UNIVERSITI PUTRA MALAYSIA

EFFECTS OF PROBIOTIC SUPPLEMENTATION ON CALCIUM ABSORPTION AND BONE HEALTH STATUS IN OVARIECTOMIZED RATS

KOLSOOM PARVANEH

FPSK(p) 2015 19

EFFECTS OF PROBIOTIC SUPPLEMENTATION ON CALCIUM ABSORPTION AND BONE HEALTH STATUS IN OVARIECTOMIZED RATS

By

KOLSOOM PARVANEH

Thesis Submitted to the School of Graduate Studies, Universiti Putra Malaysia, in Fulfillment of the Requirements for the Degree of Doctor of Philosophy

August 2015

DEDICATION

This thesis is dedicated to my family, parents, brothers and sisters who love me unconditionally and whose good examples have taught me to work hard for the things that I aspire to achieve.

This work is also dedicated to my husband, Mr. Behzad Mahdian who has been a constant source of support and love. I am truly thankful for having you in my life.

Abstract of thesis presented to the senate of Universiti Putra Malaysia in fulfillment of the requirement for the Degree Doctor of Philosophy

EFFECTS OF PROBIOTIC SUPPLEMENTATION ON CALCIUM ABSORPTION AND BONE HEALTH STATUS IN OVARIECTOMIZED RATS

By

KOLSOOM PARVANEH

August 2015

Chairman: Associate professor Rosita Jamaluddin, PhD
Faculty: Medicine and Health Sciences

Osteoporosis is a bone metabolism disorder which is explained by low bone mass and deterioration of bone tissue which is mostly due to the aging or menopause. The possibility of wrist, hip or spine fracture due to osteoporosis is estimated as parallel to the risk of heart disease. Therefore, the aim of the current study is to examine the effect of probiotics (*Bifidobacterium. longum,* and *Lactobacillis. helveticus*) as a single species or double strain on calcium absorption and bone health status in ovariectomized rats.

For this purpose, 56 female Sprague–Dawley rats of 10 weeks old were used in this study. Bilateral ovariectomy (Ovx) and sham-ovx (S-ovx) were performed on the rats. After 2 weeks of surgery the rats were randomly divided into 6 groups of Ovx and one group of S-ovx. The surgical process and diet were as follow: G1: S-ovx, G2: Ovx, G3: Ovx + Ca, G4: Ovx + *B. longum,* G5: Ovx + *L. helveticus,* G6: Ovx + mixture of *L. helveticus* and *B. longum,* and G7: Ovx + mixture of *L. helveticus* and *B. longum* + Ca. The urine, fecal, and serum of the rats were collected and apparent Ca absorption and Ca retention were measured. Serum biomarkers were evaluated. Bone mineral contents (BMC) were analysed from the femur using atomic absorption spectrophotometer (AAS). BMD, trabecular structures, percentage of bone volume/total volume (BV/TV %), and percentage of total porosity were assessed using micro CT-scan. Breaking force of the femur was analysed by three point bending test using universal testing machine. Bone cellular structures were measured as static histomorphometric measurements. The fold changes of the expression of the genes were quantified with Real-time quantitative polymerase chain reaction (RT-qPCR).

The results of the research showed combination of mixed bacteria and Ca in G7 and *L. helveticus* in G5 as a single species have increased significantly BMD of the femur compared to Ovx-non treated group (0.96±0.02, and 0.90±0.05 vs 0.74±0.05 $gr.cm^{-3}$ respectively). *B. longum* treated group showed a higher Th.Th and a lower total porosity percentage ($p<0.05$) as compared to Ovx- non treated group. In addition, a higher strength of the femur have found in G7 (mixed bacteria + Ca) ($p<0.05$) (0.88±0.16 vs 0.54±0.11 $N.m^2$). Single species bacteria have increased the expression of runt related transcription factor (RUNX-2), bone morphometric protein2 (BMP-2), secreted protein acidic and rich in cysteine (SPARC), Collagen type 1 & 2 (Col 1&2), osterix (OSX), as well as calcium sensing receptor (CaSR) genes ($p<0.05$), and have decreased parathyroid receptor (PTHr) ($p<0.05$) as compared to Ovx non-treated group. However, mixed bacteria only altered the expression of CaSR, SPARC and PTHr. In addition, liver toxicity was not reported with the

selected dosage of bacteria consumption, since treated and non-treated Ovx were not significantly different in histology changes of the liver and liver serum biomarkers.

This study found that *L. helveticus*, and *B. longum* as a probiotics supplementation, have resistance to acid and bile in gastrointestinal tract. This meant that, selected bacteria after their passage through the gastrointestinal tract were alive. In this regards, single species showed a higher potential effect on changes of BMD of the femur as compared to mixed strains. Thus, *B. longum* and *L. helveticus*, specially as a single species have shown a great potential on bone health. Therefore, choosing appropriate probiotics as a single species or mixed strains is important to act on the target organ for intervention or treatment of the specific disease, such as bone in order to prevent osteoporosis.

Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia sebagai memenuhi keperluan untuk ijazah Doktor Falsafah

KESAN SUPPLEMEN PROBIOTIK TERHADAP PENYERAPAN KALSIUM DAN STATUS KESIHATAN TULANG PADA TIKUS OVARIEKTOMI

Oleh

KOLSOOM PARVANEH

Ogos 2015

Pengerusi: Prof Madya Rosita Binti Jamaluddin, PhD
Fakulti: Perubatan dan Sains Kesihatan

Osteoporosis adalah sejenis gangguan metabolisma tulang dengan kepadatan tulang yang rendah dan kemerosotan tisu tulang kebanyakannya disebabkan oleh penuaan atau menopaus. Kemungkinan berlakunya kepatahan pada pergelangan tangan, pinggul atau tulang belakang disebabkan osteoporosis adalah dianggap selari dengan risiko penyakit jantung. Maka, tujuan kajian semasa adalah untuk mengkaji kesan probiotik (*Bifidobacterium. longum*, dan *Lactobacillis. helveticus*) sebagai spesies tunggal atau probiotik dua strain terhadap penyerapan kalsium dan status kesihatan tulang pada tikus ovariektomi.

Bagi tujuan ini, 56 ekor tikus Sprague–Dawley yang berusia 10 minggu telah zdigunakan dalam kajian ini. Ovariektomi dua hala (Ovx) dan sham-ovx (S-ovx) telah dilakukan pada tikus tersebut. Selepas 2 minggu pembedahan, tikus tersebut dibahagikan secara rawak kepada 6 kumpulan Ovx dan satu kumpulan S-ovx. Proses pembedahan dan diet adalah seperti berikut: K1: S-ovx, K2: Ovx, K3: Ovx + Ca, K4: Ovx + *B. longum*, K5: Ovx + *L. helveticus*, K6: Ovx + campuran *L. helveticus* dan *B. longum*, dan K7: Ovx + campuran *L. helveticus* dan *B. longum* + Ca. Kencing, najis dan serum tikus dikumpulkan dan penyerapan serta pengekalan Ca ketara diukur. Penanda biomarker serum telah dinilai. Kandungan mineral tulang (BMC) telah dianalisis dari tulang femur menggunakan spektrofotometer penyerapan atom (AAS). BMD, struktur trabekula, peratusan jumlah tulang/jumlah keseluruhan (BV/TV%) dan peratusan jumlah keliangan telah dinilai menggunakan imbasan tomografi terkomputasi (CT) mikro. Daya pemecahan tulang femur dianalisis pada tiga titik lenturan dengan menggunakan mesin ujian universal. Struktur sel tulang diukur sebagai ukuran histomorfometrik statik. Perubahan kuantiti replikasi ekspresi gen diukur dengan reaksi rantaian polimerasi kuantitatif masa sebenar (RT-qPCR).

Keputusan kajian menunjukkan gabungan bakteria campuran dan Ca dalam K7 dan *L. helveticus* dalam K5 sebagai spesies tunggal telah meningkat dengan ketara BMD tulang femur berbanding dengan kumpulan bukan rawatan Ovx (0.96±0.02, dan 0.90±0.05 vs 0.74±0.05 gr.cm^{-3} masing-masing). Kumpulan yang dirawat dengan *B. longum* menunjukkan Th yang lebih tinggi. Th dan jumlah peratusan keliangan dalah lebih rendah ($p<0.05$) berbanding dengan kumpulan bukan rawatan Ovx. Di samping itu, kekuatan tulang femur yang lebih tinggi telah didapati pada K7 (bakteria campuran + Ca) ($p<0.05$) (0.88±0.16 vs 0.54±0.11 N.m^{2}). Bakteria spesies tunggal telah meningkat ungkapan runt berkaitan factor transkripsi (RUNX-2), tulang morfometrik protein2 (BMP-2), protein rembesan berasid dan kaya dengan cysteine (SPARC), jenis kolagen 1 & 2 (Kol 1&2), osterix (OSX) serta gen

kalsium sensing reseptor (CaSR) ($p<0.05$) dan pengurangan resoptor paratiroid (PTHr) ($p<0.05$) berbanding dengan kumpulan bukan rawatan Ovx. Walaubagaimanapun, bakteria campuran hanya mengubah ungkapan CaSR, SPARC dan PTHr. Di samping itu, ketoksikan buah hati tidak dilaporkan dengan dos penggunaan bakteria memandangkan tiada perbezaan ketara untuk perubahan histologi pada buan hati dan biomarker serum hati antara kumpulan rawatan dan bukan rawatan Ovx.

Kajian ini mendapati bahawa *L. helveticus*, dan *B. longum* sebagai supplemen probiotik yang mempunyai daya ketahanan terhadap asid dan cecair hempedu dalam saluran gastrousus. Ini bererti bahawa bakteria terpilih masih hidup selepas melalui saluran gastrousus. Sehubungan dengan ini, spesies tunggal menunjukkan kesan potensi yang lebih tinggi ke atas perubahan BMD tulang femur berbanding dengan probiotik campuran. Oleh itu, *B. longum* dan *L. helveticus* khas sebagai spesies tunggal telahpun menunjukkan potensi yang besar terhadap kesihatan tulang. Oleh itu, pemilihan probiotik yang bersesuaian sebagai species tunggal atau jenis campuran adalah penting untuk bertindak pada organ sasaran tertentu untuk intervensi atau rawatan penyakit tertentu seperti pada bahagian tulang untuk mengelakkan osteoporosis.

ACKNOWLEDGEMENTS

First and foremost thanks to GOD for the bounties and all the abilities he had granted me in facing with difficulties during my life to complete my PhD research.

My utmost regards and deep appreciations goes to my father (MohamadKazem Parvaneh) and my mother (Fatemeh Kazhaghand), for their unending love and support during my postgraduate years and have taken me to graduate and would certainly have not existed without them. To my lovely parents no word can express my gratefulness to accept and tolerate all these years of my postgraduate studies in another country and encouraging me. I owe my deepest gratitude to my lovely brothers Alireza and Hamidraze, and sisters Arezoo and Mandana whose love and support has been and will continue to be my inspiration, and I am so blessed to have such a caring and supportive family.

Warmest thanks to my supervisor, Associate Professor Dr. Rosita Jamaluddin, who had made available her support in a number of ways at every stage of my research. This thesis would not have been possible without her kind support. I would also like to thank my supervisory committee members; Associate Professor Dr. Zuraini and Dr. Zuriati for their kindness and insightful comments. It is a pleasure and special thanks to Dr. Angela NG for her kind guidance, encourage, expert comments and her direction help.

I have to confess that I am in debt to of all my lovely friends for supporting throughout my rough time, for their guidance in completing my PhD research. They provide me a stimulating and fun environment that inspired me to overcome some of the challenges of living abroad. I would also like to acknowledge and thanks to laboratories of Biochemistry, and Nutrition from Faculty of Medicine and Health Sciences, UPM, and Tissue Engineering Centre of UKM for their help and assistance.

This thesis was submitted to the Senate of Universiti Putra Malaysia and has been accepted as fulfillment of the requirement for the degree of Doctor of Philosophy. The members of the Supervisory Committee were as follows:

Rosita binti Jamaluddin, PhD
Associate Professor
Faculty of Medicine and health Sciences
Universiti Putra Malaysia
(Chairman)

Zuraini binti Ahmad, PhD
Associate Professor
Faculty of Medicine and health Sciences
Universiti Putra Malaysia
(Member)

Zuriati Ibrahim, PhD
Senior Lecturer
Faculty of Medicine and health Sciences
Universiti Putra Malaysia
(Member)

Angela Ng Min Hwei, PhD
Senior Lecturer
Faculty of Medicine
Universiti Kebangsaan Malaysia
(Member)

BUJANG KIM HUAT, PhD
Professor and Dean
School of Graduate Studies
Universiti Putra Malaysia

Date:

Declaration by graduate student

I hereby confirm that:

- this thesis is my original work;
- quotations, illustrations and citations have been duly referenced;
- this thesis has not been submitted previously or concurrently for any other degree at any other institutions;
- intellectual property from the thesis and copyright of thesis are fully -owned by Universiti Putra Malaysia, as according to the Universiti Putra Malaysia (Research) Rules 2012;
- written permission must be obtained from supervisor and the office of Deputy Vice-Chancellor (Research and Innovation) before thesis is published (in the form of written, printed or in electronic form) including books, journals, modules, proceedings, popular writings, seminar papers, manuscripts, posters, reports, lecture notes, learning modules or any other materials as stated in the Universiti Putra Malaysia (Research) Rules 2012 ;
- there is no plagiarism or data falsification/fabrication in the thesis, and scholarly integrity is upheld as according to the Universiti Putra Malaysia (Graduate Studies) Rules 2003 (Revision 2012-2013) and the Universiti Putra Malaysia (Research) Rules 2012. The thesis has undergone plagiarism detection software.

Signature:_____ Date: _____

Name and Matric No.: Kolsoom Parvaneh, GS33466

Declaration by Member of Supervisory Committee

This is to inform that:

- the research conducted and the writing of this thesis was under our supervision;
- supervision responsibilities as stated in the Universiti Putra Malaysia (Graduate Studies) Rules 2003 (Revision 2012-2013) are adhered to.

