Article

Creation of a Databank for Content of Antioxidants in Food Products by an Amperometric Method

Yakov I. Yashin 1, Boris V. Nemzer 2,*, Vadim Yu. Ryzhnev 1, Alexandr Ya. Yashin 1, Nina I. Chernousova 1 and Polina A. Fedina 1

1 Scientific Development & Production Center “Khimavtomatika”, Selskohozyaistvennaya 12a, 129226 Moscow, Russia
2 VDF FutureCeuticals, Inc., 300 West 6th Street, Momence, IL 60954, USA

* Author to whom correspondence should be addressed; E-Mail: bnemzer@vandrunen.com; Tel.: +1-815-472-3100; Fax: +1-815-472-3850.

Received: 20 September 2010 / Accepted: 20 October 2010 / Published: 22 October 2010

Abstract: Oxidative stress, i.e. excessive content of reactionary, oxygen, and nitrogen compounds (ROAC), including free radicals, is one of the causes of various dangerous diseases as well as premature aging. The adverse effect of free radicals can be neutralized by antioxidants. In order to carry out antioxidant therapy, one needs to know the contents of antioxidants in food products. We have created the databank for the contents of antioxidants in 1,140 food products, beverages, etc. Apart from water-soluble antioxidants, fat-soluble antioxidants in dairy and fish products, cacao, chocolate, nuts etc. were determined for the first time using an amperometric method.

Keywords: water- and fat-soluble antioxidants; amperometric method; antioxidant therapy; databank

1. Introduction

Over the past decades various studies from different countries have confirmed that oxidative stress is one of the main causes of premature aging and many other diseases [1-8]. This means excessive contents of free oxygen radicals in biological liquids are in the human body such as superoxide anion, hydroxyl radical, perhydroxyl radical, etc. Free radicals are constantly formed, even with normal cell metabolism. Some data shows that 2% of total absorbed oxygen is transformed into free radicals. It is
widely held that our body requires certain amounts of active forms of oxygen to help eliminate harmful bacteria, dying cells, etc. The antioxidant system of a healthy person provides for a normal and safe level of free radicals. However, if adverse factors (radioactive and UV radiation exposures, environmental pollution or low quality food products, stress, diseases, certain powerful drugs and treatment procedures, smoking, alcoholism, drug abuse, etc.) affect the body, the level of active oxygen forms (free radicals and peroxides) may also increase. Such forms may damage DNAs, proteins, lipids, carbohydrates as well as vascular walls, which results in the disorganization of normal processes in human bodies. To put it generally, it works as follows: adverse conditions $\rightarrow$ oxidative stress $\rightarrow$ oxidation of vital molecules $\rightarrow$ development of diseases or premature aging.

Free radicals are especially reactive towards membrane lipids containing unsaturated bonds. As a result, the properties of cellular membranes are modified. The most active free radicals break bonds in the DNA molecule and injure the genetic apparatus of cells, which may lead to the development of oncological diseases. Upon oxidation, low-density lipoproteins can deposit onto the vascular walls and initiate the development of cardiovascular diseases. Presently, tens of diseases have been associated with oxidative stress.

To prevent oxidative stress, one should consume products with sufficient amounts of antioxidants, so that the harmful effect of free radicals is significantly diminished. The major natural antioxidants include flavonoids, aromatic oxyacids, vitamins C and E, carotenoids, and other compounds. The continual increase of the recommended amount of antioxidants should also be avoided, as some of them become pro-oxidants at elevated concentrations. The Russian Ministry of Health recommends consuming approximately 350 mg of different antioxidants per day. Sick and debilitated individuals, as well as those who are overworked and work in poor conditions are advised to consume around 1,000-1,300 mg. However, such recommendations specify only the general amount of antioxidants as, unfortunately, it is not yet possible to estimate the exact ratio for water- and fat-soluble antioxidants.

The aim of this study is a creation of database with results of antioxidant capacity of main food products: vegetables and greens, fruits, berries, juices, nuts, dairy products, seafood products, vegetable oils, honey, tea, coffee, different alcoholic beverages, etc.

2. Results and Discussion

Studies on evaluation of antioxidant activity (capacity) for various food products in different countries have been published [9-13]. Halvosen et al. [10] established the total content of antioxidants in fruits, berries, vegetables and other products determined by the Ferric Reducing Antioxidant Power (FRAP) method. The following berries contained the highest amount of antioxidants: rose hips, black currant, raspberry, bilberry, and cranberry. Fruits, berries and grains contribute 43.6%, 27.1%, and 11.7% of antioxidants in the diet of Norwegians, respectively. Vegetables contribute only 8% to the total content of plant antioxidants in the diet.

The USDA database of the flavonoids content in selected foods [14] provides the flavonoids composition in different foods and beverages, including raw and processed fruits, berries, vegetables, spices and such on. In particular, the following flavonoids and aromatic oxyacids have been indentified: hydroxy aromatic acids (gallic acid, dihydroxybenzoic acid, $p$-hydroxybenzoic acid), anthocyanidins (cyanidin, pelargonidin, peonidin, delphinidin, malvidin), anthocyanins, hydroxicoric acids (caffeic acid, chlorogenic acid, coumarinic acid, ferulic acid, syringic acid), flavanols (catechin,
epicatechin), flavones (luteolin, apigenin). Only data generated by acceptable procedures defined as those which lead to good separation of flavonoids compounds (e.g. column chromatography, HPLC, capillary zone electrophoresis, micellar electrokinetic capillary chromatography) have been included in this database.

Thus, although over 5,000 flavonoids have been identified [15], only 50-100 of them are present in significant amounts in fruits, berries, vegetables, and other natural products and can be separated and identified by HPLC or LC/MS systems. Flavanones (isonaringin, naringenin, naringin, hesperetin, hesperidin, neohesperidin), flavanones (quercetin, auranetin), flavones (tangeretin, nobiletin) were indentified in oranges.

Sakakibara et al. [16] classified over 100 of the most common antioxidants found in food products nd simultaneously separated by a Capcell pak C18 UG120 column (Shiseido Co., Ltd., Tokyo, Japan) and detected by a HPLC system equipped with a diode array detector. With such a column, the contents of antioxidants in food products may be expressly evaluated.

In this work the total contents of water- and fat-soluble antioxidants in 1,140 food products, beverages, dietary supplements, herbal extracts, vitamins, medicines etc. have been estimated by an amperometric method using a TsvetYauza-01-AA (NPO Khimavtomatika Inc., Moscow, Russia) amperometric detector.

