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A B S T R A C T

Study Objective: The objective was to quantify cis and trans fatty acids in maternal plasma and infant cord plasma from adolescent mothers. Design: From 80 adolescent healthy mothers, we sampled postpartum maternal blood and umbilical cord blood at birth. Trans fatty acids (tFAs), linoleic (18:2), and arachidonic acid (AA, 20:4) acids of the n-6 family, and α-linolenic (18:3), eicosapentaenoic (20:5) and docosahexaenoic (22:6) acids of the n-3 family were analyzed by gas-liquid chromatography. Results were expressed as a percentage of total fatty acids.

Results: Linoleic fatty acid was present in greater proportions in the maternal plasma than in that of the umbilical cord, whereas AA was present in greater proportions in the total lipids of umbilical cord blood. Docosahexaenoic acid was the long-chain polyunsaturated fatty acid of the n-3 family that was predominant in both maternal and umbilical cord plasma. The tFAs in the maternal plasma had a negative correlation with oleic acid and linoleic acid. Linolenic acid had a positive correlation with cephalic perimeter upon birth. A tendency for a negative correlation between trans isomers and gestational age at birth (P = .05) was observed.

Conclusions: Long-chain polyunsaturated fatty acids, which are important to fetal growth and development, were found in greater quantities in the cord blood of newborns of adolescents than in the maternal blood, indicating a priority of transfer of AA and docosahexaenoic fatty acids to the fetus. Despite the lower levels of tFAs found in maternal blood, we verified potential risk for premature birth.

Key Words: Pregnancy, Nutrition, Adolescence, Essential fatty acids, Trans isomers, Newborns, Umbilical cord

Introduction

Fetuses and newborns require an adequate supply of n-6 and n-3 fatty acids, mainly long-chain polyunsaturated fatty acids (LC-PUFA) such as docosahexaenoic acid (DHA, 22:6 n-3) and arachidonic acid (AA, 20:4 n-6) in order to sustain development of neurological structures of the brain and retina and to support normal growth and development of other tissues. Moreover, these fatty acids are the precursors of eicosanoids, which are essential constituents of the membrane lipids that maintain cellular and organelle integrity and are important intracellular mediators of gene expression. Arachidonic acid (n-6) and DHA (n-3) are derived from dietary essential fatty acids (EFAs) such as linoleic acid (LA, 18:2 n-6) and linolenic acid (ALA, 18:3 n-3) by fatty acid desaturases and elongases in the maternal organism. Because of these fundamental roles of EFA and LC-PUFA, maternal, fetal, and neonatal EFA/LC-PUFA status is an important determinant of health and disease in infancy and later life.

Adolescence is characterized by a period in which dietary behavior is based on choosing for more attractive, available, practical, and cheaper food, without much concern for nutritional quality. The habit of consuming excessive amounts of food rich in fat and industrialized products, like cookies and ice cream, containing trans fatty acids (tFAs) has been observed in Brazil. Trans fatty acids are found in a variety of processed food made with partially hydrogenated vegetable oils, and levels are higher in countries in which hydrogenated vegetable oils are extensively used by the food industry. Trans fatty acids from the diet are absorbed and incorporated into human tissue. Before and after birth, exposure to these fatty acids occurs by placental transfer and through the mother’s milk. Thus, fetal exposure is dependent on the concentration of the given fatty acid in the maternal plasma, and thus on maternal dietary intake.

Infant tissues incorporate tFAs, leading to an increase in tissue levels of linoleic acid and to a relative decrease in the levels of AA and DHA acids, which suggests that tFA have an inhibitory effect on the activity of the Δ3 and Δ6 desaturase enzymes. The industrial hydrogenation process causes the loss of LA, and, particularly of ALA, from food. Thus, food containing tFAs is often poor in EFAs. Adverse effects like reduction in weight and fetal growth and greater risk of pre-eclampsia have been reported in conjunction with elevated consumption of trans isomers during pregnancy.

