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ORIGINAL ARTICLE

Osmolality of preterm formulas supplemented with nonprotein energy supplements

L Pereira-da-Silva[1], M Pitta-Grós Dias[2,3], D Virella[1], AC Moreira[3] and M Serelha[1]

[1]*Neonatal Intensive Care Unit, Hospital Dona Estefânia, Faculdade de Ciências Médicas, Universidade Nova de Lisboa, Lisbon, Portugal;* [2]*Dietetic Service, Hospital Dona Estefânia, Lisbon, Portugal and* [3]*Escola Superior de Tecnologias da Saúde de Lisboa, Lisbon, Portugal*

Background: Addition of energy supplements to preterm formulas is an optional strategy to increase the energy intake in infants requiring fluid restriction, in conditions like bronchopulmonary dysplasia. This strategy may lead to an undesirable increase in osmolality of feeds, the maximum recommended safe limit being 400 mOsm/kg. The aim of the study was to measure the changes in osmolality of several commercialized preterm formulas after addition of glucose polymers and medium-chain triglycerides.
Methods: Osmolality was measured by the freezing point depression method. Six powdered formulas with concentrations of 14 g/100 ml and 16 g/100 ml, and five ready-to-feed liquid formulas were analyzed. All formulas, were supplemented with 10% (low supplementation) or 20% (high supplementation) of additional calories, respectively, in the form of glucose polymers and medium chain triglycerides, maintaining a 1:1 glucose:lipid calorie ratio. Inter-analysis and intra-analysis coefficients of variation of the measurements were always $< 3.9\%$.
Results: The mean osmolality (mOsm/kg) of the non-supplemented formulas varied between 268.5 and 315.3 mOsm/kg, increasing by 3–5% in low supplemented formulas, and by 6–10% in high supplemented formulas. None of the formulas analyzed exceeded 352.8 mOsm/kg.
Conclusion: The supplementation of preterm formulas with nonprotein energy supplements with up to 20% additional calories did not exceed the maximum recommended osmolality for neonatal feedings.
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Keywords: bronchopulmonary dysplasia; energy supplements; glucose polymers; medium-chain triglycerides; osmolality; preterm formulas

Introduction

The comprehensive management of infants with conditions such as bronchopulmonary dysplasia (BPD) and severe congenital heart disease usually require fluid restriction (Kurzner *et al.*, 1988; Tammela *et al.*, 1992).

In infants with BPD submitted to fluid restriction it may be difficult to meet the requirements to promote growth even by using standard preterm formulas containing a relatively high caloric density (Raffles *et al.*, 1983; Puangco and Schanler, 2000). The use of formulas with added nonprotein energy supplements, such as glucose polymers (GP) and/or medium-chain triglycerides (MCT), is a possible strategy to provide hypercaloric low-volume feedings (Raffles *et al.*, 1983; Thureen and Hay, 1993; Puangco and Schanler, 2000; Romera *et al.*, 2004). Supplementing standard formulas (Raffles *et al.*, 1983; Puangco and Schanler, 2000) or breast milk (De Curtis *et al.*, 1999; Fenton and Belik, 2002) by adding GP increases the osmolality of the feeds. Hyperosmolar feeds have been shown to empty from the stomach more slowly than isotonic solutions and are associated with an increased incidence of nausea, vomiting, diarrhea, and gastroesophageal reflux (Sutphen and Dillard, 1989; Salvia *et al.*, 2001). Considering the risks associated with hyperosmolar feedings, 400 mOsm/kg is the maximum

Correspondence: Dr L Pereira-da-Silva, Neonatal Intensive Care Unit, Faculdade de Ciências Médicas, Hospital Dona Estefânia, Universidade Nova de Lisboa, Rua Jacinta Marto, Lisbon 1169-045, Portugal.
E-mail: l.pereira.silva@netcabo.pt
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recommended safe osmolality for infant formulas (AAP CON, 1976).

Research supports many guidelines for nutritional supplemental management of infants who are growing poorly; however, some are still based on clinical practice and on undocumented methods (Reimers *et al.*, 1992; Fewtrell and Lucas, 2002). Hence, the osmolality of the feedings that are being manipulated should be determined and those caring for sick neonates should have an easy access to the measured values to prevent risks related to hyperosmolality.

