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Research Article

Therapeutic Effects of Bioquantine™ in Volunteer Patients Diagnosed With T2D, CKD, SCI, Hypothyroidism, Atypical Pyoderma Gangrenous, and Breast Cancer

Sergei Paylian[1]*, Ira Pastor[1], Tatyana Reshetnikova[1], Himanshu Bansal[2] and Nick Skiba[3]

[1]Bioquark Inc., BNY Mellon Center, 1735 Market St, #3750, Philadelphia, PA 19103, USA

[2]Institute of Spinal Injury & Stem Cell Research, Anupam Hospital, Rudrapur, Uttarakhand -263153, India

[3]Duke Eye Center, 2351 Erwin Road, DUMC 3802, AERI Room 5006 Durham, NC 27710, USA

Abstract

We have previously demonstrated that co-electroporation of Xenopus laevis frog oocytes with normal cells and cancerous cell lines induces the expression of pluripotency markers, and in experimental murine model studies that Bioquantine extract (purified from intra- and extra-oocyte liquid phases of electroporated oocytes) showed potential as a treatment for a wide range of conditions including melanoma, traumatic brain injury (TBI) and skin wrinkling. The current study observed beneficial changes with Bioquantine administration in a small group of patients with a variety of degenerative disorders. A patient with chronic kidney disease (CKD) due to polycystic kidney disease showed volumetric improvement along with a reduction in the number of renal cysts and decreases in plasma creatinine. A patient with CKD related to hypertension and type 2 diabetes also demonstrated decreases in plasma creatinine. This patient, who had been dependent on dialysis at study entry, was able to go for longer periods between dialysis to the point where he did not require dialysis for several months. A patient with type 2 diabetes and inadequate glycemic control on standard oral hypoglycemic therapy showed normalization of fasting blood glucose. A patient with hypothyroidism achieved normalization of thyroid function. A patient with pydoderma gangrenosum demonstrated considerable reduction in number of sites affected by cutaneous ulcerations with mucopurulent or hemorrhagic exudates and reduction in recurrences. Three patients with spinal cord injuries had regeneration and reconstruction of the spinal cord along all entire length of lesion as visualized using magnetic resonance imaging and considerable improvement in sensation and walking ability and coordination of movement. A patient with metastatic breast cancer showed substantial decreases in cancer-related biomarkers and improvement in body weight and vital signs. These first-in-human results indicate that Bioquantine administration may be safe and well tolerated for use in humans, that it has potential therapeutic activity including restoration of renal function, glycemic control, thyroid function, dermatologic healing, spinal cord regeneration, and anti-breast cancer activity, and deserves further clinical exploration. We propose that the mechanism of action of Bioquantine in these various diseases derives from its unique combinatorial reprogramming properties.

Introduction

Pluripotent stem cells have therapeutic and regenerative potential in clinical situations including renal failure, type 2 diabetes, dermatology, CNS disorders, and cancer [1-6]. One method of reprogramming somatic cells into pluripotent stem cells is to expose them to extracts prepared from *Xenopus laevis* oocytes [7]. We showed previously that co-electroporation of *Xenopus laevis* frog oocytes; with normal cells and cancerous cells lines, induces expression of markers of pluripotency [8]. We also observed therapeutic effects of treatment with a purified extract (Bioquantine) of intra- and extra-oocyte liquid phases derived from electroporated *X. laevisoocytes*, on experimentally induced pathologies including murine models of melanoma, traumatic brain injury, and experimental skin wrinkling induced by squalene-monohydroperoxide (Paylian et al, 2016; in development).

Taking into consideration a uniform reprogramming action of Bioquantine in different types of human cells, as well as its multi-therapeutic effects revealed in animal models, it was important to demonstrate that Bioquantine is safe and well tolerated when used in human subjects and that it possessed its ubiquitous therapeutic activity when administered in different types of human diseases. In the present, first-in-human exploratory studies, we present initial treatment results of a sampling of patients diagnosed with the following ailments: chronic kidney disease, type 2 diabetes, hypothyroidism, pyoderma gangrenosum, spinal cord injury and metastatic breast cancer.

Methods

Ethical Conduct

Each proposal was reviewed and approved by an Institutional

***Corresponding author:** Sergei Paylian[1], Bioquark Inc., BNY Mellon Center, 1735 Market St, #3750, Philadelphia, PA 19103, USA, Tel: 813-815-2099; E-mail: biomemory@gmail.com

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Review Board of each principal investigators clinical facility to make sure that research participants were protected from harm during volunteer studies that involves administration of investigational agents. Each patient signed an Informed Consent Form and a fingerprint of each participant was attached to each page of document. For ethicalreasons, no control groups were used.

Preparation of the Combinatorial Bioquantine Extract

With the aim of improving the reprogramming/therapeutic potential of the *X. laevis* oocyte-derived Bioquantine extract, a combinatorial extract Bioquantine™ (Bioquantine) was prepared using electrically-, electromagnetically- and radio frequency-activated and extracted biomaterial using oocytes from *Xenopus laevis* frogs, and related somatic tissues. We used a proprietary technology which employs activation of biomaterial using Bio-Rad Gene Pulser II system for electroporation, Polard model 1020T modulator for Extremely High Frequency (EHF) stimulation, and LW -1541 generator for Extremely Low Frequency (ELF) irradiation. Activated extracts were isolated by homogenization in Robot Coupe R401B Food Processor for 7 min stationed in Arctic Air AST48R refrigerator at 4°C followed by dual centrifugation of homogenate first at 5,000g for 25 min at 4°C and then at 10, 000g for 45 min at 4°C usinga Beckman J2-21M induction drive centrifuge. After centrifugation the collected extract underwent multi-step micro-filtration and final sterilization and filtration in Durapore® sterilizing-grade 0.1 μm hydrophilic low protein binding filter (EDM Millipore cat. # CVGL01TP3, Billerica MA, USA). The sterilized extract was distributed using Disppensetten III bottle top dispenser (GMBH BR4700240, India), aseptically sealed into 10 ml sterile amber glass vials, and was stored at 3°C for future use in subcutaneous (SC), intra-muscular (IM), intrathecal (IT) and intra-venous (IV) administration.

