

# Theory of sedimentation and centrifugation

[plc@badice.co.uk](mailto:plc@badice.co.uk)

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## Abstract

Floral honey contains pollen grains and these can be extracted from a sample of honey, diluted with water, either by sedimentation due to gravity or by the use of a centrifuge.

This paper presents the theory behind these two techniques and derives the following equations predicting the times for sedimentation using these methods.

Sedimentation

$$T = \frac{9l\eta}{2r^2(\rho_p - \rho_s)g}$$

Centrifugation

$$T = \frac{9l\eta}{2r^2(\rho_p - \rho_s)\omega^2 R}$$

However the use of these equations require knowledge of the density of the pollen, and density and viscosity of honey solution which typically are not known.

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# 1 Disclaimer

This document is written in the hope that it will be informative & useful and the author has taken some steps to ensure the information presented is accurate and reliable. However this publication is provided “as is”, without warranty of any kind. This document must not be assumed to be error free and the author will not be held liable for any loss, damages or injury arising from this document or the information contained within.

# 2 Introduction

Unless honey has undergone stringent filtering, a sample will contain pollen grains originating from the plants visited by the bees as they collected nectar. These pollen grains can be extracted, identified and used to determine the location from which the honey originated.

Typical methods for extracting pollen grains from honey [1] require some 10 g of honey be mixed with water to make up some 30 ml of solution. Pollen is thence collected from the honey solution as follows <sup>1</sup> :

- Sedimentation by gravity.  
The honey solution is added to a beaker or suitable tube, covered to prevent dirt entering, and thence left. The pollen grains then migrate to the bottom of the solution under the influence of gravity over a period of some hours. The remaining fluid (supernatant) is removed, leaving the sample of pollen.
- Sedimentation using a centrifuge.  
Pollen can be collected from the honey solution in minutes rather than hours by dividing the solution into two tubes and placing these on opposing sides of a centrifuge rotor (so they balance one another). Thence on activating the device, the centrifugal ‘force’ caused by spinning the tubes pushes the pollen through the solution to the bottom of the tubes. The the supernatant can again be removed leaving a sample of the pollen.

In practice, the times taken for pollen to be collected using these two methods are based on experience. However in the spirit of curiosity, this paper examines the theory behind these two methods of separation and derives equations predicting how long the separation might be expected to take.

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<sup>1</sup>In reality the procedure is a little more complex than described here, as an attempt is usually made to rinse the pollen collected and thence a second separation is undertaken to produce relatively sugar free samples of pollen.

### 3 Theory of sedimentation

Assume, for discussion, that we have a suspension of particles <sup>2</sup> in a solution of water and this is held in a container. Let the radius of the particles be  $r$  metres and let the solution have a depth of  $l$  metres, see figure 1.

There are three forces acting on each particle, viz

- The force of gravity,  $F_G$   
This is the gravitational attraction we are all familiar with and which causes a dropped slice of toast to fall (typically buttered side down) onto the kitchen floor. Gravity is responsible for keeping the honey solution in the container and for pulling the pollen grains to the bottom of the solution.
- Force due to buoyancy,  $F_B$   
All fluids exert a force of buoyancy on objects that are immersed within them. It is the force of buoyancy that supports a swimmer in a pool and causes a ship to float. Buoyancy acts on all bodies in a fluid, even those that sink. For example a stone thrown in a well will sink, but the weight of the stone will appear less when immersed in the water than when in the air due to the buoyancy of the water <sup>3</sup>.
- Buoyancy will act on the pollen grains in a suspension, trying to push the pollen to the surface of the solution even when the grains sink.
- Force due to viscous drag,  $F_D$   
Any particle moving through a liquid will experience a force opposing its motion due to the viscosity of the fluid. This viscous drag acts in the line of, but opposition sense to, the direction of motion.

To make progress in developing the theory of sedimentation, it is necessary to derive equations for these three forces. Fortunately this is straight forward and the details are given in the following three sections.

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<sup>2</sup>For this paper we are interested in suspension of pollen grains. However the theory is quite general and applies to all spherical bodies. Hence the word ‘particle’ is used in the derivation of equations to emphasize the generality of the approach.

<sup>3</sup>Consider the well known puzzle

Which weighs more, a pound of feathers or a pound of lead?

Both have the same mass (one pound), but the feathers have more volume, displace more air and so have greater buoyancy (the air acts like a fluid). The pound of lead therefore weighs more than the pound of feathers.

