pKa constant determination of two triazole herbicides : Tebuconazole and Penconazole

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Abstract   We determined the acidity constants of tebuconazole and penconazole, two fungicides from the group of 1,2,4-triazoles. Potentiometric titrations were performed in a 20% (v/v) acetonitrile/water mixture at 25 °C and at a fixed ionic strength (KNO$_3$, 0.1 mol·dm^{-3}). The pK_a values (representing thermodynamic constants) were determined to be 5.0 ± 0.1 and 5.2 ± 0.1 for tebuconazole and penconazole, respectively. These values could be used in pure water solutions to consider the protonated or deprotonated forms when studying the field behavior of these fungicides. Molecular modeling calculations allowed identifying the N4 atom as the protonation site.

1 Introduction

Agrochemicals include organic molecules, with various building blocks and functional groups. When they are dispersed in the field, their physical, chemical and ecotoxicological properties, mostly related to their structure, are of major importance for their environmental behavior. Among those, the acid/base properties are of special interest, because they determine the present chemical form: a neutral molecule or a charged ion (either a cation and/or an anion, depending on the molecular structure). Because the soil and its main components (clays, Fe- and Mn-oxyhydroxides, humic substances, etc.) exhibit different surface charges according to the environmental pH [1], the acidity constant K_a (or the pK_a) of the pesticides are crucial for determining their behavior in the soil environment.

Here we focused on two pesticides of the (benzo)triazole family for which some quantitative structure-properties relationships are already available [2]: tebuconazole and penconazole (Table 1, Fig. 1). They are known to behave as systemic fungicides, with both curative and protective actions, mainly applied to orchards and vineyards [3] where the soil pH is commonly within the range from 4 to 8. According to their structure (Fig. 1), both tebuconazole and penconazole can be expected to behave as weak bases, the N4 nitrogen atom in the 1, 2, 4-triazole ring being the most probable target for protonation (see section 3.1 below); unfortunately, their pK_a values are unknown (tebuconazole) or of dubious value (penconazole) [4]. The knowledge of their correct values is essential, in order to predict the nature of the species (neutral molecule or protonated one) present in the environment.

These triazole molecules are poorly water-soluble, and require water-miscible co-solvents in order to be solubilized. Acetonitrile is a very adequate cosolvent for use in RP-HPLC analysis of this class of compounds [5, 6] due to its aprotic and polar nature and because it is fully miscible with water. Therefore, a mixture of acetonitrile and water was chosen here for the potentiometric determination of the pK_a of the two fungicides.

2 Experimental

2.1 Reagents

Analytical-grade tebuconazole was supplied by the Institute of Industrial Organic Chemistry (Warsaw, Poland) with a purity of 99.9 ± 0.1 %; analytical standard penconazole was purchased from Fluka (Pestanal, 99.1 %). All reagents and acetonitrile (AN) were of analytical grade (Fluka), and dissolved in high purity de-ionized carbonate-free water (Milli-Q system: resistivity 18.2 MΩ·cm, TOC < 10 µg·dm^{-3}).

