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Photoacoustic detection of C_2H_4 emission from germinating striga seeds

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Abstract The C_2H_4 production of seeds of the parasitic weed *Striga hermontica* is observed with a laser photoacoustic detection system. The influence of the germination stimulant GR24 on the hormonal C_2H_4 production and on the germination is discussed.

Introduction

Parasitic weeds of the genus *Striga (Scrophulariaceae)* cause severe damage to graminaceous and leguminous crops in tropical and semi tropical areas. The food crop yield is directly affected in these regions due to heavy infestations by these parasites [1]. The root-parasitic and flowering plants germinate in the neighborhood of the roots of the host plant, only in the presence of a germination stimulant which is produced by the roots. Therefore, their life cycle is closely coupled to the life cycle of the host plant, which ensures that a suitable host plant is available for further development of the parasite.

The determination of the natural germination stimulants and their chemical analogues is of interest as it can be used for controlling the unbridled germination of these plants. Natural germination stimulants found so far are not suitable for controlling the germination of *Striga*; they are either too unstable (dihydrosorgoleone, fig 1) or the preparation is too difficult to produce them in large quantities (strigol) [2].

The synthetic analogue GR24 of strigol can be produced in sufficient quantities [3]. It is hypothesized that GR24 owes its effect to eliciting the biosynthesis of ethene [4].

In this contribution we study the germination of *Striga* under addition of GR24 and ACC (1-aminocyclopropane-1-carboxylic acid, the precursor of ethene in biological processes).

In previous gaschromatographic studies the time development of the ethene production could not be observed due to the low sensitivity (5-10 ppb) of the gaschromatograph for ethene.

Experimental scheme

For the detection of C_2H_4 produced during the germination of the *Striga* seeds we use our laser photoacoustic detection scheme as described earlier [5,6] (see fig 2). An infrared CO_2 waveguide

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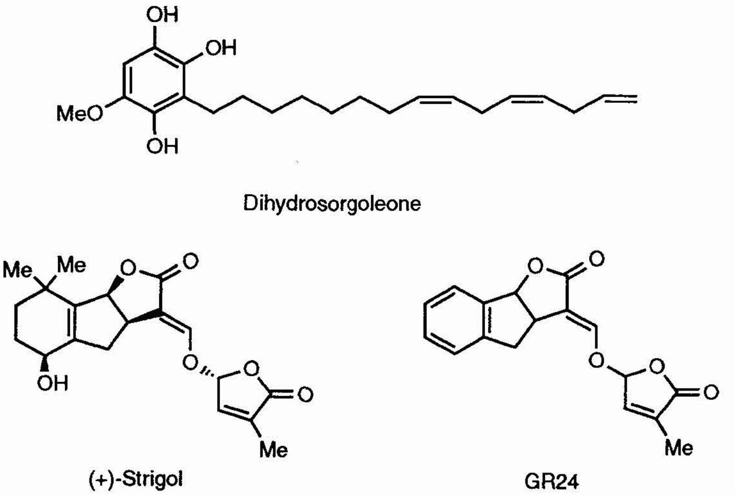

Figure 1.

laser with 90 laser transitions in the 9-11 μm wavelength region is employed as excitation source. Molecules with a vibrational absorption in the laser region like C_2H_4, NH_3,O_3,H_2O show a highly specific and unique absorption pattern [7,8]. Due to the strong absorption of ethene it has been possible to detect very low ethene concentrations in air ($1{:}10^{11}$) [5]. In our photoacoustic set up we have a three order of magnitude higher sensitivity compared to gaschromatographic methods and by flowing air (flow rate 1 l/hour) through the sampling volume we can avoid accumulation of gases in the sampling volume which could react with the biological samples[5-7].

Before air entered the sampling cell it was passed through a catalyst in order to remove hydrocarbons. Between the sampling cell and the photoacoustic cell a KOH based scrubber was placed in order to remove CO_2 from the air and an additional cooling trap (-150°C) to remove also the ethanol produced by the seeds; air passes through a wire netting at that temperature for optimal thermal contact.

The seeds of *Striga hermontica* are tiny (0.3 mm. long, 0.15 mm. wide); therefore 2400 seeds are inserted in a small cuvet to achieve an observable C_2H_4 production. The conditioning of the seeds to induce germination (14 days at 29 °C.) is described elsewhere in detail [9].