Signature: _____	Signature: _____
Name of Chairman of Supervisory Committee: Rosita binti Jamaluddin, PhD	Name of Member of Supervisory Committee: Zuraini binti Ahmad, PhD
Signature: _____	Signature: _____
Name of Member of Supervisory Committee: Zuriati Ibrahim, PhD	Name of Member of Supervisory Committee: Angela Ng Min Hwei, PhD

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF ABBREVIATIONS

AAS	Atomic absorption spectrophotometer
AC	Attenuation coefficient
ACUC	Animal Care and Use Committee
ALP	Alkaline phosphatase
ALT	Alanine aminotransferase
ANOVA	Analysis of variance
Ang II	Angiotensin II
ASBMR	American Society of Bone Mineral Research
AST	Aspartate aminotransferase
ATCC	American Type Culture Collection
B. longum	*Bifidobacterium longum*
BMC	Bone mineral content
BMD	Bone mass density
BMP	Bone morphogenetic proteins
BMPRs	Bone morphogenetic proteins receptors
BMU	Basic multicellular units
bp	Base pair
BSM	Bifidus Selective Medium
BV	Bone volume
Ca	Calcium
CA	cytosine-adenin
CaHA	Calcium hydroxyapatite
CALCR	Calcitonin receptor
CaSR	Calcium-sensing receptor
CFU	Colony forming unit
Col 1 & 2	Collagen type 1 α 2
CPP	Casein phospho peptides
CTX	C-terminal telopeptid
D2	Ergocalciferol
D3	Cholecalciferol
DEXA	Dual-energy X-ray absorptiometry
ELISA	Enzyme-linked immunosorbeny assay
ER	Estrogen receptors
ERE	Estrogen response elements
ES/BS	Eroded surface/bone surface
F	Force
FGF23	Fibroblast growth factor 23
g	Gram
GAPDH	Glyceraldehyde-3-phosphate dehydrogenase
GH	Growth hormone
GI	Gastrointestinal
GRAS	Generally regards as safe
H & E	Haematoxylin and eosin
HRT	Hormone replacement therapy
I	Moment inertia
IGF	Insulin-like growth factor
IGFs	Insulin-like growth factors
IL6	Interlukin 6
IP	Intra-peritoneally
IPP	Isoleucyl prolyl proline
L. helveticus	*Lactobacillus helveticus*

M	Moment
MAPK	Mitogen activating protein kinase
Mg	Magnesium
mg/L	Miligeram/Liter
mm	millimeter
mmol/L	Milimol/Liter
MRS	Man Rogosa and Sharpe
N	Nioton
NF-kB	Nuclear factor kappa B cells
ObS/BS	Osteoblast surface/bone surface
OC	Osteocalcin
OcS/BS	Osteoclast surface/bone surface
OD	Optical density
OPG	Osteoprotegerin
OS/BS	Osteoid surface/bone surface
OSF2	Osteoblast-specific factor 2
OSX	Osterix
OV/BV	Osteoid volume/bone volume
Ovx	Ovariectomized
P	phosphorus
POStn	Periostin
PTH	Parathyroid hormone
QC	Quality control
RANKL	Receptor activator of nuclear factor kappa-B ligand
RT-qPCR	Real-Time quantitative polymerase chain reaction
RUNX	Runt-related transcription factor
S	Stress
SCFA	Short chain fatty acid
SEM	Standard error from mean
SHBG	Sex hormone binding globulin
Sovx	Sham-ovariectomized
SPARC	Secreted protein acidic and rich in cysteine
TFTC	Too few to count
TGF-β	Tumor growth factor-beta
Th.N	Trabecular number
Th.Sp	Trabecular separation
Th.Th	Trabecular thickness
TMTC	Too many to count
TNF	Tumor necrosis factor
TV	Total volume
U	Unit
VDR	Vitamin D receptor
VPP	Valyl prolyl proline
Zn	Zinc

CHAPTER 1

INTRODUCTION

1.1 Background of Study

Osteoporosis is a bone metabolism disorder, which is explained by low bone mass and deterioration of the bone tissue, which leads to increase in bone weakness, and consequently increases the possibility of fracture (Metcalfe, 2008). Seventy five million people in Europe, Japan and the United State are affected with osteoporosis. The possibility of wrist, hip or spine fracture due to the osteoporosis is estimated to be parallel to the risk of heart disease (Kai, Anderson, & Lau, 2003). Mostly osteoporosis is due to aging or the menopause in women (Ji & Yu, 2015). The life span of the women has risen worldwide (Wong & Liyana, 2007). Therefore, one third of women's life spans are spent in menopause, thus it is necessary to reduce the inverse effect of the menopause on women's health.

Sufficient calcium intake has been reported to support bone growth and reduce bone loss during the aging process. Milk and milk products supply 75% of calcium needed for the body in Western countries, but milk consumption has reduced over the past decade (Woolf & Pfleger, 2003). Some of the specialists believes that one of the effective treatments for osteoporosis specially, is the hormone replacement therapy (HRT). However, because of the side effects of HRT, this treatment is not generally accepted due to low compliance, unwillingness and aversion of many women. Moreover, long duration usage of HRT therapy leads to increase the possibility of growing some types of cancer (Picherit et al., 2000).

Some of the alternative ways for preventing and treating osteoporosis are being developed and recommended by research scientists worldwide. Exercise, including walking and light running has been reported to influence in maintaining bone mass density (BMD) (Downey & Siegel, 2006). Another possibility way for fighting osteoporosis is probiotic consumption.

The general definition of probiotics is "live micro-organisms administered in adequate amounts which confer a beneficial physiological effect on the host'' (Reid et al., 2003). During the last decade of the 20th century, when the term of functional foods were defined clearly, probiotics became popular. Nowadays, the public aware of probiotics as dietary supplements and the components of bio-yoghurts (Woolf & Pfleger, 2003).

1.2 Problem Statement

Osteoporosis is the most prominent bone disease and it is considered as a serious public health concern, because of its prevalence worldwide. Generally, in The United States and in the Europe, 30% of all postmenopausal women suffer osteoporosis. It is estimated that, approximately one out of five American women over the age of 50 suffer from this disease. Almost 50% of all women and 15-30% of men over 50 years of age, face risk of fractures of the hip, wrist, or vertebra due to the osteoporosis. The ageing populations as well as postmenopausal condition in women are considered as a major contributor to the osteoporosis (Woolf & Pfleger, 2003).

One of the most common issues faced by osteoporosis patient is hip fracture (Kanis et al., 2007). Risk of hip fracture among women due to menopausal and osteoporosis, are more rather than men. Risk of fracture for 50 years old women was reported at 53.2% versus 20.7% risk of fracture for men for the same age (Staa, Dennison, Leufkens, & Cooper, 2001). The rate of hip fractures is rising throughout the world, and the researchers estimate this rate by 2050 to be 6.3 million worldwide. The rise will affect in particular, Asian countries. In addition, more than 50% of all hip fractures worldwide are estimated to be from Asia by 2050 (Dhanwal, Dennison, Harvey, & Cooper, 2011). The Asian population has shorter height and less body mass index (Babbar et al., 2006). It is shown that lower height and body mass index can increase the likelihood of osteoporosis disease (Munaisinghe, Botea, & Edelson, 2002). In addition, geographical variations in Asia have moderate effect in the incidence of hip fracture. In comparison of four countries in Asia, such as Singapore, Hong Kong, Thailand and Malaysia, the rates of hip fracture in Singapore and Hong Kong were similar. However, in Thailand were 60% and in Malaysia were 50% of the rates in Hong Kong (Lau et al., 2001).

One of the ways of treatment for menopausal osteoporosis is HRT, but this treatment have shown the side effects such as breast cancer (Hou et al., 2013; Marsden, 2002; Picherit et al., 2000). Therefore, an alternative way of reducing or fighting osteoporosis is needed. In this regard, probiotic is now growing in importance (Scholz-Ahrens et al., 2007). Some previous studies have shown specific changes of gut microbiota through the use of probiotics and have a possible effect on the improvement of bone health in ovariectomized (Ovx) rats which stimulate postmenopausal conditions (Jirillo et al., 2012; McCabe, Irwin, Schaefer, & Britton, 2013; Scholz-Ahrens et al., 2007). Additionally, Chiang and Pan, (2011) have shown the effect of *Lactobacillus plantarum* and *Lactobacillus paracasei* with the dosage of 10^8 CFU/ml on trabecular bone. Besides, Scholz-Ahrens et al., (2007) carried out on *Lactobacillus casei*, *Lactobacillus reuteri,* and *Lactobacillus gasseri* and reported these strain of probiotics stimulate higher apparent calcium absorption among the rats, as well as 35% higher bone weight.

Although some treatments have shown to reduce osteoporosis among menopausal women, but still osteoporosis and fracture risk is one of the main problems among post-menopausal women. This indicates a high demand for introducing prophylactic measures to reduce the inverse health effects of the menopause such as osteoporosis among women or reducing osteoporosis prevalence. Therefore, the present study aimed to determine the effect of two different probiotic strain (*L. helveticus and B. longum*) as a single or mixed strain on Ca absorption and bone health status in Ovx rats.

1.3 Significance of the Study

Although a few earlier studied reported on the effect of probiotics on bone health (Rodrigues et al., 2012; Chiang & Pan, 2011; Kim et al., 2009), but little known about the effectiveness of probiotic as a single strain compared to mixed strain. This study is the first, which assessed the potential effect of double strains of probiotic (*Bifidobacterium longum* and *Lactobacillus helveticus*), and compared with the effect of single strain on Ca bioavailability and bone health status. This study also is the first to evaluate the effectiveness of Ca supplementation on the action of *B. longum* and *L. helveticus*.

Besides, the knowledge about the genetic factors of osteoporosis are important, because, they may propose the use of genetic markers for the evaluation of the risk of osteoporosis and this gives an opportunity to recognize new targets for new drugs or supplements, or fortification of food for the prevention or treatment of osteoporosis. Due to this, the expression of some estrogenic gens was evaluated in this study. In addition, variables related to liver function were assessed and the toxicity of consumption of the selected dosage of bacteria on liver system reported. Determinant of toxicity of the proposed drug or supplement is important, that it is to be accepted as a diet supplement.

In fact, this research can bring a new knowledge on the potential effect of probiotics bacteria as a single species, and compare it with double strains on Ca absorption and bone health status as well as its safety with the selected dosage as a dietary supplement.

1.4 Objectives

1.4.1 General Objective

To investigate the effect of double strain probiotics supplementation (*L. helveticus* and *B. longum*) and compared with single strain on calcium absorption and bone health status in Ovx-induced bone loss rats.

1.4.2 Specific Objectives

I. To determine the effect of *L. helveticus* and *B. longum* as a single species or double strain and Ca supplementation on BMD, BMC and bone cellular structure changes of Ovx- induced bone loss rats.
II. To determine the effect of *L. helveticus* and *B. longum* as a single species or double strain and Ca supplementation on the strength of the bone of Ovx-induced bone loss rats.
III. To measure Ca excretion from urine and feces of the rats and evaluate apparent calcium absorption and calcium retention among control and intervention groups.
IV. To determine the effect of *L. helveticus* and *B. longum* supplementation as a single species or double strain on serum indicator of bone formation and resorption.
V. To investigate the effect of probiotics supplementation (*B. longum* and *L. heveticus*) as a single species or double strain on bone histology in Ovx-induced-bone loss rats.
VI. To determine and compare the quantity expression of some genes affected by *B. longum* and *L. heveticus* as a single species or in combination in Ovx-induced-bone loss rats.
VII. To determine the liver toxicity of selected dosage of probiotics supplementation (*B. longum* and *L. heveticus*) in Ovx induced-bone loss rats and compare with control groups.

1.5 Research Question

a) Does probiotics supplementation (*L. helveticus* or *B. longum*) increase Ca absorption and bone mass density in Ovx induced-bone loss rats?
b) Does supplementation with double strain probiotic (*L. helveticus* and *B. longum*) increase Ca absorption and bone mass density more than single strain?

1.6 Research Hypothesis

a) This study hypothesized that, *L. helveticus* or *B. longum* increase Ca absorption and bone mass density in Ovx rats and double strain of probiotic supplementation (*L. helveticus* and *B. longum*) boost calcium absorption and bone mass density more as compared to single strain
b) Supplementation of probiotics with Ca has more effect on Ca absorption and bone mass density as compared to probiotic alone.

REFERENCES

Abdou, A. M., Watabe, K., Yamane, T., Isono, T., Okamura, Y., Kawahito, S., Kim, M. (2014). Antiosteoporotic effect of orally administered yolk-derived peptides on bone mass in women. *Food Science & Nutrition*, *2*, 193–9.

Abdul-Majeed, S., Mohamed, N., & Soelaiman, I.-N. (2012). Effects of tocotrienol and lovastatin combination on osteoblast and osteoclast activity in estrogen-deficient osteoporosis. *Evidence-Based Complementary and Alternative Medicine : eCAM*, *2012*, 960742.

Abdul-Majeed, S., Mohamed, N., & Soelaiman, I.-N. (2013). A review on the use of statins and tocotrienols, individually or in combination for the treatment of osteoporosis. *Current Drug Targets*, *14*, 1579–90.

Acker, S. A., Koymans, L. M., & Bast, A. (1993). Molecular pharmacology of vitamin E: structural aspects of antioxidant activity. *Free Radical Biology & Medicine*, *15*, 311–28.

Ahmad, A. M., Hopkins, M. T., Fraser, W. D., Ooi, C. G., Durham, B. H., & Vora, J. P. (2003). Parathyroid hormone secretory pattern, circulating activity, and effect on bone turnover in adult growth hormone deficiency. *Bone*, *32*, 170–9.

Akhter, M. P., Iwaniec, U. T., Haynatzki, G. R., Fung, Y. K., Cullen, D. M., & Recker, R. R. (2003). Effects of nicotine on bone mass and strength in aged female rats. *Journal of Orthopaedic Research*, *21*, 14–9.

Alempijevic, T., & Kovacevic, N. (2007). Right liver lobe diameter:albumin ratio: a new non-invasive parameter for prediction of oesophageal varices in patients with liver cirrhosis (preliminary report). *Gut*, *56*, 1166–7.

Alma Y. Parra-Torres, M. V.-F. L. O. and R. V.-C. (2013). *Topics in Osteoporosis*. (M. Valds-Flores, Ed.). InTech. doi:10.5772/50259

Al-Majed, A. A., Al-Omar, F. A., & Nagi, M. N. (2006). Neuroprotective effects of thymoquinone against transient forebrain ischemia in the rat hippocampus. *European Journal of Pharmacology*, *543*, 40–7.

Alonso, C. G., Curiel, M. D., Carranza, F. H., Cano, R. P., & Peréz, A. D. (2000). Femoral bone mineral density, neck-shaft angle and mean femoral neck width as predictors of hip fracture in men and women. Multicenter Project for Research in Osteoporosis. *Osteoporosis International*, *11*, 714–20.

Al-Rejaie, S. S. (2013). Thymoquinone Treatment Alleviate Ovariectomy-Induced Hepatic Oxidative Damage in Rats. *Journal of Applied Pharmaceutical Science.*, *3*, 126–131.

Asemi, Z., & Esmaillzadeh, A. (2013). Effect of daily consumption of probiotic yoghurt on serum levels of calcium, iron and liver enzymes in pregnant women. *International Journal of Preventive Medicine*, *4*, 949–55.

Babazade, D., Vaddatpour, T., Nikpiran, H., Jafargholipour, M. A., & Vahdatpour, S. (2011). Effects of probiotic, prebiotic and synbiotic intake on blood enzymes and performance of Japanese quails (Coturnix Japonica). *The Indian Journal of Animal Sciences*, 81,8.

Babbar, R. K., Handa, A. B., Lo, C., Guttmacher, S. J., Shindledecker, R., Chung, W., Dixon, L. B. (2006). Bone health of immigrant Chinese women living in New York City. *Journal of Community Health*, *31*, 7–23.

Babinsky, V. N., Hannan, F. M., Youhanna, S. C., Maréchal, C., Jadoul, M., Devuyst, O., & Thakker, R. V. (2015). Association studies of calcium-sensing receptor (CaSR) polymorphisms with serum concentrations of glucose and phosphate, and vascular calcification in renal transplant recipients. *PloS One*, *10*, e0119459.

Baek, K. H., Oh, K. W., Lee, W. Y., Lee, S. S., Kim, M. K., Kwon, H. S., Kang, M. Il. (2010). Association of oxidative stress with postmenopausal osteoporosis and the effects of hydrogen peroxide on osteoclast formation in human bone marrow cell cultures. *Calcified Tissue International*, *87*, 226–35.

Baek, W.-Y., Lee, M.-A., Jung, J. W., Kim, S.-Y., Akiyama, H., de Crombrugghe, B., & Kim, J.-E. (2009). Positive regulation of adult bone formation by osteoblast-specific transcription factor osterix. *Journal of Bone and Mineral Research , 24*, 1055–65.

Bai, X., Miao, D., Li, J., Goltzman, D., & Karaplis, A. C. (2004). Transgenic mice overexpressing human fibroblast growth factor 23 (R176Q) delineate a putative role for parathyroid hormone in renal phosphate wasting disorders. *Endocrinology*, *145*, 5269–79.