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## Beaker containing suspension

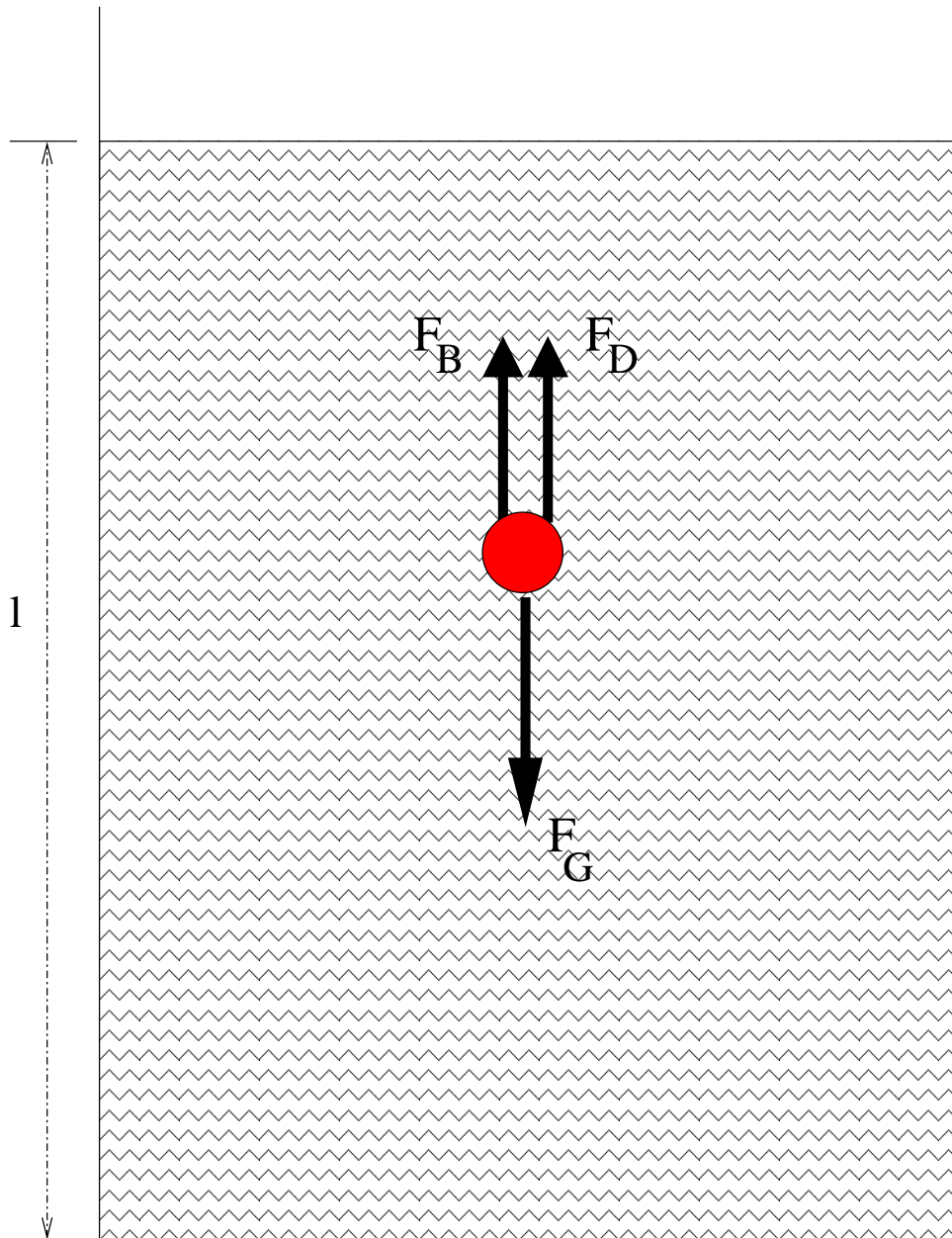


Figure 1: Forces acting on a particle in suspension

### 3.1 Force due to gravity $F_G$

The force acting on any body at the earth's surface due to the action the earth's gravity is described by the equation (1) <sup>4</sup>.

$$F_G = mg \tag{1}$$

where:-

Symbol	Represents
m	The mass of the particle (expressed in kg)
g	The acceleration due to gravity (approximately $9.81 \text{ m.s}^{-2}$ )

The above equation is not very helpful for small particles such as pollen grains as it would be quite a challenge to find scales sensitive enough to determine the mass of a single pollen grain <sup>5</sup>.

Hence we make a change using the concept of density. Density is defined as the mass per unit volume, see equation (2).

$$\rho = \frac{m}{V} \tag{2}$$

where:-

Symbol	Represents
$\rho$	density of particle (expressed in $\text{kg.m}^{-3}$ )
m	Mass (kg)
V	Volume ( $\text{m}^3$ )

If we assume the particle is a perfect sphere (typically not too far from the truth for many pollen grains) then we can calculate its volume from its radius using equation (3).

$$V = \frac{4}{3}\pi r^3 \tag{3}$$

where:-

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<sup>4</sup>Strictly the earth and any other attracted body (such as a pollen grain) both attract each other with an equal force. However the earth is so much more massive than a grain of pollen that the pollen moves to the earth rather than the earth moving towards the pollen.

<sup>5</sup>In reality it is easier to measure pollen density than pollen mass.