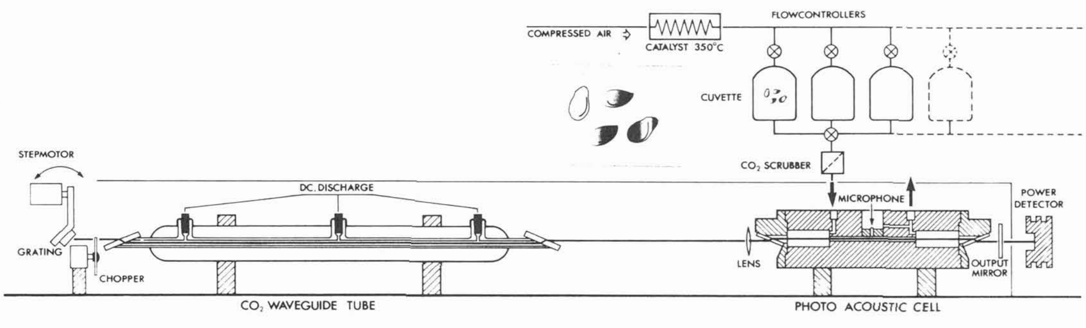

Figure 2: The photoacoustic set up

Results

Gaschromatographic (also photoacoustic) measurements have been made to determine the activity range of GR24, both for germination and stimulation of ethene production (accumulation period 24 hours, fig 3). The germination and ethene production show a very similar response from which can be concluded that both processes are very well correlated.

In order to follow the kinetics of the ethene production during germination the photoacoustic set up was used. *Striga* seeds solely treated with water did not germinate and did not show an ethene concentration level different from an empty cuvet. The seeds treated with GR24 (concentration 0.01 ppm.) showed the first sign of germination after 8 hours. In contrast to this, ethene production is directly observed showing a first peak after 3 hours and a second larger peak between 6 and 20 hours (fig 4).

During the first 6 hours the C_2H_4 production is limited due to the low EFE activity (Ethene Forming Enzyme, responsible for conversion from ACC to C_2H_4). This is shown by adding extra ACC (0.1 mM or 1mM, together with GR24) to the seeds. The production is hardly increased but lasts far beyond 20 hours.

Similarly, the C_2H_4 production during the first 6 hours does not depend on the total amount of GR24. Apparently GR24 does not induce EFE but during a limited period ACC synthase activity. This agrees with the observation of independent production rates for ACC and GR24 (fig. 4); the integrated sum of production is equal to the sum of individual productions if both chemicals are applied.

ACC itself induces germination and ethene production at relatively high concentrations, but less effective than GR24. From this can be concluded that either GR24 increases also the sensitivity of the biological tissue to C_2H_4 or GR24 induces ethene production and germination via two parallel pathways.

The intention is to use GR24 to liberate infested fields from *Striga* seeds by inducing suicidal germination.

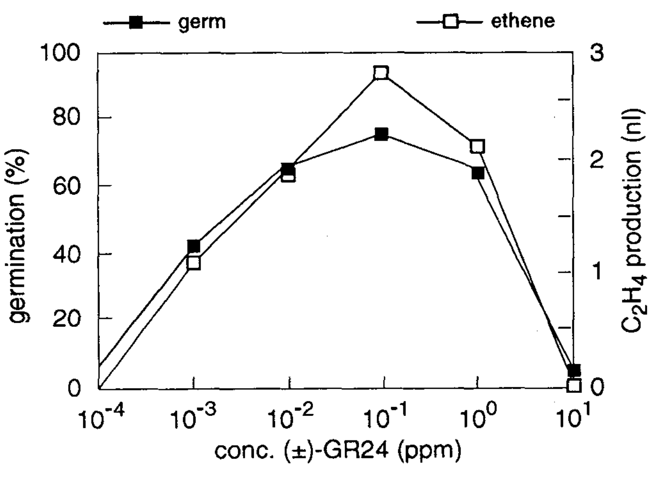

Figure 3: The effect of the GR24 concentration on the germination and the ethene production.

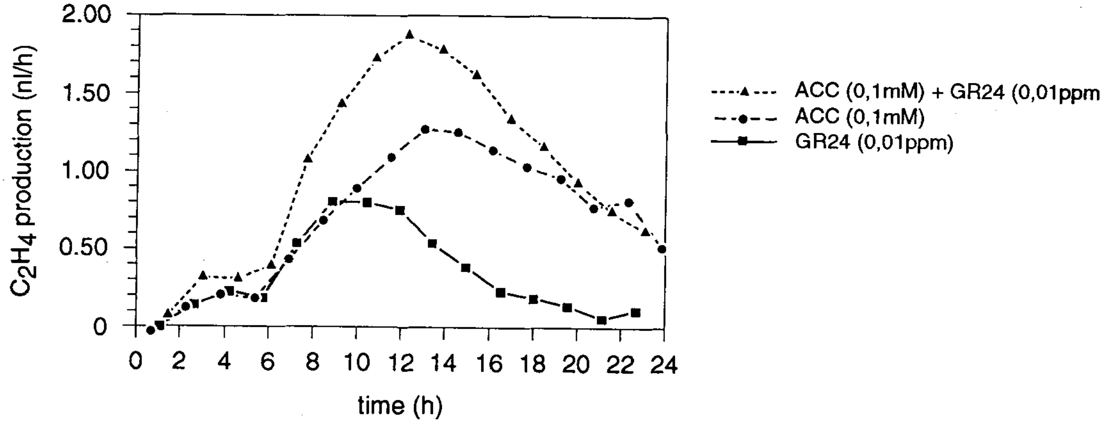

Figure 4: Evolution of the C_2H_4 production of 2400 *Striga* seeds.

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