

## Introduction: White Papers from the “First International Congress on Antioxidant Methods”

Antioxidants have become a major area of focus in papers published in the *Journal of Agricultural and Food Chemistry* over the past five years. The topics of these papers have included many aspects of antioxidants and the chemistry associated with them. Antioxidants perform many functions ranging from phytoprotectants in the plant, to protecting lipids in food products, to in vivo antioxidant activity in animals and humans, including scavenging reactive oxygen species, acting as antagonists toward oxidative enzymes such as cyclooxygenases, and influencing the expression of multiple genes. The types of studies reported have ranged from compositional studies of a variety of plant species to multiple approaches for the measurement of antioxidant or oxygen scavenging capacity. Many approaches have also been reported using various cell culture models to demonstrate activities including control of cell proliferation, changes in apoptosis, and regulation of gene expression. The editorial staff of the Journal became concerned because there was little consistency in methods that were being reported and there were few papers that used multiple methods; thus, comparison of activities and reported results was impossible. It was also clear that without consistency of methods and the ability to compare results, a regulatory agency would have a very difficult time assessing requests to support claims for a product. During the same period the number of papers received in this topic area has increased dramatically. To address these issues, the editorial staff has implemented increasingly higher standards for publication of papers in this area. These standards, which will be clearly defined in a future editorial by the Editor, include a focusing on sources of raw material that are closely associated with agriculture or agricultural products. Papers relating to herbs that are not commonly cultivated may be more appropriately directed to the *Journal of Natural Products*. Papers reporting new or different measures of antioxidant capacity should clearly describe the methods used. If the method is new, it must be clearly described, and if it is a modification of an existing method, changes need to be described clearly and justified. Papers describing antioxidant or biological activity from an agriculturally relevant plant must include chemical analysis of the starting materials, particularly on components responsible for the purported activity. The most significant effort by the Journal was to lead the convening of the First International Congress on Antioxidant Methods to discuss the state of methods related to antioxidants and attempt to drive standardization of methods. It is important to recognize that methods will evolve over time and that no one method will address all of the problems.

The First International Congress on Antioxidant Methods was held in Orlando, FL, June 16–19, 2004. One hundred and forty scientists from 18 countries participated.

The discussions included guidelines for future publication in journals, protocols for submission and conducting of Association of Official Analytical Chemists (AOAC) collaborative assays,

and current status of standard methods for antioxidants of the American Oil Chemists' Society (AOCS).

The heart of the meeting focused on chemical methods for antioxidant content and activity in model systems and foods, measurement of antioxidant capacity in food and biological systems, and the in vitro and in vivo methods used to estimate effectiveness in animals and man. There were stimulating and often intense debates around methodologies. The three papers that follow this introduction address three of the areas discussed at the meeting.

The first paper by Prior et al. addresses the measurement of total phenolics and antioxidant capacity. One method that is useful for the measurement of antioxidants in many systems is the Folin assay for total phenolics. There is an issue of interference by ascorbic acid, which must be resolved. A method is recommended in the Prior paper. This method will be validated in a single laboratory and then proposed for a collaborative study through the AOAC. When this is done, it was suggested that this should be considered a benchmark for future publications.

In the papers following, the current status of methods being used is discussed and certain methods are suggested to be evaluated for standardization. For chemical estimation of antioxidants in foods three methods are being proposed in the paper by Prior et al.: oxygen radical absorbance capacity (ORAC), the Folin–Ciocalteu method, and possibly the Trolox equivalent antioxidant capacity (TEAC). ORAC is based on a hydrogen atom transfer reaction mechanism, which is relevant to human biology. The Folin–Ciocalteu method is an electron transfer based assay and gives reducing capacity, which has normally been used to estimate phenolic contents of biological materials. The TEAC assay represents a second electron transfer based method. Although none of these assays are perfect, they will serve as a starting point. The relevance to biological systems is yet to be established for these methods. These methods also are questionable when antioxidants are at the aqueous/oil interface. As the methods become standardized we also need to guard against ORAC horsepower races between products and ingredients. These are tools for estimating, and cross-product comparisons will be possible, but the relationships to health benefits are not established.

The complexity of food matrices adds a complexity to the estimation of antioxidant activity. To accurately evaluate antioxidants in foods, models must account for the chemical, physical, and environmental conditions specific to a particular food product. The paper by Decker et al. outlines model systems for the evaluation of antioxidants in three types of foods: bulk oil; oil-in-water emulsions, and muscle foods. The three models, although not all inclusive, do measure lipid oxidation and antioxidant activity. However, when standardized, these models could allow researchers to more easily compare results from one laboratory or paper to another.

Testing for bioactivity in cell culture systems is a rapidly emerging tool in many fields of research, particularly the effects of antioxidants on particular cell types. More research is needed to develop cell-based antioxidant activity assays that account for digestion, absorption distribution within the body, metabolism, and excretion of antioxidants. Mechanism-based cell

culture models will likely become valuable tools in future antioxidant research, although the technology is too young for standardization at this time.

The follow-up for the meeting and further discussion of the papers will be at the Second International Congress on Antioxidant Methods, June 22–24, 2005, in Orlando, FL.

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JF058079K