. Col Gomory x 100

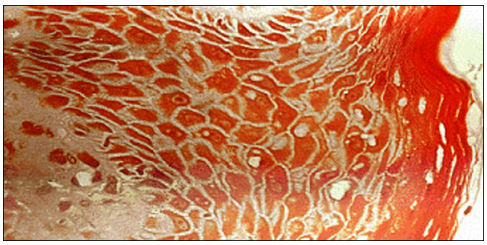


Fig. 13: Results 5 after

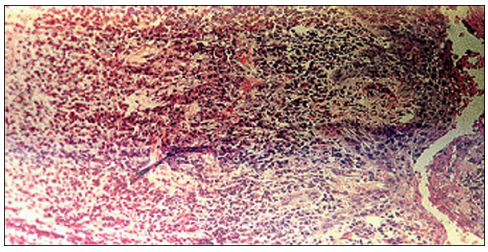


Fig. 14: Results 5 before

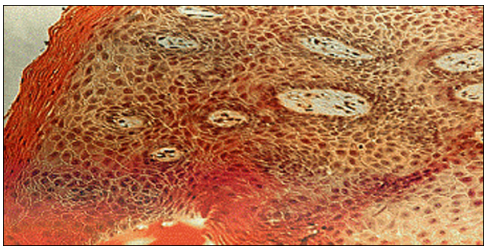


Fig. 15: Results 6 after

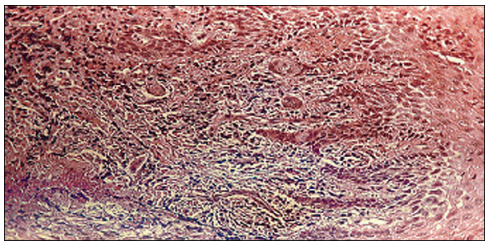
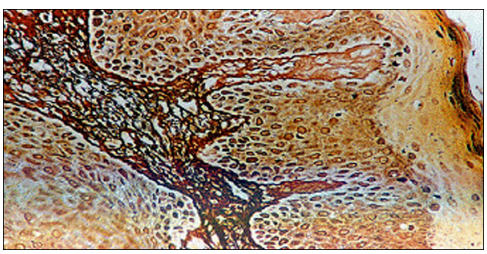


Fig. 16: Results 6 before



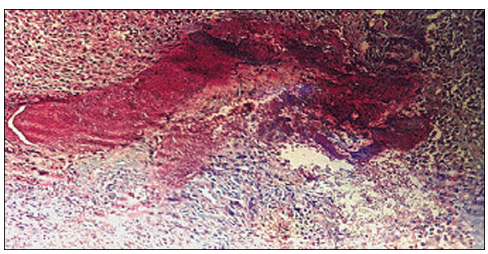


Fig. 17: Results 8 after

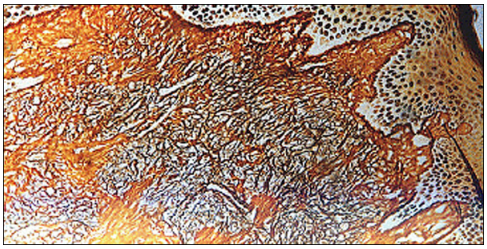


Fig. 18: Results 8 before

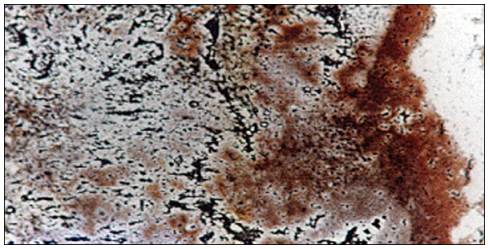


Fig. 19: Results 9 after

Fig. 20: Results 9 before

Conclusions

Because acquired immunity is reduced in 6 months after first immunomodulator cycle, reimmunization is required in 6 months, 1 year and 2 years at the end of each cycle, consisting of 9-10 inoculations of successive dilutions of ½ and 1/1 vial. If relapses occur, 3 to 4 vials of successive dilutions are to be inoculated. SVD immunomodulating therapy can be applied together with conventional periodontal treatments as hygiene, cleaning scaling, surgery, occlusion therapy etc.

