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Cancer Growth Acceleration By External Electrostatic Fields

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***Abstract*— A number of biological studies, both here and abroad, suggest that external electrostatic fields can exert influence inside the body of a living mammal. Depending on the circumstances, this influence may be beneficial or detrimental. For example, we have previously reported that medically controlled application of these fields can beneficially increase the action of chemotherapy against cancer (see PubMed: Gray, J., Frith, C., Parker, J.: Bioelectromagnetics 21, 575-583, 2000). On the other hand, some studies suggest that uncontrolled exposure to these fields may present previously unrecognized hazards. Here we report on the ability of external electrostatic fields to significantly accelerate cancer growth inside the bodies of mice.**

Female mice with transplanted murine mammary adenocarcinoma were divided into four randomly assigned groups of 11 animals each. Two of the four groups were subjected to electrostatic fields generated from charges created as the mice rubbed their fur against a polyester carpet suspended above the animals. One of the other groups also rubbed against a carpet surface (to incur the same mechanical forces), but the carpet was treated with an antistatic agent to reduce static charge generation. The final group did not rub against a carpet surface to generate static charge. The qualitative level of static charge (and thus electrostatic fields) the groups were exposed to was measured daily. All groups started the study with approximately equivalent tumor burdens. However, within 13 days the median tumor growth in the two groups exposed to higher electrostatic fields was over twice as large as that of the two minimal-field exposed groups. Statistically, *p*-values as low as 0.006 were achieved.

I. Introduction

The work we report here should not be confused with the current debate over a possible connection between electromagnetic fields and cancer. Electrostatic fields and electromagnetic fields are different in their physics, and in the way we are exposed to them. In addition, and unlike power-line frequency electromagnetic fields, electrostatic fields have shown strong biological influence in a number of previous animal studies.

Much of our current knowledge regarding electrostatic field interaction with biological systems stems from studies conducted in the late 1960's through the 1990's. This research included subjects ranging from possible dangers to possible therapeutic uses. For examples see [1-16]. In one example, some of the early work on the now common use of electric or magnetic fields to stimulate osteogenesis showed that 21-day exposure to an externally applied electrostatic field of just 2,500 V/m could significantly increase the healing speed and strength of fractured rabbit fibulas [17, 18].

More recently, we have reported that externally applied electrostatic fields can significantly increase the affect of chemotherapy against cancer cells inside the bodies of mice [19]. We believe that controlled exposure to these fields may improve some medical treatments by locally potentiating the action of applied therapeutic agents. However, in addition to this hopeful possibility, we are observing adverse effects from uncontrolled exposure to these fields. Our studies have shown that external electrostatic fields, without chemotherapy present, can accelerate cancer growth inside a mammalian body. We first observed this in studies exposing mammary tumor-bearing mice to artificially generated electrostatic fields (using a high-voltage power supply to charge an insulated metal plate above the animals). The present study was conducted to evaluate the ability of naturally generated electrostatic charges and fields (i.e. tribo, or frictional, electric charges) to influence cancer growth inside the bodies of mice.

II. Methods

Female B6C3F1 mice, approximately five weeks of age, (Charles River Laboratories) were used for the study. On day zero of the study, 44 of the mice were each implanted with an approximately 35 mg fragment of 16/C murine mammary adenocarcinoma. The implant was placed subcutaneously in the axillary region (chest) through a puncture in the inguinal region. The tumor fragments used were from a mixture of minced tumors from several donor animals maintained by Southern Research Institute, Birmingham, Alabama. This animal model, tumor, and implant method, are commonly used in chemotherapy research. After the implant an area of fur, approximately 12 mm by 25 mm, over each tumor was clipped so the site could be periodically examined to track tumor development.

Following the implant, all of the animals were placed in cages for four days to allow the individual tumors to develop to a measurable size. Suspended cages with open-grid floors were used throughout the study. The open-grid floors allowed wastes to drop into a polyethylene tray beneath each cage, and also provided good ventilation to keep humidity levels low and the animals dry. We had determined in previous studies that keeping the animals clean and dry was required for consistent application of electric fields. This has also been previously reported [20]. Urine is so conductive that even an invisible film on the animals' fur, or on cage surfaces, allows electric charges to move and distorts local electric fields. Closed cages, with the animals on litter, do not keep the animals and cage surfaces dry enough for this type of study even if the cages are individually ventilated.

