Dietary Fat Composition Alters Pulmonary Function in Pigs

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OBJECTIVES: We investigated the effect of various dietary fats on pulmonary surfactant composition and lung function changes that occur before and after endotoxin infusion in pigs.

METHODS: Eighteen pigs were assigned to three groups ($n = 6$ per group) to receive a diet of protein (20% of calories), carbohydrate (20% of calories), and fat (40% of calories). In one group the fat content consisted entirely of palmitic acid. In the second group, fat came from Intralipid, which provided predominantly linoleic acid. The third group was fed fish oil. Pigs were maintained on these diets for 21 d before the experiment. Cardiovascular and pulmonary functions were determined on day 22. Pigs then were infused with endotoxin (80 $mg \cdot kg^{-1} \cdot min^{-1}$) until the pulmonary arterial pressure reached a pressure similar to that found in trauma victims (45 to 50 mmHg). Cardiovascular and pulmonary function tests were then repeated, the animals killed, and the lungs removed for study.

RESULTS: Compliance was reduced in the linoleate and fish-oil groups compared with the palmitate group before and after endotoxin. Compliance changes in pigs fed the linoleate and fish-oil diets were consistent with significant increases in lung wet:dry weight ratios, increased CO_2 retention, histologic evidence of vascular congestion, intra-alveolar edema, and alveolar septa thickening. Changes in surfactant phosphatidylcholine composition between groups were consistent with the notion that increased unsaturated fatty acids could affect surfactant function.

CONCLUSIONS: We concluded that the common practice of providing calories in the form of polyunsaturated fatty acids to critically ill patients carries the risk of being detrimental to lung function. *Nutrition* 2002;18:647–653. ©Elsevier Science Inc. 2002

KEY WORDS: surfactant, lung compliance, dietary fatty acids

INTRODUCTION

Lung dysfunction is an important cause of morbidity and mortality in critically ill patients. A deficiency or change in the composition of the lung surfactant complex may contribute to this response. Pulmonary surfactant plays an important physiologic role by decreasing alveolar surface tension and in keeping the lungs dry. Clinical circumstances in which impaired lung function is common (e.g., sepsis and acute pancreatitis) are associated with decreased surfactant production.[1–3] Further, changes in the composition of the surfactant complex can also affect lung function. Dipalmitoylphosphatidylcholine is the most active phospholipid in this complex. The substitution of unsaturated fatty acids such as oleate or linoleate can affect the surface tension–lowering capacity of phosphatidylcholine (PC) and lung compliance.[4] The most well-recognized circumstance in which altered composition is related to respiratory distress is in premature infants who have a reduced proportion of palmitate in the lung surfactant PC.[5] Similar changes in pulmonary surfactant composition also have been reported in patients with acute respiratory distress syndrome.[6] Substitution of unsaturated fatty acids for saturated fatty acids can decrease the surface tension–lowering capacity of surfactant and its ability to keep the lungs dry.[7,8] Consequently, the amount and composition of surfactant can potentially affect lung function.

Under normal circumstances there is sufficient reserve of surfactant in the lungs so that perturbations such as normal stress or dietary manipulations that might affect surfactant quantity or composition will have a minimal effect on lung function. However, in a clinical circumstance in which the lung reserves are compromised, a deficiency in the amount or composition of surfactant might have devastating results. For example, endotoxin injection caused an almost immediate increase in the mean pulmonary artery pressure, microvascular permeability,[9] and lung compliance.[10] Thus, a change in lung surfactant superimposed on an already-compromised lung function likely would become clinically significant.

We recently reported that the plasma free fatty acids (FFAs) are the principal precursors for synthesis of pulmonary PC,[11] even when peripheral lipolysis is suppressed by high rates of glucose intake.[12] It could, therefore, be anticipated that dietary manipulations that alter the fatty acid profile in plasma also might cause corresponding changes in lung PC composition. If true, this could have significant clinical implications because conventional nutrition in critically ill patients often includes a lipid-based emulsion derived from soy beans in which over half the fatty acids are linoleate (18:2), and nutrition support with this formulation causes the proportion of total plasma FFAs represented by linoleate to increase.[13] Thus, nutrition support of critically ill patients with a lipid-based system could decrease the proportional contribution of palmitate to lung surfactant PC, thereby inadvertently contributing

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to a deficiency in lung surfactant function. In contrast, the deacylation–reacylation pathway in PC synthesis reactions enable the lung surfactant PC composition to be markedly different from the plasma FFA composition under normal conditions,[14] so it is not certain that changes in the plasma FFA profile would be directly reflected in the composition of lung surfactant PC.

