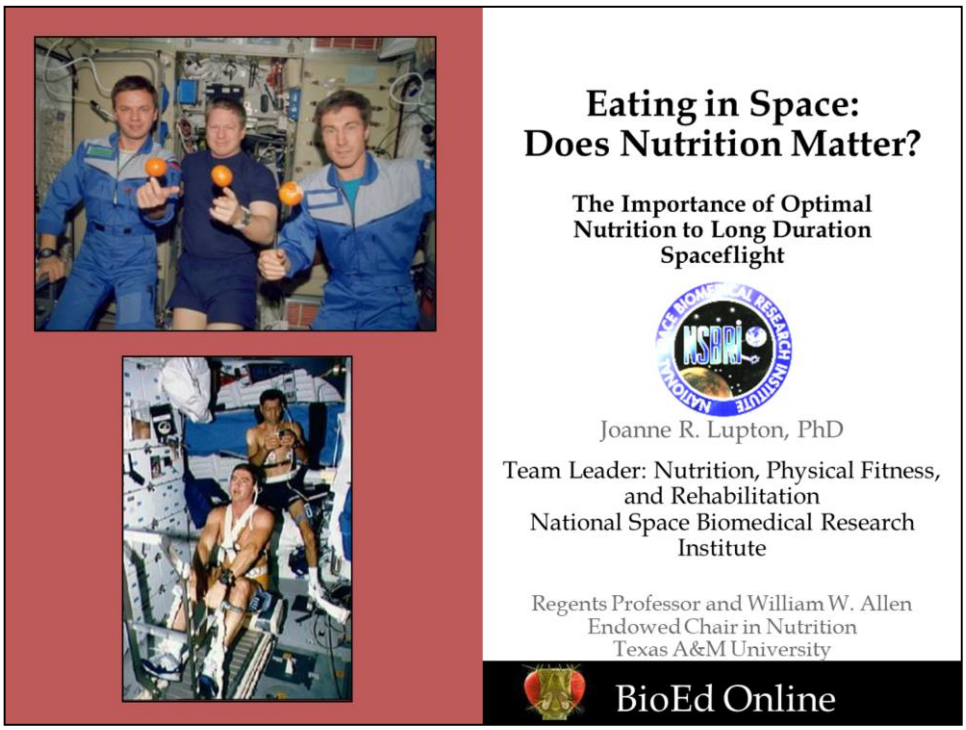


Notes from Speaker's Transcript

Hello, I'm Joanne Lupton, Professor of Nutrition at Texas A&M University and Team Leader for Nutrition and Physical Fitness at the National Space Biomedical Research Institute, or NSBRI. Long duration space flight poses a number of important challenges to human physiological systems. This presentation focuses on how nutrition and exercise can be used as countermeasures to some of the more critical risks. It also emphasizes the importance of optimal nutrition and physical fitness to health, both in space and on earth.

Image Reference:

NASA. *Photo gallery*. Retrieved 09-19-2007 from http://www.nasa.gov/multimedia/imagegallery/index.html.

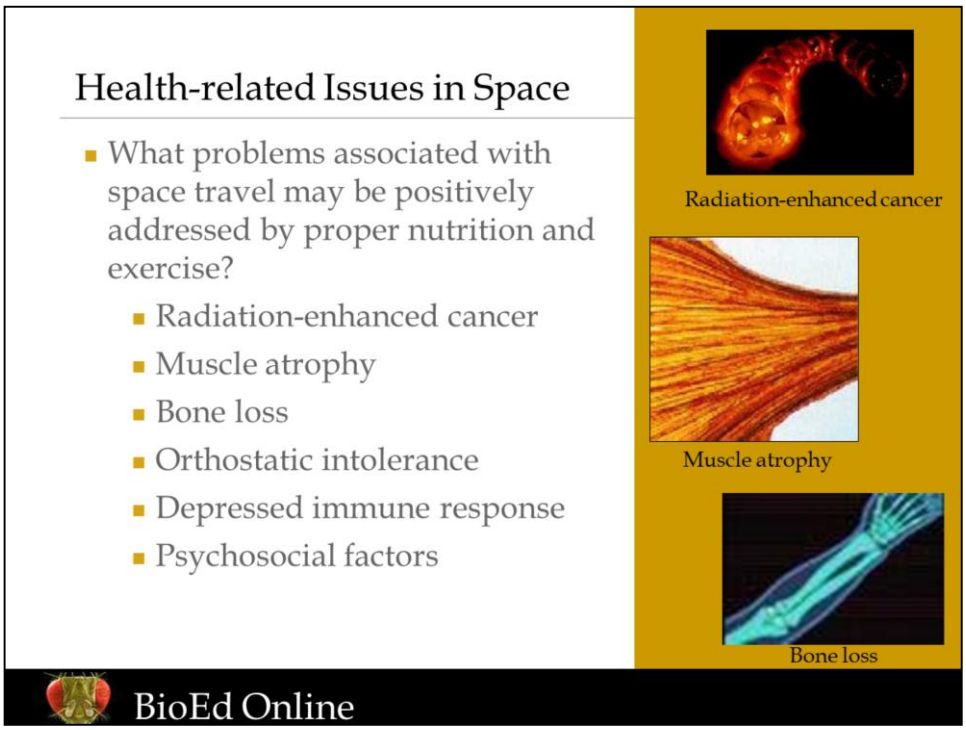
Overview

Four issues will be discussed in this presentation. First, we will address the importance of nutrition to manned spaceflight. Next, we will focus on two key areas of concern that can be impacted positively by nutritional countermeasures: muscle wasting and radiation-enhanced cancer. When nutrients are consumed they affect the entire body, not just one physiological system. So we must consider the effect of any nutritional or exercise countermeasure on the whole person. That topic will be emphasized in section three of this presentation. Finally, we will see how countermeasures developed for use in space apply equally well to health challenges here on earth.

Image References:

NASA. *Nutrition in space*. Retrieved 09-19-2007 from http://www.nasa.gov/multimedia/imagegallery/index.html.

NASA. *STS 117 Crew.* Retrieved 09-19-2007 from http://www.nasa.gov/mission_pages/shuttle/shuttlemissions/sts117/117_crew.html



Health-related Issues in Space

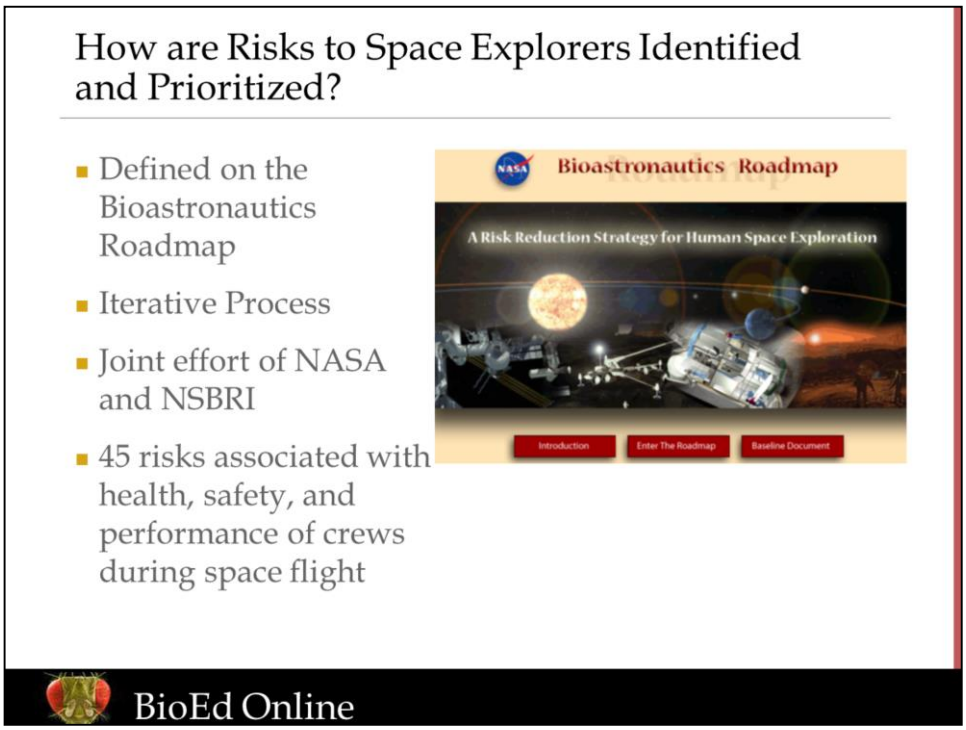
What problems associated with space travel may be positively addressed by proper nutrition and exercise? All of them can, from radiation-enhanced cancer to psychosocial factors. This presentation will concentrate on three health issues related to space travel: muscle atrophy, radiation-enhanced cancer, and to a lesser extent, bone loss.

Image References:

Lang, K. *X-ray view of the solar cycle*. Courtesy of K. Lang.

NASA. *Muscle atrophy*. Retrieved 09-19-2007 from http://weboflife.nasa.gov/images/muscles.jpg.

NASA. (2001) *NASA connect: Research on the ISS*. Retrieved 09-19-2007 from http://nasa.ibiblio.org/selection_page_new.php?action=search&query=ISS%20Research&start=40.

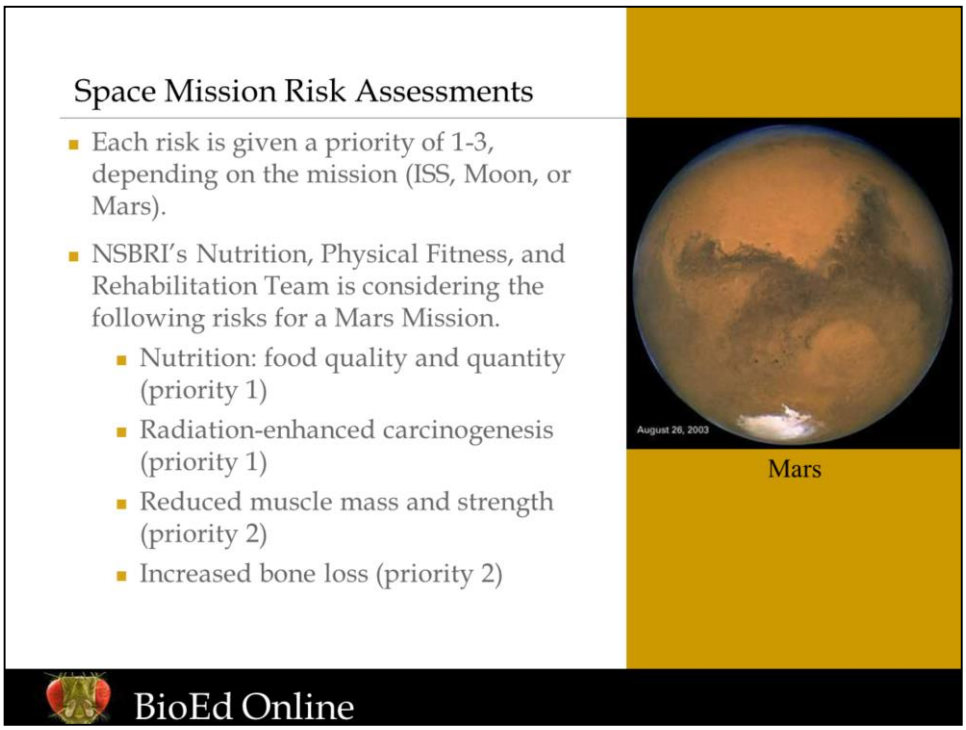


How are Risks Identified and Prioritized?