2.2 Potentiometric Measurements

The pK_a values of tebuconazole and penconazole were determined by potentiometric titration in a 20% (v/v) acetonitrile/water solvent mixture, according to a slight modification of the method described by Fikri et al. [7]. Each triazole compound (0.01 g) was dissolved in 20% AN (100 cm^3; at a fixed 0.1 mol·dm^{-3} ionic strength using KNO$_3$), then sonicated (Bransonic model 200, power = 19 W, frequency = 40 Hz) during 20 min. Nitric acid (0.1 mol·dm^{-3}) was added in equivalent amount to 20 cm^3 of the triazole solution and this mixture of strong acid and protonated weak base was subsequently titrated by 0.01 mol·dm^{-3} KOH (in the 20% AN solvent) in a thermostatted cell (25 ºC) under nitrogen atmosphere (Linde, 5.0). The exact concentration of the KOH solution was determined by titration with HNO$_3$ (0.1 mol·dm^{-3}; Normadose Prolabo). The titration was performed (Fig. 2) using the automatic titrator Metrohm 716 DMS Titrino coupled to a Metrohm 727 Ti Stand, a Metrohm 722 stirrer and equipped with a glass/calomel pH electrode (Metrohm, pH 0–14/0–80 °C; KCl 3 mol·dm^{-3}). The set parameters are the dynamic mode, a 1 mV·min^{-1} signal drift and a 5 cm^3·min^{-1} maximal flow-rate. The titration started after pH equilibrium was reached in the cell. The combined glass electrode was calibrated with pH 7.00 and 4.00 aqueous buffers and then with a 0.05 mol·kg^{-1} potassium hydrogenophthalate solution in the 20% AN solvent (pH$_S$ = 4.58 [8, 9]). All procedures were performed in triplicates. The titration curves were fitted using the ProtoFit (version 2.1) software [10] to calculate the pK_a values and using the Davies activity coefficient corrections calculated from the data in the mixed solvent system [11]. According to Fikri et al. [7], such a calculated value can be considered as numerically equal to the value in pure water, within the experimental uncertainties (± 0.05).

2.3 Molecular Modeling

111 For each compound, the geometries of the two enantiomers were optimized by molecular

112 mechanics (MM2 force field) with HyperChem [12] and further refined by the PM6 semi-

113 empirical molecular orbital method with MOPAC [13, 14]. The gas phase formation enthalpies at

114 298 K were also computed from MOPAC.

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117 **3 Results and Discussion**

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119 3.1 Protonation Site

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121 Due to the presence of a single chiral center (asymmetric carbon atom) in both molecular

122 structures, each consists of two (*R* or *S*) enantiomers (Fig. 1), whose properties can be somewhat

123 different; the studied samples were racemic mixtures, however.

124 According to our molecular theoretical quantum calculations of gas phase formation enthalpies

125 (Table 3), the N4 protonation is favored by *ca.* 50–65 kJ·mol^{-1}, compared to N2 protonation,

126 whatever the studied fungicide and its enantiomer form.[1]

127 Any further protonation on the N2 atom of the triazole ring will be hindered, due to both steric

128 hindrance and electrostatic repulsion. Moreover, there are no other protonable or deprotonable

129 site on neither of these two molecules.

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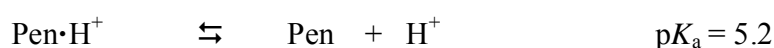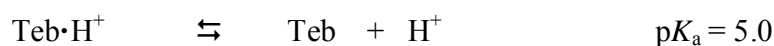131 3.2 Protonation Constants

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133 The acidity constants, pK_a, of the two triazole compounds were determined from potentiometric

134 titrations in a 20% (v/v) AN solvent mixture at 25 °C (Fig. 2). Due to the introduction of activity

135 coefficient corrections during data treatment, our calculated pK_a values can be considered as the

136 thermodynamic ones.

137 However it would be more useful to have information about the pK_a value in pure water. In order

138 to estimate the effect of the 20% AN solvent mixture, we refer to our previous study on amino-2

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[1] We have also tested the new PM7 method in MOPAC 2012. This method, which is still under development, gave enthalpy values within 5.3% of the PM6 values. The relative stabilities of the protonated forms were unchanged.

139 pyridine derivatives [7]: it appeared that the values of aqueous pK_a would be *ca*. 0.02 unit higher

140 than those determined in the 20% AN solvent, what is within the experimental uncertainties (±

141 0.05).