Several preterm formulas have labels that do not provide information on osmolality, and some only provide the calculated osmolarity values that are lower than the measured osmolalities. On the other hand, it is difficult to compare the osmolality of formulas of similar type if their manufacturers use different methods of osmometry.

In the case of powdered formulas, the osmolality theoretically changes proportionally to the reconstitution concentration. It is also assumed that the addition of GP to the formulas alters the osmolality according to their concentration and their molecular weight by contrast the influence of MCT is very low. Although the change in osmolality by addition of GP may be calculated mathematically (Anderson and Kennedy, 1986), it is not certain to what extent osmolality is changed with the simultaneous addition of GP and MCT.

The aim of this study was to measure the osmolality of several commercialized preterm formulas at different concentrations by using the same osmometry method and to measure the change in their osmolality after the addition of GP and MC, thereby contributing to a better knowledge of this method of manipulating formulas.

Methods

Six powdered preterm formulas, Aptamil Pre (Milupa, Friedrichsdorf, Germany), Enfamil Preterm Formula (Mead-Johnson, Nijmegen, Holland), Nenatal (Nutricia, Zoetermeer, Holland), Nutribén Bajo Peso (Alter, Madrid, Spain), Pre Nan (Nestlé, Frankfurt, Germany) and S26 Gold LBW (Wyeth, Georgia, Vermont, USA); and five liquid ready-to-feed preterm formulas, Aptamil Pre (Milupa, Friedrichsdorf, Germany), Humana 0 (Humana, Herford, Germany), Nenatal (Nutricia, Zoetermeer, Holland), Pre Nan (Nestlé, Frankfurt, Germany) and Similac Special Care Advance (Abbott, Ross, Columbus, OH, USA), were included in the study.

According to the manufacturers' specifications, the concentration of the reconstituted powdered formulas need to range from about 14 g/100 ml (14%) in more dilute preparations, to about 16 g/100 ml (16%) in full-strength preparations. Therefore, the powdered formulas were reconstituted at 14 and 16%. All the powdered formulas and liquid formulas were supplemented with either 10% (low supplementation; LS) or 20% (high supplementation; HS) of calories using powdered GP as maltodextrin (Moducal, Mead-Johnson, Vansville, IN, USA; 1 g = 0.95 g maltodextrin) and MCT (MCT oil Module, SHS, Liverpool, UK; 1 ml = 0.95 g MCT), always maintaining a 1:1 glucoselipid calorie ratio.

A Kern 440-43N scale (Kern & Sohn GmbH, Balingen, Germany) with resolution of 0.1 g was used to weigh the amounts of powdered formulas and of GP. To reduce errors in dilution, 500 ml was the volume chosen for the preparation of powdered formulas and supplemented liquid formulas, and a mixer was used to homogenize the solutions during preparation. Both the powdered and supplemented formulas were prepared by the same investigator (MP-GD). An automatic pipette was used to measure the volumes of MCT to be added to the formulas, and to collect samples of prepared formulas for osmolality measurement.

Energy and protein contents of the formulas

Table 1 shows energy density and protein-to-energy (P:E) ratio of the formulas as well as energy and protein provided by the formulas calculated for daily fluid intakes of 130 ml/kg (fluid restriction) and 150 ml/kg (regular fluid intake). The non-supplemented preterm formulas contain mean energy

Table 1 Energy density, P:E ratio, and energy and protein provided by the analyzed formulas

