Rapid Evaporation-driven Chemical Pre-Concentration and Separation on Paper
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Abstract
Airflow-enhanced evaporation is investigated as a method for rapid chemical pre-concentration on a thin porous substrate. The mechanism is described by combining 1D models of capillary rise, chromatography and pervaporation concentration. It is shown that the effective length of the column can be shorter than its actual length, allowing concentrate to be held at a stagnation point and then released for separation, and that the Péclet number, which determines concentration performance, is determined only by the substrate properties. The differential equations are solved dynamically, and it is shown that faster concentration can be achieved during capillary filling. Experiments are carried out using chromatography paper in a ducted airflow and concentration is quantified by optical imaging of water-soluble food dyes. Good agreement with the model is obtained, and concentration factors of $\approx 100$ are achieved in 10 mins using Brilliant Blue FCF. Partial separation of Brilliant Blue from Tartrazine is demonstrated immediately following concentration, on a single un-patterned substrate. The mechanism may provide a method for improving the sensitivity of lab-on-paper devices.

KEYWORDS: Lab-on paper, chemical concentrator, paper chromatography
1. Introduction

Great strides have been made in developing paper as a substrate for water-based lab-on-a-chip devices [1, 2], with the aim of providing low-cost medical diagnosis for the developing world. Flow is driven by capillary action, channels have been defined using wax patterning, ink-jet printing, toner printing and boundary shaping [3-6], and devices have been fabricated with hollow and closed channels [7, 8]. Detection has mainly been carried out using colorimetric assays for off-site analysis via the ubiquitous smartphone [9, 10]; however, electrochemical detection has also been investigated [11, 12]. Several reviews are available [13-15].

Several well-known methods of preconcentration have been adapted to increase sensitivity in paper devices, including isotachophoresis [16, 17], isoelectric focusing [18] and field amplified sample stacking [19]. The most commonly used method is ion concentration polarization (ICP) [20-23]. This can provide concentration factors of 100-1000 in 2-10 minutes after filling, but require nanoporous membranes, electrodes and relatively high voltages.

In this paper we explore an alternative mechanism that can operate on a bare substrate, concentrate during filling and provide a hold-up function that allows subsequent separation operations. The driving force is solvent evaporation, which is well known to concentrate non-volatile compounds at stagnation points. An everyday example, coffee staining (which involves ambient evaporation of a finite source in a semi-cylindrical geometry) has been extensively studied [24, 25], and the effect has also been exploited in paper devices using ambient evaporation [26, 27]. Here we investigate inherent limits, and show that a performance comparable to ICP can be achieved using evaporation-driven capillary pumping.

Pumps based on ambient evaporation were developed some years ago [28-30]. Following the observation of permeation-induced flows in poly(dimethyl siloxane) [31, 32], airflow-enhanced PDMS pervaporation pumps were used to induce concentration in microfluidic channels [33-35]. Similar systems have been constructed for concentration of bacteria and viruses [36, 37], and concentration has also been carried out using microstructured media [38-40] Evaporation pumps with greater capacity were subsequently developed using porous media [41-43], and used for
sample injection [44, 45].

We show that ducted airflow can provide the enhanced evaporation needed for rapid concentration of water-soluble dyes on paper. The device is extremely simple. A paper strip is filled with a dilute aqueous solution from a reservoir by capillary action. Evaporation then leads to an advective flow that concentrates non-volatile compounds near or at the tip of the column. The mechanism involves capillary rise, microfluidic concentration and paper chromatography. All are very well known. However, there are three main differences from earlier work. Firstly, we show theoretically that the effective length of the column can be shorter than its actual length if the rate of evaporation is high enough. Consequently, concentrate may be held at a stagnation point and then released for capillary-pumped separation. Secondly, we show that the Péclet number, which determines concentration performance, is defined only by the properties of the substrate. Thirdly, we solve the governing equations dynamically, and show that faster concentration can be achieved during capillary filling. Theoretical predictions are confirmed using dyes and un-patterned chromatography paper, with a fan-driven ducted airflow derived from ambient air. Rapid concentration requires relatively high fan drive power (15 W). However, there is scope to reduce power using dry air, optimised duct dimensions and local heating. A unique advantage is control of stagnation, which allows concentration to be followed by other capillary-pumped operations.

A model for capillary rise in the presence of evaporation is presented in Section 2. The solvent velocity profile is combined with a differential equation model of separation in Section 3, and concentration is simulated for different initial wetting conditions. Experimental materials and methods are described in Section 4. Concentration and separation of concentrate are demonstrated in Section 5. Conclusions are drawn in Section 6.
2. **Capillary rise**

In this Section, we derive the solvent velocity profile obtained in evaporation-dominated capillary rise, using a combination of earlier models.