Food (Purina Formulab 5008) and water were provided *ad libitum* to each group throughout the study. Central air-conditioning and an auxiliary dehumidifier maintained 67 to 73° F, and 35 to 40% relative humidity. Fluorescent lighting was on for 12 hours and off for 12 hours. Twice daily survival checks for morbidity and mortality were made.

Caliper measurements of each animal's tumor were recorded on day four, then each three days thereafter. The measurements were used to estimate tumor weight as the volume of a prolate ellipsoid using length and width measurements in mm to provide weight in milligrams with the formula: (Length x Width^2) ÷ 2. This method is recognized by the National Cancer Institute, and allowed us to track tumor development over the course of the 13-day study without having to sacrifice the animals.

On day four, as each animal's tumor was measured and converted to milligrams, the animals were divided into four groups of 11 animals each with approximately equal numbers of equivalent size tumors in each group. This ensured that all groups had approximately equivalent tumor burdens at the start of electrostatic field exposure. The study was blinded (but not double-blinded) from this point forth. The four groups were blindly randomized and assigned to one of the four study methods. In addition the identification of the study groups was masked to blind all subsequent tumor measurements.

Starting at this time each group was maintained in a cage specifically designed not to, or to, generate electrostatic charges on the animals' fur as they moved about in their normal activities. All of the cages were constructed from Sterilite Clear View™ brand 66-quart polyethylene storage boxes. The box bottoms were removed and replaced either with a wire-grid (formed from 6 mm mesh, 23-gauge galvanized hardware cloth), or two crossed layers of 12 mm open square by 9 mm thick styrene-plastic grid (Plaskolite, Inc.). Both methods provided an open-grid cage floor that measured 34.3 cm by 50.2 cm, and the resulting 1,722 square centimeter space was almost double the U.S. Department of Health and Human Services horizontal size recommendations for housing the 11 mice used in each group. Carpet was placed above the animals in three of the cages, and this restricted vertical movement of those animals on study days 4 to 13 to less than that recommended by the HHS "Guide for

the Care and Use of Laboratory Animals". However spacing the carpet above the animals so they could rub against it to generate electrostatic charges was a critical part of the study protocol, and this variance is therefore allowed under the Guide.

Figure 1 shows one of the cages with carpet suspended so the animals would rub their back fur against the carpet nap as they moved about the cage (only one animal shown). The untreated control group cage did not contain carpet. The carpet used in the other three cages was Shaw Industries, Patriarch Style, 100% polyester. All carpet used was cleaned with Tide Free™ soap and water, followed by thorough water rinsing to remove any antistatic agents. The carpet was attached to Plaskolite™ plastic-grid, and suspended by plastic spacers with the carpet nap 22 mm above the cage floor. All cages were suspended 12.5 cm above trays used to catch wastes and this spacing, along with the open-grid cage floors, provided good airflow into the cages even with the carpet in place.

Group A, serving as an untreated control group**,** was housed in a wire-grid floored cage identical to that used for Groups B and C, but without carpet suspended above the animals. This cage provided minimum electrostatic charge generation on the animals' fur. We had previously determined that, even with the animals occasionally rubbing their fur against each other, they would typically encounter less than 200 volts of electrostatic potential in this type of cage.

Group B, serving as a treated control group, was housed in a wire-grid floored cage with carpet suspended above the animals. However, in this group the carpet was treated with a conductive mixture (1% Exxon Q-14-2 cationic surfactant) to provide minimum charge generation. The animals in this group were subjected to the same mechanical forces (rubbing against the carpet) as the animals in Groups C and D, but not to the high-intensity electrostatic fields experienced by Groups C and D.

Group C was housed in an identical cage as that used for Group B (with a wire-grid floor), but with nonconductive carpet suspended above the animals. Significant electrostatic charges were generated on the animals' fur as they moved about the cage.