We performed the following experiment to test the hypothesis that changes in the dietary fatty acid composition will be reflected in the composition of pulmonary PC, and that these changes will be reflected in altering lung dynamic compliance, lung water content, and gas exchange. Our initial interest was in the comparison of diets containing predominantly palmitate or linoleate because 1) the unsaturated linoleate would be predicted to have a deleterious effect on surfactant effacy as compared with palmitate, and 2) conventional nutrition support of critically ill patients often includes a lipid-based formulation comprised of a high proportion of linoleate. However, in addition to affecting surfactant composition, linoleate might affect lung function through other mechanisms stemming from the metabolism of linoleate (ω-6 fatty acid). Therefore, a third group was included in which a caloric-equivalent amount of fish oil was given instead of palmitate or linoleate. Fish oil contains a mixture of long-chain (ω-3) polyunsaturated fatty acids that in sum have been reported to be immune enhancers.[15] Thus, if comparable effects of linoleate and fish oil are observed, it would be reasonable support for the notion that, with regard to surfactant function, the extent of saturation is the primary difference between linoleate and fats in fish oil, on the one hand, and palmitate, on the other hand. We studied the dietary effects in animals subject to endotoxin infusion to evaluate the response in animals in which the physiologic reserve in lung function was been challenged so that alterations in the surfactant complex would more likely result in physiologic changes.

RESEARCH AND PROCEDURES

Eighteen Yorkshire swine (K-Bar Live Stock, Sabinal, TX, USA) were used for the experiment. The study was approved by the University of Texas Medical Branch Committee on Animal Care and Use (ACUC 97-10-046). The animals were kept individually separated in the animal research center and acclimated for 10 d before starting experiments.

Diets

The animals were randomly assigned to three groups to be given diets with different fatty acid compositions. The diets contained the same amount of protein (70 g/d) and total daily caloric intake (1450 kcal/d) as the standard diet (Minipig Grower Diet, Purina, DMI Nutrition International, Inc., Brentwood, MO), but the proportion of fat was increased to supply 40% of the total caloric intake (65 g fat/d). The remaining balance of the diet consisted of protein (20% of calories) and carbohydrate (148 g/d, 40% of calories). Ten percent of the total caloric intake came from Purina standard diets to provide various vitamins, essential fatty acids, fiber, and minerals. The remaining caloric intake came from Pro-MOD (protein supplement), Polycose (carbohydrate supplement), palmitic acid, Intralipid (linoleic-rich supplement), or fish oil. Butylhydroxyltoluene (0.3 g/d) was used as a food preservative. All diets were made in a weekly supply and refrigerated. After baseline blood samples were taken, the animals were fed twice a day with the assigned diet for 3 wk.

Analysis of the lipid component in the diets by gas chromatography (Hewlett Packard 5890, Palo Alto, CA) showed that the palmitate diet was 98.4% palmitate and 0.2% linoleate, the linoleate diet was 16% palmitate and 61% linoleate (ω-6, 18:2), and the fish oil diet was 38% saturated and 62% unsaturated fatty acids, with 38% of the fatty acids being ω-3 (18:4, 20:5, 22:5, 22:6) and 2% being ω-6 (18:2 + 20:4).

Study Protocol

The animals were fed the assigned diets for 21 d and the night before the study and then allowed only water until the following morning, when they were given ketamine anesthesia (20 mg/kg, 1 M; Ketaset, Fort Dodge Laboratories, Fort Dodge, IA, USA) for catheter placement. A Swan Ganz catheter was inserted via the jugular vein and advanced through the heart into the pulmonary artery for pulmonary artery and pulmonary capillary wedge pressures. A second catheter was inserted via the right common carotid artery into the abdominal aorta for blood sampling and monitoring blood pressure. The catheters were externalized at the back of the neck for access during the experiment. The animals were placed in a sling and allowed to awaken from the surgical procedure. Then a pulmonary function test was performed under a smaller dose of ketamine (Abbott Laboratories, N. Chicago, IL).

After completion of the initial pulmonary function test and measurement of cardiovascular parameters, endotoxin was infused at the rate of 80 ng $\cdot$ kg^{-1} $\cdot$ min^{-1} to induce a vascular pressure stress on the lungs. The endotoxin was infused until the pulmonary artery pressure increased from the basal value of approximately 15 mmHg to 45 to 50 mmHg. This required from 18 to 24 min, with a similar average time required in all groups. When the pulmonary artery pressure reached approximately 50 mmHg, the endotoxin infusion was stopped and the animals again were infused with ketamine to induce mild anesthesia. Cardiovascular measurements were repeated and pulmonary function again determined. After completion of the pulmonary function test, the animals were killed with ketamine and saturated potassium chloride. The lungs were quickly removed, weighed, and cooled for lavage and collecting tissue samples.

