(19) Europäisches Patentamt
European Patent Office
Office européen des brevets

(11) Publication number: **0 278 103**
A2

(12) EUROPEAN PATENT APPLICATION

(21) Application number: **87119008.8**

(51) Int. Cl.⁴: **A61K 37/36** , A61K 9/10

(22) Date of filing: **22.12.87**

(30) Priority: **12.01.87 US 2536**

(43) Date of publication of application:
17.08.88 Bulletin 88/33

(84) Designated Contracting States:
AT BE CH DE ES FR GB GR IT LI LU SE

(71) Applicant: **AMERICAN CYANAMID COMPANY**
1937 West Main Street P.O. Box 60
Stamford Connecticut 06904-0060(US)

(72) Inventor: **Tyle, Praveen**
1101 Salisbury CT,
32 LINCOLN, Nebraska 68505(US)

(74) Representative: **Wächtershäuser, Günter, Dr.**
Tal 29
D-8000 München 2(DE)

(54) Sustained release growth hormone compositions for parenteral administration and their use.

(57) The invention relates to sustained release compositions of growth hormones and/or related compounds and multiple water-in oil-in water emulsions. The invention also relates to methods for increasing and maintaining increased levels of growth hormones and/or related compounds in the blood of treated animals for extended periods of time, increasing weight gains in animals and increasing milk production of lactating animals by the administration of a composition of the invention.

EP 0 278 103 A2

SUSTAINED RELEASE GROWTH HORMONE COMPOSITIONS FOR PARENTERAL ADMINISTRATION AND THEIR USE

Advances in the fields of biotechnology and genetic engineering have resulted in the availability of sufficient quantities of biologically active macromolecules such as growth hormones and/or related compounds to make the administration of these agents on a commercial scale economically feasible. Administration of growth hormones and/or related compounds to animals has been reported to provide beneficial effects such as increasing weight gains, increasing milk production in lactating animals, increasing growth rate, increasing feed efficiency, increasing muscle size, decreasing body fat and improving the lean meat to fat ratio. The above beneficial effects may be accomplished by daily injection or periodic injection of sustained release or prolonged release compositions.

Multiple water-in oil-in water emulsions, represented as W/O/W emulsions, described as suitable vehicles for the administration of chemotherapeutic agents are known in the art. The use of multiple W/O/W emulsions for oral administration of insulin has been demonstrated.

It is an object of this invention to provide injectable sustained release compositions of a growth hormone and/or a related compound, wherein the internal aqueous phase contains the growth hormone and/or a related compound, wherein the internal aqueous phase contains the growth hormone and/or related compound emulsified in an oil phase which in turn is emulsified in an aqueous phase.

SUMMARY OF THE INVENTION

The present invention is directed to novel sustained release multiple water-in oil-in water ($W_1/O/W_2$) emulsions comprising an internal aqueous phase (W_1) containing a growth hormone, growth factor, somatomedin, or biologically active fragment or derivative thereof; dispersed in a water immiscible liquid or oil phase (O); dispersed in an external aqueous phase (W_2). The invention is also directed to methods for elevating and maintaining elevated blood levels of a biologically active growth hormone, growth factor, somatomedin, or a biologically active fragment or derivative thereof for the purpose of increasing weight gains, growth rate, milk production, or muscle size, improving feed efficiency, and/or decreasing body fat and improving lean meat to fat ratio in an animal

DETAILED DESCRIPTION OF THE PREFERRED EMBODIMENTS

The compositions of the invention comprise on a weight basis an internal aqueous phase (W_1) of about 55% to 99.7% water, 0.2% to 5% salts and/or buffers, 0.1% to 40% of growth hormone, growth factor, somatomedin or a biologically active fragment or derivatives thereof, 0% to 40% polyol, glycol or sugar, and 0% to 2% preservatives and/or stabilizers, dispersed in an oil phase (O) of about 65% to 98% pharmaceutically and pharmacologically acceptable oil or water immiscible liquid, 2% to 40% non-ionic surfactant(s), 0% to 15% thickening agent, gelling agent or a mixture thereof, dispersed in a second aqueous phase (W_2) of about 38% to 98% water, 0.2% to 5% salts and/or buffers, 2% to 20% non-ionic surfactant(s), 0% to 15% thickening agent, gelling agent, or a mixture thereof, 0% to 2% preservatives and/or stabilizer, 0% to 50% polyol, glycol or a sugar. Preferred compositions of the invention comprise a $W_1/O/W_2$ emulsion on a weight ratio basis of from 1/1/1 to 1/3/8 of the various phases as described above.