Bioquantine Bio-Safety Testing

Bioquantine extracts were extensively tested for the presence of various pathogens, biologic toxins, allergens, heavy metals, and various pesticides and organic environmental contaminants. Gram-positive and gram-negative bacteria, yeast and other fungi testing was conducted using Molecular Probes Cell Culture Contamination Detection Kit (C-7028); end toxins, using Gen Script Tox-in-Sensor™ broad range detection kits: L00447, L00448, L00449, L00450 and L00451. Allergens (Purified frog muscle parvalbumin) testing was performed using Abcam ab50338 detection kit. Mutagenic capacity was tested, using EBPI AMES test kit B5051 and hepatitis viruses A, B, C and E tests using One Step Rapid Test Kits by Qinghao High top Biotech Co., Ltd. Tetrodotoxin testing was conducted by Reagen LLC using enzyme-linked immunosorbent assay (ELISA), microbial assay performed by Genysis Labs UT, and heavy metals and pesticides/organic environmental contaminants screening was conducted at ALS Laboratories, UT. All results were negative.

Administration of Bioquantine

In the beginning of treatment, patients were given sublingually 25 drops of Bioquantine, administered each day for 3 days. On the 4th day, in the absence of adverse reactions, 0.2 m l-0.4 ml of Bioquantine solution was administered by the subcutaneous (SC) route, and the patient received close observation for possible allergic reactions or other side effects. Only patients who tolerated initial Bioquantine administration very well were chosen to continue treatment with further SC, intramuscular (IM), intrathecal (IT) or intravenous (IV) administration protocols. Patients with spinal cord injuries received a modified oral Bioquantine tolerance test prior to receiving the initial SC injections; these patients held a cotton ball soaked in Bioquantine in their mouths for 5-10 minutes. All participants received Bioquantine under the direct supervision of physicians. Routine blood work and imaging procedures were conducted in accordance with treatment plans.

Patients

Chronic Kidney Disease (CKD) – Not Undergoing Dialysis

The first patient in our study was a Russian adult man, aged 64 years, with polycystic kidney disease and high plasma creatinine levels (458 μmol/L), who refused to undergo renal dialysis against medical advice. This patient received 45 IM injections of 1 ml Bioquantine, at a concentration of 10 mg/ml, every other day starting on July 4, 2015 and continuing through September 22, 2015. Renal function was assessed by measuring creatinine, electrolyte and protein levels in blood and urine samples. The patient also underwent ultrasound imaging and volumetric measurements of both kidneys and the thickness of the renal parenchyma layers (Esaote MyLab 20 Plus ultrasound system). Laboratory testing (blood and urine) was conducted by Invitro Labs (Moscow, Russia). Medical synopsis of and patient monitoring was conducted by Healthy City Group LLC, Urology Department, Moscow, Russian Federation).

CKD –Receiving Dialysis

The second patient with CKDenrolled was an adult Indian man, aged 60 years, with a history of type 2 diabetes and hypertension, who had been undergoing dialysis once weekly for last 3 months before study entry. Prior to starting dialysis treatment, his creatinine levels ranged from 790 μmol/L to 970 μmol/L. He was treated with a course of Bioquantine 1 ml (at a concentration of 5 mg/mL) diluted in 100ml normal saline IV every fortnight and 1ml IM twice a week for at a concentration of 5mg/mlfor three months. Renal function was monitored with measurements of serum creatinine levels and duration between dialysis determined by medical opinion (Medical synopsis and patient monitoring by the Institute of Brain and Spine. Delhi, India).

Type 2 Diabetes Inadequately Controlled With Conventional Therapy

The patient was an adult man from the Republic of Georgia, aged 67 years, who has been diagnosed with diabetes mellitus type 2 in 2004. The patient was receiving met for min 1000mg twice daily and glimepiride4mg twice daily. These oral therapies did not adequately control the patient's blood glucose, with the patient's fasting blood glucose readings going from 222 and 235 mg/dL to 364 mg/dL. When the patient had a fasting blood glucose (FBG) reading of

373 mg/dL, he was advised to start subcutaneous insulin therapy. The patient rejected the use of insulin therapy. In this study, the patient received 1 ml peritoneal injections of Bioquantine, at 10 mg/ml, daily for 60 days. Glycosylated hemoglobin A1C (HbA_{1c}) levels were monitored regularly (Greek Scientific Foundation "Hippocrates", Tbilisi Georgia).

Hypothyroidism

This patient was an adult woman, in the Czech Republic, who had been diagnosed with hypothyroidism in 2012 and was taking Euthyrox90µg, once daily, when she entered the study. This patient received 1 ml IM injections of 5 mg/ml Bioquantine, every other day, for 40 days (total of 20 administrations). Serum levels of thyroid-stimulating hormone (TSH), triiodothyronine (T3), thyroxine (T4) levels, and medically indicated Euthyroxdose were monitored(Medical synopsis and patient monitoring conducted by BioReprogen s.r.o (LLC) , Lazne5, Karlovy Vary, Czech Republic).

Pyoderma Gangrenous

This man, aged 44 years, from the Russian Federation, had been diagnosed with a rare and resistant form of noninfectious neutrophilic dermatosis. The patient had been diagnosed in 2005, and at study entry had painful ulcers, of varyingdepth and size, with undermined violaceous borders. During and after treatment with Bioquantine (5mg/ml IM daily for 14 days) the patient was assessed for the recurrence of cutaneous ulcerations with purulent or hemorrhagic exudates, as well as measurement of diameter of erythema surrounding painful ulcers present with undermined bluish borders (Medical synopsis and monitoring conducted by BioReprogen s.r.o (LLC) , Lazne5, Karlovy Vary, Czech Republic) .

Spinal Cord Injury (SCI)

Three adult male patients from India ages 25, 34 and 55yearswith SCIs participated in the study who had sustained the injuries during motor vehicle accidents. Two of these patients had complete spinal cord transection, and the third patient had a nearly complete spinal cord transaction. All three patients entered the studies with American Spinal Injury Association (ASIA) classification of ASIA-A, which corresponds to no sensory and no motor functions preserved below the neurological level including sacral segments S4-S5. Clinical symptoms were expressed in the temporary or permanent loss of cord function below the injured segments with poor post-SCI prognosis. The patient aged 25 years received conventional therapy for 30 days before entering this study; the patient aged 34 years started this study 42 days after the injury, and the patient aged 55 years started this study treatment 27 days after the injury. Bioquantine was introduced as adjuvant therapy as follows: on day 0, the patients underwent the modified oral tolerance test; on day 1, each patient received the subcutaneous sensitivity test; on day 2, an IM injection was given; on day 3, the patients received Bioquantine intravenously (5 ml of 5 mg/ml Bioquantine in 100 ml of saline. Patients tolerated Bioquantine well, and on day 7 and onward, patients received biweekly IT injections of Bioquantine (1ml of 5 mg/mL) using a 27 gauge needles above and below S4-S5for the period of 2 months was conducted. In addition, each patient received a single intralesional injection of 1 ml (5mg/ml) of Bioquantine at the end of the cycle of intrathecal injections. Treatment progress in SCI patients was assessed by MRI imaging and clinical observation of motor- and sensory activity. (Medical synopsis and monitoring conducted by Institute of Spinal Cord Injury, Anupam hospital, Rudrapur, India).

