Summary: Life as we know it heavily relies on biological catalysis, in fact, in a very nonromantic version of it, life could be considered as a series of chemical reactions, regulated by the guarding principles of thermodynamics. In ancient times, a beating heart was a good sign of vitality, however, to me, it is actually the presence of active enzymes that counts… Though we do not usually pay attention, the history of enzymology is as old as humanity itself, and dates back to the ancient times. This paper is dedicated to these early moments of this remarkable science that touched our lives in the past and will make life a lot more efficient for humanity in the future. There was almost always a delicate, fundamentally essential relationship between mankind and the enzymes. Challenged by a very alien and hostile Nature full of predators, prehistoric men soon discovered the medicinal properties of the plants, through trial and error. In fact, they accidently discovered the enzyme inhibitors and thus, in crude terms, kindled a sparkling area of research. These plant-derivatives that acted as enzyme inhibitors helped prehistoric men in their pursuit of survival and protection from predators; in hunting and fishing… Later in history, while the underlying purposes of survival and increasing the quality of life stayed intact, the ways and means of enzymology experienced a massive transformation, as the ‘trial and error’ methodology of the ancients is now replaced with rational scientific theories.

Keywords: enzyme, drugs, inhibitors, art, history, industry

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Kratak sadržaj: Život oko nas uglavnom se zasniva na biološkoj katalizi, pa bi se čak, u nekakvoj neromantičnoj priči, život mogao opisati kao niz hemijskih reakcija koje reguliraju glavni principi termodynamike. U davnjoj prošlosti, kućanje srca bilo je znak dobrog zdravlja, međutim, za mene, najvažniju ulogu ima upravo prisustvo enzima… Mada ne obračamo često pažnju na to, istorija enzimologije stara je koliko i sam ljudski rod i seže daleko u prošlost. Ovaj rad posvećen je baš tim počecima u ovoj veoma zanimljivoj nauči, koja je u prošlosti ostavila traga na našim životima i koja će u budućnosti učiniti da život bude mnogo produktivniji za čovečanstvo. Između čoveka i enzima postojalo je oduvek jedan delikatan, suštinski važan odnos. Suočen s nepoznatim i neprijateljskim nastrojenom prirodom punom grabljivaca, preistorijski čovek rano je otkrio lekovita svojstva biljaka, a to metodom probe i greške. Štaviše, slučajno su otkriveni inhibitori enzima, čime je, grubo rečeno, još tada otvorena nova oblast istraživanja. Ovi biljni derivati koji su imali ulogu inhibitora enzima pomagali su preistorijskom čoveku u njegovoj borbi da opstane i zaštiću se od grabljivaca; prilikom lova i ribolova… Kasnije kroz istoriju, sa nepremenjenom svrhom opstanka i poboljšanja kvaliteta života, enzimologija je iz kojena transformisana, a metodologiju »probe i greške« iz preistorije zamenile su racionalne naučne teorije.

Ključne reči: enzim, lekovi, inhibitori, umetnost, istorija, industrija
Introduction

Life begins with a catalytic activity. The importance of catalytic activity was annotated by Kleczkowski and Garncarz (1) in a very striking manner, that is, 'the original environment of life on Earth was seawater containing micrornutrients with structural, metabolic and catalytic activity'. Today, we know that germination of spores begins with macromolecular synthesis (2).

Historical relevance of clinical enzymology begins with Francis Home in 1780, through his studies on the detection of glucose in urine (3). Catalysis per se as a term was first introduced by J.J. Berzelius in 1836 (4). Richard Bright, on the other hand, dedicated his work to the investigation of 'proteinuria' by heating up urine with a candle flame in a tablespoon (1789–1858) (5). After many subsequent works by the 19th century scientists, it became evident that digestion was in fact done by the relevant enzymes (6). Scientific enquiries in clinical enzymology began with rudimental tests, mostly performed haphazardly, and this later paved the way for more sophisticated methods such as nuclear magnetic resonance and mass spectrometry (5). Nowadays, moreover, there are myriads of different, equally fascinating areas of research on enzymes; diseases, metabolic significance, catalytic mechanisms, novel roles and interactions with various chemicals, drugs, agonists, metals are only a few of them... (7–15). Applications of computational protein studies give different points of view for investigation and designing novel catalysts from scratch. Through these delicate, meticulous approaches, clarification of protein-protein interactions and inhibitors of various molecules and large oligomeric assemblies is made possible (16). With the project 'Enzyme Function Initiative', defining sequence-structure interaction/interdependence would provide the prerequisite infrastructure for accurately predicting the in vitro functions of previously unknown enzymes and open the door for much more challenging studies (17). Both in the near and distant future, life might require and depend on designing state-of-the-art application areas of enzymes for novel scientific outcomes in daily life in addition to their benefits in medicinal therapies (18–20).

Enzymes are Vital for Every Part of Life

With the limited evidence, the daily, monotonous life of ancient humans is hard to predict with precision, yet one thing known for sure is that they also required various food sources to sustain life. Based on the preexisting knowledge in addition to newly emerging data, we are putting pieces together and trying to predict and clarify which enzyme-bound products they used.

What was the role of fermentation in the emergence of enzyme products in the ancients’ life? In fact, was the first enzyme product a fermented one or not? Which came first? Bread? Wine? Vinegar? Beer? Soy sauce? Kumis? Sake? Koji? Kefir? Sour cream? Pickles? Sauerkraut? Sourdough? Yoghurt? Boza? Kimchi? Miso? Tempah? Or perhaps a completely distinct, unknown enzyme product was consumed that unfortunately left the scene with no trace for us to follow.

Neanderthal man’s food culture began to emerge approximately in 30,000 BC with the introduction of bread (21) and, accordingly, it is very hard to find any documented evidence on this subject. Bakery technology begins with the cultivation of wheat in Göbekli Tepe that is, with its 11,600 years old existence, also the world’s oldest temple. Sourdough starters or yeast was used as a food additive in bread baking, however, the exact or approximate date is not clear (22). Dairy products were another important food source in ancient societies. Cheese is an effortless candidate to claim the role of being the first enzymatic product. And the use of milk dates back to 8,000 years ago and it evolved in the ‘Fertile Crescent’ between the Tigris and Euphrates rivers (23).