The following groups of products have been studied by this method: berries, fruits, vegetables, (potato separately), greens, dairy, meat and fish products, vegetable oils, cacao and chocolate, nuts and seeds, grains, sprouted grains, condiments and spices, tea, coffee, wine, beer, cognac, whisky, juices, beverages, dietary supplements, herbal extracts, medicinal-antioxidants, etc. This report includes the data of antioxidant capacity or antioxidants content in some food products and beverages (Tables 1-12). Before each measurement, the device was calibrated according to standards; the correlation ratio for calibration curves was no less than 0.99. The used average value was based on 3-5 consecutive measurements; SD was less than 3%. Maximum permissible concentration (MPC) for major antioxidants was no higher than 10^{-8}–10^{-9} g. Based on sample selection and preparation, the total error for the measurements was within ±10%.

In Table 1 are presented data of total content of antioxidants (TCA) in berries and fruits. It is shown that lemon peel contains a significantly higher amount of antioxidants in comparison with the whole lemon or lemon pulp.

Table 2 contains TCA data for different vegetables and greens. We have separately studied more than 30 potato varieties provided by the Lorkh Institute of Potato Growing (Russia). TCA value for the best potato varieties was around 70 mg/100 g. According to the data presented at the Malta Polyphenols 2007: 4th International Conference on Polyphenols Applications [17] among vegetables, in France, potatoes contribute the highest amount of antioxidants into the diet, and among fruits—Apples.

Table 3 includes data of total content of fat soluble antioxidants (TCFA) in cheeses and dairy products. All samples have been purchased in grocery store in Moscow (Russia). In Table 4 are presented data of TCFA in seafood products and in Table 5 - TCFA results for vegetable oils. Gallic acid has been used as a standard for cheeses, dairy products and vegetable oils samples and quercetin – for seafood products. We did not find any data related to the contents of fat-soluble antioxidants in
dairy or seafood products in the literature. There is a huge difference in TCFA results between salmon caviar samples – from 136 to 6 mg/100 g.

Table 1. Total Content of Antioxidants (TCA) in Berries and Fruits (quercetin – standard).

| No. | Name                          | TCA mg/100g |
|-----|-------------------------------|-------------|
|     | **Berries**               |             |
| 1   | Chokeberry                    | 800         |
| 2   | Black currant                 | 765         |
| 3   | Black sour cherry             | 572         |
| 4   | Hawthorn                      | 570         |
| 5   | Rose hip                      | 530         |
| 6   | Viburnum                      | 495         |
| 7   | Bilberry                      | 406         |
| 8   | Blueberry                     | 335         |
| 9   | Cranberry                     | 270         |
| 10  | Barberry                      | 230         |
| 11  | Black cherry                  | 221         |
| 12  | Wild strawberry               | 210         |
| 13  | Red currant                   | 200         |
| 14  | Ashberry                      | 200         |
| 15  | Raspberry                     | 171         |
| 16  | Cowberry                      | 143         |
| 17  | Gooseberry                    | 46          |
| 18  | Black grapes                  | 42          |
| 19  | Sea buckthorn                 | 40          |
|     | **Fruits**                   |             |
| 1   | Lemon peel                    | 285         |
| 2   | Feijoa                        | 243         |
| 3   | Red orange                    | 79          |
| 4   | Lemon                         | 74          |
| 5   | Apple (Zhigulevskoye variety) | 64          |
| 6   | Pear                          | 50          |
| 7   | Kiwi                          | 45          |
| 8   | Apricot                       | 38          |
| 9   | Plum                          | 30          |
| 10  | Avocado                       | 27          |
| 11  | Nectarine                     | 17          |
| 12  | Peach                         | 12          |
| 13  | Banana                        | 7           |
Table 2. Total Content of Antioxidants (TCA) in Vegetables and Greens (quercetin – standard).

| No. | Name               | TCA mg/100g |
|-----|-------------------|-------------|
|     | Vegetables        |             |
| 1   | Garlic            | 273         |
| 2   | Sweet red pepper | 245         |
| 3   | Beet              | 217         |
| 4   | Turnip            | 135         |
| 5   | Red onion         | 117         |
| 6   | Sweet yellow pepper | 92     |
| 7   | Yellow onion      | 88          |
| 8   | Bulb onion        | 79          |
| 9   | Red cabbage       | 76          |
| 10  | White onion       | 75          |
| 11  | White cabbage     | 69          |
| 12  | Tomato            | 64          |
| 13  | Carrot            | 64          |
| 14  | Radish            | 62          |
| 15  | Eggplant          | 54          |
| 16  | Potato            | 43          |
| 17  | Cucumber          | 22          |
|     | Greens            |             |
| 1   | Parsley           | 110         |
| 2   | Watercress salad (leaves) | 100   |
| 3   | Celery leaves     | 100         |
| 4   | Cilantro          | 90          |
| 5   | Watercress salad (stems) | 70     |
| 6   | Green onion       | 50          |
| 7   | Dill              | 30          |
| 8   | Salad-premium     | 10          |
| 9   | Celery stem       | 10          |

Table 3. Total Content of Fat-Soluble Antioxidants (TCFA) in Cheese & Other Dairy Products (standard - gallic acid).

| No. | Name                                           | TCFA mg/100g | Manufacturer                                      |
|-----|------------------------------------------------|--------------|--------------------------------------------------|
|     | Cheeses                                        |              |                                                  |
| 1   | Suluguni cheese                               | 50           | JSC Giaginsky Diary Plant, Russia                |
| 2   | Parmesan Goya cheese 50%                      | 25           | Molfino Hermanos S.A., Argentina                 |
| 3   | Djigas cheese (PARM.) 40%                     | 21           | Zemaitijos Pienas, Lithuania                     |
| 4   | Svalya cheese                                 | 13           | Agroaspect, LLC, Russia                           |
| 5   | Gauda cheese 48%                              | 12           | JSC Valio, Finland                               |
| 6   | Rossiysky cheese                              | 12           | Uglich, Russia                                   |
|     | Dairy Products                                 |              |                                                  |
| 1   | Activia yogurt, enriched with ActiRe-gularis bifido-bacterium with herbs 2.2% | 75           | Danon Industry, LLC, Russia                      |
| 2   | Miracle-yogurt “Strawberry+wild strawberry” 2.4% | 75           | Vimm-Bill-Damm, LLC, Russia                      |
| 3   | Lianozovskoye milk 3.2%                       | 72           | Vimm-Bill-Damm, LLC, Russia                      |
| 4   | Curdled milk 3.2%                             | 66           | CJSC Opolye Holding Company, Russia               |
| 5   | Fromage frais cottage cheese 0%               | 54           | Senoble France, France                           |
| 6   | Dolce Vita sour cream 20%                     | 49           | CJSC Ozeretsky Dairy Complex, Russia             |
| 7   | Vologodskoye butter 82.5%                     | 24           | CJSC Ozeretsky Dairy Complex, Russia             |
| 8   | Sour cream 20%                                 | 19           | CJSC Opolye Holding Company, Russia               |
| 9   | Countryside House drinking cream 10%          | 13           | Vimm-Bill-Damm, LLC, Russia                      |
Table 4. Total Content of Fat-Soluble Antioxidants (TCFA) in Seafood Products (standard – quercetin).