Metastatic Breast Cancer

This adult woman from the Ukraine was first diagnosed with localized breast cancer in 2007 at age 63 and received treatment with everolimus. She remained cancer-free until July 2012, when she was found to have bone metastases and later, metastases in liver, spleen and brain. From July 22, 2013 till March 24, 2014, this patient receivedIM injection of 1 ml of 5 mg/ml of Bioquantine administered every other day. Treatment progress in this patient was assessed by blood levels of the cancer-associated antigen (CA 27.29), carcinoembryonic antigen (CEA) and alkaline phosphates (ALP). (Medical synopsis and monitoring by State Body I.I. Mechnikov Ukrainian Anti-Plague Research Institute, Ministry of Health of Ukraine, Odessa, Ukraine). This patient would not consent to publication of radiologic imaging data.

Results

CKD – Not on Dialysis

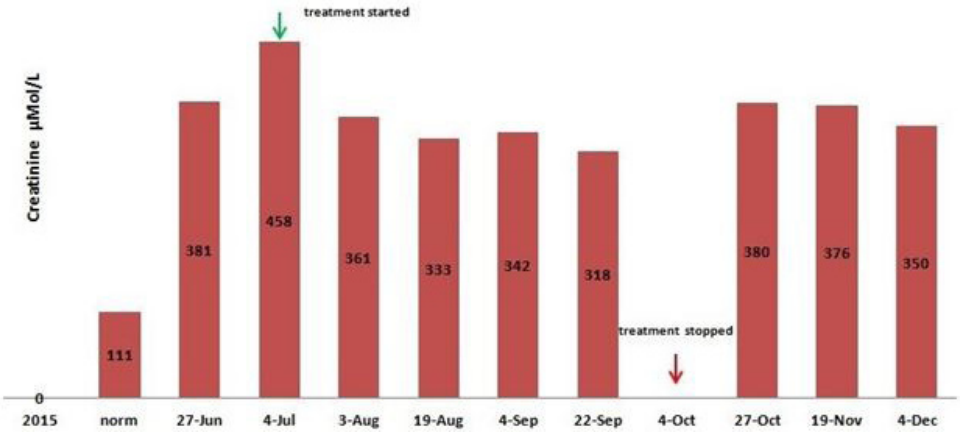


Figure 1: Drop in blood serum creatinine levels in polycystic chronic kidney disease patientafter intramuscular administration of 1 ml of 5 mg/ml of Bioquantine(45x1ml injections administered each other day).

The patient with CKD as a result of polycystic kidney disease, who had refused dialysis, received treatment with Bioquantine over 2 months and 18 days (starting on July 4, 2015 and continuing through September 22, 2015). At this point, the patient's plasma creatinine levels diminished from 438 μmol/L (5.0 mg/dL) to 318 μmol/L (3.6 mg/dL), a 27.4% decrease. We also observed a decrease in total urine protein concentration for this patient, from 3.20g/L to 1.71 g/L, a 46.6% reduction (Figure 2). This finding may indicate improvement in glomerular function. To assess whether Bioquantine had lasting therapeutic effects, we stopped treatment for a period of 23 days. The patient's plasma creatinine value rose from 318 μMol/L (4.17 mg/dL) to a high of 380 μMol/L (4.98 mg/dL) during the 23-day period of non-treatment, and the serum creatinine level fluctuated between 350 and 376 μMol/L (4.6 and 5.0 mg/dL). The serum creatinine concentration continued to fluctuate in this rangeduring the following seven months of

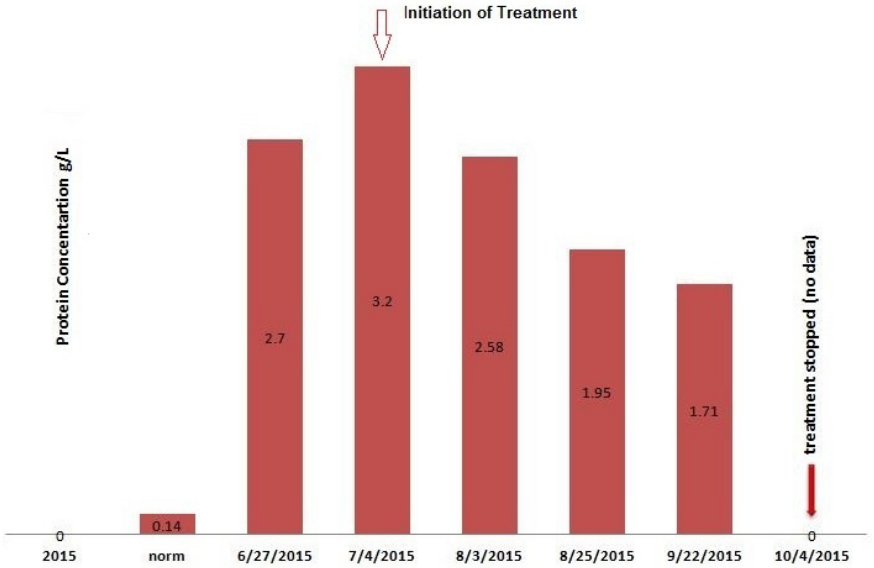
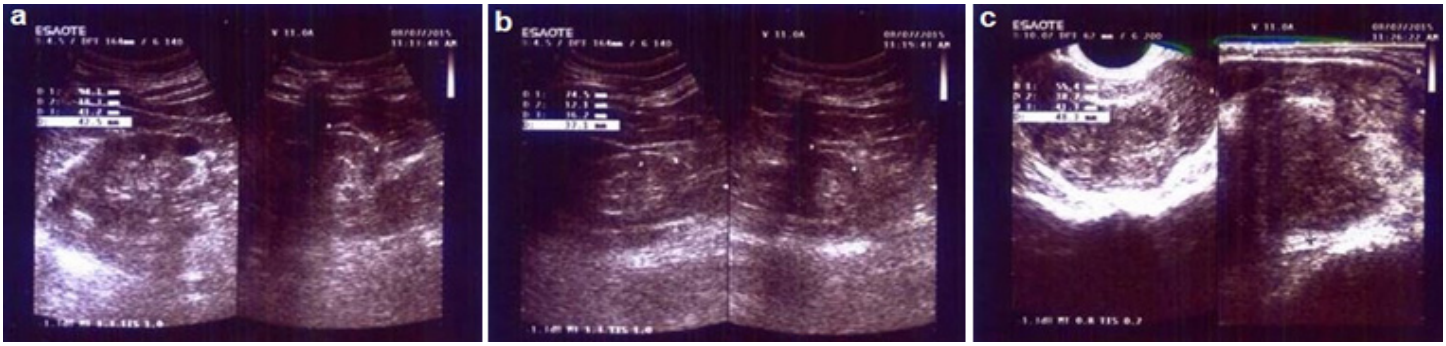