Four animals (two with palmitate and two with linoleate) served as controls for the histology examination of lung tissues. These animals were treated as described above for dietary control but killed before being given endotoxin, and samples of the lungs were prepared for histologic examination.

Pulmonary Function Test

A Smart Cath esophageal multifunction balloon was placed in the lower one-third of the esophagus to monitor the esophageal pressure continuously. A transducer was placed into the trachea to measure the air flow during the spontaneous breath. The transducer and the balloon catheter were connected to the CP-100 pulmonary monitor (Bicore Monitoring Systems, Irvine, CA, USA). Based on the measurements of esophageal pressure and airway flow, the dynamic compliance was calculated by the pulmonary monitor according to the formula

$$C_{dyn} = V_t/(P_{tp}max - P_{tp}min)$$

where C_{dyn} is the dynamic compliance, V_t is the tidal volume, $P_{tp}max$ is the transpulmonary pressure at maximum volume and zero flow, and $P_{tp}min$ is the transpulmonary pressure at minimum volume and zero flow.

Sample Analysis: Bronchoalveolar Lavage

The right lung was isolated, weighed, vacuum degassed, and suspended in a Plexiglas vacuum chamber via the trachea cannula, which passed through the top of the chamber to two 150-mL syringes connected by a three-way valve. A subatmospheric chamber pressure was externally applied to the lungs, and the lungs were filled with fluid consisting of normal saline plus 0.01 M Tris buffer. This procedure ensured that the lungs were fully filled with fluid and that any residual CO_2 was absorbed without the introduction of air bubbles. Lavage fluid was withdrawn from the lungs into a syringe by applying a positive chamber pressure to the lungs with gentle suction. This process was repeated three times and as

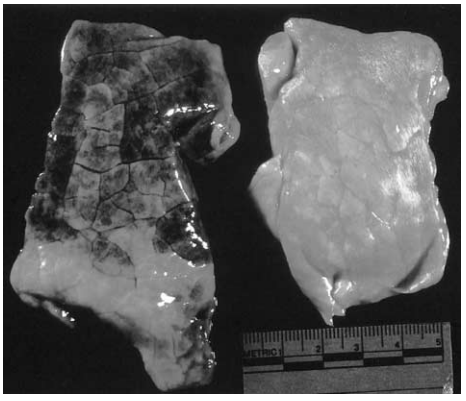

FIG. 1. Gross appearance of lungs from animals treated with palmitate (right) and linoleic acid (left). Lungs from the fish-oil group appeared similar to those from the linoleate group.

much bronchoalveolar lavage was collected as possible with positive external pressure; care was taken to not introduce any air bubbles. The entire procedure was repeated six times, each time starting with saline plus Tris buffer.

Histology

Blocks of tissue were excised from the lungs and fixed by immersion in 10% buffered formalin for 2 to 24 h. The tissue was dehydrated in ethanol and embedded in paraffin, and histologic sections were prepared at the thickness of 6 μm. Sections were stained with hematoxylin and eosin. Coded slides were examined by a pathologist, and scores were assigned for congestion, hemorrhage, edema, infiltration by neutrophils, and thickening of alveolar septa, without knowledge of the experimental group.

Analysis of Fatty Acids

The fatty acid composition of PC from the lamellar bodies (site of production of surfactant) and bronchoalveolar lavage was determined as described previously.[12] Briefly, lamellar body isolation involved isolation from tissue homogenates with the use of a discontinuous sucrose density gradient and ultracentrifugation.[16] Isolation of surfactant from bronchoalveolar lavage was accomplished by ultracentrifugation.[11] PC was isolated by extraction and separation by thin layer chromatography.[11] The composition of fatty acids from the resultant PC was measured by high-performance liquid chromatography, as described previously.[11] Plasma FFA concentrations also were measured by high-performance liquid chromatography.

Statistics

Statistical comparisons between groups was made with analysis of variance and Tukey's post hoc test. The unpaired *t* test was used to test differences between pre- and postendotoxin experiments. Significance was accepted at $P < 0.05$.

