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COMPOSITIONS AND METHODS FOR ALTERING GENE EXPRESSION

CROSS-REFERENCE TO RELATED APPLICATIONS

[0001] This application claims priority to U.S. Provisional Application No. 60/891,171, filed on February 22, 2007.

FIELD OF THE INVENTION

[0002] The present invention relates to compositions and methods regulating gene expression or transcription in a growing animal.

BACKGROUND OF THE INVENTION

[0003] Commercially available canine and feline foods include compositions specially formulated to address many different nutritional needs. These include, for example, formulations designed for different breed types, sizes and body conditions. They also include formulations designed to address the nutritional needs of animals in the different stages of their life cycle. Typically, these stages include the growth, adult and senior stages of life. For example, US Patent 5,851,573 discloses a pet food composition for large breed puppies; US Patent 6,426,100 discloses compositions to provide improved bone modeling and chondrocyte functioning in growing animals; US Patent 6,582,752 discloses gender specific puppy food. Despite the availability of such pet food formulations, however, the need remains for the development of additional formulations comprising innovative ingredients and nutrients designed to enhance the development of growing animals.

[0004] It is known in the art that certain nutrients have an effect on gene expression. Nutrigenomics is the study of such a relationship. Despite what is already known, there is a need to develop compositions and methods which may positively influence gene expression of an animal.

SUMMARY OF THE INVENTION

[0005] In certain aspects, the present invention relates to compositions that are useful to enhance the development of a growing animal. Particularly, the compositions of the present invention comprise one or more nutrients or bioactive substances that can enhance neurologic development, bone and joint health, immune function, and promote healthy body composition in a growing animal. In certain embodiments, the nutrients and bioactive substances include, but are not limited to, fatty acids, antioxidants, essential nutrients, amino acids, minerals and trace elements, vitamins and vitamin-like substances. Other aspects of the invention relate to methods to enhance the development of a growing animal comprising administration of effective amounts of the compositions of the present invention directly to a growing animal or to the dam of said animal while the animal is *in utero* or is a nursling.

[0006] Thus, in one aspect, the present invention includes Composition 1.0, a pet food composition comprising:

- about 5 to about 70% protein,
- about 0.5 to about 1.6% methionine,
- about 50 to about 200 ppm manganese,
- about 0.1 to about 0.5% DHA,
- about 0.1 to about 0.7% EPA,
- about 1200 to about 7500 ppm choline,
- about 1000 to about 2000 ppm taurine,
- about 2.5 to about 6% linoleic acid,
- about 1 to about 3% total n-3 fatty acids,
- about 50 to about 1200 IU/kg vitamin E,
- about 50 to about 500 ppm vitamin C,

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about 50 to about 500 ppm carnitine, and
and about 2.5 to about 7 g lysine/1000 kcal. Composition 1.0 may be used
for the regulation of gene expression in a canine.

[0007] The preset invention also includes the following compositions:

1.1 Composition 1.0 comprising:

0 to about 90% by weight of carbohydrates;
about 20% to about 60% by weight of protein;
about 2% to about 50% by weight of fat;
about 0.1% to about 20%, by weight of total dietary fiber;
and
0 to about 15% by weight of vitamins, minerals, and other
nutrients in varying percentages which support the
nutritional needs of the animal.

1.2 Composition 1.0 or 1.1 comprising about 5% to about 55%, by weight
of carbohydrates;

1.3 Any of the preceding compositions comprising about 20% to about
50%, by weight of protein, e.g., about 22% to about 50%;

1.4 Any of the preceding compositions comprising about 5% to about 40%,
by weight of fat, e.g., e.g., at least about 8% or about 9% to about 40%
fat;

1.5 Any of the preceding compositions comprising about 1% to about 11%,
by weight of total dietary fiber;

1.6 Any of the preceding compositions comprising about 0.1% to about
0.4% DHA, e.g., about 0.5%;

1.7 Any of the preceding compositions comprising about 200 IU/kg to
about 1200 IU/kg Vitamin E, e.g., about 200 IU/kg to about 1000
IU/kg;

1.9 Any of the preceding compositions comprising about 100 ppm to

about 500 ppm carnitine, e.g., about 200 to about 400, or about 300 ppm;

- 1.10 Any of the preceding compositions comprising about 2.5 g/1000 kcal to about 7 g/1000 kcal lysine;
- 1.11 Any of the preceding compositions comprising about 2500 ppm to about 7500 ppm choline, e.g., about 3000, about 3500, about 4000, about 4500, about 4600, about 4625, about 4650, about 4700, about 4800, or about 6000 ppm;
- 1.12 Any of the preceding compositions comprising about 0.1 % to about 0.6 % EPA;
- 1.13 Any of the preceding compositions comprising about 50 ppm to about 150 ppm manganese; and
- 1.14 Any of the preceding compositions comprising about 0.8% to about 1.6% methionine.

[0008] In another aspect, the invention relates to methods to cause a beneficial modification in gene expression in an animal, specifically, down regulation in expression of a gene or genes associated with an undesirable biological condition or pathway or disease state and/or up regulation in expression of a gene or genes associated with a desired biological condition or pathway or which may have a positive or preventive effect on a disease state for any one or more biological conditions, pathways or disease states and genes described in Tables 2 - 15, comprising administering an effective amount of a composition of according to any one of compositions 1.0 - 1.14 to an animal, either directly to the animal or to the dam while the animal is *in utero*.

[0009] In another aspect, the invention is directed to Method 2.0, a method to regulate gene expression in a canine comprising administering to the canine

any one of compositions the compositions of the present invention, e.g., compositions 1.0 – 1.14.

[0010] The present invention also includes the following methods:

- 2.1 Of method 2.0 wherein the canine is a puppy.
- 2.2 Of method 2.0 or 2.1 wherein the canine is born of a dam fed any one of compositions 1.0 – 1.14 during pregnancy.
- 2.3 Of method 2.2 wherein the puppy is *in utero*.
- 2.4 Of method 2.2 wherein the dam is fed any one of compositions 1.0 – 1.14 prior to pregnancy.
- 2.5 Of method 2.2 or 2.4 wherein the dam is fed any one of compositions 1.0 – 1.14 for a majority of the pregnancy duration.
- 2.6 Of any one of methods 2.2 – 2.5 wherein the dam is fed compositions consisting essentially of any one of compositions 1.0 – 1.14 prior to and during pregnancy.
- 2.7 Of any one of the preceding methods wherein the puppy is fed any one of compositions 1.0 – 1.14 prior to weaning, e.g., while still a nursling.
- 2.8 Of any one of the preceding methods wherein the puppy is fed any one of compositions 1.0 – 1.14 post weaning.
- 2.9 Of method 2.8 wherein the puppy is fed food compositions consisting of any one of compositions 1.0 – 1.14.
- 2.10 Of any one of the preceding methods wherein an effective amount of the composition is administered to the canine.
- 2.11 Of any one of the preceding methods wherein the composition is administered to the canine for an effective amount of time.
- 2.12 Of method 2.11 wherein the composition is administered to the canine for at least one year post partum or one year post weaning.

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2.13 Of any one of the preceding method wherein the gene is selected from those listed in Tables 2-15.

[0010a] In a further aspect, the invention relates to a pet food composition for the regulation of a gene in a canine puppy, comprising: about 5 to about 70% protein, about 0.5 to about 1.6% methionine, about 50 to about 200 ppm manganese, about 0.1 to about 0.5% DHA, about 0.1 to about 0.7% EPA, about 1200 to about 7500 ppm choline, about 1000 to about 2000 ppm taurine, about 2.5 to about 6% linoleic acid, about 1 to about 3% total n-3 fatty acids, about 50 to about 1200 IU/kg vitamin E, about 50 to about 500 ppm vitamin C, about 50 to about 500 ppm carnitine, and about 2.5 to about 7g lysine/1000 kcal, and the regulation of the gene is at least one of: (a) down-regulation of genes associated with arthritis or inflammation, wherein the genes associated with arthritis or inflammation comprise interleukin 1-beta, fibronectin, v-fos FBJ murine osteosarcoma viral oncogene homolog, dual specificity phosphatase 1, lactotransferrin, compliment component 5a receptor, xanthine dehydrogenase, and secreted phosphoprotein 1; (b) down-regulation of genes associated with DNA fragmentation, wherein the genes associated with DNA fragmentation comprise interleukin 1-beta, fibronectin, neuroregulin 1, dual specificity phosphatase 1, and cell death-inducing DFFA-like effector a; (c) up-regulation of genes associated with DNA processing control, wherein the genes associated with DNA processing control comprise budding uninhibited by benzimidazoles 1 homolog, topoisomerase II alpha, kinetochore associated 2, claspin homolog, cyclin B1, cyclin B2, human chromosome condensation protein G, leucine zipper protein 5, kinesin family member 23, and flap structure specific endonuclease I; (d) down-regulation of genes associated with cancer incidence, wherein the genes associated with cancer incidence comprise interleukin 1-beta, fibronectin, v-fos FBJ murine osteosarcoma viral oncogene homolog, dual specificity phosphatase 1, compliment component 5a receptor, xanthine dehydrogenase, cellular repressor of E1A-stimulated genes 1, chondroitin sulfate proteoglycan 2, mycrocytic anemia transcobalamin II, clade B member 10 serpin peptidase inhibitor, adenomatosis polyposis coli down-regulated 1, six membrane epithelial antigen of the prostate 2, and alanyl aminopeptidase; (e) up-regulation of genes associated with tumor suppression, wherein the genes associated with tumor suppression comprise cancer susceptibility candidate 5, paternally expressed 3,

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topoisomerase II alpha, tubulin alpha 6, cyclin B1, cell division cycle associated 1, cell division cycle 2, ribonucleotide reductase M2 polypeptide, breast cancer anti-estrogen resistance 3, tumor necrosis factor receptor superfamily member 17, and protein kinase C iota; (f) down-regulation of genes associated with cellular compromise, wherein the genes