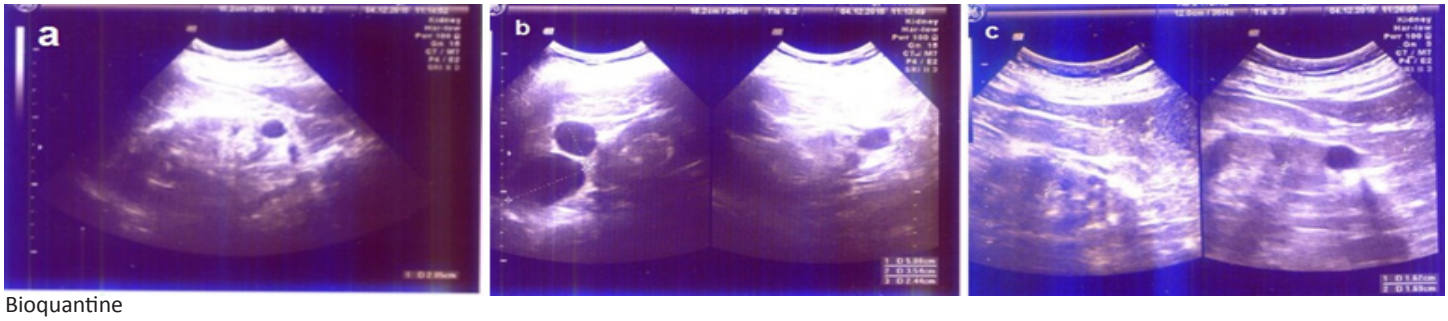
Figure 2: Drop in concentration of total protein in urine of the patient with chronic kidney disease (polycystic chronic kidney) after intramuscular administration of 1 ml of 5 mg/ml of Bioquantine (45x1ml injections administered each other day).

Figure 3: Ultrasonograms of right (a), left (b) kidneys and their corresponding parenchymal layers (c) of the patient with chronic kidney disease (polycystic kidney disease) before the start of Bioquantine treatment (June 8, 2015).



Bioquantine

Figure 4: Ultrasonograms of right (a) and left (b) kidneys and their corresponding parenchymal layers (c) of CKD patient after completion of Bioquantine treatment (December 4, 2015).



Bioquantine

Figure 5: Ultrasonogram results for the patient with chronic kidney disease (polycystic kidney disease) before and after Bioquantine treatment

Location	Right Kidney (cm^3)		Right Kidney Parenchyma (mm)		Left Kidney (cm^3)		Left Kidney Parenchyma (mm)	
	Before	After	Before	After	Before	After	Before	After
Volume/Thickness	165.7	176 .4	17 .0	16.9	120.4	160.8	13.0	16.2
Shift(%)	+ 6.8		- .59		+33.5		+24.6	+24.6
Number of Cysts	3	1	4	3	n/a		n/a	n/a
Ø of Cysts (mm)	13, 16, 16	21	42,25,17, 14	51, 35, 24	n/a		n/a	n/a