RESULTS

Gross inspection of the lungs of animals treated with endotoxin showed a hyperemic appearance in pigs fed linoleate or fish oil and a normal appearance in pigs fed a diet rich in palmitate (Fig. 1). This gross appearance was consistent with the wet:dry weight ratios, which were significantly elevated in the linoleate and fish-oil groups (mean $\pm$ standard error of the mean: 5.55 $\pm$ 0.5 and 5.23 $\pm$ 0.03 in the linoleate and fish oil groups, respectively, versus the palmitate value of 4.98 $\pm$ 0.05; Table IV)). Microscopic sections from the lungs of animals given linoleate or fish oil showed more congestion after endotoxin treatment than did those from the palmitate group, i.e., the pulmonary capillaries appeared engorged by red blood cells (Fig. 2). Focal edema, identified as pale eosinophilic fluid within groups of alveoli, was seen in the lungs of animals fed linoleate or fish oil after endotoxin challenge. A few polymorphonuclear neutrophils were seen focally in the interstitium of the lung in all animals fed fish oil pre- and postendotoxin, but in no other group. No hyaline membranes could be identified. Histology slides were scored on a scale of 0 to 4, with 4 being highly abnormal. They were scored according to the presence of congestion, edema, polymorphonuclear neutrophils, and thickened alveolar septa. The scores for congestion and edema were averaged. Before endotoxin, average scores for congestion and hemorrhage, edema, polymorphonuclear neutrophils, and alveolar thickening were 0.5, 0, 0, and 0.5 for palmitate; 1.0, 0, 0, and 0 for linoleate; and 0, 0, 1.0, and 1.0 for fish oil. After endotoxin, average scores for congestion plus hemorrhage were 1.0, 1.5, and 2.0 for palmitate, linoleate, and fish oil, respectively, and edema scores were 0, 1.0, and 2.0, respectively. The scores for extravascular polymorphonuclear neutrophils were 0 in the lungs of pigs fed palmitate or linoleate and 1.0 in the lungs of pigs fed fish oil. Alveolar septa thickening scores were 0.5, 1.5, and 1.0 for pigs fed palmitate, linoleate, and fish oil, respectively.

Pretreatment with the linoleate or fish-oil diets caused a significant reduction in the mean pulmonary artery pressure when compared with palmitate. The fish-oil group also had a lower mean arterial blood pressure before endotoxin infusion than either of the other groups. Cardiac output, body temperature, and wedge pressures were similar between groups. In all groups the mean pulmonary artery pressure increased to nearly 50 mmHg after endotoxin (Table I). There were modest increases in all groups in mean arterial pressure, wedge pressure, and cardiac output after endotoxin, but the only difference between groups was that the wedge pressure was lower in the linoleate group than in the palmitate group.

The proportionate distribution of individual fatty acids in the plasma FFA was similar between groups before the controlled feedings began (Table II). After the feeding, the palmitate group had a significant increase in the proportion of palmitate, from 27.5 $\pm$ 0.6% to 34.8 $\pm$ 1.7%, but the proportion of linoleate did not change. Feeding with the linoleate-rich diet caused the proportion of linoleate in plasma FFA to increase from 14.9 $\pm$ 0.9% to 42.7 $\pm$ 2.8% and also caused the proportion of palmitate to fall significantly. The proportion of palmitate did not change significantly in the fish-oil group, but the linoleate composition fell to only 6.2 $\pm$ 0.3%. The proportions of eicosapentaenoate (20:5) and docosahexaenoate (22:6) increased from the initial values of 1.3 $\pm$ 0.2% and 2.5 $\pm$ 0.5%, respectively, to 15.5 $\pm$ 1% and 14.7 $\pm$ 1.3%, respectively, after treatment. The palmitate diet caused no significant shift in the percentage of saturated or unsaturated fatty acids in plasma, whereas the linoleate diet caused an 18.5% increase in the percentage of unsaturated fatty acids and the fish-oil diet caused a 12.3% increase in the percentage of unsaturated fatty acids.

Changes in the plasma profile of fatty acids were reflected in corresponding changes in lung surfactant PC fatty acid composition, but the magnitude of changes was greatly blunted (Table III). The linoleate group had a significantly greater proportion of linoleate than did the other groups, and the fish oil group had significantly less linoleate than the palmitate group, but the contributions of eicosapentaenoate and docosahexaenoate increased from undetectable to values between 0.5% and 3%, respectively (Table III). The palmitate composition did not differ significantly between groups, but the contribution of palmitate in the fish-oil group to the lavage surfactant PC dropped almost 10% below the that in the palmitate group.