How are the risks to space explorers identified and prioritized? They are defined on the Bioastronautics Roadmap, which is created through an iterative process developed jointly by NASA and National Space Biomedical Research Institute (NSBRI). Forty-five important risks to the health, safety, and performance of crews during space flight have been identified. They are listed on the Bioastronautics Roadmap, which is publicly available at http://bioastroroadmap.nasa.gov/index.jsp.

Image Reference:

NASA. (2005). *Bioastronautics roadmap.* Retrieved 09-19-2007 from http://bioastroroadmap.nasa.gov.

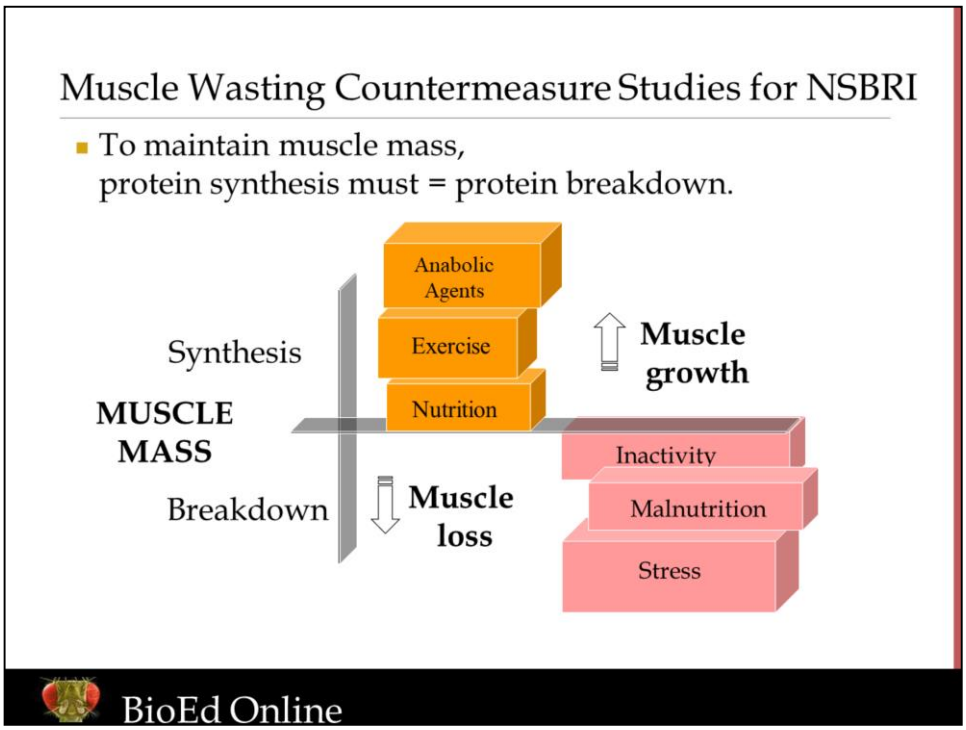


Space Mission Risk Assessments

Each of the 45 risks in the Bioastronautics Roadmap is given a priority (from 1-3, with 1 representing the highest risk), depending on the mission (International Space Station [ISS]; a trip to the Moon; or an exploratory mission to Mars). This slide shows the priorities for risks being considered by NSBRI's Nutrition, Physical Fitness, and Rehabilitation Team, in relation to a Mars mission. Nutrition and radiation-enhanced carcinogenesis are considered priority #1 risks, and muscle wasting and bone loss are considered priority #2. It should be noted that all items included among the 45 risks are of critical importance to NASA, and that the development of countermeasures against risks in that area is a high priority.

Image Reference:

NASA. (2003). *Mars, imaged by NASA's Hubble Space Telescope*. Retrieved 09-19-2007 from http://www.jpl.nasa.gov/news/features.cfm?feature=533.



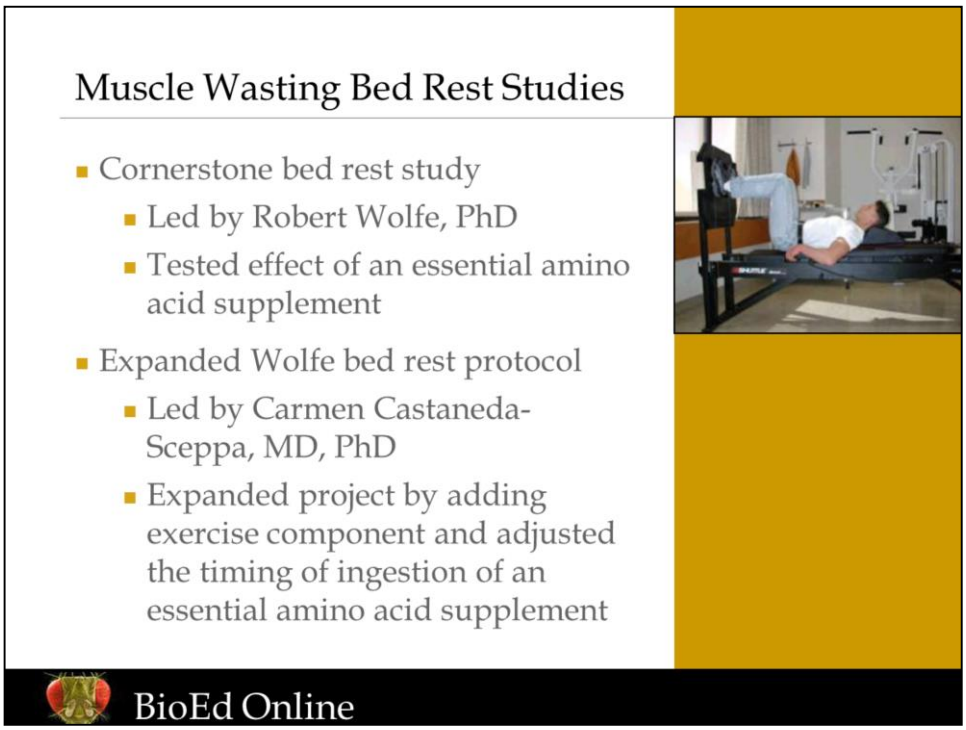
Muscle Wasting Countermeasure Studies for NSBRI

The two key areas for countermeasures covered in this presentation are radiation-enhanced cancer and muscle wasting. Let's start with countermeasures against muscle wasting.

To maintain muscle mass on Earth, muscle protein synthesis must equal muscle protein breakdown. This is not true in space, where protein breakdown exceeds protein synthesis, due to a number of factors, including inactivity (caused in large part by the microgravity environment), which does not place stress on the muscles. In addition, a depression in energy intake in space contributes to suboptimal nutrition and an increase in catabolic hormones, such as cortisol, as a reaction to stress. To counteract these negative effects on muscle mass and strength, it is necessary to develop countermeasures that supply optimal nutrition and appropriate exercise, and perhaps include anabolic agents.

Image References:

Paddon-Jones, D., Sheffield-Moore, M., Urban, E. J., Sanford, A. P., Aarsland, A., Wolfe, R.R., Ferrando, A.A. (2004). Essential amino acid and carbohydrate supplementation ameliorates muscle protein loss in humans during 28 days bedrest. *J Clin Endocrinol Metab, 89*, 4351-4358. Unpublished image by permission of D. Paddon-Jones.



Muscle Wasting Bed rest Studies

Two key projects of the NSBRI Nutrition, Physical Fitness and Rehabilitation Team are working to develop countermeasures against loss of muscle mass and strength. The first, our cornerstone bed rest study (with bed rest being an important earth-based model for some of the physiological changes occurring in space), tests the effect of a supplement containing essential amino acids. Dr. Robert Wolfe, currently at the University of Arkansas Medical School, is the principal investigator. The second project uses the same protocol as the Wolfe bed rest study so that results from both projects can be compared. But this second study, done at Tufts University, with Dr. Carmen Castaneda-Sceppa as Principal Investigator, adds an exercise component and a third component, which optimizes the timing between supplement ingestion and exercise.

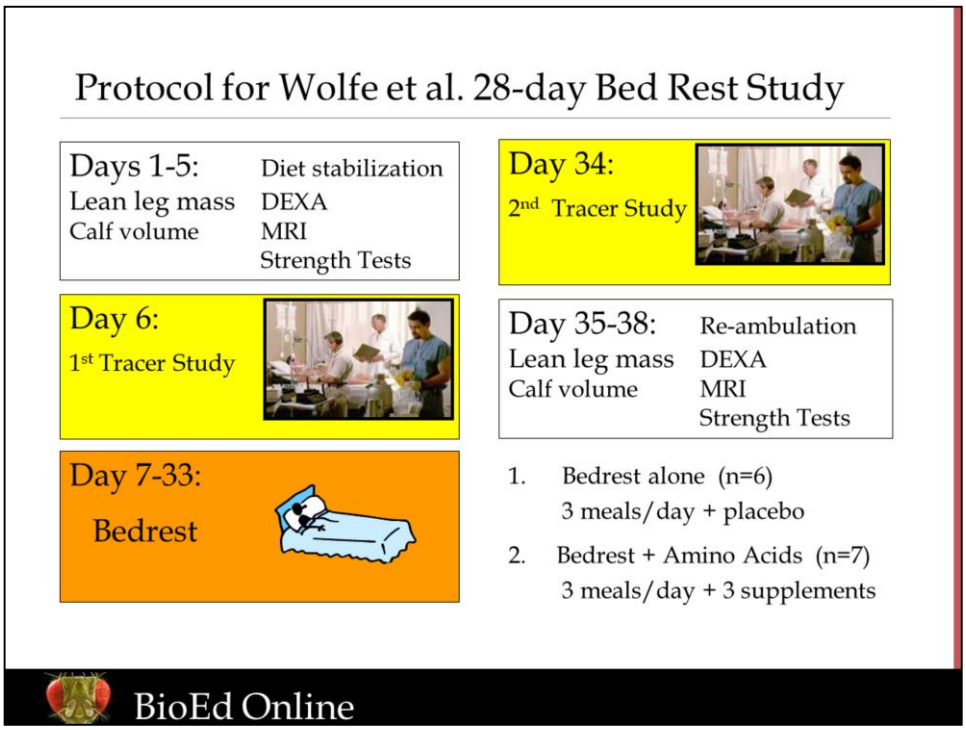
Reference:

Castaneda, C., Cloutier, G., Cadena, S., Pearlman, J., Brooks, N., Layne, J., Roubenoff, R. (2006). Resistance training and timed amino acid supplementation protects against the loss of muscle mass and strength with disuse. *FASEB, 20,* A591.

