# ASPECTS OF THE WATER RELATIONS OF FIVE MOSSES FROM SAVANNA AREA OF UPPER OGUN, SOUTHWEST NIGERIA

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Aspects of the water relations of five mosses growing in a savanna area of southwest Nigeria were studied. Experiments were conducted using *Archidium acanthophyllum* Snider, *Bryum coronatum* Schwagr., *Calymperes erosum* Mull.Hal., *Stereophyllum conterminum* Cardot. (=Entodontopsis nitens (Mitt.) W.R.Buck & Ireland and *Thuidium gratum* (P. Beauv.) A. Gaeger. The water content at field capacity ( $WC_{FC}$ ) significantly higher (P <0.05) in *C. erosum* and *T. gratum* than in other mosses and subsequently  $WC_{FT}$  was lower. The RWC (% dry wt) was significantly higher (P<0.05) in *C. erosum* (376%) and *B. coronatum* (351%). After the short term exposure (48 h) to preset relative humidity (RHs), WC was lower in all the taxa except for *T. gratum* that was significantly higher (P<0.05) at 80% RH. During the long term exposure (8 days) to preset RHs the WC of each tested taxon remained constant after 2 days of exposure. The rate of water loss peaked at 4 h of exposure in all taxa and afterward declined. The rate of decline in *A. acanthophyllum* was significantly higher (P<0.05) than in other taxa. Conclusively, mosses in drier areas survived dehydration through morphological characters while those in wetter areas survived through physiological process.

**Keywords:** Archidium acanthophyllum, Bryum coronatum, Calymperes erosum, Inselberg, Stereophyllum conterminum, Thuidium gratum

#### **INTRODUCTION**

Unlike many other land plants, biological properties of bryophytes make them generally vulnerable to environmental changes. These properties include their dependence on relative humidity, substrate moisture, temperature and shade. Mosses are poikilohydric species with thin 'leaves' that equilibrate their water content with the surrounding environment (Noakes & Longton, 1989) thus surviving periods of cytoplasmic dehydration from which they rapidly recover on rehydration. The main causes of bryophyte decline are anthropogenic activities such as direct deforestation, road construction, urbanization and bush burning by humans. In order to live successfully in a disturbed habitat, bryophytes generally have physiological adaptations. As an example, Dilks & Proctor (1979) reported that photosynthesis declines at high water content in Tortula intermedia growing in a dry habitat.

Johnson & Kokila (1970) exposed ten tropical mosses to different preset humidity levels for a few hours and observed two natural groups: those with high and those with low desiccation tolerance. They concluded that the former group may be found in an ecological habitat with low humidity. Glime (2007) defined and distinguished between

drought tolerance and desiccation tolerance. According to this author, drought tolerance is the ability of a bryophyte to survive in seasonality dry habitats while desiccation tolerance is the ability of bryophyte cells to survive water stress when all metabolic systems collapse. Hence, drought tolerance reflects environmental conditions while desiccation tolerance reflects plant internal condition in relation to water availability.

Owing to the biological importance of water for their growth and survival, mosses in dry habitats take advantage of water stress management strategy (Rychnovska & Suarez, 1988) and surprisingly bryophytes in dry habitats are fully hydrated for most of the time when they are metabolically active; they only suffer water stress as they dry out (Proctor, 1999). For instance, the morphology of the moss leaf widely aids water retention (Bazzaz, et. al., 1970; Gimingham & Smith, 1971; Bayfield, 1973). Some of the features for water storage and movement in mosses are gaps in cell walls (apoplast water), cell to cell (symplast water) and externally(capillary water). Others include colorless (hyaline) cells that serve as reservoir to supply water to photosynthetic cells as in Calymperes. The border-like elongated row of differentiated cells between the margin

and cells toward the mid rib (teniola) also act as support and help to transport water (Reese, 1993).

Comparatively, little is known about water relations of tropical sub-Saharan mosses. On water retention in mosses, Egunyomi (1979) observed in western Nigeria that *Octoblepharum albidum* can hold between 70% and 80% its own dry weight of water but loses 28% of its fresh weight in water per day.