5 associated with cellular compromise comprise interleukin 1-beta, fibronectin, neuroregulin 1, v-fos FBJ murine osteosarcoma viral oncogene homolog, dual specificity phosphatase 1, compliment component 5a receptor, xanthine dehydrogenase, and macrophage receptor with collagenous structure; (g) up-regulation of genes associated with cellular integrity, wherein the genes associated with cellular integrity comprise cyclin B1, cell division cycle 2,

10 hemoglobin epsilon 1, and protein kinase C iota; (h) down-regulation of genes associated with disruption of cellular assembly, wherein the genes associated with disruption of cellular assembly comprise interleukin 1-beta, fibronectin, neuroregulin 1, chondroitin sulfate proteoglycan 2, muscarinic 3 cholinergic receptor, xanthine dehydrogenase, and macrophage receptor with collagenous structure; (i) up-regulation of genes associated with cellular

15 organization, wherein the genes associated with cellular organization comprise cyclin B1, cell division cycle 2, leucine zipper protein 5, protein kinase C iota, kinesin family member 23, budding uninhibited by benzimidazoles 1 homolog, cyclin B2, topoisomerase II alpha, kinetochore associated 2, cell division cycle associated 1, human chromosome condensation protein G, kinesin family member 11, A kinase (PRKA) anchor protein 2, and thymopioten;

20 (j) down-regulation of genes associated with disruption of cell cycle control, wherein the genes associated with disruption of cell cycle control comprise interleukin 1-beta, fibronectin, neuroregulin 1, v-fos FBJ murine osteosarcoma viral oncogene homolog, muscarinic 3 cholinergic receptor, dual specificity phosphatase 1, and cellular repressor of E1A-stimulated genes 1; (k) up-regulation of genes associated with cell cycle regulation, wherein the genes

25 associated with cell cycle regulation comprise cyclin B1, cell division cycle 2, leucine zipper protein 5, 350/400ka mitotin centromere protein F, breast cancer anti-estrogen resistance 3, budding uninhibited by benzimidazoles 1 homolog, cyclin B2, topoisomerase II alpha, kinetochore associated 2, cell division cycle associated 1, human chromosome condensation protein G, kinesin family member 11, and thymopioten; (l) down-regulation of genes

30 associated with immune activation, wherein the genes associated with immune activation comprise lymphocyte antigen 6 complex, fibrinogen-like 2, CD1A antigen, complement

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factor D (adipsin), CD163 or macrophage-associated antigen, toll-like receptor 2, renal amyloidosis lysozyme, transcription elongation factor A (SII) 3, fibronectin 1, macrophage receptor with collagenous structure, bactericidal/permeability-increasing protein, T cell receptor alpha locus, and immunoglobulin heavy constant alpha 2; (m) down-regulation of genes associated with lipid metabolism, wherein the genes associated with lipid metabolism comprise toll-like receptor 2, cytochrome P450 family 1 subfamily B, polypeptide 1, colony stimulating factor 1 receptor, phospholipase C beta 1, fibronectin 1, neuregulin 1, macrophage receptor with collagenous structure, and CD1a molecule; (n) down-regulation of genes associated with cardiovascular development, wherein the genes associated with cardiovascular development comprise bactericidal/permeability-increasing protein, fibronectin 1, neuregulin 1, xanthine dehydrogenase, annexin A6, colony stimulating factor 1 receptor, and alanyl (membrane) aminopeptidase; (o) down-regulation of genes associated with skeletal muscular disorders, wherein the genes associated with skeletal muscular disorders comprise toll-like receptor 2, colony stimulating factor 1 receptor, fibronectin 1, and neuregulin 1; (p) up-regulation of genes associated with muscle contraction/function, wherein the genes associated with muscle contraction/function comprise exostoses, microfibrillar associated protein 5, myosin light polypeptide kinase, dystroglycan 1, and moloney leukemia virus 10-like 1; (q) down-regulation of genes associated with cellular compromise, wherein the genes associated with cellular compromise comprise neuroregulin 1, fibronectin 1, macrophage receptor with collagenous structure, xanthine dehydrogenase, bactericidal permeability increasing protein, and phospholipase C beta 1; and (r) down-regulation of genes associated with cancer, wherein the genes associated with cancer comprise fibronectin 1, xanthine dehydrogenase, neuregulin 1, colony stimulating factor 1 receptor, alanyl aminopeptidase, and S100 calcium binding protein.

[0010b] In a further aspect, the invention relates to a pet food composition for the regulation of a gene in a canine puppy, comprising: about 6% moisture, about 29% crude protein, about 18% fat, about 1.4% Ca, about 1% P, about 0.3% EPA, about 0.2% DHA, about 4% linoleic acid, about 1% total n-3 fatty acids, about 4% total n-6 fatty acids, about 0.1% taurine, about 300 ppm carnitine, about 1% methionine, about 0.4% cystine, about 95% manganese, about 800 IU/kg of vitamin E, about 170 ppm of vitamin C, and

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about 5000 ppm choline, and the regulation of the gene is at least one of: (a) down-regulation of genes associated with arthritis or inflammation, wherein the genes associated with arthritis or inflammation comprise interleukin 1-beta, fibronectin, v-fos FBJ murine osteosarcoma viral oncogene homolog, dual specificity phosphatase 1, lactotransferrin, compliment component 5a receptor, xanthine dehydrogenase, and secreted phosphoprotein 1; (b) down-regulation of genes associated with DNA fragmentation, wherein the genes associated with DNA fragmentation comprise interleukin 1-beta, fibronectin, neuroregulin 1, dual specificity phosphatase 1, and cell death-inducing DFFA-like effector a; (c) up-regulation of genes associated with DNA processing control, wherein the genes associated with DNA processing control comprise budding uninhibited by benzimidazoles 1 homolog, topoisomerase II alpha, kinetochore associated 2, claspin homolog, cyclin B1, cyclin B2, human chromosome condensation protein G, leucine zipper protein 5, kinesin family member 23, and flap structure specific endonuclease I; (d) down-regulation of genes associated with cancer incidence, wherein the genes associated with cancer incidence comprise interleukin 1-beta, fibronectin, v-fos FBJ murine osteosarcoma viral oncogene homolog, dual specificity phosphatase 1, compliment component 5a receptor, xanthine dehydrogenase, cellular repressor of E1A-stimulated genes 1, chondroitin sulfate proteoglycan 2, mycrocytic anemia transcobalamin II, clade B member 10 serpin peptidase inhibitor, adenomatosis polyposis coli down-regulated 1, six membrane epithelial antigen of the prostate 2, and alanyl aminopeptidase; (e) up-regulation of genes associated with tumor suppression, wherein the genes associated with tumor suppression comprise cancer susceptibility candidate 5, paternally expressed 3, topoisomerase II alpha, tubulin alpha 6, cyclin B1, cell division cycle associated 1, cell division cycle 2, ribonucleotide reductase M2 polypeptide, breast cancer anti-estrogen resistance 3, tumor necrosis factor receptor superfamily member 17, and protein kinase C iota; (f) down-regulation of genes associated with cellular compromise, wherein the genes associated with cellular compromise comprise interleukin 1-beta, fibronectin, neuroregulin 1, v-fos FBJ murine osteosarcoma viral oncogene homolog, dual specificity phosphatase 1, compliment component 5a receptor, xanthine dehydrogenase, and macrophage receptor with collagenous structure; (g) up-regulation of genes associated with cellular integrity, wherein the genes associated with cellular integrity comprise cyclin B1, cell division cycle 2, hemoglobin epsilon 1, and protein kinase C iota; (h) down-regulation of genes

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associated with disruption of cellular assembly, wherein the genes associated with disruption of cellular assembly comprise interleukin 1-beta, fibronectin, neuroregulin 1, chondroitin sulfate proteoglycan 2, muscarinic 3 cholinergic receptor, xanthine dehydrogenase, and macrophage receptor with collagenous structure; (i) up-regulation of genes associated with

5 cellular organization, wherein the genes associated with cellular organization comprise cyclin B1, cell division cycle 2, leucine zipper protein 5, protein kinase C iota, kinesin family member 23, budding uninhibited by benzimidazoles 1 homolog, cyclin B2, topoisomerase II alpha, kinetochore associated 2, cell division cycle associated 1, human chromosome condensation protein G, kinesin family member 11, A kinase (PRKA) anchor protein 2, and

10 thymopioten; (j) down-regulation of genes associated with disruption of cell cycle control, wherein the genes associated with disruption of cell cycle control comprise interleukin 1-beta, fibronectin, neuroregulin 1, v-fos FBJ murine osteosarcoma viral oncogene homolog, muscarinic 3 cholinergic receptor, dual specificity phosphatase 1, and cellular repressor of E1A-stimulated genes 1; (k) up-regulation of genes associated with cell cycle regulation,

15 wherein the genes associated with cell cycle regulation comprise cyclin B1, cell division cycle 2, leucine zipper protein 5, 350/400ka mitosis centromere protein F, breast cancer anti-estrogen resistance 3, budding uninhibited by benzimidazoles 1 homolog, cyclin B2, topoisomerase II alpha, kinetochore associated 2, cell division cycle associated 1, human chromosome condensation protein G, kinesin family member 11, and thymopioten; (l) down-

20 regulation of genes associated with immune activation, wherein the genes associated with immune activation comprise lymphocyte antigen 6 complex, fibrinogen-like 2, CD1A antigen, complement factor D (adipsin), CD163 or macrophage-associated antigen, toll-like receptor 2, renal amyloidosis lysozyme, transcription elongation factor A (SII) 3, fibronectin 1, macrophage receptor with collagenous structure, bactericidal/permeability-increasing protein,

25 T cell receptor alpha locus, and immunoglobulin heavy constant alpha 2; (m) down-regulation of genes associated with lipid metabolism, wherein the genes associated with lipid metabolism comprise toll-like receptor 2, cytochrome P450 family 1 subfamily B, polypeptide 1, colony stimulating factor 1 receptor, phospholipase C beta 1, fibronectin 1, neuregulin 1, macrophage receptor with collagenous structure, and CD1a molecule; (n) down-regulation of genes

30 associated with cardiovascular development, wherein the genes associated with cardiovascular development comprise bactericidal/permeability-increasing protein, fibronectin 1,

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- neuregulin 1, xanthine dehydrogenase, annexin A6, colony stimulating factor 1 receptor, and alanyl (membrane) aminopeptidase; (o) down-regulation of genes associated with skeletal muscular disorders, wherein the genes associated with skeletal muscular disorders comprise toll-like receptor 2, colony stimulating factor 1 receptor, fibronectin 1, and neuregulin 1;
- 5 (p) up-regulation of genes associated with muscle contraction/function, wherein the genes associated with muscle contraction/function comprise exostoses, microfibrillar associated protein 5, myosin light polypeptide kinase, dystroglycan 1, and moloney leukemia virus 10-like 1; (q) down-regulation of genes associated with cellular compromise, wherein the genes associated with cellular compromise comprise neuroregulin 1, fibronectin 1,
- 10 macrophage receptor with collagenous structure, xanthine dehydrogenase, bactericidal permeability increasing protein, and phospholipase C beta 1; and (r) down-regulation of genes associated with cancer, wherein the genes associated with cancer comprise fibronectin 1, xanthine dehydrogenase, neuregulin 1, colony stimulating factor 1 receptor, alanyl aminopeptidase, and S100 calcium binding protein.