Koumiss was an alcoholic beverage (V century BC) of ancient times in central Asia and was used in the treatment of various diseases such as phthisis (24). Yogurt and kefir are also among the ancient traditional dairy products and were used extensively as preventive compounds against diseases, and for curative purposes. The nomads carried fresh milk in bags, probably crudely made from animals’ stomach, and the milk, in this relatively perfect condition, fermented into yogurt or kefir (25). The origins of wine and vinegar go back to as far as 2,500 BC. Hammad-rabi tablets, casted in stone, are perhaps the first documented artifacts on fermentation of grapes. These tablets date back to 2,100 BC and again this was probably the first document on the commercial use of enzymes. The second commercial product of the enzyme origin might have been vinegar, which was widely used in those days both for medicinal purposes as a painkiller, and in disinfection of wounds and finally in food storage (26). However, C. Wang and his colleagues, using five analytical methods to identify the chemical constituents of the potteries originating from China, found out that the emergence of various fermented products dates back to as early as 7,000 BC (27). On the other hand, the origin of sake is an oblique one. Sake may be as old as wine. It goes back as far as 2,500 years when the cultivation of rice became widespread. The most mysterious and acutely necessary component of sake is koji which contains alpha-amylase, beta-amylase proteases, peptidases, sulfatases enzymes. Traditionally, koji is also used in soy sauce and in miso. Natto is a traditional, dateless Japanese meal and the enzyme found in natto has been named nattokinase (28).
Smell or the occurrence of various odors, again an interesting façade of enzyme catalyzed reactions, is another, crucial factor in survival. The first study on enzymatic catalysis and the related occurrence of odors was performed approximately four decades ago. Investigators, for instance, studied the effect of γ-irradiation on cystein sulfoxide lyase and odor of onions (29). Odors originating from the catalytic activities of the enzymes are potent regulators of biological functions. Jasmonic acid, a highly volatile terpene compound, shows its effect on protein kinases or transcription factors (30). Odor evoked behaviors or social interactions show their effect on the enzymes such as flavin containing enzyme monooxygenase-3 (31). In the determination and approximate diagnosis of various health problems, dogs and cats smell some of the diseases such as cancer, epileptic seizures, and hypoglycemia (32, 33).

Meat tenderizer, which contains the enzyme papain, has been recommended for the treatment of bee (34) and fire ant (35) stings. It was also established that these meat tenderizers were safe and effective for patients with a phytobezoar (36).

**Enzyme Inhibition**

Medicine is a cumulative pile of interactions from the collective components of science, technology, and human values, beginning from the ancient times (37). Poisons, in other words enzyme inhibitors, are widespread in plants and animals and they were used – and are still in use – for hunting as arrow poison, in fishing (38) or in even warfare (39). The history of enzyme inhibitors is as old as humanity (40). However, the very first documented experiment on enzyme inhibition was done on proteolytic enzymes with formaldehyde in 1899 (40).

**Enzymes and Art**

In his article, Irvine H. Page proclaims that ‘Scientific research is, in many ways, related to art’ (41). Enzyme inhibitors have special roles in the production and stimulation of artwork! The whole assumption might seem irrelevant and out of context, but the roads of enzymes and artifacts might effortlessly entangle. One of the known art effector molecules is Thujone, which is widely established as a severely neurotoxic compound. A few liver enzymes are responsible in the metabolism of Thujone; cytochrome P450 (CYP), CYP2A6, CYP3A4, and CYP2B6. Thujone inhibits gamma-aminobutyric acid A (GABA) receptor (42, 43). It was discovered that CYP2A6 is the key enzyme in the metabolism of Thujone in human liver (43). The most interesting and appealing point of Thujone is that it is present in the notoriously famous absinthe drink (42). A number of great artists and writers from the late 1800s used absinthe as a social drink, including Vincent van Gogh and Toulouse-Lautrec (44). There are also implications for the ailments of Vincent van Gogh that involve heavy absinthe consumption (45). Furthermore, another enzymatic product, ‘vinegar’, is the main subject of a beautiful painting in Rijksmuseum. As a matter of fact, Cleopatra’s knowledge of chemistry allowed her to win a bet over Antony (46).

**Enzyme Inhibitors and their Effects on Warfare**

In the Second World War (WW2), Nazi Germany used acetylcholinesterase inhibitors such as tabun, sarin, and soman as chemical weapons. And following this initial experience in war, widespread weapon development programmes have been initiated in the exploration of various neurophysiological and neurotoxicological chemical compounds (47). These types of studies have opened a new window on enzyme inhibitors such as medical treatment in poisoning with organophosphorus compounds (48) and provided new insights on the reactivation of acetylcholinesterases (49).

**Enzyme Inhibitors and Cosmetic Beauty**

Sirtuins are associated with cellular energy metabolism and the redox state of the cell through their interactions with multiple signaling and survival pathways. It is accepted that activation of sirtuins is a valuable therapeutic target against aging and age-related diseases, including the renal diseases (50). Aging has now been defined both as a cellular and molecular event. And sirtuin-activating and anti-glycation products are already being marketed by cosmetic and pharmaceutical companies (51). Botulinum toxin, likewise, is an enzyme that enters peripheral cholinergic nerve endings and specifically and selectively cleaves one or more SNARE proteins to induce paralysis (52). Most patients and physicians are using monotherapy by the botulinum toxin for a natural look that softens wrinkles for the upper face profile (53). Recent studies show that botulinum toxin has many other roles in medicine (54).

