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# Poly(vinyl alcohol) as a novel constituent to form aqueous two-phase systems with acetonitrile: Phase diagrams and partitioning experiments

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## A B S T R A C T

In this work it is shown, for the first time, that aqueous solutions of poly(vinyl alcohol) (PVA) and acetonitrile (ACN) undergo liquid–liquid demixing and form aqueous two-phase systems (ATPS). The ternary phase diagrams composed of PVA of different molecular weights, namely 9000–10,000, 13,000–23,000, 31,000–50,000  $\text{g mol}^{-1}$  and 85,000–124,000  $\text{g mol}^{-1}$ , acetonitrile and water, and the respective tie-lines were determined at 25 °C. In all the systems investigated, ACN is enriched in the top phase while PVA is found in the bottom phase. To explore the potentiality of these ATPS for extraction strategies, the partitioning of vanillin among the coexisting phases was further evaluated. The effects of ACN and PVA concentrations and temperature toward the vanillin partitioning were also investigated. An increased vanillin partitioning to the top phase was verified with the increase of the ACN concentration; however, the partition coefficient of vanillin for the ACN-rich phase decreases with the increase on both the PVA concentration and temperature. The highest partition coefficient of vanillin for the ACN-rich phase (2.24) was found with the system composed of 49 wt% of ACN + 9 wt% of PVA 9000–10,000  $\text{g mol}^{-1}$  at 5 °C, with a recovery of 79%. The vanillin migration to the top phase is spontaneous and an exothermic process.

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**Keywords:** Aqueous two-phase system; Acetonitrile; Poly(vinyl alcohol); Vanillin; Partition; Extraction

## 1. Introduction

Poly(vinyl alcohol) (PVA) is a non-toxic, amorphous and biodegradable synthetic high- $\kappa$ -polymer presenting, in addition, a randomly coiled and highly flexible chain when in solution (Senga and Sankhala, 2007). This polymer is obtained by hydrolysis of polyvinyl acetate and its properties depend on the polymerization and hydrolysis conditions, as well as on the drying and grinding (Krumova et al., 2000). PVA is a relatively water soluble polymer with interesting

properties, such as a low cost, good surface alignment effects, and an excellent film forming, emulsifying and adhesive properties (Chou, 2010). Due to these features, PVA is broadly used as a thickening, emulsifying or film-forming agent or as an adhesive in many household and industrial applications, especially in the paper, textile and chemical industries (Marusincová et al., 2013). In addition, PVA is usually employed as a phase forming-component in aqueous two-phase systems (ATPS) when combined with inorganic salts (Wu et al., 2001).

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Acetonitrile, CH<sub>3</sub>CN, also known as cyanomethane or methyl cyanide, is an aprotic and polar organic solvent miscible with water in all proportions (Mandal et al., 2011). The ACN molecules do not strongly interact with themselves and leave a hydrogen-bond network formed by water (Takamuku et al., 1998). ACN is also a by-product from the manufacture of acrylonitrile (Pollak et al., 2000), which is widely used by industry in the production of perfumes, rubber products, pesticides or pharmaceuticals (Zhang et al., 2011), or as a mobile phase in reverse phase high performance liquid chromatography (HPLC) (Taha et al., 2012; Gu and Shih, 2004).

ATPS have been divided into two major categories: those which contain two polymers, such as polyethylene glycol (PEG) + dextran (Karakatsanis and Liakopoulou-Kyriakides, 2007; Chen and Lee, 1995) and PEG + maltodextrin (Silva and Meirelles, 2000) systems, and those formed by a polymer and a salt, such as PEG-phosphate-based (Silva et al., 2013; Lima et al., 2002; Köhler et al., 1991) and PEG-citrate-based salts (Neves et al., 2012; Porto et al., 2008). However, many other pairs of compounds can be used to form ATPS, especially alcohol + salt (Reis et al., 2012), ionic liquid + salt (Ventura et al., 2012b; Neves et al., 2009; Gutowski et al., 2003), ionic liquid + PEG (Pereira et al., 2013a; Freire et al., 2012) and ionic liquid + carbohydrate (Freire et al., 2011) mixtures. Recently, it was demonstrated that acetonitrile (ACN) can also form ATPS when combined with carbohydrates (Cardoso et al., 2013) or polyols (Cardoso et al., 2014).

