

OSMOLALITY OF EXCIPIENTS FOR PARENTERAL FORMULATION MEASURED BY FREEZING POINT AND VAPOR PRESSURE – A COMPARATIVE ANALYSIS

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Supplemental material

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Supplemental material

Table S1 - Chemical structure, molecular weight (MW) and critical micelle concentration (CMC) of the polymers used in this study. Legend: n/a – not available.

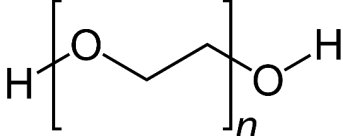
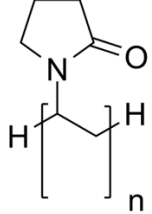
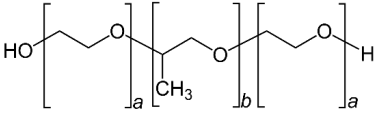
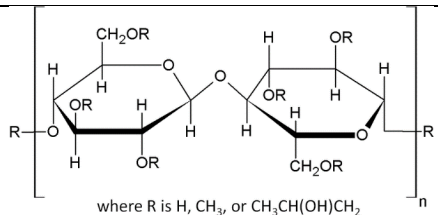
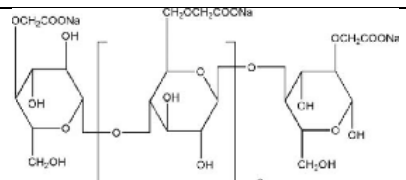
Polymer	Chemical structure	MW (g/mol)	CMC	Ref
PEG 3350		3000-3700	n/a	[1]
PEG 4000		3000-4800	n/a	[1]
PEG 6000		5400-6600	n/a	[2]
PEG 8000		7000-9000	n/a	[1]
PEG 20000		16000-28000	n/a	[3]
PVP-K12		2500	n/a	[1]
PVP-K17		10000	n/a	[1]
PVP-K30		50000	n/a	[1]
Poloxamer 188		7680-9510	0.125 mM (30 °C)	[1,4,5]
Poloxamer 338		12700-17400	1.9-2.6 mM (25 °C)	[1,6]
Poloxamer 407		9481-14600	2.3-3.6 mM (37 °C)	[1,7]
HPMC E5	 <p>where R is H, CH₃, or CH₃CH(OH)CH₂</p>	20000	n/a	n/a
Na-CMC		n/a	n/a	n/a

Table S2 - Chemical structure, molecular weight (MW) and critical micelle concentration (CMC) of the surfactants used in this study.

Surfactant	Chemical structure	MW	CMC	Ref
Polysorbate 20	 Note: R is laurate for polysorbate 20 and oleate for polysorbate 80.	1128	0.0499 mM (25 °C)	[1,8]
Polysorbate 80		1310	0.015 mM (25 °C)	[1,8]
Vitamin E-TPGS		1543	0.02 %w/v (37 °C) 0.13 mM	[1,5]
Lipoid E PG		n/a	n/a	n/a
DOSS		444.56	0.11 %w/v (25 °C) 2.5 mM	[1,5]
SDS		288	0.24%w/v (20 °C) 8.1-8.3 mM	[5,8]

Bayesian bivariate mixed model

Per each subgroup, the Bayesian bivariate mixed model with type of polymer as random effect and concentration as fixed is fitted for FPD and VP and is expressed as:

$$\begin{aligned} \begin{pmatrix} Y_{FPD} \\ Y_{VP} \end{pmatrix} &\sim MVN \left[\begin{pmatrix} \mu_{FPD} \\ \mu_{VP} \end{pmatrix}, \Sigma \right] \\ \begin{pmatrix} Y_{FPD} \\ Y_{VP} \end{pmatrix} &\sim MVN \left[\text{Concentration} + \begin{pmatrix} M_{FPD} \\ M_{VP} \end{pmatrix} + \begin{pmatrix} Polymer_{FPD} \\ Polymer_{VP} \end{pmatrix}, \Sigma \right] \\ \begin{pmatrix} Polymer_{FPD} \\ Polymer_{VP} \end{pmatrix} &\sim MVN \left[\begin{pmatrix} 0 \\ 0 \end{pmatrix}, \Theta \right] \end{aligned}$$