Baldock, P. A., Morris, H. A., Need, A. G., Moore, R. J., & Durbridge, T. C. (1998). Variation in the short-term changes in bone cell activity in three regions of the distal femur immediately following ovariectomy. *Journal of Bone and Mineral Research*, *13*, 1451–7.

Banerjee, A., Chitnis, U. B., Jadhav, S. L., Bhawalkar, J. S., & Chaudhury, S. (2009). Hypothesis testing, type I and type II errors. *Industrial Psychiatry Journal*, *18*, 127–31.

Barber, P. J. (2004). Disorders of the parathyroid glands. *Journal of Feline Medicine and Surgery*, *6*, 259–69.

Barco Romero, C. M., Manrique Arija, S., & Rodríguez Pérez, M. (2012). Biochemical markers in osteoporosis: usefulness in clinical practice. *Reumatología Clinica*, *8*, 149–52.

Baron, M. (2009). A patented strain of Bacillus coagulans increased immune response to viral challenge. *Postgraduate Medicine*, *121*, 114–8.

Barrère, F., van Blitterswijk, C. A., & de Groot, K. (2006). Bone regeneration: molecular and cellular interactions with calcium phosphate ceramics. *International Journal of Nanomedicine*, *1*, 317–32.

Ben-Dov, I. Z., Galitzer, H., Lavi-Moshayoff, V., Goetz, R., Kuro-o, M., Mohammadi, M., Silver, J. (2007). The parathyroid is a target organ for FGF23 in rats. *The Journal of Clinical Investigation*, *117*, 4003–8.

Bennett, T., Desmond, A., Harrington, M., McDonagh, D., FitzGerald, R., Flynn, A., & Cashman, K. D. (2000). The effect of high intakes of casein and casein phosphopeptide on calcium absorption in the rat. *The British Journal of Nutrition*, *83*, 673–80.

Beral, V., Bull, D., Green, J., & Reeves, G. (2007). Ovarian cancer and hormone replacement therapy in the Million Women Study. *Lancet*, *369*, 1703–10.

Bergillos-Meca, T., Navarro-Alarcón, M., Cabrera-Vique, C., Artacho, R., Olalla, M., Giménez, R., Ruiz-López, M. D. (2013). The probiotic bacterial strain Lactobacillus fermentum D3 increases in vitro the bioavailability of Ca, P, and Zn in fermented goat milk. *Biological Trace Element Research*, *151*, 307–14.

Bevilacqua, M., Dominguez, L. J., Righini, V., Valdes, V., Toscano, R., Sangaletti, O., Bianchi-Porro, G. (2005). Increased gastrin and calcitonin secretion after oral calcium or peptones administration in patients with hypercalciuria: a clue to an alteration in calcium-sensing receptor activity. *The Journal of Clinical Endocrinology and Metabolism*, *90*, 1489–94.

Bezkorovainy, A. (2001). Probiotics: determinants of survival and growth in the gut. *The American Journal of Clinical Nutrition*, *73*, 399S–405S.

Black, D. M., Steinbuch, M., Palermo, L., Dargent-Molina, P., Lindsay, R., Hoseyni, M. S., & Johnell, O. (2001). An assessment tool for predicting fracture risk in postmenopausal women. *Osteoporosis International*, 12, 519–28.

Bonjour, J.-P., Benoit, V., Payen, F., & Kraenzlin, M. (2013). Consumption of yogurts fortified in vitamin D and calcium reduces serum parathyroid hormone and markers of bone resorption: a double-blind randomized controlled trial in institutionalized elderly women. *The Journal of Clinical Endocrinology and Metabolism*, *98*, 2915–21.

Bonjour, J.-P., Chevalley, T., Ferrari, S., & Rizzoli, R. (2009). The importance and relevance of peak bone mass in the prevalence of osteoporosis. *Salud Pública de México*, *51 Suppl 1*, S5–17.

Bonnet, N., Conway, S. J., & Ferrari, S. L. (2012). Regulation of beta catenin signaling and parathyroid hormone anabolic effects in bone by the

matricellular protein periostin. *Proceedings of the National Academy of Sciences of the United States of America*, *109*, 15048–53.

Bonnet, N., & Ferrari, S. L. (2010). Exercise and the skeleton: How it works and what it really does. *IBMS BoneKEy*, *7*, 235–248.

Booth, S. L., Tucker, K. L., Chen, H., Hannan, M. T., Gagnon, D. R., Cupples, L. A., Kiel, D. P. (2000). Dietary vitamin K intakes are associated with hip fracture but not with bone mineral density in elderly men and women. *American Journal of Clinical Nutrition*, *71*, 1201–1208.

Borriello, S. P., Hammes, W. P., Holzapfel, W., Marteau, P., Schrezenmeir, J., Vaara, M., & Valtonen, V. (2003). Safety of probiotics that contain lactobacilli or bifidobacteria. *Clinical Infectious Diseases*, *36*, 775–80.

Bouxsein, M. L., Boyd, S. K., Christiansen, B. A., Guldberg, R. E., Jepsen, K. J., & Müller, R. (2010). Guidelines for assessment of bone microstructure in rodents using micro-computed tomography. *Journal of Bone and Mineral Research*, *25*, 1468–86.

Bradshaw, A. D., & Sage, E. H. (2001). SPARC, a matricellular protein that functions in cellular differentiation and tissue response to injury. *The Journal of Clinical Investigation*, *107*, 1049–54.

Brown, M. A., Haughton, M. A., Grant, S. F., Gunnell, A. S., Henderson, N. K., & Eisman, J. A. (2001). Genetic control of bone density and turnover: role of the collagen 1alpha1, estrogen receptor, and vitamin D receptor genes. *Journal of Bone and Mineral Research*, *16*, 758–64.

Burger, H. G. (2002). Hormonal Changes in the Menopause Transition. *Recent Progress in Hormone Research*, *57*, 257–275.

Burguera, B., Hofbauer, L. C., Thomas, T., Gori, F., Evans, G. L., Khosla, S., Turner, R. T. (2013). Leptin Reduces Ovariectomy-Induced Bone Loss in Rats. *Endocrinology*, *142*, 3546-3552.

Burr, D. B., Hirano, T., Turner, C. H., Hotchkiss, C., Brommage, R., & Hock, J. M. (2001). Intermittently administered human parathyroid hormone(1-34) treatment increases intracortical bone turnover and porosity without reducing bone strength in the humerus of ovariectomized cynomolgus monkeys. *Journal of Bone and Mineral Research*, *16*, 157–65.

Campbell, I. (2011). Thyroid and parathyroid hormones and calcium homeostasis. *Anaesthesia & Intensive Care Medicine*, *12*, 465–468.

Capcarova, M., Hascik, P., Kolesarova, A., Kacaniova, M., Mihok, M., & Pal, G. (2011). The effect of selected microbial strains on internal milieu of broiler chickens after peroral administration. *Research in Veterinary Science*, *91*, 132–7.

Capcarova, M., Weiss, J., Hrncar, C., Kolesarova, A., & Pal, G. (2010). Effect of Lactobacillus fermentum and Enterococcus faecium strains on internal milieu, antioxidant status and body weight of broiler chickens. *Journal of Animal Physiology and Animal Nutrition*, *94*, e215–24.

Carafoli, E. (2004). Calcium-mediated cellular signals: a story of failures. *Trends in Biochemical Sciences*, *29*, 371–9.

Carissa M. Thomas, J. V. (2010). Probiotics-host communication: Modulation of signaling pathways in the intestine. *Gut Microbes*. *1*, 148-163.

Carrillo-López, N., Román-García, P., Rodríguez-Rebollar, A., Fernández-Martín, J. L., Naves-Díaz, M., & Cannata-Andía, J. B. (2009). Indirect regulation of PTH by estrogens may require FGF23. *Journal of the American Society of Nephrology*, *20*, 2009–17.

Cashman, K. D. (2002). Calcium intake, calcium bioavailability and bone health. *The British Journal of Nutrition*, *87*, S169–77.

Cauley, J. A., Robbins, J., Chen, Z., Cummings, S. R., Jackson, R. D., LaCroix, A. Z., Watts, N. B. (2003). Effects of estrogen plus progestin on risk of fracture and bone mineral density: the Women's Health Initiative randomized trial. *The Journal of the American Medical Association*, *290*, 1729–38.

Cenesız, S., Yaman, H., Ozcan, A., Kart, A., & Karademır, G. (2008). Effects of kefir as a probiotic on serum cholesterol, total lipid, aspartate amino transferase and alanine amino transferase activities in broiler chicks. *Medycyna Weterynaryjna*, *64*, 168–170.

Chailurkit, L., Ongphiphadhanakul, B., Piaseu, N., Saetung, S., & Rajatanavin, R. (2001). Biochemical Markers of Bone Turnover and Response of Bone Mineral Density to Intervention in Early Postmenopausal Women: An Experience in a Clinical Laboratory. *Clinical Cheistry.*, *47*, 1083–1088.

Champs, C., Maroncle, N., Balestrino, D., Rich, C., & Forestier, C. (2003). Persistence of colonization of intestinal mucosa by a probiotic strain, Lactobacillus casei subsp. rhamnosus Lcr35, after oral consumption. *Journal of Clinical Microbiology*, *41*, 1270–3.

Chang, K.-L., Hu, Y.-C., Hsieh, B.-S., Cheng, H.-L., Hsu, H.-W., Huang, L.-W., & Su, S.-J. (2013). Combined effect of soy isoflavones and vitamin D3 on bone loss in ovariectomized rats. *Nutrition*, *29*, 250–7.

Chen, Y. J., Son, K. S., Min, B. J., Cho, J. H., Kwon, O. S., & Kim, I. H. (2005). Effects of Dietary Probiotic on Growth Performance, Nutrients Digestibility, Blood Characteristics and Fecal Noxious Gas Content in Growing Pigs. *Asian-Australasian Journal of Animal Sciences*, *18*, 1464–1468.

Chen, Y., Wang, S., Bu, S., Wang, Y., Duan, Y., & Yang, S. (2011). Treadmill training prevents bone loss by inhibition of PPARγ expression but not

promoting of Runx2 expression in ovariectomized rats. *European Journal of Applied Physiology*, *111*, 1759–67.

Cheng, S., Lyytikainen, A., Kroger, H., Lamberg-Allardt, C., Alen, M., Koistinen, A., Tylavsky, F. (2005). Effects of calcium, dairy product, and vitamin D supplementation on bone mass accrual and body composition in 10-12-y-old girls: a 2-y randomized trial. *American Journal of Clinical Nutrition*, *82*, 1115–1126.

Chiang, S.-S., & Pan, T.-M. (2011). Antiosteoporotic effects of Lactobacillus -fermented soy skim milk on bone mineral density and the microstructure of femoral bone in ovariectomized mice. *Journal of Agricultural and Food Chemistry*, *59*, 7734–42.

Chow, E. C. Y., Quach, H. P., Vieth, R., & Pang, K. S. (2013). Temporal changes in tissue 1α,25-dihydroxyvitamin D3, vitamin D receptor target genes, and calcium and PTH levels after 1,25(OH)2D3 treatment in mice. *American Journal of Physiology Endocrinology and Metabolism*, *304*, E977–89.

Čitar, M., Hacin, B., Tompa, G., Štempelj, M., Rogelj, I., Dolinšek, J., Matijašić, B. B. (2015). Human intestinal mucosa-associated Lactobacillus and Bifidobacterium strains with probiotic properties modulate IL-10, IL-6 and IL-12 gene expression in THP-1 cells. *Beneficial Microbes*, *6*, 325–36.

Clarke, B. (2008). Normal bone anatomy and physiology. *Clinical Journal of the American Society of Nephrology*, *3*, S131–9.

Coetzee, M., & Kruger, M. C. (2004). Osteoprotegerin-receptor activator of nuclear factor-kappaB ligand ratio: a new approach to osteoporosis treatment? *Southern Medical Journal*, *97*, 506–11.

Collado, M. C., Grześkowiak, Ł., & Salminen, S. (2007). Probiotic strains and their combination inhibit in vitro adhesion of pathogens to pig intestinal mucosa. *Current Microbiology*, *55*, 260–5.

Compston, J. (2011). Age-related changes in bone remodelling and structure in men: histomorphometric studies. *Journal of Osteoporosis*, *2011*, 108324.

Cranney, A., Guyatt, G., Griffith, L., Wells, G., Tugwell, P., & Rosen, C. (2002). Meta-analyses of therapies for postmenopausal osteoporosis. IX: Summary of meta-analyses of therapies for postmenopausal osteoporosis. *Endocrine Reviews*, *23*, 570–8.

Crews, D., & McLachlan, J. A. (2006). Epigenetics, evolution, endocrine disruption, health, and disease. *Endocrinology*, *147*, S4–10.

Cummings, S. R., Browner, W. S., Bauer, D., Stone, K., Ensrud, K., Jamal, S., & Ettinger, B. (1998). Endogenous hormones and the risk of hip and vertebral fractures among older women. Study of Osteoporotic Fractures Research Group. *The New England Journal of Medicine*, *339*, 733–8.

Dani, C., Biadaioli, R., Bertini, G., Martelli, E., & Rubaltelli, F. F. (2002). Probiotics feeding in prevention of urinary tract infection, bacterial sepsis and necrotizing enterocolitis in preterm infants. A prospective double-blind study. *Biology of the Neonate*, *82*, 103–8.

Davidson, M. K., Lindsey, J. R., & Davis, J. K. (1987). Requirements and selection of an animal model. *Israel Journal of Medical Sciences*, *23*, 551–5.

Delany, A. M., & Hankenson, K. D. (2009). Thrombospondin-2 and SPARC/osteonectin are critical regulators of bone remodeling. *Journal of Cell Communication and Signaling*, *3*, 227–38.

Dempster, D. W., Birchman, R., Xu, R., Lindsay, R., & Shen, V. (1995). Temporal changes in cancellous bone structure of rats immediately after ovariectomy. *Bone*, *16*, 157–61.

Deyhim, F., Stoecker, B. J., Brusewitz, G. H., & Arjmandi, B. H. (2003). The effects of estrogen depletion and isoflavones on bone metabolism in rats. *Nutrition Research*, *23*, 123–130.

Dhanwal, D. K., Dennison, E. M., Harvey, N. C., & Cooper, C. (2011). Epidemiology of hip fracture: Worldwide geographic variation. *Indian Journal of Orthopaedics*, *45*, 15–22.

Donato, R. (2001). S100: a multigenic family of calcium-modulated proteins of the EF-hand type with intracellular and extracellular functional roles. *The International Journal of Biochemistry & Cell Biology*, *33*, 637–68.

Downey, P. A., & Siegel, M. I. (2006). Bone biology and the clinical implications for osteoporosis. *Physical Therapy*, *86*, 77–91.

Drouault, S., Corthier, G., Ehrlich, S. D., & Renault, P. (1999). Survival, physiology, and lysis of Lactococcus lactis in the digestive tract. *Applied and Environmental Microbiology*, *65*, 4881–6.

Du, Z., Chen, J., Yan, F., Doan, N., Ivanovski, S., & Xiao, Y. (2013). Serum bone formation marker correlation with improved osseointegration in osteoporotic rats treated with simvastatin. *Clinical Oral Implants Research*, *24*, 422–7.

Duque, G., El Abdaimi, K., Macoritto, M., Miller, M. M., & Kremer, R. (2002). Estrogens (E2) regulate expression and response of 1,25-dihydroxyvitamin D3 receptors in bone cells: changes with aging and hormone deprivation. *Biochemical and Biophysical Research Communications*, *299*, 446–54.

Eastell, R., & Lambert, H. (2002). Strategies for skeletal health in the elderly. *The Proceedings of the Nutrition Society*, *61*, 173–80.

Egan H, K. R. and S. R. (1981). *Pearson's chemical analysis of food* (8th edition). Churchill Livingstone, London. 26–28.