Stabilizers, preservatives, surfactants, glycols, polyols, sugars, thickening agents, gelling agents, salts, buffers and mixtures thereof which are employed in the compositions of the invention normally comprise on a weight basis from 10% to 25% and preferably 14% to 25% of the total composition. These excipients provide maximum stability of the multiple emulsion, adjust the viscosity of the final composition and control the rate of release of the biologically active agent from the inner aqueous phase by providing the appropriate concentration gradient between the inner aqueous phase (W_1) and the outer aqueous phase (W_2).

Preferred salts and buffers employed in the aqueous phases of the invention are those which are normally used in the preparation of phosphate buffered saline (PBS), containing $NaH_2PO_4 \bullet H_2O$ (0.025 mol), Na_2HPO_4 (0.025 mol), and NaCl (0.15 mol), adjusted to pH 7.1; carbonate buffered saline (CBS), containing Na_2CO_3 (0.025 mol), $NaHCO_3$ (0.025 mol), and NaCl (0.15 mol), adjusted to pH 9.4; and saline.

Preferred stabilizers employed in the compositions of the invention include dehydroacetic acid and salts thereof, preferably the sodium salt; salicylanilide; sorbic acid, boric acid, benzoic acid and salts thereof;

sodium nitrite and sodium nitrate.

Preferred non-ionic surfactants for use in the compositions of the invention include the sorbitan oleates and stearates, polyethoxylated sorbitan oleates, and block copolymers of ethylene oxide and propylene oxide; with total amounts of from 2% to 20% on a weight basis being distributed between the oil phase (O) and the outer aqueous phase being preferred.

A preferred embodiment of this invention is the incorporation of 1% to 10% of sorbitan monooleate, sorbitan trioleate, sorbitan sesquioleate, ethoxylated (5) soya sterol or sorbitan monostearate in the oil phase (O); in conjunction with the incorporation of 1.0% to 10% of polyoxyethylene (20) sorbitan monooleate or a block copolymer of ethylene oxide and propylene oxide in the outer aqueous phase (W_2).

Thickening agents, gelling agents and sugars useful in the compositions of the invention may be naturally occuring or synthetic in origin. Thickening agents, gelling agents, suspending agents, bulking substances, tonicity modifiers, or sugars with aluminum monostearate, aluminum distearate, aluminum tristearate, gelatin, polyvinyl pyrrolidone, sodium alginate, sodium carboxymethyl cellulose, methyl cellulose, polyethylene glycol, sorbitol, mannitol, glycerol, and lactose are preferred.

Pharmaceutically and pharmacologically acceptable water immiscible liquids suitable for use as the oil phase of the invention include oils, liquid fats, water immiscible alcohols and glycols or mixtures thereof.

Preferred water immiscible liquids for use as the oil phase (O) in the compositions of the invention include fatty acid glycerides and blends thereof which are liquid at ambient temperatures. Representative examples are synthetic oils, light mineral oils, heavy mineral oils, vegetable oils, such as olive, sesame seed, peanut, sunflower seed, soybean, cottonseed, corn, safflower, palm, rapeseed and coconut; animal oils such as fish oils, fish liver oils, sperm oils; or fractions derived therefrom; and mixtures thereof.

Biologically active agents suitable for administration in the compositions of the invention include growth hormones, somatomedins, growth factors, and other biologically active fragments and derivatives thereof. Preferred agents include bovine, ovine, equine, porcine, avian, and human growth hormones. The term hormones encompasses those which are of natural, synthetic, recombinant or biosynthetic origin.

The invention is further illustrated by the following non-limiting examples.