where,

- $\begin{pmatrix} Y_{FPD} \\ Y_{VP} \end{pmatrix}$ is vector of observed osmolality measurements by FPD and VP
- $\begin{pmatrix} \mu_{FPD} \\ \mu_{VP} \end{pmatrix}$ is a mean vector of osmolality measurements by FPD and VP. The mean is a function of concentration and method
- $\Sigma = \begin{pmatrix} \sigma_{FPD}^2 & \sigma_{FPD-VP} \\ \sigma_{FPD-VP} & \sigma_{VP}^2 \end{pmatrix}$ is the residual variance-covariance matrix with residual variances σ_{FPD}^2 and σ_{VP}^2 of FPD and VP, respectively. The σ_{FPD-VP} is the covariance between the two measurement methods.
- $\Theta = \begin{pmatrix} \sigma_{Polymer-FPD}^2 & \sigma_{Polymer-FPD-VP} \\ \sigma_{Polymer-FPD-VP} & \sigma_{Polymer-FPD}^2 \end{pmatrix}$ is a variance-covariance matrix with polymer variances $\sigma_{Polymer-FPD}^2$ and $\sigma_{Polymer-VP}^2$ of FPD and VP, respectively. The polymer variability is different between the two methods. The $\sigma_{Polymer-FPD-VP}$ is the covariance between the two methods since each formulation with specific polymer was measured by both methods.

Concentration was considered as a fixed factor to account for the design of the study. Vaguely informative or non-informative prior distributions were used for all the parameters, meaning that the posterior distributions were basically driven by the data only. Based on the model, average FPD and VP measurements per concentration were reported as well as the 95% high posterior density intervals for these estimates to support the decision whether measurements are on average statistically different or not.

Table S3 - Overview of the polymer solutions prepared with respective concentrations in percentage weight per volume (% w/v) and the difference between the two measuring principles calculated based on the average values. Osmolality values were obtained by the two different osmometer principles: freezing point depression (FDP) and vapor pressure (VP).

Polymer	Concentration (% w/v)	Osmolality (mOsm/kg)		Difference between FPD and VP measurement
		FPD	VP	Δ
PEG 3350	3.5	18.3 \pm 0.6	11.7 \pm 1.5	6.7
	7	58.3 \pm 1.2	53.2 \pm 2.1	26.0
	10.5	124.0 \pm 5.0	76.7 \pm 4.5	47.3
	14	234.7 \pm 5.0	149.0 \pm 4.4	85.7
PEG 4000	3.5	16.7 \pm 0.6	7.7 \pm 2.1	9.0
	7	55.7 \pm 3.8	32.0 \pm 2.6	23.7
	10.5	114.3 \pm 0.6	75.3 \pm 2.1	39.0
	14	219.7 \pm 3.2	135.3 \pm 2.1	84.3
PEG 6000	3.5	15.3 \pm 1.2	7.7 \pm 0.6	7.7
	7	47.0 \pm 1.0	24.0 \pm 1.7	23.0
	10.5	110.0 \pm 1.7	62.0 \pm 1.0	48.0
	14	213.0 \pm 2.6	122.0 \pm 1.7	91.0
PEG 8000	3.5	11.0 \pm 0	10.0 \pm 1.0	1.0
	7	37.3 \pm 0.6	26.7 \pm 2.3	10.7
	10.5	85.0 \pm 2.6	49.0 \pm 1.7	36.0
	14	149.3 \pm 2.1	82.7 \pm 2.3	66.7
PEG 20000	3.5	9.0 \pm 0	0	9.0
	7	38.7 \pm 2.1	4.0 \pm 1.7	34.7
	10.5	99.3 \pm 1.2	37.0 \pm 2.0	62.3
	14	223.7 \pm 1.2	93.3 \pm 1.5	130.3
PVP-K12	3.5	29.3 \pm 0.6	20.7 \pm 2.3	8.7
	7	68.3 \pm 0.6	29.0 \pm 1.0	39.3
	10.5	117.0 \pm 1.0	76.0 \pm 0	41.0
	14	183.0 \pm 1.0	129.0 \pm 1.7	54.0
PVP-K17	3.5	14.0 \pm 0	16.7 \pm 0.6	2.7
	7	36.0 \pm 1.0	34.7 \pm 1.2	1.3

