14th International Nutrition & Diagnostics Conference

Correlation between plasma levels of lutein and oxidized low density lipoproteins: a short human intervention study

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BACKGROUND AND AIM

The development of vegetable functional food products that provide benefits beyond their traditional nutritional value raised increasing interest. In fact several studies have shown that diet plays a role in the development of human diseases associated with oxidative damage (diabetes, cardiovascular disease and metabolic syndrome) [1]. **Pigmented vegetables** such red spinach, red chicory and red chard contain bioactive molecules such as phytochemicals (polyphenols and carotenoids), vitamins, minerals and fibers and therefore could be considered as funtional foods [2].

Among phytonutrients, **carotenoids** have been widely investigated for their protective role against lipid peroxidation [3-5]; in fact they circulate associated to lipoproteins in particular to low density lipoproteins (LDL).

Aim of the study was to investigate the bioavailability of the main carotenoids (lutein and beta carotene) and the functional properties of a experimental ready-to-eat product containing pigmented vegetables such as red and green spinach (*Spinacia oleracea*), red and green chicory (*Cichorium intybus*) and a lower percentage of green and red chard (*Beta vulgaris L. var. cicla*).

METHODS and DESIGN OF THE STUDY

- **Study of compositional** (macronutrients, vitamins and phytochemicals) **and antioxidant properties** (ORAC assay) of experimental ready-to-eat product containing pigmented vegetables.
- **Intervention study to investigate the functional properties of the experimental ready-to-eat product containing pigmented vegetables**

Forty-eight subjects healthy volunteers were recruited. The mean age was 40.5±12.3 years and mean BMI 23.6±4.1 kg/m^2. The intervention phase consisted of a 2-week period which included daily consumption of a portion (300 g) of the vegetable product supplied by Italsur srl (Notaresco, Teramo,Italia). Each subject was asked to complete a 15-day dietary record.

At the beginning of the intervention (baseline, T0) and at the end (after 2 weeks, T15), fasting blood samples were collected. Levels of **serum lipids** (TG, triglycerides; TC, total cholesterol; LDL-C, LDL cholesterol; HDL-C, HDL cholesterol) were analyzed by commercial kits. **Plasma levels of carotenoids (beta-carotene and lutein)** were quantified by high-performance liquid chromatography (HPLC) system [4]. **Plasma total antioxidant capacity (PAT)** was measured using oxygen radical absorbance capacity (ORAC) assay [5].

As marker of lipid peroxidation, the **plasma levels of oxidized LDL** were determined in plasma by a sandwich ELISA procedure [6].

RESULTS AND DISCUSSION

Table 1 shows the values of the energy, macro and micronutrient intake of the subjects through the baseline diet and the contribution of daily dose of the product containing pigmented vegetable.

Effect on plasma levels of carotenoids**.** A portion of the vegetable product contains about 30 mg lutein and 11 mg beta carotene. Plasma carotenoid concentration increases after 2 weeks, beta-carotene by 56% and lutein by 94.5% compared with baseline (**Figure 1**). These data demonstrate that carotenoids in the product are bioavailable.

Effect on plasma lipids and oxidized LDL. The dietary treatment favorably influences serum lipid profile with a significant decrease in the levels of TC (mean percentage reduction -6.4% with respect to baseline) and LDL-C (mean percentage reduction -9.4% with respect to baseline) **(Table 2)**. Moreover a significant decrease of concentration of ox-LDL (mean percentage decrease -21.9 %, with respect to baseline) and an increase of total antioxidant capacity are observed in plasma of subjects after dietary treatment **(Table 2)**. As shown in **figure 2** subjects with higher levels of ox-LDL have a higher decrease of this variable. This variability in response to dietary treatment could be associated with genetic or other characteristics of the individuals. These results strengthen the hypothesis that the effect of dietary intervention could be modulated by baseline conditions of the subjects.