*Device geometry*

Figure 1 shows an evaporation concentrator, arranged to allow comparison with earlier pervaporation devices [46, 47]. Solutes are transferred from a reservoir at the right-hand end towards a tip at the left via a porous substrate of length $L$, thickness $d$ and uniform (but unimportant) width. The medium transports a mobile phase by capillary action. This phase is assumed to be volatile, and evaporation is enhanced by airflow on either side. In contrast, solutes are assumed to be soluble, but non-volatile. Consequently, solutes will accumulate at the tip, in a concentration profile derived from a balance between the effects of advection and diffusion.

One key difference from earlier work is the replacement of a pervaporation membrane over a channel with a porous substrate that allows increased evaporation and chromatographic effects. At low evaporation rates, solutes will concentrate at the tip; however, increased evaporation can lead to concentration at a closer point $H_e$ from the reservoir, where concentrate may be held up. Very high evaporation can also allow concentration during the wetting phase. A model may be constructed by combining concepts from capillary rise, chromatography and concentration. For simplicity, we use 1D analysis throughout. To aid comparison with standard models, we use the $Y$ co-ordinate for capillary ascent and the $X$ co-ordinate for concentration.

*Capillary transport and evaporation*

In a vertical capillary tube, rise is modelled using the Lucas-Washburn equation, which balances the effects of surface tension, gravitational force and viscous drag in Poiseuille flow [48, 49]. For media such as paper, the equation must be modified to include permeability and porosity. Early
models of wicking assume uniform filling [50], while later models consider the observed distribution of fluid [51]. Evaporation is typically ignored, except in the related phenomenon of rising damp [52]. Here the substrate is horizontal, so gravitational force can be ignored, but evaporation must now be included. However, for simplicity we assume full filling.

Assuming a fluid velocity $V$ in the $Y$ direction, a uniform volumetric rate of evaporation $u_e$ per unit area from the two sides together and a rise $H$ from the reservoir, the continuity equation at time $T$ is:

$$Vd = v_e (H - Y) + d \frac{dH}{dT}$$

(1)

Here $v_e = u_e/\varepsilon$, where $\varepsilon$ is the porosity. Equation 1 can be derived from a balance between the input fluid flow (the first term), the loss of fluid by evaporation (the second) and the change of stored fluid due to extension of the column (the third), assuming a constant column width.

In porous media, the volumetric flow rate $q$ per unit cross-sectional area is linked to the gradient in pressure $P$ using Darcy’s Law [53]:

$$q = \frac{-\kappa}{\eta \varepsilon \varepsilon} \frac{\partial P}{\partial Y}$$

(2)

Here $\kappa$ is the permeability of the stationary phase and $\eta$ is the dynamic viscosity of the mobile phase. The relation between flow and velocity is $q = \varepsilon V$. Substitution gives:

$$-\left(\frac{\kappa}{\eta \varepsilon}\right) \frac{\partial P}{\partial Y} = \left(\frac{v_e}{d}\right) (H - Y) + \frac{dH}{dT}$$

(3)

Integrating both sides between $Y = 0$ and $Y = H$ then gives:

$$-\left(\frac{\kappa}{\eta \varepsilon}\right) [P(H) - P(0)] = \left(\frac{v_e}{2d}\right) H^2 + H \frac{dH}{dT}$$
Since $P(0) - P(H) = P_c$, where $P_c$ is the capillary pressure, we then obtain:

$$H \frac{dH}{dT} = \frac{P_c \kappa}{\eta \varepsilon} - \frac{v_c H^2}{2d}$$

(4)

Noting that $H \frac{dH}{dT} = \frac{1}{2} \frac{dH^2}{dT}$, Eqn. (5) can be recast as a first-order differential equation in $H^2$, namely:

$$\frac{dH^2}{dT} + \frac{v_c H^2}{d} = \frac{2P_c \kappa}{\eta \varepsilon}$$

(5)

For the initial conditions $H = 0$ at $T = 0$ the solution can now be found analytically in terms of a complementary function and a particular integral as:

$$H = H_e \sqrt{1 - \exp\left(-\frac{T}{\tau_e}\right)}$$

(6)

Here $H_e = \sqrt{(2\tau_e P_c \kappa / \eta \varepsilon)}$ is the evaporation-dominated rise, and $\tau_e = d/v_e$ is a characteristic time constant. Assuming fixed material and geometric parameters, $H_e$ is controlled by the value of $v_e$, and consequently may be smaller than the substrate length $L$ if $v_e$ is high enough.