| No. | Name                                | TCFA mg/100g | Manufacturer                      |
|-----|-------------------------------------|--------------|-----------------------------------|
| 1   | Grainy salmon caviar                | 136          | Sakhalin, Russia                  |
| 2   | Canned Baltic sprats in oil         | 88           | Kreon, LLC, Svetly, Russia        |
| 3   | Cooked cod (cans)                   | 31           | Espersen, Denmark                 |
| 4   | Fish oil                            | 30           | Russia                            |
| 5   | Grainy salmon caviar                | 24           | JSC ICE Meridian, Russia          |
| 6   | Grainy salmon caviar                | 22           | CJSC Northeast Company, Ltd, Russia |
| 7   | Herring (filet in oil)              | 22           | Vachyunay, LLC, Russia            |
| 8   | Elite grainy salmon caviar          | 17           | JSC Fish-Processing Plant No.1, Russia |
| 9   | Red grainy salmon caviar            | 12           | CJSC ITA Northern Company, Russia |
| 10  | Grainy salmon caviar                | 6            | JSC Fish-Processing Plant No.1, Russia |

Table 5. Total Content of Fat-Soluble Antioxidants (TCFA) in Vegetable Oils (standard - gallic acid).

| No. | Name                                | TCFA mg/100g | Manufacturer                      |
|-----|-------------------------------------|--------------|-----------------------------------|
| 1   | Ethiopian black cumin oil           | 294          | Egypt                             |
| 2   | CAROTINO palm-tree oil              | 198          | Malaysia                          |
| 3   | Olive oil                           | 127          | Greece                            |
| 4   | Sea buckthorn oil                   | 123          | VitaOil, LLC, Russia              |
| 5   | Wheat-germ oil                      | 109          | DecosT Series, NPKF DecosT, LLC   |
| 6   | Shark oil                           | 73           | Kubanrybprom, LLC, Russia         |
| 7   | Inca Gold amaranthine oil           | 62           | DecosT Series, NPKF DecosT, LLC, Russia |
| 8   | Corn oil                            | 45           | Efko, LLC, Russia                 |
| 9   | Pine nut oil                        | 34           | Altay, LLC, Russia                |
| 10  | Cucurbits oil                       | 31           | DecosT Series, NPKF DecosT, LLC, Russia |
| 11  | Mustard oil                         | 22           | Life Aromas Series, NPKF DecosT, LLC, Russia |
| 12  | Flax oil                            | 18           | Life Aromas Series, NPKF DecosT, LLC, Russia |
| 13  | Celery oil                          | 18           | CJSC SPA Europe-Biopharm          |
| 14  | Unfiltered walnut oil (crumb)       | 16           | Life Aromas Series, NPKF DecosT, LLC, Russia |

Interesting data was obtained upon detection of both water- and fat-soluble antioxidants in cacao and chocolate samples (Table 6). Russian samples of cacao contain significant amounts of water- and fat-soluble antioxidants because the cacao oil is not extracted from raw cacao material. Per serving, in terms of the general level of antioxidant activity, cacao exceeds tea and red wine. Moreover, cacao is one of few products containing the entire set of required antioxidants: water-soluble antioxidants, fat-soluble antioxidants, and anthocyanins. Cacao may be considered one of the most balanced food products taking into account that cacao contains fats, proteins, carbohydrates, microelements, and vitamins. No wonder that cacao and chocolate are considered “food of the Gods.” Fat-soluble antioxidants in nuts and seeds (Table 7) are in an approximate correspondence with the published data.
Table 6. Total Content of Fat-Soluble Antioxidants (TCFA) (standard - gallic acid) and Water-Soluble Antioxidants (TCA) in Cacao, Chocolate.

| No. | Name                                | TCFA mg/100g | TCA mg/100g | Manufacturer                                      |
|-----|-------------------------------------|--------------|--------------|--------------------------------------------------|
| 1   | Golden Label powder cacao           | 522          | 420          | JSC Red October, Moscow, Russia                   |
| 2   | Smak powder cacao                   | 377          | 1770         | CJSC Presconita, Lithuania                        |
| 3   | Golden Anchor cacao                 | 325          | 870          | Bogorodskaya Confectionery Plant, LLC, Moscow, Russia |
| 4   | Alpen Gold chocolate (dark)         | 135          |              | Craft Foods, LLC, Moscow, Russia                  |
| 5   | Luker chocolate (dark)              | 115          |              | Casa Luker, Colombi                             |
| 6   | VAN fat free cacao                  | 105          | 800          | Maspex-GMV, Poland                               |
| 7   | Rossiysky powder cacao              | 97           | 1500         | JSC Rossiya Confectionery Plant, Samara, Russia  |
| 8   | Red October chocolate (bitter 80% cacao) | 66      |              | JSC Red October, Moscow, Russia                   |
| 9   | Vecherny Zvon chocolate             | 66           |              | JSC Rot Front, Moscow, Russia                     |
| 10  | NOIR AUTHENTIQUE chocolate          | 56           |              | FREY AG, Switzerland                             |
| 11  | Lux chocolate                       | 49           |              | JSC Babayevsky Confectionery Complex, Moscow, Russia |
| 12  | Ritter Sport chocolate with elite cacao from Ecuador (bitter 71% cacao) | 47     |              | Kakao Ritter GmbH & Co. KG, Germany              |
| 13  | Black chocolate (small lumps)       | 45           |              | JSC Rossiya Confectionery Plant, Samara; Nestle Russia, LLC, Russia |
| 14  | Nesquik Plus cacao                  | 45           |              | Wawel SA, Poland                                 |
| 15  | Wawel SA cacao (Naturalne cacao)     | 43           | 2570         | Craft Foods, LLC, Pokrov, Russia                  |
| 16  | Alpen Gold milk chocolate with whole hazelnuts | 34     |              | Cadbury Wedel Sp, Poland                         |
| 17  | Cacao Wedel E cacao                 | 32           | 2370         | Nestle SA, Switzerland                            |
| 18  | Hot Cocoa Mix cacao                 | 31           | 139          | JSC Rossiya Confectionery Plant, Samara, Russia  |
| 19  | Bitter chocolate 80% cacao          | 27           |              | JSC Red October, Russia                           |
| 20  | Coffee With Milk, chocolate milk with coffee | 26     |              | JSC Rossiya Confectionery Plant, Samara, Russia  |

