Effectiveness of Antioxidants in Selected Model Drugs: Mitigation Strategy and Impact of Reformulation in Their Stability

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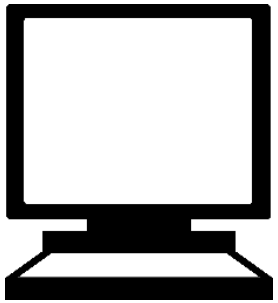
Pharmaceutical Quality

A quality product of any kind consistently meets the expectations of the user



Pharmaceutical Quality

A quality product of any kind consistently meets the expectations of the user

Drugs are no different


Patients expect safe and effective medicine with every dose they take.

Pharmaceutical quality is

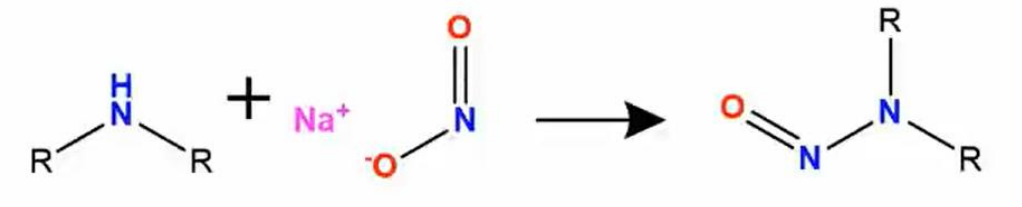
assuring *every* dose is safe and effective, free of contamination and defects.

Presentation Outline

- Nitrosamines & Drug Product Contamination
- Nitrosamine mitigation strategies
- Case study: Drug A (effect of three antioxidants and pH modifiers to mitigate nitrosamine formation)
- Conclusions

Nitrosamines & Drug Product Contamination

- NDMA and other nitrosamines are common contaminants detected in low amounts (ppm) in food, beverages, cosmetics, water, tobacco products and consumer goods



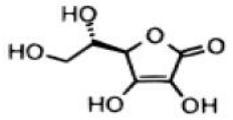
- Nitrosamines are found in drugs and their formation could be due to:
  - Drug synthesis
  - Breakdown of unstable compounds
  - Contamination from recycled solvents used in manufacturing
  - Excipients
  - Manufacturing process
  - Drug packaging

Potential Mitigation of Nitrosamine Drug Substance Related Impurities (*NDSRI*) Risk

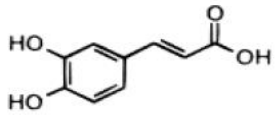
- Use of antioxidant/nitrite scavenging excipients
- Adjustment of pH
- Control of nitrite content in the formulation

Mitigation strategies

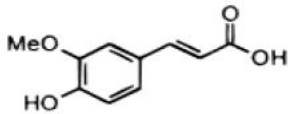
- **Antioxidants:**
  - Have been shown to be effective inhibitors in reducing formation of nitrosamine impurities
  - Ascorbic acid, Caffeic acid and Ferulic acid were chosen for this study, and they each exhibited higher inhibition than other known antioxidants



Ascorbic acid
(Vitamin C)