	10.5	66.7 ± 0.6	59.3 ± 1.5	7.3
	14	108.0 ± 3.5	84.3 ± 1.2	23.7
PVP-K30	3.5	14.3 ± 1.2	3.7 ± 1.2	10.7
	7	36.0 ± 1.7	12.0 ± 1.7	24.0
	10.5	67.3 ± 1.5	20.7 ± 1.2	46.7
	14	110.3 ± 8.5	45.3 ± 0.6	65.0
Poloxamer 188	3.5	11.7 ± 0.6	1.7 ± 1.2	10.0
	7	42.3 ± 0.6	23.0 ± 1.0	19.3
	10.5	99.0 ± 1.7	41.0 ± 0	58.0
	14	184.7 ± 4.0	93.3 ± 2.9	91.3
Poloxamer 338	3.5	9.3 ± 0.6	0.7 ± 1.2	8.7
	7	37.0 ± 0	13.3 ± 0.6	23.7
	10.5	123.0 ± 3.0	37.0 ± 1.7	86.0
	14	234.0 ± 6.1	69.0 ± 1.0	165.0
Poloxamer 407	3.5	10.3 ± 0.6	0	10.3
	7	40.3 ± 2.1	0.3 ± 0.6	40.0
	10.5	98.3 ± 3.5	22.0 ± 1.7	76.3
	14	202.0 ± 2.6	42.0 ± 1.7	160.0
HPMC E5	3.5	16.7 ± 5.5	0	16.7
	7	37.3 ± 1.2	0	37.3
	10.5	72.3 ± 4.6	4.3 ± 1.2	68.0
	14	119.0 ± 5.2	59.3 ± 5.1	59.7
Na-CMC	1	19.0 ± 0	7.3 ± 0.6	11.7
	2	43.7 ± 0.6	32.7 ± 0.6	11.0
	3	58.7 ± 1.2	51.0 ± 1.7	7.7
	4	94.3 ± 0.6	88.3 ± 1.5	6.0

Table S4 - Overview of the surfactant solutions prepared with respective concentrations in percentage weight per volume (% w/v). Osmolality values were obtained by the two different osmometer principles: freezing point depression (FDP) and vapor pressure (VP).

Surfactant	Concentration (% w/v)	Osmolality (mOsm/kg)	Difference between FPD and VP measurement
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		FPD	VP	Δ
Polysorbate 20	1	1.0 ± 0	0	1.0
	2	4.0 ± 0	0	4.0
	4	10.7 ± 0.6	0	10.7
Polysorbate 80	1	0	0	0.0
	2	1.0 ± 0	0	1.0
	4	5.3 ± 0.6	0	5.3
Vitamin E-TPGS	1	0	0	1.0
	2	0	0	0.0
	4	2.0 ± 0	0	2.0
Lipoid E PG	1	-1.0 ± 0	0	1.0
	2	-1.0 ± 0	0	1.0
	4	0.7 ± 0.6	0	0.7
DOSS	0.5	8.0 ± 0	0.3 ± 0.6	7.7
	1	10.0 ± 0	0	10.0
	2	11.0 ± 0	3.3 ± 1.2	7.7
SDS	0.5	17.0 ± 0	9.3 ± 0.6	7.7
	1	21.0 ± 0	14.3 ± 1.5	6.7
	2	31.0 ± 0	21.3 ± 1.5	9.7