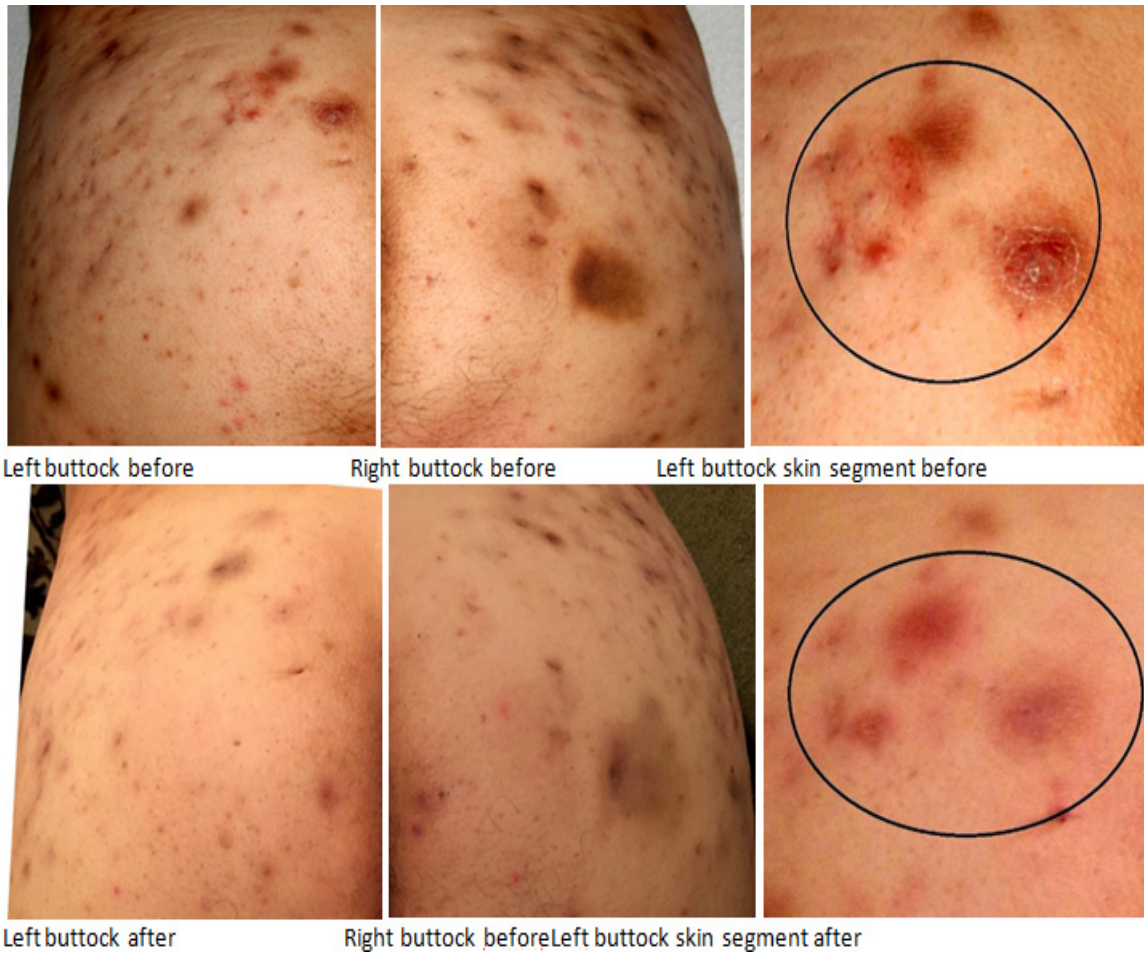


Figure 10: View of affected skin areas of pyoderma gangrenosum patient before and after 2 weeks of Bioquantine therapy.

observation (Figure 10). After this period of observation, the patient began maintenance therapy consisting of a once-weekly IM administration of Bioquantine (1 ml of 5 mg/mL), and his renal function remains stable as of September 19, 2016.

The right and left kidneys had irregular surfaces before and after Bioquantine treatment and all readings showed preservation of corticomedullary differentiation. Other renal ultrasonography findings are listed in (Table 1). The right kidney showed a minimal increase in volume of 6.8% whereas the volume of the left kidney, which was substantially smaller than that of a healthy kidney, increased in volume by 33.5% by the end of Bioquantine therapy. In addition, the thickness of renal parenchymal layer in the left kidney normalized, increasing from 13 mm to 16.2 mm. The thickness of the parenchymal layer of the right kidney, which was in the normal range before treatment, did not change. Positive changes in size and thickness of left kidney without notable changes of same parameters of the right kidney may indicate that Bioquantine and its unique physiochemical properties are capable of selectively targeting areas of pathology. Pre-treatment ultrasonography revealed the presence of multiple cysts of different sizes in both kidneys. After Bioquantine therapy, the number of cysts decreased from 3 to 1 in the right kidney and from 4 in to 3 in the left kidney.

Table 1: Renal Ultrasound Findings for the Patient With Chronic Kidney Disease not Receiving Dialysis*

	Right		Left	
Parameter	**Before Bioquantine**	**After Bioquantine**	**Before Bioquantine**	**After Bioquantine**
Dimensions (mm)	94 X 41 X 43	98 X 45 X 40	70 X 42 X 41	76 X 41 X 43
Cysts	≤13 mm (upper segment) ≤16 mm (lower edge) ≤16 mm (posterior surface level of middle third)	20 mm (lower segment)	≤42 (upper edge) ≤25 mm(external edge, middle segment) ≤17 mmm (lower edge) ≤14 mm (posterior edge, middle segment)	51 mm (upper segment) 35 mm (upper segment) 24 mm (lower segment)
Parenchymal thickness[†] (mm)	17	17	13	16
Blood flow	decreased			
Pelvicalyceal system		not dilated		not dilated
Sinus echogenicity			not increased	
Concretions		none		none
Adrenal gland		Not visualized and no mass lesions in its plane		Not visualized and no mass lesions in its plane

*This patient received Bioquantine starting on July 4, 2015 and continuing through September 22, 2015. The pre-treatment ultrasound was performed on June 8, 2015, and the post-treatment ultrasound was conducted on December 4, 2015.

[†]Measured at the level of the middle segment.

Some cysts became larger, especially those in left kidney, possibly because of the dramatic expansion of volume of left kidney which might stretch the cysts.

CKD and Dialysis

A male patient aged 60 years in New Delhi, India with a history of type 2 diabetes and hypertension and presented with CRF of two years duration. This patient had been receiving dialysis during the last 3 months before starting this study.

After one month of Bioquantine administration(5 mL of 5 mg/mL diluted in 100ml normal saline) every fortnight, followed by 1ml(5 mg/mL) intramuscularly twice a week, his plasma creatinine level stabilized at 700 µmol/L (7.9 mg/dL) and he needed the dialysis only after 15 days. Over the 3 months of therapy, the interval between dialysis improved, and his serum creatinine level has been maintained between 440 and 480 µmol/L (5.0 and 5.4 mg/dL) {Figure 6}. The patient did not required dialysis during the subsequent 10 months of observation.

Type 2 Diabetes and Inadequate Glycemic Control

A male patient aged 67 years from Georgia with type 2 diabetes had received conventional treatment with metformin and glimepiride treatment for 6 months that did not adequately control his hyperglycemia. During the conventional treatment, the patient's FBG concentration increased from 235 mg/dl to 373 mg/dl. After one month of Bioquantine administration, the patient's fasting plasma glucose level dropped from 373 mg/dl to 105 mg/dl. The patient continued receiving 1 ml of 5 mg/ml IP Bioquantine every other day, and his fasting plasma glucose concentration has stayed in the range 85 mg/dl to 125 mg/dl. Patient reported disappearance of

Figure 6: Plasma creatinine values for hemodialysis patient taken off from dialysis after Bioquantine therapy.

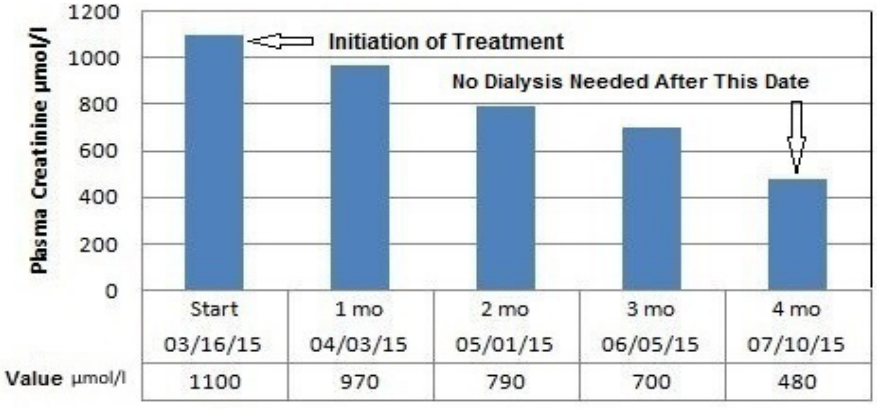
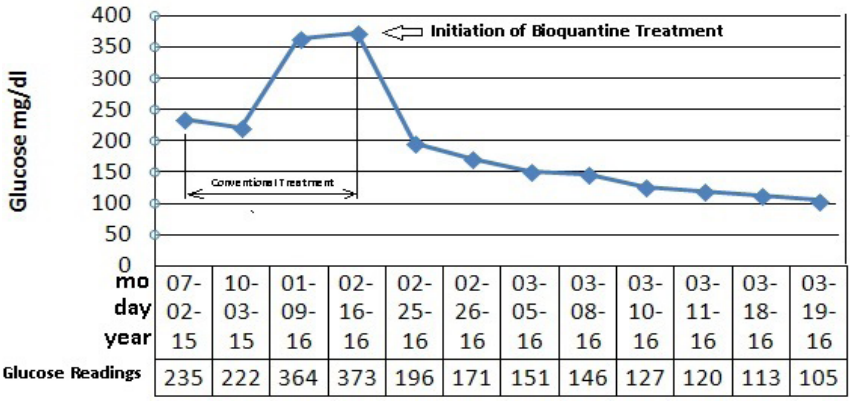