Caffeic acid

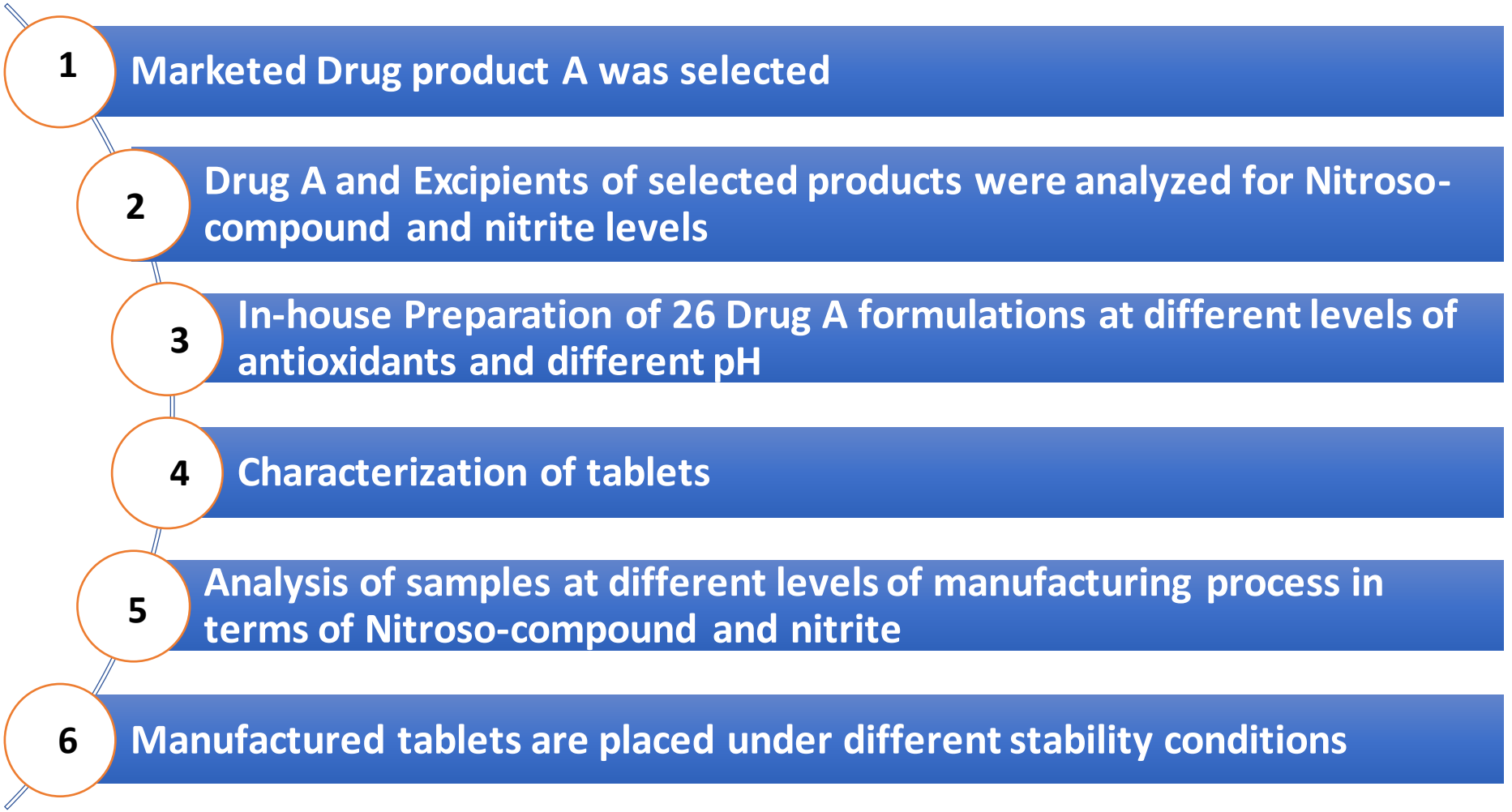


Ferulic acid

- **pH Control**
  - Acidic conditions facilitates the nitrosating reaction to form the nitrosating agent
  - Maintaining neutral pH of the drug product will serve as protection strategy against nitrosamine formation

- **Heat and Moisture Control**
  - Data suggests that formation of nitrosamine is greater under conditions of **elevated heat** and moisture
  - It has been hypothesized that nitrosamines or its main components may be introduced or formed during the **drying** of wet granulation
  - This might be due to the **presence of a secondary amine or NOx** (Oxides of nitrogen) in the wet mass which may be converted into a nitrosamine impurity during drying

Experimental Workflow (Case Study)



Screening of Drug Product

Assessment of the formation extent of nitrosamines in tested drug products

Drug product	Samples to be tested	Conditions
Drug A	Non-crushed tablet	50°C/75% RH for 12 days
	Crushed tablet	
	Crushed tablets + spiked with nitrite	
	Crushed tablets + spiked with nitrite + Antioxidant	

