P-03

Possible Metabolic Transformation of Pinenes to Ionones

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1. Introduction

The unintended consequence of the ingestion of certain foods to alter the scent or color of urine is well known [1]. Less awareness exists regarding the practice of ingestion of natural products or drugs with the intended purpose of conferring urine the scent of violets [2]. The resin of the terebinth tree and the derived turpentine were widely used in Antiquity in wine-making, both as taste enhancer and conserving agent, so the effect on urine was possibly noticed due to the presence in wines [3].

The scent altering effect requires metabolic conversion of pinene, the main turpentine component to ionone, the molecule mainly responsible for the scent of violets [2,4]. The metabolic pathway (in humans or otherwise) was (to our knowledge) not yet described [4]. Thus, we here propose a possible metabolic pathway for the conversion of pinene to ionone, explaining the scent altering effect of turpentine.

2. Materials and methods

In-silico prediction of the site of metabolism and the human CYP involvement were achieved *via* the SOMP webserver [5].

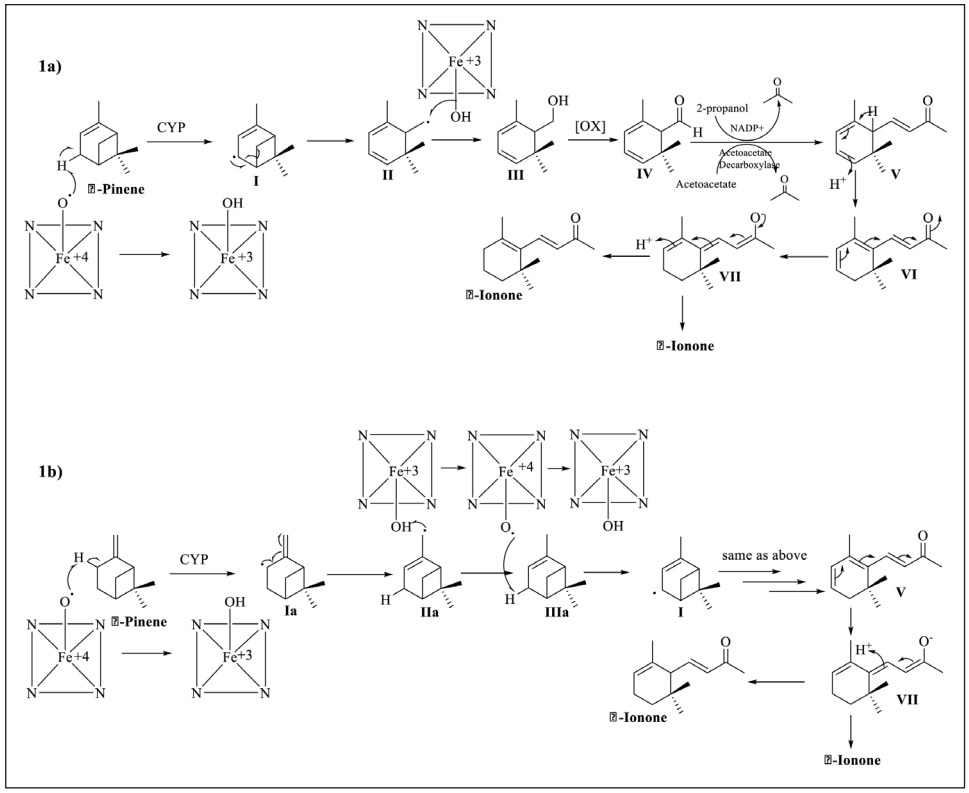

Figure 1 *Possible metabolic pathways to α-and β ionone based on the enzymatic oxidation of α-pinene and β-pinene*

3. Results

We propose a possible metabolic pathway that might lead to α-and β-ionone based on enzymatic oxidation of α-pinene and β-pinene, respectively [***Figure 1***: pathway 1a and 1b] [6].