**Enzyme Inhibitors as Therapeutic Compounds**

As a rule, most of the drugs derived from food products show their effect on enzyme inhibition (55). Target-based drugs and identifying novel and safe chemotherapy targets with particular emphasis on the inhibition of key reactions in metabolic pathways are many a researcher’s dream (56). Understanding the role of enzymes in various diseases and their detailed mechanisms is among the important aims of the enzyme inhibition studies (57). Antibiotics were first
### Table I: Analysis of the historiography of enzymes.

| Year       | Finding                                                                 | Ref |
|------------|-------------------------------------------------------------------------|-----|
| 1683–1757  | First biochemical experiment and first enzyme specificity experiment    | 81  |
| 1729–1799  | Effect of concentration, temperature, time on enzymes (were unaware of enzymes) | 81  |
| 1833       | Something converts starch into sugar and is named diastase (the suffix ‘ase’ comes from diastase) | 82  |
| 1835       | Science world gained the word ‘catalysis’                                | 83  |
| 1836       | Theodor Schwann discovered that gastric juice contains a digestive substance and he named this substance pepsin | 84  |
| 1876       | Wilhelm Friedrich Kühne used a Greek word ‘enzymos’                      | 81  |
| 1876       | Studies on glycolysis were begun in this year by Bernard. Disappearance of sugar from blood upon being left for 24 hours at room temperature | 85  |
| 1884       | Enzymes of the digestive tract of fishes. First publication in PubMed    | 86  |
| 1890       | Emil Fischer proposed the ‘lock and key’ model                          | 81  |
| 1897       | Bertrand partially purified the enzyme laccase from tree sap            | 81  |
| 1897       | The Buchner brothers investigated the first cell-free assay             | 87  |
| 1897       | Fermentation explained; homogenisation and filtration of yeast and naming of filtrate as ‘zymase’ | 88  |
| 1901       | Enzyme theory; for every vital reaction a specific enzyme exists         | 89  |
| 1902       | Victor Henri published the first successful mathematical model for describing enzyme kinetics | 90  |
| 1903       | Enzymes are realized in tumour                                           | 91  |
| 1910       | Beginning of the study of glycolysis                                     | 93  |
| 1914       | Description of the coenzyme and cofactor cozymase zymin respectively    | 92  |
| 1915       | First use of a spectrophotometer                                         | 94  |
| 1915       | Effects of acids and salts upon enzyme activity (amylase)                | 95  |
| 1923       | Synthesis of urea with urease and understanding of the importance of enzymes | 96  |
| 1925       | Use of digestive enzyme in therapeutics                                  | 97  |
| 1926       | First enzyme purification by James B. Sumner                            | 98  |
| 1940s      | Completion of the glycolysis pathway                                     | 99  |
| 1910–1940  | Citric acid cycle                                                       | 99  |
| 1943       | Biochemists began to speak on enzyme deficiencies                       | 100 |
| 1947       | Cori cycle was explained                                                | 101 |
| 1948       | Fawism was explained                                                    | 102 |
| 1952       | The C-terminal residue of lysozyme                                       | 103 |
| 1953       | N-terminal sequence of carboxypeptidase                                  | 104 |
| 1956       | Explanation of the pentose phosphate pathway                            | 105 |
| 1959       | Koshland explained the induced fit model                                | 106 |
| 1960       | Became aware of isoenzymes                                              | 107 |
| 1960       | Began to talk about the secondary or tertiary structure of trypsinogen  | 108 |
| 1962       | Partial determination of the primary structure of the enzyme            | 109 |
| 1962       | Three-dimensional structure of lysozyme                                 | 110 |
| 1963       | Completed the primary structure of bovine pancreatic ribonuclease      | 111 |
| 1965       | Allosteric regulation clarified                                          | 112 |
| 1965       | Studies began on immobilized enzymes                                    | 113 |
| 1972       | Synzyme: synthetic enzyme                                               | 114 |
| 1975       | Enzyme replacement therapy was suggested in 1950 and began in 1975      | 115 |
| 1980       | Computer programs for use in enzyme kinetic studies                     | 116 |
| 1981       | Discovery of ribozyme                                                   | 117 |
| 1981       | Use of enzymes in biosensors                                            | 118 |
| 1986       | Abzyme, a monoclonal antibody with catalytic activity                   | 119 |
| 1990       | Telomerase and aging, relationship explained                            | 120 |
| 1991       | First enzyme replacement therapy                                         | 121 |
| 1994       | Deoxyribozyme, DNA enzymes or catalytic DNA                             | 122 |
| 2011       | Conformational selection                                                | 123 |
| 2012       | Bio-energetic theory of carcinogenesis and inhibition, Krebs enzymes    | 124 |
| 2014       | Ubiquitin signaling explained                                           | 125 |
| 2014       | Artificial photosynthesis in nanobiocatalytic assemblies                | 126 |
defined by Selman Waksman in 1941. He initially described an ‘antibiotic agent’ as a microbe that antagonizes the growth of other microbes. With the initiation and discovery of many antibiotics of different classes such as penicillin, streptomycin, chloramphenicol, and tetracycline between the years 1945 and 1955, the dawn of antibiotics formally began (58). Likewise, Angiotensin Converting Enzyme Inhibitors are among the most common drugs which are widely prescribed for benign hypertension (60). Furthermore, inhibition of key, regulatory enzymes in lipid metabolism provides medicinal benefits as well. For instance, statins are inhibitors of HMG CoA reductase which have been used for treatment of high cholesterol. Another group of the enzyme inhibitors are used for treatment of obesity (61). Examining and characterizing the mechanism of actions for enzyme inhibition is one of the trending topics in biochemistry (62). From the regulation of respiration, pH balance, gluconeogenesis, ureagenesis, lipogenesis and Na+ retention; to the prevention of calcification, carcinogenesis, it is clear that enzyme inhibitors can be used for diverse therapeutic purposes (63) and this list could be extendible. Furthermore, enormous clinical success can be achieved with novel enzyme inhibitors in cancer therapy as well and every day, researchers are defining new drug targets for various, equally challenging diseases (65). On the other side of the medallion, pharmaceutical companies spend enormous amounts of money for clinical trials and marketing in novel drug development studies that use enzyme inhibitors (66). It is, thus, not hard to foretell that the current and future researchers will try to investigate novel drugs or modify preexisting drugs in new, alternative therapies for developing novel drug indications such as successfully eliminating seizures with little or no side effects (10–12, 14, 67–72).