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[0011] In a further aspects, the invention relates to the use of any of the formulae of the present invention in the manufacture of a composition to modify gene expression in an animal as described herein.

[0012] Other features and advantages of the present invention will be understood by reference to the detailed description of the examples that follow.

DETAILED DESCRIPTION OF THE INVENTION

[0013] It is contemplated that the invention described herein is not limited to the particular methodology, protocols, and reagents described as these may vary. It is also to be understood that the terminology used herein is for the purpose of describing particular embodiments only, and is not intended to limit the scope of the present invention in any way.

[0014] Unless defined otherwise, all technical and scientific terms used herein have the same meanings as commonly understood by one of ordinary skill in the art to which this invention belongs. In the event of a conflict in a definition in the present disclosure and that of a cited reference, the present disclosure controls.

[0015] As used herein and in the appended claims, the singular forms "a", "an", and "the" include plural reference unless the context clearly dictates otherwise.

[0016] The present invention relates to any animal; preferably a mammal, more preferably a companion animal. The term "companion animal" refers to any animal that lives in close association with humans and includes, but is not limited to, canines and felines of any breed. It is contemplated herein,

however, that any animal whose diet may be controlled by humans may benefit from feeding the formulations disclosed herein. These animals may include, for example, domesticated farm animals (e.g., cattle, horses, swine, etc.) as well as undomesticated animals held in captivity, e.g., in zoological parks and the like. Preferred animals include canines, e.g., dogs, including growing dogs, e.g., puppies.

[0017] “Beneficial modification in the expression of genes” as used herein includes, e.g., down regulation of genes expressing proteins associated with disease states and/or up regulation of genes expressing proteins which have a beneficial or healthful effect as compared to appropriate controls, as may be determined using conventional methods, e.g., by microarray (e.g., Affymetrix gene chip) techniques familiar to one of skill in the art. Further, one of skill in the art is familiar with the known associations between diseases and specific genes as those listed in the tables provided hereinbelow such that it is understood whether an increase or decrease in expression of a particular gene is desirable.

[0018] The “growth” life stage of an animal refers to the period from birth or weaning (approximately 8 weeks of age) to about 1 year of age or beyond, depending on the species and breed of the animal.

[0019] As used herein, the term “puppy” refers to an immature canine, typically between the ages of birth and 12 months.

[0020] “Essential amino acids” as used herein refers to those amino acids that cannot be synthesized *de novo* by an organism and thus must be supplied in the diet. It is understood by one of skill in the art that the essential amino acids varies from species to species, depending upon the organism’s metabolism. For example, it is generally understood that the essential amino

acids for dogs and cats (and humans), are phenylalanine, leucine, methionine, lysine, isoleucine, valine, threonine, tryptophan, histidine and arginine.

[0021] As understood by one of skill in the art, a “limiting amino acid” refers to an amino acid which if present in insufficient quantities in a diet, results in the limitation in usefulness of other essential amino acids, even if the other essential amino acids are present in otherwise large enough quantities.

Lysine is the limiting essential amino acid in the compositions disclosed herein. Thus, the remaining essential amino acids are quantitatively formulated or “balanced” in relationship to the amount of lysine determined critical to affect the desired biological result. As used herein, “balanced amino acids” refers to the relationship of the essential amino acid lysine to energy to assure optimal animal growth and development.

[0022] “Essential nutrients” as used herein refers to nutrients required for normal body functioning that cannot be synthesized by the body. Categories of essential nutrient include vitamin dietary minerals, fatty acid, and amino acid. It is understood by one of skill in the art that the nutrients deemed essential varies from species to species, depending upon the organism’s metabolism. For example, essential nutrients for dogs and cats include Vitamins A, D, E, K, B1, B6 B12, riboflavin, niacin, pantothenic acid, folic acid, calcium, phosphorous, magnesium, sodium, potassium, chlorine, iron, copper, zinc, manganese, selenium and iodine. Choline, generally regarded as a B complex vitamin, may be included among the semi-essential nutrients. In addition, taurine, while technically not an amino acid but a derivative of cysteine, is an essential nutrient for cats.

[0023] Carnitine, also known as L-carnitine, (levocarnitine) is a quaternary ammonium compound synthesized from the amino acids lysine and

methionine and is responsible for the transport of fatty acids from the cytosol into the mitochondria.

[0024] Without being limited to any theories or particular modes of action, the present invention is based on the surprising discovery that the addition of certain ingredients to pet food compositions and administration of these compositions to animals can enhance the development of a growing animal. For example, data indicate that animals fed the compositions of the present invention (or those whose dams were fed the compositions during gestation and prior to weaning but continued throughout growth of their litters), demonstrate enhanced neurologic development, bone and joint health, immune function, and have overall healthier body composition. Interestingly, microarray data also indicate a differential change in gene expression in these animals compared to controls which is generally reflective of a beneficial modification (i.e., up or down regulation) in the expression of many genes associated with biological processes including those involved in growth and development. Thus, in one aspect, the invention relates to compositions and methods to enhance the development of a growing animal as described in detail herein.

[0025] As contemplated herein, the compositions of the present invention comprise nutritionally complete and balanced animal feed compositions. Such compositions include, among other nutrients and ingredients, recommended healthful amounts of protein, carbohydrate and fat. "Nutritionally complete and balanced animal feed compositions", as well as nutrients and ingredients suitable for animal feed compositions, and recommended amounts thereof, are familiar to one of skill in the art (see, for example, National Research Council, 2006 Nutritional Requirements for Dogs and Cats, National Academy Press, Washington D.C. or the Official

Publication of the Association of American Feed Control Officials, Inc. Nutrient Requirements for Dogs and Cats 2006).

[0026] It is contemplated herein that the compositions disclosed herein may also comprise antioxidants, additives, stabilizers, thickeners, flavorants, palatability enhancers and colorants in amounts and combinations familiar to one of skill in the art. "Antioxidants" refers to a substance that is capable of reacting with or decreasing the production of free radicals and neutralizing them. Examples include, but are not limited to, beta-carotene, selenium, coenzyme Q10 (ubiquinone), lutein, tocotrienols, soy isoflavones, S-adenosylmethionine, glutathione, taurine, N-acetylcysteine, vitamin E, vitamin D, vitamin C, flavanoids, anthocyanindins, and lipoic acid.

[0027] While foods of any consistency or moisture content are contemplated, preferably the compositions of the present invention may be, for example, a wet, semi-dry, or dry animal food composition. "Wet" food refers to food which is sold in cans or foil bags and has a moisture content of about 70 to about a 90%. "Dry" food refers to compositions with about 5 to about 15% moisture content and is often manufactured in the form of small bits or kibbles. Semi-dry compositions refers to food which has a moisture content greater than dry foods, but less than wet foods. Also contemplated herein are compositions of intermediate moisture consistency and those that may comprise components of various consistency as well as components that may include more than one consistency, for example, soft, chewy meat-like particles as well as kibble having an outer cereal component and an inner cream component as described in, e.g., US Patent 6,517,877.

[0028] Various processes for manufacturing and packaging the compositions of the present invention may be employed and are familiar to one of skill in the art.

[0029] It is also contemplated herein that the methods of the present invention include methods to cause a beneficial modification in gene expression in an animal, specifically, down regulation in expression of a gene or genes associated with an undesirable biological condition or pathway or disease state and/or up regulation in expression of a gene or genes associated with a desired biological condition or pathway or which may have a positive or preventive effect on a disease state, as the case may be, for any one or more biological conditions, pathways or disease states and genes described in Tables 14-27, comprising administering an effective amount of a composition of the present invention to the animal, either directly to the animal or to the dam while the animal is *in utero*. Indeed, as discussed in Examples 7 and 8 here-in-below, by effecting the animal at the genomic level, the administration of the compositions of the present invention, either directly to the animal or to the dam while the animal is *in utero*, may have beneficial, even prophylactic, health effects on the animal by effecting the expression of any one or more genes listed in Tables 14-27. For example, the administration of an effective amount of a composition of the present invention to an animal may enhance bone and joint health in the animal by causing the down regulation in genes associated with cartilage and joint damage associated with arthritis, e.g. interleukin 1-beta, fibronectin, lactoferrin, etc. (see Table 14).

[0030] It is contemplated herein that the compositions of the present invention may be administered to an animal alone as a complete nutritionally balanced diet, or in conjunction with dietary supplements, vitamins and/or other nutritionally beneficial agents familiar to one of skill in the art, as part of an overall wellness program for the animal. Compositions of the invention may also be useful as a veterinary therapeutic product. As such, the

compositions may optionally contain a carrier, diluent, or an excipient, the suitability of which for the intended use being familiar to one of skill in the art.