Oxidation of α-pinene or β-pinene by CYP might lead to intermediates I and Ia, respectively. Rearrangement of the free radicals in I and Ia, should deliver intermediates II and IIa, respectively. Then, intermediate IIa should abstract an H· from CYP to deliver intermediate IIIa. The latter, will lose the allylic H· to CYP to produce

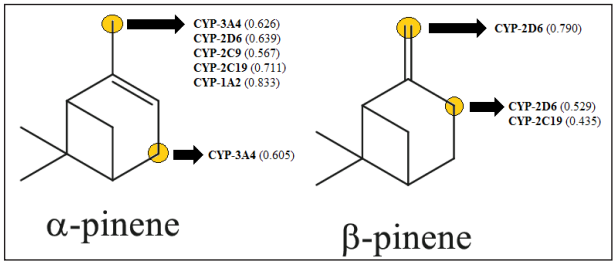

Figure 2 *Predicted site of metabolism (SOM) for α- and β-pinene monoterpenes along with the cytochrome P-450 isoforms involved, where, values between brackets indicate the probability of involvement.*

intermediate I. At this stage, the two routes (1a and 1b) converge through the formation of intermediate II. The latter is formed through free radical rearrangement and cleavage of the cyclobutyl ring leading to II. Subsequent oxidation of intermediate II should furnish the alcohol III which upon oxidation produces the aldehyde IV. At this junction, Knoevenagel condensation of IV with acetone should produce intermediate V. 1,5-sigmatropic rearrangement of V should produce intermediate VI. Rearrangement of the highly conjugated system in VI should deliver the trienol VII. The latter can easily rearranges to both α-and β-ionone as shown in ***Figure 1a*** and ***1b***. The two routes might converge as a results of 1,5-sigmatropic rearrangement of V leading to intermediate VI.

One of the key component of this proposed metabolic transformation is acetone. Acetone could be produced in organisms either by the decarboxylation of acetoacetate or the dehydrogenation of propan-2-ol according to the proposed routes in ***Figure 1a*** and ***1b***. It is well documented that acetoacetate decarboxylation is the major source of acetone in mammals, which could arise from either lipolysis or degradation of amino acid [7].

The predicted sites of metabolism (SOM) of the α- and β-pinene monoterpenes with respect to the corresponding human CYP isoform involvement are given in ***Figure 2***. The predicted sites are consistent with the known metabolic reactions especially; allylic oxidation, epoxidation and stereoselective gem-dimethyl hydroxylation [8]. Furthermore, the predicted CYP isoforms with high probabilities were; 1A2, 3A4, 2C19 and 2D6. Recently, an X-ray crystal structure of α-pinene with CYP 2B was resolved (PDB-ID: 4I91) confirming the role of CYP 2B6 and 2A6 in the metabolism α- and β-pinenes [9].

4. Conclusions

Metabolic pathways for the synthesis of α- and β-ionones from α- and β-pinenes are proposed, explaining the scent altering effect of turpentine exposure.

References

1. *Mitchell S.C., Food idiosyncrasies: beetroot and asparagus, Drug Metabolism and Disposition, 29: 539-543 (2001).*
2. *Petroianu A., Stegmeier-Petroianu A., Lorke D.E., Cleopatra: from turpentine and juniper to ionone and irone, Pharmazie, 73: 676-680 (2018).*
3. *Norton S., Pharmacology of Mithridatum: A 2000 Year Old Remedy, Molecular Interventions, 6: 60-66 (2006).*
4. *Vespermann K.A.C., Paulino B.N., Barcelos M.C.S., Pessôa M.G., Glaucia M. Pastore G.M., Molina G Biotransformation of α- and β-pinene into flavor compounds, Appl Microbiol Biotechnol, 101:1805–1817 (2017).*
5. *Rudik A., Dmitriev A., Lagunin A., Filimonov D., Poroikov V. SOMP: web server for in silico prediction of sites of metabolism for drug-like compounds. Bioinformatics. 31:2046-2048 (2015).*
6. *Negoi A., Parvulescu V.I., Tudorache M. Peroxidase-based biocatalysis in a two-phase system for allylicoxidation of pinene, Catalysis Today, 306: 199–206 (2018).*
7. *Kalapos M.P., On the mammalian acetone metabolism: from chemistry to clinical implications, Biochim Biophys Acta, 1621: 122-139 (2003).*
8. *Eriksson K., Levin J.O., Gas chromatographic-mass spectrometric identification of metabolites from alpha-pinene in human urine after occupational exposure to sawing fumes, J Chromatogr B Biomed Appl, 677:85-98 (1996).*
9. *Wilderman P. R., Shah M. B., Jang H. H., Stout C. D., Halpert J.R., Structural and thermodynamic basis of (+)-α-pinene binding to human cytochrome P450 2B6, J Am Chem Soc 135:10433-10440 (2013)*