Mapping the active sites of the enzymes began in the middle of the 1960s but designing drugs that

| Year | Finding                                                                                                                                                                                                 | Ref   |
|------|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|-------|
| 1917 | Enzymes in the fermentation industries                                                                                                                                                                   | 128   |
| 1951 | Conversion of starch to fermentable sugars, through chill-proofing of beer                                                                                                                               | 131   |
| 1957 | Pickle industry                                                                                                                                                                                          | 132   |
| 1972 | Enzymes in the detergent industry                                                                                                                                                                         | 133   |
| 1976 | Role of microbial enzymes in flavor development in foods                                                                                                                                                   | 134   |
| 1984 | Enzymes in starch industry                                                                                                                                                                                | 135   |
| 2001 | Immobilized enzymes applied to peptide synthesis, i.e., sweetener aspartame, enkephalin and other bioactive peptides                                                                                      | 136   |
| 2004 | Diagnostic kits                                                                                                                                                                                          | 137   |
| 2005 | Extracting essential oil (natural perfume) by enzymes                                                                                                                                                     | 138   |
| 2006 | Industrial application of β-galactosidase in food technology                                                                                                                                               | 139   |
| 2007 | Making diet foods                                                                                                                                                                                         | 140   |
| 2010 | Food technology (adding enzymes in for example celiac disorders)                                                                                                                                          | 141   |
| 2011 | α-Amylases applications in food, textile, paper, detergent industries                                                                                                                                     | 142   |
| 2012 | Enzymes have been used in cosmetics for more than 20 years                                                                                                                                                 | 143   |
| 2013 | Cellulase enzymes in paper, textile, and biofuels industries                                                                                                                                                | 144   |
| 2013 | Pectinase in fruit juice, greater juice extraction                                                                                                                                                         | 145   |
| 2013 | Enzymatic treatment of leather technology                                                                                                                                                                  | 146   |
| 2013 | Biosynthesis of rare hexoses                                                                                                                                                                              | 147   |
| 2014 | Commercial enzymes are used for producing biofuels                                                                                                                                                         | 148   |
interact with the active site is an important field in drug development (73). Kinetic or modelling studies provide us with the information on differentiating the active site and binding site of the enzymes, respectively. Enzyme research, or in other words biochemical research, today focuses on the molecular basis of biological processes and it applies especially to human health and diseases (80). Self-evidently, enzyme inhibitors will always be one of the dominating trends in drug development (74–79).

Enzymes in Industry

In the beginning of the current century, more than 3000 different enzymes from different organisms have been identified and many have found their respective applications in biotechnology as well as in the pharmaceutical, chemical and food industries (127). In fact, enzymes effectively changed the applications and methods in industries (128). Half a century ago, studies that involved various microorganisms began on clots formed in spoiling milk. By the year 1933, on the other hand, enzymes in bread and bakery industries started to mature with the observations of Blagoveschenski and Sossiedov on wheat flour (129). And even today, one of the most important study subjects is designing novel biocatalysts for the relevant industries (130) (Table II).

Applications that eventually ended up being used for commercial purposes initially started with the study of enzymes in daily, routine life. An effortless example, vinegar, practically had many applications in the past such as pickling of various food products both for preservation as well as for culinary purposes; carpet cleaning or removing of stains from fabrics, and as a brightening agent in dishwashing or antipyretic in villages, in Turkey. However, vinegar is still in use for similar or almost the same purposes in modern life.

Today, enzymes and the related enzyme products have many roles in industries including but not limited to pharmaceutical and food industries; perhaps most importantly, though, in medicine, enzymes can be used as valuable biomarkers in the diagnosis and management of various diseases such as heart, lung, liver, muscle, bone, pancreas, hematology, genetic diseases and malignancies as well as applications in toxicology and forensic medicine. Besides their roles as biomarkers in medicine, there are also various other applications in medicine such as; medicinal digestive enzymes that are used for therapeutic reasons, determination of parental lineage (149), and in plastic surgery (150). Examples of the industrial applications (wine, beer, cheese, bread, cosmetics, detergents, textile) are given in Table II.

Advances in the science and technology of enzymes will give birth to revolutionary progress in many distinct areas.

Conclusion

Enzymes braced their roles in pharmaceutical industry as diagnostic markers or therapeutic molecules, still a very vivid area of research in the field. Commercial, mass production of many consumer goods such as cheese is made possible by some enzymes. Similar to his prehistoric cousin who made cheese from the stomach of slaughtered animals, the modern man also uses the same enzymes to make this widely consumed, delicious dairy product, only in immobilized forms suitable for commercial use. Moreover, the food industry today relies on enzymes as well. Biological detergents containing specialized enzymes such as proteases, lipases and isomerases are widely used in consumer products. Mighty enzymes have accompanied us on our journey through history and clearly they will be with us in the future with myriads of novel and preexisting benefits.

Both from a macro and micro perspective, needless to say, enzymes played key roles in humanity as well as in the preservation of life quality for ordinary men in everyday life. Enzymes have sparkling roles in every part of our life, from an apoptotic cell on the verge of dying to the sustaining of delicate balances in metabolic pathways. In fact, enzymes solidly marked their footprints in diverse, sometimes interrelated, mostly distinct fields; medicinal therapeutics, warfare, art and food industry are among some of these fields... These small machines that have the potency to make enormous chemical reactions possible in sometimes less than milliseconds constitute the magnificent and marvelous building blocks of our cells. An insight into the history of enzymology enhances our knowledge and understanding of biochemistry. Research on enzymes has had a central indispensable role in the past, present and future of biochemistry in almost every aspect of life. Enzymes are cardinal contents of life and, therefore, exploration of the functions of enzymes and various biochemically uncharacterized proteins with undisclosed functions will continue in the experimental designs of future research. Both current and future scientists working in biochemistry will always resort to enzymes and the methods in enzymology in their pursuit of reliable, scientific remedies and solutions.