Figure 7: Blood glucose values for hyperglycemic crisis patient under conventional and Bioquantine therapy.



mo	07-	10-	01-	02-	02-	02-	03-	03-	03-	03-	03-	03-
day	02-	03-	09-	16-	25-	26-	05-	08-	10-	11-	18-	19-
year	15	15	16	16	16	16	16	16	16	16	16	16
Glucose Readings	235	222	364	373	196	171	151	146	127	120	113	105

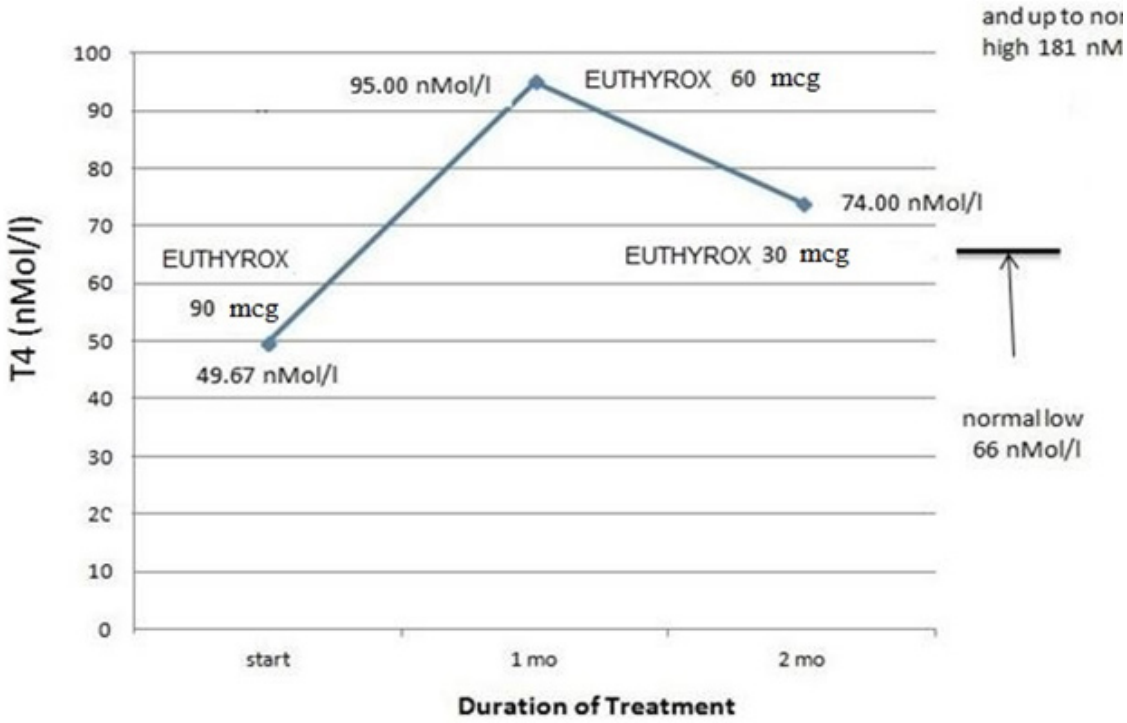
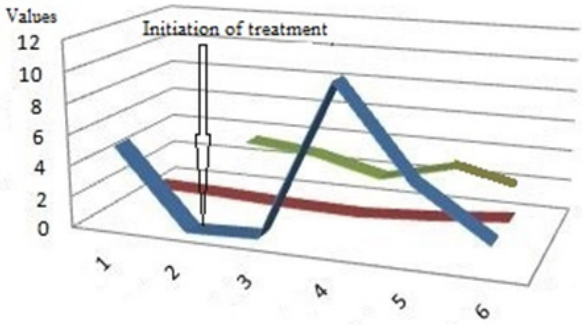


Figure 8: Example of 90µg-60µg-30µg drop in thyroid medication Euthyrox dosage and normalization of T4 values in hypothyroid patient after Bioquantine therapy(normal range 66 nMol/l-181nMol/l).

blurry vision, improvement in night time vision, and considerable relief in dry, itchy skin.

Hypothyroidism

The results of thyroid function test and dynamics of TSH/T4/T3 levels in blood plasma during Bioquantine administration for the patient with hypothyroidism are shown in (Figure 9). Before starting Bioquantine, the patient was taking 90 µg Euthyroxdaily, which allowed TSH/T4/T3 stay in relatively normal range with TSH slightly elevated (5.15 m U/ml). After Bioquantine treatment started, production of thyroid stimulating hormone decreased dramatically (0.011 m U/ml) while T4 and T 3levels stayed generally within the normal range. An approximately 500-fold reduction in TSH production during the first days of treatment may indicate beginning of crucial improvement in thyroid gland function, which by negative feedback caused sharp down regulation of TSH production by the pituitary gland. However, two months later, TSH levelsrose up almost 1000 times to 10.33mU/L, though T4 (relatively) and T3 parameters of thyroid gland stayed in the normal range: 0.64ng/L and 1.74nmol/L accordingly. The tremendous and rapid spike in TSH production was followed by normalization of all three parameters (TSH, T4 and T3) in the next six months of observation. These findings indicate that Bioquantine normalized vital physiological hormones of thyroid gland in this hypothyroid patient.