Symbol	Represents
r	Radius of a sphere (m)
$\pi$	ratio of circle circumference to diameter (3.14159265 ...)
V	Volume ( $m^3$ )

Combining equations (1), (2) and (3) gives the required result, equation (4).

$$F_G = \frac{4}{3}\pi r^3 \rho_p g \quad (4)$$

where:-

Symbol	Represents
$\rho_p$	density of a particle particle (expressed in $kg.m^{-3}$ )

### 3.2 Force due to buoyancy $F_B$

The starting point in calculating the force of buoyancy is Archimedes' Principle [2] which can be expressed as

“Any object wholly or partly immersed in a fluid is buoyed up by a force equal to the weight of the fluid displaced by the object.”

For a spherical particle of radius r immersed in a solution of density  $\rho_s$ , the buoyancy force  $F_B$  is given by equation (5). This follows as the volume of fluid displaced is  $\frac{4}{3}\pi r^3$  and this must be multiplied by the density of the fluid to give the mass of fluid displaced. Finally this quantity must be multiplied by the acceleration due to gravity g, to convert it to a force.

$$F_B = \frac{4}{3}\pi r^3 \rho_s g \quad (5)$$

where:-

Symbol	Represents
$\rho_s$	Density of the displaced fluid ( $kg.m^{-3}$ )



### 3.3 Force due to drag $F_D$

When a particle moves through a fluid, it has to expend energy in shearing the fluid as it passes through. This is because fluids are viscous, that is they resist movement, The drag on a spherical particle moving through a fluid as it overcomes the viscous drag is given by an expression termed Stokes Law [3] <sup>6</sup> see equation (6).

$$F_D = 6\pi r\eta v \quad (6)$$

where

Symbol	Represents
r	radius of the moving sphere (Units m)
$\eta$	Viscosity of the solution (Units Pa.s)
v	Velocity of the moving sphere (Units $m.s^{-1}$ )

The symbol  $\eta$  <sup>7</sup> is the viscosity of the solution which is a measure of the resistance to flow in a liquid. It has the units of Pascal seconds in the modern SI system of units.

Note, Stokes' law applies to a spherical body. As pollen grains are typically not spherical the results from using this equation will only be an approximation.

### 3.4 Sedimentation of a small sphere in water

Combining the results from the theory of sedimentation outlined above, it follows that if a particle such as pollen is suspended in a container of water, it will immediately experience a force downwards due to gravity and a force upwards due to buoyancy. Assuming the density of the particle is greater than that of the surrounding solution, gravity will 'win' and the particle will start to fall towards the bottom of the beaker.

As soon as the particle starts to move, the drag, will come into operation opposing the motion. As the particle accelerates, so the drag increases, until this drag force just balances the difference of the gravitational and buoyancy

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<sup>6</sup>Stokes law only applies to lamella flow, that is where the regions of fluid move smoothly past each other. Contrast this with turbulent flow where fluid particles move in a random fashion. The assumption of lamella flow can be shown to be valid for pollen moving through water.

<sup>7</sup>In physics it is traditional to use letters of the Greek alphabet  $\rho$ ,  $\eta$ ,  $\theta$  etc to represent physical quantities, probably an attempt by the physics students to impress classics students as the latter hold better parties.

forces. At this stage the particle will continue to fall through the solution towards the bottom of the container moving with a constant terminal velocity  $v_T$ . The value of this can be determined by equating the three forces as shown in equation (7),

$$\frac{4}{3}\pi r^3(\rho_p - \rho_s)g = 6\pi r\eta v_T \quad (7)$$

Rearranging this provides an equation for the terminal velocity of a pollen grain (8).

$$v_T = \frac{2r^2(\rho_p - \rho_s)g}{9\eta} \quad (8)$$

Referring to figure 1, we assumed the depth of the solution containing the pollen was  $l$  metres. Thus the time for all pollen grains in the solution to migrate to the bottom of the fluid under gravity,  $T$ , is given by  $\frac{l}{v_T}$  see equation (9).

$$T = \frac{9l\eta}{2r^2(\rho_p - \rho_s)g} \quad (9)$$

where

Symbol	Represents
T	Time for all particles to fall (seconds)
l	Depth of solution (metres)

### 3.5 Example calculation of sedimentation time for pollen

Given the equations developed in section 3.4, it is time to put some values into these and estimate settling times for pollen in a solution of honey.

Typical values are as follows

Symbol	Description	Value
d	depth of solution	8 cm (0.08 m)
$\eta$	viscosity of water (at 20°C)	1.002 x 10 <sup>-3</sup> Pa.s
r	Radius of (small) pollen grain	20 x 10 <sup>-6</sup> m
$\rho_p$	Density of hydrated pollen	1250 kg.m <sup>-3</sup>
$\rho_s$	Density of water	1000 kg.m <sup>-3</sup>

Substituting the values into equation (9) predicts the time for all the pollen grains to settle at the bottom of the tube to be 368 seconds or just over 6 minutes.