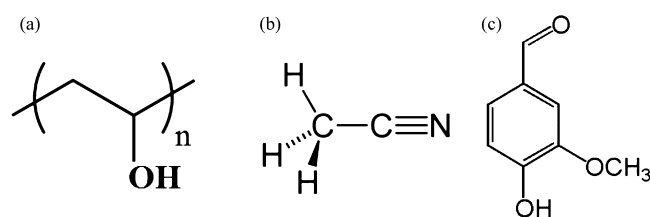
ATPS can be seen as an alternative option for extraction, separation and purification purposes over conventional systems which usually require organic solvents (Martínez-Aragón et al., 2009). Moreover, ATPS are easy to scale up, present low cost and typically lead to a high product purity as well as to a high yield, while maintaining the biological activity of the molecules due to their water-rich environment (Lu et al., 2013; Rito-Palomares et al., 2000). These systems have been used in the recovery and purification of many biomolecules, namely enzymes (lipase – Ventura et al., 2012a; Barbosa et al., 2011; Souza et al., 2010), antioxidants (rutin – Reis et al., 2014; gallic acid – Cláudio et al., 2012), alkaloids (caffeine – Cláudio et al., 2013; theobromine, theophylline, nicotine and caffeine – Passos et al., 2013), antibiotics (tetracycline – Pereira et al., 2013b; Wang et al., 2010) and antibodies (Azevedo et al., 2009; Samatou et al., 2007).

This work addresses novel ATPS based on acetonitrile and poly(vinyl alcohol) of different molecular weight. The corresponding ternary phase diagrams, tie-lines and tie-line lengths at 25 °C were firstly determined. Further, their potential application to extract or separate a model antioxidant – vanillin (3-methoxy-4-hydroxybenzaldehyde) – was evaluated. This biomolecule is used here as a standard biomolecule and representative of the phenolic compounds with antioxidant properties. Vanillin is the major component of natural vanilla and it is widely used as a flavoring material in confectionery, food products, beverages, perfumes and in pharmaceutical preparations (Walton et al., 2003).

## 2. Materials and methods

### 2.1. Materials

The ATPS studied in this work are formed by water, poly(vinyl alcohol) of different molecular weights ( $M_w = 9000\text{--}10,000\text{ g mol}^{-1}$ ;  $13,000\text{--}23,000\text{ g mol}^{-1}$ ;  $31,000\text{--}50,000\text{ g mol}^{-1}$ ;  $85,000\text{--}124,000\text{ g mol}^{-1}$ ) and acetonitrile. All



**Fig. 1 – Chemical structures of the phase-forming components used in the ATPS formation and of the biomolecule used as a partitioning solute: (a) poly(vinyl alcohol); (b) acetonitrile; (c) vanillin.**

reagents were purchased from Sigma–Aldrich: poly(vinyl alcohol) 9000–10,000 g mol<sup>-1</sup> 80% hydrolysed (>99.5 wt% pure), poly(vinyl alcohol) 13,000–23,000 g mol<sup>-1</sup> 87–89% hydrolysed, poly(vinyl alcohol) 31,000–50,000 g mol<sup>-1</sup> 98–99% hydrolysed, poly(vinyl alcohol) 85,000–124,000 g mol<sup>-1</sup> >99 wt% hydrolysed, acetonitrile (HPLC grade with a purity of 99.9 wt%) and vanillin (>99 wt% pure). The chemical structures of the phase-forming components of the ATPS investigated and of the target biomolecule used in the partitioning experiments are shown in Fig. 1. Distilled and deionized water was used in all experiments.