Screening of Excipients

- Physical mixtures of Drug A with corresponding excipients were prepared in certain ratio corresponding to their drug formulations to evaluate their effect on formation of nitroso-compound impurities

	Excipient	Q. (mg)	Stability conditions
Physical Mixtures	No excipient (only DS)	---	50°C/75% RH for 1 month
	Anhydrous Lactose	xx	
	Pregelatinized Starch	xx	
	Avicel PH 112	xx	
	Maize Starch	xx	
	Talc	xx	
	Mg. Stearate	xx	

In-House Formulations (Drug A)

Basic formulation	
Ingredient	**(%)**
Intra-granular	
Drug A	**xx**
Anhydrous Lactose	**xx**
Avicel PH-101	**xx**
Starch 1500	**xx**
Antioxidant	0, 0.1,0.5,1
Granulation	
Mixture of water and IPA	**Q.s**
Nitrite	0 , 100 ppm
Extra-granular	
Corn Starch	**xx**
Avicel PH-101	**xx**
Talc	**xx**
Magnesium Stearate	**xx**
Total	**100**

Formulation #	Antioxidant	Percentage of Antioxidant (%)	Nitrite (100 ppm)	Formulation
1	---	---	---	Control
2	---	---	Spiked	Nitrite Spiked Control
3	Ascorbic Acid	0.1	N/A	Antioxidant effect on nitrite spiked and non-spiked formulations
4			Spiked	
5		0.5	N/A	
6			Spiked	
7		1	N/A	
8			Spiked	
9	Caffeic Acid	0.1	N/A	
10			Spiked	
11		0.5	N/A	
12			Spiked	
13		1	N/A	
14			Spiked	
15	Ferulic Acid	0.1	N/A	
16			Spiked	
17		0.5	N/A	
18			Spiked	
19		1	N/A	
20			Spiked	
21	None		N/A	Acidic pH modifier (pH=3)
22			Spiked	
23			N/A	Basic pH modifier (pH=8-9)
24			Spiked	
25			Spiked	Nitrite Spiked Placebo
26			N/A	Acidic Placebo

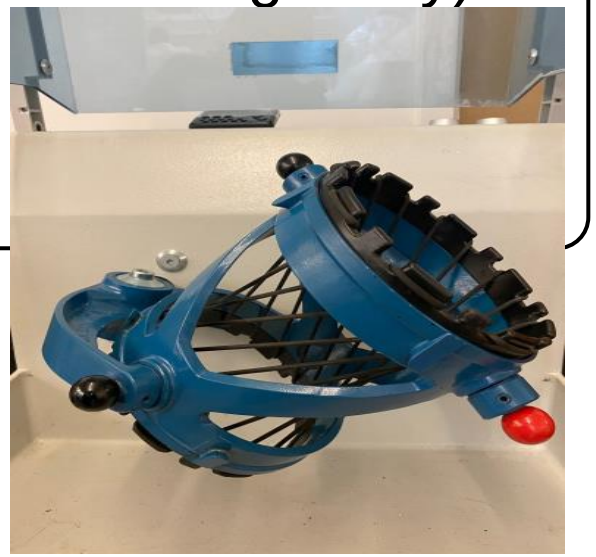
EVALUATED FACTORS	
Antioxidant Type	Ascorbic Cid Caffeic Acid Ferulic Acid
Antioxidant Level	0.1% 0.5% 1.0%
Effect of spiking nitrite	Zero 100 ppm
Effect of pH	Acidic pH = 3.0 Basic pH = 8.0
Moisture and heat	60% water 60°C Heated air
Stability Conditions	50°C/75% RH 1 month 40°C/75% RH 1, 2, 3, 6 months 25°C/60% RH 1, 2, 3, 6 months

Manufacturing Process

Sieving of Drug A (active ingredient) and single excipients

Mixing…

Drug A and antioxidants by geometric dilution (to ensure homogeneity)



Intragranular blend (homogenous and no aggregations)