Paddon-Jones, D., Sheffield-Moore, M., Urban, E. J., Sanford, A. P., Aarsland, A., Wolfe, R.R., Ferrando, A.A. (2004). Essential amino acid and carbohydrate supplementation ameliorates muscle protein loss in humans during 28 days bedrest. *J Clin Endocrinol Metab, 89,* 4351-4358.

Image Reference:

Leg exercise. Dr. Carmen Castenade-Sceppa Study: Time feeding and resistance training to prevent muscle atrophy. Prepared by Gregory Cloutier.



Protocol for Wolfe et al. 28-day Bed rest Study

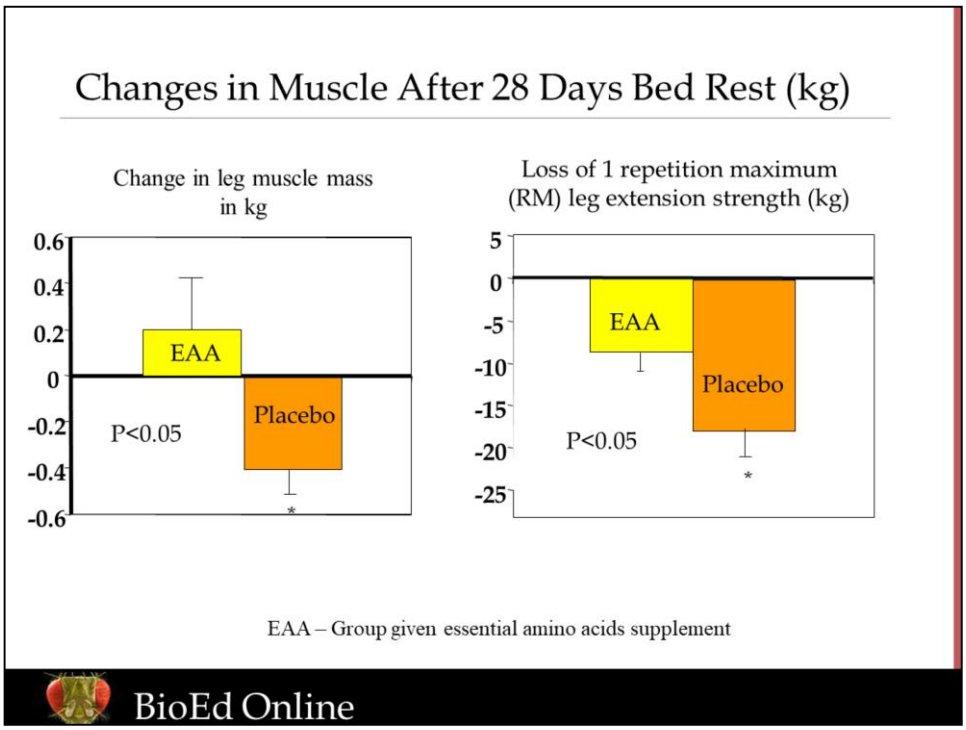
This slide shows the protocol for the Wolfe bed rest study. Days 1-5 were the diet stabilization period in which initial measurements were made: lean leg mass (by DEXA* – Dual Energy X-Ray Absorptiometry); calf volume (by MRI); and a series of strength tests. On Day 6, the first tracer study** was done with a stable isotope to measure muscle protein synthesis. During days 7-33, subjects were in bed rest (with other samples collected and food intake controlled). Day 34 was a repeat of the tracer study done on Day 6, and Days 35-38 provided time for re-ambulation and repeats of measurements made on days 1-5. There were two groups, a placebo group and the group receiving the essential amino acid supplement.

* DEXA: a precise instrument that uses energy in the form of very small doses of X-rays to determine bone mineral density.

** Tracer studies: harmless radio isotopes are attached to specific amino acids that travel through the bloodstream. Blood samples are taken to determine the amount of amino acids that enter and leave the muscle.

Image Reference:

Paddon-Jones, D., Sheffield-Moore, M., Urban, E. J., Sanford, A. P., Aarsland, A., Wolfe, R.R., Ferrando, A.A. (2004). Essential amino acid and carbohydrate supplementation ameliorates muscle protein loss in humans during 28 days bedrest. *J Clin Endocrinol Metab, 89*, 4351-4358. Unpublished image/by permission of D. Paddon-Jones.



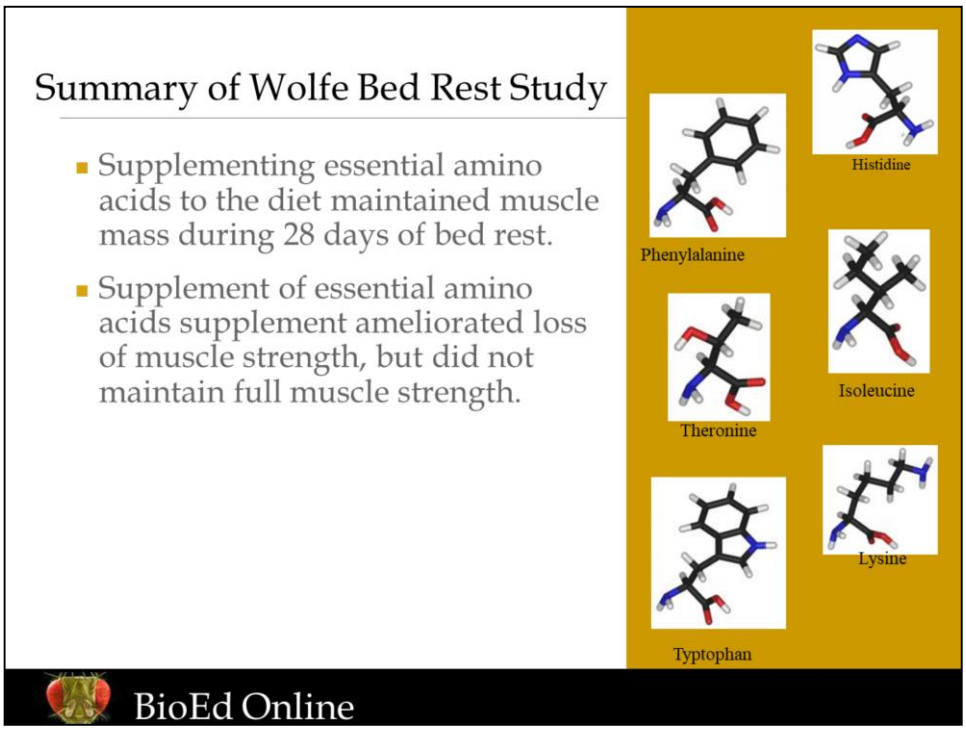
Changes in Muscle After 28 Days Bed Rest (kg)

This slide shows the effect of adding the essential amino acid supplement to the diet on leg muscle mass, and the loss of muscle strength as a result of the 28 days of bed rest. Results from the supplemented group are shown in yellow; results from the placebo group are shown in orange. The graph on the left illustrates the loss of approximately 400 g of leg muscle mass in the placebo group. In contrast, those supplemented with essential amino acids actually gained about 200 g of leg muscle mass.

Loss of leg strength after bed rest is represented by the graph on the right. Again, the amino acid-supplemented group is shown in yellow and the placebo group in orange. Both groups lost muscle strength, although the loss was somewhat ameliorated in the amino acid supplemented group.

Image Reference:

Paddon-Jones, D., Sheffield-Moore, M., Urban, E. J., Sanford, A. P., Aarsland, A., Wolfe, R.R., Ferrando, A.A. (2004). Essential amino acid and carbohydrate supplementation ameliorates muscle protein loss in humans during 28 days bedrest. *J Clin Endocrinol Metab, 89*, 4351-4358. Redrawn.

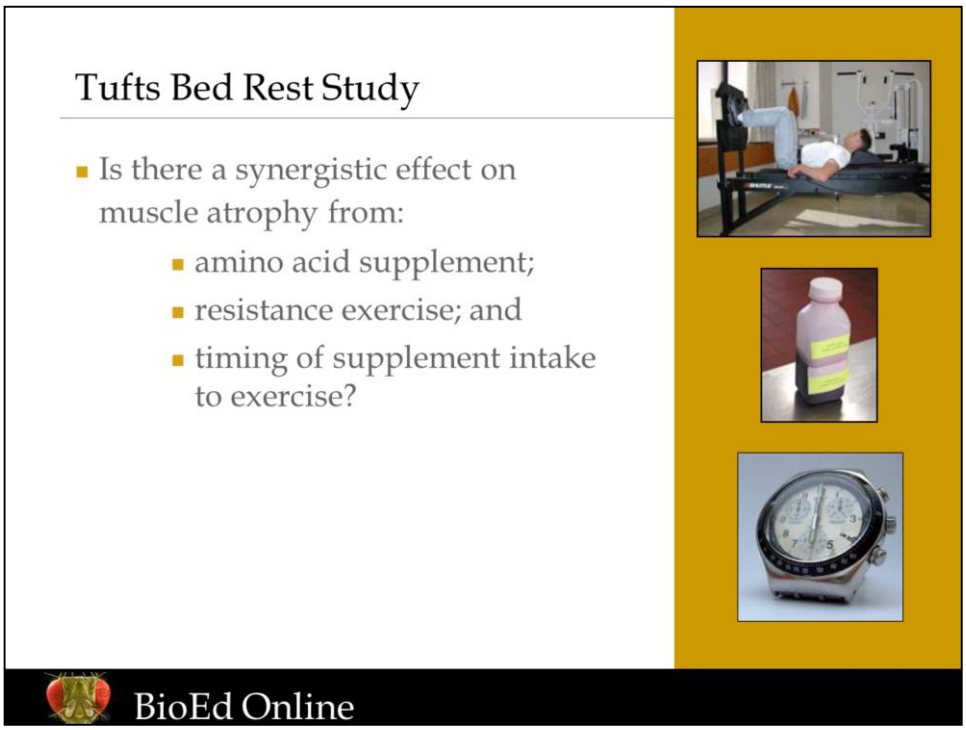


Summary of Wolfe Bed Rest Study

An essential amino acid supplement to the diet maintained muscle mass during 28 days of bed rest. This same essential amino acid supplement ameliorated loss of muscle strength, but did not maintain full muscle strength. These results suggested that we needed an additional countermeasure, together with the amino acid supplement, to maintain muscle strength. This led us to develop the next study implemented at Tufts University, with Dr. Carmen Castaneda-Sceppa.