[0031] It is also contemplated that, in addition to administering the compositions disclosed herein directly to a growing animal, e.g., to a growing puppy or kitten, the compositions may be administered to the dam of the animal while the animal is still *in utero* or while the animal is a nursling.

[0032] The following examples further illustrate the present invention and are not intended to limit the invention. As used throughout, ranges are used as shorthand for describing each and every value that is within the range. Any value within the range can be selected as the terminus of the range. It is understood that when formulations are described, they may be described in terms of their ingredients, as is common in the art, notwithstanding that these ingredients may react with one another in the actual formulation as it is made, stored and used, and such products are intended to be covered by the formulations described.

[0033] The following examples further describe and demonstrate illustrative embodiments within the scope of the present invention. The examples are given solely for illustration and are not to be construed as limitations of this invention as many variations are possible without departing from the spirit and scope thereof. Various modifications of the invention in addition to those shown and described herein should be apparent to those skilled in the art and are intended to fall within the appended claims.

[0034] Except to the extent stated otherwise, all percentages used in this specification are weight percentages on a dry matter basis. The phrase "dry matter basis" means the component concentration in the composition after any moisture in the composition is removed.

EXAMPLES

Example 1

Canine Genetic Effects: Conception to one year of age

[0035] In order to characterize the genotypic effects of feeding a composition formulated to enhance the development of a growing animal, an experiment is performed to identify changes in gene expression in puppies at one year of age from dams fed the compositions disclosed in Table 1.

Table 1. Average of analyzed nutrient profiles of foods utilized in the study

Nutrients, 100% Dry Matter Basis	Composition H	Composition P
Moisture, %	6.09	7.44
Crude Protein, %	29.4	27.9
Fat, %	17.6	14.2
Ca, %	1.43	1.46
P, %	1.2	0.96
EPA, %	0.31	<0.01
DHA %	0.19	0.01
Linoleic Acid, %	3.8	2.14
Total n-3 fatty acids, %	1.4	0.19
Total n-6 fatty acids, %	3.7	2.6
Taurine, %	0.14	0.08
Carnitine, ppm	312.1	No analysis
Methionine, %	1.3	0.52
Cystine, %	0.4	0.39
Manganese, %	94.5	64
Vitamin E, IU/kg	816	43.8
Vitamin C, ppm	168.6	<10
Choline, ppm	4876	No analysis

[0036] Composition P is a commercially available dog food. Composition H is an experimental dog food composition.

[0037] Dams are fed either Composition H or Composition P for at least 10 days prior to conception. Dams are maintained in group lodging until confirmed pregnant via palpation, and are then moved to maternity lodging. Puppies from dams are kept on the same foods fed to the dams until one year

of age (including pre-weaning and post-weaning period). Blood samples are then taken from the puppies and mRNA isolated according to conventional methods. Microarray assays are performed using the Affymetrix Canine- 2 gene chip according to conventional methods.

[0038] The tables below show the genes grouped by function and the direction of expression, wherein up regulation in Composition H ("H") vs Composition P ("P") demonstrates increased gene expression in Composition H fed puppies compared to Composition P fed puppies. Similarly, down regulation of a gene in the Composition H fed puppies vs Composition P fed puppies represents decreased gene expression in puppies fed Composition H relative to those fed Composition P.

Table 2. Genes associated with arthritis/inflammation relevant to bone and joints

Annotation	Probe	Direction of expression H vs P
Interleukin 1-beta	CfaAffx.11741.1.S1_s_at	down
Fibronectin	Cfa.3707.1.A1_s_at	down
v-fos FBJ murine osteosarcoma viral oncogene homolog	Cfa.9039.1.A1_at	down
dual specificity phosphatase 1	CfaAffx.25714.1.S1_s_at	down
Lactotransferrin	CfaAffx.21286.1.S1_s_at	down
compliment component 5a receptor	CfaAffx.7180.1.S1_s_at	down
xanthine dehydrogenase	CfaAffx.9452.1.S1_s_at	down
Secreted phosphoprotein 1	CfaAffx.15042.1.S1_s_at	down

Table 3. Genes associated with DNA replication and repair

Annotation	Probe	Direction of expression H vs P
DNA fragmentation		
Interleukin 1-beta	CfaAffx.11741.1.S1_s_at	down
Fibronectin	Cfa.3707.1.A1_s_at	down
Neuroregulin 1	CfaAffx.10523.1.S1_s_at	down
Dual specificity phosphatase 1	CfaAffx.25714.1.S1_s_at	down
Cell death-inducing DFFA-like effector a	CfaAffx.28752.1.S1_at	down
Proper DNA processing control		
Budding uninhibited by benzimidazoles 1 homolog	Cfa.1559.1.A1_at	up
Topoisomerase II alpha	Cfa.18946.1.S1_s_at	up
Kinetochore associated 2	Cfa.3066.1.S1_s_at	up
Claspin homolog	CfaAffx.6235.1.S1_s_at	up
Cyclin B1	CfaAffx.12419.1.S1_s_at	up
Cyclin B2	Cfa.11939.1.A1_s_at	up
Human chromosome condensation protein G	CfaAffx.25509.1.S1_at	up
Leucine zipper protein 5	CfaAffx.8781.1.S1_s_at	up
Kinesin family member 23	Cfa.15293.1.A1_at	up
Flap structure specific endonuclease 1	Cfa.1854.1.A1_at	up

Table 4. Genes associated with cancer

Annotation	Probe	Direction of expression H vs P
Interleukin 1-beta	CfaAffx.11741.1.S1_s_at	down
Fibronectin	Cfa.3707.1.A1_s_at	down
v-fos FBJ murine osteosarcoma viral oncogene homolog	Cfa.9039.1.A1_at	down
dual specificity phosphatase 1	CfaAffx.25714.1.S1_s_at	down
compliment component 5a receptor	CfaAffx.7180.1.S1_s_at	down
xanthine dehydrogenase	CfaAffx.9452.1.S1_s_at	down
cellular repressor of E1A-stimulated genes 1	Cfa.10558.3.A1_at	down
chondroitin sulfate proteoglycan 2	CfaAffx.13597.1.S1_s_at	down
transcobalamin II, mycrocytic anemia	CfaAffx.19852.1.S1_s_at	down
serpin peptidase inhibitor, clade B member 10	CfaAffx.1043.1.S1_s_at	down
adenomatosis polyposis coli down-regulated 1	CfaAffx.28621.1.S1_at	down
six membrane epithelial antigen of the prostate 2	Cfa.1933.1.S1_at	down
alanyl aminopeptidase	Cfa.3774.1.A1_s_at	down
Cancer susceptibility candidate 5	Cfa.1126.1.S1_at	up
Paternally expressed 3	Cfa.4482.1.S1_at	up
Topoisomerase II alpha	Cfa.18946.1.S1_s_at	up
Tubulin alpha 6	CfaAffx.13667.1.S1_at	up
Cyclin B1	CfaAffx.12419.1.S1_s_at	up
Cell division cycle associated 1	CfaAffx.20529.1.S1_at	up
Cell division cycle 2	CfaAffx.20006.1.S1_at	up
Ribonucleotide reductase M2 polypeptide	CfaAffx.6059.1.S1_at	up
Breast cancer anti-estrogen resistance 3	Cfa.651.1.S1_at	up
Tumor necrosis factor receptor superfamily member 17	CfaAffx.28790.1.S1_at	up
Protein kinase C, iota	CfaAffx.22822.1.S1_s_at	up

Table 5. Genes associated with cell compromise

Annotation	Probe	Direction of expression H vs P
Interleukin 1-beta	CfaAffx.11741.1.S1_s_at	down
Fibronectin	Cfa.3707.1.A1_s_at	down
Neuroregulin 1	CfaAffx.10523.1.S1_s_at	down
v-fos FBJ murine osteosarcoma viral oncogene homolog	Cfa.9039.1.A1_at	down
dual specificity phosphatase 1	CfaAffx.25714.1.S1_s_at	down
compliment component 5a receptor	CfaAffx.7180.1.S1_s_at	down

xanthine dehydrogenase	CfaAffx.9452.1.S1_s_at	down
Macrophage receptor with collagenous structure	Cfa.15713.1.A1_s_at	down
Cyclin B1	CfaAffx.12419.1.S1_s_at	up
Cell division cycle 2	CfaAffx.20006.1.S1_at	up
Hemoglobin epsilon 1	CfaAffx.10240.1.S1_s_at	up
Protein kinase C, iota	CfaAffx.22822.1.S1_s_at	up

Table 6. Genes associated with cellular assembly

Annotation	Probe	Direction of expression H vs P
Interleukin 1-beta	CfaAffx.11741.1.S1_s_at	down
Fibronectin	Cfa.3707.1.A1_s_at	down
Neuroregulin 1	CfaAffx.10523.1.S1_s_at	down
chondroitin sulfate proteoglycan 2	CfaAffx.13597.1.S1_s_at	down
cholinergic receptor, muscarinic 3	Cfa.8414.1.A1_s_at	down
xanthine dehydrogenase	CfaAffx.9452.1.S1_s_at	down
Macrophage receptor with collagenous structure	Cfa.15713.1.A1_s_at	down
Cyclin B1	CfaAffx.12419.1.S1_s_at	up
Cell division cycle 2	CfaAffx.20006.1.S1_at	up
Leucine zipper protein 5	CfaAffx.8781.1.S1_s_at	up
Protein kinase C, iota	CfaAffx.22822.1.S1_s_at	up
Kinesin family member 23	Cfa.15293.1.A1_at	up
Budding uninhibited by benzimidazoles 1 homolog	Cfa.1559.1.A1_at	up
Cyclin B2	Cfa.11939.1.A1_s_at	up
Topoisomerase II alpha	Cfa.18946.1.S1_s_at	up
Kinetochore associated 2	Cfa.3066.1.S1_s_at	up
Cell division cycle associated 1	CfaAffx.20529.1.S1_at	up
Human chromosome condensation protein G	CfaAffx.25509.1.S1_at	up
Kinesin family member 11	CfaAffx.12118.1.S1_s_at	up
A kinase (PRKA) anchor protein 2	Cfa.10574.1.A1_at	up
thymopioten	Cfa.18367.2.S1_s_at	up