Group D was housed in a cage with suspended nonconductive carpet like that used for Group C. However, for Group D the cage used a styrene plastic-grid floor. The floor was formed from two pieces of Plaskolite™ grid. This grid has 12 mm square openings, and two pieces were laid on top of each other with the openings out of line to cross and reduce each opening to about 6 mm square. This group was included to provide a comparison group to determine if the conductive wire-grid floor might be a factor in results seen in the Group C cage. In Group D, significant electrostatic charges were generated on the animals' fur as they moved about the cage and the nonconductive styrene floor prevented charges from flowing off of, or on to, the animals' bodies from a conductive floor as would occur in Group C.

Fig. 1. Cage with carpet suspended to rub the animals' fur (only one animal shown)

From day four (at the time carpet was first placed in the cages) forward, the suspended carpet in the cages of Groups B, C, and D was replaced every 24 hours to minimize any influence from oils or urine on the animals' fur that might be transferred to the carpet. Also, the waste trays under all cages were replaced every 24 hours, and clean cages were provided every three days. This minimized humidity and conductive film build-up in the cages that might reduce charge generation.

This mouse strain is particularly wild and does not calmly tolerate being handled. It is therefore not possible to position the animals to directly measure the potential of the electrostatic charges on their fur without creating or destroying charges while holding the animals. Instead an indirect measurement of the approximate electrostatic potential the fur of the animals in Groups B, C, and D was acquiring was possible when the suspended carpet was removed and replaced in each cage. The carpet was replaced daily, and each time the electrostatic potential at four identical locations on each carpet was measured (in reference to ground) with a Monroe #261 Digital Static Meter. Under the *Law of Charge Conservation*, an equal but opposite polarity charge could be expected to be on the fur of some of the animals in the group at that time. The 36 measurements made for each group (day 5-13) were then averaged. This provided a qualitative measurement of the relative level of electrostatic potential, and thus electrostatic field, the individual groups were experiencing.

The charge on an animal's fur was about 1 mm (approximately the thickness of the fur) away from the body. The relative level of electrostatic field intensity the animals in each group were experiencing was calculated as a point in space on an axis per-

pendicular to, and passing through the center of, a 1 cm diameter uniformly charged area of fur having the average electrostatic potential found for that group.

The 16/C tumor is fast growing but there are always a few tumors that naturally grow unusually slow or fast in any group of animals, and this results in skewed distributions. Nonparametric analysis based on group medians is the recommended metric under these conditions [21]. In addition, the interquartile range is recommended to illustrate the tumor variability between the 25^{th} and 75^{th} percentiles, thus examining the central 50% of data to minimize the influence of tumors that have naturally grown unusually slow or fast [22].

Statistical differences among the study groups' tumor weights were evaluated subsequent to day 4 in terms of each animal's change in tumor weight relative to that same animal's tumor weight on day 4. The Fisher exact test was first used with the null hypothesis assumption of the same median in Groups A and B (low electrostatic field exposure) and Groups C and D (high electrostatic field exposure) to compare the proportion of cases in each group that exceeded the combined median. This was followed by Mann-Whitney tests to indicate significant differences in absolute tumor sizes, differences in tumor size changes from day 4, and also odds of a Group C and D animal having a larger tumor size increase than a Group A and B animal. In addition, stem-and-leaf plots [23] were made for the tumors in each group on each measurement day to provide the variability of the interquartile tumor range.

III. Results

Figure 2 graphically illustrates each group's median tumor weight gain percent from day four to day 13 (end of the study). All four study groups started with approximately equivalent tumor burdens on day four. The median tumor of Group A compared to B, and also the median tumor of Group C compared to D, behaved in a very similar manner throughout the study. However, the median tumor growth rate for Groups C and D was over twice that of Groups A and B by day 13.