Several methods have been used in the past to estimate the degree of desiccation tolerance in bryophytes. Tallis (1959) used respiration method, while humidity method was used by Lee & Stewart (1971). According to Egunyomi (1979), the use of leaf regeneration was suggested for assessing desiccation tolerance in mosses.

In view of the relatively low water status in the Nigerian savanna, the present work was undertaken to quantify water relations of savanna mosses with particular reference to the relative water content, water loss rate and moisture absorption capacity of *Archidium acanthophyllum*, *Bryum coronatum*, *Calymperes erosum*, *Stereophyllum conterminum* and *Thuidium gratum*.

#### **MATERIALS AND METHODS**

### Study Area

Fresh materials of A. acanthophyllum (Saxicolous), B. coronatum (Saxicolous), C. erosum (Corticolous), S. conterminum (Corticolous) and T. gratum (Corticolous) were collected from four sites, Iseyin (Lat. 8.1 °N, Long. 3.36 °E); Oke-Iho (Lat. 8.1 °N, Long. 3.18 °E); Baasi inselberg (Lat. 8.18 °N, Long. 3.24 °E) and Saki (Lat. 8.41 °N, Long. 3.21 °E) (Fig.1). At each site (Isevin, Oke-Iho, Baasi and Saki) collection of one composite sample from three to six random points were made. Each composite sample from the four sites were combined into one composite and washed under tap water on a metal screen (mesh 20) to remove the adhered soil particles and decayed plants, spread on paper towel and dried under laboratory conditions  $31^{\circ} \pm 1^{\circ}$ C and  $75 \pm 2\%$ 

# Creation of Constant Relative Humidity Conditions

Different relative humidity levels were created inside sample glass using traditional method of varied saturated salt solutions (Winston & Bates, 1960; Johnson & Kokila, 1970; Noakes & Longton, 1989): fused Potassium hydroxide (0% RH), Glucose (55% RH), Sodium chloride (76% RH), Ammonium sulphate (80% RH), Potassium sulphate (97% RH) and deionized water (100% RH). Sample glass tubes (9.5 cm × 2.5 cm) were half-filled with the designated RH solutions and tightly sealed with push-in plastic screw cap and allowed to stand for 24 h to equilibrate.

# Determination of Water Content at Field Capacity $(WC_{FC})$

Clean air dried moss sample (0.10 g) from each composite collection was loosely bound with a thin nylon string and soaked in distilled water contained in Petri dish for 1 h, held up with forceps for 1 minute to drain, placed on sensitive digital Mettler Toledo balance (UK) and weighed as total water held both externally by capillaries (Bowen, 1933) and internally by spaces within the cytoplasm and around the cell walls ( $WC_{e+i}$ ). Afterwards, it was dried at 80°C in air-ventilated oven (Gallenkamp, England) until weight was constant. Water content at  $WC_{FC}$  was calculated using Equation 1.

$$W_{FC} = \frac{w_{s+i} - D_w}{D_w} \times 100 - equation 1$$

where  $W_{FC}$  = water content at field capacity,  $w_e$  = weight of water held externally,  $w_i$  = weight of water held internally.

# Determination of Water Content at Full Turgor $(WC_{FT})$

The internal water content ( $WC_i$ ) was determined by using fresh moss at  $WC_{FC}$ . The external capillary water was removed through centrifugal process at 5,000 rpm on 14.5 cm rotor (Gallenkamp, England) for 5 minutes (Dilks & Proctor, 1979) and carefully blotted out superficially water (Santarius, 1994) with filter paper to give better estimate of full-turgor water content especially from C. erosum with concave leaves. The moss was weighed immediately and

dried in the oven at 80°C until weight was constant. Water content at full turgor ( $WC_{FT}$ ) was calculated using Equation 2.

$$W_{FT} = \frac{[w_{\varepsilon+i} - w_{\varepsilon}] - D_W}{D_W} \times 100$$
....equation 2

### Determination of Relative Water Content (RWC)

The relative water content (*RWC*) of each moss sample was also calculated by using modified formula of Beckett (1995), using *Equation 3*.