### 2.2. Phase diagrams and tie-lines

The ternary phase diagrams for poly(vinyl alcohol) and acetonitrile were determined at  $(25 \pm 1)^\circ\text{C}$  and atmospheric pressure by the cloud point titration method. Stock solutions of poly(vinyl alcohol) ( $\approx 5\text{ wt}\%$ ) and acetonitrile ( $\approx 80\text{ wt}\%$ ) were previously prepared and used for the determination of the phase diagrams. Repetitive drop-wise addition of the ACN solution to the aqueous solution of each PVA was carried out until the detection of a cloudy solution, followed by the drop-wise addition of ultra-pure water until the detection of a monophasic region (clear and limpid solution). These additions were carried out under continuous stirring and the saturation curves were determined gravimetrically within  $\pm 10^{-5}\text{ g}$ .

The tie-lines (TLs) were obtained through a gravimetric method originally described by Merchuck et al. (1998). Several mixtures at the biphasic region of the ternary systems were prepared, vigorously stirred, and allowed to reach equilibrium and phase separation, for a minimum of 18 h at  $(25 \pm 1)^\circ\text{C}$ . Initial soluble aqueous solutions of each PVA were prepared followed by the subsequent addition of pure ACN. After the equilibration step, the top and bottom phases were carefully separated and weighted within  $\pm 10^{-5}\text{ g}$ . Each individual TL was determined by the application of the lever-arm rule, which describes the relationship between the weight of the top phase and the overall system weight and composition. For that purpose, the binodal curves were correlated using Eq. (1),

$$Y = A \exp\{(B \times X^{0.5} - (C \times X^3))\} \quad (1)$$

where Y and X are the acetonitrile and poly(vinyl alcohol) weight fraction percentages, respectively, and A, B and C are constants parameters obtained by the regression of the experimental binodal data.

The determination of the TLs ( $Y_T$ ,  $Y_B$ ,  $X_T$  and  $X_B$ ) was then accomplished by solving the a system of four equations based

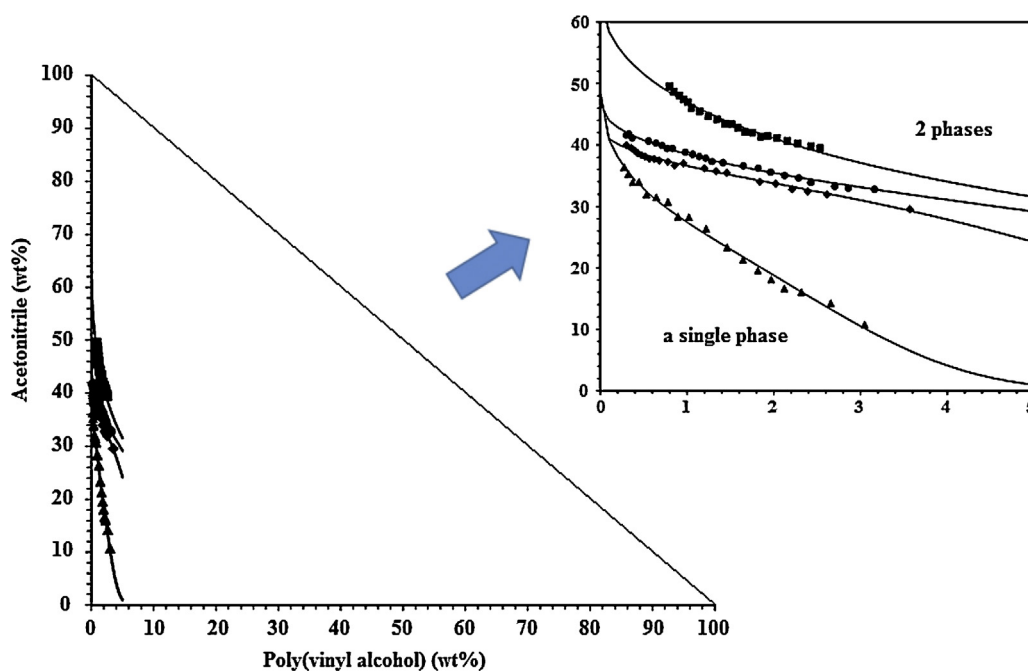


Fig. 2 – Phase diagrams for the systems composed of acetonitrile + poly(vinyl alcohol) + water at 25 °C and atmospheric pressure: poly(vinyl alcohol) 9000–10,000 g mol<sup>-1</sup> (■); 13,000–23,000 g mol<sup>-1</sup> (●); 31,000–50,000 g mol<sup>-1</sup> (◆) and 85,000–124,000 g mol<sup>-1</sup> (▲). The lines correspond to the fitting by Eq. (1).