EXAMPLE 1

Preparation of sustained release growth hormone multiple emulsions compositions

Procedures

A. Emulsification by Syringe Technique

Lyophilized recombinant bovine growth hormone is dissolved in the primary aqueous phase (W_1) and then taken up in a 10 mL all glass syringe. The oil phase is taken up into a second syringe. All air is expelled from both syringes and they are connected via a three way stopcock with Luer-Lok fittings (Pharmaseal K75). The two phases are mixed by passing them from one syringe to another for a specific number of exchanges. All of the sample (W_1/O primary emulsion) is then pushed into one syringe and the secondary aqueous phase (W_2) taken up into the second syringe. Multiple emulsification (W_1/O/W_2) is then acocmplished by once again passing the contents of the syringe back and forth. Sufficient multiple emulsion is prepared to provide dosage for testing. The emulsions are remixed prior to each injection to insure that a homogeneous dispersion of the primary emulsion is being administered.

B. Emulsification by Homogenization

Lyophilized recombinant bovine growth hormone is dissolved in the primary aqueous phase (W_1) in a beaker and oil phases added to the beaker with continuous homogenization by a Tissumizer (Tekmar, Model SDT-1810) at low speed (20-40 V). The W_1/O primary emulsion formed is then added with homogenization to the beaker containing the external aqueous phase (W_2). The multiple emulsion formed is checked by brightfield light microscopy.

Utilizing the above procedures with the materials listed in Table I below yields the multiple (W_1/O/W_2) emulsion growth hormone compositions listed in Table II below.

TABLE I

Abbreviation	Material
K. Alg	Potassium Alginate
HVO	Hydrogenated Vegetable Oil
LMO	Light Mineral Oil
HMO	Heavy Mineral Oil
CBS	Carbonate Buffered Saline
CB	Carbonate Buffer
Gel	Gelatin Type A, 150 Bloom
Corn	Corn Oil
Cot	Cotton Seed Oil
Ses	Sesame Oil
Lect	Lecithin UF-H
AMS	Aluminum Monostearate
Dextrin	Carbohydrate (Nadex 772)
BW	Beeswax
Sq	Squalene
CO	Castor Oil (Trylox-CO5, Emery)
CMC	Carboxymethyl cellulose
PG	Propylene Glycol
STO	Sorbitan trioleate
SMO	Sorbitan monooleate
SSO	Sorbitan Sesquioleate
MMO	Mannide monooleate
PSMS	Polyoxyethylene (20) sorbitan monostearate
PSMO	Polyoxyethylene (20) sorbitan monooleate
SMS	Sorbitan monosteatate
PSML	Polyoxyethylene (20) sorbitan monolaurate
PSE	Polyoxyethylene (2) stearyl ether

TABLE I (Continued)

Abbreviation	Material
POE	Polyoxyethylene (2) oleyl ether
SLI	Sodium lauriminodipropionate
BCP_1	Block copolymer of ethylene-oxide and propylene oxide Average molecular weight - 8,350
BCP_2	Block copolymer of ethylene-oxide and propylene oxide Average molecular weight - 5,000
BCP_3	Block copolymer of ethylene-oxide and propylene oxide Average molecular weight - 7,700
BCP_4	Block copolymer of ethylene-oxide and propylene oxide Average molecular weight - 10,800
BCP_5	Block copolymer of ethylene-oxide and propylene oxide Average molecular weight - 12,500
Sorb	Sorbitol aqueous solution USP (70% w/w)
EPS	Ethoxylated (5) Phytosterol