Table 7. Total Content of Fat-Soluble Antioxidants (TCFA) in Nuts and Seeds (standard - gallic acid).

| No. | Name                                | TCFA mg/100g | Manufacturer                                      |
|-----|-------------------------------------|--------------|--------------------------------------------------|
| 1   | Walnut                              | 135          | Russia                                           |
| 2   | Hazelnut                            | 99           | Rosso-M, LLC, Russia                             |
| 3   | Pine nuts                           | 86           | Russia                                           |
| 4   | Cashew                              | 64           | Rosso-M, LLC, Russia                             |
| 5   | Chestnut                            | 63           | Russia                                           |
| 6   | Sunflower seeds                     | 60           | Rosso-M, LLC, Russia                             |
| 7   | Pumpkin seeds                       | 43           | Rosso-M, LLC, Russia                             |
| 8   | Almond                              | 43           | Rosso-M, LLC, Russia                             |
| 9   | Roasted peanuts                     | 40           | Rosso-M, LLC, Russia                             |
| 10  | Roasted hazelnuts                   | 36           | Hazelnut Promotion Group, Turkey                 |
| 11  | Californian almond                  | 21           | California, USA                                  |
Our databank contains TCA-related data for all kinds of tea: green, oolong, black, and puer [18]. In Table 8 are presented TCA data for green and black teas. Table 10 includes data for TCA content in coffee samples.

**Table 8.** Total Content of Antioxidants (TCA) in Green & Black Teas (standard – quercetin).

| No. | Name                                           | TCA mg/g | Manufacturer                      |
|-----|------------------------------------------------|----------|-----------------------------------|
| 1   | T-sips Ceylon tea in bags                      | 190      | Sri Lanka                         |
| 2   | Alokozay                                       | 171      | Dubai, UAE                        |
| 3   | Riston Green Exotic in bags                    | 155      | Sri Lanka                         |
| 4   | Lipton in bags                                 | 143      | Unilever Foodsolutions, USA       |
| 5   | Minamoto (pelleted leaves 100% of ecologically safe Japanese green tea) | 143   | Yunako Company, Japan             |
| 6   | Nama Cha, live green tea from Japan            | 140      | Japan                             |
| 7   | Tea Tang Sour Sap in bags                      | 138      | Sri Lanka                         |
| 8   | Azercay (Yastl cay) in bags                    | 133      | Azerbaijan                        |
| 9   | Greenfield flying Dragon in bags               | 130      | London, UK                        |
| 10  | Merlin                                         | 127      | CJSC Brand Tea, Sri Lanka         |
| 11  | Gift green tea                                 | 125      | Mirax Pharma                      |
| 12  | Nadin Super AOH Verbena                        | 125      | Vitali Tea                        |
| 13  | Green Elephant                                 | 125      | Sri Lanka                         |
| 14  | Impra Blackcurrant Green Tea (rich in antioxidants) | 124   | Sri Lanka                         |
| 15  | Selenium Green Tea                             | 124      | Wahan Mingcha Tea Industry Ltd, Hubei, China |

| No. | Name                                           | TCA mg/g | Manufacturer                      |
|-----|------------------------------------------------|----------|-----------------------------------|
| 1   | Darjeeling Tea Premium                         | 187      | Mlesna tea Naturally, bagged in Sri Lanka |
| 2   | Mabroc 1001 Nights in bags (mixture of black and green teas etc.) | 155   | Sri Lanka                         |
| 3   | Tea No.1 Darjeeling Rare Tea Flavor from Darjeeling | 150   | India                             |
| 4   | Darjeeling Tea                                 | 147      | Djukpana Plantation, India        |
| 5   | AKBAR Premium Quality Tea in bags              | 139      | Ceylon Tea, Sri Lanka             |
| 6   | Crown Gold Hilltop Collection Tea              | 124      | English Tea Collection, Sri Lanka |
| 7   | Beta Tea Black Tea (selected quality) in bags   | 118      | Beta Tea Groups, Turkey           |
| 8   | MLESNA Presidential Ceylon Baikhovi Tea in bags | 118   | MLESNA, Sri Lanka                 |
| 9   | Tea Lisma in bags                              | 113      | May Ltd., Russia                  |
| 10  | Heyleys                                        | 109      | Regeney Teas (PVT) Ltd., Sri Lanka |
| 11  | Bahar in bags                                  | 106      | Cornill Exports (PVT) LTD, Colombo – Sri Lanka British Blend, Sri Lanka |
| 12  | Greenfield fine Darjeeling in bags             | 106      | Greenfield Tea Ltd., UK           |
| 13  | Beseda                                         | 106      | Unilever Foodsolutions, Russia    |
| 14  | Darjeeling                                    | 104      | Chaygorod, Ltd., Russia           |
| 15  | Alokozay                                       | 102      | Dubai, UAE                        |

Table 9 demonstrates TCA values for natural ground coffee from various manufacturers. The major antioxidants in coffee are aromatic oxyacids: chlorogenic acid, caffeic acid, ferulic acid, protocatechic
Chlorogenic acid has the highest content in coffee, and a standard 200 mL cup with 10 g of brewed coffee may contain up to 300 mL of chlorogenic acid [19,20]. The degrees of coffee roasting vary from one country to another (low, medium, strong). When brewed, the antioxidant activity of coffee decreases; this is especially true for Robusta coffee.

Table 9. Total Content of Antioxidants (TCA) in Coffee (Ground) (standard – quercetin).

| No. | Name                          | TCA mg/g |
|-----|-------------------------------|----------|
| 1   | Maragogipe, Guatemala         | 32       |
| 2   | Brazil                        | 31       |
| 3   | Colombia                      | 31       |
| 4   | Maragogipe Chocolate, Santo-Domingo | 30     |
| 5   | Ethiopia, Yirgacheffe          | 29       |
| 6   | Puerto Rico Elite             | 29       |
| 7   | Decaffeinated coffee          | 27       |
| 8   | Kenya                         | 27       |
| 9   | Cuba                          | 27       |
| 10  | Jamaica elite                 | 26       |
| 11  | Sumatra                       | 25       |
| 12  | Mexico                        | 24       |
| 13  | Nicaragua                     | 24       |
| 14  | Guatemala                     | 24       |
| 15  | Honduras                      | 24       |
| 16  | Maragogipe Nicaragua          | 23       |
| 17  | Maragogipe Mexico             | 23       |
| 18  | Tanzania                      | 23       |
| 19  | Yemen Elite                   | 23       |
| 20  | Costa Rica                    | 23       |
| 21  | Australia Skyberry Elite      | 22       |
| 22  | Yava                          | 21       |
| 23  | Tchibo Exclusive              | 14       |