$$RWC = \frac{(w_{FC} - w_{FT}) - D_w}{D_W} \times 100....equation 3$$

# Determination of Short Term Absorption Capacity of Samples at a Preset Constant RH

Fresh samples (5 g) at full turgor (internally held water) were suspended with the aid of thin plastic string half way above the preset RH solutions (0%, 55%, 75%, 80%, 97% and 100%) and sealed with push-in plastic screw cap, allowed to stand for 48 h. Afterwards, each sample was weighed ( $W_{i+a}$ ) and immediately oven-dried at 80°C until weight was constant to determine amount of absorbed water. Oven dried samples were cooled in a desiccator containing CaCl<sub>2</sub> and their dry weights ( $D_w$ ) were determined. A filter paper of similar weight was used as control and treated the same way as the tested samples. Absorbed water ( $W_A$ ) was determined using Equation 4.

$$W_A = \frac{[(w_i + a) - w_i) - D_w]}{D_w} \times 100 \dots equation 4$$

Where  $w_a$  = weight of water absorbed  $D_w$  = dry weight,  $W_A$  = absorbed water,  $w_i$  = weight of water held internally.

Eight replicates of samples for each determination were set up under ambient conditions (31°  $\pm$  1°C and 75  $\pm$  2% RH).

### Determination of Long Term Absorption Capacity of Samples at a Preset Constant RH

Air-dried sample shoots (0.1 g) were suspended in a humidity tube with the aid of thin plastic string half way above the RH solutions and resealed. Changes in total fresh weight (water content at field capacity ( $WC_{FC}$ ) were recorded every 24 h, repeatedly by removing and replacing the samples in the sealed tubes. Eight replicates were set up under ambient conditions (31°  $\pm$  1°C and 75  $\pm$  2% RH).

### Determination of Water Loss Rate (WLR)

Discs (dia. 6 cm) of cleaned samples were soaked in Petri dish, which contained distilled water, for 1 h. Each sample disc was suspended with the aid of forceps for 1 minute to drain and carefully blotted in pads of filter paper to attain full turgor state then placed in pre-weighed evaporating glass disc (9 cm dia.) and weighed immediately on Mettler Toledo balance (UK). Subsequent weight losses were recorded at one hour interval for 6 hours and at every other day to the eleventh day. Eight replicates were set up under laboratory conditions (31° ± 1°C and 75 ± 2% RH).

### Statistical Analysis

Data obtained were summarized using mean and standard deviation. Comparison of means were done using One-way ANOVA (SPSS13 Software Package)

#### **RESULTS**

# Water Content at Field Capacity ( $WC_{FC}$ ) and Full Turgor ( $WC_{FT}$ )

The water content at field capacity and full turgor of the five species of mosses is shown in Fig 2. The  $(WC_{FC})$  of C. erosum and T. gratum was significantly higher (P < 0.05) compared with other three mosses. In contrast, water content at full turgor was significantly higher (P < 0.05) in A. acanthophyllum and S. conterminum compared with the other three mosses.

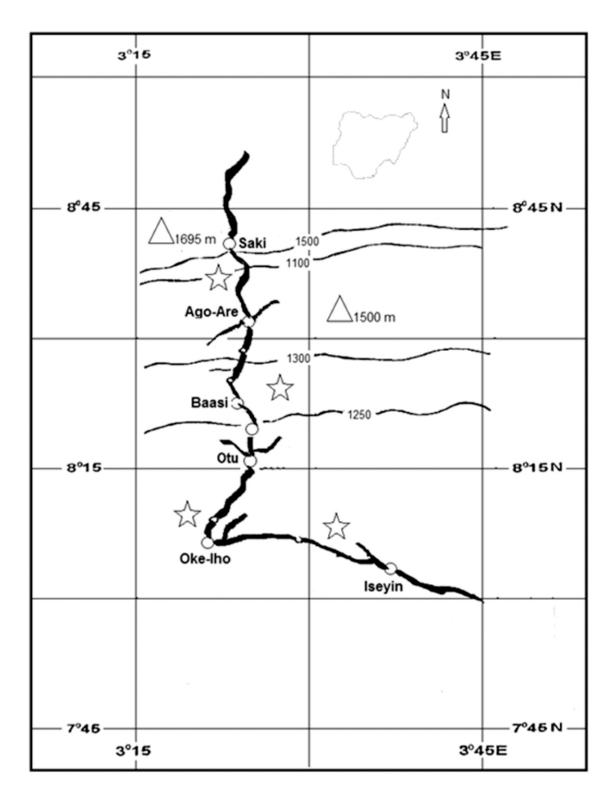


Figure 1. Route to natural habitat of Archidium acanthophyllum, Bryum coronatum, Calymperes erosum, Stereophyllum conterminum and Thuidium gratum. Collection site ( $\bigstar$ ), Mountain peak ( $\Delta$ )