Granulation…

Binder 40% IPA in H_2O. (60% w/w to intragranular blend)

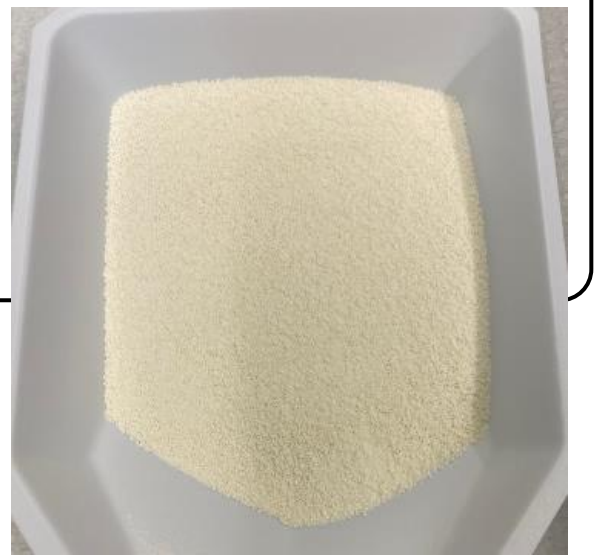
Nitrite was spiked here

Manufacturing Process (Continued)

Semi-Drying…
Granules were sifted thru #20 sieve and placed in 60°C oven (45 mins.)

Drying…
Using a fluidized bed dryer.
airflow: 20m^3/h
temp : 60°C

Granules…
Sifted thru #30 sieve.
Moisture content (< 2%)



Tableting…
Granules were mixed with EG mixture (15 mins.)
And ready for compression.