Table 7. Genes associated with cell cycle regulation

Annotation	Probe	Direction of expression H vs P
Interleukin 1-beta	CfaAffx.11741.1.S1_s_at	down
Fibronectin	Cfa.3707.1.A1_s_at	down
Neuroregulin 1	CfaAffx.10523.1.S1_s_at	down
v-fos FBJ murine osteosarcoma viral oncogene homolog	Cfa.9039.1.A1_at	down

cholinergic receptor, muscarinic 3	Cfa.8414.1.A1_s_at	down
dual specificity phosphatase 1	CfaAffx.25714.1.S1_s_at	down
cellular repressor of E1A-stimulated genes 1	Cfa.10558.3.A1_at	down
Cyclin B1	CfaAffx.12419.1.S1_s_at	up
Cell division cycle 2	CfaAffx.20006.1.S1_at	up
Leucine zipper protein 5	CfaAffx.8781.1.S1_s_at	up
centromere protein F, 350/400ka (mitosin)	CfaAffx.19534.1.S1_at	up
Breast cancer anti-estrogen resistance 3	Cfa.651.1.S1_at	up
Budding uninhibited by benzimidazoles 1 homolog	Cfa.1559.1.A1_at	up
Cyclin B2	Cfa.11939.1.A1_s_at	up
Topoisomerase II alpha	Cfa.18946.1.S1_s_at	up
Kinetochore associated 2	Cfa.3066.1.S1_s_at	up
Cell division cycle associated 1	CfaAffx.20529.1.S1_at	up
Human chromosome condensation protein G	CfaAffx.25509.1.S1_at	up
Kinesin family member 11	CfaAffx.12118.1.S1_s_at	up
thymopioten	Cfa.18367.2.S1_s_at	up

Table 8. Genes associated with various functions

Annotation	Probe	Direction of expression H vs P
calcyphosine-like	Cfa.21214.1.S1_s_at	down
ELKS/RAB6 interacting/CAST family member 2	Cfa.21214.1.S1_at	down
glutamyl tRNA synthetase 2	Cfa.2057.1.A1_at	down
Coiled-coil containing domain 3	Cfa.9084.1.A1_at	down
chromosome 16 clone RP11-26P10	Cfa.3834.1.S1_at	down
SAM domain- and HD domain-containing protein 1	CfaAffx.13689.1.S1_at	down
AJ420591	Cfa.15196.1.A1_at	down
family with sequence similarity 46, member A	CfaAffx.5194.1.S1_at	down
CD1a antigen	Cfa.18390.1.S1_s_at	down
epidermal dendritic cell marker (CD1a)	CfaAffx.17796.1.S1_at	down
ANKYRIN MOTIF	CfaAffx.6104.1.S1_s_at	down
RP11-549H3 on chromosome X	Cfa.9597.1.S1_at	down
La ribonucleoprotein domain family, member 1	Cfa.14608.1.A1_at	down
adenomatosis polyposis coli down-regulated 1	CfaAffx.28621.1.S1_at	down
Chromosome 3 open reading frame 21	CfaAffx.20763.1.S1_at	down
EGF-like module containing, mucin-like, hormone receptor-like 1	CfaAffx.28381.1.S1_s_at	down
acyltransferase like 1	CfaAffx.14833.1.S1_at	down
RP11-95J15 from 7	Cfa.6138.1.A1_at	down
CD8 antigen, beta polypeptide 2	Cfa.21011.1.S1_at	down
multimerin 1	CfaAffx.15515.1.S1_s_at	down
none	Cfa.12500.1.A1_at	down
Hypothetical protein FLJ21062	Cfa.1933.2.A1_at	down
normal mucosa of esophagus specific 1	Cfa.11815.1.A1_at	down
regulator of G-protein signalling 7	CfaAffx.24021.1.S1_s_at	down
Islet cell auto-antigen 1	Cfa.11691.1.A1_at	down
normal mucosa of esophagus specific 1 (NMES1), transcript variant 2	CfaAffx.851.1.S1_s_at	down
none	Cfa.5989.1.A1_s_at	down
eukaryotic translation initiation factor 2, subunit 3 gamma	Cfa.9060.1.A1_at	down
Serine/threonine kinase 17A	CfaAffx.21987.1.S1_at	down
RP11-542F9 on chromosome 6	CfaAffx.21067.1.S1_at	up
chromosome 20 open reading frame 172	CfaAffx.13625.1.S1_at	up
RP11-111I12 on chromosome 1	CfaAffx.16431.1.S1_at	up
BAT2 domain containing 1	Cfa.15064.1.S1_at	up
Rho GTPase activating protein 11A	CfaAffx.13079.1.S1_s_at	up

polo-like kinase 4	CfaAffx.6762.1.S1_s_at	up
NADH dehydrogenase (ubiquinone) flavoprotein 3	CfaAffx.16432.1.S1_s_at	up
3 BAC RP11-223L18	Cfa.3066.1.S1_at	up
par-3 partitioning defective 3 homolog B	Cfa.12402.1.S1_a_at	up
Hypothetical protein MGC24039	Cfa.19506.1.S1_at	up

TABLE 8 (cont.)

Annotation	Probe	H vs P
immunoglobulin mu heavy chain	CfaAffx.539.1.S1_x_at	up
origin recognition complex, subunit 1-like	Cfa.8552.1.A1_s_at	up
RP11-653G16 from 4	CfaAffx.15773.1.S1_at	up
abnormal spindle-like microcephaly-associated protein	Cfa.103.1.A1_s_at	up
Solute carrier family 22	Cfa.10558.2.S1_at	up
BCSynL32 immunoglobulin lambda light chain	CfaAffx.346.1.S1_at	up
Purinergic receptor P2Y, G-protein coupled 10	Cfa.1521.1.S1_at	up
DEP domain containing 1	CfaAffx.31279.1.S1_at	up
chromosome 17, clone RP11-160L11	Cfa.16355.1.S1_at	up
Hypothetical protein LOC644115	Cfa.15220.2.A1_at	up
isolate MRPS17P1 mitochondrial ribosomal protein S17	Cfa.12402.1.S1_s_at	up
KIAA0101 (L5)	CfaAffx.26141.1.S1_s_at	up
chromosome 13 open reading frame 3	CfaAffx.11656.1.S1_at	up
SHC SH2-domain binding protein 1	CfaAffx.6499.1.S1_at	up
CDC20 cell division cycle 20 homolog	CfaAffx.8850.1.S1_s_at	up
Chromosome 14 open reading frame 32	CfaAffx.23135.1.S1_s_at	up
3-oxoacyl-ACP synthase, mitochondrial	Cfa.10049.1.A1_s_at	up
MAD2 mitotic arrest deficient-like 1	CfaAffx.25205.1.S1_s_at	up
thioredoxin domain containing 5	Cfa.10753.1.A1_at	up
Apla tubulin	Cfa.6991.1.A1_at	up
none	CfaAffx.32.1.S1_at	up
Melanin-concentrating hormone receptor 1	Cfa.6032.1.A1_at	up
Aquaporin 7	Cfa.21549.1.S1_s_at	up
cathepsin E	CfaAffx.15966.1.S1_at	up
EF hand domain (C-terminal) containing 2	Cfa.1885.1.A1_at	up
bone morphogenetic protein 6	Cfa.15702.1.S1_at	up
hemoglobin, alpha 2	Cfa.3973.2.A1_s_at	up
SH3-domain GRB2-like 3	Cfa.10644.1.A1_at	up
Acidic nuclear phosphoprotein 32 family, member A	Cfa.31.1.S1_s_at	up
none	CfaAffx.20953.1.S1_x_at	up
Rh family, B glycoprotein	Cfa.13613.1.A1_at	up
putative protein STRF8	CfaAffx.15063.1.S1_s_at	up
fatty acid desaturase 1	Cfa.13257.1.A1_at	up
Ligand dependent nuclear receptor corepressor-like	Cfa.1476.1.A1_at	up
F-box only protein 2	CfaAffx.9335.1.S1_at	up
Ring finger protein 152	Cfa.15414.1.A1_at	up
immunoglobulin lambda constant 1	CfaAffx.22878.1.S1_at	up

G-protein coupled receptor 158	Cfa.9162.1.A1_at	up
Hypothetical protein MGC42105	CfaAffx.28387.1.S1_at	up

[0039] Gene expression profiles from puppies fed Composition H and P are obtained and compared. Results indicate that 143 genes are differentially expressed between the study groups and, in general, their functions may be associated with cell assembly, cell cycle regulation, DNA replication and repair, cell compromise, arthritis and cancer. At a minimum of 1.5 fold change, 143 genes are differentially expressed in the two groups.

[0040] Genes associated with arthritis/inflammation are down-regulated in puppies fed Composition H compared to puppies fed Composition P, which suggests reduced cartilage/joint damage in Prototype pups. A separate study performed also indicates that levels of bone alkaline phosphatase are lower in puppies fed composition H (data not shown). See Table 2.

[0041] Genes associated with DNA fragmentation are down-regulated and genes associated with DNA processing control are up-regulated in puppies fed Composition H compared to puppies fed Composition P, which suggests reduced DNA damage and improved DNA protection in Prototype pups (also supported by elevated Vitamin E levels in blood, data not shown). See Table 3.

[0042] Genes associated with cancer incidence are down-regulated and genes associated with tumor suppression are up-regulated in puppies fed Composition H compared to puppies fed Composition P, which suggests the possibility of reduced cancer susceptibility in these Prototype animals; see Table 4.