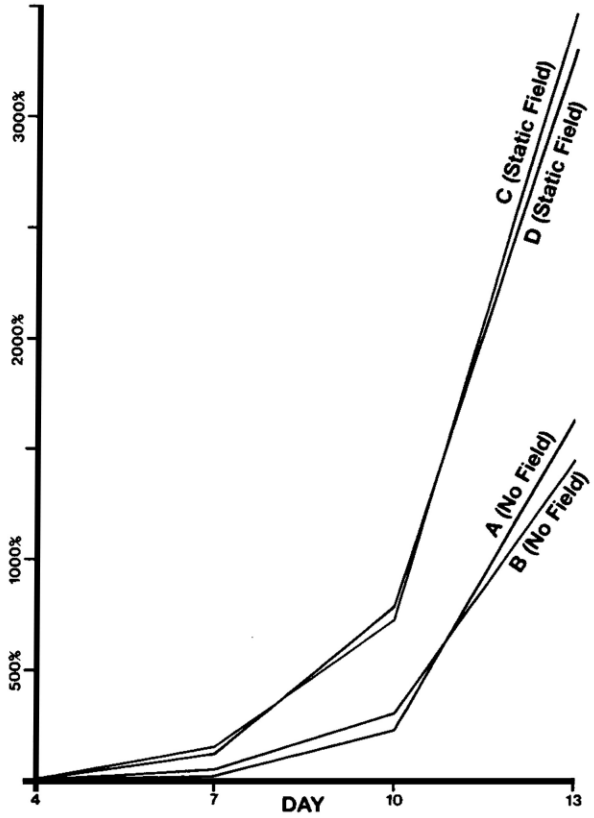

Fig. 2. Median tumor weight gain percent from the start of field exposure on day 4

By weight, food and water consumption for all groups was approximately equivalent throughout the study. There was no significant difference in mean body weight among the groups during the study (Table 1).

The average electrostatic potentials found on the carpets of the groups are shown in Table 2. Note that the Group A cage did not have carpet suspended above the animals, so they were not exposed to significant static charges. Table 2 also shows the electrostatic field intensity an animal would experience from the average potentials measured. Groups C and D were subjected to significantly higher electrostatic potentials, and thus electrostatic fields, than were Groups A and B.

TABLE 1: GROUP MEAN BODY WEIGHT (g)

Group	Day 4	Day 7	Day 10	Day 13
A	18.3	19.7	20.3	22.2
B	18.3	19.3	20.0	21.3
C	18.5	19.4	20.5	23.4
D	18.6	19.6	21.1	23.1

TABLE 2: AVERAGE ELECTROSTATIC POTENTIAL AND FIELD

Group	Potential	Field at 1 mm
A	< 200 V*	*
B	-300 V	48,233 V/m
C	-1,250 V	200,971 V/m
D	-2,350 V	377,825 V/m

* = Group A did not rub against carpet to create electrostatic charges

Table 3 shows the median tumor weight (mg), on each measurement day, for each group. The interquartile range of tumor size is shown in parentheses, and the overall range is shown in braces.

TABLE 3: MEDIAN TUMOR WEIGHT (mg)

Group	Day 4	Day 7	Day 10	Day 13
A	36.5 (30-48) {12-85}	37.0 (32-59) {25-294}	119.8 (110-292) {37-1130}	636.0 (425-956) {310-3514}
B	39.5 (30-61) {12-103}	59.6 (45-115) {35-184}	161.9 (103-284) {54-921}	619.0 (517-1409) {406-2160}
C	43.5 (35-60) {15-80}	98.4 (59-112) {41-311}	358.2 (249-525) {140-1331}	1548.0 (877-1880) {512-2954}
D	41.6 (36-68) {15-184}	104.6 (67-158) {36-240}	362.4 (229-496) {112-841}	1459.0 (993-2085) {429-3361}

(Numbers in parentheses = Interquartile tumor range)
{Numbers in braces = overall tumor range}

Statistical analysis (with a p-value of less than 0.05 considered significant) revealed:

1) There was not a significant difference among the group tumor sizes on day four at the start of electrostatic field exposure.
2) Groups A and B were not significantly different at any time during the study.
3) Groups C and D were not significantly different at any time during the study.
4) The Fisher exact test indicated there was a significant difference between Groups A and B (minimal static field exposure) compared to Groups C and D (intense static field exposure) on all three measurement days after static field exposure was started: Day 7 p=0.034, Day 10 p=0.006, and Day 13 p=0.034.
5) In terms of absolute tumor size, the Mann-Whitney test indicated significant differences between Groups A and B compared to Groups C and D: Day 7 p=0.012, Day 10 p=0.007, and Day 13 p=0.010.
6) In terms of tumor size changes from day 4, the Mann-Whitney test indicated significant differences between Groups A and B compared to Groups C and D: Day 7 p=0.023, Day 10 p= 0.006, Day 13 p= 0.012.
7) The Mann-Whitney U scores for tumor size changes were then used to calculate the odds that an animal from Groups C and D would have a larger tumor size increase than an animal from Groups A and B: 2.34 to 1 odds on day 7, 2.84 to 1 on day 10, and 2.86 to 1 on day 13.

The different statistical significances found on days 7, 10, and 13 are most likely the result of the tendency of this fast growing tumor to converge in size in all groups over time. This occurs because the faster growing (larger) tumors are eventually unable to acquire enough nutrients to support the same growth rate as the smaller tumors. The larger tumors are therefore forced to slow their fast growth at some point, and this allows the smaller tumors to demonstrate a relatively faster growth rate.

IV. CONCLUSION

The mechanism by which electrostatic fields exert influence inside a mammalian body is not currently understood. Only zero electric field can exist inside a static conductive object at equilibrium. However a living body is obviously much different than a static conductive object, and the differences should be the key to ultimately understanding the mechanism of influence.

For example, it is known that:

1. Far from being static, mammals are electrochemical systems that depend on countless, and constantly changing, electric fields for the simplest to the most complex cellular operations [24].
2. A mammalian body is not a simple electrical conductor. For example a human body is around 60% water by weight, but only about $1/3^{rd}$ of that is available as an electrically conductive volume since the rest is isolated within electrically insulated cells at any given time [25].
3. It is known that external electrostatic fields exert an electrically polarizing influence on a mammalian body [26]. The materials forming a living mammal are highly heterogeneous, and have a wide range of different permittivities and conductivities.

They also exhibit Maxwell-Wagner polarization; "When an electric field is applied to such material, the mobility of charge carriers, such as ions, migrating through the material can be significantly higher in some regions (for example a aqueous phase) compared with other regions (for instance a lipid phase). This inevitably leads to a build up of charge carriers at non-conducting boundaries and results in a non-uniform charge distribution in this region" [27].

Regardless of the mechanism, the degree of biological influence seen from these fields [1-19] (also see our companion paper "Static Fields: Possible Therapeutic benefit --- Possible Danger" in the current 2004 ESA-IEEE Joint Meeting Proceedings) has led us to believe that uncontrolled exposure to them may present a significant, and previously unrecognized, hazard. Endogenous electric charges and fields cause and control almost all of our cell operations, including DNA assembly. Any external force capable of influencing these natural charges and fields could result in a range of disease possibilities.

Humans are often exposed to intense electrostatic fields. These charges and fields are produced when two materials are rubbed together (or are in contact then separated). This commonly happens when our clothing rubs together or against another surface such as upholstery, etc. For example, Department of Defense Handbook #263 lists some typical measurements of electrostatic potential generated by various activities. Walking 20 feet across carpet can generate 35,000 volts at 10-20% relative humidity (RH), and 1,500 volts at 65-90% RH. Simple movement of a worker at a bench can generate 6,000 volts at 10-20% RH, and 100 volts at 65-90% RH. A worker moving around in a chair padded with polyurethane foam can generate 18,000 volts at 10-20% RH, and 1,500 volts at 65-90% RH.

Because these electrostatic charges are often very close to our bodies, the fields we encounter from them can be intense. For example, we found that rubbing a polyester blouse across a nylon bra can easily leave an electrostatic potential of several thousand volts on the fabric surfaces. If the fabric is a common 1 mm thick, a .01 m diameter charged area of just 1,250 V would expose the breast tissue to the same 200,971 V/m electrostatic field averaged by Group C in the present study. Under low humidity conditions these charges can stay in place on the garments, with the electrostatic field connecting to body tissue, for long periods of time.

The body of evidence presented by this, and previous studies, points to the importance of determining if these fields may be a factor in the increase in certain cancer rates in the United States over the past 30 years [28-35].

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