	4/18/12 1	8/12/14 2	9/12/14 3	10/1/14 4	2/14/15 5	3/12/16 6
TSH mIU/ml	5.15	0.011	0.183	10.33	4.61	1.47
T4 ng/dl	1.1	0.95	0.74	0.64	0.95	1.4
T3 nmol/l	no data	3.4	2.9	1.75	2.9	1.89

TSH Reference Values: 0.4mU/L-4.5mU/L
T4 Reference Values: 0.8ng/dL-1.8ng/dL
T3 Reference Values: 1.0 nmol/L- 2.8nmol/L

Figure 9: Thyrotropin (TSH), T-levothyroxine (T4) and T3-triiodothyronine (T3) levels in hypothyroid patient before and after Bioquantine therapy.

Pyoderma Gangrenosum

A patient in the Russian Federation with pydoderma gangrenosum, a rare and resistant form of noninfectious neutrophilic dermatosis,received both subcutaneous/intramuscular injections of Bioquantine in close vicinity to most painful ulcerated areas located both in upper and lower parts of patient's buttocks. This patient received 14 x1 ml mixed SC/IM injections of Bioquantine. The patient demonstrated considerable reduction in number of sites affected by cutaneous ulcerations with mucopurulent or hemorrhagic exudates (Figure 10).

Recurrence of cutaneous ulcerations after Bioquantine therapy was markedly reduced and new sites of ulceration was practically absent. The diameter of erythema caused by hyperemia (increased blood flow) in superficial capillaries surrounding ulcers and redness of the skin was reduced dramatically.

Spinal cord injury

Magnetic resonance imaging presented in (Figure 11) is obtained from patient's ages 25 years (Figure 11A), 34 years (Figure 11B) and 55 years (Figure C). who suffered from complete and near complete tear within the spinal cord accompanied by its dislocation due to

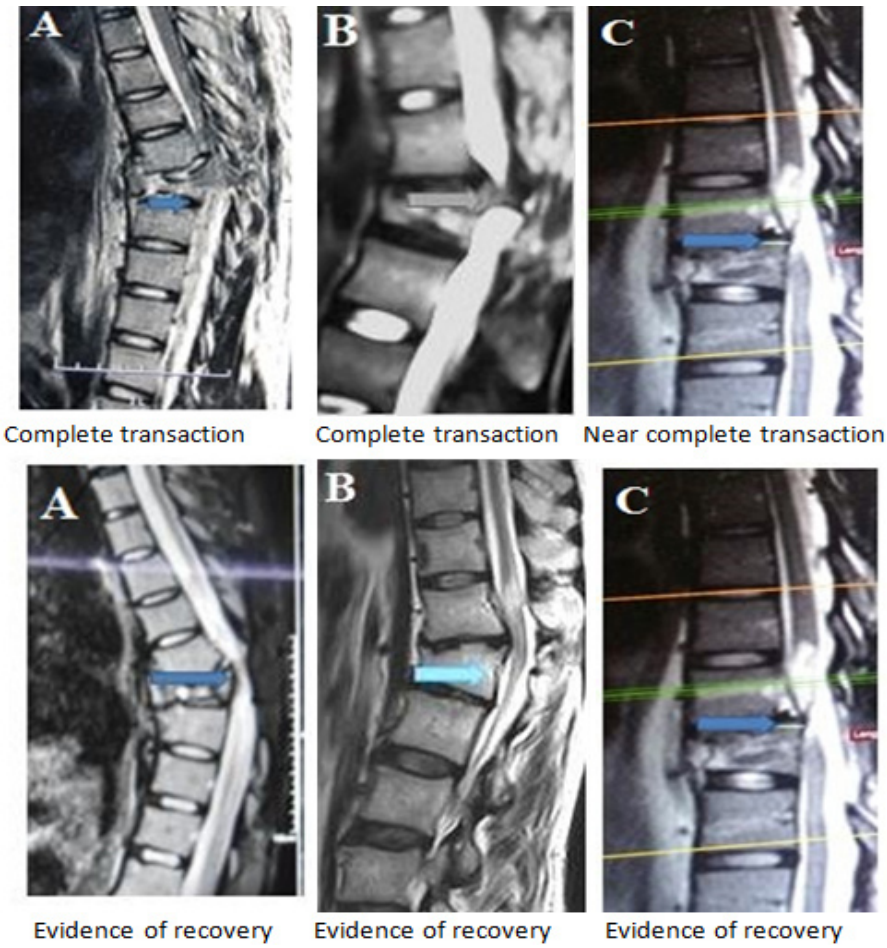


Figure 11: Magnetic Resonance Imaging of three SCI patients before (A, B, C) and after Bioquantine therapy (D, E, F).

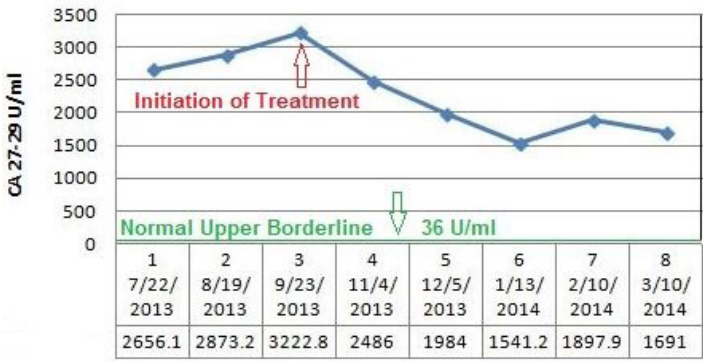
significant traumatic injury. All patients taken into studies were treated for the period of approximately 2 months.

Treatment progress for the three patients with spinal cord injuries (two with complete transection and one near complete spinal cord transection) were assessed by MRI imaging and clinical observation of their motor- and sensory activity. Adjuvant Bioquantine was introduced by mixed IM, IV, intrathecal and intralesional routes. High resolution magnetic resonance images obtained after completion of Bioquantine administration show visible regeneration and reconstruction of spinal cord along all entire length of lesion (Figure. 11).

After two month of administration, patients demonstrated considerable improvement in sensation and walking ability and coordination of movement. All three patients subsequently were moved from ASIA-A to ASIA-B neurological classification (sensory but not motor function is preserved below the neurological level including sacral segments S4-S5).

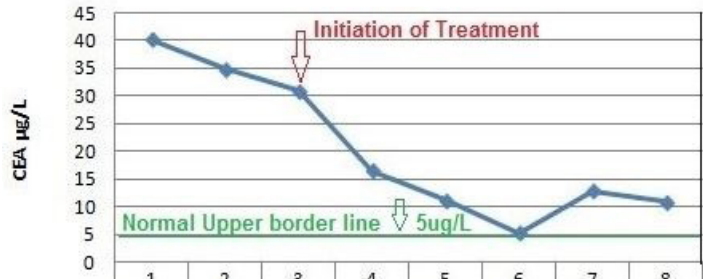
Figure. 12 Down-regulation of cancer antigens CA 27-29 (A), carcinoembryonic antigen (B), and alkaline phosphatase activity (C) in the patient with metastatic breast cancer patient. CEA, carcinoembryonic antigen.