TABLE II

Multiple Emulsion Growth Hormone Compositions

Composition	Phase W_1 containing growth hormone	Phase O	Phase W_2	$W_1/O/W_2$
	Components (% w/w)	Components (% w/w)	Components (% w/w)	ratio
1	CBS (100)	LMO(90),STO(10)	CBS(93),Sorb(5),PSMO(2)	1/1/1.33
2	CBS (100)	HMO(96),SMO(10),AMS(2),PSMO(2)	CBS(93),PSMO(2),Sorb(5)	1/1/1.33
3	CBS (100)	HMO(92.3),SMS(7.7)	CBS(97),BCP_1(3)	1/1/1.33
4	CBS (100)	HMO(89),EPS(11)	CBS(97), BCP_1(3)	1/1/1.33
5	CBS (100)	HMO(90),MMO(10)	CBS(93),PSMO(2),Sorb(5)	1/1/2
6	CBS (100)	HMO(82),Lect.(13),PSMO(5)	CBS(91),SMO(2),PSMO(7)	1/1/2
7	CBS (100)	LMO(88),AMS(1),MNO(10),PSMO(1)	CBS(97.8),Gel(0.2),PSMO(2)	1/1/1
8	CBS (100)	LMO(76),AMS(2),MMO(20),PSMO(2)	CBS(97),BCP_1(3)	1/1/2
9	CBS (100)	LMO(89),AMS(1),STO(10)	CBS(93),Sorb(5),PSMO(2)	1/1/2

TABLE II (Continued)

Composition	Phase W_1 containing growth hormone — Components (% w/w)	Phase O — Components (% w/w)	Phase W_2 — Components (% w/w)	$W_1/O/W_2$ ratio
10	K.Alg(0.36),Sorb.(5),PSMO(2),CBS	HVO(67),MMO(33),	CBS(93),Sorb(5),PSMO(2)	1/1/1
11	Dextrin(3),CBS(90)	HMO(87.2),SSO(10.5),PSMS(2.3)	CBS(96),Gel(2),PSMO(2)	1/3/2
12	CBS (100)	LMO(89),AMS(10),STO(1)	CBS(96),Gel(2),PSMO(2)	1/3/8
13	CBS (100)	LMO(89),AMS(1),STO(10)	CBS(93),Sorb(5),PSMO(2)	1/1/1.33
14	CBS (100)	LMO(89),AMS(1),STO(10)	CBS(93),Sorb(5),PSMO(2)	1/1/1.33
15	CBS (73),Sorb(25),BCP_1(2)	BCP_2(12.5),Sq(50),BW(37.5)	CBS(18.75),Sorb(67.5), PSMO(13.75)	1/1/2
16	CBS(67), Sorb (33)	Corn (83.4),CO(16.6)	CBS(95.15),CMC(2),PSMO(1), PSML(1), NaCl(0/85)	1/2/2
17	CBS(67), Sorb(33)	Cot(83.75),PSE(11.25),POE(5)	CBS(83.3)CO(16.6)	1/2/2
18	CBS(100),	Ses(95),SMO(5),	CBS(90),BCP_3(5),BCP_4(5)	1.5/2.5

0 278 103

EXAMPLE 2

Effectiveness of injectable compositions of the invention

The efficacy of injectable compositions of this invention is demonstrated utilizing a hypophysectomized (hypox) rat assay. The hypophysectomized rat does not produce its own growth hormone and is sensitive to injected bovine growth hormone. The response measured is growth over a period of time such as ten days.

Each of the hypox albino rats (Taconic Farms, Sprague Dawley derived) is injected with a sufficient quantity of representative compositions prepared in Example 1 to provide a dose of 2400 micrograms of bovine growth hormone in 0.2 mL of $W_1/O/W_1$) multiple emulsion.

Test Procedure

Prior to the test, the animals are weighed and the animals to be used for the test are selected based on body weight. Only those animals whose body weights are one standard deviation from the mean body weight of the group are selected. The resulting group is then randomly divided into treatment groups consisting of eight rats/group by a computer generated randomization procedure. The test animals are then transferred to a clean cage and housed four rats/cage. On the initial day of the study the test animals are weighed and any animals with excessive weight gain or loss (± grams) are replaced. The animals are then assigned to test groups and treated.

At the end of the ten-day test period, total weight gain for each animal is recorded and the average weight gain per rat for each treatment determined. The results of these experiments, which are summarized in Table III below, demonstrate the effectiveness of injectable compositions of this invention.