We have measured the total contents of antioxidants (TCA) in tens of samples of red wines obtained directly from the manufacturers at elite wine expositions or purchased in Moscow stores. The wines which have been studied included ones from Chile, France, Italy, Argentina, South Africa, Macedonia, Romania, Austria, Greece, Russia, Ukraine, Georgia, and Moldova. The study showed that the total content of antioxidants in natural good wines (in reference to quercetin as a standard) varies from 250 mg/100 mL to 100 mg/100 mL (Table 10). The contents of antioxidants in some wines purchased in stores were much lower (five-fold) than the indicated values, thereby suggesting possible product adulteration. Thus, by measuring the content of antioxidants, one may obtain additional information not only about the quality or the benefits of a red wine, but also about its authenticity. According to our measures, the antioxidant activity of white wines was 5-10 times lower than that of red wines; other data showed that it was 5-20 times lower.
Table 10. Total Content of Antioxidants (TCA) in Red & White Wines (standard – quercetin).

| No. | Name                          | TCA mg/100 mL | Manufacturer                        |
|-----|-------------------------------|---------------|-------------------------------------|
|     |                               |               |                                     |
|     | **Red Wines**                 |               |                                     |
| 1   | Vin de Perys D’OO             | 234           | France                              |
| 2   | Selection Cabernet Saperavi 2008 | 220         | Chateau Le Grand Vostok, Russia     |
| 3   | Don Segundo                   | 216           | Agricola Kantalehos, Chile          |
| 4   | Sunrise Merlo                 | 192           | Vina Concha y Toro, Chile           |
| 5   | Ruby Cabernet/Pinotage        | 185           | Winecorp PLS, RSA                   |
| 6   | Cabernet-Sauvignon Tamani     | 176           | Southern Wine Company, CJSC MPBK Ochakovo, Russia |
| 7   | Cabernet                      | 175           | Bovine, Macedonia                    |
| 8   | Merlo                         | 169           | Bovine, Macedonia                    |
| 9   | Blue Pat                      | 163           | Curico Valley, Chile                 |
| 10  | Mukuzani                      | 162           | Tamada, Georgia                      |
| 11  | Merlo-Cabernet                | 159           | Cape Town, Southern Africa           |
| 12  | Merlo                         | 159           | JV Lion Gri, Moldova                 |
|     | **White Wines**               |               |                                     |
| 1   | Stirbey                       | 63            | Provinum, Romania                    |
| 2   | Pino Gris                     | 45            | VJ Lion-Gri, LLC, Moldova            |
| 3   | Mtsevane                      | 36            | Tamada, Georgia                      |
| 4   | Mtsevane                      | 27            | Old Tbilisi, Georgia                 |
| 5   | STOPBANKS Savignon Blanc, 2007 | 18           | New Zealand                         |

Cognacs contain antioxidants originating from the source cognac spirit as well as tannins and other compounds which find their way into the final product by extraction from oak casks (Table 11). Whisky also contains antioxidants by means of extraction from oak casks. We have measured the total contents of antioxidants (TCA) in cognac samples from various countries (Table 11). TCAs for many cognacs which are not specified in the Table were very low, giving reason to believe that the data was misrepresented.

Table 11. Total Content of Antioxidants (TCA) in Various Cognacs, Brandy & Other Hard Liquors (standard – quercetin).

| No. | Name                                      | TCA mg/100 mL | Manufacturer                        |
|-----|-------------------------------------------|---------------|-------------------------------------|
|     |                                           |               |                                     |
|     | **Brandy/Cognac**                          |               |                                     |
| 1   | Guerin Freres (cognac)                     | 41            | Merle and Son, France               |
| 2   | Vartsikhe (brandy)                         | 33            | JSC David Sarajshvili & Eniseli, Georgia |
| 3   | Rossisiy (brandy)                          | 30            | CJSC Novokubanskoie, Krasnodor Region |
| 4   | Kutuzov (brandy)                           | 27            | Moscow Inter-republic Winery, Moscow |
| 5   | Henessy (cognac)                           | 26            | Ja Hennessy, France                 |
| 6   | Sarajshvili (brandy)                       | 25            | JSC David Sarajshvili & Eniseli, Georgia |
| 7   | Remi Martin V.S.O.P. (cognac)              | 22            | France                             |
| 8   | Akhtamar (brandy)                          | 22            | CJSC Yerevan Cognac Distillery, Armenia |
| 9   | Janneau V.S.O.P. Armanagnc, 7-year-old     | 19            | Janneau S.A., France               |
| 10  | Dacia, 25 years (brandy)                   | 17            | JSC Aroma, Moldova                  |
| 11  | Martell V.S.O.P. MEDAILLON (old fine cognac) | 15         | Martell &Co, France                |
| 12  | Baku (brandy)                              | 15            | Azerbaijan                         |
| 13  | Kievsky, 6-year-old (brandy)               | 14            | Moscow Wine & Cognac House KiN, Russia |
|     | **Whisky, special vodkas**                 |               |                                     |
| 1   | Ksenta Absinth                            | 53            | Torino, Italy                       |
| 2   | The Macallan Whisky, 12-year-old           | 11            | Scotland                           |
| 3   | Bocardi Rum, 8-year-old                   | 8             | Bahamas                            |
| 4   | MAGONY MEZES Meggy Palinka (Cherry with honey) | 7          | Hungary                            |
| 5   | GlenfiddichWhisky                         | 5.7           | Scotland                           |
| 6   | Black Label Whisky                         | 5.0           | Scotland                           |
| 7   | Scottish Collie Whisky                    | 4.9           | Scotland                           |
| 8   | Chivas Regal Premium Whisky               | 3.6           | Scotland                           |
| 9   | Red Label Whisky                           | 3.0           | Scotland                           |
| 10  | Tequila                                   | 1.6           | Mexico                             |
| 11  | Vinogradov, Special Limon Vodka           | 0.04          | CJSC Stolichniy Trest, Russia      |
| 12  | Stolichniy Doctor Recipe No.1 No Hangover Vodka | 0.03      | CJSC Stolichniy Trest, Russia      |
| 13  | Zelyonaya Marka Vodka                      | 0.00          | CJSC Topaz Distillery, Moscow Region, Pushkino |
| 14  | Bombay Gin                                | 0.00          | England                            |
Quite interesting data was obtained when detecting TCA in sprouted grains (Table 12). Some of these measurements have been performed for the first time [18]. Several conclusions can be made based on the data obtained. In all cases, without exception, the amount of antioxidants in sprouted seeds increases significantly over time. When evaluating a group of grain varieties based on this parameter, it may be noted that TCA in dry seeds is not high, and the data is related to the measurements of the same order. The content of antioxidants in naked oat (Tyumensky Naked-2, author V.V. Novokhatin) is slightly higher than in wheat or rye. It may be related to the higher immunity level of this culture. On Day 5, the amount of antioxidants increases significantly in the sprouts of all three grain varieties, and, although TCA of wheat sprouts is lower than that for naked oat, the intensity of its accumulation is somewhat higher. The significant amount of antioxidants in dry buckwheat seeds, most probably due to the presence of rutin in the plant. TCA increases only twofold during the germination process; however, it reaches a certain average level. TCA in beans is higher than in cereals, particularly, in chickpea and Chickasano pea. It turned out that the 5-day sprouts of these two cultures contain very high amounts of antioxidants (503 mg/100 g and 517 mg/100 g), and this amount increases, upon germination, by 6- and 5-fold, respectively. Black sesame seeds have the highest amount of antioxidants. TCA in 5-day sprouts of this culture increases by 1.7 times when compared to dry seeds, and it reaches a significant level (490 mg/100 g). TCA in squash and flax sprouts also significantly increases, although the level of antioxidants in seeds of these cultures is not significantly higher.