Conflict of interest statement

The authors stated that they have no conflicts of interest regarding the publication of this article.
References

1. Kleczkowski M, Garncarz M. The role of metal ions in biological oxidation – the past and the present. Pol J Vet Sci 2012; 15: 165–73.

2. Setlow P. Summer meeting 2013-when the sleepers wake: the germination of spores of Bacillus species. J Appl Microbiol 2013; 115: 1251–68.

3. Böttner J. Evolution of Clinical Enzymology. J. Clin. Chcm Clin Biochem 1981; 19: 529–38.

4. Robertson JB. The Early History of Catalysis. Platinum Metals Rev 1975; 19: 64–9.

5. D’Alessandro A, Giardina B, Gevi F, Timperio AM, Zolla L. Clinical metabolomics: the next stage of clinical biochemistry. Blood Transfus 2012; 10: 19–24.

6. Kohler RE. The Enzyme theory and the origin of biochemistry. Chic J 1975: 64: 181–96.

7. Jovičić S, Ignjatović S, Majkić-Singh N. Biochemistry and metabolism of vitamin D. J Med Biochem 2012; 31: 309–15.

8. Lazarević D, Dordević VV, Čosić V, Vlahović P, Tošić-Golubović S, Ristić T, et al. Increased lymphocyte caspase-3 activity in patients with schizophrenia. J Med Biochem 2011; 30: 55–61.

9. Karaman K, Tirnaksiz MB, Ulusu N, Dincer N, Sener B, Demirezer LÖ, Ulusu NN. In vitro effects of rosmarinic acid on glutathione reductase. Folia Biol (Praha) 2011; 57: 57–64.

10. Demirezer LÖ, Ulusu NN. Comparative in vitro effects of isoorientin, forsythoside B, and verbascoside on bovine kidney cortex glutathione reductase. Int Chem Kinet 2013; 45: 574–9.

11. Tandogan B, Guvenc A, Calis I, Ulusu NN. In vitro effects of imatinib on glucose-6-phosphate dehydrogenase and glutathione reductase. Folia Biol (Praha) 2011; 57: 57–64.

12. Tandogan B, Kuruüzüm-Uz A, Sengerze C, Ulusu NN. In vitro effects of rosmarinic acid on glutathione reductase and glucose 6-phosphate dehydrogenase. Pharm Biol 2011; 49: 587–94.

13. Tandogan B, Ulusu NN. Comparative in vitro effects of metal ions on bovine kidney cortex glutathione reductase. Prep Biochem Biotechnol 2010; 40: 405–11.

14. Tandogan B, Ulusu NN. A comparative study with colchicine on glutathione reductase. Protein J 2010; 29: 380–5.

15. Tandogan B, Ulusu NN. Inhibition of purified bovine liver glutathione reductase with some metal ions. J Enzyme Inhib Med Chem 2010; 25: 68–73.

16. Khare Sagar D, Fleishman Sarel J. Emerging themes in the computational design of novel enzymes and protein-protein interfaces. FEBS Lett 2013; 587: 1147–54.

17. Gerl JA, Allen KN, Almo SC, Armstrong RN, Babbitt PC, Cronan JE, et al. The Enzyme Function Initiative. Biochem 2011; 22: 9950–62.

18. Roncada P, Piras C, Soggiu A, Turk R, Urbani A, Bonizzi L. Farm animal milk proteomics. J Proteomics 2012; 19: 4259–74.

19. He H, Qin Y, Chen G, Li N, Liang Z. Two-step purification of a novel β-glucosidase with high transglycosylation activity and another hypothetical β-glucosidase in Aspergillus oryzae HML356 and enzymatic characterization. Appl Biochem Biotechnol 2015; 169: 870–84.

20. Natarajan P, Ray KK, Cannon CP. High-density lipoprotein and coronary heart disease: current and future therapies. J Am Coll Cardiol 2010; 30: 1283–99.

21. Reay Tannahill, editors, Food in history. (1973) New York Three Rivers Press p. 16, 68, 69.

22. Mithen SJ, Finlayson B, Smith S, Jenkins E, Najjar M, Maričević D. Göbekli tepe: An 11 600 year-old communal structure from the Neolithic of southern Jordan. Antiquity 2011; 85: 350–64.

23. Fox PF, editors, Cheese: An overview. Cheese: Chemistry, Physics and Microbiology, 2ed. Chapman and Hall, London (1993), pp. 1–36.

24. Salimei E, Fantuz F. Equid milk for human consumption. Int dairy J 2012; 24: 130–42.

25. Rosell JM. Yoghourt and kefir in their relation to health and therapeutics. Can Med Assoc J 1932; 26: 541–5.

26. Copeland Robert A, editor. Enzymes, A. Practical Introduction to Structure, Mechanism, and Data Analysis. John Wiley & Sons, Inc., 2000: 2–10.

27. McGovern PE, Zhang J, Tang J, Zhang Z, Hall GR, Moreau RA, et al. Fermented beverages of pre- and proto-historic China. Proc Natl Acad Sci USA 2004; 21: 17593–8.

28. Bambaugh CW, editors, Food, Fermentation and Micro-organisms, Blackwell Science: Oxford, UK, 2005: 143–153.

29. Kawakish S, Namiki K, Shirouzu H, Namiki M. Effects of .gamma.-irradiation on the enzyme relating to the development of characteristic odor of onions. J Agric Food Chem 1971; 19: 166–9.

30. Xu Y, Zhang Z, Wang M, Wei J, Chen H, Gao Z, et al. Identification of genes related to agarwood formation: transcriptome analysis of healthy and wounded tissues of Aquilaria sinensis. BMC Genomics 2013; 8: 1–16.

31. Li Q, Korzan WJ, Ferrero DM, Chang RB, Roy DS, Buchi M, et al. Synchronous evolution of an odor biosynthesis pathway and behavioral response. Curr Biol 2013; 7: 11–20.

