changing the image of the fungus.

The Watershed operator was tested on images obtained from the Top-Hat; however, they presented excessive fragmentation of the mycelium, which are already naturally fragmented.

After the Top-Hat, a Threshold operation was carried out transforming the image into a two-level one, by restricting the range of tones from 256 to 2. The threshold makes a cut on the tones of the image, attributing 0 to tones below the cut level, and 255 to those equals or above the cut level. This binarization results in some small noises, which can not be completely eliminated by the Top-Hat operator, and could interfere with the measurement process of the mycelium.

In order to eliminate the remaining noise, an opening operation by reconstruction was used. A marker was used whose effect is to eliminate small objects, without deforming the larger objects [Banon-Barrera94]. The final result is an image, which contains only any mycelium.

3. Morphological Results
After having completed the previous operations, a skeletonization by thinning was applied, which resulted in the representation of the mycelium in the form of lines (skeleton); this representation was useful for obtaining the number of hyphal of the mycelium and its total length [BWMM96].

The total number of hyphal on the mycelium was obtained from the end-points of the skeletonized image, which in turn were obtained by applying the Inf-canonical morphological operator. These points represent the extremities of the hyphal on mycelium; however, as the main hyphal has two extremities, the number of end-points that were obtained was decremented by one.

Figure 3(a) shows a photographed image of the mycelium. Figure 3(b) shows the same image after the segmentation. Figure 3(c) in turn shows the skeletonization by thinning of that image. Figure 3(d) shows the End-Points that identify the number of hyphal (subtracting one from the number of end-points). These images were obtained by using the Khoros environment.

Figure 3a - Original fungus, 3b - Segmented image., 3c - Skeletonized image, and 3d - End-points of the mycelium.
4. Geometric Measurements

The euclidean distance \( d(x, X) \) of a non-empty subset \( X \) in \( E \), is obtained by the minimal distance between the \( x \) and \( y \) points; that is:

\[
    d(x, X) = \inf \{ d(x, y) \mid x, y \in X \} \tag{9}
\]

Considering that a structuring isotropic element of \( \rho \) radius centered on the origin, denoted by \( B(0, \rho) \), the set of eroded points of \( X \) in relation to \( B(0, \rho) \) is given by:

\[
    X \ominus B(0, \rho) = \{ x \in E \mid B(x, \rho) \subseteq X \} \tag{11}
\]

Let \( X^c \) be the complement of \( X \), then:

\[
    X \ominus B(0, \rho) = \{ x \in E \mid d(x, X^c) > \rho \} \tag{12}
\]

Using \( \rho = 1 \) for measurements in pixels, there are two kinds of structuring elements \( B_1(0, \rho), B_2(0, \rho) \) and two euclidean measures forming angles of 45° and 90°; that is, the measurement of the length is obtained considering the connection of points; the points that connect perpendicularly are separated from the points that are connected through inclination.

In Figure 4 the length of the point is obtained by taking into consideration the angle of connection of a pixel with its neighbor; considering a squared form, the perpendicular points will have the same length as each side of the square measuring 1, on the other hand, the diagonal of the square will be the squared root of 2.

The length of the hyphals is obtained in the following way: the number of diagonally connected points are counted, the resulting value is multiplied by \( \sqrt{2} \), and then the number of the other points is added; the obtained value corresponds to the length of the mycelium. Applying two erosions to the skeletonized image carries out the separation of points. First, the points vertically connected are eliminated. Second, those points horizontally connected are eliminated. In this way, the only remaining points are those diagonally connected. Then, the image of the diagonal points is subtracted from the skeletonized image, giving the points horizontally and vertically connected.

From the new obtained images, the calculation of the total length of the hyphals, becomes quite simple. By obtaining the number of points of each image, and multiplying this value by its corresponding coefficient (\( \sqrt{2} \) for 45° and 1 for 90°), then from the sum of the obtained values, the length is obtained.

5. Three Dimensional Visualization

The area was obtained from the number of white pixels in the segmented image, as shown in Figure 3-b. With the area and length values, the value of the mycelium volume was estimated considering the area of the projection of the image as the area of a rectangle:

\[
    V = A_t L \tag{13}
\]

\[
    A_t = \frac{\pi D^2}{4}
\]

where: \( V \) represents the mycelium volume, \( L \) the total length, \( A_t \) represents the transversal area, and \( D \) the mycelium’s diameter. Replacing \( A_t \) in the formula of the volume:

\[
    V = \frac{\pi D L}{4} \quad \text{and} \quad V = \frac{\pi D}{4} A \tag{14a}
\]

Replacing DL by A (the mycelium’s area) and multiplying the resulting equation by L/L, then:

\[
    V = \frac{\pi D}{4} A L \tag{14b}
\]

\[
    V = \frac{A^2}{4} L \tag{15a}
\]

\[
    V = \frac{\pi A^2}{4 L} \tag{15b}
\]

The equation above is therefore an