Correlation between plasma lutein and oxidized LDL. A significant negative correlation has been established between plasma levels of lutein and ox-LDL before and after daily intake of the vegetable product **(Figure 3)**. These results suggest that lutein may play a role in the protective effect against oxidation of LDL. However, the synergistic effects of phytochemicals (dietary fibers, polyphenols, carotenoids) in the frozen ready to eat product could be responsible for the protective effects.

In conclusion an accurate selection of pigmented vegetables results in palatable and healthy product with physiological effects on plasma lipids and lipid peroxidation.

TABLE 2. Effect of dietary treatment on plasma lipid profile, levels of ox-LDL and total antioxidant capacity . *p<0.05 vs baseline

	BASELINE	AFTER 2-WEEK PERIOD WHICH INCLUDED A DAILY PORTION OF VEGETABLE PRODUCT
TC (mmol/L)	4.87±0.56	4.55±0.52*
LDL-C (mmol/L)	2.80±0.56	2.53±0.52*
HDL-C (mmol/L)	1.65±0.28	1.65±0.30
TG (mmol/L)	0.91±0.41	0.87±0.37
Total antioxidant capacity (mmolTE/L)	14634±3492	16218±3834*
ox-LDL (U/L)	41.7±16.1	32.6±12.2*
ox-LDL/LDL-C (U/mmol)	15.2±5.6	13.2±5.2*

TABLE 1. Energy, macronutrient and micronutrient intake of subjects through the baseline diet and contribution of the vegetable product daily dose evaluated by 15-day dietary record.

	BASELINE DIET	CONTRIBUTION OF A PORTION OF VEGETABLE PRODUCTS TO DAILY OVERALL INTAKE
ENERGY (kcal)	1614 ±311	18±2
PROTEIN (g)	60.2±16.3	7.8±1.1
CARBOHYDRATE(g)	211±46	11±2.2
FIBER(g)	17.7	8.5±0.18
FAT(g)	51.5±12.7	0.63±0.01
Vitamin C	62.2±23.2	85±3.7
Folate(mg/day)	0.17±0.03	0.33±2.7
Alpha-tocopherol(mg/day)	7.4±1.7	3.9±0.30
Niacin(mg/day)	14.1±3.2	1.2±0.05
Retinol(mg/day)	0.7±0.2	3.2±0.20
Riboflavin(mg/day)	1.24±0.23	0.27±0.03
Thiamine(mg/day	0.99±0.16	0.18±0.01
Total POLYPHENOLS(mg/day)		530±82
CAROTENOIDS		
Beta-carotene(mg/day)	3.1-±2.1	11.1±0.8
Zeaxanthin(mg/day)		0.7±0.1
Lutein (mg/day)		29.6±0.30
Total ORAC (mmol(TE)/day)	4.7±1.6	5.8±0.1

FIGURE 1. Effect of dietary treatment on plasma levels of carotenoids (beta-carotene and lutein); ( ) levels at baseline and ( ) after 2-weeks period which included a daily portion of the ready-to-eat product.*p<0.05 *vs* baseline