Image Reference:

Wikipedia. (2006). *Amino acids*. Retrieved 09-20-2007 from http://en.wikipedia.org/wiki/Strecker_amino_acid_synthesis.



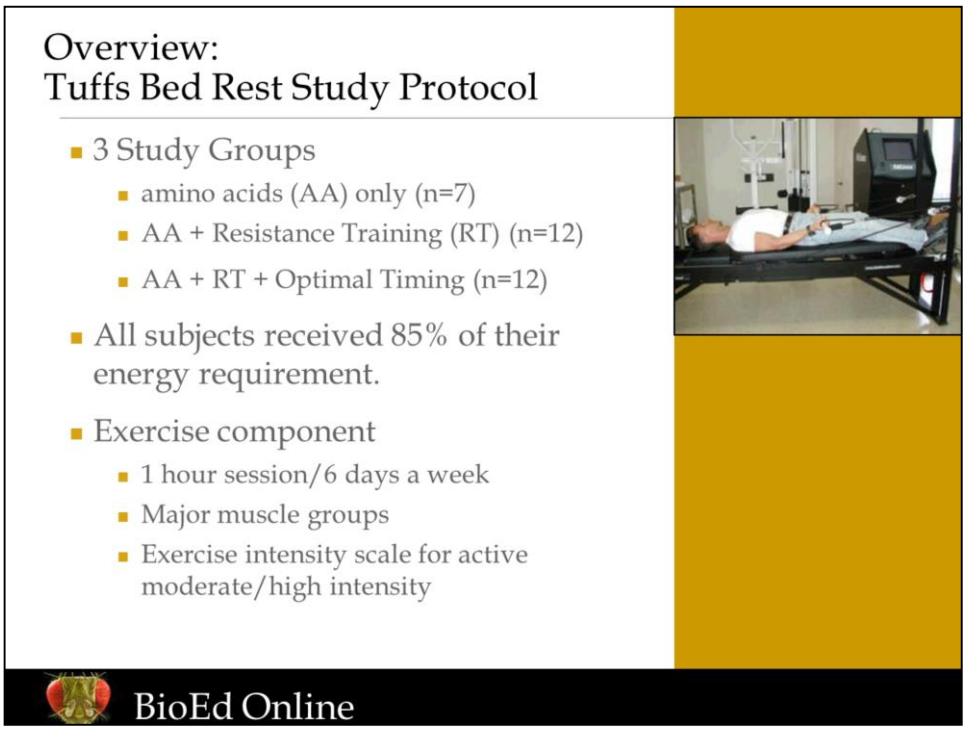
Tufts Bed Rest Study

Using the Wolfe bed rest study as our prototype, and keeping the same protocol so that we could compare between studies, a second study was designed and implemented at Tufts University, with Dr. Carmen Castaneda-Sceppa as Principle Investigator. This study expanded on the original project by adding both an exercise component and a timing component (optimal time between ingestion of the supplement and performance of the exercise). The purpose of the study was to determine if an amino acid supplement, plus a resistance exercise program, plus optimized timing of supplement intake to exercise, would have a synergistic effect on muscle atrophy.

Image Reference:

Leg exercise. Dr. Carmen Castenade-Sceppa Study: Time feeding and resistance training to prevent muscle atrophy. Prepared by Gregory Cloutier.

Watch. Retrieved 09-21-2007 from http://en.wikipedia.org/wiki/Time.



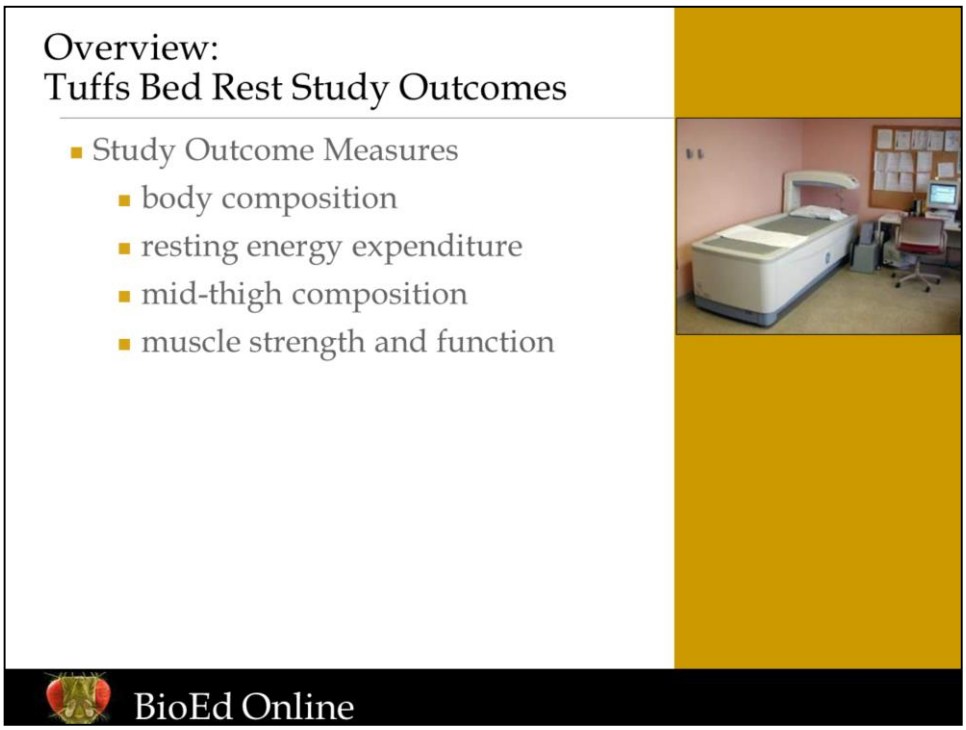
Overview of Tuffs Bed Rest Study Protocol

There were three groups in the study. One group received amino acids only; one group received an amino acid supplement plus resistance training; and one group combined the amino acid supplement with resistance training and the optimal timing between ingestion of the supplement and initiation of the exercise. It is important to note that all subjects received only 85% of their energy requirements, in order to mimic the usual depressed energy intake of astronauts.

The exercise component consisted of one-hour sessions which used all of the major muscle groups, six days per week. Sessions focused on upper body one day and lower body the next. The exercise intensity was at an active moderate/high level.

Image Reference:

Strength exercise. Dr. Carmen Castenade-Sceppa Study: Time feeding and resistance training to prevent muscle atrophy. Prepared by Gregory Cloutier.

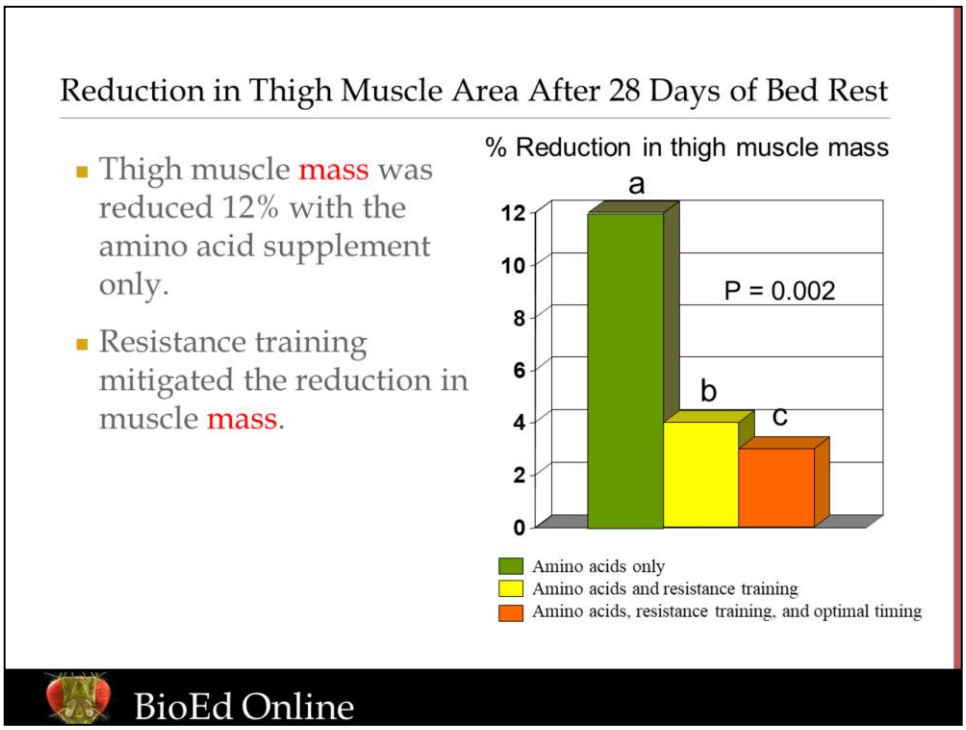


Overview of Tuffs Bed Rest Study Outcomes

Some of the important study outcome measures were analyses of body composition, resting energy expenditure, mid-thigh composition, and muscle strength and function. The following two slides illustrate some of our findings.

Image Reference:

Dr. Carmen Castenade-Sceppa Study: Time feeding and resistance training to prevent muscle atrophy. Prepared by Gregory Cloutier.