Source: Contour map of Saki, Nigeria 1967 \_\_\_\_\_\_10 km



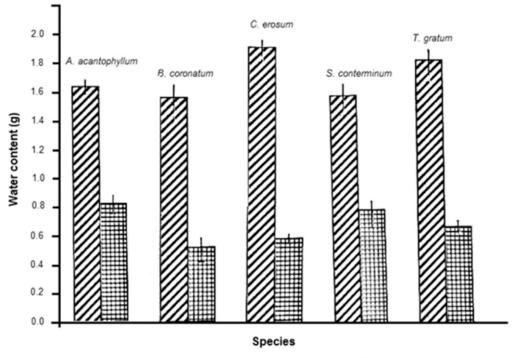


Figure 2 Water content at Field Capacity (FC) and Full Turgor (FT) of Archidium acanthophyllum, Bryum coronatum, Calymperes erosum, Stereophyllum conterminum and Thuidium gratum. Initial wt of sample =  $0.10 \, \mathrm{g}$ , n = 4, mean  $\pm \, \mathrm{SD}$ 

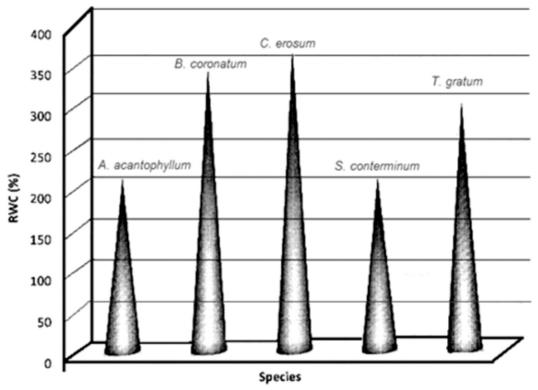


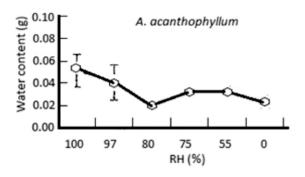
Figure 3 Relative water content of Archidium acanthophyllum, Bryum coronatum, Calymperes erosum, Stereophyllum conterminum and Thuidium gratum . n = 4

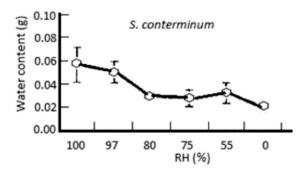
Although relative water content (% d.w.) ranged between 200% and 380% and 60% of the moss samples contained well above 250% relative water content. *S. conterminum and A. acanthophyllum* were below 200% RWC (Fig 3).

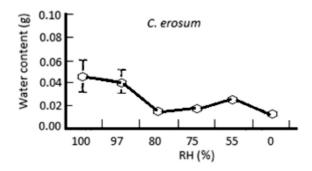
### Short-term Absorption of Water (% d.w.) in a Preset RH

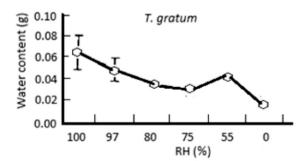
Figure 4 shows a general trend of decrease in internal water content (%d. w.) of the moss

sample at varying preset relative humidity levels after 48 h of exposure. Among the mosses, *T. gratum* absorbed water sufficiently higher from atmosphere of 100% and 50% RH and showed higher significant difference (P <0.05) compared with other three mosses. In contrast, *T. gratum* and the control (filter paper) showed similar ability to absorb water from atmosphere of 100% RH.