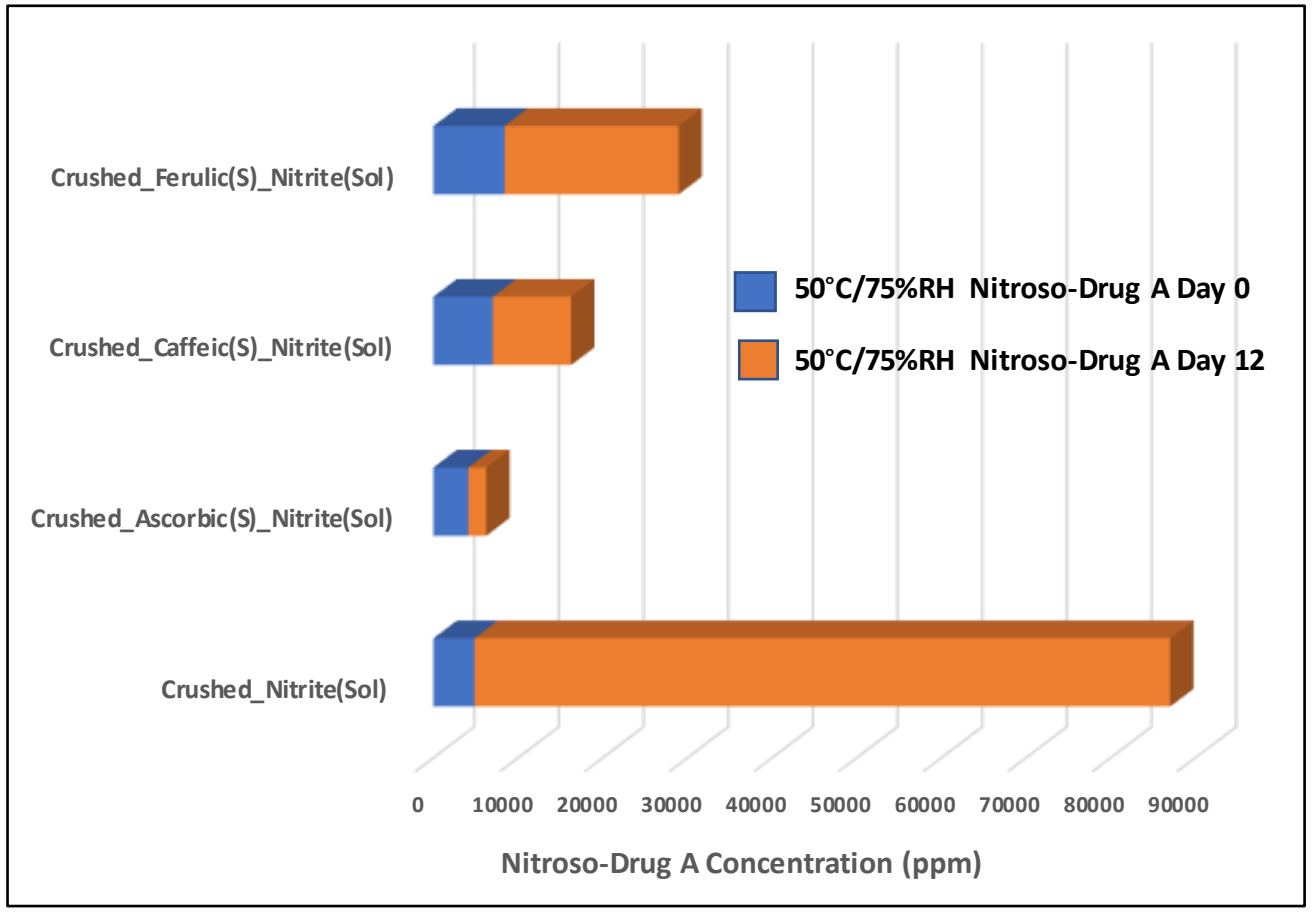
Characterization

- **Pre-granulation blend:**
  - pH was measured
  - Nitroso-drug A and nitrite were analyzed

- **Granules:**
  - pH was measured
  - Moisture content (< 2%) was measured
  - Nitroso-drug A and nitrite were analyzed

- **Tablets:**
  - pH was measured
  - Weight and dimensions were measured: 335-345 mg, 10 mm tablets
  - Hardness (30 – 90 N) and Friability (< 1 w/w%) were measured
  - Nitroso-drug A, nitrite and % drug content were analyzed

Screening of Marketed Drug Product



- Compared to the control "Crushed tablet and spiked with nitrite (in solution) and stored under 50°C/75%RH", spiked formulation with 1% ascorbic acid provided the greatest mitigation when compared to the other two tested antioxidants (caffeic acid & ferulic acid)

- This indicates that **ascorbic acid can be used as a mitigation excipient** in drug A product formulations