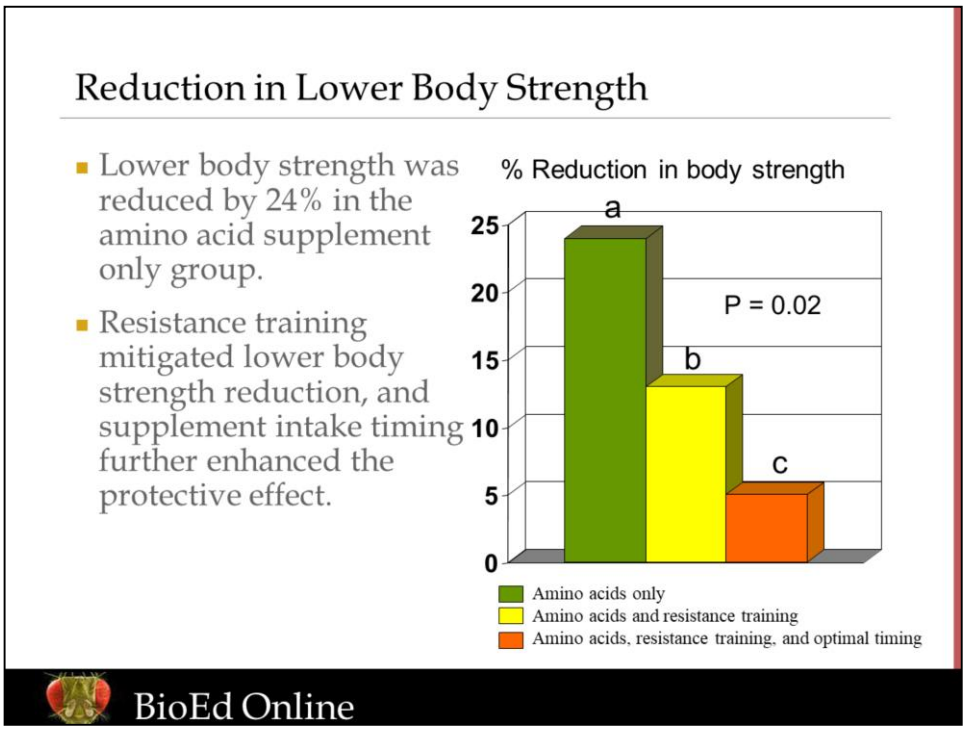


Reduction in Thigh Muscle Area After 28 Days of Bed Rest

The graph in this slide shows the reduction in thigh muscle mass after 28 days of bed rest. The green bar represents the group that received only the amino acid supplement. The yellow bar represents the group that received the amino acid supplement plus resistance training. The orange bar represents the group that received the amino acid supplement plus resistance training, and also followed optimal timing between the supplement and training. As indicated, there was a highly significant loss of muscle mass in the supplement-only group. The addition of resistance training and resistance training plus optimal timing mitigated the reduction in muscle area.

Image Reference:

Castaneda, C., Cloutier, G., Cadena, S., Pearlman, J., Brooks, N., Layne, J., Roubenoff, R. (2006). Resistance training and timed amino acid supplementation protects against the loss of muscle mass and strength with disuse. *FASEB, 20,* A591. Redrawn.

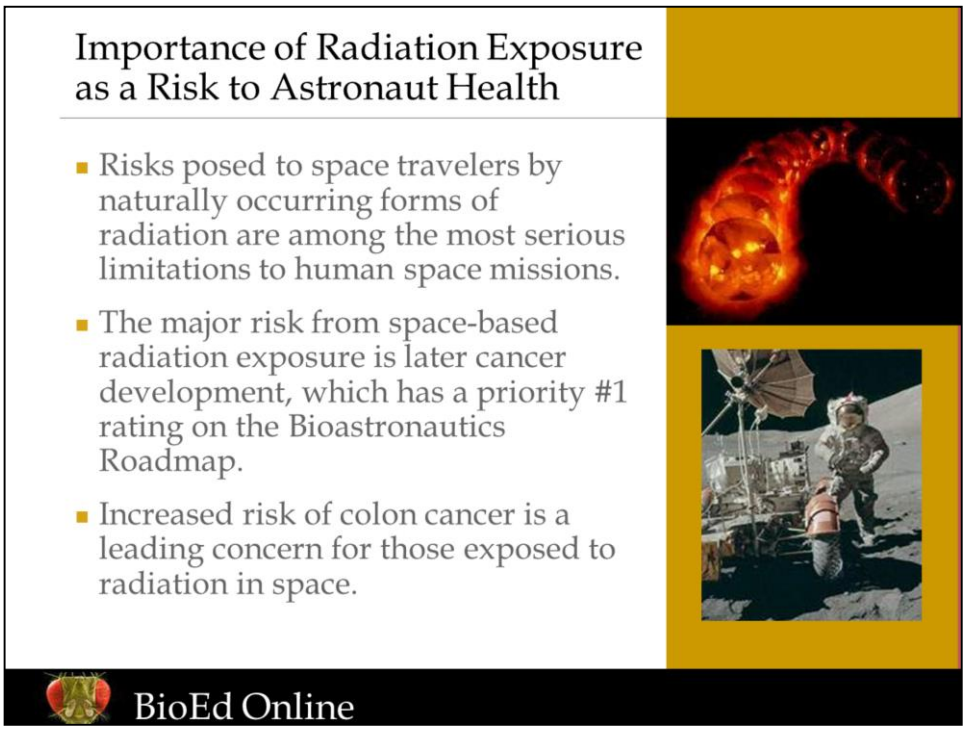


Reduction in Lower Body Strength

The graph on this slide illustrates the reduction in lower body strength after 28 days of bed rest. As on the previous slide, the green bar represents the group that received the amino acid suupplement only. The group represented by the yellow bar received the amino acid supplement plus resistance training; and group represented by the orange bar had a combination of the amino acid supplement, resistance training and timing. Lower body strength was reduced by 24% in the amino acid only group. Resistance training mitigated the reduction of strength, and timing further enhanced the protective effect. An important difference between this bed rest study (Tufts) and the Wolfe bed rest study was that all subjects in the Tufts study received only 85% of their daily energy requirement. The effectiveness of the amino acid supplement alone was much less in this study than was observed in the Wolfe study.

Image Reference:

Castaneda, C., Cloutier, G., Cadena, S., Pearlman, J., Brooks, N., Layne, J., Roubenoff, R. (2006). Resistance training and timed amino acid supplementation protects against the loss of muscle mass and strength with disuse. *FASEB, 20,* A591.



Importance of Radiation Exposure as a Risk to Astronaut Health

Now let's turn to nutritional countermeasures against radiation-enhanced cancer. As noted earlier, risks posed to personnel in space by naturally occurring radiations are among the most serious limitations to human space missions, with the primary risk from radiation exposure being later cancer development. Radiation-enhanced cancer has a priority #1 on the Bioastronautics Roadmap. Of all the cancer risks likely to be enhanced by radiation, colon cancer is among the leading concerns. It is the second leading cause of cancer deaths in the United States today and strikes men and women more or less equally.

References:

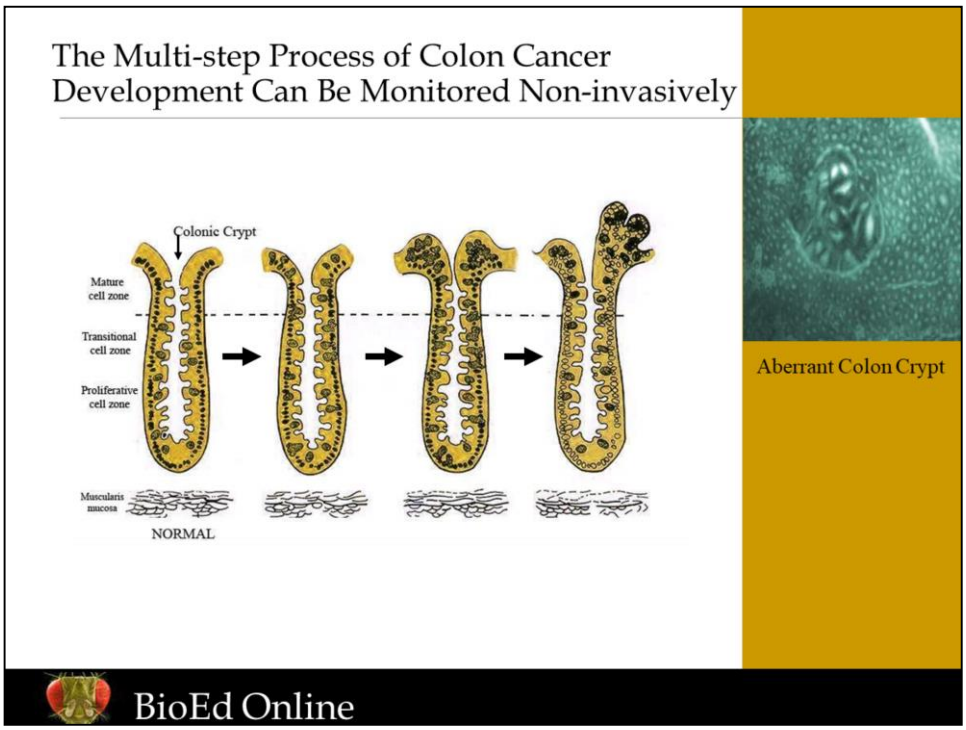
National Research Council. (1990). BEIR V Health effects of Exposure to low levels of ionizing radiation. Washington, DC: National Academy Press.

National Research Council. (1998). BEIR V Health effects of Exposure to low levels of ionizing radiation. Washington, DC: National Academy Press.

Image References:

Lang, K. *X-ray view of the solar cycle*. Courtesy of K. Lang.

NASA. *Lunar expedition*. Retrieved 09-19-2007 from http://www.nasa.gov/multimedia/imagegallery/index.html.



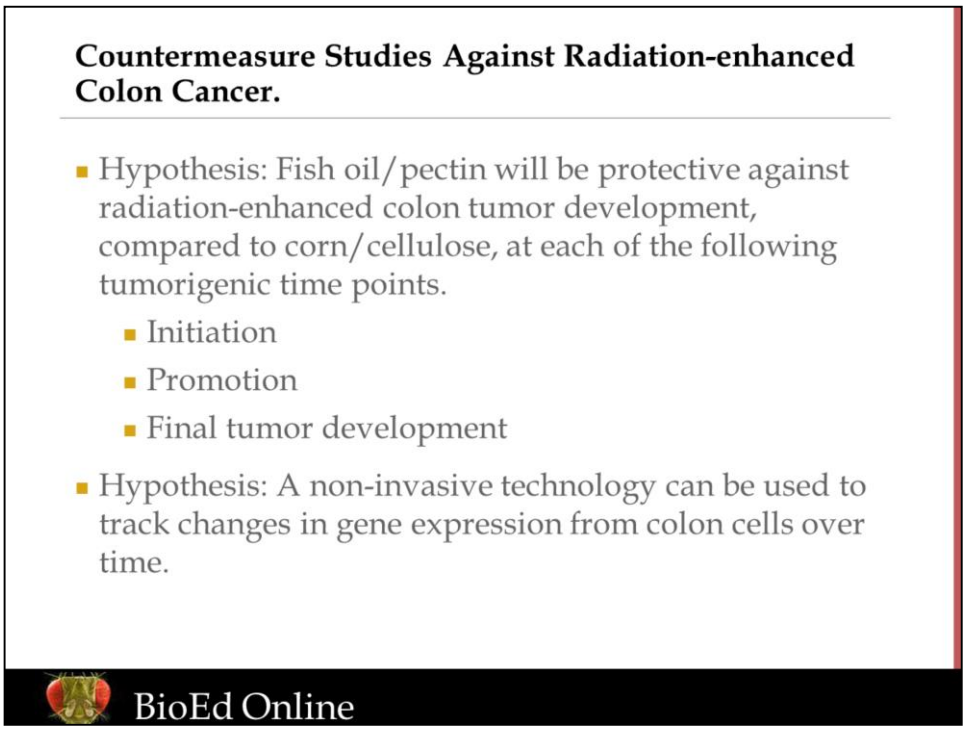
The Multi-step Process of Colon Cancer Development Can Be Monitored Non-invasively

The development of colon cancer from a normal cell to a malignant tumor is a multi-step process. Colon epithelial cells are only one layer deep and they are arranged in patterns called "crypts." Stem cells are located at the bottom of the crypts and they give birth to daughter cells, which migrate up the side of the crypt, dividing several times while in transit. As they migrate, they differentiate, and towards the top of the crypt, they undergo apoptosis – or programmed cell death – and are exfoliated into the fecal stream. We are able to recapture these colon cells from fecal material and analyze the mRNA in the cells to track changes in colon gene expression patterns. We can do this at any stage of the carcinogenic process, from normal healthy cells, to cells that have become dysplastic, to the development of what are called aberrant crypts (shown here), to colon polyps and through to carcinomas.