Elli, M., Callegari, M. L., Ferrari, S., Bessi, E., Cattivelli, D., Soldi, S., Antoine, J.-M. (2006). Survival of yogurt bacteria in the human gut. *Applied and Environmental Microbiology*, *72*, 5113–7.

Engelbrektson, A. L., Korzenik, J. R., Sanders, M. E., Clement, B. G., Leyer, G., Klaenhammer, T. R., & Kitts, C. L. (2006). Analysis of treatment effects on the microbial ecology of the human intestine. *FEMS Microbiology Ecology*, *57*, 239–50.

Eriksen, E. F. (2010). Cellular mechanisms of bone remodeling. *Reviews in Endocrine & Metabolic Disorders*, *11*, 219–27.

Ettinger, B., Genant, H. K., & Cann, C. E. (1987). Postmenopausal bone loss is prevented by treatment with low-dosage estrogen with calcium. *Annals of Internal Medicine*, *106*, 40–5.

Fang, Y., van Meurs, J. B. J., Bergink, A. P., Hofman, A., van Duijn, C. M., van Leeuwen, J. P. T. M., Uitterlinden, A. G. (2003). Cdx-2 polymorphism in the promoter region of the human vitamin D receptor gene determines susceptibility to fracture in the elderly. *Journal of Bone and Mineral Research*, *18*, 1632–41.

Farinha, J. R.-M. (2005). *Counting Bacteria*. *Richland College*.

Faroqui, S., Levi, M., Soleimani, M., & Amlal, H. (2008). Estrogen downregulates the proximal tubule type IIa sodium phosphate cotransporter causing phosphate wasting and hypophosphatemia. *Kidney International*, *73*, 1141–50.

Felsenfeld, A. J., Rodríguez, M., & Aguilera-Tejero, E. (2007). Dynamics of parathyroid hormone secretion in health and secondary hyperparathyroidism. *Clinical Journal of the American Society of Nephrology*, *2*, 1283–305.

Filipov, O. (2014). Femoral neck fractures – biological aspects and risk factors. *Journal of IMAB*, *20*, 513–515.

Finkelstein, J. S., Brockwell, S. E., Mehta, V., Greendale, G. A., Sowers, M. R., Ettinger, B., Neer, R. M. (2008). Bone mineral density changes during the menopause transition in a multiethnic cohort of women. *The Journal of Clinical Endocrinology and Metabolism*, *93*, 861–8.

Fleet, J. C., & Schoch, R. D. (2010). Molecular mechanisms for regulation of intestinal calcium absorption by vitamin D and other factors. *Critical Reviews in Clinical Laboratory Sciences*, *47*, 181–95.

Foureaux, R. de C., Messora, M. R., de Oliveira, L. F. F., Napimoga, M. H., Pereira, A. N. J., Ferreira, M. S., & Pereira, L. J. (2014). Effects of probiotic therapy on metabolic and inflammatory parameters of rats with ligature-induced periodontitis associated with restraint stress. *Journal of Periodontology*, *85*, 975–83.

Freere, R. H., & Weibel, E. R. (1967). Stereologic techniques in microscopy. *Journal of the Royal Microscopical Society*, *87*, 25–34.

Fu, Q., Jilka, R. L., Manolagas, S. C., & O'Brien, C. A. (2002). Parathyroid hormone stimulates receptor activator of NFkappa B ligand and inhibits osteoprotegerin expression via protein kinase A activation of cAMP-response element-binding protein. *The Journal of Biological Chemistry*, *277*, 48868–75.

Furrie, E. (2005). Probiotics and allergy. *The Proceedings of the Nutrition Society*, *64*, 465–9.

Furusyo, N., Ihara, T., Hayashi, T., Ikezaki, H., Toyoda, K., Ogawa, E., Hayashi, J. (2013). The serum undercarboxylated osteocalcin level and the diet of a Japanese population: results from the Kyushu and Okinawa Population Study (KOPS). *Endocrine*, *43*, 635–42.

Gaffney-Stomberg, E., Sun, B., Cucchi, C. E., Simpson, C. A., Gundberg, C., Kerstetter, J. E., & Insogna, K. L. (2010). The effect of dietary protein on intestinal calcium absorption in rats. *Endocrinology*, *151*, 1071–8.

Gallagher, J. C. (2007). Effect of early menopause on bone mineral density and fractures. *Menopause*, *14*, 567–71.

Gao, X. W., Mubasher, M., Fang, C. Y., Reifer, C., & Miller, L. E. (2010). Dose-response efficacy of a proprietary probiotic formula of Lactobacillus acidophilus CL1285 and Lactobacillus casei LBC80R for antibiotic-associated diarrhea and Clostridium difficile-associated diarrhea prophylaxis in adult patients. *The American Journal of Gastroenterology*, *105*, 1636–41.

Garnero, P., Sornay-Rendu, E., Claustrat, B., & Delmas, P. D. (2000). Biochemical markers of bone turnover, endogenous hormones and the risk of fractures in postmenopausal women: the OFELY study. *Journal of Bone and Mineral Research*, *15*, 1526–36.

Garrett, I. R., Chen, D., Gutierrez, G., Zhao, M., Escobedo, A., Rossini, G., … Mundy, G. R. (2003). Selective inhibitors of the osteoblast proteasome stimulate bone formation in vivo and in vitro. *The Journal of Clinical Investigation*, *111*, 1771–82.

Genant, H. K., Lucas, J., Weiss, S., Akin, M., Emkey, R., McNaney-Flint, H., … Nolan, J. C. (1997). Low-dose esterified estrogen therapy: effects on bone, plasma estradiol concentrations, endometrium, and lipid levels. Estratab/Osteoporosis Study Group. *Archives of Internal Medicine*, *157*, 2609–15.

Ghanem, K. Z., Badawy, I. H., & Abdel-Salam, A. M. (2004). Influence of yoghurt and probiotic yoghurt on the absorption of calcium, magnesium, iron and bone mineralization in rats. *Milchwissenschaft*, *59*, 472–475.

Giannini, E. G., Testa, R., & Savarino, V. (2005). Liver enzyme alteration: a guide for clinicians. *Canadian Medical Association Journal*, *172*, 367–79.

Gilman, J., & Cashman, K. D. (2006). The effect of probiotic bacteria on transepithelial calcium transport and calcium uptake in human intestinal-like Caco-2 cells. *Current Issues in Intestinal Microbiology*, *7*, 1–5.

Glass, D. A., Bialek, P., Ahn, J. D., Starbuck, M., Patel, M. S., Clevers, H., Karsenty, G. (2005). Canonical Wnt signaling in differentiated osteoblasts controls osteoclast differentiation. *Developmental Cell*, *8*, 751–64.

Glimcher, M. J. (2006). Bone: Nature of the Calcium Phosphate Crystals and Cellular, Structural, and Physical Chemical Mechanisms in Their Formation. *Reviews in Mineralogy and Geochemistry*, *64*, 223–282.

Goda, T., Kishi, K., Ezawa, I., & Takase, S. (1998). The Maltitol-induced Increase in Intestinal Calcium Transport Increases the Calcium Content and Breaking Force of Femoral Bone in Weanling Rats. *Journal of Nutrition*, *128*, 2028–2031.

Gom, I., Fukushima, H., Shiraki, M., Miwa, Y., Ando, T., Takai, K., & Moriwaki, H. (2007). Relationship between serum albumin level and aging in community-dwelling self-supported elderly population. *Journal of Nutritional Science and Vitaminology*, *53*, 37–42.

Gomes da Cruz, A., Alonso Buriti, F. C., Batista de Souza, C. H., Fonseca Faria, J. A., & Isay Saad, S. M. (2009). Probiotic cheese: Health benefits, technological and stability aspects. *Trends in Food Science & Technology*, *20*, 344–354.

Gopal, P. K., Prasad, J., & Gill, H. S. (2003). Effects of the consumption of Bifidobacterium lactis HN019 (DR10TM) and galacto-oligosaccharides on the microflora of the gastrointestinal tract in human subjects. *Nutrition Research*, *23*, 1313–1328.

Gowen, M., Stroup, G. B., Dodds, R. A., James, I. E., Votta, B. J., Smith, B. R., Fox, J. (2000). Antagonizing the parathyroid calcium receptor stimulates parathyroid hormone secretion and bone formation in osteopenic rats. *The Journal of Clinical Investigation*, *105*, 1595–604.

Grajek, W., Olejnik, A. and Sip, A. (2005). Probiotics, prebiotics and antioxidants as functional foods. *Acta Biochim*, *52*, 665–671.

Greene, J. D., & Klaenhammer, T. R. (1994). Factors involved in adherence of lactobacilli to human Caco-2 cells. *Applied and Environmental Microbiology*, *60*, 4487–94.

Greer, F. R., & Krebs, N. F. (2006). Optimizing bone health and calcium intakes of infants, children, and adolescents. *Pediatrics*, *117*, 578–85.

Gregory, M. H., Capito, N., Kuroki, K., Stoker, A. M., Cook, J. L., & Sherman, S. L. (2012). A review of translational animal models for knee osteoarthritis. *Arthritis*, *2012*, 764621.

Gropper, & SareenSmith, J. (2013). *Advanced Nutrition and Human Metabolism, 6th Edition - Sareen S. Gropper | Jack L. Smith - Cengage Learning - 978-1133104056. college bookstore*.

Guaraldi, F., & Salvatori, G. (2012). Effect of breast and formula feeding on gut microbiota shaping in newborns. *Frontiers in Cellular and Infection Microbiology*, *2*, 94.

Guglielmi, G., Muscarella, S., & Bazzocchi, A. (2011). Integrated imaging approach to osteoporosis: state-of-the-art review and update. *Radiographics*, *31*, 1343–64.

Guo, Y., Ren, L., Liu, C., Yuan, Y., Lin, X., Tan, L., Mei, X. (2013). Effect of implantation of biodegradable magnesium alloy on BMP-2 expression in bone of ovariectomized osteoporosis rats. *Materials Science & Engineering. C, Materials for Biological Applications*, *33*, 4470–4.

Hadjidakis, D. J., & Androulakis, I. I. (2006). Bone remodeling. *Annals of the New York Academy of Sciences*, *1092*, 385–96.

Hagiwara, H., Hiruma, Y., Inoue, A., Yamaguchi, A., & Hirose, S. (1998). Deceleration by angiotensin II of the differentiation and bone formation of rat calvarial osteoblastic cells. *The Journal of Endocrinology*, *156*, 543–50.

Hamilton-Miller, J. M. T. (2004). Probiotics and prebiotics in the elderly. *Postgraduate Medical Journal*, *80*, 447–51.

Hancock, R. D., & Viola, R. (2001). The use of micro-organisms for L-ascorbic acid production: current status and future perspectives. *Applied Microbiology and Biotechnology*, *56*, 567–76.

Hankenson, K. D., James, I. E., Apone, S., Stroup, G. B., Blake, S. M., Liang, X., Bornstein, P. (2005). Increased osteoblastogenesis and decreased bone resorption protect against ovariectomy-induced bone loss in thrombospondin-2-null mice. *Matrix Biology*,*24*, 362–70.

Hao, Q., Lu, Z., Dong, B. R., Huang, C. Q., & Wu, T. (2011). Probiotics for preventing acute upper respiratory tract infections. *The Cochrane Database of Systematic Reviews*, *3*, CD006895.

Harada, S., & Rodan, G. A. (2003). Control of osteoblast function and regulation of bone mass. *Nature*, *423*, 349–55.

Harrison, E., Adjei, A., Ameho, C., Yamamoto, S., & Kono, S. (1998). The effect of soybean protein on bone loss in a rat model of postmenopausal osteoporosis. *Journal of Nutritional Science and Vitaminology*, *44*, 257–68.

Harvie, M., Howell, A., Vierkant, R. A., Kumar, N., Cerhan, J. R., Kelemen, L. E., Sellers, T. A. (2005). Association of gain and loss of weight before and after menopause with risk of postmenopausal breast cancer in the Iowa women's health study. *Cancer Epidemiology, Biomarkers & Prevention, 14*, 656–61.

Hassan, H. A., El Wakf, A. M., & El Gharib, N. E. (2013). Role of phytoestrogenic oils in alleviating osteoporosis associated with ovariectomy in rats. *Cytotechnology, 65*, 609–19.

Heaney, R. P., Abrams, S., Dawson-Hughes, B., Looker, A., Marcus, R., Matkovic, V., & Weaver, C. (2001). Peak Bone Mass. *Osteoporosis International*, *11*, 985–1009.

Hedlund, L. R., & Gallagher, J. C. (1989). The effect of age and menopause on bone mineral density of the proximal femur. *Journal of Bone and Mineral Research, 4*, 639–42.

Hendrix, S. L., Wassertheil-Smoller, S., Johnson, K. C., Howard, B. V, Kooperberg, C., Rossouw, J. E., Torner, J. (2006). Effects of conjugated equine estrogen on stroke in the Women's Health Initiative. *Circulation, 113*, 2425–34.

Hennequin, C., Kauffmann-Lacroix, C., Jobert, A., Viard, J. P., Ricour, C., Jacquemin, J. L., & Berche, P. (2000). Possible role of catheters in Saccharomyces boulardii fungemia. *European Journal of Clinical Microbiology & Infectious Diseases, 19*, 16–20.

Heshmati, H. M., Khosla, S., Robins, S. P., O'Fallon, W. M., Melton, L. J., & Riggs, B. L. (2002). Role of low levels of endogenous estrogen in regulation of bone resorption in late postmenopausal women. *Journal of Bone and Mineral Research, 17*, 172–8.

Hinoi, E., Fujimori, S., Wang, L., Hojo, H., Uno, K., & Yoneda, Y. (2006). Nrf2 negatively regulates osteoblast differentiation via interfering with Runx2-dependent transcriptional activation. *The Journal of Biological Chemistry, 281*, 18015–24.

Horiuchi, K., Amizuka, N., Takeshita, S., Takamatsu, H., Katsuura, M., Ozawa, H., Kudo, A. (1999). Identification and characterization of a novel protein, periostin, with restricted expression to periosteum and periodontal ligament and increased expression by transforming growth factor beta. *Journal of Bone and Mineral Research, 14*, 1239–49.

Hou, N., Hong, S., Wang, W., Olopade, O. I., Dignam, J. J., & Huo, D. (2013). Hormone replacement therapy and breast cancer: heterogeneous risks by race, weight, and breast density. *Journal of the National Cancer Institute, 105*, 1365–72.

Houillier, P., Froissart, M., Maruani, G., & Blanchard, A. (2006). What serum calcium can tell us and what it can't. *Nephrology, Dialysis, 21*, 29–32.

Hoveyda, N., Heneghan, C., Mahtani, K. R., Perera, R., Roberts, N., & Glasziou, P. (2009). A systematic review and meta-analysis: probiotics in the treatment of irritable bowel syndrome. *BMC Gastroenterology*, *9*, 15.

Hruska, K. (2000). Pathophysiology of renal osteodystrophy. *Pediatric Nephrology*, *14*, 636–640.

Hsu, J.-T., Chen, Y.-J., Ho, J.-T., Huang, H.-L., Wang, S.-P., Cheng, F.-C., Tsai, M.-T. (2014). A comparison of micro-CT and dental CT in assessing cortical bone morphology and trabecular bone microarchitecture. *PloS One*, *9*, e107545.

Hsu, J.-T., Chen, Y.-J., Tsai, M.-T., Lan, H. H.-C., Cheng, F.-C., Chen, M. Y. C., & Wang, S.-P. (2012). Predicting cortical bone strength from DXA and dental cone-beam CT. *PloS One*, *7*, e50008.

Hun, L. (2009). Bacillus coagulans significantly improved abdominal pain and bloating in patients with IBS. *Postgraduate Medicine*, *121*, 119–24.

