

Radiation In **Microgravity**

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Table I.

1. Introduction

Spaceflight results in unavoidable exposure of astronauts or experimental organisms to the complex space radiation environment while simultaneously subjecting the individuals to gravity unloading and other stresses such as vibroacoustic stimuli. A natural question is whether these independent environmental factors interact in significant ways to compromise the health of spaceflight crews or to significantly modify the outcome of biological experiments focussed on an independent variable. Access to time in space and the ability to control acceleration levels of spacecraft have been limited so important experimental data are limited. Despite these constraints there are data to suggest that exposure to microgravity may modify organisms' responses to radiation exposure and there are physiological changes during **spaceflight** that could plausibly explain such changes.

In this chapter I will first discuss the nature of the space radiation environment with emphasis on exposure to charged particle ionizing radiation, and then review results of selected experiments that have investigated modification of radiation responses by other variables, especially microgravity. Emphasis will be given to findings from animal experiments where the microgravity plus radiation interaction is most directly applicable to human physiology and organization will be by experimental protocol.

2. Characteristics of the Space Radiation Environment

2.1 Composition and Distribution

The radiation environment of space is extremely complex. Electromagnetic radiation from energetic **gamma-rays** and X-rays through ultraviolet and visible light to microwaves and radio waves all permeate the solar system and are dominated overwhelmingly by solar activity. Of importance for human health under normal shielding conditions inside spacesuits and spacecraft are the penetrating X-rays and gamma-rays emitted directly by the sun or resulting from the interactions of charged particles with spacecraft hardware (Bremmstrahlung). Superimposed on the electromagnetic spectrum are a myriad of subatomic particles such as **pions**, muons, electrons

and neutrons derived from solar and galactic sources or from interactions of nuclei with the **Earth's upper** atmosphere and spacecraft materials. Finally, and most importantly, there are large numbers of atomic nuclei of all elements from hydrogen through uranium which travel to relativistic velocities [31]. These nuclei are derived from galactic sources and the sun and, with the exception of protons (H nuclei), are collectively referred to as cosmic rays or high atomic number [Z] energetic (**HZE**) particles. Stellar synthesis can only support the production of elements through iron so that the **fluence** (incident particles per unit area) of elements above $Z = 26$ drops dramatically and represents the galactic cosmic ray (**GCR**) contribution derived from stellar explosions (novae and supernovae) **modified** by magnetic fields. The most abundant charged particles are protons (85% of **fluence**), followed by helium nuclei or alpha particles (13% of **fluence**); nuclei of higher atomic number make up the remaining 2% of the **fluence**. But these later nuclei contribute nearly 50% of the absorbed dose from particles with iron constituting the single greatest contribution to absorbed dose (energy absorbed per unit mass). This is measured in Gray, where $1 \text{ Gy} = 1 \text{ Joule/kg}$; **older** terminology used rad, where $1 \text{ rad} = 10^{-2} \text{ Gy}$ or 100 erg/g [33].

The **fluence** of GCR is isotropic except for shielding by the earth for spacecraft in low earth orbit. The flux (**fluence** per unit time) is generally constant except for long term variations due to the solar cycle. The **fluence** of solar particles is less constant and is significantly modified by the magnetic fields of the sun and earth. During solar flares the **fluence** may increase dramatically over hours or a few days and tends to localize in "flux tubes" which may or may not intersect the path of the earth. Solar flares consist primarily of relatively low energy protons which fail to penetrate the earth's magnetosphere at low latitudes due to magnetic focussing effects that divert them back into space. All of these particles penetrate to low altitudes above the earth's magnetic poles and down to regions of about 50 degrees of latitude as the particles can follow open field lines [31].

Generally speaking, low earth orbital spacecraft

receive little dose from cosmic rays due to their localization in low inclination orbits. Most U.S. spacecraft orbit at inclinations of 28.5 degrees, but some U.S. and most Soviet missions spacecraft have used 50 to 65 degree inclinations. Of all manned missions, only the Apollo lunar flights have left the earth's magnetic envelope for exposure to the "free space" cosmic ray environment.

The major contribution of radiation to manned spacecraft comes from protons and electrons confined to the trapped **radiation** belts (Van Allen belts) which occupy toroidal regions of space from altitudes above 500 km to several earth radii. They are distorted to lower altitudes at polar latitudes where they are said to form "horns", by the appearance of their **fluence** density in meridional planar sections. Due to asymmetries in the **earth's** magnetic field, there exists a region over the South Atlantic Ocean called the South Atlantic Anomaly (SAA) where the magnetic field is slightly weaker which allows particles to penetrate to lower altitudes than at other locations around the earth. Thus, for orbits at any altitude the **fluence** of trapped protons is maximum during traversal of the SAA. The SAA has been the main exposure source for most manned missions and is expected to be the major source of radiation for space station freedom [32].

3.0 Properties of Ionizing and Non-Ionizing Radiation

Non-ionizing electromagnetic radiation excites vibrational or rotational motions in molecules or promotes electronic transitions that, in turn, lead to chemical reactions. These interactions do not usually lead to strand breaks in DNA, but cause specific classes of lesions and elicit repair responses from living cells. The effect of ultraviolet light in causing pyrimidine dimers in DNA is perhaps the best known example [9]. By contrast, ionizing radiation extracts electrons from atoms in a relatively nonspecific pattern and leads to the production of highly reactive chemical species with unpaired electrons, *i.e.* free radicals. The most abundant biological molecule is water whose major ionization product is the **hydroxyl** radical. Such radicals may diffuse to nearby nucleic acids or proteins where they can cause or **lead** to strand breaks or the formation of adducts that are difficult to repair. Macromolecules may

also be ionized directly without water as an intermediate. The presence of oxygen can accentuate free-radical damage [33].

X-rays and gamma-rays are low linear energy transfer (LET) forms of radiation, which means they lose (transfer to the target) only a small amount of energy per unit of track length (LET = $-dE/dx$ expressed in **keV/micron**). For these radiation species small clusters of 1 to 3 ionization events are formed every micron or so. The large average distance between ionizations makes DNA strand breakage relatively inefficient. However, as the dose builds, the distribution of ionization events throughout the target volume becomes more uniform; the mean distance between events decreases such that macromolecule strand break probability rises [19]. An important consequence of this uniform pattern of dose deposition is that all regions of a biological target are exposed equally, and the large distance between lesions at low doses allows repair mechanisms to keep pace with damage formation [9].

3.1 Unique Properties of Charged Particle Radiation

Charged particles are capable of interacting with electrons as well as with nuclei of target atoms. Most interactions are with electrons, but fragmentation of primary and target nuclei is significant for thick targets, (human bodies and spacecraft walls) and results in the creation of multiple secondary particles. The consequence is that particles produce dense linear tracks of ionization with little change in direction. The LET is relatively high and follows the Bethe-Bloch relation as a function of range in the target; This means that $-dE/dx$ varies as the primary **particle's** charge squared divided by its velocity squared. This behavior is manifested as a so-called Bragg peak in which relative ionization versus track position rises sharply at the end of a particle's range. Radially, energy loss varies as the inverse square of the off-track distance, which is related to the range of scattered electrons or delta rays. However, delta-rays display structured energy deposition themselves leading to "**spurs**" and "**blobs**" of ionization emanating from primary tracks which "**thin down**" at the limit of their range [19,34].