This seems a very short time as the usual practice is to leave honey solution a period of some hours to allow the enclosed pollen grains to sediment. However the above values are not very realistic, specifically

- Viscosity of the solution  
The value of viscosity was taken to be equal to that of pure water at a temperature of  $20^{\circ}C$ . However a honey solution is much more viscous than this as it contains substantial amounts of dissolved sugar. In addition the value of viscosity is very temperature dependent, rapidly rising as temperature falls. Thus a solution of honey left overnight in a typical UK house during the winter months will have a temperature substantially less than  $20^{\circ}$ .
- Density of the solution  
Looking at equation (9) it can be seen that the time for pollen grains to settle is inversely proportional to the difference in the density of the pollen and that of the solution, and thus for an accurate prediction of sedimentation time, it is very important to use accurate values of these two densities. The density of the solution was taken to equal that of water. However this is wrong as the honey solution will have a density greater than this value.
- Density of the pollen  
Pollen density is variable and not well documented. The value taken was that of hydrated corn pollen [4] but it is not known (at least by the author) how representative this is of the densities of floral pollen in honey.

Hence the production of a more accurate prediction of sedimentation time will require more investigation to quantify how viscosity and density of a solution vary with amount of dissolved honey and with changes in temperature. However these are matters for another time and perhaps another paper!

## 4 The laboratory centrifuge

A laboratory centrifuge is a device (possibly hand cranked but typically driven by a motor) which is used to spin fluid samples at high speeds. The resultant centrifugal effect causes particles suspended in the fluid to migrate

to the bottom of the tube as a precipitate (the pellet). The remaining fluid is termed the supernatant.

Centrifuges come in different sizes, with different maximum speed and different uses. Two common types are identified by their types of rotor viz:

- Fixed angle

These hold the tubes in the centrifuge at a fixed inclination (typically about 35 degrees) to the vertical. The most common devices hold eight tubes and they have the advantage of not having moving parts on the rotor (so less to go wrong). This arrangement means that the solute is forced against the side of the tube. This leads to a faster separation of the solute from fluid, but risks abrasion of the particles as they are forced down the wall of the centrifuge tube. Also the end result is a smear along the side of the tube rather than the precipitate forming a neat pellet.

- Swinging bucket

This form of rotor allows the centrifuge tubes to freely swing out towards the horizontal as the device operates. This gives the longest path of particle movement as the centrifugation proceeds and has the advantage that the solid forms in a clear pellet at the bottom of the centrifuge tube.

The following section of paper outlines some of the theory of centrifuge use, and develops equations predicting how long it should take for particles (such as pollen) to separate from a solution.

## 4.1 Theory of centrifuge operation

An example of a fixed angle centrifuge is the Hettich EBA 20 device [5]. This is illustrated in figure 2 which shows the device with the lid raised and two centrifuge tubes placed in the rotor.

The theory of centrifuge operation can be developed as follows. Let a centrifuge tube of length  $l_0$  metres and cross sectional area  $A$   $m^2$  be filled to a height of  $l$  metres with fluid of density  $\rho_s$ . Let the fluid contains a suspension of spherical particles of radius  $r$  metres and density  $\rho_p$   $kg.m^{-3}$ . See figure 3.

The tube is inserted into the centrifuge and the device operated <sup>8</sup>. As the rotor spins, an apparent centrifugal force acts on the sample of fluid and

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<sup>8</sup>In practice a centrifuge must always be loaded with an even number of tubes, diametrically opposed containing the same mass of liquid so that the rotor is balanced. Running a centrifuge with a single tube would certainly make the centrifuge vibrate and could quite



Figure 2: Small centrifuge with fixed angle rotor

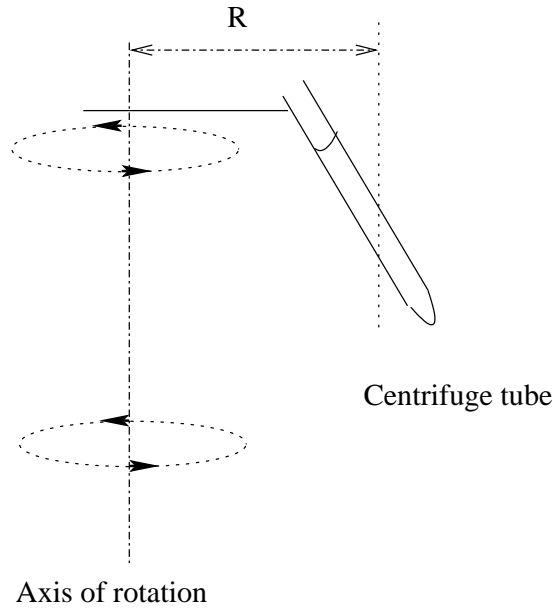


Figure 3: Theory of centrifugation

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the particles inside it, pushing both radially outwards towards the side of the centrifuge tube <sup>9</sup>.

