Evaluation of Biofield Treatment Dose and Distance in a Model of Cancer Cell Death

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Abstract

Objective: This study assessed the potential influence of biofield treatment on cultured human cancer cells and whether such influence was affected by varying the duration of the treatment (dose) or the distance between the biofield practitioner and the target cells.

Design: Biofield treatment dosage was assessed from a short distance (0.25 meters) in three independent experiments involving 1, 2, or 5 treatments, along with another set of three independent and comparable mock experiments. Biofield treatment distance was assessed at 0.25, 25, and ~ 2000 meters involving two treatments in three independent experiments along with another set of three mock experiments.

Intervention: Biofield treatments were delivered by a highly acclaimed biofield practitioner with the intention of diminishing growth of the cells or inducing cancer-cell death.

Outcome measure: Cell viability was quantified 20 hours after treatments, using a spectrophotometric assay for live-cell counting. The dependent measure for each experiment was the log ratio of the cell viability values of treated samples (biofield or mock) over the values of untreated control samples.

Results: A trend of decreasing cell viability with increasing biofield dose was evident in the first set of experiments assessing dose–response; however, no such effect was evident in the second set of experiments evaluating biofield treatment distance. Mock experiments yielded relatively stable viability ratios in both sets of experiments. Linear regression analysis and hypothesis testing of the data taken as a whole did not yield statistical significance at $p < 0.05$.

Conclusions: These results represent the first indication of a biofield treatment dose–response in a controlled laboratory setting. The data are inconclusive because of the inability of reproduce the cellular response in a replicate experiment.

Introduction

WITHIN THE BURGEONING FIELD of complementary and alternative medicine, the domain of biofield treatments, or energy medicine, is replete with both faith and skepticism. Biofield treatments involve a putative bioenergy that is intentionally channeled by a practitioner to heal patients from a distance. A common assumption among biofield practitioners is that increased dosage, typically in the form of repeated treatments—and even increased distance over which the treatments are delivered¹—can result in increased efficacy. This trial sought to examine this assumption, using human cancer cells cultured *in vitro* as the target of biofield treatments. Two sets of experiments were conducted under strict laboratory conditions: one evaluating varying dosage and a second evaluating biofield treatment delivered from varying distances.

Materials and Methods

Cell culture and handling

Human U87 glioblastoma cells were expanded in standard culture media (RPMI-1640, Sigma-Aldrich, St. Louis, MO) supplemented with 10% fetal bovine serum and divided into aliquots for storage in liquid nitrogen. At the start of each experiment, an aliquot was thawed and cells were seeded into four 96-well culture plates at 1000 cells/well. The plates were each labeled with an identifying code and placed in a humidified incubator (designated as incubator #1) set at 37°C and 5% CO₂. After 4 hours of incubation, two of the plates were selected randomly by a second scientist to receive treatment (biofield or mock), transferred to a treatment room, and placed in a humidified incubator with clear walls (incubator #2). The remaining two plates served as controls and received no treatment but were taken out of the

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incubator for 2 minutes at the beginning and end of the treatment period to mimic the change in ambient temperature that occurred with the treatment plates during their transfer. At the completion of the treatment period, the two treatment plates were returned to incubator #1. All four plates remained undisturbed overnight and cell viability was assessed the following day.

Cell viability analysis

Cell viability was quantified 20 hours after the initiation of experimental treatments (biofield or mock) using the Cell Count Kit-8 (Dojindo Laboratories, Santa Clara, CA), a colorimetric assay based on a tetrazolium salt that is reduced by dehydrogenase activities within cells to produce a yellow-color formazan dye. The amount of the formazan dye generated in the cell-culture media is directly proportional to the number of living cells. The absorbance was read, using a spectrophotometer with a test wavelength of 450 nm. Data were acquired from a total of 30 independent wells in each plate, avoiding wells on the edge of the culture plate. Background absorbance was compensated for, using measurements obtained from six wells that contained only media (no cells). The average absorbance of these wells was subtracted from the other wells.

Because of natural variation in cell-viability measurement from day to day, the dependent measure for each experiment was the ratio of the cancer-cell viability values from treated cultures (biofield or mock) over the values from control cultures. Cell viability ratios were computed by dividing the mean values from the duplicate plates kept in incubator #2 during the treatment period (a total of 60 wells) by the mean values from the duplicate plates in the control

incubator (a total of 60 wells). Because ratios are inherently asymmetrical, a log transform was applied to the ratios. A log ratio value less than zero indicates cancer cell death or diminished growth following treatment compared to the control condition and a value greater than zero indicates the opposite.