There are several consequences of this structured energy deposition. First, concentrated doses are localized to small target regions surrounded by unaffected regions. For example, cells traversed by a heavy ion may sustain the equivalent of many hundreds of rads while neighboring cells are unaffected. Second, the mean distance between ionizations along the track is small so that multiple radicals or ionizations may be created on the scale of a few DNA base pairs leading to efficient strand breakage. This is true whether the macroscopic dose is large or small. Third, the concepts of dose and dose rate break down when single-particle, single target (cell) interactions are considered; the fundamental deposition event is from a particle moving at nearly the speed of light so that the time associated with traversal of a 3 micron cell nucleus is about 10^{-14} sec. Fourth, direct ionization of targets becomes more important than for photons and water and the relative role of active oxygen species in mediating damage is reduced *vis-a-vis* low LET radiation [19].

3.2 Risk Assessment

Risk assessments for charged particles have generally been based on the assumption that it is possible to identify a dose of low LET radiation, such as X-rays or gamma rays, which produces the same biological effect as exposure to a dose of charged particles. This has the convenience of allowing risks to be interpreted in the context of familiar hazards, such as medical diagnostic procedures. When effects **unique** to high LET radiation species are not present the relative biological effectiveness is often calculated: $RBE = (\text{dose of standard radiation}) \div (\text{dose of test radiation})$ at the same frequency of biological effect. The RBE for a particular endpoint (mutation, tumor incidence, survival) may vary substantially with dose and dose rate and is generally varies 2- to 3--fold with LET; that is, $RBE_{\text{endpoint}} = RBE_{\text{endpoint}}(\text{dose, dose rate, I, ET})$ so inferences must be carefully considered. Infinite RBE values would occur if an endpoint were only observed after high LET radiation exposure. The concept of dose equivalence is used to express the enhanced effectiveness of high LET radiation. Dose equivalent [Sievert (100 rem)] = absorbed dose [Gray (100 rad)] X RBE. When many endpoints and experimental

systems are involved in estimating risk for particular radiation species a combined RBE value is usually desired. This value, called the quality factor, is established by a panel of experts and is based on the body of published experimental observations. The National Council on Radiation Protection and Measurements (NCRP) is the organization in the **United States** usually charged with establishing risk estimates and exposure limit recommendations, and it establishes quality factors for its own use. The NCRP has determined that of all the potential health risks associated with exposure to space radiation, radiation-induced cancer is the dominant risk component for long-term space missions [10]. **Cataractogenesis**, mutation, damage to gametes, behavioral deficits, and other problems rank far behind.

For charged particle-dominated radiation environments a more useful approach than dose equivalent versus response for describing the kinetics of damage induction by radiation would be the use of **fluence** versus response or probability of response per unit **fluence** (action cross section, σ) relationships for particles of particular structures and energies. The dose is related to the **fluence** in the following way: Dose (Gray) = $1.6 \times 10^{-9} \times \text{LET (keV}/\mu\text{m}) \times \text{fluence (cm}^{-2})$. This method would allow the prediction of biological effects from mixed populations of particles in a natural environment by weighting the probabilities according to the abundance of the particles of a given LET (or charge state and energy range). High LET-unique endpoints would not be a factor and, for space missions, dose rate would not be an issue because of relatively low flux. This approach is currently under consideration by NASA [10].

Numerous biological effects have been examined resulting from charged particle exposure and these vary significantly with the details of track structure. Ground-based studies are carried out at particle accelerators (e.g. Lawrence Berkeley Laboratory's **Bevalac**) and provide data from biological systems as diverse as mammals, invertebrates, cultured cells, and microorganisms. Cell survival, cell transformation, **tumorigenesis**, cataractogenesis, mutation, chromosome aberration, gene expression, life shortening, developmental defects, and virus inactivation are the biological models most **fre-**

quently used [19]. Some of these well characterized laboratory systems have been examined in the **spaceflight** environment, but alternate species, media, incubation conditions, etc. have been developed to accommodate restrictions of **spaceflight** experimentation (e.g., pocket mouse and stick insect) .

4.0 Radiobiological Experiments in Space

The various **radiobiological** responses of organisms have been studied extensively in the laboratory and the results applied to human health issues. However, there is no satisfactory way to simulate the complex, low dose rate, omnidirectional, multi-component environment of space with its complicating feature of **microgravity**. Therefore, exploratory investigations and experiments to verify ground-based predictions for well-characterized model systems must be done **in space**.

Some of the earliest space experiments were **radio-biological**. These were of simple design and typically consisted of inert biological objects not likely to be gravity sensitive. Most used natural radiation exposures and compared data from the **microgravity** satellite sample with that from matched ground controls. Later, **inflight** radioisotope sources were used to enhance the frequency of the biological endpoint to detect negative or positive influences from "**space** flight factors". More recent experiments have also utilized on-board centrifuges as acceleration controls to examine combined radiation and microgravity as controlled independent variables. The sophistication of the more recent investigations has increased as the supporting flight hardware has improved. Table 1 summarizes the most conclusive **spaceflight** experiments which address the issue of radiation effects in microgravity. The details are discussed below in sections 4.2 and 4.3.

4.1 Exploratory Studies

A number of unmanned and manned spacecraft launched by the United States and the Soviet Union from the late **1950's** through the **1970's** carried biological specimens consisting primarily of bacteria, bacteriophages, fungal spores, **microalgae**, plant seeds, arthropod cysts or eggs, and some mammalian cultured cells. Most satellites were

placed in low earth orbits for several hours to two weeks allowing exposure to natural cosmic rays and trapped proton belts. **Controls** consisted of matched samples incubated in duplicate flight hardware subjected to vibration and acceleration profiles that simulated rocket launches. In these experiments the natural space radiation induced mutations and chromosome aberrations and effectively disrupted the normal development of invertebrate and plant embryos [24,32].

Results from exploratory experiments in **this** genre have been reviewed by Horneck [13] and Antipov [1]. They include findings from the Discoverer, NERV, Sputnik, Cosmos, Vostok, and Voskhod flights. The biological specimens included: T4, T7, ECHO 1, and PR8 viruses; *Escherichia*, *Clostridium*, *Bacillus*, and *Pseudomonas* bacteria; *Chlorella* algae; *Neurospora* fungus; *Arabidopsis*, *Nicotiana*, *Lactuca*, *Oryza*, *Crepis*, *Zea* and *Tradescantia* plant seeds and microspore; and *Artemia*, *Tribolium*, and *Drosophila* arthropods.