Table 12. Total Content of Antioxidants (TCA) in Seeds & Sprouts of Various Cultures Based onAbsolutely Dry Weight (standard – quercetin).

| No. | Culture                        | In Dry Seeds TCA mg/100 g | In Sprouts on Day 2 TCA mg/100 g | In Sprouts on Day 5 TCA mg/100 g |
|-----|--------------------------------|---------------------------|----------------------------------|----------------------------------|
| 1   | Black sesame                   | 291                       | 150                              | 490                              |
| 2   | Saint-Mary-thistle             | 235                       | 334                              | 896                              |
| 3   | Buckwheat                      | 182                       | 203                              | 383                              |
| 4   | Chickasano pea                 | 102                       | 263                              | 517                              |
| 5   | Chickpea                       | 84                        | 190                              | 503                              |
| 6   | Flax                           | 56                        | 201                              | 526                              |
| 7   | Corn                           | 42                        | -                                | -                                |
| 8   | Lentil                         | 42                        | 72                               | 90                               |
| 9   | Naked oat                      | 34                        | 65                               | 334                              |
| 10  | Squash                         | 33                        | 65                               | 333                              |
| 11  | Rye                            | 29                        | 102                              | 320                              |
| 12  | Zhigulevskaya Niva (rye)   germinated wheat (rye) in flakes, Meta-Lux, LLC, Zhigulevsk | 29 | - | - |
| 13  | Wheat                          | 24                        | 69                               | 275                              |
| 14  | Amaranth                       | 10                        | 17                               | 200                              |

Surprising results were obtained when analyzing amaranth seeds and sprouts. TCA in these seeds is only 10 mg/100 g – the lowest amount when compared to other cultures; however, by Day 5, this amount increases up to 200 mg/100 g, which is still not that high. However, the amount of antioxidants increases by 20 times during the germination process, which is twice as high when compared to the
rates of accumulation of such substances for other cultures. The results related to holy thistle are of particular interest. It is known that the seeds of this wonderful plant serve as a raw material for the production of various flavonoids – in particular, quercetin, – which are to be found in several pharmacological preparations. The amount of antioxidants in dry holy thistle seeds was sufficiently high (235 mg/100 g) but lower than, let’s say, in black sesame seeds. By Day 2, it increased and was already higher than for other cultures (334 mg/100 g); by Day 5, it became significantly higher (896 mg/100 g). The experiment was extended till Day 13, and by that time, the amount of antioxidants in the holy thistle sprouts increased up to 1,000 mg/100 g, getting quite ahead of black currant.

It should be also noted that 12 different species of plants whose sprouts were studied for the total content of water-soluble antioxidants are related to different species and varieties as well as to different families with distant phylogenetic properties. It gives us reason to believe that it is common for all higher plants to increase their amounts of antioxidants with seed germination. Apart from the seeds and their sprouts, we have also compared several products obtained from grains and buckwheat seeds in terms of the content of antioxidants. These products are widely used in daily diet. For this reason, it is especially important to measure them correctly.

3. Experimental

3.1. Methods of Antioxidant Activity Measurement

For the past decades, many methods aimed at determining the level of antioxidant activity have been proposed, including new reagents, model systems and devices. Many studies related to the antioxidant activity measurement methods have been published [15,21-28]. Various chemical and physicochemical methods are applied to measure the level of antioxidant activity (AA). Such methods are primarily based on direct or indirect measurement of the reaction rate or reaction completeness.

Three types among such methods, based on the following measurements, may be noted:

– Oxygen intake;
– Formation of oxidation products;
– Uptake (or bonding) of free radicals.

For the first and second cases, AA is determined based on the inhibition of the degree or rate of the intake of reagents or the products formation. There are several main methods for AA measurements: ORAC – oxygen radical absorbance capacity; TRAP – total radical trapping antioxidant parameter; FRAP – ferric reducing antioxidant power; TEAC (Randox) – Trolox equivalent antioxidant capacity; ABTS - [2,2-azinobis (3-ethylbenzthiazoline)-6-sulfonic acid; TBARS – thiobarbituric acid reactive substance.

In all of these methods, AA is the function of numerous parameters, including time, temperature, nature of the substance, the concentration of antioxidants and other compounds etc. The antioxidant activity cannot be measured directly; what is generally measured is the impact of antioxidants on the oxidation rate. All of these methods are not inter-correlated.

The disadvantage of many antioxidant activity measurement methods is the lack of proper substrates during the measurement process. Often, the antioxidant activity towards free synthetic long-living radicals (ABTS, DPPH, AAPH etc.) is measured. Various synonymous terms have been
proposed in the literature: “antioxidant ability”, “antioxidant power,” “antioxidant activity,” “antioxidant capacity” [26]. All of these terms are related to the antioxidant concentration (the activity of substances or substance groups). Many known methods (TEAC, TRAP, FRAP etc.) are based on reduction reactions of long-living free radicals or Fe (III) complex. Undoubtedly, these methods have their shortcomings as they all use synthetic free radicals, which have nothing to do with free radicals in human bodies.

3.2. Amperometric Method for Measurement of Total Content of Antioxidants

The amperometric method [29] is based on measurement of electric current resulting from oxidation of the substance (or the mixture) being studied on the surface of a working electrode at a certain voltage potential. The nature of the working electrode as well as the voltage potential applied determine the sensitivity of the amperometric method. The following are used as working electrode materials: glass carbon, gold, platinum, silver, copper, nickel, palladium etc. The voltage potential may be set from 0 to 2.5 V.