The magnitude of the centrifugal force,  $F_C$  can be show to be [6] given by equation (10).

$$F_C = m\omega^2 R \quad (10)$$

where

Symbol	Represents
$F_C$	Magnitude of the centrifugal force (units Newton)
$\omega$	Angular velocity of the centrifuge (units radian $s^{-1}$ )
R	Radius of the circular motion of the centrifuge (unit metres)
m	Mass of the particle being spun. This could be either the mass of the total solution, or the mass of a pollen grain depending on the context of the discussion.

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easily break the device. However for the point of developing theory, only the forces acting on a single tube will be considered.

<sup>9</sup>Strictly centrifugal force is an illusion caused by the inertia of the spun object which naturally tends to travel in a straight line. However as anyone who has ridden a fair-ground centrifuge can testify, it is a very powerful illusion.

The linear velocity  $v$  of the tube as it moves in a circle is given by equation (11).

$$v = \omega R \quad (11)$$

The force  $F_C$  acts on the particles in the fluid sample, pushing them towards the outside of the centrifuge tube and thence causing them to slide down the tube to form a pellet at the bottom. This force plays the same role as that played by gravity in the sedimentation described in the previous section <sup>10</sup>. Again there is a force of Buoyancy  $F_B$  given by an equation analogous to (5) already derived. However in this case the buoyancy force acts in the opposite direction to the centrifugal force rather than towards the fluid surface. Also there is again a drag force  $F_D$ , (6) as the solution resists the flow of particles and again this acts to oppose the motion of the particles. Equating all forces, we find that when the suspended particles attain terminal velocity, they behave as predicted by equation (12).

$$\frac{4}{3}\pi r^3(\rho_p - \rho_s)\omega^2 R = 6\pi r\eta v_T \quad (12)$$

The time for all particles to collect into the pellet in the centrifuge tube  $T$  is thus given by dividing  $l$ , the depth of fluid in the centrifuge tube by  $v_T$ , the terminal velocity, see equation 13.

$$T = \frac{9l\eta}{2r^2(\rho_p - \rho_s)\omega^2 R} \quad (13)$$

where

Symbol	Represents
$T$	Time taken to precipitate all suspended particles (unit seconds)
$\omega$	Angular velocity of the centrifuge (units $\text{radian } s^{-1}$ )
$r$	Radius of suspended particles (m)
$\eta$	Viscosity of the solution (units Pa.s)
$\rho_p$	Density of suspended particles ( $\text{kg } m^{-3}$ )
$\rho_x$	Density of solution ( $\text{kg } m^{-3}$ )
$R$	Radius of the circular motion of the centrifuge (unit metres)

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<sup>10</sup>Actually gravity also acts on the suspended particles pulling them vertically downwards. However the centrifugal force is typically a thousand or more times that of gravity and so the influence of gravity can be ignored.

## 4.2 Details of Centrifuge Operation

Given a centrifuge, it is interesting to see how the equations derived so far affect its use.

### 4.2.1 Relative Centrifugal Force

The force exerted onto a particle in solution when centrifuged depends both on the speed of the rotation  $\omega$  and on the radius of the centrifuge rotor  $R$ , see equation (10). Since the latter varies with different models of centrifuge, this makes it difficult to compare centrifuge times and speeds across different devices.

This issues is resolved by quoting centrifugation in terms of relative centrifugal force (RCF) rather than rotational speeds. The RCF is defined as the ratio of the force acting on a particle by centrifugation to the force acting on the particle by gravity. Thus the value of RCF says how many g forces a particle experiences when processed in a centrifuge.

The equation for the force on a particle due to gravity was given in equation (1) and centrifugal force is given by equation (10). Dividing these gives the equation for the RCF, see equation (14).

$$RCF = \frac{\omega^2 R}{g} \quad (14)$$

where

Symbol	Represents
$\omega$	Rotational speed (units $\text{rad s}^{-1}$ )
$R$	Radius of the centrifuge rotor (units metres)
$g$	Acceleration due to gravity (units $\text{metres s}^{-2}$ )

However equation (14) is usually rearranged to the form shown (15).

$$RCF = 1.118r \left( \frac{RPM}{1000} \right)^2 \quad (15)$$

where

Symbol	Represents
RPM	Rotational speed (units revolutions per minute)
$r$	Radius of the centrifuge rotor (units millimetres)



Given the EBA 20 centrifuge has a rotor radius of 86 mm and a maximum rotational speed of 6000 rpm, it follows from equation (15) that the relative centrifugal force exerted is 3461g, that is almost 3500 times the force of gravity.

Now the EBA 20 centrifuge holds eight sample tube and the limit of operation is that each tube can hold 15 ml of a fluid of density  $1200 \text{ kg m}^{-3}$ , that is 18 gram of sample. Since the mass of a centrifuge tube is 12.2 gram, this means the total mass of each sample can be up to 30.2 gram. The radius of the rotor is 86 mm. Thus from equation (10) it follows that the centrifugal force experience by a single tube and its contents is 1025 newton, or approximately 105 kg force <sup>11</sup>.