In this 1950 through 1970 time period access to particle accelerators (cyclotrons, **linacs**, synchrotrons) capable of generating heavy ions in the same energy ranges as cosmic rays was also very limited, so baseline dose versus response relationships (except for X-rays) were not well known for many systems. Vibration, acceleration, and noise, which could modify **radiobiological** responses, were not common clinical or scientific variables. They became operational issues in military aviation and manned space flight. Thus, a data-base of these and other uncommon environmental effects, which could potentially modify **radiobiological** responses, had to be developed *de novo* for each experiment [2].

4.2 Natural Radiation Exposures

For experiments utilizing the natural radiation environment to provide exposure, acquisition of large data-bases were always limited by the volume, mass and power constraints of life supporting flight payload hardware because biological sample sizes had to be large to detect rare genetic and cellular events. Correlations of biological responses with mission duration (proportional to dose) have large variability. These constraints persist.

A second generation of studies arose in which individual cosmic rays were correlated with **radiobiological** endpoints in space and in which in-flight acceleration controls were provided. The Biostack, **Biobloc** and **Exobloc** series of experiments used ray-tracing techniques to localize cosmic ray tracks to immobilized spores, seeds, and eggs sandwiched between nuclear track detector materials such as nuclear emulsions, cellulose nitrate, and **polycarbonate** plastic. In this way the rare cosmic rays (typically at 4 to **10/cm²•day fluence** for $Z \geq 6$) could be identified first and then followed by analysis of "hit" biological samples without wasting time and resources on "unhit" specimens. Details of experiments of this type with the brine shrimp, **Artemia**, the stick insect, **Carausius**, the nematode, **Caenorhabditis**, the pocket mouse, **Perognathus**, and the rat, **Rattus**, are presented below.

4.2.1 Experiments Using the Brine Shrimp, **Artemia salina**

Perhaps the most popular organism for space **radio-biological** studies is the brine shrimp **Artemia salina**. This organism can exist as a desiccated **gastrula** cyst for years inside an eggshell thereby requiring very modest life support provisions. Upon return to saline solution the shrimp recover and resume development. Following experimental irradiation or exposure to space, rates of hatching, developmental progression, and incidence of developmental anomalies have been recorded. The results from Cosmos 1887 and ten previous **spaceflight** experiments have been reviewed by **Gaubin** [11]. Rates of inactivation and anomaly scale with dose but no obvious influence of microgravity on radiation-induced lesions in **Artemia** has been reported. This may be due to the fact that the cysts were essentially dry and the shrimps' structures were rigid and gravity resistant. Therefore regulatory and repair processes based on soluble enzymes and other **biochemical** mediators were probably not active.

4.2.2 Experiments Using the Stick Insect, **Carausius morosus**

The stick insect **Carausius morosus** has been used on

several **spaceflights** to investigate the effects of gravity and charged particle radiation on development. These insects **undergo** incomplete metamorphosis in eggs within 75 to 105 days at 18 to 22 degrees C. This property permits eggs to be integrated into parallel layers of nuclear track detectors such that ray tracing methods can be used to identify the eggs "**hit**" by HZE particles. This hardware arrangement was integrated into "**Biostack**" experiments on several missions, most notably **Spacelab D1** and Biocosmos 1887. In each of these missions eggs in five developmental stages experienced 7 to 12.6 days of microgravity and were separated into groups "**hit**" or "**unhit**" by HZE particles during flight and an "**unhit**" ground control. On the **D1** mission some eggs were also incubated inflight in a 1XG centrifuge. Hatching rate from eggs, growth rate, and frequency of developmental anomalies were measured. The anomalies were primarily fused, tilted, or stunted segments of antennae, abdomens, and legs [13,14,20,27,28].

The results from the **D1** mission indicated that both microgravity and HZE particle strikes disrupted development in a stage specific fashion. Hatching rate was reduced and the frequency of anomalies was enhanced. Results from the in-flight centrifuge control insects indicated that **microgravity** and HZE effects were synergistic, suggesting a role for gravity in the ability of embryos to manage or repair damage by HZE particles or vice versa [27]. The sample sizes were limited on **D1** so a second experiment on Cosmos 1887 (without the centrifuge) was performed to improve the statistical base, especially for sensitive stage embryos; approximately 4,300 eggs were flown. Hatching rate was reduced significantly by microgravity exposure (81% control vs 50% flight for stage II embryos) and a slight additional impairment (additive, not synergistic) was associated with HZE hits. However, developmental anomalies were substantially increased in "**hit**" eggs, especially in stage II embryos where "**unhit**" flight samples showed approximately a 2% rate of anomaly and flight "**hit**" samples showed a 17% rate of anomaly [28]. This compares with ground control rates of less than 0.15% on both **D1** and Cosmos 1887 flights. These findings are provocative with respect to the apparent interaction of gravity and radiation on sensitive, stage-specific, developmental processes. They raise the possibility that microgravity

and radiation may interact at the level of repair processes, but no specific mechanisms have been proposed.

4.2.3 Experiments Using the Nematode, *Caenorhabditis elegans*.

To isolate cosmic ray-induced mutations, the microscopic nematode, *Caenorhabditis elegans*, was flown in 1992 on shuttle **STS-42**, as part of the Biorack investigations on the International Microgravity Laboratory # 1, Spacelab mission [22,23]. Approximately 60 **autosomal** recessive **lethals** in a 350 gene region, and 12 mutations in the muscle protein gene, *uric-22*, were isolated from animals held in a dormant larval state or growing in logarithmic culture. Dormant animals were incubated in a laminated assembly next to CR-39 plastic nuclear track detectors so that a ray tracing procedure could be used to correlate specific cosmic rays with specific mutants. An extensive series of ground experiments using accelerated charged particles provided an interpretive framework for these mutants whose structural properties were being analyzed at the molecular level. Unique or characteristic features of authentic cosmic ray-induced mutants were identified [22]. The experiment design did not permit an explicit test of gravity effects on mutation kinetics because of the low **fluence** of particles in space, the small responding fraction of animals, and the requirement for low temperature to immobilize animals for ray tracing which precluded on-board centrifugation. However, in complementary experiments development and chromosome behavior (segregation and recombination) were measured in rapidly growing cultures as a function of gravity level and found no obvious dependence on gravity [22]. The main findings from this experiment were the correlation of molecular structures with authentic cosmic ray "hits", and the observation of normal chromosome mechanics and development for two successive generations in an animal exposed to microgravity.

4.2.4 Apollo Lunar Flight Experiments with the Pocket Mouse, *Perognathus*, and Associated Investigations.

Experimental animals carried in Apollo 16 and 17 spacecraft were unique in that, along with Zond 5 and 6 lunar flyby missions, they were exposed to radiation

fields outside the earth's geomagnetic envelope, and they completely traversed the trapped proton and electron belts. The **fluence** of particles on these missions included the full energy range for GCR. In earlier Apollo missions crew members observed light flashes that were caused by passage of cosmic ray particles through their dark-adapted retinas [18]. The possibility of behavioral decrements and destruction of nervous tissue, including photoreceptors, increased interest in HZE effects. Severe constraints on mass, volume, and power limited **radiobiological** experiments to passive tests [18].