Evaporation depends on temperature and relative humidity. However, the former is simple to control using air conditioning. At constant temperature, the rate of evaporation is a linear function of relative humidity (RH), falling to zero at 100% RH [54]. A decrease in RH from (say) 50% to 0% will therefore increase $v_e$ by 2, reduce $\tau_e$ by the same factor, and reduce $H_e$ by $\sqrt{2}$. Evaporation can also be enhanced by airflow, which continually replaces the boundary layer between the substrate and its surround with less saturated air.

To model similar phenomena with the substrate arranged vertically, $P_c$ is simply replaced by $P_c - \rho g H$, where $\rho$ is the fluid density and $g$ is the gravitational constant. Equation 5 must then generally be solved numerically. However, in the absence of evaporation, it is simple to show...
that this leads to a steady state capillary rise height \( H_c = \frac{P_c}{\rho g} \), allowing \( P_c \) to be determined experimentally from a measurement of \( H_c \).

**Velocity profile**

Differentiating Equation 7 and re-arranging, we may then obtain \( \frac{dH}{dT} \) as:

\[
\frac{dH}{dT} = \frac{(H_e^2 - H^2)}{2\tau_e H}
\]

(Equation 8)

Equations 1 and 8 then allow the general expression for the fluid velocity to be found as:

\[
V(Y, T) = \left( H - Y + \frac{(H_e^2 - H^2)}{2H} \right) / \tau_e \text{ for } Y \leq H
\]

(Equation 9)

Equation 9 implies that \( V(0, 0) \) is infinite, a result that in reality is prevented by additional flow restrictions. Ignoring this aspect, the velocity profile is an offset, truncated linear variation, which tends gradually to the equilibrium profile \( V_e(Y) = \frac{1}{\tau_e} (H_e - Y) \). In the \( X \) co-ordinate system of the following section, the velocity is \( U(X) = -V(H_e - X) \). Introducing the normalised time \( t = T/\tau_e \) and position \( x = X/H_e \), the normalised velocity \( u = U\tau_e/H_e \) is:

\[
u(x, t) = -(x - (1 - h) + (1 - h^2)/2h) \text{ for } x \geq 1 - h
\]

(Equation 10)

with \( h = \frac{H}{H_e} = (1 - \exp(-t))^{1/2} \).

Figure 2 shows normalised velocity profiles \( |u| \) calculated from Equation 10 for different normalised times \( t \). At short times, the substrate is only partly wetted; however, as \( t \) increases the liquid gradually fills the column from the right hand end. In all cases, the velocity profile is linear in the wetted region and zero elsewhere. In addition, each profile has an offset that gradually tends to zero, and at long times the profile tends to \( u = -x \).
Figure 2. Theoretical variation of normalised velocity $|u|$ with normalised position $x$ during capillary fill, at different normalised times $t$ as indicated.

**Substrate length**

The results above have been derived without considering the finite length $L$ of the substrate. If $L > H_e$, flow will clearly be held up at an intermediate stagnation point where concentration may occur. The disadvantage of this approach is that unsteady evaporation (for example, due to variations in ambient humidity) may lead to ‘wandering’ of the stagnation point and reduced concentration. However, if concentration is carried out for a fixed time, and evaporation is then suppressed, further capillary transport can be used for chromatographic separation. If $L < H_e$, on the other hand, a two-stage model must be used, the first describing rise to the tip, and the second relaxation to equilibrium. Here we shall ignore the details, and simply note that the equilibrium profile must then be $V_e'(Y) = (1/\tau_e) (L - Y)$, as is generally assumed directly. The advantage now is that the stagnation point is fixed, simplifying subsequent analysis. However, if $L$ is significantly less than $H_e$, concentration may be reduced. The most controllable approach is therefore likely to involve evaporation into dry air from a substrate length just less than $H_e$. 
3. **Concentration**

In this Section, we present a 1D theoretical model of concentration, based on standard differential equation models of chromatographic separation [55]. Such models consider transport, diffusion and concentration, but ignore transverse velocity profiles arising from shear forces and associated effects such as Taylor’s dispersion.