The energy E stored in a single sample of mass m spinning at top speed of in the EBA 20 centrifuge is calculated with equation (16) as 44.09 Joule. Hence the energy stored in all eight samples is 352.7 Joule. If it were possible to capture all this energy and apply it to a single 1kg mass it would be sufficient to fling it some 35 metres vertically into the air.

$$E = \frac{1}{2}m(\omega r)^2 \quad (16)$$

where

Symbol	Represents
E	Kinetic energy of rotation (unit Joule)
r	Radius of the centrifuge rotor (unit metre)
$\omega$	Rotational speed of centrifuge (unit $\text{rad s}^{-1}$ )

#### 4.2.2 Centrifugation of dense samples

A centrifuge will typically be rated with the following information

- Maximum number of samples that can be held  
This will be determined with the number of holes on the rotor, a typical value being 8.
- Maximum allowed volume of each sample V  
For the EBA 20 centrifuge with an E1624 rotor this value is 15 ml.
- Maximum allowed density of the fluid sample,  $\rho_0$   
For the EBA 20 this is  $1200 \text{ kg m}^{-3}$ .

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<sup>11</sup>Considerably more than an English gentleman with interests in microscopy could lift.

- Maximum angular velocity that the centrifuge can operate,  $\omega_0$   
For the EBA 20 this is 6000 rpm or  $200 \pi$  radian  $s^{-1}$ .

The question arises, if there is a need to centrifuge a liquid with density  $\rho_l$  such that  $\rho_l > \rho_0$ , what is the maximum speed  $\omega_l$  that the centrifuge can safely operate. This is given by calculating the centrifugal force exerted by the higher density fluid and ensuring this does not exceed the safe working limit of the device, that is

$$\rho_0 V \omega_0^2 r = \rho_l V \omega_l^2 r \quad (17)$$

Which on re-arranging gives equation (18).

$$\omega_l = \sqrt{\frac{\rho_0}{\rho_l}} \omega_0 \quad (18)$$

where

Symbol	Represents
$\omega_l$	Maximum rotational speed of rotor with high density fluid
$\omega_0$	Maximum rotational speed for unrestricted centrifuge use
$\rho_0$	Maximum density of sample for unrestricted centrifuge use
$\rho_l$	Density of sample where $\rho_l > \rho_0$

Thus if there were a need to centrifuge a sample of liquid with a density of  $1600 \text{ kg m}^{-3}$ , the maximum safe speed for operation  $\omega_l$  would be given by

$$\omega_l = \sqrt{\frac{1200}{1600}} 6000 \quad (19)$$

That is 5196 rpm.

### 4.2.3 Inclination of a ‘swinging bucket’

The theory developed so far relates to a device which holds the centrifuge tubes at a fixed inclination. Thus the radius of the centrifuge is fixed and one can use equation (14) to calculate RCF. However with a swinging bucket centrifuge the effective radius varies with the speed of the device. This is illustrated in figure 4 where, as the rotational speed increases, the ‘buckets’ tend to the horizontal and the value of radius R increases.

We need to derive an equation predicting the inclination of the buckets to the vertical as a function of speed of centrifugation. Referring to figure 5, consider a centrifuge tube filled with a fluid sample and being spun at a