The Apollo Biostack I and II experiments provided correlation of cosmic ray tracks with inactivation of several biological objects. The experiments assessed the efficacy of specific ions and their proximity to bacterial spores, plant seeds and **radiculae**, brine shrimp cysts, eggs of the flour beetle, *Tribolium*, and the stick insect, *Carausius*. The three animal systems proved to be most sensitive when assayed for inhibition of hatching and the occurrence of developmental abnormalities. These tests, and another experiment (MEED) which provided exposure of organisms to the natural solar light spectrum through quartz windows, did not address interactions of other **spaceflight** factors [18].

One mammalian experiment was included on the 13 day Apollo 17 flight. In this "**Biocore**" study, five pocket mice (*Perognathus longimembris*) were fitted with plastic nuclear track detectors under their scalps to provide a comprehensive map of cosmic rays passing through brain and eye tissues and to assess histologically any damage; in addition, a comprehensive study of all body tissues complemented the main objective [12]. Animals were housed separately in cylindrical containers positioned in a relatively heavily shielded location in the command module; four of the five mice survived the flight. When dosimetry and the elaborate stereotactic measurements were completed, 71 cosmic rays of known LET ($Z \geq 6$) were traced into head structures and the direction of deceleration was established for 39 of the tracks. Only five particles intersected retinal tissue of the four mice. Histological examination of serial sections failed to detect lesions in eye or brain tissue which correlated with cosmic ray tracks. The absence of

microlesions [34] does not prove that cells were not damaged, but it argues against catastrophic destruction of columns of CNS cells from ions of intermediate atomic number. These **Biocore** results are illustrative of the difficulties in assessing biological effects of naturally occurring cosmic rays.

4.2.5 **Biocosmos** 110 Experiments with Dogs and Microorganisms.

One early space biology experiment which set the stage for mammalian studies was the 22-day Cosmos 110 mission launched in 1966 using an unmanned 2.3m diameter spherical Voskhod spacecraft which flew at 51.9 degrees inclination in a 187 by 904 km orbit [2,17]. The satellite contained two terrier dogs (Veterok and **Ugolek**) kept in separate compartments within the cabin, which were maintained with the human life support system. The animals were largely immobilized, were fed by gastrostomy tubes, and were monitored extensively with physiological sensors. Veterok was given a radiation protestant drug intravenously during the mission, and two additional dogs were treated similarly and simultaneously on the ground as controls. The dogs were recovered in "good conditional but experienced significant calcium loss and had impaired movement for 8 to 10 days. **Radiobiological** endpoint data in the dogs was not available, but the measured dose was 10.5 cGy (approximately 0.5 **cGy/day**) which was due to protons which the spacecraft encountered as it transitted the lower Van Allen belt at apogee.

Accompanying the dogs were a variety of other organisms including a **lysogenic** strain of *E. coli* K12 (λ) used with and without an **aminothioliol** radioprotectant, plant seeds, *Tradescantia* microspore, and an intact plant. Phage induction was significantly enhanced in flight and suppressed by the radioprotectant. *Tradescantia* microspore showed a variety of chromosome aberrations and **mitotic** disturbances while mutation in *Chlorella* was not different from ground controls [2].

4.2.6 **Biocosmos** 782 and 936 Experiments with Rats.

Interest in effects of cosmic rays on mammalian central nervous systems and eyes led to investigations with rats aboard the Soviet **biosatellites** Cosmos 936 and 782.

Cosmos 936

Cosmos 936 was launched 3 August 1977 for 18.5 days in a 224 x 429 km orbit at 62.8 degrees inclination and 90.7 min period. Thirty specific pathogen free male Wistar rats, fed on a paste diet were flown. Twenty were exposed to microgravity and ten were incubated in an onboard **1.05XG centrifuge**. Five rats from each group were used for **radiobiological** studies and were sacrificed after 25 days of recovery on the ground. Parallel ground control rats were utilized with a 4-day lag time, they were subjected to acoustic noise, mechanical vibration, and acceleration profiles simulating those during launch and recovery. For example, launch noise was 110 dB for 10 min with 50-70 Hz vibration at **0.4mm** amplitude followed by acceleration for 10 min to 4XG for a 7 min plateau period. Reentry and recovery involved a 5 min acceleration to 6XG for a 3 min plateau, followed by a 10 msec pulse at 50XG [16,29].

Because of the postulated existence of "**microlesions**" in the highly organized retinal tissues, the rats' eyes were fixed and examined with light and electron microscopy for linear tracks of damage in the retinas. This examination was prompted by results of accelerated particle experiments at the **Bevalac**, using neon ions on C57 Black mice and pocket mice, which identified microscopic lesions in cells of the eye and nervous system. HZE particle **fluences** of 1.75 **particles/cm²•day** ($Z \geq 3$ and range in **lexan** $\geq 180\mu\text{m}$) were measured on Cosmos 936 rats using **lexan** and **nitrocellulose** track detectors. Total doses of 4.24 to 5.23 mGy were measured using TLDs. Neutron **fluences**, stopping protons and nuclear decay stars were also quantified using activation foils and nuclear emulsions.

Necrotic retinal cells were swollen and had dense nuclei; membrane debris in widely-scattered regions of

the outer retinal layer was observed, especially in the rods. Occasional. **phagocytic** activity in pigmented **epithelial** cells was seen. **Macrophages** were not present near damaged areas; but the rats were not sacrificed until 25 days after landing. Findings from flight centrifuged and microgravity-exposed rats were similar with respect to retinal lesions. Necrotic lesions were not observed in the ground control material.

Cosmos 782

Similar necrotic cells were observed in rats flown at OXGon the 25 November 1975 Cosmos 782 mission (19.5 days in a 226 X405 km orbit at 62.8 degree inclination) with slightly higher **fluence** of HZE particles (4.05 **particles/cm²•day**). One difference on this mission was that a subset of animals was sacrificed several hours after recovery and in these animals macrophages were seen near lesions. This was interpreted as evidence of passage of HZE particles through retinal tissues with the formation of **microlesions**; there was no evidence of a gravity interaction [16].

4.3 Artificial Radiation Exposures

It is clear from the preceding section that deleterious radiation effects from natural exposures are very rare and mitigate against interpretation of gravity interactions for practical reasons. A different strategy in performing such experiments is to orbit samples in the trapped proton belts to provide exposures of 1 - 5 Gy, but this would require development of a dedicated unmanned spacecraft. Alternatively, onboard sources of radiation from radioisotopes or irradiation just before or after flight could be used to determine whether flight conditions, especially **microgravity**, modify **radiobiological** responses. The latter strategies have been applied in manned and unmanned **spaceflights** and are discussed in this section.

4.3.1 The S4 Experiments on Gemini III and XI Using Blood Cells and Fungi.

An important early experiment to address **radiobiological** responses in human cells was conducted in 1965

and 1966 on Gemini spacecraft. The S4 experiment was flown twice to detect possible interactions of micro gravity and radiation on human nucleated blood cells [4,5]. On the second mission bread mold spores were also included to provide independent confirmation of genetic effects [6,8].