Formulas	*Energy density (kcal/ 100 ml)*	*P:E ratio (g/100 kcal)*	*Energy intake (kcal/kg/day)*		*Protein intake (g/kg/day)*	
			130 ml/kg/day	*150 ml/kg/day*	*130 ml/kg/day*	*150 ml/kg/day*
	Mean (s.d.)	*Mean (s.d.)*	*Mean (s.d.)*	*Mean (s.d.)*	*Mean (s.d.)*	*Mean (s.d.)*
14%	71.1 (2.2)	3.0 (0.4)	92.5 (7.9)	106.7 (9.1)	2.7 (0.2)	3.2 (0.3)
LS 14%	78.4 (1.9)	2.7 (0.2)	101.9 (2.4)	117.6 (2.8)	2.7 (0.2)	3.2 (0.3)
HS 14%	85.8 (2.0)	2.4 (0.2)	111.6 (2.6)	128.8 (3.1)	2.7 (0.2)	3.2 (0.3)
16%	81.3 (1.9)	3.0 (0.2)	105.7 (2.4)	121.9 (2.8)	3.1 (0.2)	3.6 (0.3)
LS 16%	89.6 (2.1)	2.7 (0.2)	116.4 (2.8)	134.4 (3.2)	3.1 (0.3)	3.6 (0.3)
HS 16%	98.1 (2.3)	2.4 (0.2)	127.5 (3.0)	147.2 (3.5)	3.1 (0.3)	3.6 (0.3)
Liquid	79.3 (2.5)	2.9 (0.1)	103.1 (3.2)	118.9 (3.7)	3.0 (0.2)	3.5 (0.3)
LS liquid	87.6 (2.7)	2.6 (0.1)	113.9 (3.5)	131.4 (4.1)	3.0 (0.2)	3.5 (0.3)
HS liquid	96.0 (3.0)	2.4 (0.1)	124.7 (3.9)	143.9 (4.5)	3.0 (0.2)	3.5 (0.3)

HS, high supplementation; LS, low supplementation; P:E ratio, protein-to-energy ratio.

densities ranging from 71 to 81 kcal/100 ml, providing mean daily energy intakes from 107 to 123 kcal/kg with regular fluid intake, and only 92.5 to 105.7 kcal/kg in case of fluid restriction. The formulas with added energy supplements contain mean energy densities from 78.4 to 98.1 kcal/100 ml, providing the minimum recommended 110 kcal/kg/day for premature infants (Klein, 2002) at both fluid intakes, except for the LS 14% formulas in the case of fluid restriction. The HS formulas contain less than the minimum 2.5 recommended P:E ratio for preterm infants formulas (Klein, 2002), and HS 16% formulas and HS liquid formulas may provide more than the recommended 135 kcal/kg/day for premature infants (Klein, 2002).

Osmometry

Using the previously reported methodology (Pereira-da-Silva *et al.*, 2004), osmolality was measured by freezing point depression using an Osmomat 030 (Gonotec GmbH, Berlin, Germany) automatic cryoscopic osmometer. This osmometer is programmed to sample volumes of 50 μl with reproducibility $< \pm 2\%$. After every 30 measurements, the osmometer was calibrated, using standard solutions. Three samples of all analyzed formulas were measured in triplicate and measurements were compared to determine intraassay and interassay coefficients of variation. All the samples were blindly measured by the same investigator (LPdS.). Inter-analysis and intra-analysis coefficients of variation of measurements were always $<$ 3.9%.

Statistical analysis

Statistical analysis was performed with Microsoft Excel 2000™ and SPSS 6.1.3™ (SPSS Inc., Chicago, IL, USA) statistical packages. The osmolality was described using mean and standard deviation and 95% confidence interval of the mean (for graphic analysis). The daily energy intakes, daily protein intakes, and the P:E ratio were described using mean and standard deviation. Statistical difference on the osmolality of formulas after addition of energy supplements was analyzed using the Student's *t*-test, to compare the means of pairs of supplemented formulas, and Kruskal–Wallis one-way ANOVA, for analysis of formulas (14%, 16% and liquid formulas) according to energy supplementation (no added energy supplements, LS and HS). The usual rule for statistic significance was used ($P<0.05$).

Results

The mean osmolality (mOsm/kg) of the analyzed non-supplemented formulas was 268.5 for 14% formulas, 305.8 for 16% formulas, and 315.3 for liquid formulas (Table 2).

With LS, the osmolality increased by 3% in 14% formulas (mean; 277.5 mOsm/kg) (NS), 5% in 16% formulas (mean; 322.1 mOsm/kg) ($P=0.056$), and 3% in liquid formulas (mean; 325.6 mOsm/kg) (NS) (Figure 1).