**Concentration**

We model a single solute with diffusion coefficient $D$ and concentration $C_L$ and $C_S$ in the liquid and solid phases, in the $X$ co-ordinate system. Assuming a variable flow velocity $U$, the continuity equation is:

$$
e D \frac{\partial^2 C_L}{\partial X^2} - \frac{\partial (UC_L)}{\partial X}$$

(11)

As usual, the first two terms describe accumulation, and the last two model diffusion and transport. Now, $C_S$ and $C_L$ are related by the adsorption isotherm $C_S = f(C_L)$. Writing $\partial C_S/\partial T = f' \partial C_L/\partial T$ and expanding the final term, we obtain:

$$(1 + FF') \frac{\partial C_L}{\partial T} = D \frac{\partial^2 C_L}{\partial X^2} - (U \frac{\partial C_L}{\partial X} + C_L \frac{\partial U}{\partial X})$$

(12)

Here $F = (1 - \varepsilon)/\varepsilon$ is the ratio of the volumes of the stationary and mobile phases. The function $f$ depends on the adsorption process, which may involve monolayer or multilayer adsorption; common models include the linear, Langmuir, Freundlich and BET isotherms [56]. Once $C_L$ is known, $C_S$ can be found from the isotherm, and the average concentration $C_{AV}$ (which is optically measurable) can be evaluated as $C_{AV} = \varepsilon C_L + (1 - \varepsilon)C_S$. Here for simplicity we assume the linear isotherm $C_S = a C_L$, where $a$ is Henry’s coefficient. In this case, $1 + FF' = 1/R_f$, where $R_f = 1/(1 + Fa)$ is the retardation or retention factor, and $C_{AV}$ is proportional to $C_L$.

In terms of the normalised time $t = T/\tau_e$ and normalised position $x = X/H_e$ we then obtain:
Here $Pe = H_e^2/D\tau_e$ is the Péclet number, which denotes the relative significance of advection and diffusion [57]. To reduce the absolute time taken to achieve concentration effects, $\tau_e$ should be small, so large values of $v_e$ and small values of $d$ are required. $R_f$ also affects timescales directly. It is well known from studies of pervaporation concentrators that $Pe$ controls the normalised width of the concentration peak [47]. However, since $H_e^2 = 2\tau_eP_c/\eta\varepsilon$, $Pe = 2P_c\kappa/\eta\varepsilon D$ here, and is therefore a function only of material parameters; variations in $v_e$ will not affect its value. Large values require substrates with high permeability, high capillary pressure and low porosity, solvents with low viscosity and solutes with low diffusion coefficients. Different solutes will have different $R_f$ and $Pe$. Consequently, each solute in a multi-component solution will concentrate differently.

**Wetting conditions**

There are now two different velocity profiles that may be assumed. For pre-wetted columns, we may assume that steady flow conditions have already been reached before the introduction of any solute, so $u = -x$. With this assumption, Equation 13 reduces to:

$$R_f^{-1} \frac{\partial C_L}{\partial t} = Pe^{-1} \frac{\partial^2 C_L}{\partial x^2} - (u \frac{\partial C_L}{\partial x} + C_L \frac{\partial u}{\partial x})$$

(13)

When $R_f = 1$, the model therefore corresponds exactly to previous descriptions of pervaporation concentrators [47]. For a single solute, the evolution of local concentration when the input is connected to a reservoir with a concentration $C_{L0}$ at $t = 0$ can be found by solving Equation 14 subject to the conditions $C_L(x, 0) = 0$, $C_L(1, t) = C_{L0}$, $\partial C_L/\partial x = 0 \mid_{x=0}$. Partial differential equations may easily be solved numerically for fixed conditions; here we have used the MATLAB® solver 'pdepe'. Conversely, if the column is initially dry, the full velocity profile in Equation 10 must be used, and the equation integrated with a moving boundary condition $\partial C_L/\partial x = 0 \mid_{x=0}$. It is now
harder to obtain a solution. Here, we have circumvented the problem by retaining the condition \( \partial C_L/\partial x = 0 \rvert_{x=0} \) so the same solver may be used. We then modify Equation 9 so that there is no flow \((u = 0)\) and no diffusion \((Pe^{-1} = 0)\) in the 'dry' part of the column, namely \(0 \leq x \leq h\). The price paid for the simplicity of this approach is a reduction in accuracy caused by discontinuity.

**Numerical results**

Figure 3 shows the spatial variation of the concentration factor \(CF = C_L/C_{L0}\) near \(x = 0\) at different normalised times \(t\) as indicated, for low values of \(R_f\) (0.25) and \(Pe\) (1,000). The full and dashed lines show results for substrates that are initially wet and dry, respectively. Here, the retardation factor and Péclet number are both so small that the column has filled before any significant concentration takes place, and similar variations of \(CF\) are obtained in each case. The concentration profile gradually evolves until the column is filled with solute, and then stabilises into the so-called ‘hyperbolic ramp’ \(CF = 1/x\) for large values of \(x\); however, for small values of \(x\), the profile never stabilises, but instead tends to the Gaussian variation \(CF = R_f(t - t_f) \sqrt{2Pe/\pi} \exp(-Pe x^2/2)\), where \(t_f\) is a normalised filling time. The width of the peak clearly depends on \(1/\sqrt{Pe}\), while its maximum increases linearly, at a rate dependent on \(\sqrt{Pe}\) [47].

