the systematic application of their art to produce products pleasing to the increasingly knowledgeable consumer base that enjoys wine as part of its civilized society.

The process of making wine involves a series of concerns for the grower, as well as the winemaker. The first concerns are viticultural, including delivery of sound, high-quality fruit at optimal maturity. Upon arrival at the winery, fruit quality is assessed, the grapes are processed, and fermentation is begun. Almost immediately, and in many instances simultaneously, chemical and microbiological stability of the young and/or aging wine becomes important. Finally, problems occur on occasion requiring utilization of remedial techniques to produce an acceptable product.

Production considerations serve as the framework in organizing this book. Within each chapter is information culled from the authors' collective years of experience, as well as from the literate wine community around the world. These chapters provide numerous practical, as well as fundamental, insights into the various aspects of the process. Winemakers will benefit from these insights while still maintaining (and gaining further insight into) their own.

Analytical techniques have become valuable tools of modern winemakers wishing to better understand their product. These analytical tools are another major feature of this text. The authors have gathered numerous procedures commonly used for grape, juice, and wine analysis. These procedures are presented as they are generally practiced in the industry around the world. We have formatted them into an easy-to-follow recipestyle to make them more useful to the winery technician. Our procedures provide instructions for preparing required reagents, stains, and media, and then outline the analyses in detail. To make these procedures more accessible, we have gathered them into a single chapter at the end of the "text" chapters. In addition to the "standard" laboratory procedures we have included a section of rapid "diagnostic" tests to assist in identifying problems encountered during winemaking. Several frequently used conversion and correction tables have also been collected into one section for the reader's convenience. Finally, we have provided some information on the safety aspects of the various reagents employed in conducting the laboratory analyses.

In developing material for this text, the authors have emphasized analyses as they would be carried out in a production laboratory. Realizing that different laboratories have different analytical capabilities, personnel, and equipment, we have in many instances provided several different approaches to the same analysis. Throughout this book we have given special attention to practical considerations and their importance in the total spectrum of winery operations. We have done the same with the laboratory Also forthcoming are improved and simplified analytical techniques required for the analysis of 2,4,6-trichloroanisol and other compounds that may be implicated in causing cork taint. Sulfides present another analytical challenge in need of a solution. Wines may be bottled with no apparent sensory problems and develop perplexing and annoying sulfide-type aromas some months later. Although some analytical methods are available for the determination of these compounds at the detection limits required, such methods are generally too complex for use on a day-to-day basis.

Another issue of concern in the wine industry is increasing demands for regulatory compliance, thus increasing the demand for analyses of trace metals and agricultural chemical residues. Attempts to characterize wines using "signature" or "fingerprint" techniques have captured the attention of those in the wine industry. The signature concept holds the potential for allowing the analyst to trace authenticity of a wine to grape variety (including clonal selection), appellation, and vintage.

Historically, characterization of wines has been (and still is) conducted using sensory analysis. The nose remains the most sensitive analytical tool. However, at present quality control and regulatory requirements are forcing wineries to perform more analyses for both major and minor components. The necessity of testing for residues will be a powerful driving force toward more sophisticated instrumental methods in the industry. Finally, it is the interest in producing even higher quality products, and the search for more definitive information that cause winemakers to ask for better analytical measurements. It is safe to speculate that the number and quality of wine analyses performed will increase and these analyses will be accomplished using increasingly advanced analytical techniques. pathogens (Malamy and Klessig 1992; Muller et al. 1994; Raskin 1992a, 1992b; Sieman and Creasy 1992; Yalpani and Raskin 1993).

Of these defense chemicals, phenolics, both simple and complex, are ubiquitous in plants; they are plethoric in grapes and wine (Bachman 1978; Cartoni et al. 1991; Gorinstein et al. 1993; Kanner et al., 1994; Kinsella et al. 1992; Mahler et al. 1988; Muller et al. unpublished data; Salagoity-Auguste and Bertrand 1984; Scholten and Kacprowski 1993; Shahidi et al. 1992; Singleton and Esau 1969; Steffany et al. 1988; Stich and Rosin 1984; Torle et al. 1986). However, defense is not the only function of some of these phenolics. Skin pigments, for instance, also have the specific function of attracting useful predators to the fruit for propagation of the species.