Image Reference:

Lipkin, M. (1974). *Cancer Crypts* (re-drawn with permission of M. Lipkin) from Phase 1 and Phase 2 proliferative lesions of colonic epithelial cells in diseases leading to colonic cancer. *Cancer,* 34, 878-888.

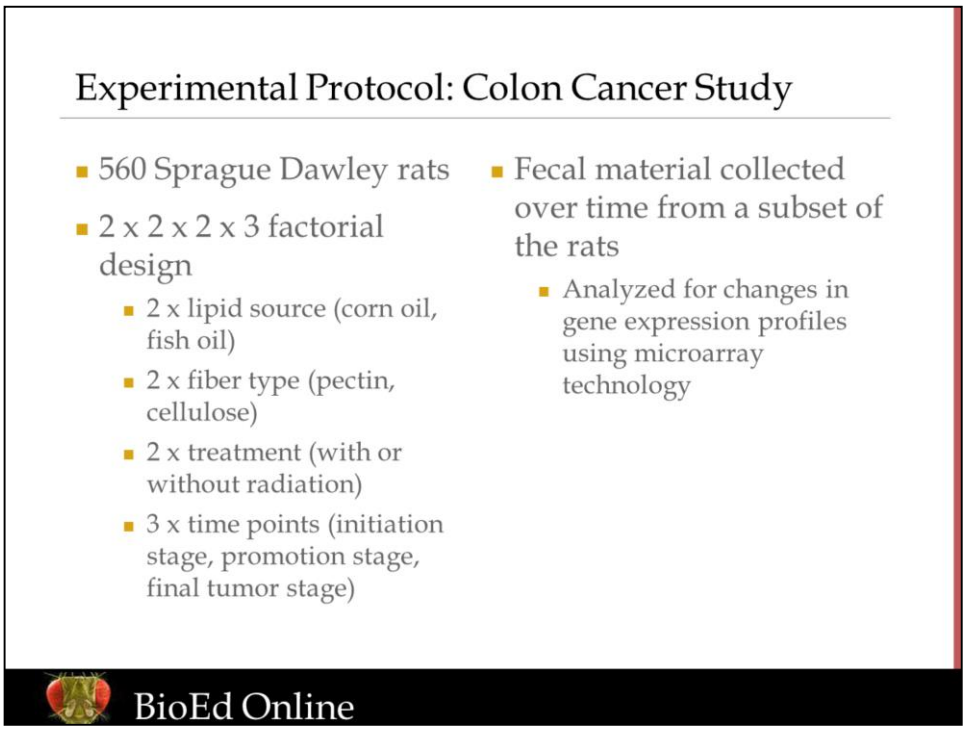
Lupton, J. (2007). *Aberrant colon crypts.*



Radiation-enhanced Countermeasure Studies for NSBRI

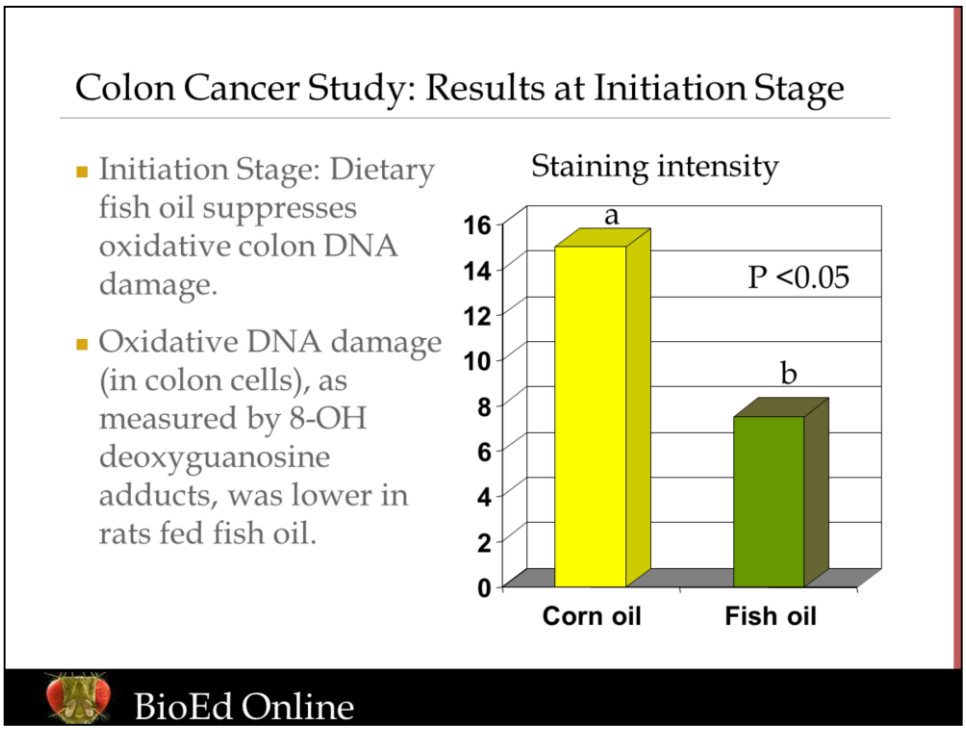
We developed the following hypotheses for countermeasure studies against radiation-enhanced colon cancer.

1. A diet high in fish oil (a good source of omega 3 fatty acids) and pectin (a fermentable fiber), which we had shown to be protective against chemically-induced colon cancer, also would protect against radiation-enhanced colon cancer at three tumor development points and further.
2. A non-invasive technology (recovering mRNA from fecal material) can be used to track changes in gene expression from colon cells over time.



Experimental Protocol: Colon Cancer Study

To test the hypothesis (given in the previous slide), we used the following experimental protocol. Five hundred and sixty male Sprague Dawley rats were allocated to a 2 by 2 by 2 by 3 factorial design with two sources of lipid (corn oil or fish oil), two types of fiber (pectin or cellulose), with or without irradiation (1 Gray iron ions, at Brookhaven National Laboratory), and terminated at three different time points–the initiation stage, the promotion stage and at the final tumor endpoint. Fecal material was collected throughout the study from a subset of rats, and mRNA was analyzed for changes in gene expression profiles using microarray technology.

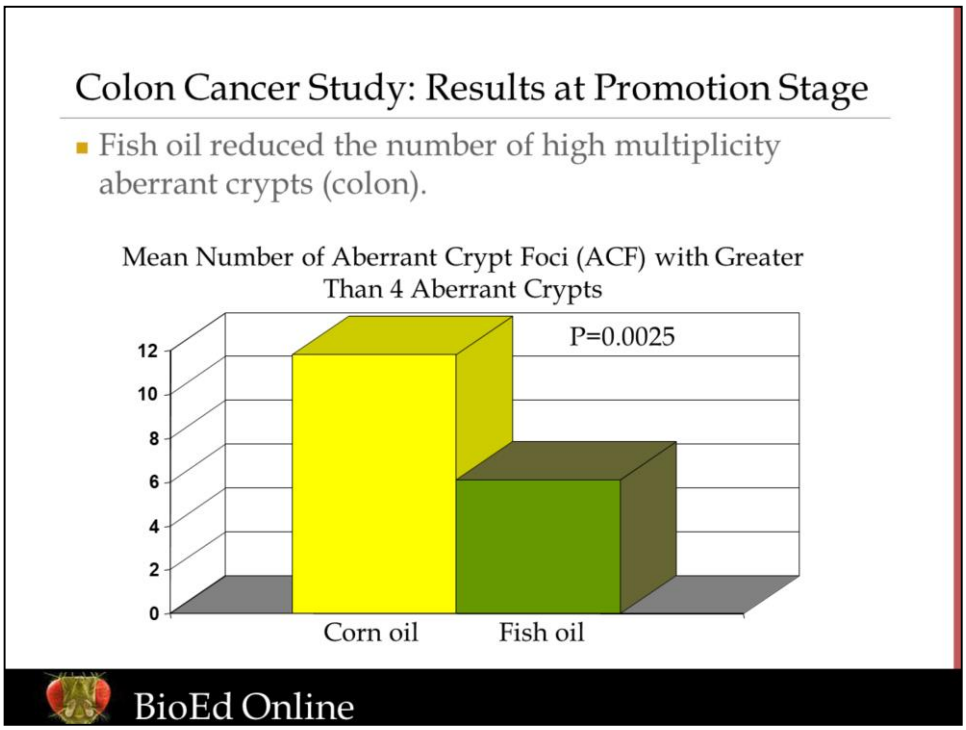


Colon Cancer Study: Results at Initiation Stage

The graph in this slide shows the effect of diet at the initiation stage, when colon DNA is damaged. A marker for oxidative damage to DNA (the type of damage that occurs from radiation) is the DNA adduct 8-OH deoxyguanosine. As shown here, fish oil feeding reduced DNA damage more than corn oil feeding did.

Image Reference:

Lupton, J., Turner, N.D., Braby, L.A., Chapkin, R.S., Ford, J.R., & Carroll, R.J. (not published). Nutritional countermeasures to radiation-enhanced colon cancer.