Excipients Screening Study

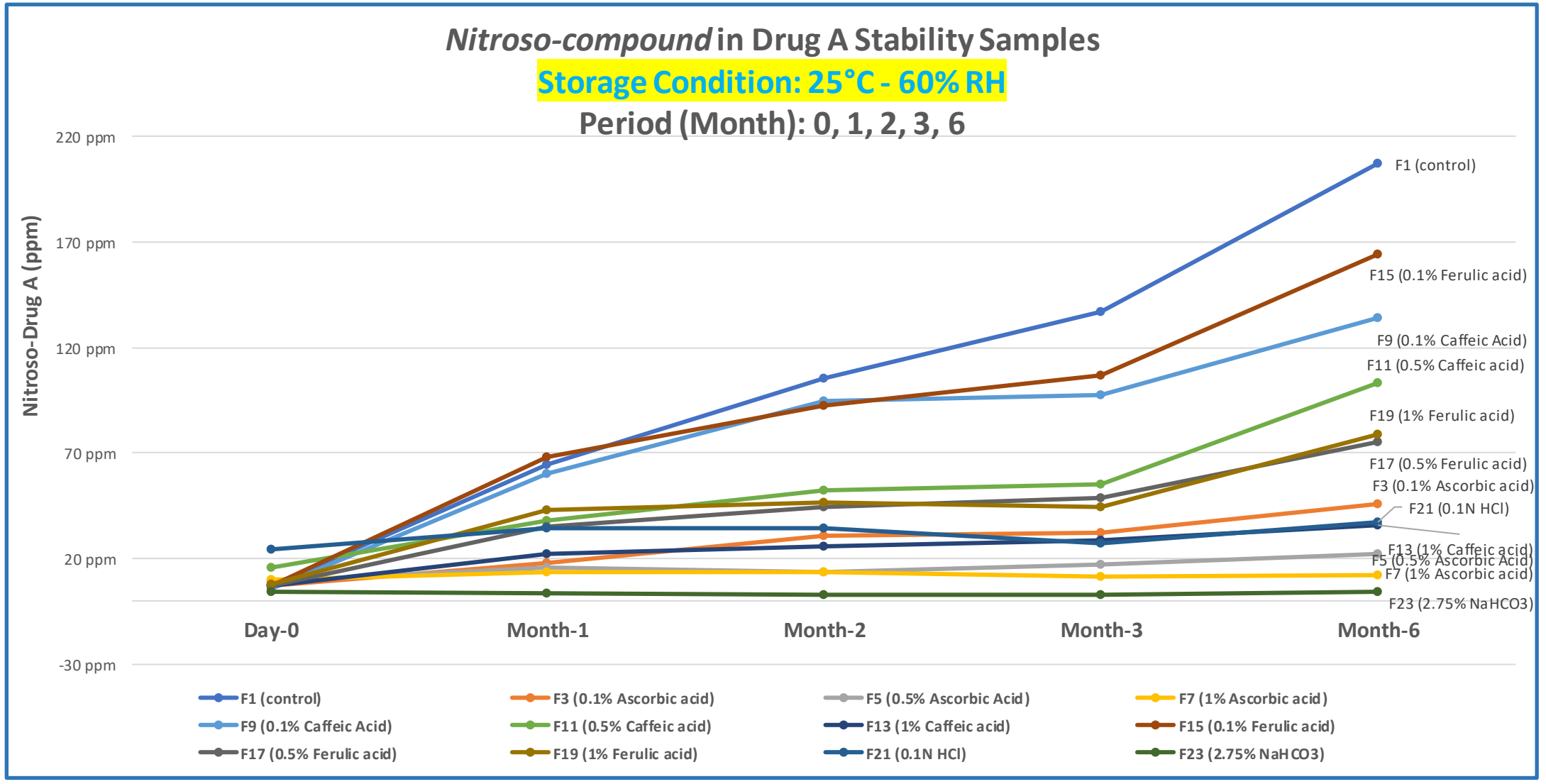
Sample Name	Nitroso-Drug A (µg/g; ppm)		
	Day 0	Room Temp_30-Days	50C/75%RH_30-Days
Drug-A DS	1.468	1.623	1.850
Drug-A DS+Lactose	2.958	23.447	34.154
Drug-A DS+Starch 1500	1.553	5.410	34.984
Drug-A DS+Avicel PH101	2.115	9.097	224.142
Drug-A DS+Corn Starch	1.763	3.084	36.312
Drug-A DS+Talc	2.150	3.588	16.372
Drug-A DS+Magnesium Stearate	2.215	1.526	3.997
Drug-A DS+All Exipients	1.308	2.453	390.760
Placebo	ND	<LOD	<LOQ
Lactose	ND	ND	ND
Starch 1500	ND	<LOD	ND
Avicel PH 101	ND	ND	<LOD
Corn Starch	ND	ND	<LOD
Talc	ND	<LOD	<LOD
Magnesium Stearate	ND	ND	<LOD

pH Measurement

- pH of powdered samples of pre-granulation, granules and final blend were measured using Solids Pro probe (Special probe for solids)
- 6 g of water was added to 2 g of each sample to form a slurry (33%)
- Then, mixed for 5 mins in a vortex mixer and 10 mins in a TURBULA® 3D shaker mixer.