Table III

Efficacy of sustained release compositions of the invention for increasing weight gains in hypox rats

Composition	Average body weight (g)/animal					Average weight gain (g)/animal				
	Day 0	Day 2	Day 4	Day 7	Day 10	Days 0-2	Days 2-4	Days 4-7	Days 7-10	Days 0-10
1	90.3	93.4	98.9	103.4	105.6	3.1	5.4	4.6	2.1	15.2
2	90.0	94.4	98.1	100.3	102.6	4.4	3.8	2.1	2.4	12.7
3	84.8	89.5	92.0	92.5	93.0	4.8	2.5	0.5	0.5	6.3
4	86.4	89.6	93.4	97.4	95.5	3.3	3.8	4.0	-1.9	9.1
5	90.8	93.0	96.8	96.1	98.1	2.3	3.8	-0.6	2.0	7.5
6	86.0	94.1	95.4	95.4	97.4	6.1	1.3	0.0	2.0	9.4
7	93.8	104.7	105.5	108.3	110.0	10.8	0.8	2.8	1.7	16.1
8	86.9	91.7	91.7	93.1	95.1	4.9	0.0	1.4	2.0	7.3
9	89.3	92.1	94.8	99.6	102.6	2.9	2.6	4.9	3.0	13.4
10	92.9	95.9	99.3	100.1	101.6	3.0	3.4	0.9	1.5	9.8
11	94.3	103.1	102.3	100.8	100.4	8.9	-0.9	-1.5	-0.4	6.1
12	91.1	94.0	94.6	98.4	99.1	2.9	0.6	3.8	0.8	8.1

EXAMPLE 4

Table III (Continued)

Composition	Average body weight (g)/animal					Average weight gain (g)/animal				
	Day 0	Day 2	Day 4	Day 7	Day 10	Days 0-2	Days 2-4	Days 4-7	Days 7-10	Days 0-10
13	94.3	98.0	100.5	105.5	103.6	3.8	2.5	5.0	-1.9	9.4
14	94.6	97.3	103.8	106.4	104.8	2.6	6.5	2.6	-1.6	10.1
15	92.3	94.3	96.9	98.6	98.9	2.0	2.6	1.8	0.3	6.7
16[1]	90.9	91.0	92.7	96.4	94.9	0.1	1.7	3.7	-1.6	3.9
17[1]	89.3	89.6	92.4	92.5	92.0	0.4	2.8	0.1	-0.5	2.8
18	91.9	97.9	97.6	102.4	104.3	6.0	-0.3	4.8	1.9	12.4

[1]Bovine growth hormone dose 1200 micrograms.

Effectiveness of compositions of the invention for increasing and maintaining increased levels of growth hormone in blood

Groups of three wether lambs weighing approximately 35 kg each are treated with the compositions described in Table IV below.

Prior to injecting the formulation, one pretreatment blood sample is obtained from each animal at 24 hours before treatment. These animals are acclimated to the facilities and fed daily at 8:00 a.m. Care is taken so as not to excite the sheep any more than necessary, as this may stimulate a natural release of growth hormone.

On the day of treatment, blood samples are taken just prior to injection. Each sheep then receives a single injection of the formulation. Blood samples are collected at 0, 2, 4, 6, 24, 48, 72, 96 hours and periodically thereafter.

The serum is separated from the clot by centrifugation and the serum frozen and delivered to the Analytical Laboratory for growth hormone by radioimmunoassay procedures.

The results of these experiments which are summarized in Table V below demonstrate the effectiveness of the compositions of the invention for increasing and maintaining increased blood levels of growth hormones. Comparable results are obtained with other compositions of the invention.