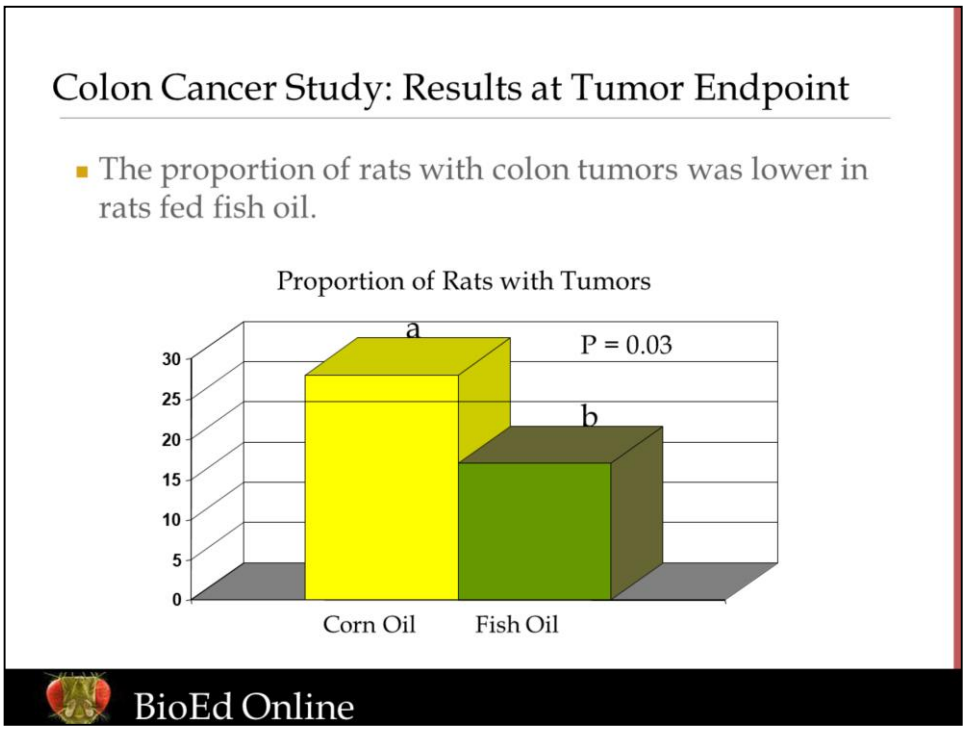


Colon Cancer Study: Results at Promotion Stage

Aberrant crypts are precursors to later tumor development in both rats and in humans. A focus with four or more aberrant crypts is considered highly predictive of later tumor development. As shown here, fish oil reduced the number of high multiplicity aberrant crypts more than corn oil did.

Image Reference:

Lupton, J., Turner, N.D., Braby, L.A., Chapkin, R.S., Ford, J.R., & Carroll, R.J. (not published). Nutritional countermeasures to radiation-enhanced colon cancer.

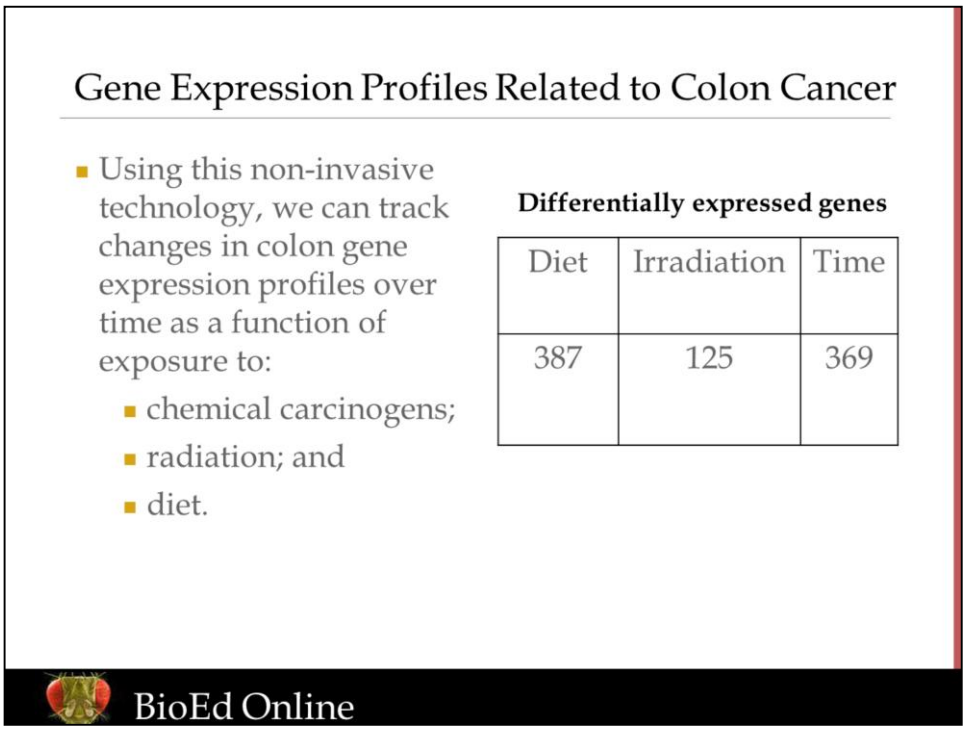


Colon Cancer Study: Results at Tumor Endpoint

Finally, at the tumor stage, a lower proportion of rats fed fish oil had colon tumors, as compared to rats that were fed corn oil. Thus, the diet intervention was an effective countermeasure at each stage of the tumorigenic process.

Image Reference:

Lupton, J., Turner, N.D., Braby, L.A., Chapkin, R.S., Ford, J.R., & Carroll, R.J. (not published). Nutritional countermeasures to radiation-enhanced colon cancer.

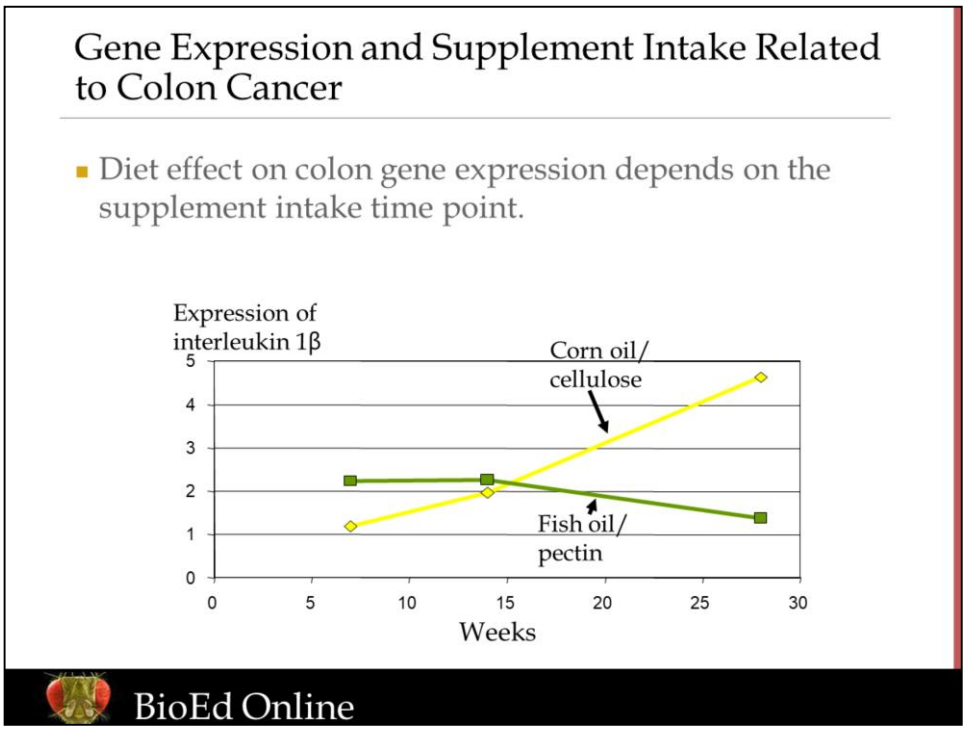


Gene Expression Profiles Related to Colon Cancer

We also were able to use our non-invasive technology (recovering mRNA from fecal material) to track changes in colon gene expression profiles over time. Using this technique, we discovered 387 genes that were significantly differentially expressed—either over- or under-expressed—as a function of diet. Similarly, we discovered 125 genes differentially expressed as a function of irradiation, and 369 as a function of time.

Image Reference:

Lupton, J., Turner, N.D., Braby, L.A., Chapkin, R.S., Ford, J.R., & Carroll, R.J. (not published). Nutritional countermeasures to radiation-enhanced colon cancer.



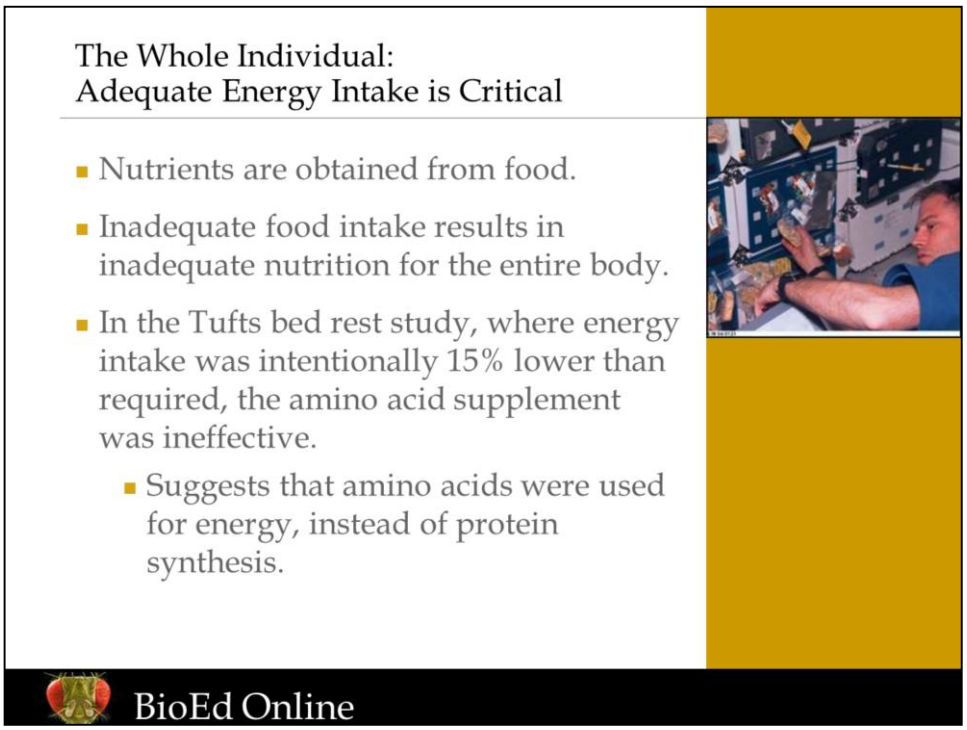
Gene Expression and Supplement Intake Related to Colon Cancer

When we looked at specific genes either up- or down-regulated as a function of diet, radiation, or time, the importance of monitoring these changes over time became apparent. This slide shows the expression of one gene over time (interleukin 1 beta–a pro-inflammatory protein that promotes colon cancer). If we ask, "What is the effect of diet on the expression of interleukin 1 beta?" the answer is, "It depends." As depicted in the graph, at the earliest time point (week 7), gene expression is higher in rats fed fish oil. At 15 weeks, expression levels are the same for both diets, and at the final tumor endpoint (30 weeks), the corn oil/cellulose diet resulted in much higher expression levels than did the fish oil/pectin diet. This finding emphasizes the point that if one monitors the effect of diet on gene expression, it is more accurate to do so in a time-dependent manner than as a single snapshot.