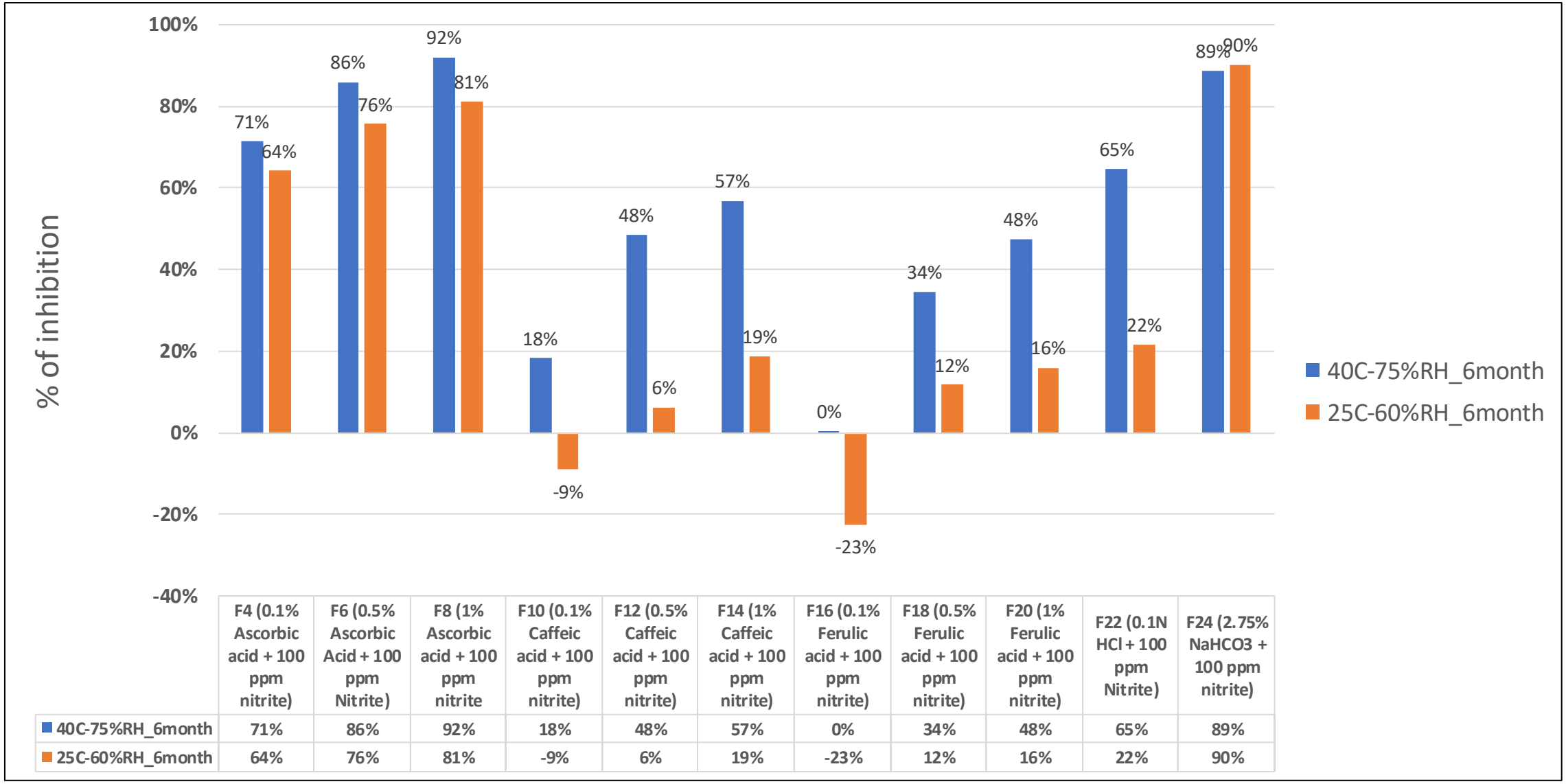
F #	Description	pH		
		Pre-granules	Granules	Final blend (same as tablets)
F1	Original Formulation	5.12	4.93	6.23
F2	100 ppm nitrite	5.13	5.35	6.26
F3	0.1% Ascorbic Acid	4.11	4.27	6.17
F4	0.1% Asc. Acid + 100 ppm nitrite	3.98	4.97	6.18
F5	0.5% Asc. Acid	3.43	3.31	5.78
F6	0.5% Asc. Acid + 100 ppm nitrite	3.43	3.44	6
F7	1% Asc. Acid	3.15	3.16	4.2
F8	1% Asc. Acid + 100 ppm nitrite	3.19	3.35	4.46
F9	0.1% Caffeic Acid	4.27	4.27	6.19
F10	0.1% Caf. Acid + 100 ppm nitrite	4.28	4.72	6.2
F11	0.5% Caf. Acid	3.79	3.74	5.11
F12	0.5% Caf. Acid + 100 ppm nitrite	3.87	4	5.3
F13	1% Caf. Acid	3.84	3.79	4.73
F14	1% Caf. Acid + 100 ppm nitrite	3.84	3.98	4.83
F15	0.1% Ferulic Acid	4.27	4.27	6.15
F16	0.1% Fer. Acid + 100 ppm nitrite	4.21	4.2	6.21
F17	0.5% Fer. Acid	3.8	3.65	4.9
F18	0.5% Fer. Acid + 100 ppm nitrite	3.81	3.7	4.95
F19	1% Fer. Acid	3.81	3.63	4.64
F20	1% Fer. Acid + 100 ppm nitrite	3.85	3.69	4.65
F21	1N HCl + Na_2HPO_4	4.3	2.72	2.95
F22	1N HCl + Na_2HPO_4 + 100 ppm nitrite	4.3	2.72	3.26
F23	NaHCO3	8.06	9.33	9.33
F24	$NaHCO_3$ + 100 ppm nitrite	8	9.52	9.5
F25	100 ppm nitrite (Placebo)	5.48	5.3	7.71
F26	1N HCl + Na_2HPO_4 (Placebo)	4.34	1.72	2.19