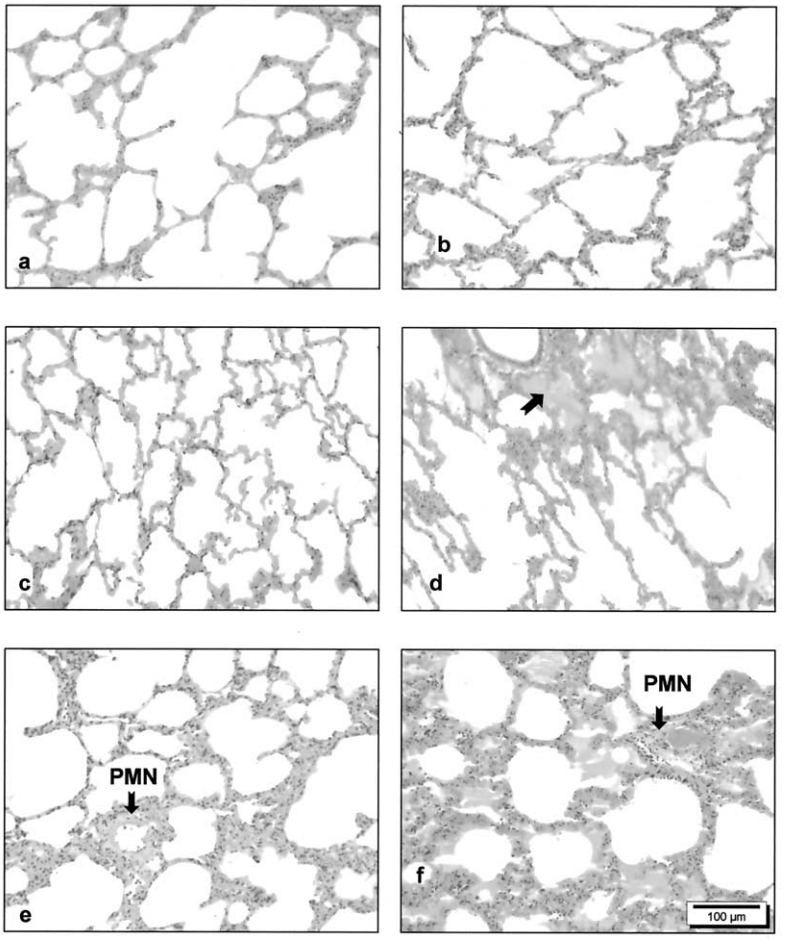

FIG. 2. (a) Histology of the lung of a pig fed a diet rich in palmitate (control group without endotoxin treatment). Alveolar septa are uniform and relatively delicate, although there is slight congestion. Hematoxylin and eosin stain. (b) The lung of a pig fed a diet rich in palmitate and treated with endotoxin. The capillaries in the alveolar septa are filled with red blood cells. No inflammatory infiltrate or edema is seen. (c) The lung of a pig fed a diet rich in linoleate. There is some congestion, but no edema is seen. (d) The lung of a pig fed a diet rich in linoleate and treated with endotoxin. Vascular congestion is present, and there are focal accumulations of eosinophilic edema fluid (arrow). No significant inflammatory infiltrate is seen. (e) The lung of a pig fed a diet rich in fish oil. There is mild, variable widening of alveolar septa. No edema is seen. (f) The lung of a pig fed a diet rich in fish oil and treated with endotoxin. Vascular congestion and focal edema are present. There is also a diffuse mild infiltrate of PMNs (arrow). PMN, polymorphonuclear neutrophil.

Feeding with linoleate or fish oil caused lung compliance to fall before endotoxin infusion as compared with feeding with palmitate (Table IV). The compliance changes corresponded to changes in wet:dry weight ratios, which were significantly elevated in the linoleate and fish-oil groups (Table IV). Endotoxin infusion caused a significant fall in all groups, but the compliance fell to lower levels in the linoleate and fish-oil groups than in the palmitate group. Changes in arterial blood gases generally corresponded with the changes in lung function. Arterial partial pressure of CO_2 increased after endotoxin in the linoleate group, with little change in the palmitate group or the fish-oil group, but the pressure in the fish-oil group was significantly higher than in the palmitate group before and after endotoxin (Table V). The ratio of arterial partial pressure to the fractional inspired concentration decreased significantly below those in the other two groups before and after endotoxin (Table V).

TABLE I.