on Eq. (1) (Merchuck et al., 1998). The respective tie-line lengths (TLLs) were determined through the application of Eq. (2),

$$\text{TLL} = \sqrt{(X_T - X_B)^2 + (Y_T - Y_B)^2} \quad (2)$$

### 2.3. Partitioning of vanillin

The liquid-liquid systems for the partitioning of vanillin were prepared in graduated glass centrifuge tubes weighing the appropriate amounts of poly(vinyl alcohol) 9000–10,000 g mol<sup>-1</sup>, acetonitrile and an aqueous solution containing vanillin. Vanillin was at 0.4 g L<sup>-1</sup> in the initial aqueous solution. After the complete mixing of all components for a given mixture composition, each system was centrifuged at 2000 × g for 10 min to favor the phase separation, and then each tube was placed in a thermostatic bath at 5–35 °C for at least 18 h. The volume of each phase was measured and both phases were further separated for the quantification of vanillin and for the determination of their pH values. At least three independent replicates were made and the average partition coefficients and associated standard deviations were therefore determined.

The concentration of vanillin at each aqueous phase was quantified through UV-spectroscopy, using a Varian Cary 50 Bio UV-vis spectrophotometer, and at a wavelength of 280 nm using a calibration curve previously established.

The partition coefficient of vanillin was determined taking into account the concentration of the antioxidant in each phase and according to,

$$K_{\text{van}} = \frac{C_T}{C_B} \quad (3)$$

where  $K_{\text{van}}$  is the partition coefficient of vanillin, C represents the vanillin concentration, and the subscripts T and B denote the top (acetonitrile-rich) and bottom (poly(vinyl alcohol)-rich) phases, respectively.

The recovery of vanillin ( $R_T$ ) in the top phase was evaluated using Eq. (4),

$$R_T = \frac{100}{1 + (1/(K_{\text{van}} \times R_v))} = \frac{C_T V_T}{C_T V_T + C_B V_B} \quad (4)$$

where  $R_v$  is the ratio between the volumes of the top ( $V_T$ ) and bottom ( $V_B$ ) phase.

At least three independent ATPS were prepared and the vanillin quantified in each phase for a particular set of conditions, allowing thus to determine the associated standard deviations associated to the partition coefficient and recovery of vanillin.

The standard molar Gibbs free energy of transfer ( $\Delta_{\text{tr}}G_o^m$ ), the standard molar enthalpy of transfer ( $\Delta_{\text{tr}}H_o^m$ ) and standard molar entropy of transfer ( $\Delta_{\text{tr}}S_o^m$ ) associated with the vanillin partition coefficient were determined by the van't Hoff approach at different temperatures (from 5 to 35 °C). The following isochors were used to determine the molar thermodynamic functions of transfer (Eq. (5)):

$$\Delta_{\text{tr}}G_o^m = \Delta_{\text{tr}}H_o^m - T\Delta_{\text{tr}}S_o^m = -RT \ln(K_{\text{van}}) \quad (5)$$

where T is the temperature (Kelvin) and R is the ideal gas constant. The enthalpy and entropy contributions can be directly deduced from the linear approximation of  $\ln(K_{\text{van}})$  versus  $T^{-1}$ .

## 3. Results and discussion

### 3.1. Phase diagrams and tie-lines

A remarkable number of phase diagrams can be found in literature for ATPS; however, this is the first evidence for ATPS based on acetonitrile and PVA of different molecular weight and their further application in the extraction of vanillin.