Table IV

		% w/w of Phase	% of Total
Composition			
A.	W_1 Phase		
	Recombinant bovine growth hormone	12.5	3.75
	CBS	87.5	26.3
	O Phase		
	LMO	89.0	27.1
	AMS	1.0	0.03
	STO	10.0	3.0
	W_2 Phase		
	CBS	93.0	37.1
	PSMO	2.0	0.8
	Sorb(70%)	5.0	2.0
Composition			
B.	W_1 Phase		
	Recombinant bovine growth hormone	7.3	2.8
	Gel	13.3	5.1
	Water	79.4	30.4
	O Phase		
	SES	91.9	24.9
	CO	1.8	0.5
	SSO	7.3	1.97
	W_2 Phase		
	Gel	1.0	0.65
	Water	79.0	26.9
	BCP_5	20.0	6.74

Table IV (Continued)

Composition	% w/w of Phase	% of Total
C. W_1 Phase		
Recombinant bovine growth hormone	13.25	2.65
CB	86.75	17.35
O Phase		
SES	95.0	28.5
SMO	5.0	1.5
W_2 Phase		
BCP_3	5.0	2.5
BCP_4	5.0	2.5
Water	90.0	45.0

TABLE V

Bovine growth hormone blood levels in sheep

Composition A

(2 mL)

Time	(ng/mL) Sheep # 1	2	3	Average
- 24 hrs	7.1	8.0	6.6	-
- 23 hrs	5.1	4.0	4.8	4.6
- 22 hrs	5.4	4.5	3.8	4.6
0 hr	9.6	6.0	7.5	7.7
2 hrs	222.0	816.0	979.0	672.3
4 hrs	177.0	505.0	689.0	457.0
6 hrs	166.0	368.0	468.0	334.0
1 day	146.0	49.1	77.3	90.8
2 days	27.9	19.8	21.5	23.1
3 days	27.9	13.7	28.0	23.2
4 days	21.7	11.0	22.0	11.6
6 days	6.6	6.6	7.4	6.9
8 days	7.8	6.9	9.3	8.0
10 days	10.1	7.5	7.7	8.4
13 days	4.7	5.3	7.1	5.7
15 days	6.5	4.7	4.7	5.3
17 days	5.3	3.9	8.3	5.8
20 days	6.4	6.6	5.8	6.3
22 days	5.7	6.1	7.1	6.3
24 days	2.8	4.5	6.0	4.4

TABLE V (Continued)

Composition B

(5 mL)

Time	(ng/mL) Sheep # 1	2	3	Average
- 24 hrs	2.6	4.0	2.3	3.0
0 hr	1.5	4.0	2.9	2.8
1 hr	13.8	10.3	10.4	11.5
	11.1	8.3	8.0	9.2
2 hrs	376.0	66.4	38.5	160.3
	20.7	24.6	20.9	22.1
4 hrs	109.8	102.6	53.3	88.6
6 hrs	171.8	119.8	156.1	149.2
1 day	65.1	176.4	319.4	187.0
2 days	17.1	38.9	67.8	41.3
3 days	9.9	22.0	31.7	21.2
4 days	9.6	14.6	38.9	21.0
5 days	5.1	9.4	28.5	14.3
6 days	2.2	7.7	48.2	19.4
8 days	2.0	18.3	98.1	39.5
10 days	1.5	13.7	80.9	63.4
13 days	1.7	11.7	81.0	31.5
15 days	1.8	15.1	73.7	30.2
17 days	4.0	13.7	73.1	30.3

TABLE V

Composition C

(2.5 mL)

Time	(ng/mL) Sheep # 1	2	3	Average
- 24 hrs	2.7	2.4	1.8	2.3
0 hr	2.9	1.8	2.2	2.3
1 hr	315.5	168.0	196.3	226.6
2 hrs	551.2	280.6	296.9	376.2
4 hrs	756.8	462.2	466.7	561.9
6 hrs	1007.1	593.1	624.6	741.6
1 day	70.4	91.5	142.5	101.5
2 days	29.0	36.0	41.1	35.4
3 days	21.3	23.8	26.2	23.8
4 days	15.3	11.4	18.5	15.1
5 days	19.2	8.3	14.3	13.9
6 days	22.0	5.4	11.9	13.1
8 days	21.7	8.5	8.7	12.9
10 days	21.6	12.3	7.2	13.7
13 days	16.3	19.9	4.1	13.4
15 days	17.0	19.2	3.1	13.1
17 days	14.5	17.5	2.2	11.4
20 days	16.0	14.4	2.3	10.9

Claims

1. A biologically active sustained release multiple water-in oil-in water, emulsion characterized by an internal aqueous phase W_1 containing a growth hormone, growth factor, somatomedin, or a biologically active fragment or derivative thereof; dispersed in a water immiscible or oil phase O; dispersed in an external aqueous phase W_2.