With HS, the osmolality increased by 6% in 14% formulas (mean; 283.4 mOsm/kg) ($P=0.003$), 10% in 16% formulas (mean; 336.2 mOsm/kg) ($P=0.000$), and 7% in liquid formulas (mean; 336.2 mOsm/kg) ($P=0.01$) (Figure 1).

None of the formulas analyzed exceeded 352.8 mOsm/kg.

Table 2 Osmolality of the analyzed formulas. None of the formulas exceeded 352.8 mOsm/kg

Formulae		*Osmolality (mOsm/kg)*		
		Non-supplemented	*LS*	*HS*
14%	Mean	268.5	277.5	283.4
	(s.d.)	(19.9)	(21.0)	(18.8)
16%	Mean	305.8	322.1	336.2
	(s.d.)	(18.7)	(22.2)	(20.7)
Liquid	Mean	315.3	325.6	336.2
	(s.d.)	(17.3)	(18.8)	(16.3)

HS, high supplementation; LS, low supplementation.

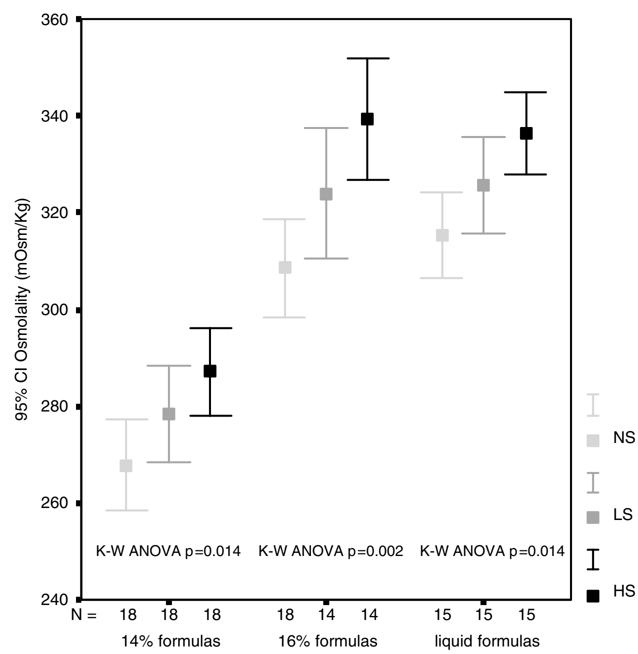

Figure 1 Osmolality of the analyzed formulas. None of the formulas exceeded the maximum 400 mOsm/kg recommended for infant formulas. K–W: Kruskal–Wallis one-way ANOVA analysis among energy supplements, for each formula. NS, no supplementation; LS, low supplementation; HS, high supplementation.

Discussion

The mean osmolality of the analyzed non-supplemented and supplemented formulas varied between 268.5 and 336.2 mOsm/kg. By adding energy supplements in the form of GP and MCT the osmolality increased by 3–5% in LS formulas and 6–10% in HS formulas. Although addition of energy supplements resulted in a significant increase in the osmolality of the HS formulas, none of the formulas exceeded 352.8 mOsm/kg (Figure 1). Until evidence-based data are made available, the Committee on Nutrition of the American Academy of Pediatrics has recommended that infant formulas have concentrations no greater than 400 mOsm/kg (AAP CON, 1976). To the best of our knowledge, no further evidence-based data has as yet been published on this subject.

The rationale for measuring the osmolalities of the preterm formulas at different concentrations, without and with nonprotein energy supplements, was to provide clinicians with evidence of a wide range of energy and protein intakes and respective P:E ratios, in infants managed with regular fluid intake or with fluid restriction (Table 1), without exceeding the maximum recommended safe osmolality of neonatal feedings (Table 2).