Pregnancy during adolescence accounts for up to one-fifth of all births worldwide, and it is associated with...
Materials and Methods

Study Design and Participants

A cross-sectional survey of a convenience sample of pregnant adolescents was conducted from August 2010 to June 2011 in the maternity ward of the Institute Fernandes Figueira/Fundaçao Oswaldo Cruz (FIOCRUZ), Rio de Janeiro, Brazil, a public health unit of the municipality of Rio de Janeiro, Brazil, which attends pregnant adolescents at high, medium, and low risk. The ethics committee of FIOCRUZ approved the study protocol.

Eighty healthy pregnant adolescents, who met the eligibility criteria and were willing to participate, were enrolled in the study. The eligibility criteria for participation of the volunteers in the study were: (1) ages varying from 15–19 years (owing to the high proportion of pregnant teens in this age group in Brazil); (2) first and singleton pregnancy; and (3) not smokers or users of illicit drugs or alcohol. Exclusion criteria were maternal gestational diabetes, pregnancy-induced hypertension, metabolic or genetic disorders, infection, and complications during delivery, including a newborn with congenital malformation. All participants and/or 1 of their parents or legal guardians signed an informed consent form prior to enrolment.

A questionnaire was administered to obtain sociodemographic, anthropometric, and obstetric data and information regarding lifestyle. The local nursing staff carried out anthropometric measurements of infants at delivery. All other birth outcome variables mentioned in Table 1 were measured according to standard hospital procedures.

Anthropometric Assessment of Pregnant Adolescents and Newborns

Pre-gestational weight and height measurements were taken on the first visit to antenatal care, prior to the 14th gestational week. Body mass index (BMI: weight [kg]/height[m]^2) cutoff points specific to adolescents and the female sex were used for the assessment of nutritional status of the adolescents.

The total gestational weight gain was calculated by subtracting the pre-pregnancy weight from the final weight immediately before delivery. The adequacy of weight gain was classified according to recommendations from the Institute of Medicine, following pre-gestational BMI categories, as proposed by Gutierrez and King.

To classify the nutritional status of the newborn, the gestational weight/age ratio at birth was used according to criteria defined by Alexander et al. The classification of low birth weight was defined according to the World Health Organization established cutoff point of weight less than 2500 g.

Collection of Samples, Preparation, and Analysis

During labor, which lasted 6–12 hours, the women were not allowed to eat and had an intravenous infusion of normal saline (no more than 50 mL/h). Epidural analgesia was not used. Samples of maternal blood were taken from the antecubital vein (which was not used for infusion) immediately after delivery (within 30 min after delivery). Blood samples from the umbilical cord of newborns were
collected immediately after the placenta had been expelled. The samples were collected in tubes containing 1 g Na2-EDTA/L. The plasma was separated within 30 minutes following sampling and kept at −80°C until analyzed. All determinations were performed in duplicate.

The lipids in the samples were extracted, saponified, and methylated according to the method described by Lepage and Roy. Fatty acid methyl esters were separated (SP2560 column, Supelco, Bellefonte, PA, USA) and quantified by gasliquid chromatography. The chromatographic conditions were similar to those described by Tinoco et al. The esters were identified by comparing their retention times to known standards (Sigma, Supelco). Results were expressed as mean ± standard deviation of weight percentage (g/100 g of total fatty acids). The coefficients of variation for the analyses were below 5.0% for all FA.

Statistical Analysis

Statistical analysis was performed using the Statistical Program for the Social Sciences (version 13.0, SPSS, Chicago, IL, December 2004) and Epi Info (version 6.04, Centers for Disease Control and Prevention/World Health Organization, Atlanta, GA, 1996) software.

To evaluate correlations between continuous variables, the Pearson correlation coefficient was used to investigate associations between maternal and neonatal FAs and between characteristics of the newborn infants (gestational age, weight, length, and head circumference at birth) and FAs. The continuous variables of the newborns and adolescents were presented in mean, minimum, and maximum values, and, for categorical variables, in frequencies. For comparison of the average FA content in the umbilical cord plasma of neonates and the plasma of adolescent mothers, the Student t-test was used. Differences were considered statistically significant at P < .05.

Results

The mean age of the adolescent mothers was 16.8 ± 2.2 years, and the mean age of menarche was 11.7 ± 1.7 years (Table 1). Around 10% of the population studied was overweight or obese and 4% was underweight, according to their pre-gestational BMI. Of the 80 participants, 41% gained more weight than recommended during gestation.