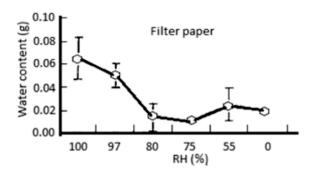


Figure 4. Water content (dry wt) of *Archidium acanthophyllum*, *Calymperes erosum*, filter paper (control), *Stereophyllum conterminum and Thuidium gratum* at different humidity equilibrium constant, n = 8, , mean ± SD

### Long Term Absorption of Water (% f.w.) in a Preset RH

Figures 5-8 show trends of exposure of samples to long-term constant relative humidity conditions. All the four samples absorbed similar

amount of water from the varying RH conditions and attained optimal equilibrium by the third day. There were no significant differences in optimal water content among the samples. ( $P \ge 0.05$ )

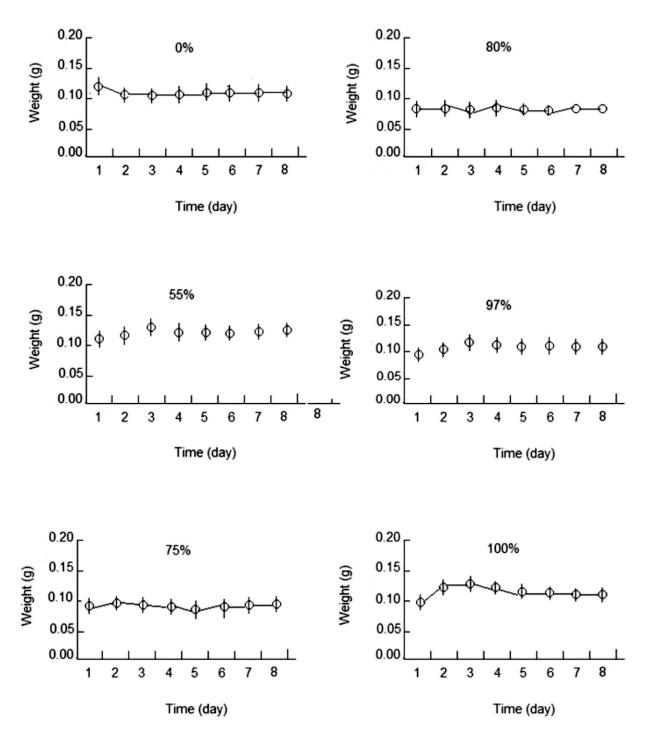


Figure 5. Exposure of Archidium acanthophyllum to different constant relative humidity conditions, n = 8, mean  $\pm$  SD.

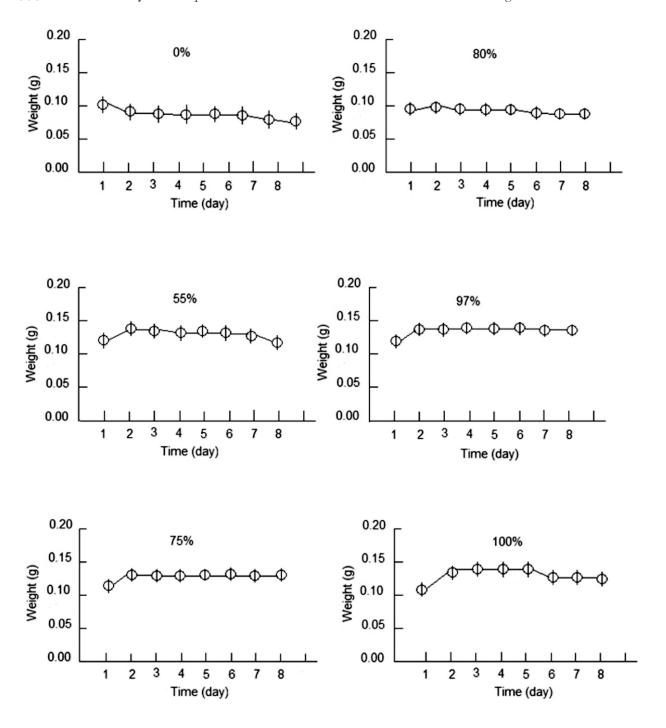


Figure 6 Exposure of Calymperes erosum to different constant relative humidity conditions, n = 8, mean ± SD