The solubility of a given solute in water is affected by the presence of other species that can improve or decrease such solubility in the aqueous medium. The relatively high

**Table 1 – Correlation parameters used to describe the experimental binodal data by Eq. (1) and respective standard deviations ( $\sigma$ ) and correlation coefficients ( $R^2$ ).**

PVA ( $\text{g mol}^{-1}$ )	$A \pm \sigma$	$B \pm \sigma$	$C \pm \sigma$	$R^2$
9,000–10,000	$64.72 \pm 3.46$	$-0.321 \pm 0.057$	$1.87\text{E}-13 \pm 4.13\text{E}-3$	0.9764
13,000–23,000	$46.80 \pm 0.36$	$-0.195 \pm 0.008$	$0.0003 \pm 0.0003$	0.9896
31,000–50,000	$43.50 \pm 0.31$	$-0.174 \pm 0.008$	$0.0014 \pm 0.0003$	0.9908
85,000–124,000	$48.77 \pm 1.42$	$-0.554 \pm 0.037$	$0.0213 \pm 0.0028$	0.9921

solubility of PVA in water (taking into account that it is a polymer) is mainly due to hydrogen-bonding between water and the –OH groups of the polymeric chain (Senga and Sankhala, 2007).

The experimental phase diagrams for acetonitrile and different molecular weight PVA polymers were determined at 25 °C and atmospheric pressure. The experimental weight fraction data are provided in Supporting Information (Table S1). The corresponding phase diagrams are depicted in Fig. 2 and allow the analysis of the PVA molecular weight influence to induce the liquid–liquid demixing. In general, for any mixture within the biphasic envelope it is visible that relatively high amounts of acetonitrile and low content of PVA are present. This is a major result of the complete solubility of ACN and lower solubility of PVA in water. However, small amounts of PVA are enough to lead to the phase separation. In general, an increase on the molecular weight of PVA leads to a higher ability for liquid–liquid demixing. The aptitude of the polymer to form ATPS decreases in the order:  $9000\text{--}10,000 \text{ g mol}^{-1} < 13,000\text{--}23,000 \text{ g mol}^{-1} < 31,000\text{--}50,000 \text{ g mol}^{-1} < 85,000\text{--}124,000 \text{ g mol}^{-1}$ . As the molecular weight of the polymer is increased, the binodal curves shift to lower PVA and CAN concentrations. This is a main result of the decreasing solubility of the polymer in water when its molecular weight increases. Therefore, species with low affinity for water are more able to be phase separated and to form an ATPS. This trend is in agreement with other experimental results regarding ATPS composed of polymers and ionic liquids (Freire et al., 2012), polymers and salts or two polymers (Zafarani-Miattar and Sadeghi, 2001; Diamond and Hsu, 1989). It should be remarked that in all systems investigated, the PVA is enriched at the bottom phase while acetonitrile is enriched at the top phase.

Supplementary Table S1 related to this article can be found, in the online version, at doi:10.1016/j.chemd.2014.08.009.

Contrarily to more conventional ATPS composed of polymer-salt or polymer-polymer combinations, the PVA-ACN-based ATPS proposed here allow the simple recovery of the phase-forming components since ACN and water are volatile and the polymer can be easily recovered after an evaporation step. This possibility opens the door for a deeper investigation on these systems as alternative extraction/separation techniques when envisaging a more sustainable process with a combined recovery and reuse of the phase-forming components of a given ATPS. Furthermore, and although ACN presents a non-negligible toxicity, it's a major byproduct of the acrylonitrile manufacture and it is commonly used in the manufacture of pharmaceuticals (Carey et al., 2006). In this context, the novel ATPS presented here make use of a low-cost residue and deserve to be studied in pharmaceuticals purification steps.

The experimental binodal data were fitted using the empirical relationship described by Eq. (1). The fitted parameters A, B and C (estimated by least-squares regression) and the corresponding standard deviations ( $\sigma$ ), as well as the regression

coefficient ( $R^2$ ), are presented in Table 1. As can be seen by the correlation coefficients obtained, equation 1 provides an accurate description of the experimental binodal saturation curves. Furthermore, the representation of the fitting of the experimental data by Eq. (1) is shown in Fig. 2.