To summarize the studies on diet and radiation-enhanced colon cancer: a diet high in fish oil and pectin can protect against radiation-enhanced colon cancer and a noninvasive technique can be used to monitor colon gene expression over time.

Image Reference:

Lupton, J., Turner, N.D., Braby, L.A., Chapkin, R.S., Ford, J.R., & Carroll, R.J. (not published). Nutritional countermeasures to radiation-enhanced colon cancer.



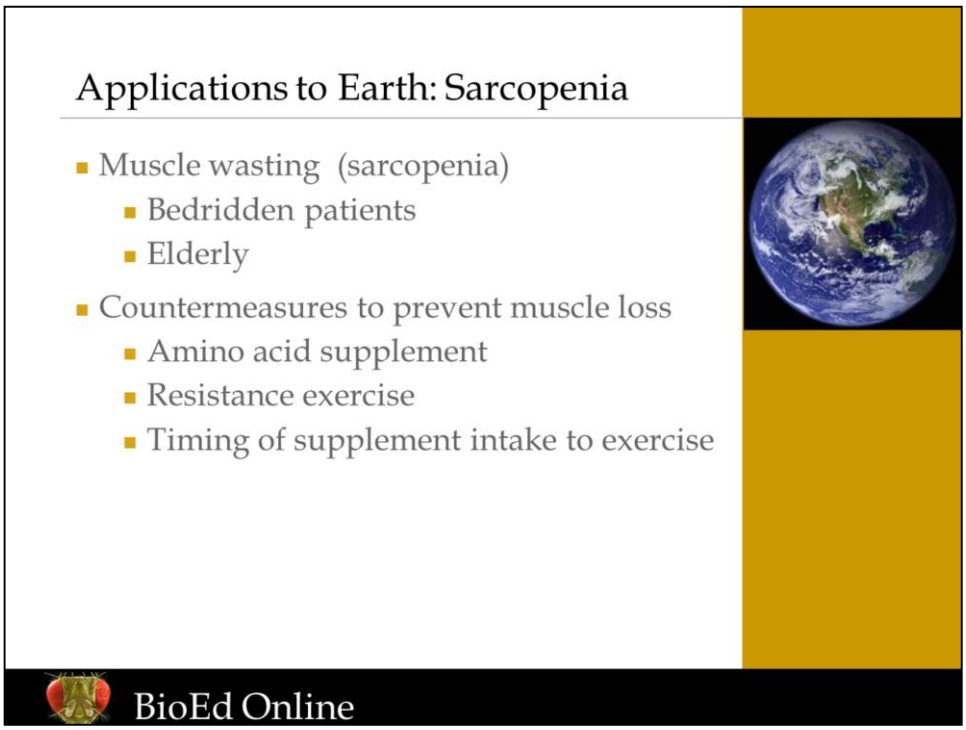
The Whole Individual: Adequate Energy Intake is Critical

Nutrients are delivered through foods. When foods are digested, nutrients are disbursed and absorbed into all parts of the body not just our site of interest, such as muscle or the colon. Thus, we must be concerned with the effect of any single countermeasure on ALL systems, not just our system of interest. For example, some preliminary data suggest that an amino acid supplement provided to enhance muscle protein synthesis may exacerbate bone loss. Certain amino acids produce a lower metabolic pH in the body, which, in turn, can cause reabsorption of calcium from bone. This issue must be considered. In contrast, fish oil, which we have shown to protect against radiation—enhanced colon cancer, also may enhance muscle mass through an effect on decreasing protein breakdown.

The single most important overall effect of suboptimal nutrition is inadequate energy intake—not eating enough food—which is characteristic of individuals in space. Inadequate food intake results in inadequate nutrition. We saw this in the Tufts bed rest study, where energy intake was intentionally kept 15% lower than required (to mimic the intake observed in spaceflight). As we saw, under these conditions, the amino acid supplement was not effective in protecting against loss of muscle mass. The amino acids probably were being used for energy instead of protein synthesis. Finding a solution to promote adequate energy intake is an important goal of our team.

Image Reference:

NASA. *Nutrition in space*. Retrieved 09-19-2007 from http://www.nasa.gov/multimedia/imagegallery/index.html.

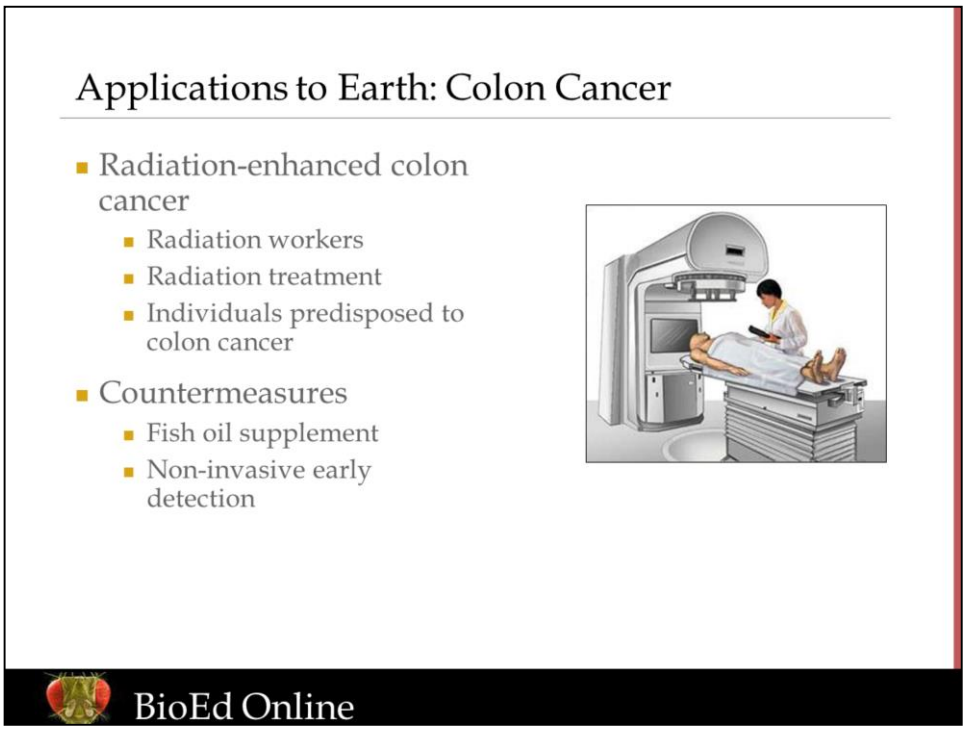


Applications to Earth: Sarcopenia

Countermeasures developed for use in space often are equally important on Earth. Sarcopenia, or muscle wasting, is a serious condition, particularly for bedridden and elderly individuals. An amino acid supplement, combined with resistance exercise and timing of the supplement intake to the exercise, should be beneficial for individuals on Earth.

Image Reference:

NASA. *Nutrition in space*. Retrieved 09-19-2007 from http://www.nasa.gov/multimedia/imagegallery/index.html.

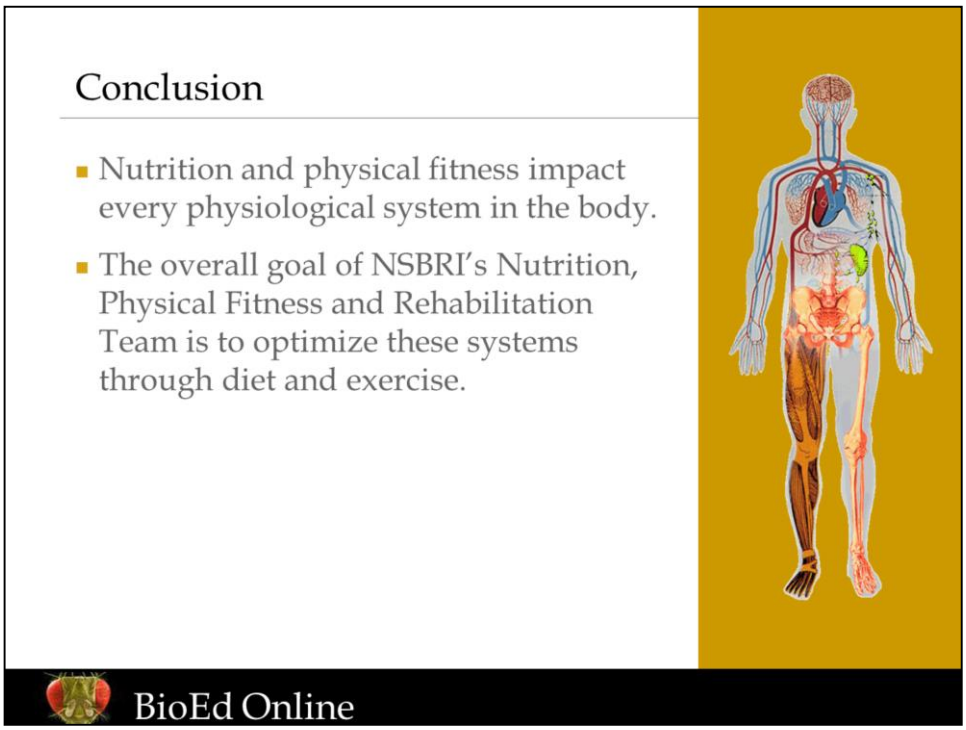


Applications to Earth: Colon Cancer

As mentioned before, colon cancer is the second leading cause of death from cancer in the United States today. A diet intervention to protect against its development should work equally well on Earth as in space. Also, certain groups, such as radiation workers, those undergoing radiation treatment for colon cancer, and individuals genetically predisposed to colon cancer, who are at higher risk for this cancer could benefit both from the diet discussed here, and from the non-invasive early detection of this disease.

Image Reference:

National Cancer Institute. (2007). *External beam radiation therapy*. Retrieved 09-21-2007 from http://www.cancer.gov/cancertopics/radiation-therapy-and-you/page3.



Conclusion

Nutrition and physical fitness impact every physiological system in the body. The overall goal of NSBRI's Nutrition, Physical Fitness and Rehabilitation Team is to optimize these systems through diet and exercise.

Image Reference:

NASA. *Photo gallery*. Retrieved 09-19-2007 from http://www.nasa.gov/multimedia/imagegallery/index.html.