Only 5% of the neonates weighed under 2500 g at birth, 77% were born at a weight that was adequate for their gestational age, and 7.5% were premature (gestational age < 37 wk).

The tFA content present in the mothers’ blood and that of the umbilical cord was under 2% (Table 2). The predominant n-6 FA in maternal plasma is LA, whereas in umbilical cord plasma it is AA. Docosahexaenoic acid is the predominant n-3 FA, both in maternal and in umbilical cord plasma. In relation to total saturated FAs, greater proportions were found in the total lipids of the umbilical cord plasma than in the maternal plasma (P < .001; Table 2).

<table>
<thead>
<tr>
<th>Fatty acids</th>
<th>Maternal plasma</th>
<th>Umbilical Cord Plasma</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mono-unsaturated</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>C18:1 (n-9) cis</td>
<td>16.3 ± 0.4</td>
<td>12.8 ± 0.3</td>
<td>.06</td>
</tr>
<tr>
<td>C18:1 (n-9) trans</td>
<td>0.7 ± 0.09</td>
<td>0.5 ± 0.07</td>
<td>.07</td>
</tr>
<tr>
<td>Essential polyunsaturated</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>C18:2 n-6 (linoleic)</td>
<td>31.5 ± 0.4</td>
<td>12.9 ± 0.3</td>
<td>.00</td>
</tr>
<tr>
<td>C18:3 n-3 (linolenic)</td>
<td>0.16 ± 0.04</td>
<td>0.5 ± 0.1</td>
<td>.00</td>
</tr>
<tr>
<td>Long-chain polyunsaturated</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>C20:4 n-6 (AA)</td>
<td>5.6 ± 0.1</td>
<td>15.3 ± 0.3</td>
<td>.00</td>
</tr>
<tr>
<td>C22:4 n-6</td>
<td>0.7 ± 0.04</td>
<td>0.9 ± 0.09</td>
<td>.00</td>
</tr>
<tr>
<td>C20:5 n-3 (EPA)</td>
<td>0.3 ± 0.03</td>
<td>0.6 ± 0.05</td>
<td>.00</td>
</tr>
<tr>
<td>C22:5 n-3</td>
<td>0.2 ± 0.03</td>
<td>0.3 ± 0.1</td>
<td>.41</td>
</tr>
<tr>
<td>C22:6 n-3 (DHA)</td>
<td>2.0 ± 0.08</td>
<td>3.1 ± 0.1</td>
<td>.01</td>
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<tr>
<td>Total MFAe</td>
<td>19.4 ± 0.3</td>
<td>20.7 ± 0.5</td>
<td>.04</td>
</tr>
<tr>
<td>Total EPAf</td>
<td>31.8 ± 0.4</td>
<td>13.4 ± 0.3</td>
<td>.00</td>
</tr>
<tr>
<td>Total n-6 PUFAG</td>
<td>37.9 ± 0.7</td>
<td>26.8 ± 0.3</td>
<td>.00</td>
</tr>
<tr>
<td>Total n-3 PFUA</td>
<td>4.5 ± 0.2</td>
<td>2.9 ± 0.1</td>
<td>.00</td>
</tr>
<tr>
<td>Total LC-PUFAG</td>
<td>9.5 ± 0.1</td>
<td>18.5 ± 0.4</td>
<td>.00</td>
</tr>
<tr>
<td>Total saturatedf</td>
<td>36.0 ± 0.3</td>
<td>43.9 ± 0.4</td>
<td>.00</td>
</tr>
<tr>
<td>Total transg</td>
<td>0.7 ± 0.08</td>
<td>0.5 ± 0.08</td>
<td>.08</td>
</tr>
</tbody>
</table>

Note. The differences which were considered statistically significant at P < .05 are shown in bold.