1. Staphylococcus is present in a significant proportion (45%) of periodontal pockets of purulent exudation.
2. Immunotherapy with staph vaccine, administered by proposed inoculation scheme is effective to reduce inflammation and the symptoms of periodontal disease, curing and regenerating periodontal structures by epithelial hyperplasia and metaplasia of granulation tissue into fibrous tissue.
3. Immunomodulating effect of the proposed method depends on general reactivity of patient and periodontal disease needs to be treated together with improvement of its general health status (holistic approach).
4. Before treatment staph vaccine should be immune to a thorough consultation carried out by GP or specialist and correct treatment of identified pathology.
5. Staph vaccine immunotherapy is effective, operative, low-cost, well tolerated by the patient, successfully completing the usual therapeutic procedures.

Literature

1. Gafar M, Georgescu T, Dumitriu H, Codiţă I, Neguţ M.: The role of the staphylococcal infectious-allergic process in chronic marginal periodontal diseases, and specific therapeutic possibilities with staphylococcal vaccine. Preliminary note. Rev Chir Oncol Radiol O R L Oftalmol Stomatol Ser Stomatol. 1987 Jan-Mar;34(1):1-10. PMID: 2977820 [PubMed - indexed for MEDLINE]
2. Georgescu T.: Treatment method of chronic marginal periodontal disease. - OSIM Romanian patent no. CL 111,022 / 1996
3. Orlando S.: Polyvalent heterovaccine injectable in inflammatory periodontopathies: clinical and therapeutic contribution with microbiologic and immunologic studies. Riv Ital Stomatol. 1967 Jan;22(1):31-43. PMID: 5336487 [PubMed - indexed for MEDLINE]

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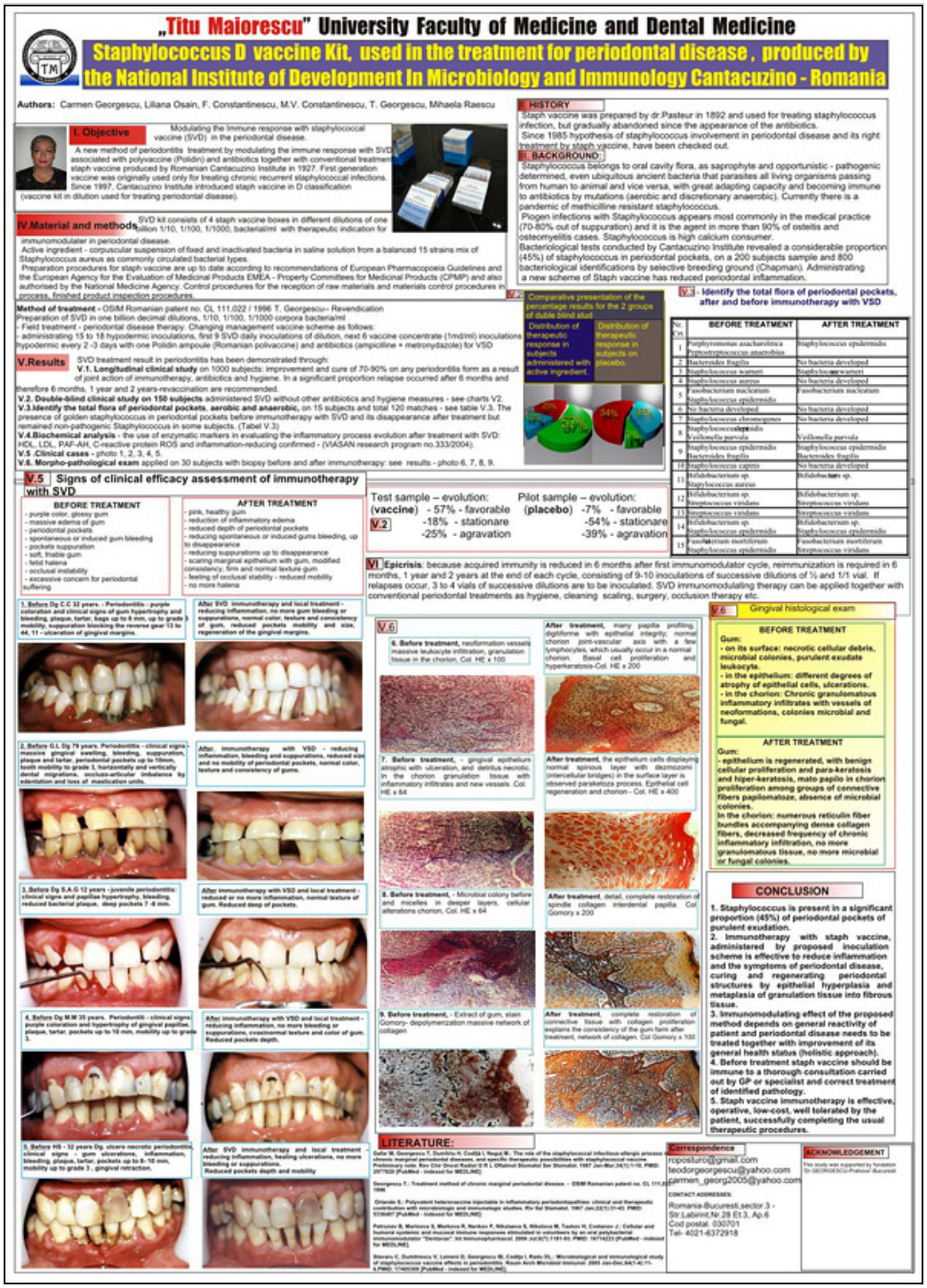
Poster Faksimile:



„Titu Maiorescu" University Faculty of Medicine and Dental Medicine

Staphylococcus D vaccine Kit, used in the treatment for periodontal disease , produced by the National Institute of Development In Microbiology and Immunology Cantacuzino - Romania

Authors: Carmen Georgescu, Liliana Osain, F. Constantinescu, M.V. Constantinescu, T. Georgescu, Mihaela Raescu

I. Objective

Modulating the Immune response with staphylococcal vaccine (SVD) in the periodontal disease.

A new method of periodontitis treatment by modulating the immune response with SVD associated with polyvaccine (Polidin) and antibiotics together with conventional treatment. staph vaccine produced by Romanian Cantacuzino Institute in 1927. First generation vaccine was originally used only for treating chronic recurrent staphylococcal infections. Since 1997, Cantacuzino Institute introduced staph vaccine in D classification (vaccine kit in dilution used for treating periodontal disease).

II. HISTORY

Staph vaccine was prepared by dr Pasteur in 1892 and used for treating staphylococcus infection, but gradually abandoned since the appearance of the antibiotics.
Since 1985 hypothesis of staphylococcus involvement in periodontal disease and its right treatment by staph vaccine, have been checked out.

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Active ingredient - corpuscular suspension of fixed and inactivated bacteria in saline solution from a balanced 15 strains mix of Staphylococcus aureus as commonly circulated bacterial types.
Preparation procedures for staph vaccine are up to date according to recommendations of European Pharmacopoeia Guidelines and the European Agency for the Evaluation of Medicinal Products EMEA - Property Committees for Medicinal Products (CPMP) and also authorised by the National Medicine Agency. Control procedures for the reception of raw materials and materials control procedures in process, finished product inspection procedures.