Huynh, M. H., Sage, E. H., & Ringuette, M. (1999). A calcium-binding motif in SPARC/osteonectin inhibits chordomesoderm cell migration during Xenopus laevis gastrulation: evidence of counter-adhesive activity in vivo. *Development, Growth & Differentiation*, *41*, 407–18.

Iki, M., Morita, A., Ikeda, Y., Sato, Y., Akiba, T., Matsumoto, T., Yoneshima, H. (2006). Biochemical markers of bone turnover predict bone loss in perimenopausal women but not in postmenopausal women-the Japanese Population-based Osteoporosis (JPOS) Cohort Study. *Osteoporosis International*, *17*, 1086–95.

Ilich, J. Z., Brownbill, R. A., & Tamborini, L. (2003). Bone and nutrition in elderly women: protein, energy, and calcium as main determinants of bone mineral density. *European Journal of Clinical Nutrition*, *57*, 554–65.

Imam, M. U., & Ismail, M. (2012). Effects of brown rice and white rice on expression of xenobiotic metabolism genes in type 2 diabetic rats. *International Journal of Molecular Sciences*, *13*, 8597–608.

Ioannidis, J. P. A., Ralston, S. H., Bennett, S. T., Brandi, M. L., Grinberg, D., Karassa, F. B., Uitterlinden, A. G. (2004). Differential genetic effects of ESR1 gene polymorphisms on osteoporosis outcomes. *JAMA*, *292*, 2105–14.

Ishibashi, N., & Yamazaki, S. (2001). Probiotics and safety. *American Journal of Clinical Nutrition*, *73*, 465S–470.

Ivaska, K. K., Hentunen, T. A., Vääräniemi, J., Ylipahkala, H., Pettersson, K., & Väänänen, H. K. (2004). Release of intact and fragmented osteocalcin molecules from bone matrix during bone resorption in vitro. *The Journal of Biological Chemistry*, *279*, 18361–9.

Iwamoto, J., Takeda, T., & Ichimura, S. (2002). Increased bone resorption with decreased activity and increased recruitment of osteoblasts in osteogenesis imperfecta type I. *Journal of Bone and Mineral Metabolism*, *20*, 174–9.

Iwaniec, U. T., Samnegård, E., Cullen, D. M., & Kimmel, D. B. (2001). Maintenance of cancellous bone in ovariectomized, human parathyroid hormone [hPTH(1-84)]-treated rats by estrogen, risedronate, or reduced hPTH. *Bone*, *29*, 352–60.

Iwaniec, U. T., Yuan, D., Power, R. A., & Wronski, T. J. (2006). Strain-dependent variations in the response of cancellous bone to ovariectomy in mice. *Journal of Bone and Mineral Research*, *21*, 1068–74.

Jacobs, P. A., Hyland, M. E., & Ley, A. (2000). Self-rated menopausal status and quality of life in women aged 40-63 years. *British Journal of Health Psychology*, *5*, 395–411.

Ji, M.-X., & Yu, Q. (2015). Primary osteoporosis in postmenopausal women. *Chronic Diseases and Translational Medicine*, *1*, 9–13.

Jirillo, E., Jirillo, F., & Magrone, T. (2012). Healthy effects exerted by prebiotics, probiotics, and symbiotics with special reference to their impact on the immune system. *International Journal for Vitamin and Nutrition Research*, *82*, 200–8.

Joseph, F., Ahmad, A. M., Ul-Haq, M., Durham, B. H., Whittingham, P., Fraser, W. D., & Vora, J. P. (2008). Effects of growth hormone administration on bone mineral metabolism, PTH sensitivity and PTH secretory rhythm in postmenopausal women with established osteoporosis. *Journal of Bone and Mineral Research*, *23*, 721–9.

Juntunen, M., Kirjavainen, P. V, Ouwehand, A. C., Salminen, S. J., & Isolauri, E. (2001). Adherence of probiotic bacteria to human intestinal mucus in healthy infants and during rotavirus infection. *Clinical and Diagnostic Laboratory Immunology*, *8*, 293–6.

Kaastad, T. S., Reikerås, O., Halvorsen, V., Falch, J. A., Obrant, K. J., & Nordsletten, L. (2001). Vitamin D deficiency and ovariectomy reduced the strength of the femoral neck in rats. *Calcified Tissue International*, *69*, 102–8.

Kaastad, T. S., Reikerås, O., Narum, S., Madsen, J. E., Haug, E., Obrant, K. J., & Nordsletten, L. (1997). Effect of intensive training on lower leg structural strength: an in vivo study in ovariectomized rats. *Scandinavian Journal of Medicine & Science in Sports*, *7*, 220–5.

Kabeir, B. M., Yazid, A. M., Stephenie, W., Hakim, M. N., Anas, O. M., & Shuhaimi, M. (2008). Safety evaluation of Bifidobacterium pseudocatenulatum G4 as assessed in BALB/c mice. *Letters in Applied Microbiology*, *46*, 32–7.

Kaburagi, T., Yamano, T., Fukushima, Y., Yoshino, H., Mito, N., & Sato, K. (2007). Effect of Lactobacillus johnsonii La1 on immune function and serum albumin in aged and malnourished aged mice. *Nutrition*, *23*, 342–50.

Kadooka, Y., Sato, M., Imaizumi, K., Ogawa, A., Ikuyama, K., Akai, Y., Tsuchida, T. (2010). Regulation of abdominal adiposity by probiotics (Lactobacillus gasseri SBT2055) in adults with obese tendencies in a randomized controlled trial. *European Journal of Clinical Nutrition*, *64*, 636–43.

Kai, M. C., Anderson, M., & Lau, E. M. C. (2003). Exercise interventions: defusing the world's osteoporosis time bomb. *Bulletin of the World Health Organization*, *81*, 827–30.

Kailasapathy, K., & Chin, J. (2000). Survival and therapeutic potential of probiotic organisms with reference to Lactobacillus acidophilus and Bifidobacterium spp. *Immunology and Cell Biology*, *78*, 80–8.

Kalu, D. N., Liu, C. C., Hardin, R. R., & Hollis, B. W. (1989). The aged rat model of ovarian hormone deficiency bone loss. *Endocrinology*, *124*, 7–16.

Kanis, J. A. (2002). Diagnosis of osteoporosis and assessment of fracture risk. *Lancet*, *359*, 1929–36.

Kanis, J. A., Oden, A., Johnell, O., Johansson, H., De Laet, C., Brown, J., Yoshimura, N. (2007). The use of clinical risk factors enhances the performance of BMD in the prediction of hip and osteoporotic fractures in men and women. *Osteoporosis International*, *18*, 1033–46.

Kantner, I., & Erben, R. G. (2012). Long-term parenteral administration of 2-hydroxypropyl-β-cyclodextrin causes bone loss. *Toxicologic Pathology*, *40*, 742–50.

Kanzler, B., Foreman, R. K., Labosky, P. A., & Mallo, M. (2000). BMP signaling is essential for development of skeletogenic and neurogenic cranial neural crest. *Development*, *127*, 1095–104.

Kelsey, J. L., Prill, M. M., Keegan, T. H. M., Quesenberry, C. P., & Sidney, S. (2005). Risk factors for pelvis fracture in older persons. *American Journal of Epidemiology*, *162*, 879–86.

Kharode, Y. P., Sharp, M. C., & Bodine, P. V. N. (2008). Utility of the ovariectomized rat as a model for human osteoporosis in drug discovery. *Methods in Molecular Biology*, *455*, 111–24.

Khassawna, T., Böcker, W., Govindarajan, P., Schliefke, N., Hürter, B., Kampschulte, M., Heiss, C. (2013). Effects of multi-deficiencies-diet on bone parameters of peripheral bone in ovariectomized mature rat. *PloS One*, *8*, e71665.

Khosla, S., Atkinson, E. J., Melton, L. J., & Riggs, B. L. (1997). Effects of age and estrogen status on serum parathyroid hormone levels and biochemical markers of bone turnover in women: a population-based study. *The Journal of Clinical Endocrinology and Metabolism*, *82*, 1522–7.

Kifor, O., McElduff, A., LeBoff, M. S., Moore, F. D., Butters, R., Gao, P., Brown, E. M. (2004). Activating antibodies to the calcium-sensing receptor in two patients with autoimmune hypoparathyroidism. *The Journal of Clinical Endocrinology and Metabolism*, *89*, 548–56.

Kim, J. G., Ku, S. Y., Jee, B. C., Suh, C. S., Kim, S. H., Choi, Y. M., & Moon, S. Y. (2004). The relationship between calcium sensing receptor gene (CA) polymorphism, bone mineral density and bone responsiveness to hormone replacement therapy in postmenopausal Korean women. *Korean Journal of Obstetrics and Gynecology*, *47*, 2430–2438.

Kim, J. G., Lee, E., Kim, S. H., Whang, K. Y., Oh, S., & Imm, J.-Y. (2009). Effects of a Lactobacillus casei 393 fermented milk product on bone metabolism in ovariectomised rats. *International Dairy Journal*, *19*, 690–695.

King, J. C. (2002). Evaluating the impact of plant biofortification on human nutrition. *The Journal of Nutrition*, *132*, 511S–513S.

Kitada, H., Miyata, M., Nakamura, T., Tozawa, A., Honma, W., Shimada, M., Yamazoe, Y. (2003). Protective role of hydroxysteroid sulfotransferase in lithocholic acid-induced liver toxicity. *The Journal of Biological Chemistry*, *278*, 17838–44.

Klappenbach, J. A., Saxman, P. R., Cole, J. R., & Schmidt, T. M. (2001). rrndb: the Ribosomal RNA Operon Copy Number Database. *Nucleic Acids Research*, *29*, 181–4.

Komori, T. (2010). Regulation of bone development and extracellular matrix protein genes by RUNX2. *Cell and Tissue Research*, *339*, 189–95.

Kovacs, C. S. (2001). Calcium and bone metabolism in pregnancy and lactation. *The Journal of Clinical Endocrinology and Metabolism*, *86*, 2344–8.

Krishnamoorthy, D., Frechette, D. M., Adler, B. J., Green, D. E., Chan, M. E., & Rubin, C. T. (2015). Marrow adipogenesis and bone loss that parallels estrogen deficiency is slowed by low-intensity mechanical signals. *Osteoporosis International*, doi:10.1007/s00198-015-3289-5

Krivosíková, Z., Krajcovicová-Kudlácková, M., Spustová, V., Stefíková, K., Valachovicová, M., Blazícek, P., & Němcová, T. (2010). The association between high plasma homocysteine levels and lower bone mineral density in Slovak women: the impact of vegetarian diet. *European Journal of Nutrition*, *49*, 147–53.

Kroll, M. H. (2000). Parathyroid hormone temporal effects on bone formation and resorption. *Bulletin of Mathematical Biology*, *62*, 163–88.

Kruger, M. C., Fear, A., Chua, W.-H., Plimmer, G. G., & Schollum, L. M. (2009). The effect of Lactobacillus rhamnosus HN001 on mineral absorption and bone health in growing male and ovariectomised female rats. *Dairy Science and Technology*, *89*, 219–231.

Kumar, R., & Thompson, J. R. (2011). The regulation of parathyroid hormone secretion and synthesis. *Journal of the American Society of Nephrology*, *22*, 216–24.

Kumar, Sachin; Verma, A. K.; Mondal, S. K.; Gupta, Mahesh; Patil, A. K.; Lal Jangir, B. (2012). Effect of live Saccharomyces cerevisiae feeding on serum biochemistry in early weaned cross bred piglets. *Vet. World*,, *5*, 663–666.

Kunz, A. N., Noel, J. M., & Fairchok, M. P. (2004). Two cases of Lactobacillus bacteremia during probiotic treatment of short gut syndrome. *Journal of Pediatric Gastroenterology and Nutrition*, *38*, 457–8.

Kuroda, S., Mukohyama, H., Kondo, H., Aoki, K., Ohya, K., Ohyama, T., & Kasugai, S. (2003). Bone mineral density of the mandible in ovariectomized rats: analyses using dual energy X-ray absorptiometry and peripheral quantitative computed tomography. *Oral Diseases*, *9*, 24–8.

Kuwata, F., Yao, K. L., Sodek, J., Ives, S., & Pulleyblank, D. (1985). Identification of pre-osteonectin produced by cell-free translation of fetal porcine calvarial mRNA. *The Journal of Biological Chemistry*, *260*, 6993–8.

La Rosa, M., Bottaro, G., Gulino, N., Gambuzza, F., Di Forti, F., Inì, G., & Tornambè, E. (2003). Prevention of antibiotic-associated diarrhea with Lactobacillus sporogens and fructo-oligosaccharides in children. A multicentric double-blind vs placebo study. *Minerva Pediatrica*, *55*, 447–52.

Lamberg-Allardt, C. J., Outila, T. A., Kärkkainen, M. U., Rita, H. J., & Valsta, L. M. (2001). Vitamin D deficiency and bone health in healthy adults in Finland: could this be a concern in other parts of Europe? *Journal of Bone and Mineral Research,16*, 2066–73.

Lan, G. Q., Abdullah, N., Jalaludin, S., & Ho, Y. W. (2002). Efficacy of supplementation of a phytase-producing bacterial culture on the performance and nutrient use of broiler chickens fed corn-soybean meal diets. *Poultry Science*, *81*, 1522–32.

Laschke, M. W., Witt, K., Pohlemann, T., & Menger, M. D. (2007). Injectable nanocrystalline hydroxyapatite paste for bone substitution: in vivo analysis of biocompatibility and vascularization. *Journal of Biomedical Materials Research*, *82*, 494–505.

Lau, E. M., Lee, J. K., Suriwongpaisal, P., Saw, S. M., Das De, S., Khir, A., & Sambrook, P. (2001). The incidence of hip fracture in four Asian countries: the Asian Osteoporosis Study (AOS). *Osteoporosis International*, *12*, 239–43.

Lau, K., Benitez, P., Ardissone, A., Wilson, T. D., Collins, E. L., Lorca, G., Larkin, J. (2011). Inhibition of type 1 diabetes correlated to a Lactobacillus johnsonii N6.2-mediated Th17 bias. *Journal of Immunology*, *186*, 3538–46.

Leali, P. T., Muresu, F., Melis, A., Ruggiu, A., Zachos, A., & Doria, C. (2011). Skeletal fragility definition. *Clinical Cases in Mineral and Bone Metabolism*, *8*, 11–3.

Ledoux, D., Labombardi, V. J., & Karter, D. (2006). Lactobacillus acidophilus bacteraemia after use of a probiotic in a patient with AIDS and Hodgkin's disease. *International Journal of STD & AIDS*, *17*, 280–2.

Lee, H.-J., Kim, S.-Y., Kim, G. S., Hwang, J.-Y., Kim, Y.-J., Jeong, B., Lee, J.-Y. (2010). Fracture, bone mineral density, and the effects of calcitonin receptor gene in postmenopausal Koreans. *Osteoporosis International*, *21*, 1351–60.

Legrand, E., Chappard, D., Pascaretti, C., Duquenne, M., Krebs, S., Rohmer, V., Audran, M. (2000). Trabecular bone microarchitecture, bone mineral density, and vertebral fractures in male osteoporosis. *Journal of Bone and Mineral Research*, *15*, 13–9.

Lei, Z., Xiaoying, Z., & Xingguo, L. (2009). Ovariectomy-associated changes in bone mineral density and bone marrow haematopoiesis in rats. *International Journal of Experimental Pathology*, *90*, 512–9.

Levin, R. J. (2002). The physiology of sexual arousal in the human female: a recreational and procreational synthesis. *Archives of Sexual Behavior*, *31*, 405–11.