32. Fischer-Tenhagen C, Tenhagen BA, Heuwieser W. Short communication: Ability of dogs to detect cows in estrus from sniffing saliva samples. J Dairy Sci 2013; 96: 1081–4.

33. Wells DL. Dogs as a diagnostic tool for ill health in humans. Altern Ther Health Med 2012; 18: 12–7.

34. Agostinucci W, Cardoni AA, Rosenberg P. Effect of papain on bee venom toxicity. Toxicol 1981; 19: 851–5.
35. Ross Jr EV, Badame AJ, Dale SE. Meat tenderizer in the acute treatment of imported fire ant stings. J Am Acad Dermatol 1987; 16: 1189–92.

36. Baker EL, Baker WL, Cloney DJ. Resolution of a phytobezoar with Aldoph's Meat Tenderizer. Pharmacotherapy 2007; 27: 299–302.

37. Surico N, Codecà C, Caccia S. Medical humanities in gynecology and obstetrics. Minerva Ginecol 2012; 64: 447–53.

38. Philippe G, Angenot L, Tits M, Frédéric M. About the toxicity of some Styrchnos species and their alkaloids. Toxicicon 2004; 15: 405–16.

39. Norman G. Bisset War and hunting poisons of the New World. Part 1. Notes on the early history of curare. J Ethnopharmacol 1992; 36: 1–26.

40. Bliss CL, Noy FG. Action of formaldehyde on enzymes and on certain proteins. J Exp Med 1899; 1: 47–80.

41. Cao L. Immobilised enzymes: science or art? Current Opinion in Chemical Biology 2005; 9: 217–26.

42. Pelkonen O, Abass K, Wiesner J. Thujone and thujone-containing herbal medicinal and botanical products: Toxicological assessment. Regul Toxicol Pharmacol 2015; 65: 100–7.

43. Abass K, Reponen P, Mattila S, Pelkonen O. Metabolism of α-thujone in human hepatic preparations in vitro. Xenobiota 2011; 41: 101–11.

44. Holstege CP, Baylor MR, Rusyniak DE. Absinthe: return of the Green Fairy. Semin Neurol 2002; 22: 89–93.

45. Bonkovsky HL, Cable EE, Cable JW, Donohue SE, White EC, Greene YJ, et al. Porphyrogenic properties of the terpenes camphor, pinene, and thujone (with a note on historic implications for absinthe and the illness of Vincent van Gogh). Biochem Pharmacol 1992; 43: 2359–68.

46. Jones PJ. Cleopatra's cocktail. Class World 2010; 103: 207–20.

47. Smaltz F. Neurosciences and research on chemical weapons of mass destruction in Nazi Germany. J Hist Neurosci 2006; 15: 186–209.

48. Jokanović M, Prostran M. Pyridinium oximes as cholinesterase reactivators. Structure-activity relationship and efficacy in the treatment of poisoning with organophosphorus compounds. Curr Med Chem 2009; 16: 2177–88.

49. Kuca K, Juna D, Musilek K. Structural requirements of acetylcholinesterase reactivators. Mini Rev Med Chem 2006; 6: 269–77.

50. Kitada M, Kume S, Takeda-Watanabe A, Kanasaki K, Koya D. Sirtuins and renal diseases: relationship with aging and diabetic nephropathy. Clin Sci (Lond) 2013; 124: 153–64.

51. Farris PK. Innovative cosmeceuticals: sirtuin activators and anti-glycation compounds. Semin Cutan Med Surg 2011; 30: 163–6.

52. Lebeda FJ, Cee RZ, Mudunuri U, Stephens R, Singh BR, Adler M. The zinc-dependent protease activity of the botulinum neurotoxins. Toxins (Basel) 2010; 2: 978–97.
and liver of streptozotocin-diabetic rats: comparison with vitamin E. Biochim Biophys Acta 2002; 9: 71–8.

69. Ulusu NN, Ercil D, Sakar MK, Tezcan EF. Abietic acid inhibits lipoxygenase activity. Phytother Res 2002; 16: 88–90.

70. Guz G, Demirogullari B, Ulusu NN, Dogu C, Demirtola A, Kavutcu M, et al. Sotobadine protects rat kidney against ischaemia/reperfusion injury. Clin Exp Pharmacol Physiol 2007; 34: 210–6.

71. Ulusu NN, Sahilli M, Avci A, Canbolat O, Ozansoy G, Ari N, et al. Pentose phosphate pathway, glutathione-dependent enzymes and antioxidant defense during oxidative stress in diabetic rodent brain and peripheral organs: effects of sotobadine and vitamin E. Neurochem Res 2003; 28: 815–23.

72. Tandogan B, Guvenc A, Çalış İ, Ulusu NN. In vitro effects of compounds isolated from Sideritis brevibracteata on bovine kidney cortex glutathione reductase. Acta Biochim Pol 2011; 58: 471–5.

73. Schechter I. Mapping of the active site of proteases in the 1960s and rational design of inhibitors/drugs in the 1990s. Curr Protein Pept Sci 2005; 6: 501–12.

74. Sohier JS, Laurent C, Chevigné A, Pardon E, Srinivasan V, et al. Pentose phosphate pathway, glutathione-dependent enzymes and antioxidant defense during oxidative stress in diabetic rodent brain and peripheral organs: effects of sotobadine and vitamin E. Neurochem Res 2003; 28: 815–23.

75. Ulusu NN, Tandogan B. Purification and kinetics of sheep kidney cortex glucose-6-phosphate dehydrogenase. Comp Biochem Physiol B Biochem Mol Biol 2006; 143: 249–55.

76. Ulusu NN, Tandogan B, Tezcan FE. Kinetic properties of glucose-6-phosphate dehydrogenase from lamb kidney cortex. Biochimie 2005; 87: 187–90.

77. Ulusu NN, Kus MS, Acan NL, Tezcan EF. A rapid method for the purification of glucose-6-phosphate dehydrogenase from bovine lens. Int J Biochem Cell Biol 1999; 31: 787–96.