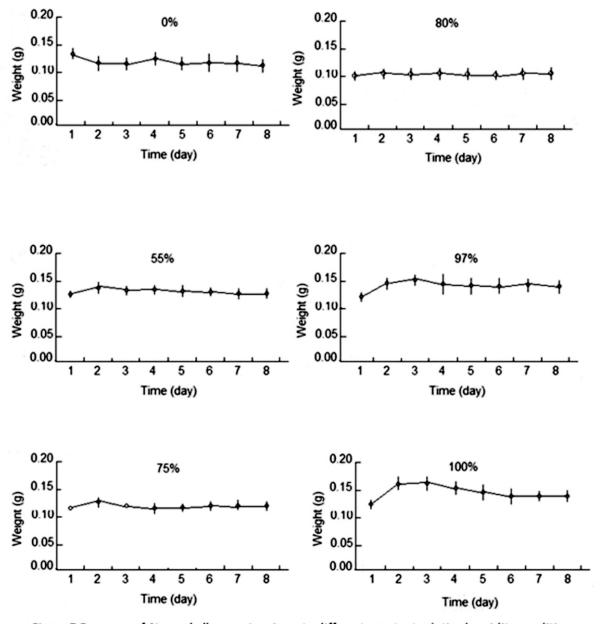


Figure 7 Exposure of Stereophyllum conterminum to different constant relative humidity conditions. n = 8, mean  $\pm$  SD

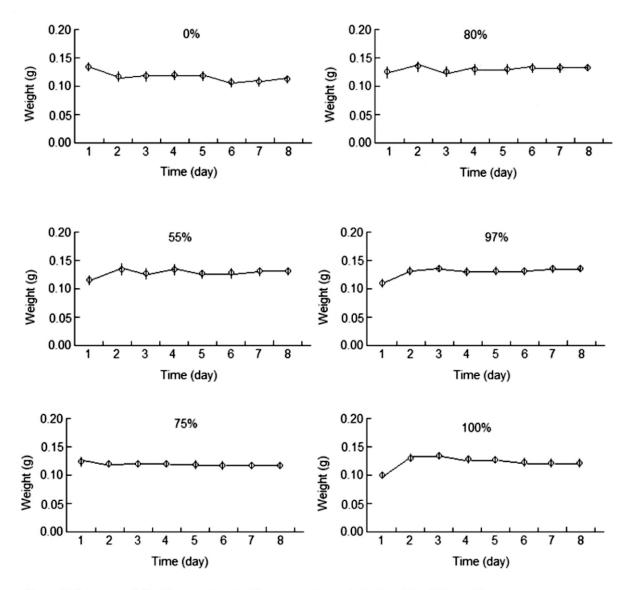


Figure 8. Exposure of *Thuidium gratum* to different constant relative humidity (%) conditions,  $n = 8 \text{ mean} \pm \text{SD}$ 

### Water Loss Rate (WLR)

Figure 9 shows rate of water loss from initial full turgor state of the samples. The maximum rate of water loss occurred the fourth day in each of the samples significant by difference (P<0.05) compared with other days.

Figure 10 shows that continuous long term exposure resulted in sharp decline of loss between the third and seventh day. The rate of water loss in A. acanthophyllum was significantly higher (P < 0.05) than in the other four samples.

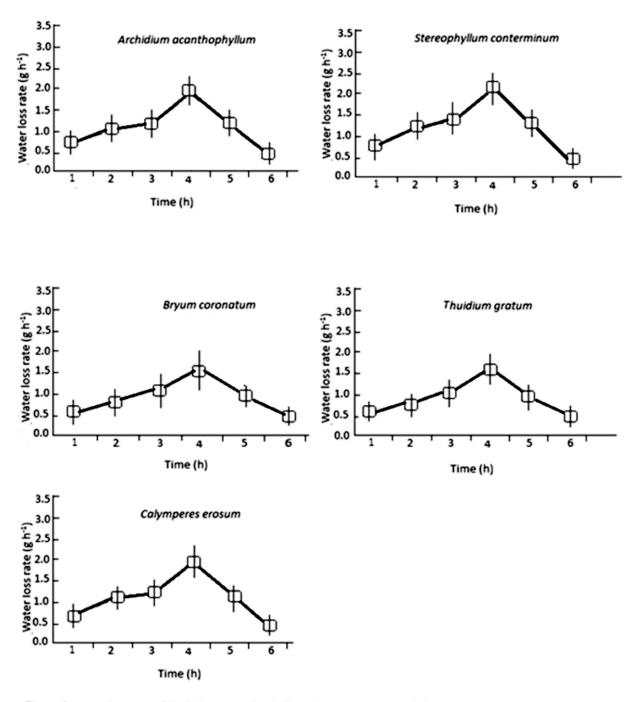


Figure 9. Water loss rate of Archidium acanthophyllum, Bryum coronatum, Calymperes erosum, Stereophyllum conterminum and Thuidium gratum. Moss disc dia. 6 cm, n = 8, mean  $\pm$  SD