The Gemini III mission (23 March 1965) consisted of only three orbits (0.20 days, 32.5 degrees inclination, perigee of 161 km and apogee of 224 km) and received 0.20 to 0.45 mGy radiation, measured with thermoluminescent detectors (TLD), depending upon location. The 2.97 day Gemini XI mission (12 September 1966) flew in an elliptical orbit (29 degrees inclination, perigee of 159 km, and apogee of 298 km) except for two orbits with an apogee over Australia of 1370 km and a perigee of 298 km for a rendezvous and docking maneuver which took the spacecraft briefly into the lower Van Allen proton belt. The total TLD dose was 0.23 to 0.39 mGy depending upon location.

Fresh blood samples were placed into 3mm thick glass chambers which astronauts could activate by moving them to an irradiation area in the shielded flight hardware where they were exposed to a series of doses of 0.7 MeV B-particles from ^{32}P ; exposures ranged from 0 to 1.74 Gy. Single and multiple break chromosome aberrations were measured in leukocytes held in the G_1 phase of their cell cycle. Survival and mutations at the *ad-3A* and *ad-3B* loci were measured in *Neurospora conidial* spores in suspension, or held on millipore filters and irradiated like the leukocytes but at exposures up to 144 Gy. All results were compared with simultaneous ground control responses. No differences between flight and ground samples were detected for mutation or survival in *Neurospora* irradiated on filters, or in leukocytes examined for multibreak aberrations. Differences between flight and ground samples for single break leukocyte aberrations were observed on Gemini III but were not seen again on Gemini XI. It was suggested that sampling error may explain the earlier results. Differences in survival levels and mutation rates for suspended fungal spores which were metabolically active, unlike inactive filtered spores, could be explained by anoxia. Thus, no obvious radiation + gravity interaction was detected.

4.3.2 **Biosatellite II.** Microorganisms, plants and insects irradiated in orbit with gamma rays.

On September 7, 1967 the unmanned **Biosatellite II** was launched into a 190 mi circular orbit at 28.5 degrees inclination for 45 hours, after which it was recovered (one day earlier than planned) in midair over the Pacific ocean and its biological payload, consisting of microorganisms, plants, and insects, was sent to Hawaii for analysis. Several of the **radiobiological** experiments provided known doses of gamma rays to the specimens in orbit, prior to launch, and to matched ground controls to detect the effect of gravity on radiation-induced genetic and developmental lesions. The radiation source was ^{85}Sr in a tungsten holder which could be opened and **closed** by ground command. The source had an activity of approximately 1.2 Ci and produced 0.513 MeV gamma rays measured by thermoluminescent detectors. The findings provided evidence for a modifying effect of gravity on the radiation effects. Environmental control was acceptable except for formaldehyde vapor (0.8 to 2 ppm), which exceeded the design specification of 1 ppm and could conceivably have contributed to mutagenesis, especially in the *Drosophila* experiment.

Experiment P-1135 investigated the induction of P22 virus production from a **lysogenic** strain of *Salmonella typhimurium* BS-5 (P22)/P22, and lambda phage from *E. coli* C-600 (lambda)/lambda [Mattoni, et al. In: 30]. Because of early reentry of the satellite, only the **Salmonella** experiment could be performed. Small **aliquots** of 120 cells/ml in broth were placed near the ^{85}Sr source and received 0 to 16.30 Gy exposures. After flight, differences in cell density and growth rate were detected between ground and flight samples, and a slightly increased resistance to **γ -rays** in the flight samples was observed. The critical measurement of induced phage production per viable cell immediately after recovery was also measured. A 38 to 42% decrease in yield in the flight samples relative to ground controls which was significant ($P < 0.02$) for doses of 2.65 and 6.45 Gy and at the $P < 0.06$ level for 16.30 Gy. Thus exposure to microgravity appears to show an antagonistic effect on radiation induction of phage.

Experiment P-1037 investigated mutagenesis in heterokaryon **conidiospores** of the bread mold, *Neurospora crassa* [de Serres, et al. In: 30]. Mutation at the *ad-3A* and *ad-3B* loci were measured from stationary culture cells filtered onto **nitrocellulose** filters under conditions where they were metabolically inactive. The **conidiospores** received exposures from 3.40 to 36.00 Gy in space. No significant differences between flight and ground control cells were observed for point mutations or for chromosome deletions. These results agree with those of the previous S4 experiment on Gemini XI for cells held on filters. This is in contrast to the metabolically active cell suspension cultures on Gemini where an antagonistic effect of **microgravity** on mutagenesis was measured.

Experiment P-1079 examined mutagenesis and genetic recombination in the parasitic wasp *Habrobrachon juglandis* Ashmead = *Bracon hebetor* (Say) as a function of **γ-ray** dose [Von Borstel, et al. In: 30]. Young wasps were placed in culture chambers where they received 20.00 Gy just before launch or in-flight doses of 0.07 to 24.25 Gy. (Brine shrimp cysts and yeast cells were also flown in conjunction with this experiment but showed no differences between flight and ground samples for development and recombination, respectively.) Analysis of sperm from XO males after recovery revealed no significant difference between flight and ground samples for dominant lethality, recessive lethality, or partial sterility; but there was a slight increase in fertilizing ability. A higher spontaneous rate of recessive lethality was observed, but it was reproduced by vibration on the ground. **Oocytes** from XX females in the first stage of meiotic **metaphase** in flight showed a reduction in post fertilization viability from 0.89 - 0.93 to 0.49 - 0.56, whereas other stages of **oogenesis** revealed no differences except for a modest increase of fertility. This **oocyte** effect correlated with its position in the spacecraft, but not with radiation dose or microgravity, suggesting that other stress such as vibration may have been involved. No effect on recombination in females was observed between the *lemon*, *honey*, and *cantaloupe* genes on chromosome I. Two additional observations suggested increased longevity of flight females, and disorientation of males leading to decreased male mating behavior in flight, which were not affected by radiation. Thus ,

little or no interaction between **microgravity** and radiation was observed with respect to production of inherited genetic changes. Only a slight enhancement of sperm and egg fertility was detected.

Experiment P-1159 used *Drosophila melanogaster* larvae, irradiated with 8.32 Gy of γ -rays, to investigate **chromosomal** alterations and mutation in germ cells and somatic tissues [Oster. In: 30]. One strain of flies of a specific genotype was used to detect alterations in karyotype in cerebral ganglion cells of adults after being irradiated as larvae (P_0 generation). Other strains, including Ring-X and multiply marked flies enabled the detection of point mutations at the *dumpy* locus, recombination, alterations in sex ratio (X-linked lethality), translocations, and non-disjunction or loss of chromosomes following breakage in the F_1 through F_6 generations. The frequency of sex-linked **lethals** ($0.71 \pm 0.19\%$, $N = 1961$ VS $0.35 \pm 0.14\%$, $N = 1724$) and recombination between X and Y chromosomes ($0.55 \pm 0.17\%$, $N = 2005$ vs $0.29 \pm 0.09\%$, $N = 3412$) was increased in flight samples when compared with ground controls. A rare finding was the appearance of seven somatic translocations in the flight flies both with and without radiation. Normally such events are very rare. This surprised the authors who further noted that these translocations were not correlated with radiation dose.