[0043] Genes associated with cellular compromise are down-regulated and genes associated with cellular integrity are up-regulated in puppies fed Composition H compared to puppies fed Composition P, which suggests the possibility of reduced cell damage and enhanced cellular protection in these

Prototype animals (also supported by elevated Vitamin E levels in blood, data not shown). See Table 5.

[0044] Genes associated with disruption of cellular assembly are down-regulated and genes associated with proper cellular organization are up-regulated in puppies fed Composition H compared to puppies fed Composition P, which suggests improved cellular organization and function in Prototype pups. See Table 6.

[0045] Genes associated with proper cell cycle regulation are up-regulated and genes associated with disruption of cell cycle control are down-regulated in puppies fed Composition H compared to puppies fed Composition P, which suggests proper control of cell cycle progression and cell survival. See Table 7.

[0046] Thus, it is contemplated herein that the nutritional benefits of the compositions of the present invention as described herein may involve modification in gene expression which results in the enhancement of the development of a growing animal. In addition, feeding proper nutrients during early development may have a prophylactic effect and influence disease processes later in life, i.e., lessen the chance of disease in the animal, as the expression of genes associated with common diseases and disorders may be influenced due to the given nutritional compositions of the foods provided maternally and during early development.

Example 2

[0047] An experiment is performed to determine the effects on gene expression in puppies fed Composition H vs. Composition P from weaning until one year of age.

[0048] Dams utilized in this study are fed Composition P prior to, and during pregnancy. Following weaning, puppies are divided into two groups and

provided with either Composition H or maintained on Composition P until one year old. Blood samples are then taken from the puppies and mRNA isolated according to conventional methods. Microarray assays are performed using the Affymetrix Canine- 2 gene chip according to conventional methods.

[0049] Results indicate that 99 genes are differentially expressed between the two study groups, and are presented in Tables 9 - 15. Of the genes identified, many are related to biological functions or pathways such as, e.g., immune activation, lipid metabolism, cardiovascular development, skeletal and muscular disorders, contraction and function and cell compromise and cancer.

Table. 9 Genes associated with immune activation

Annotation	Probe	Direction of expression
		H v. P
lymphocyte antigen 6 complex	CfaAffx.1839.1.S1_at	down
Fc fragment of IgG, receptor, transporter, alpha	Cfa.17806.2.S1_s_at	up
fibrinogen-like 2	Cfa.10303.1.S1_at	down
CD1A antigen	Cfa.18390.1.S1_s_at	down
complement factor D (adipsin)	Cfa.21381.1.S1_s_at	down
CD163 antigen; macrophage-associated antigen	Cfa.9647.1.A1_at	down
toll-like receptor 2	CfaAffx.13248.1.S1_s_at	down
lysozyme (renal amyloidosis)	CfaAffx.1598.1.S1_s_at	down
transcription elongation factor A (SII), 3	Cfa.12580.1.S1_s_at	down
fibronectin 1	Cfa.3707.2.S1_at	down
macrophage receptor with collagenous structure	Cfa.15713.1.A1_s_at	down
bactericidal/permeability-increasing protein	CfaAffx.14056.1.S1_s_at	down
T cell receptor alpha locus	Cfa.10333.1.A1_at	down
immunoglobulin heavy constant alpha 2	Cfa.4556.3.A1_a_at	down
nuclear factor I/B	Cfa.4487.1.S1_at	up

Table. 10 Genes associated with Cancer formation

Annotation	Probe	Direction of expression
		P vs C
fibronectin 1	Cfa.3707.3.S1_s_at	down
xanthine dehydrogenase	CfaAffx.9452.1.S1_s_at	down
neuregulin 1	CfaAffx.10523.1.S1_s_at	down
colony stimulating factor 1 receptor	CfaAffx.27899.1.S1_at	down
alanyl aminopeptidase	Cfa.3774.1.A1_s_at	down
S100 calcium binding protein	CfaAffx.22128.1.S1_at	down
dystroglycan 1	CfaAffx.17467.1.S1_at	up

Table. 11 Genes associated with Lipid metabolism

Annotation	Probe	Direction of expression
		H v. P
Toll-like receptor 2	CfaAffx.13248.1.S1_s_at	Down
cytochrome P450, family 1, subfamily B, polypeptide 1	CfaAffx.10229.1.S1_at	Down
colony stimulating factor 1 receptor	CfaAffx.27899.1.S1_at	Down
phospholipase C, beta 1	Cfa.10853.1.A1_at	Down
fibronectin 1	Cfa.3707.3.S1_s_at	Down
neuregulin 1	CfaAffx.10523.1.S1_s_at	Down
macrophage receptor with collagenous structure	Cfa.15713.1.A1_at	Down
CD1a molecule	Cfa.18390.1.S1_s_at	Down

Table. 12 Genes associated with Cardiovascular development

Annotation	Probe	Direction of expression
		H v. P
bactericidal/permeability-increasing protein	CfaAffx.14056.1.S1_s_at	Down
fibronectin 1	Cfa.3707.3.S1_s_at	Down
neuregulin 1	CfaAffx.10523.1.S1_s_at	Down
xanthine dehydrogenase	CfaAffx.9452.1.S1_s_at	Down
annexin A6	CfaAffx.27471.1.S1_s_at	Down
colony stimulating factor 1 receptor	CfaAffx.27899.1.S1_at	Down
alanyl (membrane) aminopeptidase	Cfa.3774.1.A1_s_at	Down

Table. 13 Genes associated with Skeletal/Muscular disorders and contraction/function

Table 15 Genes associated with skeletal muscular disorders and contractility/function		
Annotation	Probe	Direction of expression
		H v. P
Skeletal/Muscular disorders		
Toll-like receptor 2	CfaAffx.13248.1.S1_s_at	down
colony stimulating factor 1 receptor	CfaAffx.27899.1.S1_at	down
fibronectin 1	Cfa.3707.3.S1_s_at	down
neuregulin 1	CfaAffx.10523.1.S1_s_at	down
Muscle contraction/function		
exostoses	Cfa.11001.1.A1_at	up
microfibrillar associated protein 5	CfaAffx.21449.1.S1_s_at	up
myosin light polypeptide kinase	Cfa.13192.1.S1_at	up
dystroglycan 1	CfaAffx.17467.1.S1_at	up
Moloney leukemia virus 10-like 1	CfaAffx.1982.1.S1_s_at	up

Table. 14 Genes associated with Cellular Compromise

Annotation	Probe	Direction of expression
		H v. P
neuroregulin 1	CfaAffx.10523.1.S1_s_at	down
fibronectin 1	Cfa.3707.3.S1_s_at	down
macrophage receptor with collagenous structure	Cfa.15713.1.A1_at	down
xanthine dehydrogenase	CfaAffx.9452.1.S1_s_at	down
bactericidal permeability increasing protein	CfaAffx.14056.1.S1_s_at	down
phospholipase C, beta 1	Cfa.10853.1.A1_at	down
hemoglobin epsilon 1	CfaAffx.10240.1.S1_s_at	up

Table. 15 Genes associated with various functions

Annotation	Probe	Direction of expression
		H v. P
CCR4-NOT transcription complex, subunit 10	Cfa.2403.1.A1_at	up
ubiquitin carboxy-terminal hydrolase L5	Cfa.20277.1.S1_at	up
SH3-domain GBR2-like 3	Cfa.10644.1.A1_at	up
DEAH (Asp-Glu-Ala-His) box polypeptide 8	CfaAffx.22452.1.S1_s_at	up
Signal-induced proliferation-associated 1 like 2	Cfa.14345.1.A1_at	up
Cathepsin E	CfaAffx.15966.1.S1_at	up
ephrin-B3	Cfa.15231.1.A1_at	up
Fc fragment of IgG, receptor, transporter, alpha	Cfa.17806.2.S1_s_at	up
Chromosome 20 open reading frame 172	Cfa.9662.1.A1_at	up
Inositol(myo)-1(or 4) monophosphatase 2	Cfa.13029.1.A1_at	up
Ferrochelatase (protoporphyria)	Cfa.365.3.A1_x_at	up
G-protein coupled receptor 158	Cfa.9162.1.A1_at	up
dual specificity phosphatase 19	CfaAffx.22249.1.S1_at	up
Ring finger protein 152	Cfa.15414.1.A1_at	up
Chromosome 12 open reading frame 49	Cfa.11298.1.S1_at	up
hemoglobin, epsilon 1	CfaAffx.10240.1.S1_s_at	up
Crystallin lambda 1	Cfa.4354.1.S1_a_at	up
Sorting nexin 6	Cfa.10261.1.S1_at	up
microfibrillar associated protein 5	CfaAffx.21449.1.S1_s_at	up
coiled-coil domain containing 46	CfaAffx.17557.1.S1_s_at	up
Nuclear factor I/B	Cfa.4487.1.S1_at	up
carboxypeptidase A3	CfaAffx.13076.1.S1_at	up
chromosome 18, clone RP11-396D4	CfaAffx.26602.1.S1_at	up

coxsackie and adenovirus receptor protein	CfaAffx.13148.1.S1_at	up
WW domain containing oxidoreductase, transcript variant 4	Cfa.9201.1.A1_at	up
EH domain binding protein 1-like 1	Cfa.17641.1.S1_s_at	Down
RP4-760G15 on chromosome 11p13	Cfa.17214.1.S1_at	Down
phospholipase C beta 1 (phosphoinositide-specific)	Cfa.10853.1.A1_at	Down