2. The composition according to claim 1, wherein the internal aqueous phase W_1 is characterized on a weight basis of about 55.0% to 99.7% water, 0.2% to 5.0% salts and/or buffers, 0.1% to 40.0% growth hormone, growth factor, somatomedin or a biologically active fragment or derivatives thereof; 0% to 40.0% polyol, glycol or sugar, and 0% to 2.0% preservatives and/or stabilizers, the oil phase O is comprised on a weight basis of about 65.0% to 98% pharmaceutically and pharmacologically acceptable oil or water immiscible liquid, 0% to 15.0% thickening agent, gelling agent or a mixture thereof, 2.0% to 40.0% non-ionic surfactant(s), the external aqueous phase W_2 is comprised on a weight basis of about 38.0% to 98.0% water, 0.2% to 5.0% salts and/or buffers, 2.0% to 20.0% non-ionic surfactant(s), 0% to 15.0% thickening agent, gelling agent or a mixture thereof, 0% ot 2.0% preservative and/or stabilizer and 0% to 60% polyol, glycol, or a sugar.

3. The composition according to claim 2 wherein the ratio of the phases $W_1/O/W_2$ is in the range of from 1/1/1 to 1/3/8 on a weight basis.

4. The composition according to claim 3, wherein the internal aqueous phase contains a bovine, porcine, ovine, equine, avian or human growth hormone of natural, synthetic, recombinant or biosynthetic origin.

5. A method for elevating and maintaining elevated blood levels of a biologically active growth hormone, growth factor, somatomedin, or a biologically active fragment thereof for the purpose of increasing weight gains, growth rate, milk production, or muscle size, improving feed efficiency, and/or decreasing body fat and improving lean meat to fat ratio in an animal characterized by parenterally administering to the animal a biologically active sustained release multiple water-in oil-in water emulsion comprising an internal aquous phase W, containing a growth hormone, growth factor, somatomedin, or a biologically active fragment or derivative thereof; dispersed in a water immiscible or oil phase O; dispersed in an external aqueous phase W_2.

6. The method according to claim 5, wherein the internal aqueous phase W, is characterized on a weight basis of about 55.0% to 99.7% water, 0.2% to 5.0% salts and/or buffers, 0.1% to 40.0% growth hormone, growth factor, somatomedin or a biologically active fragment or derivatives thereof; 0% to 40.0% polyol, glycol or sugar, and 0% to 2.0% preservatives and/or stabilizers, the oil phase O is comprised on a weight basis of about 65.0% to 98% pharmaceutically and pharmacologically acceptable oil or water immiscible liquid, 0 to 15.0% thickening agent, gelling agent or a mixture thereof, 2.0% to 40.0% non-ionic surfactant(s); the external aqueous phase W_2 is comprised on a weight basis of about 38.0% to 98.0% water, 0.2% to 5.0% salts and/or buffers, 2.0% to 20.0% non-ionic surfactant(s), 0% to 15.0% thickening agent, gelling agent or a mixture thereof, 0% to 2.0% preservative and/or stabilizer, and 0% to 60% polyol, glycol, or a sugar.

7. The method according to claim 6, wherein the ratio of the phases $W_1/O/W_2$ is in the range of from 1/1/1 to 1/3/8 on a weight basis.

8. The method according to claim 7, wherein the internal aqueous phase contains a bovine, porcine, ovine, equine, avian or human growth hormone of natural, synthetic, recombinant or biosynthetic origin.

9. A method for preparing a biologically active sustained release multiple water-in oil-in water, emulsion characterized by (a) dissolving a growth hormone, growth factor, somatomedin, or a biologically active fragment or derivative thereof in an internal aqueous phase W, (b) dispersing the phase of step (a) in a water immiscible or oil phase O and (c) dispersing the phase of step (b) in an external aqueous phase W_2.