Li, G.-W., Xu, Z., Chang, S.-X., Nian, H., Wang, X.-Y., & Qin, L.-D. (2014). Icariin prevents ovariectomy-induced bone loss and lowers marrow adipogenesis. *Menopause*, *21*, 1007–16.

Liang, H., Yu, F., Tong, Z., & Zeng, W. (2012). Lycopene effects on serum mineral elements and bone strength in rats. *Molecules*, *17*, 7093–102.

Lick, S., Drescher, K., & Heller, K. J. (2001). Survival of Lactobacillus delbrueckii subsp. bulgaricus and Streptococcus thermophilus in the terminal ileum of fistulated Göttingen minipigs. *Applied and Environmental Microbiology*, *67*, 4137–43.

Lim, D. W., Kim, J. G., & Kim, Y. T. (2013). Effects of dietary isoflavones from Puerariae radix on lipid and bone metabolism in ovariectomized rats. *Nutrients*, *5*, 2734–46.

Litvin, J., Selim, A.-H., Montgomery, M. O., Lehmann, K., Rico, M. C., Devlin, H., Safadi, F. F. (2004). Expression and function of periostin-isoforms in bone. *Journal of Cellular Biochemistry*, *92*, 1044–61.

Liu, M.-L., Xu, X., Rang, W.-Q., Li, Y.-J., & Song, H.-P. (2004). Influence of ovariectomy and 17beta-estradiol treatment on insulin sensitivity, lipid metabolism and post-ischemic cardiac function. *International Journal of Cardiology*, *97*, 485–93.

Liu, S. F., & Malik, A. B. (2006). NF-kappa B activation as a pathological mechanism of septic shock and inflammation. *American Journal of Physiology*, *290*, L622–L645.

Liu, W., Toyosawa, S., Furuichi, T., Kanatani, N., Yoshida, C., Liu, Y., Komori, T. (2001). Overexpression of Cbfa1 in osteoblasts inhibits osteoblast maturation and causes osteopenia with multiple fractures. *The Journal of Cell Biology*, *155*, 157–66.

Loh, K. Y., & Shong, H. K. (2007). Osteoporosis: primary prevention in the community. *The Medical Journal of Malaysia*, *62*, 355–7.

Lopez, H. W., Coudray, C., Levrat-Verny, M. A., Feillet-Coudray, C., Demigné, C., & Rémésy, C. (2000). Fructooligosaccharides enhance mineral apparent absorption and counteract the deleterious effects of phytic acid on mineral homeostasis in rats. *The Journal of Nutritional Biochemistry*, *11*, 500–8.

Lu, Y., Chai, W., & Lin, X. (2007). Effect of milk basic protein on rat bone mineral density. *Journal of Hygiene Research*, *36*, 37–40.

Macdonald, H. M., New, S. A., Campbell, M. K., & Reid, D. M. (2003). Longitudinal changes in weight in perimenopausal and early postmenopausal women: effects of dietary energy intake, energy expenditure, dietary calcium intake and hormone replacement therapy. *International Journal of Obesity and Related Metabolic Disorders*, *27*, 669–76.

Macdonald, H. M., New, S. A., Golden, M. H., Campbell, M. K., & Reid, D. M. (2004). Nutritional associations with bone loss during the menopausal transition: evidence of a beneficial effect of calcium, alcohol, and fruit and vegetable nutrients and of a detrimental effect of fatty acids. *American Journal of Clinical Nutrition*, *79*, 155–165.

Macfarlane, S., & Macfarlane, G. T. (2006). Composition and metabolic activities of bacterial biofilms colonizing food residues in the human gut. *Applied and Environmental Microbiology*, *72*, 6204–11.

Mach, T. (2006). Clinical usefulness of probiotics in inflammatory bowel diseases. *Journal of Physiology and Pharmacology*, *9*, 23–33.

MacKay, D., & Miller, A. L. (2003). Nutritional support for wound healing. *Alternative Medicine Review : A Journal of Clinical Therapeutic*, *8*, 359–77.

Macpherson, C., Audy, J., Mathieu, O., & Tompkins, T. A. (2014). Multistrain probiotic modulation of intestinal epithelial cells' immune response to a double-stranded RNA ligand, poly. *Applied and Environmental Microbiology*, *80*, 1692–700.

Mahendru, A. A., & Morris, E. (2013). Cardiovascular disease in menopause: does the obstetric history have any bearing? *Menopause International*, *19*, 115–20.

Malik, V. S., Schulze, M. B., & Hu, F. B. (2006). Intake of sugar-sweetened beverages and weight gain: a systematic review. *The American Journal of Clinical Nutrition*, *84*, 274–88.

Mann, V., Hobson, E. E., Li, B., Stewart, T. L., Grant, S. F., Robins, S. P., Ralston, S. H. (2001). A COL1A1 Sp1 binding site polymorphism predisposes to osteoporotic fracture by affecting bone density and quality. *The Journal of Clinical Investigation*, *107*, 899–907.

Manolagas, S. C. (2000). Birth and death of bone cells: basic regulatory mechanisms and implications for the pathogenesis and treatment of osteoporosis. *Endocrine Reviews*, *21*, 115–37.

Maradonna, F., Gioacchini, G., Falcinelli, S., Bertotto, D., Radaelli, G., Olivotto, I., & Carnevali, O. (2013). Probiotic supplementation promotes calcification in Danio rerio larvae: a molecular study. *PloS One*, *8*, e83155.

Marino, M., Galluzzo, P., & Ascenzi, P. (2006). Estrogen signaling multiple pathways to impact gene transcription. *Current Genomics*, *7*, 497–508.

Marsden, J. (2002). Hormone-replacement therapy and breast cancer. *The Lancet Oncology*, *3*, 303–311.

Martin, T., Gooi, J. H., & Sims, N. A. (2009). Molecular mechanisms in coupling of bone formation to resorption. *Critical Reviews in Eukaryotic Gene Expression*, *19*, 73–88.

Martin, T. J., & Seeman, E. (2008). Bone remodelling: its local regulation and the emergence of bone fragility. *Best Practice & Research. Clinical Endocrinology & Metabolism*, *22*, 701–22.

Matsumoto, H. N., Yamamoto, A., Iimura, T., Oida, S., Ezawa, I., Sasaki, S., & Goseki-Sone, M. (1997). Effects of ovariectomy on intestinal alkaline phosphatase expression in rats. *Journal of Nutritional Science and Vitaminology*, *43*, 529–39.

McCabe, L. R., Irwin, R., Schaefer, L., & Britton, R. a. (2013a). Probiotic use decreases intestinal inflammation and increases bone density in healthy male but not female mice. *Journal of Cellular Physiology*, *228*, 1793–8.

McCabe, L. R., Irwin, R., Schaefer, L., & Britton, R. A. (2013b). Probiotic use decreases intestinal inflammation and increases bone density in healthy male but not female mice. *Journal of Cellular Physiology*, *228*, 1793–8.

McFarland, J. (1970). The Nephelometer:An Instrument for Estimating the Number of Bacteria in Suspensions Used for Calculating the Opsonic Index and for Vaccines. *JAMA*, *14*, 1176–78.

McManus, J., & Mowry, R. (1960). *Staining Methods: Histolgic and Histochemical*. doi:New York (NY): Paul B. Hoeber Inc.

Medici, M., van Meurs, J. B., Rivadeneira, F., Zhao, H., Arp, P. P., Hofman, A., Uitterlinden, A. G. (2006). BMP-2 gene polymorphisms and osteoporosis: the Rotterdam Study. *Journal of Bone and Mineral Research*, *21*, 845–54.

Mehrotra, M., Gupta, S. K., Tiwari, S., Agarwal, A., Kumar, K., Awasthi, P. K., & Godbole, M. M. (2003). Effect of oophorectomy on expression of calcium sensing receptor mRNA in rat duodenal mucosa. *Indian Journal of Experimental Biology*, *41*, 41–6.

Ménard, O., Butel, M.-J., Gaboriau-Routhiau, V., & Waligora-Dupriet, A.-J. (2008). Gnotobiotic mouse immune response induced by Bifidobacterium sp. strains isolated from infants. *Applied and Environmental Microbiology*, *74*, 660–6.

Messaoudi, M., Violle, N., Bisson, J.-F., Desor, D., Javelot, H., & Rougeot, C. (2011). Beneficial psychological effects of a probiotic formulation (Lactobacillus helveticus R0052 and Bifidobacterium longum R0175) in healthy human volunteers. *Gut Microbes*, *2*, 256–61.

Metcalfe, D. (2008). The pathophysiology of osteoporotic hip fracture. *McGill Journal of Medicine*, *11*, 51–7.

Minellia Elisa Bertazzoni,, Beninia Anna, Marzottob Marta, Andrea Sbarbatic, Orazio Ruzzenented, Rossano Ferrarioe, Henno Hendriksf, F. D. (2004). Assessment of novel probiotic Lactobacillus casei strains for the production of functional dairy foods. *International Dairy Journal*, *14*, 723–736.

Miquel, S., Beaumont, M., Martín, R., Langella, P., Braesco, V., & Thomas, M. (2015). A proposed framework for an appropriate evaluation scheme for microorganisms as novel foods with a health claim in Europe. *Microbial Cell Factories*, *14*, 48.

Moe, S. M. (2008). Disorders involving calcium, phosphorus, and magnesium. *Primary Care*, *35*, 215–37, v–vi.

Muhammad, S. I., Ismail, M., Mahmud, R. B., Salisu, A. M., & Zakaria, Z. A. (2013). Germinated brown rice and its bioactives modulate the activity of uterine cells in oophorectomised rats as evidenced by gross cytohistological

and immunohistochemical changes. *BMC Complementary and Alternative Medicine*, *13*, 198.

Muhammad, S. I., Maznah, I., Mahmud, R., Zuki, A. B. Z., & Imam, M. U. (2013). Upregulation of genes related to bone formation by γ-amino butyric acid and γ-oryzanol in germinated brown rice is via the activation of GABAB-receptors and reduction of serum IL-6 in rats. *Clinical Interventions in Aging*, *8*, 1259–71.

Munaisinghe, R. L., Botea, V., & Edelson, G. W. (2002). Association among age, height, weight, and body mass index with discordant regional bone mineral density. *Journal of Clinical Densitometry*, *5*, 369–73.

Murugan, R., & Ramakrishna, S. (2005). Development of nanocomposites for bone grafting. *Composites Science and Technology*, *65*, 2385–2406.

Mutuş, R., Kocabagli, N., Alp, M., Acar, N., Eren, M., & Gezen, S. S. (2006). The effect of dietary probiotic supplementation on tibial bone characteristics and strength in broilers. *Poultry Science*, *85*, 1621–5.

Nabeshima, Y., & Imura, H. (2008). alpha-Klotho: a regulator that integrates calcium homeostasis. *American Journal of Nephrology*, *28*, 455–64.

Naito, E., Yoshida, Y., Makino, K., Kounoshi, Y., Kunihiro, S., Takahashi, R., Ishikawa, F. (2011). Beneficial effect of oral administration of Lactobacillus casei strain Shirota on insulin resistance in diet-induced obesity mice. *Journal of Applied Microbiology*, *110*, 650–7.

Nakatani, T., Sarraj, B., Ohnishi, M., Densmore, M. J., Taguchi, T., Goetz, R., Razzaque, M. S. (2009). In vivo genetic evidence for klotho-dependent, fibroblast growth factor 23 (Fgf23) -mediated regulation of systemic phosphate homeostasis. *Federation of American Societies for Experimental Biology*, *23*, 433–41.

Narva, M., Collin, M., Lamberg-Allardt, C., Kärkkäinen, M., Poussa, T., Vapaatalo, H., & Korpela, R. (2004). Effects of long-term intervention with Lactobacillus helveticus-fermented milk on bone mineral density and bone mineral content in growing rats. *Annals of Nutrition & Metabolism*, *48*, 228–34.

Narva, M., Halleen, J., Väänänen, K., & Korpela, R. (2004). Effects of Lactobacillus helveticus fermented milk on bone cells in vitro. *Life Sciences*, *75*, 1727–34.

Narva, M., Nevala, R., Poussa, T., & Korpela, R. (2004). The effect of Lactobacillus helveticus fermented milk on acute changes in calcium metabolism in postmenopausal women. *European Journal of Nutrition*, *43*, 61–8.

Näse, L., Hatakka, K., Savilahti, E., Saxelin, M., Pönkä, A., Poussa, T., Meurman, J. H. (2001). Effect of long-term consumption of a probiotic bacterium, Lactobacillus rhamnosus GG, in milk on dental caries and caries risk in children. *Caries Research*, *35*, 412–20.

National research. (1995). *Nutrient Requirements of Laboratory Animals* (4th editio., pp. 26–28). Washington, DC: National Academic Press.

New, S. A., Robins, S. P., Campbell, M. K., Martin, J. C., Garton, M. J., Bolton-Smith, C., Reid, D. M. (2000). Dietary influences on bone mass and bone metabolism: further evidence of a positive link between fruit and vegetable consumption and bone health? *The American Journal of Clinical Nutrition*, *71*, 142–51.

Nguyen, T. V, Center, J. R., & Eisman, J. A. (2000). Osteoporosis in elderly men and women: effects of dietary calcium, physical activity, and body mass index. *Journal of Bone and Mineral Research*, *15*, 322–31.

Niel, C. W. (2005). Probiotics: not just for treatment anymore. *Pediatrics*, *115*, 174–7.

Nieves, J. W. (2005). Osteoporosis: the role of micronutrients. *American Journal of Clinical Nutrition*, *81*, 1232S–1239.

Nordin, B. E. C., Wishart, J. M., Clifton, P. M., McArthur, R., Scopacasa, F., Need, A. G., Horowitz, M. (2004). A longitudinal study of bone-related biochemical changes at the menopause. *Clinical Endocrinology*, *61*, 123–30.

Norek, A., Sands, D., Sobczyńska-Tomaszewska, A., Chmielewski, D., Szamotulska, K., Czerska, K., & Bal, J. (2010). Genetic risk markers of low bone mineral density in cystic fibrosis children. *Medycyna Wieku Rozwojowego*, *14*, 334–43.

Ohlsson, C., Engdahl, C., Fåk, F., Andersson, A., Windahl, S. H., Farman, H. H., Sjögren, K. (2014). Probiotics protect mice from ovariectomy-induced cortical bone loss. *PloS One*, *9*, e92368.

Oral, A., Unal, D., Halici, Z., Cadirci, E., Sengul, O., Ozaltin, S., Atamanalp, S. S. (2012). Bilateral Ovariectomy in Young Rats: What Happens in Their Livers during Cecal Ligation and Puncture Induced Sepsis? *Journal of Pediatric and Adolescent Gynecology*, *25*, 371–379.

Ouwehand, A. C., Kirjavainen, P. V., Shortt, C., & Salminen, S. (1999). Probiotics: mechanisms and established effects. *International Dairy Journal*, *9*, 43–52.

Ouwehand, A. C., Salminen, S., & Isolauri, E. (2002). Probiotics: an overview of beneficial effects. *Antonie van Leeuwenhoek*, *82*, 279–89.

Oz, O. K., Hajibeigi, A., Howard, K., Cummins, C. L., van Abel, M., Bindels, R. J., Zerwekh, J. E. (2007). Aromatase deficiency causes altered expression of

molecules critical for calcium reabsorption in the kidneys of female mice. *Journal of Bone and Mineral Research*, *22*, 1893–902.

Oztekin, E., Tiftik, A. M., Baltaci, A. K., & Mogulkoc, R. (2007). Lipid peroxidation in liver tissue of ovariectomized and pinealectomized rats: effect of estradiol and progesterone supplementation. *Cell Biochemistry and Function*, *25*, 401–5.