It has been established that the amperometric method has many advantages: low detection limit, high selectivity (only those compounds whose molecules can be oxidized are determined; other compounds which are present even in large concentrations are not determined), low cell volume (0.1-5 µL), and easy maintenance.

Compounds containing hydroxyl groups are oxidized well under amperometric detection conditions. The major and the most active natural antioxidants are phenolic compounds. These are natural polyphenols, various flavonoids, phenolic oxyacids, vitamins, etc. Thus, the amperometric method is best suited for the evaluation of antioxidant activity as well as the total content of antioxidants.

When detecting polyphenol compounds, the working electrode made of glass carbon is the most universal choice. The voltage potential may vary from 0 to 2.5 V; the ionization potential for phenol compounds varies from 100 to 1,200 mV. Electrochemical oxidation may be used as a model when measuring the activity level of free radical uptake as follows:

\[ \text{flavonoid-O-H} \rightarrow \text{flavonoid-O}^+ + \bar{e} + \text{H}^+ \] (oxidation at a maximum voltage potential)

The antioxidant activity may be measured by using the value of oxidation of such compounds on the working electrode of the amperometric detector. The signal is registered as differential dynamic curves. Special software calculates the peak areas (of the differential curves) of the analyzed and the standard substances. The average value of 3-5 consecutive measurements are used for analysis. The following common antioxidants may be used as standard substances: rutin, quercetin, dihydroquercetin, mexidol, trolox, gallic acid, etc. The amperometric device has many advantages when measuring the level of antioxidant activity. Without regard to sample prep, it only takes a few minutes to carry out a measurement. The analysis (registration and processing of the results) is performed in real time; the precise dosing by a six-port valve provides for correctness and precision of the analysis; the volume of the dosing loop may vary from 20 to 500 µL; SD for valve dosing is less than 0.5%; SD for consecutive measurements for the samples being analyzed < 3%; the detection level of the amperometric detector for polyphenols, flavonoids at the level of nano-picograms (10^{-9} – 10^{-12} g). With such low amounts, the probability of a mutual impact of various antioxidants, when they are collectively present, diminishes, in particular, the manifestation of synergy. Upon detection, the
The electrode surface is cleaned at a high positive voltage potential for 50-200 milliseconds, then it is recovered at a negative voltage potential for 100-400 milliseconds before the new cycle begins. The operation of the amperometric detector in the pulse mode remains stable for a long time. The behavior of signals towards the same antioxidant at various voltage potentials may also be used for identification purposes. No chemical agents (save for the standards) are needed for the analysis. For this reason, the cost of the analysis is very low. The most relevant objective related to the determination of antioxidant activity is to create a universal and relatively low cost method.

In our opinion, the amperometric technique meets such requirements more effectively than any other existing technique. The amperometric technique is especially good when comparing the antioxidant activities (AOA) of food products, various medications, beverages, and dietary supplements.

The amperometric method is the only one directly measuring the content of all antioxidants in a sample. Other methods are indirect; they measure the inhibition of reaction mixtures (particularly, free radicals), which have been generated through certain reactions [15, 21-24]. The amperometric method has been successfully applied to define the antioxidant capacities of various wines [30]. This study has shown that the method is direct, precise, objective, and quick. In [31], this method was applied to determine the antioxidant capacities of olive oils obtained from various Mediterranean countries, i.e. fat-soluble samples. The method allows evaluating the quality and authenticity of olive oil. Study [32] established the antioxidant activity of lipophilic compounds in vegetables, such as carotenoids, chlorophyll, tocopherol, and capsaicin. Based on pure compounds, their antioxidant activity was evaluated (in a decreasing order): lycopene > β-carotene > zeaxanthin > α-carotene > β-crytoxanthin > lutein > α-tocopherol > capsaicin > chlorophyll α > chlorophyll β > astaxanthin > santaxanthin. The measured antioxidant activity levels of extracts from five vegetable and two fruit species were compared by using ABTS. The good correlation between the methods was obtained, save for spinach. The authors of this paper have concluded that the amperometric method can be successfully used for direct, quick, and reliable monitoring of the antioxidant capacity of lipophilic extracts of food products. The amperometric (electrochemical) method is also applied to determine the antioxidant status of a person [33].

The general principles of the electrochemical detection of natural antioxidants are outlined in [34]. The amperometric method, in combination with high performance liquid chromatography, is wildly used to detect polyphenols in food products and beverages [35].

We have compared results of antioxidant capacity of freeze dried products and extracts using the ORAC assay and amperometric methods (total 23 products) [17,18]. We have obtained a good correlation for these products (0.96). Such a correlation was obtained for the first time with a large amount of various products (fruits, berries, etc.). It is probable that the correlation could have been better if the same substance, i.e. Trolox, were used in both methods. In case of positive results, when comparing these methods based on more data, the amperometric method may be used along with ORAC.

3. Conclusions

The original novel methodology by amperometric detection has been used to measure antioxidant capacity of different commercial available foodstuff sourced from different countries. We obtained a
large volume of data about antioxidants content in foodstuffs and beverages daily used, accessible to various strata of society. All results are presented in Tables 1-12 and allow the user to undertake a comparative analysis of antioxidant capacity of food from different sources. In many cases this can be correlated to the quality of products. This data can be used by experts of dietology, doctors for appointment of antioxidant therapy to different patients, and also to the people working in hard and harmful conditions, people who are exposed to frequent stresses, overloads (cosmonauts, miners), and to athletes before competitions, etc.

References

1. Fearon, I.M.; Faux, S.P. Oxidative stress and cardiovascular disease: Novel tools give (free) radical insight. *J. Mol. Cell. Cardiol.* **2009**, *47*, 372-381.

2. Valko, M.; Rhodes, C.J.; Moncol, J.; Izakovich, M.; Mazur, M. Free radicals, metals and antioxidants in oxidative stress – induced cancer. *Chem.-Biol. Inter.* **2006**, *159*, 1-40.

3. Knak Jensen, S.J. Oxidative stress and free radicals. *J. Mol. Struct.: Theor. Chem.* **2003**, *666-667*, 387-392.

4. Butterfield, D.A.; Lauderback, C.M. Lipid peroxidation and protein oxidation in Alzheimer’s disease brain: potential causes and consequences involving amyloid β-peptide-associated free radical oxidative stress. *Free Radic. Biol. Med.* **2002**, *32*, 1050-1060.

5. Cadenas, E.; Davies, K.J. Mitochondrial free radical generation, oxidative stress, and aging. *Free Radic. Biol. Med.* **2000**, *29*, 222-230.