EFFECT OF ENDOTOXIN ON CARDIAC OUTPUT AND PULMONARY VASCULAR PRESSURES IN PIGS FED DIFFERENT DIETARY FATTY ACIDS*

	MPAP (mmHg)	Wedge (mmHg)	Cardiac output (L/min)
Palmitate			
Preendotoxin	20.0 ± 1.1†	7.8 ± 0.8†	3.6 ± 0.3
Postendotoxin	47.5 ± 0.8	15.8 ± 3.1	4.3 ± 0.6
Linoleate			
Preendotoxin	15.8 ± 1.0†	5.7 ± 0.9†	3.8 ± 0.1†
Postendotoxin	46.7 ± 0.2	8.8 ± 0.3	5.0 ± 0.1
Fish oil			
Preendotoxin	14.3 ± 1.3†	5.3 ± 1.6	4.0 ± 0.2†
Postendotoxin	47.0 ± 0.9	10.8 ± 2.5	4.8 ± 0.4

* Values are means ± standard error of the mean.
† Significant difference between pre- and postendotoxin at $P < 0.05$.
MPAP, mean pulmonary artery pressure.

DISCUSSION

The most important observation of this study was that diets rich in saturated fatty acid, as presented in Intralipid or fish oil, significantly impairs lung compliance compared with a diet rich in palmitate. Although the decrease in compliance was evident at

TABLE II.

DISTRIBUTION OF PLASMA FREE FATTY ACIDS*

	% Palmitate (16:0)	% Stearate (18.0)	% Oleate (18.1)	% Linoleate (18:2)	% Other unsaturated
Palmitate					
Prefed	27.5 ± 0.6	22.6 ± 1.9	25.7 ± 3.6	14.9 ± 1.2	9.3 ± 0.6
Postfed (21 d)	34.8 ± 1.7	19.3 ± 2.1	26.5 ± 1.4	14.8 ± 1.0	4.6 ± 0.5
Linoleate					
Prefed	26.7 ± 1.1	24.4 ± 1.5	25.4 ± 1.9	14.9 ± 0.9	8.6 ± 0.7
Postfed (21 d)	19.0 ± 1.1	13.6 ± 0.9	21.7 ± 2.5	42.7 ± 2.8	3.0 ± 0.4
Fish oil					
Prefed	24.2 ± 1.0	23.8 ± 3.3	24.7 ± 2.7	15.5 ± 0.7	11.8 ± 0.6
Postfed (21 d)	23.6 ± 7.0	12.1 ± 1.0	24.7 ± 1.4	6.2 ± 0.3	32.5 ± 1.4

* Values are means ± standard error of the mean.

rest, the compliance became evident as a functional deficiency when endotoxin stress was superimposed. After endotoxin, the animals fed linoleate or fish oil were less able than those fed palmitate to eliminate CO_2 as indicated by elevated values of arterial partial pressure of CO_2. The physiologic changes caused by the linoleate and fish-oil diets were consistent with gross and microscopic examinations and the corresponding changes in wet: dry weight ratios.

The rationale for our study was predicated on our previous observation that the plasma FFA pool was the principal source of fatty acids for lung PC synthesis,[11] so that changes in the plasma profile of fatty acids should be reflected in changes in PC composition. Further, a modest reduction in the palmitate component of PC can impair the surface tension–reducing capacity of surfactant.[6] In particular, substitution of longer chained, unsaturated fatty acids for palmitate is particularly deleterious to surfactant function.[6,17] To distinguish the effects of chain length and extent of saturation from the metabolic response to a specific fatty acid (e.g., linoleate), we compared two different sources of long-chain unsaturated fatty acids. Whereas modification of dietary intake of fatty acids greatly modified the profile of plasma FFA, changes in PC composition were relatively minor. This reflects a robust mechanism for maintaining surfactant PC composition. Nonetheless, there were significant changes in the relative abundances of different long-chain unsaturated fatty acids in the linoleate and fish-oil groups (Table III). It is unclear whether these modest changes in lung surfactant PC composition were enough to significantly alter surfactant function and thus lung compliance. The number of double bonds in the fatty acids in PC can directly affect surfactant function (e.g., four) and the increase in total double levels was similar in the linoleate and fish-oil groups. However, the physiologic significance of changes in composition as subtle as those observed in this study has not been assessed. However, the physiologic data (i.e., decreased compliance) and histologic data (i.e., edema within alveoli) were consistent with a change in surfactant function.[7,8] Further, the lack of apparent infiltration of inflammatory cells into the lungs of the linoleate group lends support to the notion that the observed change in compliance was due to changes in surfactant composition rather than to products of lipid peroxidation. In any case, there is little doubt that lung function was impaired when animals were given diets high in long-chain unsaturated fatty acids, and nutrition support with compounds such as Intralipid or fish oil that are rich in such fatty acids could lead to impaired lung function.