![Figure 3. Spatial variation of normalised concentration $C_L/C_{L0}$, at different normalised times $t$ as indicated, for $R_f = 0.25$, $Pe = 1000$. Full and dashed lines show pre-wetted and un-wetted cases, respectively.](image)

Figure 4 shows the associated time-variations of maximum concentration. The results for initially wet and dry substrates (full and dashed lines, respectively) are again almost indistinguishable: in each case, the relative concentration rises slowly until the column is filled with solute, and then rises more rapidly until the linear regime is reached.
Figure 4. Variation of peak normalised concentration $C_L/C_L0$ with normalised time $t$, for $R_f = 0.25$, $Pe = 1,000$. Full and dashed lines show pre-wetted and un-wetted cases, respectively.

Figure 5 shows results corresponding to Figure 3, but for larger values of $R_f$ (1.00) and $Pe$ (16,000). The timescale has been reduced by a factor of four, to compensate for the fourfold increase in $R_f$. At long times, the Gaussian concentration peak has become sharper and higher, reflecting the fourfold increase in $\sqrt{Pe}$. In addition, there is now a clear difference between the results for initially wet and dry substrates. In the latter case, the retardation factor and Péclet number are both so large that concentration can take place during filling itself.

Figure 5. Spatial variation of normalised concentration $C_L/C_L0$, at different normalised times $t$ as indicated, for $R_f = 1$, $Pe = 16,000$. Full and dashed lines show pre-wetted and un-wetted cases, respectively.

Figure 6 shows results corresponding to Figure 4. As expected, the slope of the curve has risen by a factor of four in the linear regime, and once again, there is a difference between initially wet and dry substrates. However, the advantage of immediate concentration clearly becomes less significant at longer times.
Figure 6. Variation of peak normalised concentration $C_l/C_{LO}$ with normalised time $t$, for $R_f = 1, Pe = 16,000$. Full and dashed lines show pre-wetted and un-wetted cases, respectively.
4. Materials and methods

Concentration was investigated using cellulose paper and water-soluble food dye. In this Section, we describe materials characterisation and the equipment used to enhance evaporation. All experiments were performed in ambient laboratory conditions, which were monitored using a temperature and humidity meter (Type 971, Fluke), but not controlled. During 85 experiments performed over 1 year the mean temperature was 23.5°C, with a standard deviation of 1.5°C. The mean relative humidity was 46%, with a standard deviation of 5.5%. The final experiments were performed at 22°C, 50% RH.

Substrate properties

The substrate was 1 Chr paper (3001-845, Whatman, thickness and pore size 0.18 mm and 11 µm). Its properties were investigated using separate measurements of capillary rise, carried out with long strips of paper arranged vertically. The points in Figure 7 shows two sets of data, which were obtained firstly with a saturated surround to suppress evaporation. In the first case, the potential rise height is greater than the available length of paper (300 mm), while in the second case evaporation has limited the rise to \( H_e = 114 \) mm. The properties of the substrate were extracted from the first set of data by matching to the prediction of Equation 5 (full line), including the additional gravitational term but suppressing evaporation. Matching was carried out by adjusting the parameters \( \kappa / \eta \varepsilon \) and \( H_c \) to obtain best fits at the start and end of the curve, yielding \( \kappa / \eta \varepsilon = 7.7 \times 10^{-10} \text{ m}^3 \text{ s kg}^{-1} \) and \( H_c = 600 \text{ mm} \) (corresponding to a capillary pressure \( P_c = 5886 \text{ N/m}^2 \)).

Figure 7. Comparison between experimental and theoretical variation of capillary rise with time for Whatman Chr1 paper, with and without evaporation.
**Solute properties and Péclet number**

Concentration was carried out using water-soluble dyes, the triphenylmethane dye Brilliant Blue FCF and the synthetic lemon yellow dye Tartrazine. Brilliant Blue (erioglaucine disodium salt or C_{37}H_{34}N_{2}Na_{2}O_{9}S_{3}, also known as acid blue 9, E133 and CI 42090) is used as a food dye, despite some effects to health [58], and as a tracer in soil science. Its diffusion coefficient is known to be $D = 5.68 \times 10^{-10}$ m$^2$/s [59]. Maximum optical absorption occurs at 630 nm wavelength, allowing relative concentration to be deduced from changes in the red channel of an [R G B] image [60, 61]. Dye was obtained at $\geq 97\%$ purity (80717, Sigma Aldrich) and dissolved in de-ionised water, and a concentration maximising the experimental dynamic range ($6.25 \times 10^{-6}$ g/ml) was used. Its retardation factor on Whatman paper is quoted as close to unity [25], and we have verified it as $R_f \approx 0.98$. From the data above, a Péclet number $Pe = 2P_c\kappa/\eta\varepsilon D$ approaching 16,000 can be estimated, corresponding to the higher value used for earlier simulations. Tartrazine (C_{16}H_{9}N_{4}Na_{3}O_{9}S_{2}, acid yellow 23, E102, C.I. 19140) is also used as a food dye, but has been blamed for allergic reactions [62]. Its diffusion coefficient is $D = 4.9 \times 10^{-10}$ m$^2$/s$^{-1}$, and maximum absorption occurs at 425 nm wavelength [63]. Dye was obtained at 85% purity (TO388, Sigma Aldrich), and again dissolved in DI water at low concentration ($2.5 \times 10^{-5}$ g/ml). We have measured its retardation factor as $R_f \approx 0.88$.