Drug A Tablet Analysis: Not Spiked with Nitrite



Assay content for all the manufactured formulations under tested storage conditions were within the allowable USP acceptable limit of 90-110

% of Inhibition Efficiency by The Antioxidants – 6 Month Stability



	F4 (0.1% Ascorbic acid + 100 ppm nitrite)	F6 (0.5% Ascorbic Acid + 100 ppm Nitrite)	F8 (1% Ascorbic acid + 100 ppm nitrite	F10 (0.1% Caffeic acid + 100 ppm nitrite)	F12 (0.5% Caffeic acid + 100 ppm nitrite)	F14 (1% Caffeic acid + 100 ppm nitrite)	F16 (0.1% Ferulic acid + 100 ppm nitrite)	F18 (0.5% Ferulic acid + 100 ppm nitrite)	F20 (1% Ferulic acid + 100 ppm nitrite)	F22 (0.1N HCl + 100 ppm Nitrite)	F24 (2.75% NaHCO3 + 100 ppm nitrite)
40C-75%RH_6month	71%	86%	92%	18%	48%	57%	0%	34%	48%	65%	89%
25C-60%RH_6month	64%	76%	81%	-9%	6%	19%	-23%	12%	16%	22%	90%

Summary & Conclusion

- **Antioxidants:**
  - For the drug A the highest inhibition of NDSRI formation among the antioxidants was observed: ascorbic acid > caffeic acid > ferulic acid
  - Antioxidants need to be fit-for-purpose and their effectiveness depends on the drug substance, manufacturing and formulation
  - The increase in antioxidant concentration improved the NDSRI mitigation

- **pH Control:**
  - Acidic conditions facilitated nitrosating reactions
  - Maintaining neutral pH of the drug product served as a protective strategy against nitrosamine formation. An alkali modifier (sodium bicarbonate) had the most effective inhibition of NDSRI formation for the tested drug A

- **Heat and Moisture Control:**
  - The data suggested ***that formation of NDSRI was greater under conditions of elevated heat and moisture during the drying step of wet granulation***
  - Continuous manufacturing with direct compression may help in nitrosamine mitigation

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