We used the endotoxin model in our experiments to create a situation in which the physiologic importance of dietary-induced changes in lung compliance might be amplified, as might be found in sepsis or trauma. Although endotoxin may not represent the best model of clinical sepsis, it is a useful model because of the well-established response of a rapid increase in mean arterial pressure and decrease in the compliance of the lungs.[10] Our findings are pertinent to any clinical circumstance in which lung compliance is compromised, which may include patients who are critically ill because of any number of reasons.[6,18–20] In such patients, nutrition support is routinely provided to promote immune function, wound healing, and ameliorate the loss of lean

TABLE III.

FATTY ACID COMPOSITION OF PHOSPHATIDYLCHOLINE IN THE LUNGS OF PIGS GIVEN DIETS HIGH IN PALMITATE, LINOLEATE OR FISH OIL*

	% Palmitate (16:0)	% Linoleate (18:2)	% Eicosapentaenoate (20:5)	% Docosahexdenoate (22:6)
Lamellar bodies				
Palmitate	64.2 ± 1.6	4.8 ± 0.2	<0.1	<0.1
Linoleate	60.8 ± 2.3	11.1 ± 1.3†	<0.1	<0.1
Fish oil	64.7 ± 2.4	1.5 ± 0.5	0.6 ± 0.1	3.0 ± 0.6
Lavage				
Palmitate	75.4 ± 1.9	3.7 ± 0.2	<0.1	<0.1
Linoleate	72.5 ± 1.1	8.4 ± 0.6†	<0.1	<0.1
Fish oil	66.9 ± 3.0	1.5 ± 0.5†	1.6 ± 0.8	2.9 ± 0.4

* Values are means ± standard error of the mean.

† $P < 0.05$ versus palmitate.

TABLE IV.

LUNG COMPLIANCE (ml/cm H_2O) AND WET:DRY WEIGHT RATIOS

Group	Pre-endotoxin	Postendotoxin	Wet:dry
Palmitate	111 ± 13	65 ± 7‡	4.98 ± 0.05
Linoleate	73 ± 11†	44 ± 5†‡	5.55 ± 0.05†
Fish oil	75 ± 7†	48 ± 5†‡	5.23 ± 0.03†§

* Values are means ± standard error of the mean.
† Significant difference versus palmitate at $P < 0.05$.
‡ Significantly lower versus pre-endotoxin at $P < 0.05$.
§ Significant difference versus linoleate at $P < 0.05$.

body mass.[21] Historically, in the 1970s, nutrition support generally was provided in the form of glucose and amino acids. However, a series of studies established that, for multiple reasons, high glucose intake results in excessive rates of CO_2 production and, hence, a greater work load for stressed lungs. There are several reasons for this. 1) The respiratory quotient of glucose is one, so a diet that promotes carbohydrate oxidation will result in a higher rate of CO_2 production than a diet in which a significant portion of the non-protein calories is provided as fat, which is oxidized with a respiratory quotient close to 0.7. 2) Metabolic rate is stimulated by excessive glucose intake, resulting in increased rates of oxygen consumption and CO_2 production.[22] 3) When carbohydrate is provided at rates well in excess of caloric requirements (which often occurred clinically), hepatic triacylglycerol synthesis is stimulated.[23] This process has a respiratory quotient greater than five,[24] so that the storage of the excess glucose results in extra CO_2 production. As a result, the practice of substituting fat in the diet for carbohydrate became commonplace, with the result that some patients could be weaned from ventilators because of decreases in arterial partial pressure of CO_2.[25]

In contrast to the reports of the beneficial effects of substituting fat for carbohydrate in the nutrition support of patients with impaired ventilatory capacity, recent evidence suggested that a diet high in fat causes clinical problems in certain patients. A recent meta-analysis indicated that, in critically ill patients, complications are more frequent when lipid emulsions are used.[26] In preterm neonates, there is a likelihood that use of intervenous lipid emulsions causes chronic neonatal lung disease.[27] Also, lipid emulsions

TABLE V.