TABLE 15 (cont)

cathepsin L	CfaAffx.2868.1.S1_at	Down
Transcription elongation factor A, 1	Cfa.12580.1.S1_s_at	Down
glypican 6	Cfa.9523.1.A1_at	Down
Hypothetical protein FLJ21062	Cfa.1933.2.A1_at	Down
G-protein coupled receptor 101	CfaAffx.12632.1.S1_at	Down
Leucine-rich repeat kinase 2	CfaAffx.15613.1.S1_s_at	Down
Six transmembrane epithelial anitgen of the prostate 2	Cfa.9430.1.A1_at	Down
KIAA1074 protein	Cfa.12143.1.A1_at	down
LOC284395	Cfa.4802.1.S1_at	down
elastin microfibril interfacer 2	CfaAffx.28185.1.S1_at	down
Sulfatase 2	CfaAffx.17034.1.S1_s_at	down
IBR domain containing 2	Cfa.1882.1.A1_at	down
Leucyl/cystinyl aminopeptidase	CfaAffx.12483.1.S1_s_at	down
acyltransferase like 1	CfaAffx.14833.1.S1_at	down
NAD kinase	CfaAffx.29324.1.S1_at	down
Immunoglobulin heavy constant alpha 2 transcription factor EC (TFEC), transcript variant 2	Cfa.4556.3.A1_x_at	down
Acyl-CoA synthetase short-chain family member 1	Cfa.1175.1.A1_s_at	down
Syntaxin binding protein 6	CfaAffx.12483.1.S1_at	down
Opioid growth factor receptor-like 1	Cfa.9054.1.A1_at	down
None	Cfa.583.1.S1_at	down
None	Cfa.10106.1.A1_at	down
ELK/RAB6-interacting/CAST family member 2	Cfa.4556.3.A1_s_at	down
adipsin/complement factor D precursor	Cfa.21214.1.S1_at	down
Hypothetical protein FLJ22662	Cfa.21381.1.S1_s_at	down
solute carrier family 2 (facilitated glucose transporter), member 9	CfaAffx.20271.1.S1_at	down
Homogentisate 1,2 dioxygenase	Cfa.7132.1.A1_at	down
serpin peptidase inhibitor, clade B (ovalbumin), member 10	CfaAffx.9076.1.S1_s_at	down
None	CfaAffx.1043.1.S1_s_at	down
Fibrinogen-like 2	Cfa.13203.1.S1_at	down
None	CfaAffx.7369.1.S1_s_at	down
None	Cfa.12500.1.A1_at	down
None	Cfa.4556.2.S1_s_at	down
None	Cfa.4555.1.S1_s_at	down
None	Cfa.280.1.S1_at	down
Lymphocyte antigen 6	CfaAffx.1839.1.S1_at	down
Neuronal PAS domain protein 3	Cfa.12905.1.A1_a_at	down

[0050] Gene expression profiles from puppies fed Composition H or P from weaning to one year of age are obtained and compared. Microarray data indicate that, at a minimum of 1.5 fold change, 99 genes are differentially expressed in the two study groups.

[0051] Genes associated with immune activation are down-regulated in puppies fed Composition H when compared to puppies fed Composition P, which suggests improved immune system function (Table 9).

[0052] Genes associated with lipid metabolism are down-regulated in puppies fed Composition H when compared to puppies fed Composition P, which suggests reduced lipid processing (Table 11).

[0053] Genes associated with cardiovascular development are also down-regulated in puppies fed Composition H when compared to puppies fed Composition P, which suggests enhanced cardiovascular health (Table 12).

[0054] Genes associated with skeletal muscular disorders are down-regulated and genes associated with muscle contraction/function are up-regulated in puppies fed Composition H when compared to puppies fed Composition P, which suggests reduced skeletal disorder risk and improved muscle contraction (Table 13).

[0055] Genes associated with cellular compromise are down-regulated in puppies fed Composition H when compared to puppies fed Composition P, which suggests reduced cellular damage (Table 14).

[0056] Genes known to have an association with cancer are down-regulated in puppies fed Composition H when compared to puppies fed Composition P, which may suggest reduced cancer susceptibility. (Table 10).

[0057] As discussed above, genetic data such as these indicate that the nutritional benefits of the compositions of the present invention include the beneficial modification of gene expression in the animal such that there is an

overall enhancement in the development of the animal. In addition, the beneficial modification of gene expression may also result in a decrease in the incidence of disease in the animal due to an inhibition in expression of disease related genes and/or an increase in the expression of genes which play a role in disease prevention.

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CLAIMS:

1. A pet food composition for the regulation of a gene in a canine puppy,
comprising:

about 5 to about 70% protein,

5 about 0.5 to about 1.6% methionine,

about 50 to about 200 ppm manganese,

about 0.1 to about 0.5% DHA,

about 0.1 to about 0.7% EPA,

about 1200 to about 7500 ppm choline,

10 about 1000 to about 2000 ppm taurine,

about 2.5 to about 6% linoleic acid,

about 1 to about 3% total n-3 fatty acids,

about 50 to about 1200 IU/kg vitamin E,

about 50 to about 500 ppm vitamin C,

15 about 50 to about 500 ppm carnitine, and

about 2.5 to about 7g lysine/1000 kcal,

and the regulation of the gene is at least one of:

(a) down-regulation of genes associated with arthritis or inflammation, wherein
the genes associated with arthritis or inflammation comprise interleukin 1-beta, fibronectin,

20 v-fos FBJ murine osteosarcoma viral oncogene homolog, dual specificity phosphatase 1,

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lactotransferrin, compliment component 5a receptor, xanthine dehydrogenase, and secreted phosphoprotein 1;

(b) down-regulation of genes associated with DNA fragmentation, wherein the genes associated with DNA fragmentation comprise interleukin 1-beta, fibronectin,
5 neuroregulin 1, dual specificity phosphatase 1, and cell death-inducing DFFA-like effector a;

(c) up-regulation of genes associated with DNA processing control, wherein the genes associated with DNA processing control comprise budding uninhibited by benzimidazoles 1 homolog, topoisomerase II alpha, kinetochore associated 2, claspin homolog, cyclin B1, cyclin B2, human chromosome condensation protein G, leucine zipper
10 protein 5, kinesin family member 23, and flap structure specific endonuclease I;

(d) down-regulation of genes associated with cancer incidence, wherein the genes associated with cancer incidence comprise interleukin 1-beta, fibronectin, v-fos FBJ murine osteosarcoma viral oncogene homolog, dual specificity phosphatase 1, compliment component 5a receptor, xanthine dehydrogenase, cellular repressor of E1A-stimulated
15 genes 1, chondroitin sulfate proteoglycan 2, mycrocytic anemia transcobalamin II, clade B member 10 serpin peptidase inhibitor, adenomatosis polyposis coli down-regulated 1, six membrane epithelial antigen of the prostate 2, and alanyl aminopeptidase;

(e) up-regulation of genes associated with tumor suppression, wherein the genes associated with tumor suppression comprise cancer susceptibility candidate 5,
20 paternally expressed 3, topoisomerase II alpha, tubulin alpha 6, cyclin B1, cell division cycle associated 1, cell division cycle 2, ribonucleotide reductase M2 polypeptide, breast cancer anti-estrogen resistance 3, tumor necrosis factor receptor superfamily member 17, and protein kincase C iota;

(f) down-regulation of genes associated with cellular compromise, wherein the
25 genes associated with cellular compromise comprise interleukin 1-beta, fibronectin, neuroregulin 1, v-fos FBJ murine osteosarcoma viral oncogene homolog, dual specificity phosphatase 1, compliment component 5a receptor, xanthine dehydrogenase, and macrophase receptor with collagenous structure;

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(g) up-regulation of genes associated with cellular integrity, wherein the genes associated with cellular integrity comprise cyclin B1, cell division cycle 2, hemoglobin epsilon 1, and protein kinase C iota;

5 (h) down-regulation of genes associated with disruption of cellular assembly, wherein the genes associated with disruption of cellular assembly comprise interleukin 1-beta, fibronectin, neuroregulin 1, chondroitin sulfate proteoglycan 2, muscarinic 3 cholinergic receptor, xanthine dehydrogenase, and macrophage receptor with collagenous structure;

10 (i) up-regulation of genes associated with cellular organization, wherein the genes associated with cellular organization comprise cyclin B1, cell division cycle 2, leucine zipper protein 5, protein kinase C iota, kinesin family member 23, budding uninhibited by benzimidazoles 1 homolog, cyclin B2, topoisomerase II alpha, kinetochore associated 2, cell division cycle associated 1, human chromosome condensation protein G, kinesin family member 11, A kinase (PRKA) anchor protein 2, and thymopioten;

15 (j) down-regulation of genes associated with disruption of cell cycle control, wherein the genes associated with disruption of cell cycle control comprise interleukin 1-beta, fibronectin, neuroregulin 1, v-fos FBJ murine osteosarcoma viral oncogene homolog, muscarinic 3 cholinergic receptor, dual specificity phosphatase 1, and cellular repressor of E1A-stimulated genes 1;

20 (k) up-regulation of genes associated with cell cycle regulation, wherein the genes associated with cell cycle regulation comprise cyclin B1, cell division cycle 2, leucine zipper protein 5, 350/400ka mitotin centromere protein F, breast cancer anti-estrogen resistance 3, budding uninhibited by benzimidazoles 1 homolog, cyclin B2, topoisomerase II alpha, kinetochore associated 2, cell division cycle associated 1, human chromosome condensation protein G, kinesin family member 11, and thymopioten;

25 (l) down-regulation of genes associated with immune activation, wherein the genes associated with immune activation comprise lymphocyte antigen 6 complex, fibrinogen-like 2, CD1A antigen, complement factor D (adipsin), CD163 or macrophage-associated antigen, toll-like receptor 2, renal amyloidosis lysozyme, transcription elongation

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factor A (SII) 3, fibronectin 1, macrophage receptor with collagenous structure, bactericidal/permeability-increasing protein, T cell receptor alpha locus, and immunoglobulin heavy constant alpha 2;

(m) down-regulation of genes associated with lipid metabolism, wherein the
 5 genes associated with lipid metabolism comprise toll-like receptor 2, cytochrome P450 family 1 subfamily B, polypeptide 1, colony stimulating factor 1 receptor, phospholipase C beta 1, fibronectin 1, neuregulin 1, macrophage receptor with collagenous structure, and CD1a molecule;

(n) down-regulation of genes associated with cardiovascular development,
 10 wherein the genes associated with cardiovascular development comprise bactericidal/permeability-increasing protein, fibronectin 1, neuregulin 1, xanthine dehydrogenase, annexin A6, colony stimulating factor 1 receptor, and alanyl (membrane) aminopeptidase;

(o) down-regulation of genes associated with skeletal muscular disorders,
 15 wherein the genes associated with skeletal muscular disorders comprise toll-like receptor 2, colony stimulating factor 1 receptor, fibronectin 1, and neuregulin 1;

(p) up-regulation of genes associated with muscle contraction/function,
 wherein the genes associated with muscle contraction/function comprise exostoses, microfibrillar associated protein 5, myosin light polypeptide kinase, dystroglycan 1, and
 20 moloney leukemia virus 10-like 1;

(q) down-regulation of genes associated with cellular compromise, wherein the genes associated with cellular compromise comprise neuroregulin 1, fibronectin 1, macrophage receptor with collagenous structure, xanthine dehydrogenase, bactericidal permeability increasing protein, and phospholipase C beta 1; and

25 (r) down-regulation of genes associated with cancer, wherein the genes associated with cancer comprise fibronectin 1, xanthine dehydrogenase, neuregulin 1, colony stimulating factor 1 receptor, alanyl aminopeptidase, and S100 calcium binding protein.