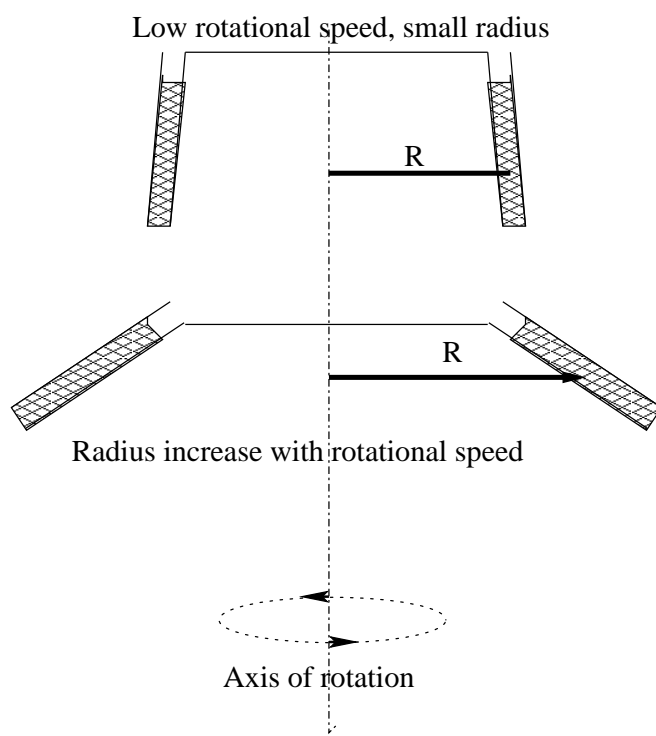


Figure 4: Radius  $R$  varies with rotation speed

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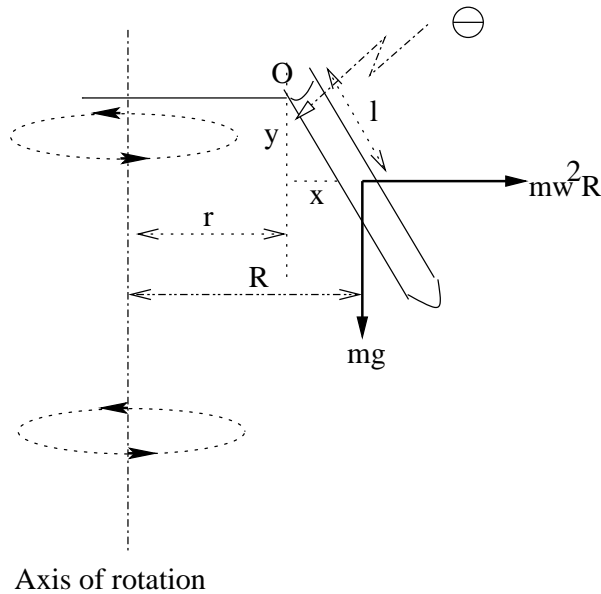


Figure 5: Calculate angle of swing of centrifuge

rotational speed of  $\omega \text{ rad.s}^{-1}$ . As a result the tube makes an angle  $\theta$  to the vertical. This is due to the action of two forces acting at the centre of mass of the tube namely the gravitational force  $mg$  acting vertically downwards and the ‘centrifugal’ force  $m\omega^2 R$  acting horizontally. Note the radius of centrifugation  $R$  now has two components namely  $r$ , the radius of the rotor and  $x$  the displacement due to the inclination of the tube. However from the diagram, it can be seen that

$$x = l \sin \theta$$

Where  $l$  is the distance of the centre of mass of the centrifuge tube from the point of suspension,  $O$ . At equilibrium, the turning moments of these two forces are equal about the point  $O$ , where the centrifuge tube is connected to the rotor. That is the clockwise turning moment from the gravitational force

$$mgx$$

is equal to the anti-clockwise turning moment of the centrifugal force

$$m\omega^2 Ry$$

Where the values  $x$  and  $y$  are the displacement of the lines of action of the forces from  $O$ .

Equating these forces and noting again from figure 5 that  $y = l \cos \theta$  gives equation (20).

$$mgl \sin \theta = m\omega^2(r + l \sin \theta)l \cos \theta \quad (20)$$

Rearranging (and remembering that  $\frac{\sin \theta}{\cos \theta} = \tan \theta$ ) leads to the following expression (21)

$$g \tan \theta = \omega^2(r + l \sin \theta) \quad (21)$$

The author could not immediately come up with an analytic solution to this equation <sup>12</sup> and hence a numerical solution was obtained using the Newton Raphson iterative method [7], which is normally expressed as equation (22) below.

$$x_{n+1} = x_n - \frac{f(x)}{f'(x)} \quad (22)$$

However in this analysis, the independent variable is  $\theta$  rather than  $x$  and so the equation will be written as (23).

$$\theta_{n+1} = \theta_n - \frac{f(\theta)}{f'(\theta)} \quad (23)$$

An iterative method of solving an equation proceeds as follows

- The equation to be solved (21) is re-arranged to equal zero, and this is included in formula (23) as the term  $f(\theta)$ . Rearranging gives the function  $f(\theta)$  as

$$f(\theta) = g \tan \theta - \omega^2(r + l \sin \theta) = 0$$

- The equation is differentiated with respect to  $\theta$ , to give the resultant term  $f'(\theta)$

$$f'(\theta) = g \sec^2 \theta - \omega^2 l \cos \theta$$

- A guess  $\theta_0$  is made of the solution of the equation  $f(\theta)$ . Thence formula (23) is used to generate a better guess of the solution  $\theta_1$ . For a rapid convergence, it makes sense to choose the initial guess as near to

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<sup>12</sup>If the reader can, please let me know.

the solution as possible. Since the centrifugal force is typically much greater than the gravitational force, it seems likely that the angle  $\theta$  will approach  $90^\circ$  (that is the centrifuge tube will be almost horizontal). Hence we will take an angle of  $\theta$  as  $89.99^\circ$ <sup>13</sup>. Given we need to work with  $\theta$  in an iterative equation, we need to convert this value to radians and  $89.99^\circ \approx 1.5706$  radian.

- The improved guess is thence again substituted in the iterative formula, to generate an even better guess, and so on until a solution is obtained to the required level of accuracy.