BLOOD GASES AND Pa_{O_2}/Fi_{O_2} RATIOS*

Group	Pre-endotoxin	Postendotoxin
Pa_{CO_2} (mmHg)		
Palmitate	32.5 ± 2.1	34.5 ± 1.3
Linoleate	37.1 ± 1.2	46.5 ± 2.4†§
Fish oil	41.1 ± 0.7†	42.5 ± 2.5†
Pa_{O_2}/Fi_{O_2} ratios		
Palmitate	459 ± 14	384 ± 16
Linoleate	427 ± 12†‡	335 ± 8†‡§
Fish oil	455 ± 15	400 ± 21

* Values are means ± standard error of the mean.
† Significantly different from palmitate at $P > 0.05$.
‡ Significantly different from fish oil at $P > 0.05$
§ Significant difference between pre- and postendotoxin at $P > 0.05$.
Fi_{O_2}, fractional inspired concentration; Pa_{O_2}, arterial partial pressure

as part of total parenteral nutrition in rats caused pulmonary hypertension and microvascular injury compared with total parenteral nutrition including only amino acids and glucose.[28] Our data suggest that one cause of such problems stems from the specific nature of the fatty acids conventionally provided in nutrition support. Most formulations containing fat for nutrition support (i.e., Intralipid) are derived from soy beans, meaning that more than 50% of the fatty acid content is linoleate. Although this source of fat is ideal for preventing essential fatty acid deficiency, only about 1% of the dietary calories is needed from linoleate to accomplish that goal. Clinically, it is not unusual to provide as much as 40% of the diet in the form of Intralipid or a comparable product rich in linoleate. The current study demonstrated that such an approach carries the risk of exacerbating any pre-existent deficiency in pulmonary compliance directly stemming from the illness. In some patients, this could become clinically significant. In contrast, a pathophysiologic response would not be expected in all patients treated with linoleate-rich diets because, although lung compliance might be affected in all cases, if such a dietary-induced decrement occurred in a patient with adequate pulmonary reserve, it would not necessarily result in a decrease in blood gas exchange.

We also tested the response to fish oil because (like Intralipid) fish oil is rich in polyunsaturated fatty acids but contains largely ω-3 fatty acids as opposed to the ω-6 fatty acid linoleate that is predominant in Intralipid. Fish oil was included in nutritional interventions in patients with acute lung injury or acute respiratory distress syndrome based on the rationale that fatty acids (i.e., EPA) in fish oil decrease the synthesis of proinflammatory eicosanoids and increase the anti-inflammatory eicosanoids.[15,29] However, the similarity of impairment in pulmonary function of the linoleate and fish-oil groups in the current study supported the notion that the most important feature with regard to lung surfactant PC is that both diets contained predominantly polyunsaturated fatty acids. Consistent with our observation, Murry et al. also found no demonstrable differences in surfactant function and pulmonary compliance during endotoxemia in pigs fed with fish oil or corn oil (enriched with linoleic acids).[10] In contrast, Gadek et al. found that a fish-oil diet improves lung function in patients with acute respiratory distress syndrome.[30] However, those patients were given the experimental diets for several days after the onset of pulmonary distress, which was sufficient time for an effect on the inflammatory response to be reflected in improved pulmonary function. In our study, there was insufficient time after endotoxin infusion (approximately 2 min) for differences in the inflammatory response to affect lung compliance. Also, the fish-oil diet given to the patients with acute respiratory distress syndrome contained amounts of antioxidants, and the antioxidants may have been the reason for the improved lung function in patients given polyunsaturated fatty acids.

Given the potential problems excessive carbohydrate can cause in patients with impaired lung function and the potential problems demonstrated in this study resulting from the administration of a significant amount of long-chain polyunsaturated fatty acids, the challenge arises as to the optimal approach to nutrition support in patients at risk for the development of pulmonary problems. Our results indicated that nutrition support should be aimed at minimizing the availability of unsaturated fatty acids for incorporation into surfactant. The more effective way to accomplish this goal is to provide a high proportion of the energy as carbohydrate. In fact, when glucose alone was given to pigs, the contribution of de novo synthesized palmitate to lung surfactant PC increased,[12] reflecting that palmitate is the principal product of de novo lipid synthesis from exogenous carbohydrate.[3] Thus, provision of a diet high in carbohydrate would be predicted to be preferable to one containing a large amount of unsaturated fatty acids. Further, the potentially detrimental effects of carbohydrate intake on CO_2 production can be minimized if caloric requirements are not exceeded. This can be monitored by indirect calorimetry: if the respiratory quotient is less than one, then the carbohydrate intake is not excessive. Although

this approach may provide less than the caloric requirement in some cases and therefore less than optimal in terms of maximal preservation of muscle mass,[31] maintenance of lung function is more important in critically ill patients with potential pulmonary problems.

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