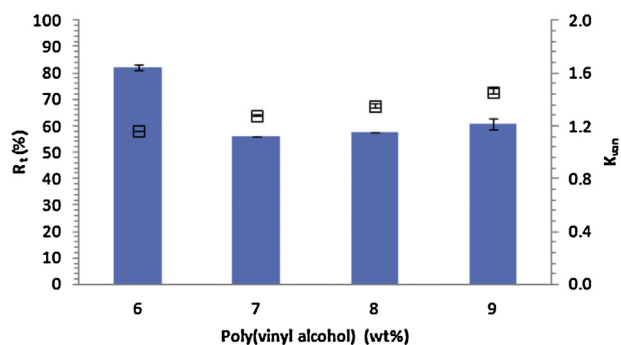
To complete the phase diagram, several TLs and the respective TLLs were further calculated and their values are given in Table 2. The TL data allow the knowledge of the coexisting phases' compositions for any mixture point along the same TL. In general, an increase in the molecular weight of the polymers leads to a reduction of the ACN content at the polymer phase. In fact, for sufficiently high molecular weight PVA polymers it is visible a complete exclusion of ACN at the PVA-rich phase for the initial mixture compositions considered.

### 3.2. Partitioning of vanillin

In order to evaluate the potential of the studied ATPS to be applied as extractive systems, it was further determined the partition coefficient and the recovery of vanillin at the acetonitrile-rich phase for biphasic systems composed of water, ACN and PVA with the molecular weight  $9000\text{--}10,000 \text{ g mol}^{-1}$ . At this stage, the effects of poly(vinyl alcohol) and acetonitrile concentrations and temperature were evaluated.

#### 3.2.1. Influence of the PVA concentration

The effect of the PVA ( $M_w = 9000\text{--}10,000 \text{ g mol}^{-1}$ ) initial concentration (6–9 wt%) on the partitioning of vanillin was studied, namely regarding its partition coefficient or recovery, and the results obtained are depicted in Fig. 3. For this study, the concentration of ACN was maintained constant at 45 wt%, as well as the temperature at 25 °C. It can be observed that under the conditions evaluated, vanillin preferentially partitions for the ACN-rich phase ( $K_{\text{van}} > 1.0$ ). Furthermore, the partition coefficient of vanillin increases with an increase in the PVA concentration. This trend is in close agreement with



**Fig. 3 – Influence of the poly(vinyl alcohol)  $9000\text{--}10,000 \text{ g mol}^{-1}$  concentration in the partition coefficient of vanillin ( $K_{\text{van}}$ ) (symbols) and in the recovery of vanillin ( $R_t$ ) (bars) in the ATPS composed of 45 wt% of acetonitrile at 25 °C.**



**Table 2 – Mass fraction composition for the TLs and respective TLLs, at the top (T) and bottom (B) phases, and initial biphasic composition of the mixture (M), for the systems composed of acetonitrile (Y) and poly(vinyl alcohol) (X) at 25 °C.**

Weight fraction composition (wt%) Acetonitrile + poly(vinyl alcohol) + water systems							
PVA (g mol <sup>-1</sup> )	Y <sub>M</sub>	X <sub>M</sub>	Y <sub>T</sub>	X <sub>T</sub>	Y <sub>B</sub>	X <sub>B</sub>	TLL
9000	51.78	6.28	55.93	0.21	3.92	76.22	78.07
–10,000	50.18	7.20	55.01	0.26	4.13	73.38	66.50
13,000	37.72	1.51	39.22	0.82	8.02	14.99	14.99
–23,000	35.58	2.03	36.44	1.63	12.40	12.81	30.02
31,000	40.00	0.50	41.34	0.08	0.51	13.12	42.86
–50,000	40.42	0.76	42.26	0.02	0.00	17.08	45.57
85,000	20.68	3.13	26.77	1.07	0.00	10.12	28.26
–124,000	30.25	1.02	31.27	0.63	0.00	12.58	33.48