Method of treatment - OSIM Romanian patent no. CL 111,022 / 1996 T. Georgescu– Revendication
Preparation of SVD in one billion decimal dilutions, 1/10, 1/100, 1/1000 corpora bacteria/ml
- Field treatment - periodontal disease therapy. Changing management vaccine scheme as follows:
- administrating 15 to 18 hypodermic inoculations, first 9 SVD daily inoculations of dilution, next 6 vaccine concentrate (1ml/ml) inoculations hypodermic every 2 -3 days with one Polidin ampoule (Romanian polivaccine) and antibiotics (ampicilline + metronydazole) for VSD

V.Results

SVD treatment result in periodontitis has been demonstrated through:
V.1. Longitudinal clinical study on 1000 subjects: improvement and cure of 70-90% on any periodontitis form as a result of joint action of immunotherapy, antibiotics and hygiene. In a significant proportion relapse occurred after 6 months and therefore 6 months, 1 year and 2 years-revaccination are recommended.
V.2. Double-blind clinical study on 150 subjects administered SVD without other antibiotics and hygiene measures - see charts V2.
V.3.Identify the total flora of periodontal pockets, **aerobic and anaerobic,** on 15 subjects and total 120 matches - see table V.3. The presence of golden staphylococcus in periodontal pockets before immunotherapy with SVD and its disappearance after treatment but remained non-pathogenic Staphylococcus in some subjects. (Tabel V.3)
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V.5 .Clinical cases - photo 1, 2, 3, 4, 5.
V.6. Morpho-pathological exam applied on 30 subjects with biopsy before and after immunotherapy: see results - photo 6, 7, 8, 9.

V.2 Comparative presentation of the percentage results for the 2 groups of duble blind stud

V.3 - Identify the total flora of periodontal pockets, after and before immunotherapy with VSD

Nr. Crt.	BEFORE TREATMENT	AFTER TREATMENT
1	Porphyromonas asaccharolitica Peptostreptococcus anaerobius	staphylococcus epidermidis
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12	Bifidobacterium sp. Streptococcus viridans	Bifidobacterium sp. Streptococcus viridans
13	Streptococcus viridans	Streptococcus viridans
14	Bifidobacterium sp. Staphylococcus epidermidis	Bifidobacterium sp. Staphylococcus epidermidis
15	Fusobacterium morbiferum Staphylococcus epidermidis	Fusobacterium morbiferum Streptococcus viridans

V.5 Signs of clinical efficacy assessment of immunotherapy with SVD

BEFORE TREATMENT	AFTER TREATMENT
- purple color, glossy gum - massive edema of gum - periodontal pockets - spontaneous or induced gum bleeding - pockets suppuration - soft, friable gum - fetid halena - occlusal instability - excessive concern for periodontal suffering	- pink, healthy gum - reduction of inflammatory edema - reduced depth of periodontal pockets - reducing spontaneous or induced gums bleeding, up to disappearance - reducing suppurations up to disappearance - scaring marginal epithelium with gum, modified consistency, firm and normal texture gum - feeling of occlusal stability - reduced mobility - no more halena

Test sample – evolution:
(**vaccine**) - 57% - favorable
-18% - stationare
-25% - agravation

Pilot sample – evolution:
(**placebo**) -7% - favorable
-54% - stationare
-39% - agravation

V.2

V.1 Epicrisis: because acquired immunity is reduced in 6 months after first immunomodulator cycle, reimmunization is required in 6 months, 1 year and 2 years at the end of each cycle, consisting of 9-10 inoculations of successive dilutions of ½ and 1/1 vial. If relapses occur, 3 to 4 vials of successive dilutions are to be inoculated. SVD immunomodulating therapy can be applied together with conventional periodontal treatments as hygiene, cleaning scaling, surgery, occlusion therapy etc.

1. Before Dg C.C 32 years. - Periodontitis - purple coloration and clinical signs of gum hyperthrophy and bleeding, plaque, tartar, bags up to 6 mm, up to grade 3 mobility, suppuration blocking the reverse gear 12 to 44, 11 - ulceration of gingival margins.

After SVD immunotherapy and local treatment - reducing inflammation, no more gum bleeding or suppurations, normal color, texture and consistency of gum, reduced pockets mobility and size, regeneration of the gingival margins.

2. Before G.L Dg 79 years. Periodontitis - clinical signs - massive gingival swelling, bleeding, suppuration, plaque and tartar, periodontal pockets up to 10mm, tooth mobility to grade 3, horizontally and vertically dental migrations, occluso-articular imbalance by edentation and loss of mastication units.