According to more recent recommendations (Klein, 2002), preterm formulas should provide a daily energy intake of 110–135 kcal/kg for appropriate catch-up growth in premature infants. This is easily achieved with fluid intakes of approximately 180 ml/kg/day (Kashyap *et al.*, 2001). The analyzed non-supplemented preterm formulas contain between 71 and 81 kcal/100 ml, providing daily mean energy intakes between 107 and 122 kcal/kg with fluid intake of 150 ml/kg/day, but only 92.5–105.7 kcal/kg if fluid intake is restricted to 130 ml/kg/day. This suboptimal energy intake is a major factor contributing to growth impairment and alterations in body composition in infants with BPD (Kurzner *et al.*, 1988; Tammela *et al.*, 1992; Huysman *et al.*, 2003). About 82–91 kcal/100 ml may be necessary for lung repair and adequate growth in infants with BPD, but approximately 98 kcal/100 ml may be needed when fluid intake is restricted to $\leqslant$130 ml/kg/day (Puangco and Schanler, 2000). This represents a significant additional energy requirement above that delivered by human milk or by most of standard preterm formulas (Thureen and Hay, 1993; Puangco and Schanler, 2000).

When breast milk is not sufficient, hypercaloric low-volume feedings may be provided by simply concentrating powdered preterm formulas above the concentration recommended by the manufacturers. Concentrating powdered formulas by reducing the amount of added water increases the level of all macro and micronutrients and results in a more balanced formulation. Once the maximum levels of limiting nutrients may be reached using this method, energy modules, either carbohydrate or fat, may be added to the powdered formulas at standard concentrations or to the ready-to-feed liquid formulas, to increase further energy alone (Romera *et al.*, 2004; O'Connor and Brennan, 2006).

The practice of simply concentrating standard formulas may involve several risks, including increased incidence of nausea, vomiting, diarrhea, and gastroesophageal reflux due to high osmolality feedings (Sutphen and Dillard, 1989; Salvia *et al.*, 2001), nitrogen and other solute overload and insufficient water for growth (Pereira *et al.*, 1994), hypertonic dehydration by exceeding the potential renal solute load (Ziegler and Fomon, 1989), and excessive mineral intake predisposing to nephrocalcinosis (Puangco and Schanler, 2000).

Addition of modular nutrient components to standard formulas is an alternative to simply concentrating preterm formulas (Raffles *et al.*, 1983; Thureen and Hay, 1993; Puangco and Schanler, 2000; Romera *et al.*, 2004). Glucose polymers are preferred as modular supplement because they are rapidly cleared from the stomach and absorbed in neonates (Costalos *et al.*, 1980; Klenoff-Brumberg and Genen, 2003). Medium-chain triglycerides are also easily accessible to the immature digestive system (Brooke, 1983). Considering the theoretical risk of excessive carbon dioxide production by solely adding GP to the formulas in patients with chronic lung disease, MCT were also added at appropriate glucose:lipid calorie ratio in the present study, since fat has a lower respiratory quotient and may therefore help offset this potential problem while still providing sufficient energy (Thureen and Hay, 1993; Pereira *et al.*, 1994; Romera *et al.*, 2004). Nevertheless, it should be emphasized that addition of nonprotein supplements to standard formulas may compromise their nutrient integrity by changing the optimal calorie-to-nitrogen ratio (Brooke, 1983; Puangco and Schanler, 2000). The analyzed formulas contain a P:E ratio between 2.4 and 3.0 g/100 kcal (Table 1), in the lower range of the current recommendations for preterm infants, between 2.5 and 3.6 g/100 kcal (Klein, 2002). However, recent evidence based on short-term outcomes suggests that formulas designed for rapidly growing very low-birth-weight infants should contain a P:E ratio as high as 3.3–3.6 g/100 kcal (Cooke *et al.*, 2006; Rigo and Senterre, 2006). For long-term outcomes lower protein densities appear adequate, but further studies are needed to define the optimal protein concentration in these cases (Cooke *et al.*, 2006).

To summarize, several preterm formulas were analyzed to evaluate the modification of feeding osmolality using formula concentration or the addition of nonprotein energy supplements thereby contributing to a better knowledge of this method. Enriching preterm formulas with GP and MCT up to 20% additional calories may be a possible strategy for increasing energy intake in infants requiring fluid restriction without exceeding the maximum recommended osmolality safe for neonatal feedings. As enrichment of formulas with nonprotein supplements may compromise the optimal calorie-to-nitrogen ratio, the better preterm formula should be judiciously chosen before using this method.

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