Abbreviations: AA, arachidonic fatty acids; AGM, monounsaturated fatty acids; AGPI-CL, long-chain polyunsaturated fatty acids; DHA, docosahexaenoic fatty acids; EFA, essential fatty acids; EPA, eicosapentaenoic fatty acid; PUFA, polyunsaturated fatty acids

a Includes cis isomers of position; b Includes C18:3 n-3 and C18:2 n-6; c Includes C18:2 n-6, C18:3 n-6, C20:2 n-6, C20:3 n-6, C20:4 n-6 e C22:4 n-6; d Includes C18:3 n-3, C20:5 n-3, C22:5 n-3 and C22:6 n-3; e Includes EPA, DHA and ARA; f Total saturated includes C14:0, C15:0, C16:0 and C18:0. g Total trans includes C 18:1 n-9 and C 18:2 n-6 trans

acid (EPA; r = −0.35, P < .001). In the umbilical cord plasma, a negative correlation was found between AA and LA (r = 0.31, P < .001; Table 3). Pre-gestational BMI correlated negatively with oleic acid content (r = 0.28, P < .02; Table 4). The LA content and total saturated FAs correlated positively with the cephalic perimeter and length at birth, respectively (r = 0.29, P < .03 and r = 0.33, P < .01). Total tFA tended to correlate negatively with the gestational age at birth (r = −0.25, P < .05; Table 5).

Discussion

Compared with international studies, in which these isomers were investigated in human milk and/or blood from adult mothers, lower levels of tFAs were found in our study. Moreover, we found no difference between concentrations of tFAs in maternal and umbilical cord plasma, although lower levels had been found in the plasma of the umbilical cord. Other authors also noted the presence of tFAs in the cord plasma at lower percentages than those observed in the mothers’ blood. One can suppose selective placental transfer of tFAs may have occurred, since they are not synthesized in fetal tissue and originate from the mother’s diet.

Children and adolescents have consumption patterns that are different from those of adults. Most of the types of food in which tFAs are available, such as french fries, snacks, cookies, and breads, are also popular among children and adolescents. On the other hand, our recent findings indicate qualitative modifications in the dietary intake of...
adolescents during pregnancy, since 77% reported that they had changed their eating habits. Of this number, 59% reported having reduced their consumption of processed products (data not shown). This finding offers a possible explanation for the reduced levels of tFA observed in plasma of the pregnant adolescents studied.

One other possible reason for the lower levels of tFAs in maternal and umbilical cord plasma observed in the present study may be associated with the reduction in tFA content in processed products in Brazil. A recent reduction in the use of partially hydrogenated fats that has been adopted by the food industry has resulted in a drop in consumption of tFAs in many countries.13,30 In Brazil, in seeking to contribute to the reduced consumption of these isomers, the National Sanitary Surveillance Agency (Agência Nacional de Vigilância Sanitária/ANVISA) published, on December 23, 2003, RDC Resolution no. 360 (Technical regulation on the nutritional labeling of packaged foods), which amounts to the obligatory declaration of the quantity of trans fats on food labels; the established deadline for meeting the requirements of the new legislation was July 31, 2006.31 Recent implementation of proper technological processing in Brazilian industries with the aim of reducing the formation of tFAs was probably a direct consequence of the requirements of the present legislation. Thus, RDC resolution 360 constitutes an important instrument for implementing public policy destined to promote the population’s consumption of healthier food.

On the other hand, the technical regulation about nutritional labeling in Brazil (RDC 360/2003),31 in effect since July 2006, is not applied to companies that commercialize prepared and ready-to-eat food. Therefore, fast food may be a potential source of tFAs, but companies are not obligated to inform consumers. The analysis carried out by Aued-Pimentel et al identified that total or partial consumption of those food items is capable of providing tFA consumption above the maximum values recommended by the World Health Organization,24 which suggests that daily tFA consumption should be kept below 1% of total energy intake. For adults who consume 2000 kcal/d, the maximum consumption above the maximum values recommended by the World Health Organization should be kept below 2 g/d. These recommendations aim to contribute to the reduction in risk of developing chronic diseases.33