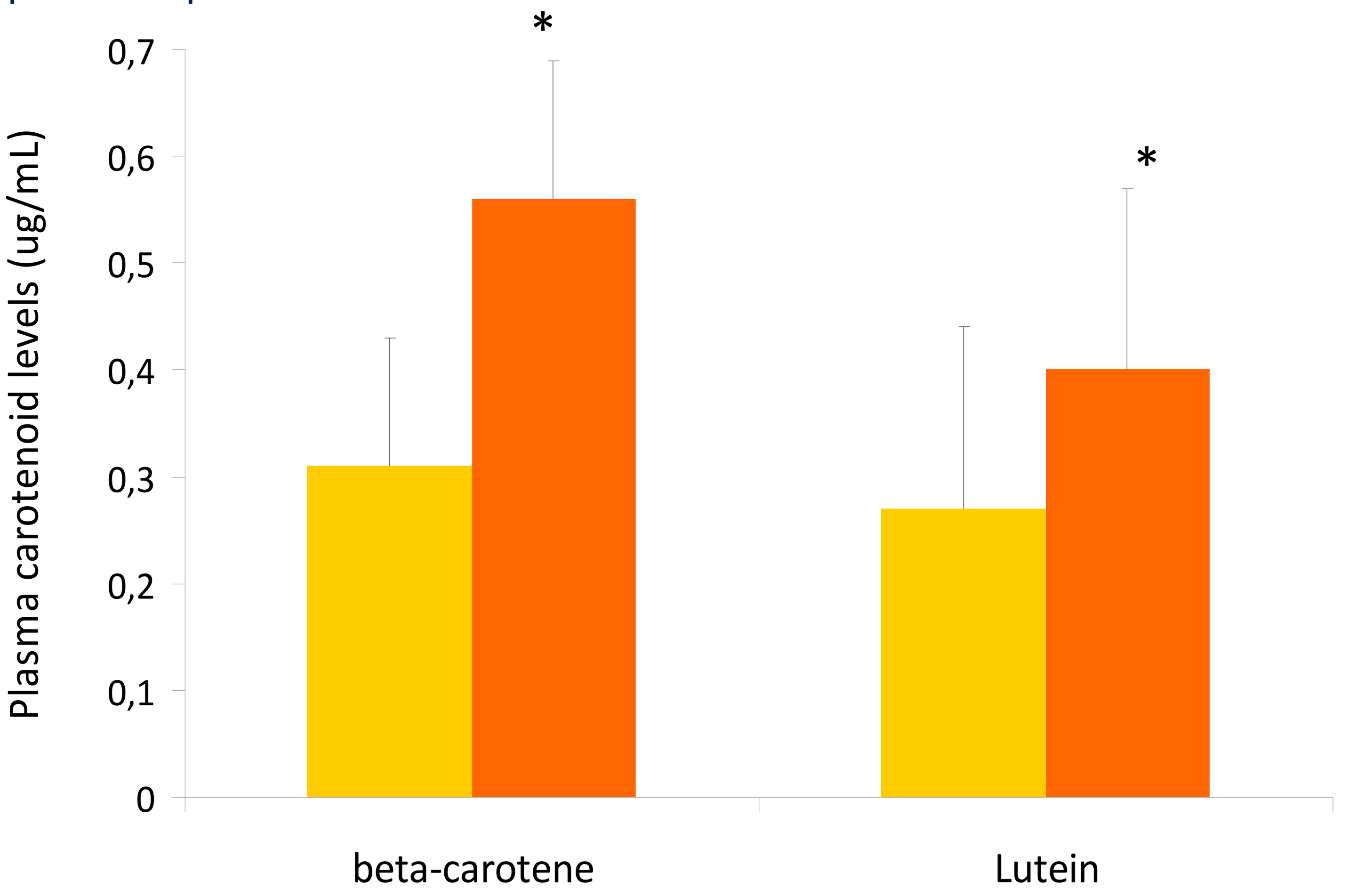


FIGURE 2. Effect of dietary treatment on levels of ox-LDL in plasma of subjects with low levels of ox-LDL (basal levels below the median value 39.7U/mL) **or high levels of ox-LDL** (basal levels above the median value 39.7U/mL). ( ) ox-LDL at baseline and ( ) after 2-weeks period which included a daily portion of the vegetable product .*p<0.05 *vs* baseline; # p<0.05 vs subjects with low levels of ox-LDL