142 This is why we consider that our values are a good estimate (better than ± 0.1 unit) of the

143 aqueous pK_a of these two fungicides, *i.e.* for the equilibrium between the protonated and the free

144 molecular triazole (± 0.1; at 25 °C):

145 $\quad$ Teb·H$^+$ $\quad$ ⇆ $\quad$ Teb + H$^+$ $\qquad$ pK_a = 5.0

146 $\quad$ Pen·H$^+$ $\quad$ ⇆ $\quad$ Pen + H$^+$ $\qquad$ pK_a = 5.2

147 Until now, there has been limited information about the dissociation constant of tebuconazole and

148 penconazole (Tables 1 and 2). According to the IUPAC Pesticides Properties Database [4],

149 tebuconazole is considered as a very weak base, without any numerical value; for penconazole,

150 the low value given (1.51, see Table 1) is not in accordance with the structure of the compound,

151 while it would correspond to a medium-strength acid. Few other data are cited in the literature

152 (see Table 2); in all cases, they are lower than our experimental values, indicating a somewhat

153 weaker basic behavior for the corresponding molecule, what is once more not coherent with its

154 structure. Furthermore, the SPARC-calculated values [15] are too low to be realistic, and should

155 be ignored.

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158 **4 Conclusion**

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160 The aqueous pK_a values, determined for the first time in this study, allow predicting the

161 environmental behavior of either tebuconazole or penconazole according to the acid/base

162 properties of the soil solution and of the soil sorption complex. If stability constants data become

163 available, their possible complexation with metal cations present in the soil solution or added as

164 pesticides (*e.g.*, Cu [17, 18]) can now be estimated, for a better knowledge of the speciation of

165 these triazole fungicides.

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174 **References**

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[223]
[224]

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Table 1 Physico-chemical properties of the fungicides tebuconazole and penconazole,
according to IUPAC Pesticides Properties Database [4]

	Tebuconazole	**Penconazole**
Chemical name	(*R,S*)-1-*p*-chlorophenyl-4,4-dimethyl-3-(1*H*-1,2,4-triazol-1-ylmethyl)pentan-3-ol	(*R,S*)-1-[2-(2,4-dichlorophenyl)pentyl]-1*H*-1,2,4-triazole
CAS Number	107534-96-3	66246-88-6
SMILES	Clc1ccc(cc1)CCC(O)(C(C)(C)C)Cn2ncnc2	Clc1ccc(c(Cl)c1)C(CCC)Cn2ncnc2
Chemical formula	$C_{16}H_{22}ClN_3O$	$C_{13}H_{15}Cl_2N_3$
Molecular mass	307.82	284.18
pK_a	not available 'very weak base'	1.51 (at 25 °C) 'very weak base'
Water solubility (g·dm^{-3} at 20 °C)	0.036	0.073
Melting point (ºC)	105	60.3
log K_{ow} [a]	3.7 (at pH 7 and 20 °C)	3.72 (at pH 7 and 20 °C)
K_{oc} (dm^3·kg^{-1}) [b]	769	2205
Degradation in soils (d) [c]	55.8	90.0
GUS leaching potential index [d]	2.00	1.51
BCF [e]	78	320

[a] K_{ow} is the octanol/water partition coefficient
[b] Partition coefficient normalized to organic carbon content, Freundlich model
[c] Aerobic soil half-life, field conditions (days)
[d] Groundwater Ubiquity Score (estimated)
[e] BioConcentration Factor

Table 2 pK_a values for the two fungicides

	Experimental results [a]	Estimated value [b]	Literature data
Tebuconazole	5.0 ± 0.1	1.56	3.39 [c]
Penconazole	5.2 ± 0.1	1.36	2.83 ± 0.12 [d]

a Potentiometric; 25 °C

b According to SPARC [15]

c Cited by Chimuka et al. [16]

d Cited by Arias et al. [17]

244 **Table 3** Gas phase formation enthalpies (kJ·mol^{-1}) calculated with MOPAC/PM6

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	Neutral molecule	N2 protonated	N4 protonated
Tebuconazole *R*	− 54.17	612.16	547.23
Tebuconazole *S*	− 46.90	602.16	551.14
Penconazole *R*	181.15	854.37	793.08
Penconazole *S*	183.27	850.82	795.17