Experiment P-1160 used both pupae and adult *Drosophila melanogaster* [Browning. In: 30]. Four types of flies of complex genotype were irradiated with 40.00 Gy just before launch or with 14.32 Gy in orbit. First, females were mated just **prior** to flight with males of a complementary genotype so that a homogeneous population of target sperm could be used to identify recessive lethal mutations and translocations (in the F_2 generation) and nondisjunction of the Y chromosome (in the F_1 generation). Second, males were used for matings **postflight** to detect induced recombination in sperm (normally absent), as well as X chromosome **lethals** and **translocations**. Third, young males were irradiated preflight and allowed to mate during flight with complementary virgin females to detect alterations in rejoining of broken chromosomes. Finally, late third instar larvae and prepupae were irradiated inflight, and males were bred to

females to detect X-linked recessive **lethals**.

Recessive lethal mutations in sperm were observed at a slightly elevated frequency ($P = 0.05$) in flight samples over ground controls, but the difference was matched by rates observed in ground controls which recycled internal satellite gases. This implicated an effect from formaldehyde and **glutaraldehyde** fixatives used in other experiments. No significant mutation differences were observed at five visible loci (*alp*, *bw*, *st*, *y*, and *p^p*), or the loss of dominant Y chromosome markers *y⁺* and *B*. Several translocations were detected from pupal samples and a slight elevation in recessive lethal mutation was seen in pre-irradiated males when compared to pooled controls, but control responses suggested that vibration may have played a role. Taken together, these observations did not indicate significant interaction between microgravity and radiation for modifying chromosome behavior or structure.

Experiment P-1039 utilized the flour beetle, *Tribolium confusum* Duval, to study a particular radiation-induced syndrome of abnormal development [Buckhold, et al., In: 30]. Damage to pupal cells in a well localized region caused a misproliferation and deformation of spikes on the membranous wings of the beetle which prevented closure of the overlying **elytra**, a feature easily measured. Seven hundred twenty pupae were flown, and they received either no dose or were preirradiated with 13.50 Gy of 180 keV X-rays to bring them into the appropriate dose range for the flight. They were placed into the spacecraft as 19 to 27 hr old pupae; half would be shielded and half would be irradiated in orbit with an additional 7.55 or 9.69 Gy of γ -rays. The flight irradiation significantly ($P < 0.025$) enhanced the incidence of wing abnormalities of flown beetles ($44.8 \pm 3.2\%$) over ground controls ($29.9 \pm 3.0\%$) for pupae receiving approximately 23 Gy. However, After postflight vibration tests and repetitions of ground control procedures, it was concluded that variations in circadian rhythms and vivarium lot differences, but not vibration effects, probably accounted for these results. Thus, it appears again that microgravity and radiation do not interact.

Experiment P-1123 used the radiation sensitive

Spiderwort plant, *Tradescantia* clone 02, to measure inactivation of pollen, microspore death, spindle defects, somatic mutation in heterozygous petals and stamen hairs from blue to pink or to colorless, as well as stunting of stamen hair-s [Sparrow, et al., In: 30]. Thirty two young plants bearing several flowers each were obtained from **axillary** cuttings and rooted. They were placed in nutrient tubes, irradiated with 2.23 Gy in orbit, and examined postflight. *Tradescantia* is very sensitive to radiation and showed high frequencies of radiation-induced changes. Pollen abortion occurred at 66% in irradiated flight samples versus 48% in ground controls. Unirradiated flight and ground samples had a spontaneous pollen abortion incidence of 37 to 39% (this high rate is normal for clone 02). Stunting of stamen hairs occurred at 26.6% for flight irradiated plants versus 12.9% for ground controls; unirradiated plants had 10.1 to 10.5% spontaneous abortion rates. These responses, as well as altered nuclei and microspore death, showed clear synergism between radiation and microgravity. By contrast, hair color mutation was clearly antagonized in microgravity with flight irradiated samples showing a 4.4% incidence when compared with a ground control rate of 7.3%. Unirradiated samples had spontaneous mutation rates of 0.2 to 0.3%.

4.3*3 Cosmos 605 and Cosmos 690 Matched Flights with Rats ± Gamma Irradiation

The most direct mammalian experiment to date which investigated the potential interaction of radiation and microgravity was performed with male Wistar rats on two Soviet satellites, Cosmos 605 and Cosmos 690 [17,25]. The rats were individually housed in cylindrical wire mesh cages 20 cm long by 10 cm in diameter and were fed pelleted food (carrots and beets). Cosmos 605 was launched on 31 October 1973 (21.5 days, 62.8 degrees inclination, 214 km perigee and 424 km apogee) and orbited 27 rats which served as a microgravity control with only low dose radiation exposure from the natural environment. Cosmos 690 was launched on 22 October 1974 and orbited 15 rats (20.5 days, a similar orbit of 62.9 degrees inclination, 223 km perigee and 389 km apogee). It included a 320 ± 38 Curie ^{137}Cs 0.661 MeV gamma radiation source which provided an 8 Gy exposure over 24 hours

beginning on flight day 10. Filtering of the source, contained in a tungsten sphere with a collimation cone opening, provided a uniform exposure $\pm 10\%$. Simultaneous ground control studies using rats from the same **vivarium** lots rats were conducted for both missions. Most analyses were histological or biometric, but some enzymatic and transplanted studies were performed. Most organ systems were studied for alterations due to **microgravity** or radiation exposure. It was concluded that radiation and **microgravity** combined led to decreased recovery rates for various reversible alterations induced by **microgravity**, but there were no significant effects on the development of radiation-induced lesions. The results for selected organ systems are summarized as follows:

Testes: Examinations of testes from both Cosmos flights and ground control rats demonstrated no effects of **microgravity**. The pattern of spermatogenesis, frequency of intermediate cell types for gametes, and the histology of seminiferous tubule components was unaffected. Testes weight decrease in irradiated animals was the most dramatic effect, but it was unaffected by **microgravity**.

Hematopoietic system: Bone marrow and lymph organs of Cosmos 605 rats showed "a slight inhibition of **erythropoiesis** in bone marrow and spleen, significant involution of the **thymus**, marked **hypoplasia** of lymph tissue of the spleen and, to a smaller extent, lymph **nodes**" [25] due mostly to reduction of lymphocyte populations. The same responses were affected by irradiation. Histological examinations of bone marrow of Cosmos 690 flight and simulation rats on

. . the second post-experimental day (12th post-radiation day) revealed well developed **aplasia** with discrete foci of **hemopoiesis**. The pattern of hemopoietic changes was typical of radiation induced bone marrow lesions. However, an exposure of animals to weightlessness influenced the after effect; on the 27th postflight day (37th post-radiation day) the recovery of the hemopoietic tissue was delayed compared with that in ground-based simulation rats. Thus, bone marrow showed radiation-induced changes, and weightlessness affected the course of reparative processes in bone marrow. [25].