For the solution of equation (21), we need to substitute the expressions derived above to give equation (24)

$$\theta_{n+1} = \theta_n - \frac{g \tan \theta - \omega^2(r + l \sin \theta)}{g \sec^2 \theta - \omega^2 l \cos \theta} \quad (24)$$

To derive a solution, take the following set of values

Symbol	Description	Value
$\omega$	rotational speed	1000 rpm = 104.7 rad.s <sup>-1</sup>
l	half length of centrifuge tube	5.5 cm = 0.055 m
r	radius of rotor	8 cm = 0.08 m
g	acceleration due to gravity	9.81 m.s <sup>-2</sup>

Hence equation (24) reduces to equation (25) and this can be solved iteratively using the initial assumption of  $\theta_0 = 1.5706$ . The working is shown below.

$$\theta_{n+1} = \theta_n - \frac{9.81 \tan \theta - 877.28 - 603.13 \sin \theta}{9.81 \cos^2 \theta - 603.13 \cos \theta} \quad (25)$$

$\theta_n$	$f(\theta)$	$f'(\theta)$	$\theta_{n+1}$
1.5706	48487.3	254512922	1.5704
1.5704	23271.9	62454231.5	1.5700
1.5700	10838.7	15469861.4	1.5693
1.5693	5075.64	4381434.6	1.56814
1.56814	2212.65	1390293.6	1.56655
1.56655	829.8	544054.9	1.56502
1.56502	217.9	294012.1	1.56428

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<sup>13</sup>It is not possible to have  $\theta = 90^\circ$  as the initial guess as  $\tan \theta \rightarrow \infty$  as  $\theta \rightarrow 90^\circ$ .

1.56428	25.0	231026.6	1.56417
1.56417	0.04	223419.9	1.56417

Hence using typical values for rotor radius and length of centrifuge tube, it can be seen that even for quite modest angular speeds (1000 rpm) the centrifuge tube settles at an angle of  $89^{\circ} 37'$  to the vertical. Thus we can assume that for all practical speeds the swinging arms of a centrifuge are horizontal and the effective radius of the device is that of the rotor plus half the length of the centrifuge tube.

### 4.3 Example calculation of centrifugation time for pollen

Given the equations developed predicting the time for segregation of pollen (13), it is interesting to insert some typical values and estimate time for segregation of pollen grains from a honey solution.

Typical values are as follows

Symbol	Description	Value
$l$	depth of solution	8 cm (0.08 m)
$\eta$	viscosity of water (at $20^{\circ}C$ )	$1.002 \times 10^{-3}$ Pa.s
$r$	Radius of (small) pollen grain	$20 \times 10^{-6}$ m
$\rho_P$	Density of hydrated pollen	$1250 \text{ kg.m}^{-3}$
$\rho_S$	Density of water	$1000 \text{ kg.m}^{-3}$
$R$	Radius of rotation	86 mm
$\omega$	Angular velocity	$209 \text{ rad s}^{-1}$ (2000 rpm)

The value for angular revolution of the centrifuge  $\omega$  was derived by taking a typical speed for centrifuge operation (2000 rpm) and converting this to radians per second. The formula for converting revolutions per minute to radians per second is (26).

$$\text{radian} = \frac{2\pi RPM}{60} \quad (26)$$

as one revolution is  $2\pi$  radians and dividing by 60 converts minutes to seconds. Thus 2000 rpm is  $209 \text{ radian.s}^{-1}$ .

Substituting values gives the time to extract pollen from the honey solution is just over a second. However, in practice it takes some 15 seconds for the centrifuge to accelerate to the required speed and the same amount of time to slow down. Thus the prediction is that some 30 seconds of operation should be sufficient to separate the pollen from solution.

As in section 3.5, this value seems very small and typically a solution of honey is centrifuged for some minutes rather than seconds. However once again the values used for density and viscosity are not very realistic and a better prediction waits on better estimates of these parameters.

## Nomenclature

$\eta$	viscosity of a fluid in Pascal seconds
$\pi$	ratio of a circles circumference to its diameter
$\rho_p$	Density of a particle or pollen grain ( $\text{Kg}m^{-3}$ )
$\rho_s$	Density of a solution ( $\text{Kg } m^{-3}$ )
$v_T$	Terminal velocity of a particle ‘falling’ through a solution
g	Acceleration due to gravity $\approx 9.8 \text{ } ms^{-2}$
g	gram
m	Mass of a particle (Kg)
ml	millilitres, a volume equal to one thousandth of a litre
Pa	Pascal, one newton per square metre
r	Radius of particle or pollen grain ( $m^2$ )
RCF	Relative Centrifugal Force
s	seconds
S.I.	Le Systeme Internation d’Unites which form the modern metric system
supernatant	clear liquid left after a suspension has been removed
Viscosity	A fluid property that relates shear stress in a fluid to angular rate of deformation



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## 5 Document History

Date	Author	Notes
18 July 2009	Peter Clarke	First release for comment