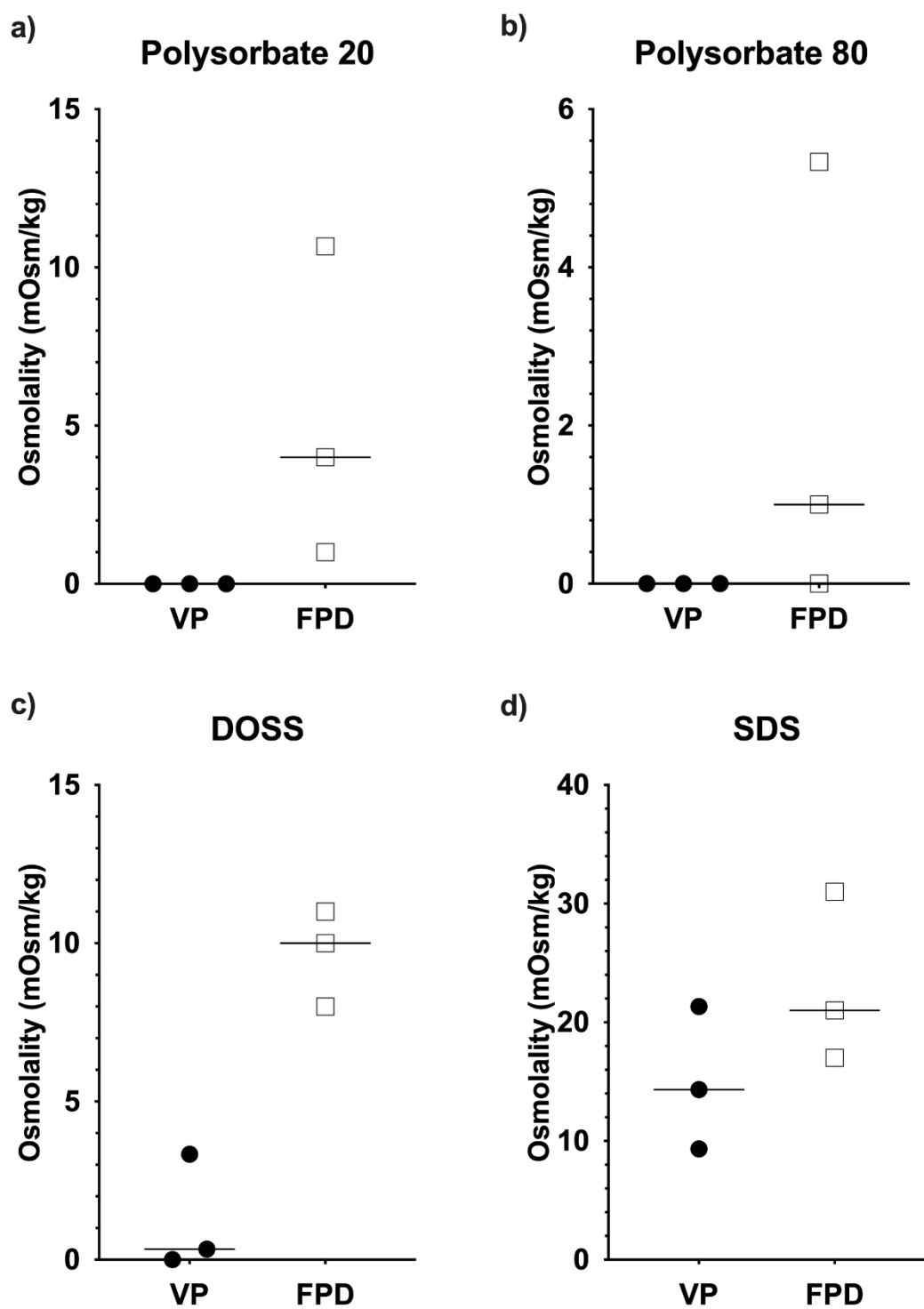


Fig. S2 - Graphs show the mean osmolality grouped as vapor pressure (VP) measurements or freezing point depression (FPD) measurements for three different excipients and its range of concentrations: a) polysorbate 20, b) polysorbate 80, c) DOSS, and d) SDS.

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