LC-PUFAs (DHA and AA) were shown to be in greater proportions in plasma of the umbilical cord in relation to the levels present in maternal plasma (Table 2). On the other hand, total EFA (LA and ALA) was significantly lower in the umbilical cord blood than in the maternal plasma. These results suggest that placental transfer is selectively greater for LC-PUFAs than for EFAs. Similar data have been
Table 4
Pearson Correlation Coefficient for Ratio between Fatty Acids, Stature, Pre-gestational Body Mass Index, Gestational Ponderal Gain, and Gestational Age in Maternal Plasma of Pregnant Adolescents

<table>
<thead>
<tr>
<th>Fatty Acids</th>
<th>Stature</th>
<th>PG BMI</th>
<th>Ponderal Gain</th>
<th>Gestational Age</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>r</td>
<td>P</td>
<td>r</td>
<td>P</td>
</tr>
<tr>
<td>Mono-unsaturated</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>C18:1 (n-9) cis (Oleic)</td>
<td>0.07</td>
<td>.55</td>
<td><strong>-0.28</strong></td>
<td>.02*</td>
</tr>
<tr>
<td>C18:1 (n-9) trans</td>
<td>0.03</td>
<td>.77</td>
<td>0.05</td>
<td>.66</td>
</tr>
<tr>
<td>Essential polyunsaturated</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>C18:2 n-6 (Linoleic)</td>
<td>-0.02</td>
<td>.86</td>
<td>0.00</td>
<td>.99</td>
</tr>
<tr>
<td>C18:3 n-3 (Linolenic)</td>
<td>-0.07</td>
<td>.54</td>
<td>0.21</td>
<td>.08</td>
</tr>
<tr>
<td>Long-chain polyunsaturated</td>
<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>C20:4 n-6 (AA)</td>
<td>-0.02</td>
<td>.87</td>
<td>0.18</td>
<td>.12</td>
</tr>
<tr>
<td>C22:4 n-6</td>
<td>0.03</td>
<td>.81</td>
<td>0.17</td>
<td>.14</td>
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<tr>
<td>C20:5 n-3 (EPA)</td>
<td>0.09</td>
<td>.44</td>
<td>-0.03</td>
<td>.79</td>
</tr>
<tr>
<td>C22:5 n-3</td>
<td>0.01</td>
<td>.96</td>
<td>0.19</td>
<td>.11</td>
</tr>
<tr>
<td>C22:6 n-3 (DHA)</td>
<td>-0.04</td>
<td>.72</td>
<td>-0.06</td>
<td>.59</td>
</tr>
<tr>
<td>Total MFAa</td>
<td>0.14</td>
<td>.25</td>
<td><strong>-0.33</strong></td>
<td>.00**</td>
</tr>
<tr>
<td>Total EFAb</td>
<td>-0.01</td>
<td>.96</td>
<td>0.04</td>
<td>.75</td>
</tr>
<tr>
<td>Total n-6 PUFAc</td>
<td>-0.02</td>
<td>.88</td>
<td>0.07</td>
<td>.55</td>
</tr>
<tr>
<td>Total n-3 PUFAd</td>
<td>0.05</td>
<td>.67</td>
<td>0.12</td>
<td>.33</td>
</tr>
<tr>
<td>Total LC-PUFAe</td>
<td>-0.01</td>
<td>.94</td>
<td>0.18</td>
<td>.13</td>
</tr>
<tr>
<td>Total saturatedf</td>
<td>-0.11</td>
<td>.34</td>
<td>0.19</td>
<td>.11</td>
</tr>
<tr>
<td>Total transg</td>
<td>0.01</td>
<td>.92</td>
<td>0.07</td>
<td>.54</td>
</tr>
</tbody>
</table>

Note. The differences which were considered statistically significant at P < .05 are shown in bold.
Abbreviations: AA, arachidonic fatty acid; DHA, docosahexaenoic fatty acid; EFA, essential fatty acids; EPA, eicosapentaenoic fatty acid; LC-PUFA, long-chain polyunsaturated fatty acids; PUFA, polyunsaturated fatty acids

* P < .05
** P < .01
a Includes all cis isomers of position
b Includes C18:3 n-3 and C18:2 n-6
c Includes C18:2 n-6, C18:3 n-6, C20:2 n-6, C20:3 n-6, C20:4 n-6 and C22:4 n-6
d Includes C18:3 n-3, C20:5 n-3, C22:5 n-3 and C22:6 n-3
e Includes: EPA, DHA and AA
f Total saturated includes C14:0, C15:0, C16:0 and C18:0
g Total trans includes C 18:1 n-9 trans and C 18:2 n-6 trans