Paik, M.-K., Lee, H.-O., Chung, H.-S., Yang, S.-O., Kim, J.-H., & Om, A.-S. (2003). Genistein may prevent cadmium-induced bone loss in ovariectomized rats. *Journal of Medicinal Food*, *6*, 337–43.

Pannemans, D. L., Schaafsma, G., & Westerterp, K. R. (1997). Calcium excretion, apparent calcium absorption and calcium balance in young and elderly subjects: influence of protein intake. *The British Journal of Nutrition*, *77*, 721–9.

Papadimitropoulos, E., Wells, G., Shea, B., Gillespie, W., Weaver, B., Zytaruk, N., Guyatt, G. (2002). Meta-analyses of therapies for postmenopausal osteoporosis. VIII: Meta-analysis of the efficacy of vitamin D treatment in preventing osteoporosis in postmenopausal women. *Endocrine Reviews*, *23*, 560–9.

Pappa, H. M., Saslowsky, T. M., Filip-Dhima, R., DiFabio, D., Lahsinoui, H. H., Akkad, A., Gordon, C. M. (2011). Efficacy and harms of nasal calcitonin in improving bone density in young patients with inflammatory bowel disease: a randomized, placebo-controlled, double-blind trial. *The American Journal of Gastroenterology*, *106*, 1527–43.

Parfitt, A. M. (2002). Misconceptions (2): turnover is always higher in cancellous than in cortical bone. *Bone*, *30*, 807–9.

Parvaneh, K., Jamaluddin, R., Karimi, G., & Erfani, R. (2014). Effect of probiotics supplementation on bone mineral content and bone mass density. *TheScientificWorldJournal*, *2014*, 595962.

Peacock, M. (2010). Calcium metabolism in health and disease. *Clinical Journal of the American Society of Nephrology*, *5*, S23–30.

Pearce, A. I., Richards, R. G., Milz, S., Schneider, E., & Pearce, S. G. (2007). Animal models for implant biomaterial research in bone: a review. *European Cells & Materials*, *13*, 1–10.

Pelleymounter, M. A., Cullen, M. J., Healy, D., Hecht, R., Winters, D., & McCaleb, M. (1998). Efficacy of exogenous recombinant murine leptin in lean and obese 10- to 12-mo-old female CD-1 mice. *The American Journal of Physiology*, *275*, R950–9.

Perwad, F., Zhang, M. Y. H., Tenenhouse, H. S., & Portale, A. A. (2007). Fibroblast growth factor 23 impairs phosphorus and vitamin D metabolism in

vivo and suppresses 25-hydroxyvitamin D-1alpha-hydroxylase expression in vitro. *American Journal of Physiology. Renal Physiology*, *293*, F1577–83.

Picherit, C., Coxam, V., Bennetau-Pelissero, C., Kati-Coulibaly, S., Davicco, M. J., Lebecque, P., & Barlet, J. P. (2000). Daidzein is more efficient than genistein in preventing ovariectomy-induced bone loss in rats. *The Journal of Nutrition*, *130*, 1675–81.

Pioszak, A. A., Harikumar, K. G., Parker, N. R., Miller, L. J., & Xu, H. E. (2010). Dimeric arrangement of the parathyroid hormone receptor and a structural mechanism for ligand-induced dissociation. *The Journal of Biological Chemistry*, *285*, 12435–44.

Pioszak, A. A., & Xu, H. E. (2008). Molecular recognition of parathyroid hormone by its G protein-coupled receptor. *Proceedings of the National Academy of Sciences of the United States of America*, *105*, 5034–9.

Pluijm, S. M. F., van Essen, H. W., Bravenboer, N., Uitterlinden, A. G., Smit, J. H., Pols, H. A. P., & Lips, P. (2004). Collagen type I alpha1 Sp1 polymorphism, osteoporosis, and intervertebral disc degeneration in older men and women. *Annals of the Rheumatic Diseases*, *63*, 71–7.

Poole, K. E. S., & Reeve, J. (2005). Parathyroid hormone - a bone anabolic and catabolic agent. *Current Opinion in Pharmacology*, *5*, 612–7.

Prentice, A. (2001). The relative contribution of diet and genotype to bone development. *The Proceedings of the Nutrition Society*, *60*, 45–52.

Pulkkinen, P., Partanen, J., Jalovaara, P., & Jämsä, T. (2004). Combination of bone mineral density and upper femur geometry improves the prediction of hip fracture. *Osteoporosis International*, *15*, 274–80.

Raggatt, L. J., & Partridge, N. C. (2010). Cellular and molecular mechanisms of bone remodeling. *The Journal of Biological Chemistry*, *285*, 25103–8.

Raisz, L. G. (2005). Pathogenesis of osteoporosis: concepts, conflicts, and prospects. *The Journal of Clinical Investigation*, *115*, 3318–25.

Raschka, L., & Daniel, H. (2005). Mechanisms underlying the effects of inulin-type fructans on calcium absorption in the large intestine of rats. *Bone*, *37*, 728–35.

Rawadi, G., Vayssière, B., Dunn, F., Baron, R., & Roman-Roman, S. (2003). BMP-2 controls alkaline phosphatase expression and osteoblast mineralization by a Wnt autocrine loop. *Journal of Bone and Mineral Research*, *18*, 1842–53.

Reid, G., Gaudier, E., Guarner, F., Huffnagle, G. B., Macklaim, J. M., Munoz, A. M., Walter, J. (2010). Responders and non-responders to probiotic interventions: how can we improve the odds? *Gut Microbes*, *1*, 200–4.

Reid, G., Sanders, M. E., Gaskins, H. R., Gibson, G. R., Mercenier, A., Rastall, R., Klaenhammer, T. R. (2003). New scientific paradigms for probiotics and prebiotics. *Journal of Clinical Gastroenterology*, *37*, 105–18.

Renkema, K. Y., Alexander, R. T., Bindels, R. J., & Hoenderop, J. G. (2008). Calcium and phosphate homeostasis: concerted interplay of new regulators. *Annals of Medicine*, *40*, 82–91.

Rhodes, E. C., Martin, A. D., Taunton, J. E., Donnelly, M., Warren, J., & Elliot, J. (2000). Effects of one year of resistance training on the relation between muscular strength and bone density in elderly women. *British Journal of Sports Medicine*, *34*, 18–22.

Riggs, B. L., Melton Iii, L. J., Robb, R. A., Camp, J. J., Atkinson, E. J., Peterson, J. M., Khosla, S. (2004). Population-based study of age and sex differences in bone volumetric density, size, geometry, and structure at different skeletal sites. *Journal of Bone and Mineral Research*, *19*, 1945–54.

Riggs, B. L., Wahner, H. W., Melton, L. J., Richelson, L. S., Judd, H. L., & O'Fallon, W. M. (1987). Dietary calcium intake and rates of bone loss in women. *The Journal of Clinical Investigation*, *80*, 979–82.

Roberfroid, M. B. (2000a). Prebiotics and probiotics: are they functional foods? *American Journal of Clinical Nutrition*, *71*, 1682S–7S.

Roberfroid, M. B. (2000b). Prebiotics and probiotics: are they functional foods? *American Journal of Clinical Nutrition*, *71*, 1682S–1687.

Rodrigues, F. C., Castro, A. S. B., Rodrigues, V. C., Fernandes, S. A., Fontes, E. A. F., de Oliveira, T. T., de Luces Fortes Ferreira, C. L. (2012). Yacon flour and Bifidobacterium longum modulate bone health in rats. *Journal of Medicinal Food*, *15*, 664–70.

Roschger, P., Rinnerthaler, S., Yates, J., Rodan, G. A., Fratzl, P., & Klaushofer, K. (2001). Alendronate increases degree and uniformity of mineralization in cancellous bone and decreases the porosity in cortical bone of osteoporotic women. *Bone*, *29*, 185–91.

Rosen, H. N., Moses, A. C., Garber, J., Iloputaife, I. D., Ross, D. S., Lee, S. L., & Greenspan, S. L. (2000). Serum CTX: a new marker of bone resorption that shows treatment effect more often than other markers because of low coefficient of variability and large changes with bisphosphonate therapy. *Calcified Tissue International*, *66*, 100–3.

Ross, A. C., Taylor, C. L., Yaktine, A. L., & Valle, H. B. Del. (2011). Overview of Calcium. National Academies Press (US). Retrieved from http://www.ncbi.nlm.nih.gov/books/NBK56060/

Rouach, V., Katzburg, S., Koch, Y., Stern, N., & Somjen, D. (2011). Bone loss in ovariectomized rats: dominant role for estrogen but apparently not for FSH. *Journal of Cellular Biochemistry*, *112*, 128–37.

Round, J. L., & Mazmanian, S. K. (2009). The gut microbiota shapes intestinal immune responses during health and disease. *Nature Reviews. Immunology*, *9*, 313–23.

Ruiz, J. C., Mandel, C., & Garabedian, M. (1995). Influence of spontaneous calcium intake and physical exercise on the vertebral and femoral bone mineral density of children and adolescents. *Journal of Bone and Mineral Research*, *10*, 675–82.

Saarela, M., Mogensen, G., Fondén, R., Mättö, J., & Mattila-Sandholm, T. (2000). Probiotic bacteria: safety, functional and technological properties. *Journal of Biotechnology*, *84*, 197–215.

Sakurai, T. (2007). The neural circuit of orexin (hypocretin): maintaining sleep and wakefulness. *Nature Reviews. Neuroscience*, *8*, 171–81.

Salive, M. E., Cornoni-Huntley, J., Phillips, C. L., Guralnik, J. M., Cohen, H. J., Ostfeld, A. M., & Wallace, R. B. (1992). Serum albumin in older persons: relationship with age and health status. *Journal of Clinical Epidemiology*, *45*, 213–21.

Sanchez-Mateos, S., Alonso-Gonzalez, C., Gonzalez, A., Martinez-Campa, C. M., Mediavilla, M. D., Cos, S., & Sanchez-Barcelo, E. J. (2007). Melatonin and estradiol effects on food intake, body weight, and leptin in ovariectomized rats. *Maturitas*, *58*, 91–101.

Sanders, M. E. (2008). Probiotics: definition, sources, selection, and uses. *Clinical Infectious Diseases*, *46*, S58–61.

Schinke, T., Schilling, A. F., Baranowsky, A., Seitz, S., Marshall, R. P., Linn, T., Amling, M. (2009). Impaired gastric acidification negatively affects calcium homeostasis and bone mass. *Nature Medicine*, *15*, 674–81.

Schoenau, E., Neu, C. M., Beck, B., Manz, F., & Rauch, F. (2002). Bone mineral content per muscle cross-sectional area as an index of the functional muscle-bone unit. *Journal of Bone and Mineral Research*, *17*, 1095–101.

Scholz-Ahrens, K. E., Açil, Y., & Schrezenmeir, J. (2002). Effect of oligofructose or dietary calcium on repeated calcium and phosphorus balances, bone mineralization and trabecular structure in ovariectomized rats. *The British Journal of Nutrition*, *88*, 365–77.

Scholz-Ahrens, K. E., Ade, P., Marten, B., Weber, P., Timm, W., Açil, Y., Schrezenmeir, J. (2007). Prebiotics, probiotics, and synbiotics affect mineral absorption, bone mineral content, and bone structure. *The Journal of Nutrition*, *137*, 838S–46S.

Seeman, E. (2002). Pathogenesis of bone fragility in women and men. *Lancet*, *359*, 1841–50.

Seibel, M. J. (2005). Biochemical markers of bone turnover: part I: biochemistry and variability. *The Clinical Biochemist. Reviews*, *26*, 97–122.

Senok, A. C., Ismaeel, A. Y., & Botta, G. A. (2005). Probiotics: facts and myths. *Clinical Microbiology and Infection*, *11*, 958–66.

Sharma, R., Callaway, D., Vanegas, D., Bendele, M., Lopez-Cruzan, M., Horn, D., Herman, B. (2014). Caspase-2 maintains bone homeostasis by inducing apoptosis of oxidatively-damaged osteoclasts. *PloS One*, *9*, e93696.

Shen, V., Dempster, D. W., Birchman, R., Xu, R., & Lindsay, R. (1993). Loss of cancellous bone mass and connectivity in ovariectomized rats can be restored by combined treatment with parathyroid hormone and estradiol. *The Journal of Clinical Investigation*, *91*, 2479–87.

Sheng, L., Cao, W., Cha, B., Chen, Z., Wang, F., & Liu, J. (2013). Serum osteocalcin level and its association with carotid atherosclerosis in patients with type 2 diabetes. *Cardiovascular Diabetology*, *12*, 22.

Sherman, P. M., Ossa, J. C., & Johnson-Henry, K. (2009). Unraveling mechanisms of action of probiotics. *Nutrition in Clinical Practice*, *24*, 10–4.

Shim, K.-S., Kim, T., Ha, H., Lee, K. J., Cho, C.-W., Kim, H. S., Ma, J. Y. (2013). Lactobacillus fermentation enhances the inhibitory effect of Hwangryun-haedok-tang in an ovariectomy-induced bone loss. *BMC Complementary and Alternative Medicine*, *13*, 106.

Shimada, T., Kakitani, M., Yamazaki, Y., Hasegawa, H., Takeuchi, Y., Fujita, T., Yamashita, T. (2004). Targeted ablation of Fgf23 demonstrates an essential physiological role of FGF23 in phosphate and vitamin D metabolism. *The Journal of Clinical Investigation*, *113*, 561–8.

Shuke, N., Aburano, T., Okizaki, A., Zhao, C., Nakajima, K., Yokoyama, K., Tonami, N. (2003). Estimation of fractional liver uptake and blood retention of 99mTc-DTPA-galactosyl human serum albumin: an application of a simple graphical method to dynamic SPECT. *Nuclear Medicine Communications*, *24*, 503–11.

Simkin-Silverman, L. R., Wing, R. R., Boraz, M. A., & Kuller, L. H. (2003). Lifestyle intervention can prevent weight gain during menopause: results from a 5-year randomized clinical trial. *Annals of Behavioral Medicine*, *26*, 212–20.

Sims, N. A., Clément-Lacroix, P., Minet, D., Fraslon-Vanhulle, C., Gaillard-Kelly, M., Resche-Rigon, M., & Baron, R. (2003). A functional androgen receptor is not sufficient to allow estradiol to protect bone after gonadectomy in estradiol receptor-deficient mice. *The Journal of Clinical Investigation*, *111*, 1319–27.

Sims, N. A., & Martin, T. J. (2014). Coupling the activities of bone formation and resorption: a multitude of signals within the basic multicellular unit. *BoneKEy Reports*, *3*, 481.

Sinha, K. M., & Zhou, X. (2013). Genetic and molecular control of osterix in skeletal formation. *Journal of Cellular Biochemistry*, *114*, 975–84.

Siu, W. S., Qin, L., & Leung, K. S. (2003). pQCT bone strength index may serve as a better predictor than bone mineral density for long bone breaking strength. *Journal of Bone and Mineral Metabolism*, *21*, 316–22.

Sjögren, K., Engdahl, C., Henning, P., Lerner, U. H., Tremaroli, V., Lagerquist, M. K., Ohlsson, C. (2012). The gut microbiota regulates bone mass in mice. *Journal of Bone and Mineral Research*, *27*, 1357–67.

Snydman, D. R. (2008). The safety of probiotics. *Clinical Infectious Diseases*, *46*, S104–11.

Soleman, N., Laferl, H., Kneifel, W., Tucek, G., Budschedl, E., Weber, H., Mayer, H. K. (2003). How safe is safe?--a case of Lactobacillus paracasei ssp. paracasei endocarditis and discussion of the safety of lactic acid bacteria. *Scandinavian Journal of Infectious Diseases*, *35*, 759–62.