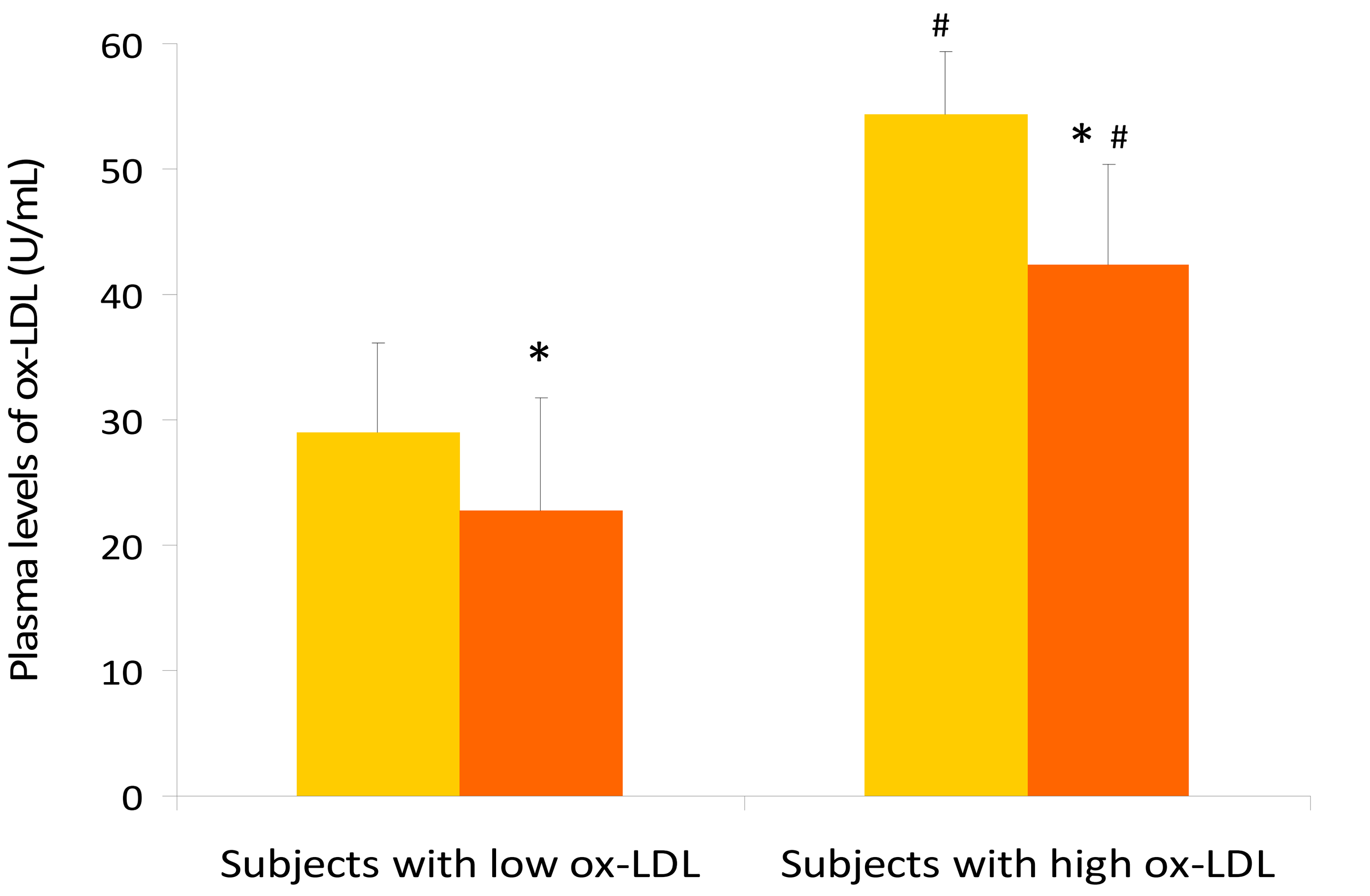
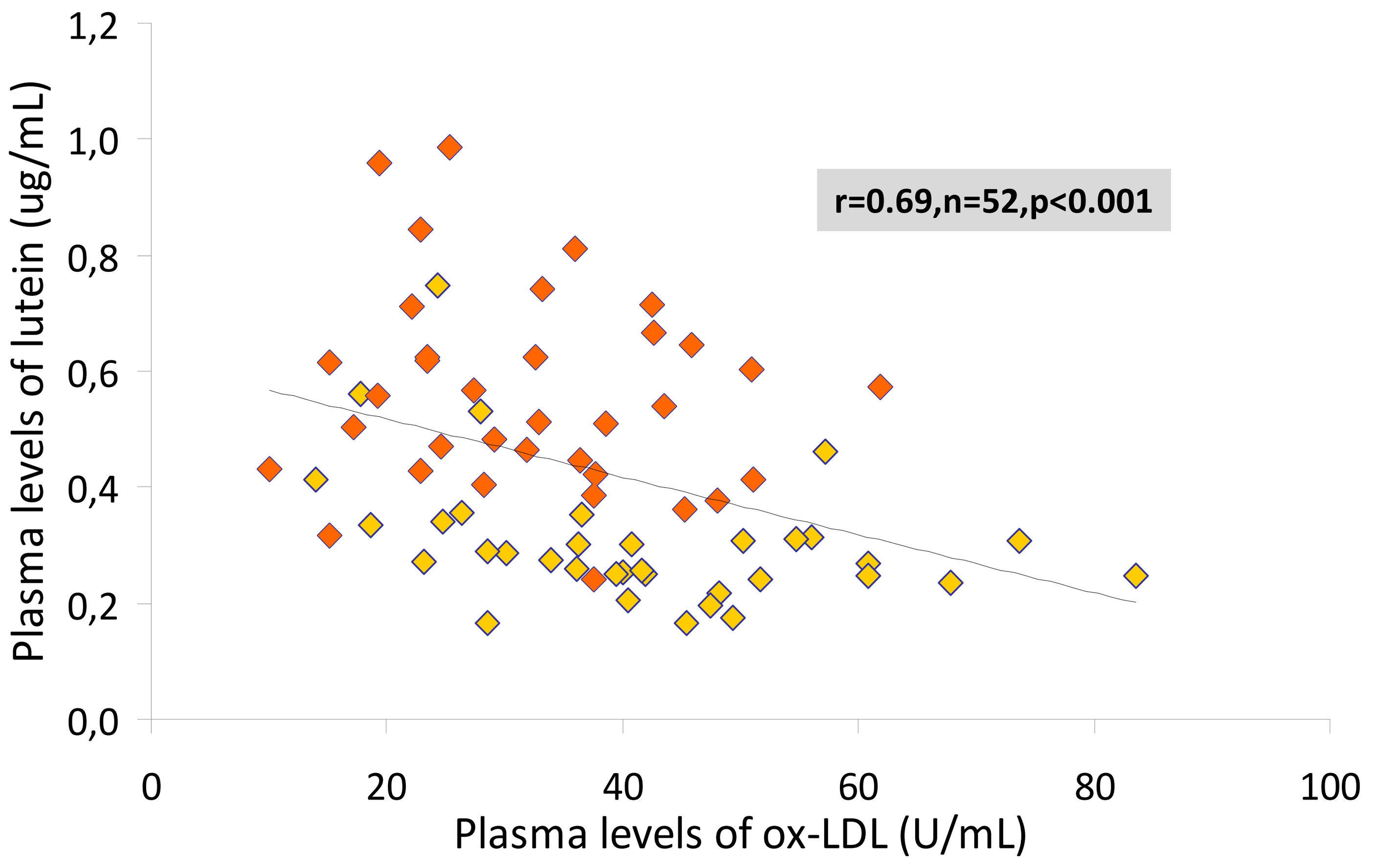


FIGURE 2. Correlation between levels of lutein and ox-LDL in plasma of subjects at baseline ( ) and after 2-weeks period which included a daily portion of vegetable product ( ).



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