Hematopoietic potential was also examined by transplantation of marrow cells to spleens and marrow of lethally irradiated recipients (colony formation assays). Cosmos 690 and control rats showed a reduction to 13-17% of normal potential for the **CFU-spleen** test, and to 40-50% for the **CFU-marrow** test as expected for the 8 Gy exposure. In spite of the diminished number of stem cells in bone marrow of flight rats, their differentiation pattern did not change. The Cosmos 690 animals showed a trend for enhanced erythroid potencies of stem cells transplanted to marrow which may be explained by the more rapid recovery of the erythroid precursor population known to follow irradiation. Thus the qualitative aspects of the recovery may be slightly affected by the combination of microgravity and radiation.

Central Nervous System: There were no effects from any of the treatments except some **"morphological signs of enhanced functional activity"** in the hypothalamus and pituitary of Cosmos 605 animals which was absent in Cosmos 690 rats [25] suggesting a small modifying influence of radiation.

Heart, Liver and Kidneys: Cosmos 690 specimens exhibited changes in four cardiac enzyme levels consistent with an alteration in carbohydrate metabolism and stimulation of lipid utilization pathways. Some lipid accumulation was noted in the liver along with some polymorphism in hepatocyte nuclei. Cosmos 605 rats showed no such changes, and the kidneys were unaffected in animals from both flight.

Muscle: Muscle mass loss in hind limbs was a feature of all flight animals, and lactate dehydrogenase **isozyme** levels showed variations. **"Inflight** irradiation aggravated weightlessness-induced changes in skeletal muscles and led to a delay in reparative processes and incomplete structural restoration of muscles" post flight [25]. This effect was localized in muscles showing **microgravity-induced** pathologies and is probably a consequence of slowed connective tissue resorption during recovery which, in turn, delays **myofibril** regeneration.

Thus, responses of tissues sensitive to **microgravity**, radiation, or both, were obtained and one unifying concept emerged: *recovery from a pathological condition*

induced by either radiation or **microgravity** may be delayed by the presence of the other environmental factor. Data from **hematopoietic** tissue are probably the most significant.

4.3.4 Cosmos 368 Experiments with Microorganisms Irradiated on the Ground

The Cosmos 368 satellite was launched 8 October 1970 for 6.0 days in a 411 X 211 km orbit at 65 degrees inclination. A variety of microorganisms and plant seeds were studied for any potential interaction between radiation and microgravity [32]. Samples were irradiated with a series of doses of gamma rays either immediately before or immediately after flight and results were compared with those from ground controls. Doses to 1,600 Gy were employed. Diploid and haploid yeast in suspension or on agar showed no obvious effects of microgravity with respect to colony formation or growth after one to four generations. Similarly, the hydrogen bacteria *Hydrogenomonas eutrophus* (now classified as *Alcaligenes*) in suspension showed no perturbations with respect to plating. Air-dried chick pea and lettuce seeds were scored for chromosome aberrations and meiotic defects (anaphase bridges) with no obvious interference from microgravity. Only a slight difference in **rootlet** growth and **catalase** levels could be ascribed to **spaceflight** factors.

4.3.5 Preirradiation of DNA repair deficient yeast flown on Spacelab, STS-42.

The design of this Biorack experiment was to **pre-irradiate** yeast cells with a series of known radiation doses, incubate them for one week with and without gravity to allow repair, and observe recovery from radiation-induced damage from measurements of colony forming ability [26]. *Saccharomyces cerevisiae* cells in stationary phase were filtered onto a monolayer at $5 \times 10^6/\text{cm}^2$ and held on supporting agar blocks. They were irradiated preflight with X-rays at five doses from 0 to 1.40 Gy and held at

4 degrees C for transport, launch, and recovery. In flight, the yeast were placed in **Biorack** incubators at 22 and 36 degrees C for seven days. Matched cultures were incubated in the ground control (**1XG**) **Biorack**. The strain used bore a temperature sensitive allele of the radiation sensitive mutation (**rad-54-3**) which is defective in repair of DNA double strand breaks at the restrictive temperature (**36°C**), but has normal repair at the permissive temperature (**22°C**). Measurements of colony forming ability versus dose showed that the survival fraction of flight samples incubated at the permissive temperature was approximately two-fold lower than ground samples which were independent of dose. Restrictively grown cells were insensitive to gravity and did not repair their **X-ray** induced damage; these cells served as a control for unexpected effects of **spaceflight**.

The conclusion was that the repair pathway for radiation-induced DNA double strand breaks is sensitive to gravity levels. These results are important for two reasons. First, a biochemical pathway was identified in a eukaryote which is affected by gravity; this could help to identify a mechanism at the molecular level. Second, they show that microgravity effects may be manifested at the level of single independent cells under conditions where external environmental effects (e.g., convection mediated mass transport of oxygen or nutrients) were minimized.

5.0 Possible Mechanisms for Microgravity and Radiation Interaction.

There is no substantive influence of gravity on the physical deposition of energy in biological targets. This process is over in less than a picosecond for heavy charged particles, and the relative magnitudes of electrostatic or nuclear forces to gravity are enormous. Once ionizations are produced in a target, however, chemical reactions (e.g., free radical attacks) ensue whose rates are limited by diffusion and convective mixing [19]. It is these chemical processes that are susceptible to gravitational influences. Though **intracel-**

lular convection may be overwhelmed by specific transport mechanisms and the cytoskeleton, convective mixing in suspended cells is critical for exchange of oxygen, carbon dioxide, and nutrients which regulate their metabolism.

Damage to biological macromolecules can be repaired by enzyme **complexes whose expression** can be **induced** by radiation exposure, hyperthermia, anoxia or exposure to heavy metals; Examples are the so-called SOS response (coordinated **recA** and **lexA** mediated gene expression phenomenon) in bacteria and the heat shock (stress protein induction) responses in most organisms [9,19,21]. The repair often requires ATP and is therefore tied to the metabolic state of the affected cells.

Direct control of repair gene expression is also likely. Results from a recent MASER sounding rocket experiment with human **epithelial** cells indicated that **microgravity** could alter the expression of protooncogenes **c-fos** and **c-jun**, which act to regulate cell proliferation and differentiation [7]. **Cogoli et al.** (*cf.* **Gmünder and Cogoli**, this volume) have also shown that differentiation and proliferation of immune cells are rapidly modulated by exposure to microgravity. Many of the enzymes involved in the synthesis of new DNA during cell proliferation have repair roles as well [9,19]; so repair capacity may well be coupled to these functions. The findings of **Press et al.** [26] may represent a first step in identifying the nature of gravity's action on a DNA repair pathway for double strand breaks in yeast.