In this sense, preferential transfer of particular LC-PUFAs from the maternal to the fetal circulation in relation to the transfer of EFAs may represent a determining factor in adequate fetal growth and development. Maintaining adequate transfer of DHA to fetal circulation in relation to the transfer of EFAs may represent a determining factor in adequate fetal growth and development. Maintaining adequate transfer of DHA to fetal blood seems to be an important physiological process, since humans, even newborns and children, convert less than 1% of ALA n-3 into DHA n-3.

Despite the lower levels of tFAs found in maternal blood, we verified that there was a negative association between these isomers and oleic acid (r = -0.30, P < .001), LA (r = -0.39, P < .001), ALA (r = -0.41, P < .001), and EPA (r = -0.35, P < .001). Similar results were also revealed by other authors. The most likely hypotheses justifying these associations involve: (1) a possible inhibiting effect of EFAs in the desaturation and elongation processes of the n-6 family of FAs, or (2) lower consumption of lipid sources rich in n-6 and n-3, owing to greater consumption of food rich in tFAs. On the other hand, in cord plasma, a negative correlation was found between AA and LA (r = -0.31, P = .01). Koletzko found inverse associations between tFA and LC-PUFAs (AA and DHA) in umbilical cord plasma and between tFAs and birth weight of premature babies.

Similar to the results reported by Decsi et al., our results revealed no correlations between tFA levels and newborns' anthropometric parameters (weight, length, and cephalic perimeter at birth). However, tFAs tended to correlate negatively with gestational age at birth (P = .05), Hornstra et al., while investigating associations between elaidic tFA (C18:1 trans) levels in plasma, arterial, and venous blood from the umbilical cord and weight, length, and cephalic perimeter upon birth, found negative and statistically significant associations between plasmatic tFAs, cephalic perimeter, and length upon birth. Elias and Innis examined 70 newborns from adult pregnancy and investigated the relationship between birth weight, birth length, and length of gestation and the levels of tFAs in umbilical cord plasma; those authors found a significant inverse relationship between the newborn infant plasma concentration of tFAs in cholesteryl esters and the length of gestation. Thus, based on the literature, and in accordance with our findings, it is suggested that even in low concentrations, tFAs may contribute to premature birth.

The results of the present study do not suggest an important correlation between tFAs and gestational age, but because dietary trans intake in the studied population was relatively low (59% reported reduced consumption of processed products), we cannot exclude a possible strong negative association at higher intake. Pregnancy in young mothers is considered to be of the most important risk conditions, not only because of young age, but also because of economic problems, lack of education, and insufficient prenatal care. In this group, the risk of maternal or neonatal complications is increased and nutrition is an important aspect of prenatal care. Presence of tFAs in blood may indicate a high risk of nutritional problems in the young mother and therefore requires in-depth nutritional assessment, counseling, and follow-up. Maternal health habits during pregnancy may be...
a major factor for clinical intervention. Moreover, considering the adolescent’s desire to maximize the health of her developing fetus, poor habits and behaviors may be changed more easily during pregnancy than at other times.

In summary, our study demonstrates higher plasma LC-PUFAs (AA and DHA) in the umbilical cord plasma of newborns of adolescent mothers compared to maternal plasma, indicating the occurrence of priority transfer of AA n-6 and DHA n-3 FAs to the fetus. There was no difference between tFAs in maternal plasma and in umbilical cord, and a tendency was observed for these FAs to have a negative correlation with gestational age at birth. Given the harmful effects of tFAs on health, action should be taken to reduce consumption of these fatty acids during pregnancy.

Acknowledgments

This research is based on data from the Brazilian study “Trans fatty acids in pregnant adolescents and its consequences for maternal and child health,” which was funded primarily by the Brazilian Research Council (Conselho Nacional de Desenvolvimento Científico e Tecnológico-CNpq) and Fundação de Amparo a Pesquisa do Rio de Janeiro (FAPERJ).

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