After immunotherapy with VSD - reducing inflammation, bleeding and suppurations, reduced size and no mobility of periodontal pockets, normal color, texture and consistency of gums.

3. Before Dg S.A.G 12 years - juvenile periodontitis: clinical signs and papillae hypertrophy, bleeding, reduced bacterial plaque, deep pockets 7 -8 mm.

After immunotherapy with VSD and local treatment - reduced or no more inflammation, normal texture of gum. Reduced deep of pockets.

4. Before Dg M.M 35 years. Periodontiti - clinical signs: purple coloration and hypertrophy of gingival papillae, plaque, tartar, pockets up to 10 mm, mobility up to grade 3.

After immunotherapy with VSD and local treatment - reducing inflammation, no more bleeding or suppurations, cvasinormal texture and color of gum. Reduced pockets depth.

5. Before HS - 32 years Dg. ulcero necrotic periodontitis, clinical signs - gum ulcerations, inflammation, bleeding, plaque, tartar, pockets up to 8- 10 mm, mobility up to grade 3 , gingival retraction.

After SVD immunotherapy and local treatment - reducing inflammation, healing ulcerations, no more bleeding or suppurations. Reduced pockets depth and mobility

V.6

6. Before treatment, neoformation vessels massive leukocyte infiltration, granulation tissue in the chorion, Col. HE x 100.

After treatment, many papilla profiling, digitiforme with epithelial integrity; normal chorion joint-vascular axis with a few lymphocytes, which usually occur in a normal chorion. Basal cell proliferation and hyperkeratosis-Col. HE x 200

7. Before treatment, - gingival epithelium atrophic with ulceration, and detritus necrotic. In the chorion granulation tissue with inflammatory infiltrates and new vessels .Col. HE x 64

After treatment, the epithelium cells displaying normal spinous layer with dezmozomi (intercellular bridges) in the surface layer is observed parakeratoza process. Epithelial cell regeneration and chorion - Col. HE x 400

8. Before treatment, - Microbial colony before and micelles in deeper layers, cellular alterations chorion, Col. HE x 64

After treatment, detail, complete restoration of spindle collagen interdental papilla. Col Gomory x 200

9. Before treatment, - Extract of gum, stain Gomory- depolymerization massive network of collagen

After treatment, complete restoration of connective tissue with collagen proliferation explains the consistency of the gum farm after treatment, network of collagen. Col Gomory x 100

V.6 Gingival histological exam

BEFORE TREATMENT

Gum:
- on its surface: necrotic cellular debris, microbial colonies, purulent exudate leukocyte.
- in the epithelium: different degrees of atrophy of epithelial cells, ulcerations.
- in the chorion: Chronic granulomatous inflammatory infiltrates with vessels of neoformations, colonies microbial and fungal.

AFTER TREATMENT

Gum:
- epithelium is regenerated, with benign cellular proliferation and para-keratosis and hiper-keratosis, mato papilo in chorion proliferation among groups of connective fibers papillomatoze, absence of microbial colonies.
In the chorion: numerous reticulin fiber bundles accompanying dense collagen fibers, decreased frequency of chronic inflammatory infiltration, no more granulomatous tissue, no more microbial or fungal colonies.

CONCLUSION

1. Staphylococcus is present in a significant proportion (45%) of periodontal pockets of purulent exudation.
2. Immunotherapy with staph vaccine, administered by proposed inoculation scheme is effective to reduce inflammation and the symptoms of periodontal disease, curing and regenerating periodontal structures by epithelial hyperplasia and metaplasia of granulation tissue into fibrous tissue.
3. Immunomodulating effect of the proposed method depends on general reactivity of patient and periodontal disease needs to be treated together with improvement of its general health status (holistic approach).
4. Before treatment staph vaccine should be immune to a thorough consultation carried out by GP or specialist and correct treatment of identified pathology.
5. Staph vaccine immunotherapy is effective, operative, low-cost, well tolerated by the patient, successfully completing the usual therapeutic procedures.

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