**Ambient evaporation velocity and time constant**

The high Péclet number of Brilliant Blue suggests that sharp concentration profiles can be obtained in open air without special measures, and we have verified this experimentally. However, the time-scales are relatively long, and may be estimated as follows. The ambient evaporation velocity can be extracted from the second set of data in Figure 7, by matching again to the prediction of Equation 5 but now also including the evaporation term. The result is $v_{e,amb} \approx 100$ nm/s, yielding a time constant $\tau_{e,amb} = 1800$ sec, or 30 mins. From Figure 6, we would expect a 100-fold relative increase in concentration to be achievable in a time $T = 6\tau_{e,amb}$, or around 3 hours, in agreement with observations. Reducing this time to (say) 3 minutes would require a 60-fold increase in $v_e$. If this could be achieved, the evaporation-dominated rise would also reduce to $H_e \approx 114/\sqrt{(60)} \approx 15$ mm, allowing concentration to be achieved rapidly in a compact column.

**Apparatus for enhanced evaporation**
The necessary increase in $v_e$ was obtained using a rapid air stream. Figure 8a shows a schematic and 8b a photograph of the experimental apparatus. The substrate consisted of a 2 mm wide paper strip, attached to a plastic support using PVC tape. The longer left-hand (LH) section was arranged as a horizontal cantilever, while the RH section was bent through a right angle to allow immersion in a small, machined reservoir used to contain the source. This arrangement allowed an evaporation section up to 40 mm long, fed by a shorter (5 mm) capillary transport section. The cantilever was surrounded by a transparent laminar flow duct, which had an air intake at the RH end and was driven by an axial-flow fan at the LH end. Airflow was induced using a 22 CFM axial fan (D481T-012KA-3, Micronel) driven by a DC supply. We were unable to measure the air velocity induced by the fan directly. However, we have measured its rotational speed using a light source and a photodetector, and found a linear variation with drive voltage. Consequently, we expect a similar variation for the air velocity. At low airflow, long wetted cantilevers tended to collapse. Such experiments were therefore performed with apparatus rotated by $90^\circ$. However, as the airflow rose, cantilevers experienced aerodynamic lift.

![Diagram](image)

Figure 8. a) Schematic and b) realisation of apparatus for rapid concentration, showing Brilliant Blue concentrated at an intermediate point along a strip of chromatography paper.

**Airflow-enhanced evaporation**

In separate experiments, the evaporation rate was quantified by measurement of mass loss using a microbalance (BasBal 240, Mettler). The inset to Figure 9 shows the apparatus used, which had a larger fluid reservoir, a vertical column with a larger duct, and a wider (20 mm) paper column.
Figure 9 shows the results. The rate of mass loss varies linearly with fan drive voltage, being enhanced approximately ten-fold compared with the background rate using a 10 V drive. Greater enhancement was achieved using larger voltages and the smaller duct of the final rig.

Evaporation velocity was estimated by measuring the capillary rise at different fan drive voltages and carrying out a best fit to the theoretical variation. To do so, proportionality was assumed between $v_e$ and $V$ as described above, and the column length was corrected to include the capillary transport section. Figure 10 shows the comparison between experiment and theory thus obtained. There is clearly reasonable qualitative agreement, and the results shown that an evaporation velocity $\approx 50$ times the ambient value can indeed be achieved if the effective column length can be reduced below $\approx 2$ cm. The main feature preventing further increase in $v_e$ is that the tape seal attaching the cantilever tended to lift in very rapid airflow, opening parallel capillary transport channels along the sides of the paper strip.
5. **Concentration and separation**

In this section, we describe the characterisation of experimental concentration by image processing and present results for concentration and separation.