## **Antioxidant Function**

Whether a compound functions as an antioxidant in biological systems depends not only on its intrinsic chemical structure but also, and perhaps more important, on the biochemical environment in which it operates. It is also crucial that a putative antioxidant be capable of reaching the target cells or tissues and, in addition, that once there, be present in sufficient concentration to effectively take care of the biological insult (Halliwell 1990). Further, it is also imperative that once it has performed its beneficial function, the now spent antioxidant be efficiently removed to prevent further interactions with surrounding species.

Thus, in characterizing an antioxidant in grapes or wine, one must be careful and cognizant of the above constraints before making claims on a compound as an antioxidant in humans simply because such compound might work in vitro or ex vivo (Halliwell 1993e; Muller and Fugelsang 1994c).

## Antioxidant-Pro-oxidant

Another question that must be asked is: What happens to an antioxidant once it has performed its defensive duties? In most instances, unfortunately, our knowledge of their biological fate is not fully understood. Neither is the fate of other potential target molecules that could be attacked by the now spent (oxidized) antioxidant. Many antioxidants might thus become pro-oxidants (Aruoma et al. 1993; Freisleben and Packer 1993; Halliwell 1990; Laughton et al. 1989; Smith et al. 1992; Snyder and Bredt 1992; Stich 1991). Whether a compound acts as an antioxidant or as a pro-oxidant depends on its oxidation state and that of other species with which it can interact. In reactions of this type, electrons move according to a rigorous hierarchy of electrochemical potentials (Koppenol 1985, 1990). Under these circumstances, iron-initiated damage by hydroxy- (HO·), peroxy- (O<sub>2</sub>·), nitric oxide (NO), and other active radicals might be considerable (Beckman and Crow 1993; Choi 1993; Cochrane 1991; Graf et al. 1984; Halliwell and Gutteridge 1992; Puppo and Halliwell 1988).

## Antioxidant Effect on Glutathione

Antioxidants might also work by helping retain the integrity of certain crucial sulfhydryl (thiol) groups such as those in glutathione (Asmus 1990; Beutler 1989; Butler and Hoey 1992). Glutathione is required in a myriad of homeostatic mechanisms in the body as varied as protection of mitochondrial DNA in the human brain (Ames 1989), intestinal absorption of amino acids (Meister 1973), and prevention of cataract formation (Bando and Obazawa 1988). Further, glutathione is required in many detoxification mechanisms including those of medicines (e.g., acetaminophen) and other xenobiotics. Glutathione is easily oxidized by endogenous and exogenous substances such as activated oxygen and radiation (including light) to the disulfide, a compound that offers lessened, if any, protection against oxidative insults (Asmus 1990; Beutler 1989; Meister 1992; Yu 1994).

In the eye, glutathione protects by scavenging free radicals formed photochemically. Formation of free radicals there eventually causes cross-linking of sulfhydryl groups in proteins with resultant formation of opacities and eventually blindness, unless protection in the form of an antioxidant such as glutathione is present in both the lens and in the vitreous humor of the eye (Bando and Obazawa 1988; Blondin et al. 1987; Spector et al. 1993).

Youngsters (and adults) often watch endless hours of television. Their proclivity to remain in close proximity to a source of strong photic stimulation might endanger their visual elements prematurely, unless their diet includes significant amounts of antioxidants. Dietary antioxidant requirements may be higher for people who spend a significant portion of their working time in front of a computer screen.

By their action on glutathione, if not directly, wine antioxidants might help prevent diabetes and associated sequelae such as visual loss. Antioxidants might also protect against diabetes caused by iron overload (hemochromatosis), a condition to which several ethnic groups are genetically predisposed (Halliwell and Gutteridge 1985a, 1985b). Some antioxidants that possess vicinal enediol structures (such as ascorbic acid and 2,3-DHB) might be useful in removing excess iron in conditions of iron overload (Muller, unpublished data). In addition, antioxidants might help prevent certain forms of liver damage, including cirrhosis and cancer (Kennedy and Tipton 1990; Poli et al. 1993). Protecting the integrity of reduced