our previous findings in ATPS composed of ACN and carbohydrates or polyols and where an increase on their molecular weight and concentration in aqueous media favors the partitioning of vanillin for the opposite phase (Cardoso et al., 2014, 2013). According to Babu et al. (2008) the volume occupied by the polymer increases with an increase in the polymer concentration and which results in a reduced space to accommodate biomolecules – volume exclusion effect. Bassani et al. (2007) observed the volume exclusion effect when partitioning lipase using an ATPS formed by PEG 10,000 g mol<sup>-1</sup> and potassium phosphate at pH 7.0 and 8 °C. The recovery of vanillin in the ACN-rich phase ranges between 56.27 and 82.17%. Since the recovery values are dependent on the volume of the phases, the partition coefficients trend does not directly translate on the recoveries. In fact, the highest recovery of vanillin in the ACN-rich phase was observed with the system composed of 6 wt% of PVA.

### 3.2.2. Influence of the acetonitrile concentration

To analyze the effect of the ACN concentration, a series of ATPS containing 9 wt% of poly(vinyl alcohol) ( $M_w = 9000\text{--}10,000\text{ g mol}^{-1}$ ) and 45–51 wt% of ACN at 25 °C, were assessed. The results obtained are depicted in Fig. 4.

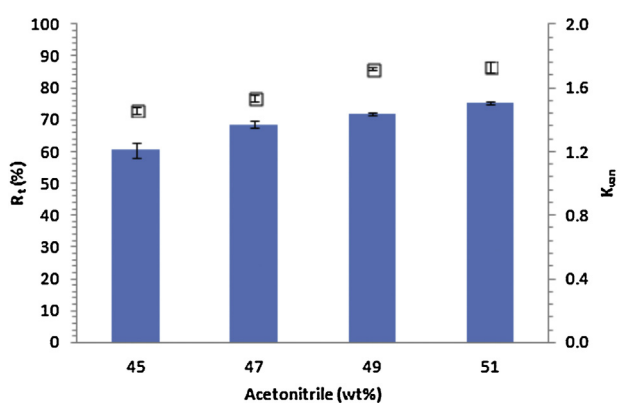
When the concentration of ACN in the total mixture composition increases, the partition coefficient of vanillin also increases. The  $K_{\text{van}}$  reaches a value of 1.5 when using 51 wt% of ACN (Fig. 4), a value very close to that found using 49 wt% of ACN. The recoveries of vanillin in the top phase follow the same trend of the partition coefficient and reach a maximum

value of about 80% for 51 wt% of ACN. Cardoso et al. (2013) also previously demonstrated that vanillin preferentially migrates for the more hydrophobic phase (acetonitrile-rich phase over the carbohydrate-rich phase) with  $K_{\text{van}} > 3.0$  and recoveries of vanillin ranging between 73 and 95%, agreeing with the results obtained here.

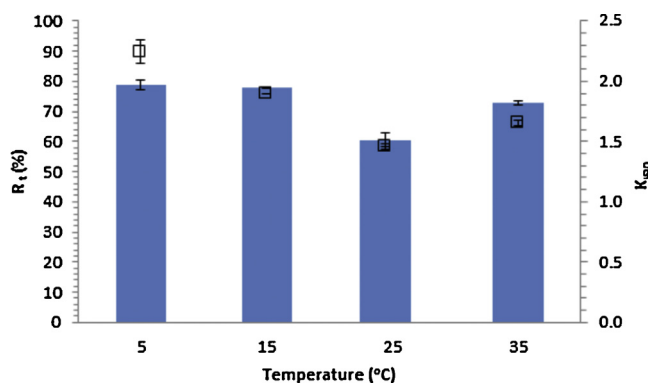
### 3.2.3. Influence of temperature

Based on the previous results, a system composed of 49 wt% of ACN + 9 wt% PVA ( $M_w = 9000\text{--}10,000\text{ g mol}^{-1}$ ) was selected for the study of the vanillin partitioning at different temperatures (from 5 to 35 °C). The results obtained are presented in Fig. 5.