78. Ulusu NN, Tandogan B. Purification and kinetic properties of glutathione reductase from bovine liver. Mol Cell Biochem 2007; 303: 45–51.

79. Tandogan B, Ulusu NN. Purification and kinetics of bovine kidney cortex glutathione reductase. Protein Pept Lett 2010; 17: 667–74.

80. Bradshaw RA, Hancock CC, Krege N. 100 years of chemistry of life. 1 ed. Columbia, Maryland 1999, 5pp.

81. Copeland RA. A Brief History of Enzymology. In Enzymes: A Practical Introduction to Structure. New York, Wiley-VCH, Inc. 2000: 1–11.

82. Tipton K, Boyce S. The history of enzyme nomenclature system. Bioinformatics 2000; 16: 34–40.

83. Ben Kilani Cö, Batis H, Chastrette M. Development of the ideas concerning catalysis at the beginning of the XIXth century. Actual Chim 2001; 7–8: 44–50.

84. Schadewaldt H. Nutrition and individual defense – historical considerations. Zentralbl Hyg Umweltmed 1991; 191: 302–6.

85. Pavy FW, Siau RL. An experimental enquiry upon glycolysis in drawn blood. J Physiol 1902; 31: 451–6.

86. Stirling W. On the Ferments or Enzymes of the Digestive Tract in Fishes. J Anat Physiol 1884; 18: 426–35.

87. Warren G. A vital assay. Nat Rev Mol Cell Biol 2012; 13: 754.

88. Madeira VM. Overview of mitochondrial bioenergetics. Methods Mol Biol 2012; 810: 1–6.

89. Kohler RE. The reception of Eduard Buchner’s discovery of cell-free fermentation 1972, 5: 327–53.

90. Henri V. General theory of the action of some glycoside hydrolases. CR Acad Sci Paris 1902; 135: 919.

91. Harden A, Macfadyen A. Enzymes in tumour. The Lancet 1903; 162: 224–5.

92. Michaelis L, Menten ML, Johnson KA, Goody RS. The original Michaelis constant: translation of the 1913 Michaelis-Menten paper. Biochem 2011; 4: 8264–9.

93. Harden A, Norris RV. The enzymes of washed zymcin and dried yeast (Lebedeff). II. Reductase. Biochem J 1914; 8: 100–6.

94. Hartridge H. An improved spectrophotometer. J Physiol 1915; 24: 101–13.

95. Sherman HC, Thomas AW, Hinck CF. Studies on amylases. VIII. The influence of certain acids and salts upon the activity of malt amylase. JACS 1915; 37: 623–43.

96. Mack E, Villars DS. Synthesis of urea with the enzyme urease. JACS 1923; 45: 501–5.

97. Bastedo WA. The use and utility of digestive enzymes in therapeutics: A summary of the replies to a questionnaire submitted to the members of the American gastroenterological association. JAMA 1925; 85: 743–4.

98. Sumner JB. The isolation and crystallization of the enzyme urease. JACS 1925; 47: 327–53.

99. Madeira VM. Overview of mitochondrial bioenergetics. Methods Mol Biol 2012; 810: 1–6.

100. Roskelley RC, Mayer N, Horwitt BN, Salter WT. Studies in cancer. VII. Enzyme deficiency in human and experimental cancer. J Clin Invest 1943; 22: 743–51.

101. Young A. Effects on plasma glucose and lactate. Adv Pharmacol 2005; 52: 193–208.

102. Marcus M. A contribution to the history of favismo. Harefuah 1948; 15: 24.

103. Thompson A. The c-terminal residue of lysozyme. Nature 1952; 169: 495–6.

104. Thompson EO. The N-terminal sequence of carboxypeptidase. Biochim Biophys Acta 1953; 10: 633–4.

105. Marks PA. A newer pathway of carbohydrate metabolism; the pentose phosphate pathway. Diabetes 1956; 5: 276–83.

106. Koshland DE, Jr. Enzyme flexibility and enzyme action. J Cell Comp Physiol 1959; 54: 245–58.
107. Wróblewski F, Ross C, Gregory K. Isoenzymes and myocardial infarction. N Engl J Med 1960; 15: 531–6.

108. Viswanatha T, Lawson WB, Witkop B. The action of N-bromosuccinimide on trypsinogen and its derivative. Biochimica et Biophysica Acta 1960; 40: 216–24.

109. Smyth DG, Stein WH, Moore S. On the sequence of residues 11 to 18 in bovine pancreatic ribonuclease. J Biol Chem 1962; 237: 1845–50.

110. Stanford Jr RH, Marsh RE, Corey RB. Structure of lysozyme: An x-ray investigation of lysozyme chloride crystals containing complex ions of niobium and tantalum: Three-dimensional fourier plot obtained from data extending to a minimum spacing of 5. Å. 1962; 196: 1176–8.

111. Smyth DG, Stein WH, Moore S. The Sequence of Amino Acid Residues in Bovine Pancreatic Ribonuclease: revisions and confirmations. J Biol Chem 1963; 238: 227–34.

112. Changeux JP. Allosterity and the Monod-Wyman-Changeux model after 50 years. Annu Rev Biophys 2012; 41: 103–35.

113. Guilbault GG. Fluorometric system employing immobilized cholinesterase for assaying anticholinesterase compounds. Anal Chem 1965; 37: 1675–80.

114. Kiefer HC, Congdon WI, Scarpa IS, Klotz IM. Catalytic accelerations of 10-fold by an enzyme-like synthetic polymer. Proc Natl Acad Sci USA 1972; 69: 2155–9.

115. Brady RO. The lipid storage diseases: new concepts and revisions and confirmations. J Biol Chem 1963; 238: 227–34.

116. Kinderlerer J, Ainsworth S, Gregory RB. Computer programs for use in enzyme kinetic studies. Biochem Soc Trans 1980; 8: 652–52.

117. Cech TR, Zaug AJ, Grabowski PJ. In vitro splicing of the ribosomal RNA precursor of Tetrahymena: involvement of a guanosine nucleotide in the excision of the intervening sequence. Cell 1981; 27: 487–96.