Biofield and mock treatments

All biofield treatments were delivered by one practitioner who is recognized internationally as a healer and who has developed a technique of mental energy transmission based on innate healing abilities. Following published guidelines,² the practitioner remains anonymous to avoid any potential conflict of interest. Each treatment lasted for 5 minutes. For increased dosage, treatments were repeated with 10-minute rest periods. During treatments from a close distance, the practitioner was escorted into the treatment room and seated in front of incubator #2. Distant treatments were delivered remotely, aided by a webcam link to a live image of the cells in incubator #2. During mock treatments, the duplicate plates were placed in incubator #2, and the room remained empty for the treatment time without the practitioner being involved.

Systematic negative controls

The experiments involving mock treatments served as systematic negative controls throughout the study; that is, each set of experiments included an equal number of experiments in which all of the physical manipulations of the cells and subsequent analyses were carried out except for the

FIG. 1. Viability plots for cancer cells exposed to either mock or biofield treatments. Cell viability ratios are plotted for (A) a set of experiments evaluating biofield dosage and (B) biofield treatment distance. Points represent the log-transform of the ratios of mean viability values from treated samples divided by mean values from untreated control samples. Positive values indicate an increase in cell viability, compared to control; negative values represent a decrease in cell viability. Dosage is cumulative duration of a number of 5-minute treatments (1, 2, and 5 treatments). Error bars represent log-transformed standard deviations. The dagger symbol (†) indicates replicate experimental conditions (same dosage and distance).

experimental intervention. This method of assessing variability intrinsic to the experimental system was considered applicable based on its usefulness in the study of nonionizing electromagnetic fields.³

Blinding and randomization

The experimental protocol was divided between two scientists to insure that the scientist handling the cells and analyzing the data was not aware of whether the cells had received biofield treatment, following published guidelines.⁴ Identifying codes and group assignments were determined using the random number generator function of a scientific calculator.

Results

Six independent experiments were conducted to evaluate the potential for varying biofield treatment dosage: three involving increasing numbers of biofield treatments (1, 2, and 5 treatments with a total cumulative duration of 5, 10, and 25 minutes, respectively); and three involving only mock treatments. The cell-viability ratios for the three mock/control experiments were all close to zero while those involving biofield treatments of increasing dosage appeared to be monotonically decreasing (see Fig. 1A). Thus, the greatest cancer-cell inhibition was observed when the practitioner was closest and delivered the highest dose.

A second set of six experiments was conducted: three involving the same biofield treatment dosage (2 treatments with a total cumulative duration of 10 minutes) from various distances (0.25, 25, and ~ 2000 meters); and three involving

mock treatments. None of the resulting cell-viability ratios in this second set of experiments were significantly different than zero when considering their standard deviations (see Fig. 1B). Notably, the experiment involving the treatment from the closest distance in these experiments is a replication of the experiment in the first set of experiments involving the 10-minute biofield treatment but the results were in opposite directions.

A weighted linear regression was performed to assess the monotonically decreasing dose-response effect using the model: $Y = B_0 + B_1X$, for which X is biofield treatment time, Y is log ratio of cell viability, and the transformed standard deviations are used as weights. The data point from the replicate experiment in the second set of experiments (treatment from closest distance) was included. Although the estimated slope (B_1) was -0.039 (and this is consistent with a trend of decreasing cell viability as a function of treatment duration) the parameter estimation was not well-determined ($p=0.348$). Linear hypothesis testing against the null hypothesis ($Y=0$), using the estimated parameters and covariance from the linear regression analysis did not yield statistical significance ($p=0.410$). Nonparametric hypothesis testing of the data considered as a whole (six biofield/control ratios versus six mock/control ratios) did not yield significance via the Wilcoxon rank-sum test ($p=0.699$) or via the Kruskal-Wallis test ($p=0.631$). Table 1 summarizes the underlying data.