Specker, B., & Binkley, T. (2003). Randomized trial of physical activity and calcium supplementation on bone mineral content in 3- to 5-year-old children. *Journal of Bone and Mineral Research*, *18*, 885–92.

Staa, T. P., Dennison, E. M., Leufkens, H. G., & Cooper, C. (2001). Epidemiology of fractures in England and Wales. *Bone*, *29*, 517–22.

Stachenfeld, N. S. (2014). Hormonal changes during menopause and the impact on fluid regulation. *Reproductive Sciences*, *21*, 555–61.

Stanislaus, D., Yang, X., Liang, J. D., Wolfe, J., Cain, R. L., Onyia, J. E., Hock, J. M. (2000). In vivo regulation of apoptosis in metaphyseal trabecular bone of young rats by synthetic human parathyroid hormone (1-34) fragment. *Bone*, *27*, 209–18.

Styrkarsdottir, U., Cazier, J.-B., Kong, A., Rolfsson, O., Larsen, H., Bjarnadottir, E., Stefansson, K. (2003). Linkage of osteoporosis to chromosome 20p12 and association to BMP2. *PLoS Biology*, *1*, E69.

Sullivan, A., & Nord, C. E. (2005). Probiotics and gastrointestinal diseases. *Journal of Internal Medicine*, *257*, 78–92.

Swain, M. R., Anandharaj, M., Ray, R. C., & Parveen Rani, R. (2014). Fermented fruits and vegetables of Asia: a potential source of probiotics. *Biotechnology Research International*, *2014*, 250424.

Szulc, P. (2006). Bone density, geometry, and fracture in elderly men. *Current Osteoporosis Reports*, *4*, 57–63.

Szulc, P., & Delmas, P. D. (2008). Biochemical markers of bone turnover: potential use in the investigation and management of postmenopausal osteoporosis. *Osteoporosis International*, *19*, 1683–704.

Tabb, M. M., Sun, A., Zhou, C., Grün, F., Errandi, J., Romero, K., Blumberg, B. (2003). Vitamin K2 regulation of bone homeostasis is mediated by the steroid and xenobiotic receptor SXR. *The Journal of Biological Chemistry*, *278*, 43919–27.

Tabbers, M. M., de Milliano, I., Roseboom, M. G., & Benninga, M. A. (2011). Is Bifidobacterium breve effective in the treatment of childhood constipation? Results from a pilot study. *Nutrition Journal*, *10*, 19.

Taguchi, H., Chen, H., Yano, R., & Shoumura, S. (2006). Comparative effects of milk and soymilk on bone loss in adult ovariectomized osteoporosis rat. *Okajimas Folia Anatomica Japonica*, *83*, 53–9.

Tanizawa, T., Yamaguchi, A., Uchiyama, Y., Miyaura, C., Ikeda, T., Ejiri, S., Nagai, Y. (2000). Reduction in bone formation and elevated bone resorption in ovariectomized rats with special reference to acute inflammation. *Bone*, *26*, 43–53.

Tannock, G. W. (1997). Probiotic properties of lactic-acid bacteria: plenty of scope for fundamental R & D. *Trends in Biotechnology*, *15*, 270–4.

Tenover, F. C. (2006). Mechanisms of antimicrobial resistance in bacteria. *The American Journal of Medicine*, *119*, S3–10; discussion S62–70.

Theman, T. A., & Collins, M. T. (2009). The role of the calcium-sensing receptor in bone biology and pathophysiology. *Current Pharmaceutical Biotechnology*, *10*, 289–301.

Theobald, T. M., Cauley, J. A., Gluer, C. C., Bunker, C. H., Ukoli, F. A., & Genant, H. K. (1998). Black-white differences in hip geometry. Study of Osteoporotic Fractures Research Group. *Osteoporosis International*, *8*, 61–7.

Thys-Jacobs, S., McMahon, D., & Bilezikian, J. P. (2007). Cyclical changes in calcium metabolism across the menstrual cycle in women with premenstrual dysphoric disorder. *The Journal of Clinical Endocrinology and Metabolism*, *92*, 2952–9.

Toba, Y., Kajita, Y., Masuyama, R., Takada, Y., Suzuki, K., & Aoe, S. (2000). Dietary Magnesium Supplementation Affects Bone Metabolism and Dynamic Strength of Bone in Ovariectomized Rats. *Journal of Nutrition.*, *130*, 216–220.

Tomofuji, T., Ekuni, D., Azuma, T., Irie, K., Endo, Y., Yamamoto, T., Morita, M. (2012). Supplementation of broccoli or Bifidobacterium longum-fermented broccoli suppresses serum lipid peroxidation and osteoclast differentiation on alveolar bone surface in rats fed a high-cholesterol diet. *Nutrition Research*, *32*, 301–7.

Tsuchita, H., Goto, T., Shimizu, T., Yonehara, Y., & Kuwata, T. (1996). Dietary casein phosphopeptides prevent bone loss in aged ovariectomized rats. *The Journal of Nutrition*, *126*, 86–93.

Tuomola, E., Crittenden, R., Playne, M., Isolauri, E., & Salminen, S. (2001). Quality assurance criteria for probiotic bacteria. *The American Journal of Clinical Nutrition*, *73*, 393S–398S.

Turner, A. S. (2001). Animal models of osteoporosis--necessity and limitations. *European Cells & Materials*, *1*, 66–81.

Turner, R. T. (2000). Invited review: what do we know about the effects of spaceflight on bone? *Journal of Applied Physiology*, *89*, 840–7.

Usman, & Hosono, A. (1999). Viability of Lactobacillus gasseri and its cholesterol-binding and antimutagenic activities during subsequent refrigerated storage in nonfermented milk. *Journal of Dairy Science*, *82*, 2536–42.

Valeur, N., Engel, P., Carbajal, N., Connolly, E., & Ladefoged, K. (2004). Colonization and immunomodulation by Lactobacillus reuteri ATCC 55730 in the human gastrointestinal tract. *Applied and Environmental Microbiology*, *70*, 1176–81.

Vanh, Outen, J. N., & Wysolmerski, J. J. (2003). Low estrogen and high parathyroid hormone-related peptide levels contribute to accelerated bone resorption and bone loss in lactating mice. *Endocrinology*, *144*, 5521–9.

Verna, E. C., & Lucak, S. (2010). Use of probiotics in gastrointestinal disorders: what to recommend? *Therapeutic Advances in Gastroenterology*, *3*, 307–19.

Viguet-Carrin, S., Garnero, P., & Delmas, P. D. (2006). The role of collagen in bone strength. *Osteoporosis International*, *17*, 319–36.

Voort, D. J., Geusens, P. P., & Dinant, G. J. (2001). Risk factors for osteoporosis related to their outcome: fractures. *Osteoporosis International*, *12*, 630–8.

Voreades, N., Kozil, A., & Weir, T. L. (2014). Diet and the development of the human intestinal microbiome. *Frontiers in Microbiology*, *5*, 494.

Wagner, R. D., Pierson, C., Warner, T., Dohnalek, M., Farmer, J., Roberts, L., Balish, E. (1997). Biotherapeutic effects of probiotic bacteria on candidiasis in immunodeficient mice. *Infection and Immunity*, *65*, 4165–72.

Wakamatsu, H., Nagamachi, S., Kiyohara, S., Fujita, S., Kamimura, K., Futami, S., Tamura, S. (2010). Predictive value of Tc-99m galactosyl human serum albumin liver SPECT on the assessment of functional recovery after partial hepatectomy: a comparison with CT volumetry. *Annals of Nuclear Medicine*, *24*, 729–34.

Wang, F., Wang, P., Wu, X., Dang, S., Chen, Y., Ni, Y., Pang, X. (2013). Deficiency of adiponectin protects against ovariectomy-induced osteoporosis in mice. *PloS One*, *8*, e68497.

Wassertheil-Smoller, S., Hendrix, S. L., Limacher, M., Heiss, G., Kooperberg, C., Baird, A., Mysiw, W. J. (2003). Effect of estrogen plus progestin on stroke in postmenopausal women: the Women's Health Initiative: a randomized trial. *The Journal of the American Medical Association*, *289*, 2673–84.

Wendlova, J. (2008). Bone quality. Elasticity and strength. *Bratislavské Lekárske Listy*, *109*, 383–6.

Wilfinger, W. W., Mackey, K., & Chomczynski, P. (1997). Effect of pH and ionic strength on the spectrophotometric assessment of nucleic acid purity. *BioTechniques*, *22*, 474–6, 478–81.

Wilkins, B. J., & Lewis, J. S. (2009). Non-functional parathyroid carcinoma: a review of the literature and report of a case requiring extensive surgery. *Head and Neck Pathology*, *3*, 140–9.

Wing, R. R. (1991). Weight Gain at the Time of Menopause. *Archives of Internal Medicine*, *151*, 97.

Wojciech, R. M. and Ś. (2002). Effect of ovariectomy on biochemical markers of bone turnover (alp, acp) and calcium content in rat mandible and teeth. *Bull. Vet. Inst. Pulawy*, *46*, 281–287.

Wolf, R. L., Cauley, J. A., Baker, C. E., Ferrell, R. E., Charron, M., Caggiula, A. W., Kuller, L. H. (2000). Factors associated with calcium absorption efficiency in pre- and perimenopausal women. *The American Journal of Clinical Nutrition*, *72*, 466–71.

Wong, L. P., & Nur Liyana. (2007). A Survey of Knowledge and Perceptions of Menopause among Young to iddle-Aged Women in Federal Territory, Kuala Lumpur, Malaysia. *Journal of the University of Malaya Medical Centre*, *10*, 22–30.

Woolf, A. D., & Pfleger, B. (2003). Burden of major musculoskeletal conditions. *Bulletin of the World Health Organization*, *81*, 646–56.

Wronski, T. J., Lowry, P. L., Walsh, C. C., & Ignaszewski, L. A. (1985). Skeletal alterations in ovariectomized rats. *Calcified Tissue International*, *37*, 324–8.

Wronski, T. J., Walsh, C. C., & Ignaszewski, L. A. (1986). Histologic evidence for osteopenia and increased bone turnover in ovariectomized rats. *Bone*, *7*, 119–123.

Wu, Y., Ackerman, J. L., Strawich, E. S., Rey, C., Kim, H.-M., & Glimcher, M. J. (2003). Phosphate ions in bone: identification of a calcium-organic phosphate complex by 31P solid-state NMR spectroscopy at early stages of mineralization. *Calcified Tissue International*, *72*, 610–26.

Xiao, J. Z., Kondo, S., Yanagisawa, N., Takahashi, N., Odamaki, T., Iwabuchi, N., Enomoto, T. (2006). Effect of probiotic Bifidobacterium longum BB536 [corrected] in relieving clinical symptoms and modulating plasma cytokine levels of Japanese cedar pollinosis during the pollen season. A randomized double-blind, placebo-controlled trial. *Journal of Investigational Allergology & Clinical Immunology*, *16*, 86–93.

Yamauchi, H., Kushida, K., Yamazaki, K., & Inoue, T. (1995). Assessment of spine bone mineral density in ovariectomized rats using DXA. *Journal of Bone and Mineral Research*, *10*, 1033–9.

Yan, F., & Polk, D. B. (2006). Probiotics as functional food in the treatment of diarrhea. *Current Opinion in Clinical Nutrition and Metabolic Care*, *9*, 717–21.

Yang, X., Chan, Y. H., Muthukumaran, P., & Lee, T. (2010). Morphological and Mechanical Changes in Ovariectomized Rat Tibia with Treatments of Ibandronate and Parathyroid Hormone. *Osteoporosis*, *8*, 255–265.

Yildiz, M., & Oral, B. (2003). Effects of menopause on bone mineral density in women with endemic fluorosis. *Clinical Nuclear Medicine*, *28*, 308–11.

Young, S. L., Simon, M. A., Baird, M. A., Tannock, G. W., Bibiloni, R., Spencely, K., Woodcock, A. (2004). Bifidobacterial species differentially affect expression of cell surface markers and cytokines of dendritic cells harvested from cord blood. *Clinical and Diagnostic Laboratory Immunology*, *11*, 686–90.

Yuehuei, Robert, D. (1999). *Mechanical testing of bone and the bone-implant interface* (pp. 207–217). CRC press, Boca Raton London New York Washington, DC.

Zaiss, M. M., Sarter, K., Hess, A., Engelke, K., Böhm, C., Nimmerjahn, F., David, J.-P. (2010). Increased bone density and resistance to ovariectomy-induced bone loss in FoxP3-transgenic mice based on impaired osteoclast differentiation. *Arthritis and Rheumatism*, *62*, 2328–38.

Zárate, G., Morata De Ambrosini, V., Perez Chaia, A., & González, S. (2002). Some factors affecting the adherence of probiotic Propionibacterium acidipropionici CRL 1198 to intestinal epithelial cells. *Canadian Journal of Microbiology*, *48*, 449–57.

Zareie, M., Johnson-Henry, K., Jury, J., Yang, P.-C., Ngan, B.-Y., McKay, D. M., Sherman, P. M. (2006). Probiotics prevent bacterial translocation and improve intestinal barrier function in rats following chronic psychological stress. *Gut*, *55*, 1553–60.

Zhang, F., Chang, J., Lu, J., Lin, K., & Ning, C. (2007). Bioinspired structure of bioceramics for bone regeneration in load-bearing sites. *Acta Biomaterialia*, *3*, 896–904.

Zhang, Y., Lai, W.-P., Wu, C.-F., Favus, M. J., Leung, P.-C., & Wong, M.-S. (2007). Ovariectomy worsens secondary hyperparathyroidism in mature rats during low-Ca diet. *American Journal of Physiology. Endocrinology and Metabolism*, *292*, E723–31.

Zhang, Y., Venugopal, J. R., El-Turki, A., Ramakrishna, S., Su, B., & Lim, C. T. (2008). Electrospun biomimetic nanocomposite nanofibers of hydroxyapatite/chitosan for bone tissue engineering. *Biomaterials*, *29*, 4314–22.

Zhang, Y., Wang, L., Zhang, J., Li, Y., He, Q., Li, H., Zhang, H. (2014). Probiotic Lactobacillus casei Zhang ameliorates high-fructose-induced impaired glucose tolerance in hyperinsulinemia rats. *European Journal of Nutrition*, *53*, 221–32.

Zhou, J. S., Shu, Q., Rutherfurd, K. J., Prasad, J., Birtles, M. J., Gopal, P. K., & Gill, H. S. (2000). Safety assessment of potential probiotic lactic acid bacterial strains Lactobacillus rhamnosus HN001, Lb. acidophilus HN017, and Bifidobacterium lactis HN019 in BALB/c mice. *International Journal of Food Microbiology*, *56*, 87–96.

Zhou, J. S., Shu, Q., Rutherfurd, K. J., Prasad, J., Gopal, P. K., & Gill, H. S. (2000). Acute oral toxicity and bacterial translocation studies on potentially probiotic strains of lactic acid bacteria. *Food and Chemical Toxicology*, *38*, 153–61.

Zhou, X., Zhang, Z., Feng, J. Q., Dusevich, V. M., Sinha, K., Zhang, H., de Crombrugghe, B. (2010). Multiple functions of Osterix are required for bone growth and homeostasis in postnatal mice. *Proceedings of the National Academy of Sciences of the United States of America*, *107*, 12919–24.

Zioupos, P., Cook, R. B., & Hutchinson, J. R. (2008). Some basic relationships between density values in cancellous and cortical bone. *Journal of Biomechanics*, *41*, 1961–8.

Zoppi, G., Cinquetti, M., Benini, A., Bonamini, E., & Minelli, E. B. (2001). Modulation of the intestinal ecosystem by probiotics and lactulose in children during treatment with ceftriaxone. *Current Therapeutic Research*, *62*, 418–435.