A)



1 7/22/ 2013	2 8/19/ 2013	3 9/23/ 2013	4 11/4/ 2013	5 12/5/ 2013	6 1/13/ 2014	7 2/10/ 2014	8 3/10/ 2014
2656.1	2873.2	3222.8	2486	1984	1541.2	1897.9	1691

B)



1 7/22/ 2013	2 8/19/ 2013	3 9/23/ 2013	4 11/4/ 2013	5 12/5/ 2013	6 1/13/ 2014	7 2/10/ 2014	8 3/10/ 2014
40.1	34.8	30.9	16.5	11.2	5.2	12.8	10.8

Alkaline phosphatase activity in blood samples also substantially decreased during this period, from 297 U/L to 135 U/L which represents 54.5% decrease (Figure 12C). During the period of Bioquantine administration, this patient gained body weight and showed considerable improvement in all vital signs.

Metastatic Breast Cancer

The female patient with metastatic breast cancer patient received IM Bioquantine for 8 months. Down-regulation of three major tumor markers: cancer-associated antigen (CA 27.29), carcinoembryonic antigen (CEA) and hydrolysis enzyme Alkaline Phosphatase (ALP) occurred during Bioquantine administration. From September 23, 2013, when Bioquantine treatment commenced, until its completion on March 10, 2014CA27-29 levels decreased from 3222.8 U/ml to 1691.0 U/ml which represents 47% drop (Figure12A). During this same period, CEAlevels decreased from 40.1µg/L to 10.8 µg/L, a 33% drop (Figure 12B).

C)

1 7/22/ 2013	2 8/19/ 2013	3 9/23/ 2013	4 11/4/ 2013	5 12/5/ 2013	6 1/13/ 2014	7 2/10/ 2014	8 3/10/ 2014
283	276	297	229	191	156	153	135

Discussion

These first-in-human studies all observed beneficial changes with Bioquantine administration in a small group of patients with a variety of disorders. The patient with CKD due to polycystic kidney disease showed volumetric improvement along with reduction in the number of renal cysts and decreases in plasma creatinine. The patient with CKD related to hypertension and type 2 diabetes also demonstrated decreases in plasma creatinine. This patient, who had been dependent on dialysis at study entry, was able to go for longer periods between dialysis to the point where he did not require dialysis for many months. The patient with type 2 diabetes and inadequate glycemic control on standard oral therapy showed normalization of fasting blood glucose and improvements in retinal neuropathy. The patient with hypothyroidism achieved normalization of thyroid function. The patient with pydoderma gangrenosum demonstrated considerable reduction in number of sites affected by cutaneous ulcerations with mucopurulent or hemorrhagic exudates and reduction in recurrences. The three patients with spinal cord injuries had regeneration and reconstruction of spinal cord along all entire length of lesion as

visualized using MRI and considerable improvement in sensation and walking ability and coordination of movement. The patient with metastatic breast cancer showed substantial decreases in cancer-related biomarkers and improvement in body weight and vital signs.

The positive human findings for spinal cord injury, pydoderma gangrenosum, and metastatic breast cancer are consistent with the results from previous animal studies with experimental models of traumatic brain injury, skin wrinkling, and melanoma, respectively (Paylian et al, 2016, in development). The improvements shown in renal function in CKD and glycemic control in type 2 diabetes extend the range of documented therapeutic effects of Bioquantine.

Because of ethical reasons, legal restrictions, and a limited numbers of volunteers, we were able to treat only a very small number of patients. These results indicate that Bioquantine may be safe and well tolerated for use in humans, and deserves further study in a range of degenerative disorders. We propose that the mechanism of action of Bioquantine in these various diseases derives from its unique pharmacology and combinatorial reprogramming properties. The dramatic changes in TSH dynamics in the patient with hypothyroidism are consistent with this reprogramming hypothesis. We observed the same normalizing pattern for thyrotropin by the 5th month following an increase of TSH production as is seen during normal development. During infancy, TSH values reach 18.1 mU/L from age 0 to 1 months and 8.21 mU/L from age 1 to 12 months. Then gradually, by age 15 to 18 years, TSH level stays approximately at 4.33 mU/L mark [9].

In conclusion, these preliminary findings suggest that Bioquantine is safe and well tolerated and contains ubiquitous therapeutic activity including restoration of renal function, glycemic control, and thyroid function, dermatologic healing, spinal cord regeneration, and anti-breast cancer activity.

References

1. Kim JS, Choi HW, Choi S, Do JT (2011) Reprogrammed pluripotent stem cells from somatic cells. Int J Stem Cells 4(1): 1-8.
2. Dziedzic K, Pleniceanu O, Dekel B (2014) Kidney stem cells in development, regeneration and cancer. Semin Cell Dev 36: 57-65.
3. De Groef S, Staels W, Van Gassen N, Lemper M, Yuchi Y, et al.(2016) Sources of beta cells inside the pancreas. Diabetologia 59(9): 1834-1837.
4. Bilousova G, Roop DR (2014) Induced pluripotent stem cells in dermatology: potentials, advances, and limitations 4(11): 15164.
5. Tso D, McKinnon RD (2015) Cell replacement therapy for central nervous system diseases. Neural Regen 10(9): 1356-1358.
6. Kim WH, Shen H, Jung DW, Williams DR (2016) Some leopards can change their spots: potential repositioning of stem cell reprogramming compounds as anti-cancer agents. Cell Bio Toxicol 32(3): 157-168.
7. Tokmakov AA, Iwasaki T, Sato KI, Kamada S (2016) Reprogramming of somatic cells and nuclei by Xenopus oocyte and egg extracts. Int J Dev Biol 60(7-8-9): 289-296.
8. Paylian S (2015) Co-electroporation with Xenopus laevis oocytes reprograms normal and cancerous human cells to resembel induced human pluripotent stem cells. BAOJ Cancer Res Ther 1: 1-13.
9. Kapelari K, Kirchlechner C, Hogler W, Schweitzer K, Virgolini I, et al. (2008) Pediatric reference intervals for thyroid hormone levels from birth to adulthood: a retrospective study. BMC Endocr Disord 8-15.