*Image processing*

Concentration was quantified optically. A 40 mm long white InGaN LED array (KWB-R3912W-1W, LuckyLight) behind the substrate was used for illumination and a camera for photography, allowing relative dye concentrations to be extracted via MATLAB-based image processing [60]. Figure 11 shows an example panel display, for a concentration experiment involving Brilliant Blue in isolation. A baseline image (Figure 11a) is compared with a later image (Figure 11b). Guidelines are used to select areas for analysis and correct registration. Variations of R, G and B pixel brightness are extracted along the strip (Figures 11c and 11d). Here, the R channel is significantly reduced, while the G and B channels are largely unaffected. The transmission variation is found from the ratio of R channel brightness in the two images (Figure 11e). Relative concentration (referenced to the value at $x = 1$) is extracted using the Beer-Lambert law (Figure 11f), and compared to the $1/x$ variation of the hyperbolic ramp (dashed line).

![Figure 11. Image processing used to extract dye concentration: a) and c) initial image and initial spatial variation of the R, G, and B channels; b) and d) corresponding results for a later image; e) and f) spatial variation in relative transmission and concentration.](image)

In practice, the extracted value is sensitive to experimental factors: small variations strip position caused by airflow (which may be corrected by monitoring brightness in dry regions) and by the
change in transmission caused by wetting (which reduces scattering at short wavelengths). For example, the B and G channel values are higher in the wetted region between the dashed lines in Figure 11c than in Figure 11d. This effect can be corrected by calibration. Similar processing was carried out to extract the concentration of Tartrazine in isolation, using the B channel. Quantitative extraction of concentration was less effective for mixtures, because of residual overlap of absorption spectra.

Concentration

Experiments were carried out with substrate mounted both vertically and horizontally, and using both pre-wetted and un-wetted substrates. Here we present results only for dry, horizontal substrates, which gave the fastest concentration and simply required injection of dye through the tape covering the reservoir. However, at low airflows, when the filling time was a fraction of the concentration time, the results obtained from wetted and un-wetted substrates became indistinguishable, and too slow for practical use. Concentration factors were deliberately limited to avoid saturating the substrate. Figure 12 shows typical images of a 30 mm long strip (rotated through 90° degrees for presentation purposes) recorded at 1.5 minute intervals during an experiment carried out using Brilliant Blue at a fan voltage of 20 V (which required a drive power of 15 W). There is little colour change during the filling stage. However, after around 6 minutes, the dye has reached the evaporation-dominated limit (here, $H_e \approx 20$ mm). At this point, the flow is held up, and a blue band of increasing strength provides evidence of concentration. After 12 minutes, the airflow is switched off, and capillary action drives the concentrate to the tip of the strip ($L \approx 30$ mm), dispersing as it propagates.

Figure 12. Spatial distribution of Brilliant Blue FCF at different absolute times $T$ (in minutes) as indicated.

Figures 13 shows the variation of concentration factor $C_L/C_{L0}$ with normalised position $x$, at different absolute times $T$. The concentration profiles are clearly similar to the dashed-line variations in Figure 5. At short times, the point of maximum concentration advances as the
solvent front propagates; at longer times, it has stabilised at $x = 0$ and a maximum concentration factor of $\approx 120$ is obtained at $T = 12$ mins. However, several departures from the theoretical predictions may be seen. Firstly, the concentration does not apparently fall to zero in unwetted regions of the substrate (A), because the transmission through dry paper is slightly higher than that of wet paper. Secondly, there is a shallow hump in the hyperbolic ramp (B). Possible explanations for this effect include variations in the ducted airflow, paper thickness or water content with position. Thirdly there is a small rise in concentration at $x = 1$ (C), almost certainly due to parasitic capillary flow around the edges of the substrate. Despite this, the concentration profiles are largely as expected during both the filling and the concentration phases.

![Graph showing concentration profiles](image)

**Figure 13.** Measured spatial variations of concentration factor of Brilliant Blue FCF at different absolute times $T$ (in minutes) during the experiment shown in Figure 11.

The circles in Figure 14 show the time variation of peak concentration factor. The concentration rate is initially low, but rises on completion of filling in qualitative agreement with the dashed line theoretical prediction of Figure 6. The crosses show results obtained at lower fan voltages (10 V and 5 V, which required drive powers of 2.5 W and 0.5 W, respectively). Similar characteristics are obtained, confirming that the concentration rate can be controlled by the evaporation velocity $v_e$.

At 20 V, the time taken to achieve a 100-fold concentration (10 mins) is comparable to that required using ion concentration polarization, for which the filling time is conventionally ignored. Unfortunately, this time is approximately three times larger than the previous theoretical estimate ($\approx 3$ mins). The obvious explanation is the 5 mm long capillary pumping section between the evaporation region and the reservoir, which acts as flow restrictor. Faster concentration would therefore be achieved if the length of this section could be reduced, for example by patterning a reservoir directly on the paper substrate. The use of dry air, a more
volatile solvent or a thinner substrate would have a similar effect.