Discussion

The apparent dose-response observed in the first set of experiments is provocative, particularly in light of the results of the systematic negative control experiments

TABLE 1. CELL VIABILITY VALUES AND SUBSEQUENT CALCULATIONS UNDERLYING THE RATIO VALUES FOR ALL 12 INDEPENDENT EXPERIMENTS

Condition	Treatment time (minutes)	Treatment distance (meters)	Viability measures by plate				Viability measures across plates		Natural log ratio	Natural log RatioSD
			Control (incubator #1)		Biofield/mock (incubator #2)		Control (incubator #1)	Biofield/mock (incubator #2)		
			Plate 1 mean/SD	Plate 2 mean/SD	Plate 1 mean/SD	Plate 2 mean/SD	Mean/SD	Mean/SD		
Mock	5	0.25	0.131/0.023	0.146/0.023	0.141/0.026	0.166/0.025	0.139/0.024	0.153/0.028	0.099	0.185
	10	0.25	0.083/.013	0.090/0.014	0.075/0.015	0.101/0.023	0.087/0.014	0.088/0.023	0.014	0.275
	25	0.25	0.095/0.017	0.111/0.012	0.098/0.011	0.102/0.044	0.103/0.017	0.100/0.032	-0.027	0.326
	10	0.25	0.568/0.084	0.598/0.099	0.537/0.108	0.597/0.127	0.583/0.092	0.567/0.121	-0.028	0.178
	10	300	0.354/0.064	0.384/0.045	0.338/0.055	0.367/0.087	0.369/0.057	0.352/0.074	-0.046	0.215
	10	2000	0.396/0.131	0.412/0.064	0.316/0.067	0.466/0.077	0.404/0.102	0.391/0.104	-0.032	0.341
Biofield	5	0.25	0.145/0.034	0.132/0.043	0.158/0.051	0.148/0.036	0.139/0.039	0.153/0.044	0.099	0.373
	10	0.25	0.107/0.057	0.139/0.027	0.065/0.031	0.073/0.040	0.123/0.047	0.069/0.036	-0.579	0.553
	25	0.25	0.180/0.029	0.191/0.032	0.079/0.025	0.097/0.032	0.185/0.031	0.088/0.030	-0.741	0.339
	10	0.25	0.210/0.073	0.410/0.084	0.384/0.057	0.419/0.066	0.310/0.128	0.402/0.064	0.260	0.382
	10	300	0.580/0.115	0.667/0.080	0.671/0.074	0.646/0.136	0.624/0.108	0.658/0.110	0.054	0.199
	10	2000	0.399/0.129	0.514/0.062	0.483/0.070	0.452/0.068	0.457/0.116	0.468/0.070	0.023	0.308

Viability measures represent average values of spectrophotometric readings of individual wells corrected for background intensity (media only). Average values are shown for individual duplicate plates (30 wells) as well as for both plates (60 wells) along with their standard deviations. Logarithmic transforms of the ratios of biofield treatment/mock to control are also shown along with the transformed standard deviations.

SD, standard deviation.

conducted in parallel that indicated a relatively stable baseline variability in the data output from the model system. However, the interpretation of these results is complicated by the lack of replicability observed in the second set of experiments using the same model system and the same participants. Part of the struggle is reconciling the centuries of precedent in which the test of replicability has been a fundamental tenet of the scientific method, and the acknowledgment that the experimental intervention in this model system derives from a human being and may thus be inherently changing with time and situation (e.g., mental and emotional state at a particular time). Lack of replicability has been a consistent element in a series of *in vitro* studies of various biofield treatment modalities conducted by the current authors' group⁵⁻⁷ and seems to be consistent with the extant data across the field. For example, the lack of independent replications was noted as an important issue in critical reviews that assessed *in vitro* studies of external *qigong*, another form of biofield treatment.^{8,9} *In vitro* models can be advantageous in terms of minimizing confounding variables; yet, these models may be sub-optimal for investigating biofield treatment efficacy for a number of reasons. Just one example is that biofield therapies may require the presence of an organized biofield in the target system for optimal effect, and this element would be disrupted or dissipated in cell cultures separated from the body. While there are encouraging data indicating that some disadvantages of *in vitro* models might be overcome through experimental design considerations specific to biofield treatment models,¹⁰ the current authors believe that *in vivo* models are more appropriate for investigating biofield treatments at the current stage of this research where resources are limited and evidence of efficacy remains controversial. Questions regarding the underlying mechanisms of action may ultimately be addressed *in vitro* better, but *in vivo* models offer the most hope for evaluating the efficacy of biofield treatments and would thus serve the most urgent public health needs better.

Conclusions

The results of this trial represent the first indication of a biofield treatment dose-response in a controlled laboratory setting. The data are inconclusive because of the inability to reproduce the cellular response in a replicate experiment.

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Disclosure Statement

No competing financial interests exist.

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