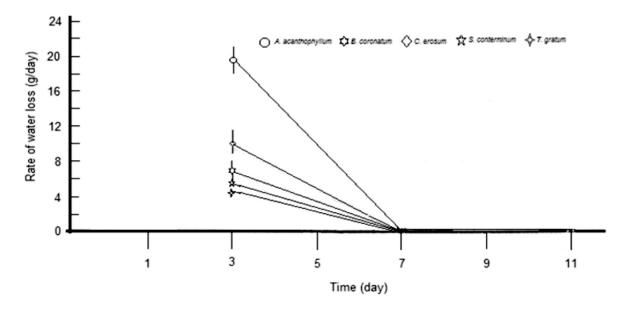


Figure 10. Water loss rate equilibrium point for Archidium acanthophyllum, Bryum coronatum, Calymperes erosum, Stereophyllum conterminum and Thuidium gratum

#### **DISCUSSION**

According to Bates (1998), life form is a critical parameter in bryophyte ecology because of their dependence on the capillary water supplies. Mosses in the study area clearly appeared to have evolved water management strategy. In *A. acanthophyllum* growing in the open inselberg located in the savanna, the arrangement of leaves on the stem and their V-shape facilitates the trapping of atmospheric water vapor and channels it down into the substrate for either storage or to avoid too much of light (teleological evidence).

Apart from the external capillary movement and storage, water may be stored within the cell walls (apoplast water), and in cytoplasm (symplast water). This was evident in A. *acanthophyllum* and S. conterminum that retained 50% of their  $WC_{FC}$  at full turgor. This observation is in agreement with the findings of many authors that leaves of mosses aid water retention (Bazzaz, et. al., 1970; Gimingham & Smith, 1971; Bayfield, 1973). In this work, the lower **RWC** of A. acanthophyllum and S. conterminum (200%) was a hint that these taxa are poikilohydric i.e. they dry up as their habitat dried up and resume normal metabolism after rehydration (Richardson 1981; Proctor 1990). Similarly, A. acanthophyllum and S. conterminum were indicators of xeric habitat (Hernández-Garciaetal., 1999). The rock dweller, A. acanthophyllum, had the lowest **RWC** and fastest **WLR** among the five taxa.

Clausen (1952) observed positive correlation between relative humidity and the distribution of some liverworts. The influence of relative humidity on the distribution of mosses in this work was not surprising. The ability of *C. erosum* to absorb no water at lower humidity level restricted the taxon to wet and waterlogged substrates especially in trunk pockets of Elaeis guineensis Jacq. The lower WC at FT state was an indication that the species utilized water in the substrate mainly by capillary method and thus restricting it to wet or waterlogged habitats exclusive of those with higher (A. acanthophyllum and S. conterminum) that displayed ability to retain more water within the cells and usually established in moist to dry habitats as observed in the field. For these reasons, taxa with higher WC at FC (C. erosum and T. gratum) usually have lower WC at FT.

According to Oliver *et al.*, (2000) desiccation tolerance is a worldwide mechanism in which conserving water and recovering from its loss makes bryophytes more unique than vascular plants. The intermediate taxon, *B. coronatum* used both external capillary water from the substrate and internal water of the cells. The two fold sources of water for *B. coronatum* explain waste ground as its natural habitat. Johnson & Kokila (1970) reported that *Calymperes dozyanum* had lower resistance to desiccation while *Bryum coronatum* had higher resistance. In the present

work, *C. erosum*, *B. coronatum* and *T. gratum* were predisposed to avoiding environmental drought by efficiently holding external capillary water while *A. acanthophyllum* and *S. conterminum* survived better with higher internal water content. The present work thus helps in separating the taxa on the basis of their water relations.

#### Conclusion

Mosses in drier areas survived dehydration by morphological features while those in wetter areas survived through physiological process.

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