Other departures from ideal behaviour have been noted. Variations in airflow cause the stagnation point to wander. Depletion of the reservoir level results in an increase in the length of the capillary pumping section, and a steady reduction in $H_e$. Finally, high concentrations of adsorbate saturate the substrate. All three effects cause a reduction in the apparent concentration. The first two should be controllable by careful design. However, the third is inherent, especially for mixtures containing solutes with very different retardation factors. The solute with the largest $R_f$ will concentrate first, and preferentially occupy adsorption sites. Saturation effects may then lead to a ‘banded’ concentration profile.

Figure 14. Variation of relative concentration of Brilliant Blue FCF with absolute time $T$ at different fan drive voltages.

Concentration data are subject to several errors. The concentration factor is affected by inaccuracy in optical measurement of relative transmission, due primarily to curvature of the flow front and motion of the paper cantilever. From variations in the saturation concentration measured in a small number of experiments we estimate this error as ± 15%. The time taken to achieve a given concentration is affected by any variation in ambient conditions. From the variation in relative humidity in our experiments, we estimate this as ± 100 x 5.5/46 = 12%.

Separation

Separation experiments were carried out using a light green mixture of Brilliant Blue and Tartrazine. Figure 15 shows images of a 40 mm long strip (again, rotated through 90° for presentation) recorded at different times $T$ in minutes. Brilliant Blue reaches the hold-up point (here, at $H_e \approx 17$ mm) slightly faster than Tartrazine, and concentrates more sharply. After 10 minutes, the airflow is switched off, reducing the evaporation rate almost to zero. Capillary
action then drives the concentrate away from original stagnation point towards the tip of the strip ($L = 40$ mm), separating the blue and yellow dyes as it propagates due to the difference in retardation factors noted in Section 4 ($R_f \approx 1$ and $R_f \approx 0.88$ for Brilliant Blue and Tartrazine, respectively). Of course, the solvent here is itself a dilute dye mixture, so complete separation will never be possible. In addition, ambient evaporation allows further concentration.

![Figure 16](image)

Figure 16. Spatial distribution of Brilliant Blue FCF and Tartrazine at different times $T$ (in minutes) as indicated. Despite this, extracted concentrations of the individual dye allow clear identification of the two components. For example, Figure 16 shows concentration profiles at $T = 15$ mins. Here the original stagnation point is at $x = 0$, and capillary flow has driven the concentrate towards the substrate tip at around $x = (17 \cdot 40)/17 = -1.35$. These results show that at least partial separation is possible and confirm the ability of controlled evaporation to allow complex operations on the same unpatterned substrate. However, further work is required to control separation and quantify the two dye concentrations more accurately.

![Figure 16](image)

Figure 16. Measured spatial variations of concentration factor of Brilliant Blue FCF and Tartrazine at $T = 15$ minutes during the experiment shown in Figure 15.
6. **Conclusions**

We have demonstrated rapid concentration of water-soluble, nonvolatile solutes on paper using airflow-enhanced evaporation. The results show good agreement with 1D theory for a two-phase system with a linear adsorption isotherm. In contrast to pervaporation-based microfluidic concentration, the introduction of a stationary phase renders concentration solute-dependent. However, the elimination of the membrane increases the rate of evaporation, which in turn increases the speed of concentration. We have shown that the maximum theoretical value of $Pe$ depends only on material parameters, and that chromatography paper offers excellent performance. Thin-layer substrates would also be suitable, especially if these may be nanostructured to allow large capillary rise heights. Potential applications include concentration of biofluids, when detection might be carried out using colorimetric indicators located near the tip, or by paper spray mass spectrometry. However, in contrast to ion concentration polarization, the concentrate may also be held up at an intermediate stagnation point if the evaporation rate is high enough, and then released to allow capillary pumped operations such as analysis or separation.

Incorporation of preconcentration into a lab-on-paper device such as a lateral flow assay (LFA) would require several modifications. The sample pad must be enlarged to store a larger fluid volume, and sealed to prevent direct evaporation. A short open zone must be provided before the conjugate pad to allow evaporative concentration, with the LFA substrate mounted as a cantilever in a ducted airflow. Operation would then take place much as shown in Figure 12, with the sample being first concentrated and then released for analysis. However, careful control and calibration would be required for consistent performance.

Further work is required to investigate non-linear effects, especially saturation (which limits concentration of single analytes) and competitive adsorption (which might prevent concentration of weak components of a multi-analyte system). This work is in progress.

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