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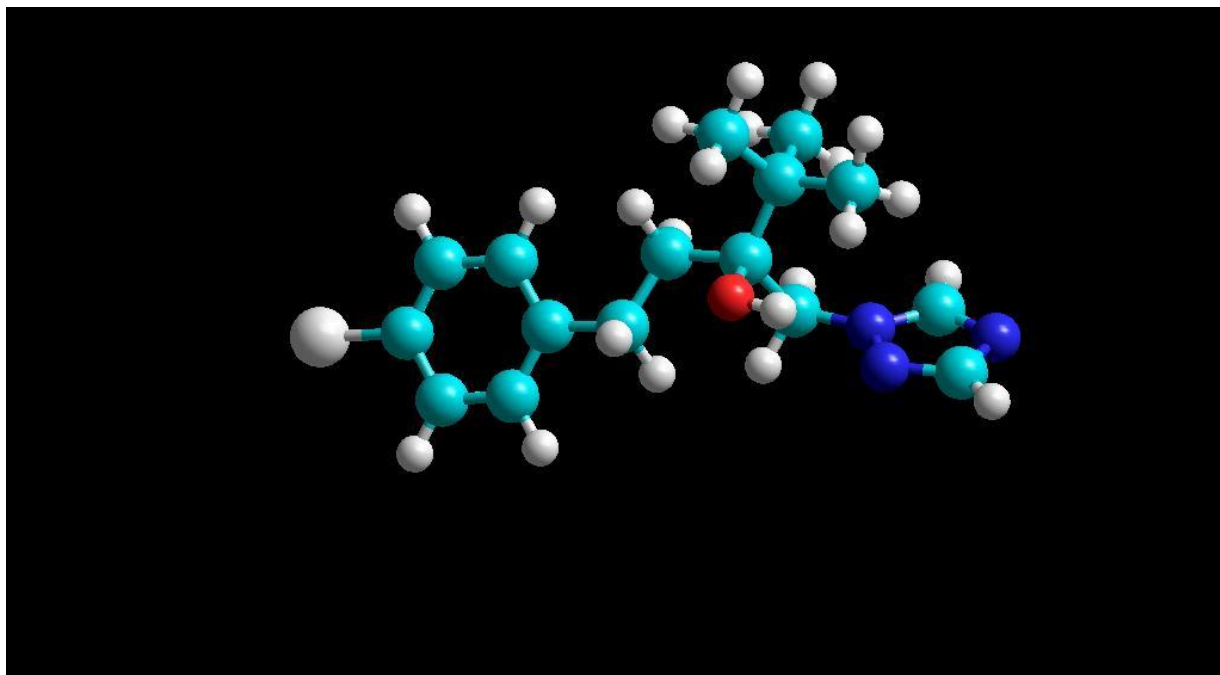**Fig. 1** Structures of Tebuconazole (a, b) and Penconazole (c, d) enantiomers (sticks and balls
presentation).
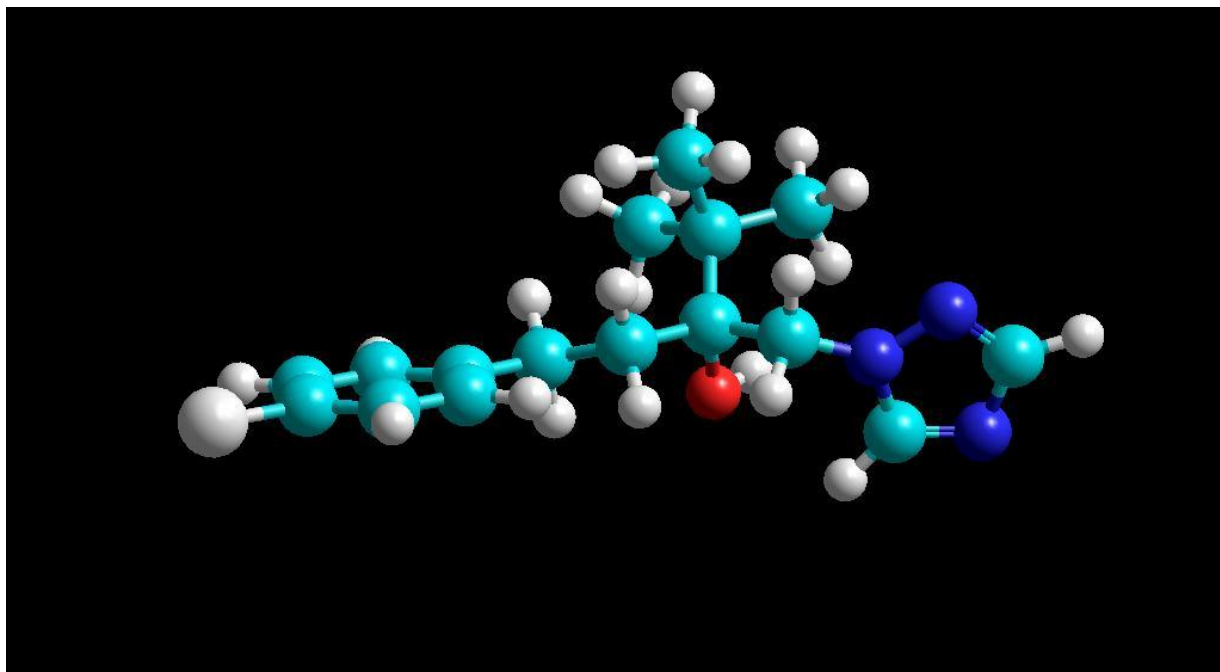
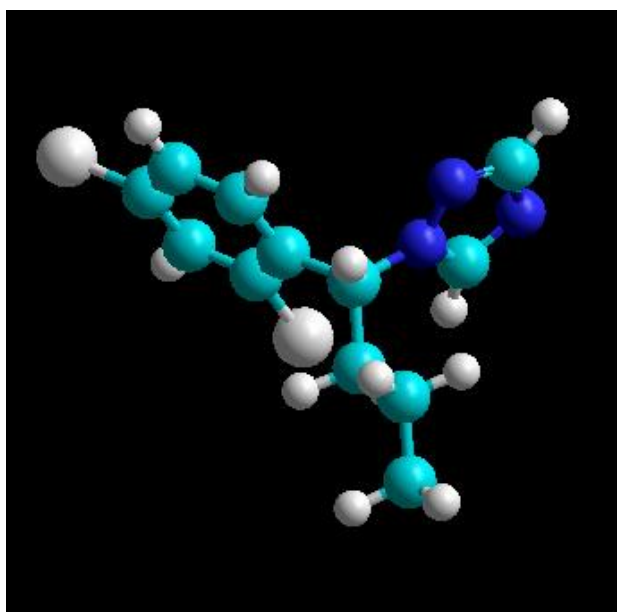(a) Tebuconazole *R*