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2. The composition of claim 1 comprising:
 - 0 to about 90% by weight of carbohydrates;
 - about 20% to about 60% by weight of protein;
 - about 2% to about 50% by weight of fat;
 - 5 about 0.1% to about 20%, by weight of total dietary fiber; and
 - 0 to about 15% by weight of vitamins, minerals, and other nutrients.
3. The composition of claim 1 or 2 comprising about 0.1% to about 0.4% DHA.
4. The composition of any one of claims 1 to 3 comprising about 100 ppm to about 500 ppm carnitine.
- 10 5. The composition of any one of claims 1 to 4 comprising about 0.1% to about 0.6% EPA.
6. The composition of any one of claims 1 to 5 comprising about 50 ppm to about 150 ppm manganese.
7. The composition of any one of claims 1 to 6 comprising about 0.8% to
15 about 1.6% methionine.
8. The composition of claim 1 wherein the canine puppy is born of a dam fed the composition as defined in any one of claims 1 to 7 during pregnancy.
9. The composition of claim 8 wherein the puppy is *in utero*.
10. The composition of claim 8 wherein the dam was fed a composition as defined
20 in any one of claims 1 to 7 prior to pregnancy.
11. The composition of claim 8 or 10 wherein the dam was fed a composition as defined in any one of claims 1 to 7 for a majority of the pregnancy duration.

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12. The composition of any one of claims 8 to 11 wherein the dam was fed compositions consisting essentially of a composition as defined in any one of claims 1 to 7 prior to and during pregnancy.

13. The composition of any one of claims 1 to 12, wherein the regulation of gene
5 expression is prior to weaning.

14. The composition of any one of claims 1 to 12, wherein the regulation of gene expression is post weaning.

15. A pet food composition for the regulation of a gene in a canine puppy,
comprising:

10 about 6% moisture,
about 29% crude protein,
about 18% fat,
about 1.4% Ca,
about 1% P,
15 about 0.3% EPA,
about 0.2% DHA,
about 4% linoleic acid,
about 1% total n-3 fatty acids,
about 4% total n-6 fatty acids,
20 about 0.1% taurine,
about 300 ppm carnitine,

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about 1% methionine,

about 0.4% cystine,

about 95% manganese,

about 800 IU/kg of vitamin E,

5 about 170 ppm of vitamin C, and

about 5000 ppm choline,

and the regulation of the gene is at least one of:

(a) down-regulation of genes associated with arthritis or inflammation, wherein the genes associated with arthritis or inflammation comprise interleukin 1-beta, fibronectin, v-fos FBJ murine osteosarcoma viral oncogene homolog, dual specificity phosphatase 1, lactotransferrin, compliment component 5a receptor, xanthine dehydrogenase, and secreted phosphoprotein 1;

10

(b) down-regulation of genes associated with DNA fragmentation, wherein the genes associated with DNA fragmentation comprise interleukin 1-beta, fibronectin, neuroregulin 1, dual specificity phosphatase 1, and cell death-inducing DFFA-like effector a;

15

(c) up-regulation of genes associated with DNA processing control, wherein the genes associated with DNA processing control comprise budding uninhibited by benzimidazoles 1 homolog, topoisomerase II alpha, kinetochore associated 2, claspin homolog, cyclin B1, cyclin B2, human chromosome condensation protein G, leucine zipper protein 5, kinesin family member 23, and flap structure specific endonuclease I;

20

(d) down-regulation of genes associated with cancer incidence, wherein the genes associated with cancer incidence comprise interleukin 1-beta, fibronectin, v-fos FBJ murine osteosarcoma viral oncogene homolog, dual specificity phosphatase 1, compliment component 5a receptor, xanthine dehydrogenase, cellular repressor of E1A-stimulated genes 1, chondroitin sulfate proteoglycan 2, mycrocytic anemia transcobalamin II, clade B

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member 10 serpin peptidase inhibitor, adenomatosis polyposis coli down-regulated 1, six membrane epithelial antigen of the prostate 2, and alanyl aminopeptidase;

(e) up-regulation of genes associated with tumor suppression, wherein the genes associated with tumor suppression comprise cancer susceptibility candidate 5, 5 paternally expressed 3, topoisomerase II alpha, tubulin alpha 6, cyclin B1, cell division cycle associated 1, cell division cycle 2, ribonucleotide reductase M2 polypeptide, breast cancer anti-estrogen resistance 3, tumor necrosis factor receptor superfamily member 17, and protein kinase C iota;

(f) down-regulation of genes associated with cellular compromise, wherein the 10 genes associated with cellular compromise comprise interleukin 1-beta, fibronectin, neuroregulin 1, v-fos FBJ murine osteosarcoma viral oncogene homolog, dual specificity phosphatase 1, compliment component 5a receptor, xanthine dehydrogenase, and macrophase receptor with collagenous structure;

(g) up-regulation of genes associated with cellular integrity, wherein the genes 15 associated with cellular integrity comprise cyclin B1, cell division cycle 2, hemoglobin epsilon 1, and protein kinase C iota;

(h) down-regulation of genes associated with disruption of cellular assembly, wherein the genes associated with disruption of cellular assembly comprise interleukin 1-beta, fibronectin, neuroregulin 1, chondroitin sulfate proteoglycan 2, muscarinic 3 cholinergic 20 receptor, xanthine dehydrogenase, and macrophage receptor with collagenous structure;

(i) up-regulation of genes associated with cellular organization, wherein the genes associated with cellular organization comprise cyclin B1, cell division cycle 2, leucine zipper protein 5, protein kinase C iota, kinesin family member 23, budding uninhibited by benzimidazoles 1 homolog, cyclin B2, topoisomerase II alpha, kinetochore associated 2, cell 25 division cycle associated 1, human chromosome condensation protein G, kinesin family member 11, A kinase (PRKA) anchor protein 2, and thymopioten;

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(j) down-regulation of genes associated with disruption of cell cycle control, wherein the genes associated with disruption of cell cycle control comprise interleukin 1-beta, fibronectin, neuroregulin 1, v-fos FBJ murine osteosarcoma viral oncogene homolog, muscarinic 3 cholinergic receptor, dual specificity phosphatase 1, and cellular repressor of
5 E1A-stimulated genes 1;

(k) up-regulation of genes associated with cell cycle regulation, wherein the genes associated with cell cycle regulation comprise cyclin B1, cell division cycle 2, leucine zipper protein 5, 350/400ka mitotin centromere protein F, breast cancer anti-estrogen resistance 3, budding uninhibited by benzimidazoles 1 homolog, cyclin B2, topoisomerase II
10 alpha, kinetochore associated 2, cell division cycle associated 1, human chromosome condensation protein G, kinesin family member 11, and thymopioten;

(l) down-regulation of genes associated with immune activation, wherein the genes associated with immune activation comprise lymphocyte antigen 6 complex, fibrinogen-like 2, CD1A antigen, complement factor D (adipsin), CD163 or macrophage-associated antigen, toll-like receptor 2, renal amyloidosis lysozyme, transcription elongation
15 factor A (SII) 3, fibronectin 1, macrophage receptor with collagenous structure, bactericidal/permeability-increasing protein, T cell receptor alpha locus, and immunoglobulin heavy constant alpha 2;

(m) down-regulation of genes associated with lipid metabolism, wherein the
20 genes associated with lipid metabolism comprise toll-like receptor 2, cytochrome P450 family 1 subfamily B, polypeptide 1, colony stimulating factor 1 receptor, phospholipase C beta 1, fibronectin 1, neuregulin 1, macrophage receptor with collagenous structure, and CD1a molecule;

(n) down-regulation of genes associated with cardiovascular development,
25 wherein the genes associated with cardiovascular development comprise bactericidal/permeability-increasing protein, fibronectin 1, neuregulin 1, xanthine dehydrogenase, annexin A6, colony stimulating factor 1 receptor, and alanyl (membrane) aminopeptidase;

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(o) down-regulation of genes associated with skeletal muscular disorders, wherein the genes associated with skeletal muscular disorders comprise toll-like receptor 2, colony stimulating factor 1 receptor, fibronectin 1, and neuregulin 1;

(p) up-regulation of genes associated with muscle contraction/function, wherein the genes associated with muscle contraction/function comprise exostoses, microfibrillar associated protein 5, myosin light polypeptide kinase, dystroglycan 1, and moloney leukemia virus 10-like 1;

(q) down-regulation of genes associated with cellular compromise, wherein the genes associated with cellular compromise comprise neuroregulin 1, fibronectin 1, macrophage receptor with collagenous structure, xanthine dehydrogenase, bactericidal permeability increasing protein, and phospholipase C beta 1; and

(r) down-regulation of genes associated with cancer, wherein the genes associated with cancer comprise fibronectin 1, xanthine dehydrogenase, neuregulin 1, colony stimulating factor 1 receptor, alanyl aminopeptidase, and S100 calcium binding protein.