Phytobase™

Stability Study of Hormones in Phytobase™

The formulation samples were assayed by validated stability indicating HPLC analytical method at predetermined time points to verify the stability of the APIs in the tested vehicle. The API's were tested at high and low concentrations individually (bracketed) and in one combination formula (bracketed). Sampling times were: initial (T = 0), 30 days (T = 30), 60 days (T= 60), 90 days (T = 90), 120 days (T = 120), 150 days (T= 150), and 180 days (T=180). Formulations were immediately assayed at each time point. The evaluation parameter is the percent recovery with respect to T = 0, using the HPLC method. Results are given as percentages. Microbial stability testing USP <61>, <62> and antimicrobial effectiveness testing USP <51> were performed on each product. All samples were stored in Topi-CLICK®* 35 gram closure device at standard room temperature (15-25 oC).

Formulation Tested:
Assay results at time points of indicated formulation amount are displayed in percentages, as compared to the standards for each API. The results presented suggest that the APIs studied are stable alone or in combination in Fitalite/Phytobase for the time period indicated.

Chemical	ºC	T = 0	T = 30	T = 60	T = 90	T = 120	T = 150	T = 180
Estriol (20mg/g)	15-25	103.75%	102.48%	100.18%	100.15%	101.53%	101.03%	101.54%
Estriol (0.5mg/g)	15-25	99.37%	100.79%	103.96%	98.80%	108.87%	101.32%	107.53%
Estradiol (20 mg/g)	15-25	98.01%	97.36%	97.0%	96.71%	93.36%	95.83%	98.10%
Estradiol (0.5mg/g)	15-25	109.56%	108.85%	109.56%	107.62%	109.55%	108.55%	108.77%
Estrone (10 mg/g)	15-25	101.31%	101.47%	101.19%	104.30%	102.77%	103.7%	———
Estrone (1mg/g)	15-25	102.69%	102.47%	102.85%	100.75%	101.73%	103.89%	———
Progesterone (200 mg/g)	15-25	107.76%	107.45%	106.71%	106.44%	106.25%	106.21%	106.38%
Progesterone (1mg/g)	15-25	101.28%	103.62%	102.32%	102.70%	102.04%	102.04%	102.21%
Testosterone (200 mg/g)	15-25	104.17%	104.12%	104.25%	104.83%	104.09%	104.09%	104.34%
Testosterone (1mg/g)	15-25	105.14%	103.09%	103.83%	103.12%	102.70%	102.70%	102.85%

Assay Results at Time Points of Indicated Formulation Amount: Estriol 0.5mg/gm, Estradiol 0.5mg/gm, and Progesterone 5mg/gm

Chemical	ºC	T = 0	T = 30	T = 60	T = 90	T = 120	T = 150	T = 180
Low Concentration Combo Estriol	**15-25**	**109.89%**	**109.49%**	**109.89%**	**109.10%**	**109.43%**	**109.57%**	**108.43%**
Low Concentration Combo Estradiol	**15-25**	**108.01%**	**105.72%**	**105.91%**	**106.47%**	**108.38%**	**107.27%**	**108.06%**
Low Concentration Combo Progesterone	**15-25**	**98.33%**	**92.45%**	**92.39%**	**97.51%**	**99.77%**	**97.50%**	**98.04%**

Assay Results at Time Points of Indicated Formulation Amount: Estriol 10mg/gm, Estradiol 20mg/gm, Progesterone 200mg/gm

Chemical	ºC	T = 0	T = 30	T = 60	T = 90	T = 120	T = 150	T = 180
High Concentration Combo Estriol	**15-25**	**102.60%**	**105.48%**	**106.96%**	**106.53%**	**105.94%**	**106.91%**	**106.83%**
High Concentration Combo Estradiol	**15-25**	**104.17%**	**106.83%**	**108.16%**	**105.27%**	**106.49%**	**107.97%**	**107.79%**
High Concentration Combo Progesterone	**15-25**	**104.69%**	**106.53%**	**108.21%**	**104.24%**	**104.76%**	**106.63%**	**107.40%**

Analyses for microbial growth were performed according USP chapters <61> microbiological examination of nonsterile products microbial enumeration tests and <62> microbiological examination of nonsterile products tests for specified microorganisms.