(b) Tebuconazole *S*
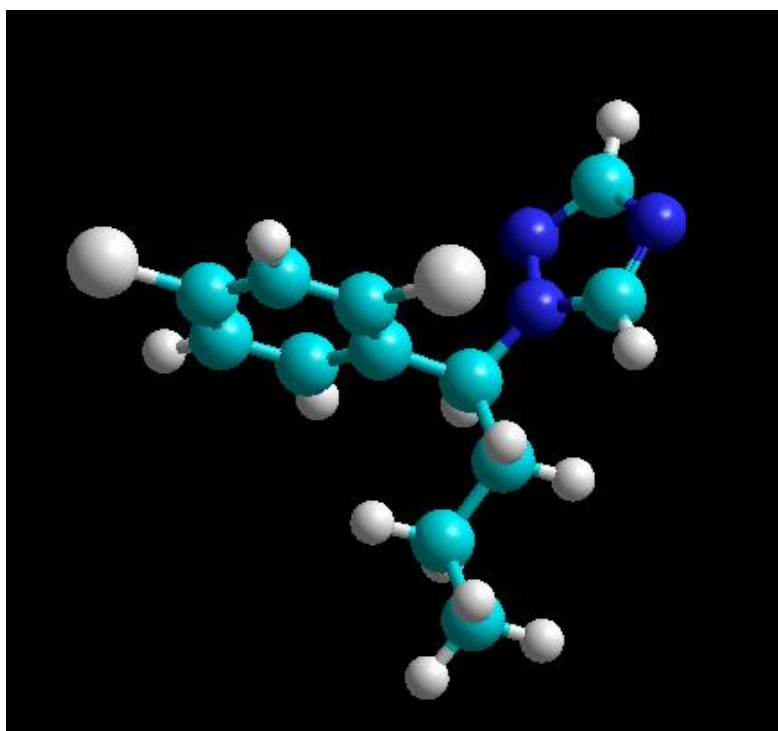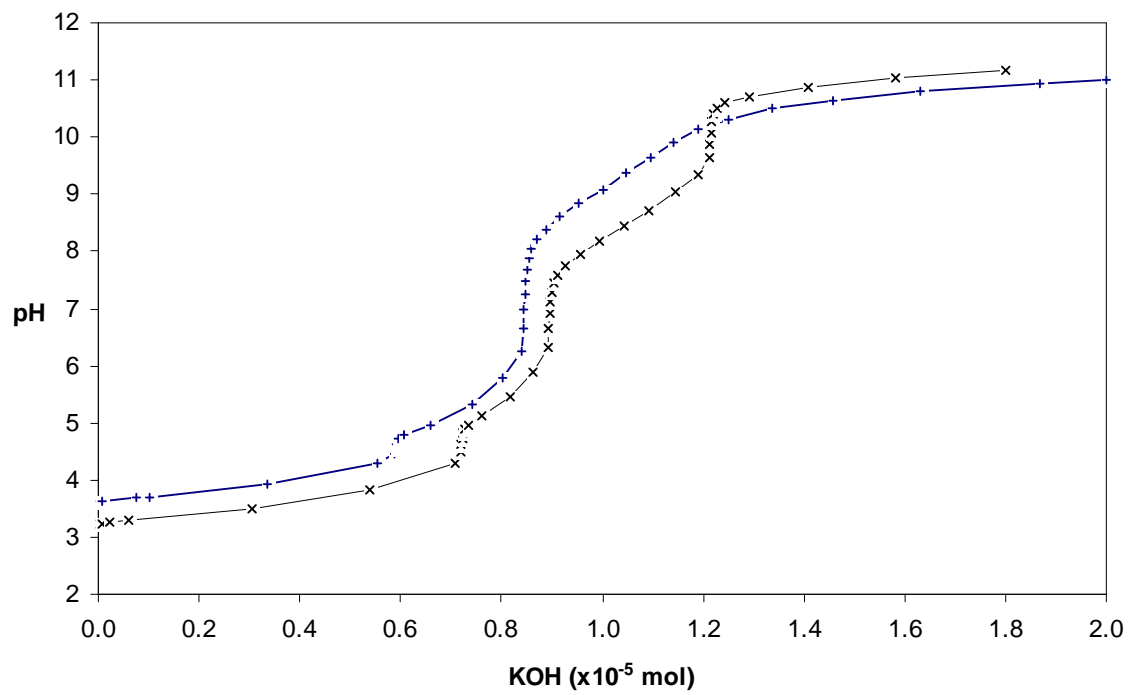
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257 (c) Penconazole *R*

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260 (d) Penconazole *S*

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Fig. 2 Titration curves for 2 mg Tebuconazole (×) and Penconazole (+) with 0.01 mol·dm^{-3}

KOH in the 20% (v/v) AN/Water, 0.1 mol·dm^{-3} KNO$_3$ medium, after acidification with an

equimolar amount of HNO$_3$.