A variety of fluid redistribution effects and hormonal responses occur in microgravity (*cf.* this handbook) which may, in turn, influence, cellular damage induction and repair systems directly, or by controlling the state of oxygenation and hydration of tissues. Another indirect effect of microgravity may occur through modification of circadian rhythms. Radiosensitivity of intestinal crypt cells and bone marrow cells follows a circadian rhythm [15,33]. These responses are thought to reflect entrainment of cell cycles with diurnal rhythms; radio-sensitivity correlates with the cell cycle and the corresponding state of DNA, which is less protected by proteins from free radical attack during replication.

Thus, a variety of direct and indirect effects of gravity unloading could, in theory, modify cellular radiosensitivity and repair.

6.0 Summary and Conclusions.

A variety of experimental approaches has been used to address the effects of radiation in space. Many investigators have attempted to capture rare cosmic ray interactions *in situ*, whereas others have employed standardized sources of low LET gamma rays and electrons (Beta particles) and have varied the gravity levels systematically. All research teams have had to adjust to the frustrating constraints of spaceflight experimentation, which impose limits on protocols that would be unacceptable to laboratory workers on the ground. In spite of these constraints, the efficacy of space radiation to induce genetic and developmental lesions has been demonstrated clearly. Further, there are statistically significant and reproducible differences in the incidence or severity of radiation-induced lesions as a function of gravity.

Several unifying trends emerge. First, the magnitude of microgravity effects on **radiobiological** endpoints is small and almost never exceeds a 2-fold difference either higher or lower than controls. Second, metabolically active or developing systems are much more likely to be affected than inactive systems. The process of development, may amplify small differences in embryonic tissues into large observable differences in **the morphology** of older organisms. Third, the direction and magnitude of radiation or microgravity effects was specific to the biological feature or response measured (endpoint), even when two endpoints were measured in the same sample; e.g., reduction of mutation and enhancement of pollen abortion in *Tradescantia*. Fourth, radiation and microgravity effects are stage specific in developing cellular systems. Thus, nonspecific effects on all cells are not seen at all times. Finally, those genetic lesions which seem most likely to be affected are those exhibiting **chromosomal** breakage. This in turn suggests a role for **cytoskeletal** elements in conferring microgravity sensitivity as these protein complexes are required to move and align chromosomes and enzymes which ligate broken strands. Many of these processes **require** ATP;

inappropriate shunting of energy into different cellular compartments, as a consequence of gravity-unloading, could also explain some of the phenomena. To understand the mechanisms of these interactions, more carefully controlled experiments are needed which employ sufficiently large statistical samples and precisely defined endpoints.

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Table I

Summary of Representative **Radiobiology** Experiments in Space

Mission(s) [Experiments]	Organism (Common Name)	Endpoint(s) Measured	Exposure Mode ^a	Flight Accel. Control Yes/No	μ Gravit. Inter- action + - 0 ^b
Cosmos 1887 Apollos Spacelabs Others	Brine shrimp	Hatching, Development	Natural	0	0
Spacelab D1 IML-1 Cosmos 1887	Stick Insect	Hatching, Development, Growth	Natural with Ray Tracing ^c	Yes & No	+ ^d
IML-1	Nematode	Mutation, Development	Natural	Yes & No	o
Apollo 16 Apollo 17	Micro- organisms	Sporulation, Viability	Natural with Ray Tracing ^c	No	o
[Biostack Experiment]	Flour Beetle	Development		No	o
	Stick Insect	Development		No	o
Apollo 17	Pocket Mouse	Brain and Eye Histology	Natural with Ray Tracing ^c	No	o
Biocosmos 110	Micro- organisms	Phage Induction, Mutation	Natural	No	o
	Spiderwort	Chromosome Aberrations	± Radio- protec- tant ^e	No	0
	Dog	Physiology		No	of
cosmos 936 ^f	Rat	Retina Histology	Natural	Yes	o
cosmos 782 ^f	Rat	Retina Histology	Natural	No	o
Gemini III	Human Leukocytes	Chromosome Aberrations	Artif. ³² P- β	No	o
Gemini XI	Human Leukocytes	Chromosome Aberrations	Artif. ³² P- β	No	o
	Bread Mold	Viability, Mutation		No	o

Mission(s) [Experiment]	Organism (Common Name)	Endpoint(s) Measured	Exposure Mode	Flight Accel . control Yes/No	μ Gravit Inter- action + - 0
Biosatellite II	Bacteria	Phage Induction	Artif. ⁸⁵ Sr- γ	No	
	Bread Mold	Mutation		No	0
	Parasitic Wasp	Mutation, Fertility, Recombina- tion		No	0
	Fruit Fly	Somatic Chromosome Aberrations, Mutation, Recombina- tion, Nondisjunc- tion		No	0, +, +, 0
	Flour Beetle	Development		No	0
	Spiderwort	Pollen Abortion, Stamen Hair Stunting, Stamen Hair Mutation		No	+, +,
Cosmos 605 ^h	Rats	Testis Mass & Spermato- genesis, Haematopoi- esis in Marrow & Spleen, Colony Formation in Allografts, Muscle Mass, General Organ Morphology	Natura 1	No	0, 0, 0, 0, 0

Mission(s) [Experiment]	Organism (Common Name)	Endpoint(s) Measured	Exposure Mode	Flight Accel . Control Yes/No	μ Gravity Inter- action + - 0
Cosmos 690 ^h	Rat	Testis Mass & Spermato- genesis, Haematopoi- esis in Marrow & Spleen, Colony Formation in Allografts , Muscle Mass, General Organ Morphology	Artif. ¹³⁷ Cs- γ	No	0, + ⁱ , - ^j , + ^k . 0
Cosmos 368	H ₂ Bacteria, Yeast, Lettuce & Chick Pea Seeds	Viability, Viability, Chromosome Aberration	Artif. Pre- flight Gamma Rays	No	0
IML-1	Yeast	Viability in DNA Repair Conditional Mutant	Artif. Pre- flight X-rays	No	+ ^l

- ^a Natural refers to cosmic ray and proton environment of particular mission as attenuated by **spacecraft** materials. Artificial refers to radioisotope or X-ray exposure superposed on natural environment.
- ^b Interaction of radiation and microgravity. +, - & 0 refer to enhancement of effect, antagonism of effect or no interaction, respectively.
- ^c Ray tracing indicates that individual cosmic ray tracks were correlated with biological targets.
- ^d Microgravity and cosmic ray strikes synergistically inhibited development in a stage specific fashion.
- ^e An **aminothiols** was administered to some specimens.
- ^f Details of dog physiology are not reported but general condition was described as "**good**". Interaction was not assessable.
- ^g Cosmos 782 and 936 are best interpreted as paired experiments.
- ^h Cosmos 605 and 690 are best interpreted as paired experiments.
- ⁱ Microgravity inhibited recovery of radiation-induced pathology.
- ^j A slight enhancement of stem cell recovery normally follows irradiation and in the table context is an antagonistic effect.
- ^k Radiation inhibited recovery of microgravity-induced pathology.
- ^l Microgravity inhibited repair and recovery of permissively grown cells but had no effect on restrictively grown cells.