6. Andreoli, T.E. Free radicals and oxidative stress. *Amer. J. Med.* **2000**, *108*, 650-651.

7. McCord, J.M. The evolution of free radicals and oxidative stress. *Amer. J. Med.* **2000**, *108*, 652-659.

8. Benett, J. P. Free radicals, oxidative stress and the origin of Parkinson’s disease. Journal of the Neurological Sciences. **1999**, *170*, 75-76.

9. Pellegrini, N.; Serafini, M.; Colombi, B.; Del Rio, D.; Salvatore, S.; Bianchi, M.; Brighenti, F. Total antioxidant capacity of plant foods, beverages and oils consumed in Italy assessed by three different in vitro assays. *J. Nutr.* **2003**, *133*, 2812-2819.

10. Halvorsen, B.L.; Holte, K.; Myhrstad, M.C.W.; Barikmo, I.; Hvattum, E.; Remberg, S. F.; Wold, A-B.; Haffner, K.; Baugerød, H.; Andersen, L. F.; Moskaug, J. Ø.; Jacobs, Jr. D. R.; and Blomhoff, R. A systematic screening of total antioxidants in dietary plants. *J. Nutr.* **2002**, *132*, 461-471.

11. Wu, X.; Gu, L.; Holden, J.; Haytowitz, D.B.; Gebhardt, S.E.; Beecher, G.; Prior, R.L. Development of a database for total antioxidant capacity in foods: preliminary study. *J. Food Compos. Anal.* **2004**, *17*, 407-422.

12. Wu, X.; Beecher, G.R.; Holden, J.M.; Haytowitz, D. B.; Gebhardt, S. E.; and Prior, R. L. Lipophilic and hydrophilic antioxidant capacities of common foods in the United States. *J. Agr. Food Chem.* **2004**, *52*, 4026-4037.

13. Holden, J.M.; Bhagwat, S.A.; Haytowitz; D.B. et. al. Development of a database of critically evaluated flavonoids data: application of USDA’s data quality evaluation system. *J. Food Compos. Anal.* **2005**, *18*, 829-844.
14. USDA Database for the Flavonoids Content of Selected Foods. Release 2.1. http://www.ars.usda.gov/nutrientdata/, accessed online January 2007.
15. Robards, K.; Antolovich, M. Analytical chemistry of fruit bioflavonoids. A review. Analyst 1997, 122, 11R-34R.
16. Sakakibara, H.; Honda, Y.; Nakagawa, S.; Ashida, H.; Kanazawa, S. Simultaneous determination of all polyphenols vegetables, fruits, and teas. J. Agr. Food Chem. 2003, 51, 571-581.
17. Nemzer, B.V.; Yashin, A.Y.; Yashin, Y.I.; Chernousova, N.I. Comparison of the study of the antioxidant activities of fruits and vegetables by oxygen radical absorbance capacity and amperometric methods. In Malta Polyphenols 2007, Proceedings of 4-th International Conference on Polyphenols Application, La Valetta, Malta, November 14-16, 2007.
18. Yashin, Y.I.; Ryzhnev, V.Y.; Yashin, A.Y.; Chernousova, N.I. Natural Antioxidants. Content in Food Products and Its Influence on Human Health and Aging; TransLit: Moscow, Russia, 2009; p. 212.
19. Richelle, M.; Tovazzi, I.; Offord, E. Comparison of the antioxidant activity of commonly consumed polyphenolic beverages (coffee, cocoa, and teas) prepared per cup serving. J. Agr. Food Chem. 2001, 49, 7, 3438-3442.
20. Yashin, A.Y.; Chernousova, N.I.; Fedina, P.A.; Levin, D.A.; Mironov, S.A. Determination of content of antioxidants in coffee by amperometric method. Beer Bever. 2009, 2, 45-47.
21. Perez, D.; Leighton, F.; Aspee, A.; Aliaga, C.; Lissi, E. A Comparison of methods employed to evaluate antioxidant capabilities. Biol. Res. 2000, 33, 1-10.
22. Llesuy S.; Evelson P.; Campos A.M.; Lissi E. Methodologies for evaluation of total antioxidant Activities in complex mixtures. A critical review. Biol. Res. 2001, 34, 1-22.
23. Khasanov, V.V.; Ryzhova, G.L.; Maltseva, E.V. Antioxidants research methods. Chem. Plant Raw Mater. 2004, 3, 63-95.
24. Roginsky, V.; Lissi, E.A. Review of methods to determine chain-breaking antioxidant activity in food. Food Chem. 2005, 92, 235-254.
25. Cao, G.; Prior, R.L. Comparison of different analytical methods for assessing total antioxidant capacity of human serum. Clin. Chem. 1998, 44, 1309-1315.
26. Braynina, K.Z.; Ivanova, A.V.; Sharaftudinova, E.K. News from universities. Evaluation of antioxidant activity of food products by using potentiometry method. Food Technol. 2004, 4, 73-75.
27. Korotkova, E.I. New method of determining antioxidant activity. J. Phys. Chem. 2000, 74, 1544-1546.
28. Korotkova, E.I.; Karbainov, Y.A.; Shevchuk, A.V. Study of antioxidant properties by voltammetry. J. Electroanal. Chem. 2002, 518, 56-60.
29. Yashin, A.Y. Use of HPLC with ABP in Vital Areas: Medicine, Analysis of Food Products, Ecology. In Chromatography for the Benefit of Russia; Moscow, Granitsa Publishing: Moscow, Russia, 2007; pp. 390-420.
30. Mannino, S.; Brenna, O.; Buratti, S.; Cosio, M.S. A new method for the evaluation of the “antioxidant power” of wines. Electroanalysis 1998, 10, 908-912.
31. Mannino, S.; Buratti, S.; Cosio, M.S.; Pellegrini, N. Evaluation of the “antioxidant power” of olive oils based on a FiA system with amperometric detection. Analyst 1999, 124, 1115-1118.
32. Buratti, S.; Pellegrini, N.; Brenna, O.V.; Mannino, S. Rapid electrochemical method for the evaluation of the antioxidant power of some lipophilic food extracts. *J. Agr. Food Chem.* **2001**, *49*, 5136-5141.

33. Hansley, K. Evaluation of oxidative stress of HPLC by electrochemical detection. *J. High Res. Chromatogr.* **1999**, *22*, 429-437.

34. Kilmartin, P.A. Electrochemical detection of natural antioxidant: principles and protocols. *Antioxid. Redox Signal.* **2001**, *3*, 941-955.

35. Dapkevicius, A.; Van Beek, T.A.; Niederlander, H.A.G. Evaluation and comparison of two improved techniques for the on-line detection of antioxidants in liquid chromatography eluates. *J. Chromatogr.* **2001**, *912*, 73-82.

*Sample Availability: Samples are available from the authors.*

© 2010 by the authors; licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution license (http://creativecommons.org/licenses/by/3.0/).