Hormones	ºC	Pseudomonas Aeruginosa	Staphylococcus Aureus	Total aerobic microbial count	Total combined yeast and mold count
Estriol (20mg/g)	**15-25**	No growth for all time points	No growth for all time points	<1,0 X 10UFC/g for all time points	<1,0 X 10UFC/g for all time points
Estriol (0.5mg/g)	**15-25**	No growth for all time points	No growth for all time points	<1,0 X 10UFC/g for all time points	<1,0 X 10UFC/g for all time points
Estradiol (20 mg/g)	**15-25**	No growth for all time points	No growth for all time points	<1,0 X 10UFC/g for all time points	<1,0 X 10UFC/g for all time points
Estradiol (0.5mg/g)	**15-25**	No growth for all time points	No growth for all time points	<1,0 X 10UFC/g for all time points	<1,0 X 10UFC/g for all time points
Estrone (10 mg/g)	**15-25**	No growth for all time points	No growth for all time points	<1,0 X 10UFC/g for all time points	<1,0 X 10UFC/g for all time points
Estrone (1mg/g)	**15-25**	No growth for all time points	No growth for all time points	<1,0 X 10UFC/g for all time points	<1,0 X 10UFC/g for all time points
Progesterone (200 mg/g)	**15-25**	No growth for all time points	No growth for all time points	<1,0 X 10UFC/g for all time points	<1,0 X 10UFC/g for all time points
Progesterone (1mg/g)	**15-25**	No growth for all time points	No growth for all time points	<1,0 X 10UFC/g for all time points	<1,0 X 10UFC/g for all time points

Testosterone (200 mg/g)	**15-25**	No growth for all time points	No growth for all time points	<1,0 X 10UFC/g for all time points	<1,0 X 10UFC/g for all time points
Testosterone (1mg/g)	**15-25**	No growth for all time points	No growth for all time points	<1,0 X 10UFC/g for all time points	<1,0 X 10UFC/g for all time points
Low Concentration Combo Progesterone	**15-25**	No growth for all time points	No growth for all time points	<1,0 X 10UFC/g for all time points	<1,0 X 10UFC/g for all time points
High Concentration Combo Progesterone	**15-25**	No growth for all time points	No growth for all time points	<1,0 X 10UFC/g for all time points	<1,0 X 10UFC/g for all time points

Antimicrobial effectiveness testing (AET) was performed per USP <51> guidelines. Analysis and results of acceptable limits are described in below chart.

Test	Time	Result	Acceptable Limit
Candida albicans	0 hour	3.6 log	N/A
Candida albicans	14 day	0	< 3.6 log increase in the number of CFU initially inoculated
Candida albicans	28 day	0	< 3.6 log increase in the number of CFU initially inoculated
Aspergillus brasiliensis	0 hour	3.6 log	N/A
Aspergillus brasiliensis	14 day	0	< 3.6 log increase in the number of CFU initially inoculated
Aspergillus brasiliensis	28 day	0	< 3.6 log increase in the number of CFU initially inoculated
Escherichia coli	0 hour	4.4 log	N/A
Escherichia coli	14 day	0	< 3.6 log increase in the number of CFU initially inoculated
Escherichia coli	28 day	0	< 3.6 log increase in the number of CFU initially inoculated
Pseudomonas aeruginosa	0 hour	4.0 log	N/A
Pseudomonas aeruginosa	14 day	0	< 3.6 log increase in the number of CFU initially inoculated
Pseudomonas aeruginosa	28 day	0	< 3.6 log increase in the number of CFU initially inoculated
Staphylococcus aureus	0 hour	4.0 log	N/A
Staphylococcus aureus	14 day	0	< 3.6 log increase in the number of CFU initially inoculated
Staphylococcus aureus	28 day	0	< 3.6 log increase in the number of CFU initially inoculated