118. Vadgama P. Enzyme electrodes as practical biosensors. J Med Eng Technol 1981; 5: 293–8.

119. Pollack SJ, Jacobs JW, Schultz PG. Selective chemical catalysis by an antibody. 1986; 234: 1570–3.

120. Harley CB, Futcher AB, Greider CW. Telomeres shorten during ageing of human fibroblasts. Nature 1990; 345: 458–60.

121. Kay AC, Saven A, Garver P, Thurston DW, Rosenbloom BF, Beutler E. Enzyme replacement therapy in type-I Gaucher disease. Trans Assoc Am Physicians 1991; 14: 256–64.

122. Breaker RR, Joyce GF. A DNA enzyme that cleaves RNA. Chem Biol 1994; 1: 223–9.

123. Changeux JP, Stuart ES. Conformational selection or induced fit? 50 years of debate resolved. F1000 Biol Rep 2011; 3: 19–24.

124. Gonzalez MJ, Miranda M, JR, Duconege J, Riordan NH, Ichim T, Quintero-Del-Rio AI, Ortiz N. The bio-energetic theory of carcinogenesis. Medical Hypotheses 2012; 79: 433–9.

125. Weller CE, Pilkerton ME, Chatterjee C. Chemical strategies to understand the language of ubiquitin signaling. Biopolymers 2014; 101: 144–55.

126. Kim JH, Nam DH, Park CB. Nanobiocatalytic assemblies for artificial photosynthesis. Current Opinion in Biotechnology 2014; 28: 1–9.

127. Breitbart H. The hunt for living gold. The search for organisms in extreme environments yields useful enzymes for industry. EMBO reports 2001; 21: 968–71.

128. Wallerstein L. Enzymes in the fermentation industries JFI 1917; 183: 531–56.

129. Blagoveschenski AV, Sossiedov MP. On the changes of wheat proteins under the action of flour and yeast enzymes. Biochem J 1935; 29: 805–10.

130. Bommarius AS, Blum JK, Abrahamsson MJ. Status of protein engineering for biocatalysts: how to design an industrially useful biocatalyst. Current Opinion in Chemical Biology 2011; 15: 194–200.

131. Smythe CV. Microbiological production of enzymes and their industrial applications 1951; 5: 126–144.

132. Demain AL, Phaff HJ. Cucumber curing, softening of cucumbers during curing. J Agr Food Chem 1957; 5: 60–4.

133. Göthe CJ, Westlin A, Sundquist S. Air-borne B. subtilis enzymes in the detergent industry. Internationales Archiv für Arbeitsmedizin 1972; 29: 201–8.

134. Shahani KM, Arnold RG, Kilara A, Dwivedi BK. Role of microbial enzymes in flavor development in foods. Biotechnol Bioeng 1976; 18: 891–907.

135. Miwa T. Saccharides from starch hydrolysis and their derivatives for foodstuffs. Yuki Gosei Kagaku Kyōshi/J Synthetic Org Chem 1984; 42: 597–601.

136. Liu P, Tian GL, Ye YH. Progress in the Study on Peptide Synthesis Catalyzed by Immobilized Enzyme. Chem J Chinese U 2001; 22: 1347–8.

137. Mason P. Basic concepts in clinical testing. Pharm J 2004; 272: 384–6.

138. Liu XG, Ju XR, Mao XD, Zhang Z. Studies on enzymic extraction of essential oil from pine needles. Linchan Huaxue Yu Gongye/Chem Ind Forest Prod 25 (3), pp. 101–114, 2005.

139. Milchová Z, Rosenberg M. Current trends of β-galactosidase application in food technology (Review). J Food Sci Nutr 2006; 45: 47–54.

140. Hsieh YHP, Ofori JA. Innovations in food technology for health. Asia Pac J of Clin Nutr 2010; 44: 4–8.

141. Selimo M, Karabiber H. Celiac disease: Prevention and treatment (Review) J Clin Gastroenterol 2010; 44: 4–8.

142. Sreedhar RP, Jhansi RD, Sulthana S. Amylases as a tool for industrial application: A review (Review). J Pure Appl Microbiol 2011; 5: 167–71.
143. Galonde N, Nott K, Debuigne A, Deleu M, Jerôme C, Paquot M, et al. Use of ionic liquids for biocatalytic synthesis of sugar derivatives (Review). J Chem Technol Biot 2012; 8: 451–71.

144. Hadden JA, French AD, Woods RJ. Unraveling cellulose microfibrils: A twisted tale. 2013; 99: 746–56.

145. Adelakun OE, Metcalfe D, Tshabalala P, Stafford B, Oni B. The effect of pectinase enzyme on some quality attributes of a Nigerian mango juice. Nutr Food Sci 2013; 43: 374–85.

146. Dettmer A, Dos APS, Gutterres M. Special review paper: Enzymes in the leather industry. J Am Leather Chem Assoc 2013; 108: 146–58.

147. Li Z, Gao Y, Nakanishi H, Gao X, Cai L. Biosynthesis of rare hexoses using microorganisms and related enzymes. Beilstein J Org Chem 2013; 12: 2434–45.

148. Khawla BJ, Sameh M, Imen G, Donyes F, Dhouha G, Raoudha EG, et al. Potato peel as feedstock for bioethanol production: A comparison of acidic and enzymatic hydrolysis. Ind Crop Prod 2014; 52: 144–9.

149. Tabita FR, Hanson TE, Satagopan S, Witte BH, Kreel NE. Phylogenetic and evolutionary relationships of RubisCO and the RubisCO-like proteins and the functional lessons provided by diverse molecular forms. Philos Trans R Soc Lond B Biol Sci 2008; 27: 2629–40.

150. Cavallini M, Gazzola R, Metalla M, Vaienti L. The role of hyaluronidase in the treatment of complications from hyaluronic Acid dermal fillers. Aesthet Surg J 2013; 1: 1167–74.

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