The  $K_{\text{van}}$  and  $R_t$  decrease from 2.24 to 1.46 and from 78.84% to 60.43%, respectively, with the temperature increase from 5 °C to 35 °C. In general, an increase in temperature is not favorable for the partitioning of vanillin for the ACN-rich phase. This trend is in agreement with the data provided by Cláudio et al. (2010) where an increase in temperature also reduces the partitioning of vanillin for an ionic-liquid-rich phase. Saravanan et al. (2008) reported that the partition coefficients of proteins, such as myoglobin and ovalbumin, also decrease with an increase in the temperature of the system. However, temperature has a small influence on the distribution behavior of mulberry anthocyanins ( $2.0 \leq K \leq 2.3$ ) in the temperature range from 25 °C to 50 °C (Wu et al., 2011). Therefore, depending on the solute being partitioned, the increase on temperature can favor or not its preferential migration for a given phase.



**Fig. 4 – Influence of the acetonitrile concentration on the partition coefficient of vanillin ( $K_{\text{van}}$ ) (symbols) and in the recovery of vanillin ( $R_t$ ) (bars) in the ATPS composed of 9 wt% of poly(vinyl alcohol) 9000–10,000 g mol<sup>-1</sup> at 25 °C.**



**Fig. 5 – Influence of temperature on the partition coefficients of vanillin ( $K_{\text{van}}$ ) (symbols) and in the recovery of vanillin ( $R_t$ ) (bars) in ATPS composed of 9 wt% of poly(vinyl alcohol) 9000–10,000 g mol<sup>-1</sup> and 45 wt% of acetonitrile.**

The thermodynamic parameters involved in the partitioning of vanillin where obtained from the linear equation fitted to the experimental data. The obtained regression coefficient of  $\ln(K_{\text{van}})$  versus  $T^{-1}$  is 0.946 and can thus support a reasonable determination of  $\Delta_{\text{tr}}H_m^\circ$  and  $\Delta_{\text{tr}}S_m^\circ$ .

The  $\Delta_{\text{tr}}G_m^\circ$  at 5 °C (highest  $K_{\text{van}}$  and  $R_t$  values for vanillin partitioning) is negative ( $-1.81 \text{ kJ mol}^{-1}$ ) meaning that the transfer process of vanillin from the PVA-rich to the ACN-rich phase is spontaneous. In fact, in all situations,  $K_{\text{van}} > 1$ . In addition, this migration of vanillin is an exothermic process ( $\Delta_{\text{tr}}H_m^\circ = -6.94 \text{ kJ mol}^{-1}$ ). However, it should be remarked that the entropic contribution is also relevant for the partitioning process ( $\Delta_{\text{tr}}S_m^\circ = -18.46 \text{ J mol}^{-1} \text{ K}^{-1}$ ).

#### 4. Conclusions

In this work it was shown that novel ATPS can be formed by combining acetonitrile and PVA of different molecular weights, at specific concentrations, in aqueous media. The ternary phase diagrams, and respective tie-lines and tie-line lengths were determined at 25 °C and at atmospheric pressure. Different system compositions of the ATPS formed by PVA ( $M_w = 9000\text{--}10,000 \text{ g mol}^{-1}$ ) and ACN were also employed for the study of the partitioning behavior of vanillin. In all situations, vanillin preferentially migrates for the acetonitrile-rich phase (top phase). An increase in the PVA concentration leads to higher partition coefficients of vanillin where an increase in the ACN concentration results in the opposite behavior. Furthermore, an increase in temperature leads to a decrease on  $K_{\text{van}}$ . The highest  $K_{\text{van}}$  and  $R_t$  values occurred in the ATPS composed of 49 wt% of ACN+9 wt% of PVA ( $M_w = 9000\text{--}10,000 \text{ g mol}^{-1}$ ) and at 5 °C ( $K_{\text{van}} = 2.24 \pm 0.10$  and  $R_t = 78.84 \pm 1.52\%$ ). The vanillin migration to top phase is spontaneous and an exothermic process.

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