

are commercially available UV lasers which operate at around 60 kilohertz and with a laser light pulse duration of around 350 picoseconds. Due to their low power, they are very compact. At 60 kilohertz, i.e. with six to twelve laser shots for a mass spectrum, the ion source then provides around one thousand to five thousand analyte ions for one scan. The high mass resolution of these devices means that the most intensive ion signals lie just below the saturation threshold of the ion detector. At the present time, a scanning rate of two gigahertz and a digitization bandwidth of eight bits are normally used. In scanning times of between one tenth of a second and one second, it is thus certainly possible to measure approximately one million to ten million analyte ions; this results in a high dynamic range for this type of measurement.

**[0030]** If, in the future, further developments in electronics lead to significantly higher acceptance rates and larger digitizing bandwidths, corresponding to a higher saturation level, for example eight gigahertz with 12-bit bandwidth, then optical systems could also be used here to focus the pulses of laser light, said systems providing more than one spot per laser light shot by splitting the beam of laser light and therefore considerably increasing the generation rate for ions according to the number of spots.

**[0031]** A time-of-flight mass spectrometer with orthogonal ion introduction is schematically shown in FIG. 1 in combination with an ion source according to the invention. A UV pulsed laser (1) with 60 kilohertz repetition rate delivers finely focused laser light pulses (2) onto samples located on a movably mounted sample plate (4). The beam of laser light is focused to a spot diameter of less than twenty micrometers, preferably less than ten micrometers, onto the sample by a lens system, which is not shown here. It is guided by a movable mirror (3), which allows the vaporization spot to be directed to a different location on the sample between laser shots. This generates plasma clouds (5) containing not only background ions, which stem from the matrix material, but also, importantly, the analyte ions, and which expand continuously into the surrounding vacuum.

**[0032]** The ions can be drawn into an ion funnel (6) and fed to a time-of-flight mass analyzer in the form of a narrow beam (12) via lens systems (7, 9, 11) and ion guides (8, 10). The pulser (13) of the analyzer accelerates sections of the ion beam (12) via a reflector (15) to an ion detector (16). The ions arriving in sequence according to their mass form a time profile of the ion current, whose peaks reflect the current profiles of distinct ion masses. The digitization produces sequences of values, each corresponding to a time-of-flight spectrum. It is quite feasible to scan around 5,000 to 10,000 time-of-flight spectra per second in these time-of-flight mass spectrometers with orthogonal ion injection. Successive time-of-flight spectra are added together to form a sum spectrum. The sum spectrum is then processed with a peak recognition computer program and the flight times of the peaks are converted into a mass spectrum with the aid of a calibration curve.

**[0033]** MALDI ionization is also popular for other types of mass spectrometers, for example ion cyclotron resonance Fourier-transform mass spectrometers (ICR-FT-MS) or electrostatic ion traps, because it scans many samples in a short time and because it is decoupled from separation methods. Although these types of mass spectrometer operate in a pulsed mode, a constant ion current is favorable for them, too. The type of MALDI according to the invention—with short

pulses of laser light with a very high repetition rate and small amounts of material vaporized—can also be used to advantage here.

**[0034]** UV lasers with a repetition rate of 60 kilohertz, a laser light pulse duration of only 350 picoseconds and relatively low power are commercially available and are ideally suited to these requirements if only a single spot per laser light pulse is to be irradiated. They are very compact compared to other UV pulsed lasers used up to now for MALDI.

**[0035]** The processes in the plasma clouds generated by very short pulses of laser light are apparently very different to those in the laser plasmas previously generated for MALDI. Matrix molecules are, for example, decomposed to a far lesser degree and are far less restructured to highly complex ions with widely differing masses. Significantly less chemical background noise is produced from ions formed from matrix molecule fragments than is the case with conventional MALDI. The ions of the unfragmented matrix substances and their dimers and trimers can be recognized much more clearly in the background noise than is the case with conventional MALDI. The background noise, which exerts strong interference up to a mass of approx. 1,000 Daltons with conventional MALDI, does not reach nearly as far into the mass range of the mass spectra when the short laser light pulses are used. The low level of background noise means that the detection limit is shifted favorably to lower concentrations.

**[0036]** FIG. 2 shows an ion source according to the invention in slightly more detail. The beam guidance for the pulses of laser light (25) is slightly different to FIG. 1: the laser light beam here passes through additional apertures in the apertured diaphragms (28) of the ion funnel. It impacts on the sample (23) on the sample support plate (21), which contains a large number of samples (22, 23) overall. The sample support plate can be made of any material; it is favorable, however, if the sample support plate is electrically conductive, or if a metallic core, a metallic backing or an electrically conducting surface can carry an electric potential, which can be used to create a potential difference between sample support plate (21) and ion funnel (28). Moreover, the sample support plate (21) must be made in such a way that the samples (22, 23) can be firmly held and later desorbed without large lumps of sample breaking off. Samples on the basis of thin layers of the matrix material are favorable. Since the desorption is carried out using laser light, the surface of the sample support plate should be reasonably resistant to ablation by the pulses of laser light. The sample support plate (21) can be moved in two directions parallel to the surface which holds the samples (22, 23), so that all the samples (22, 23) in succession can be brought into the spot of the laser light beam (25). In FIG. 2, the specially labeled sample (23) is in the focus spot of the laser light beam (25).

**[0037]** As is the case with normal vacuum MALDI, the MALDI samples here (22, 23) consist of a coating of matrix substance with a small proportion of analyte molecules, only one hundredth of one percent or so. The dilution means that the analyte molecules are not desorbed in the form of dimers or trimers; this is favorable because, once formed, dimers and trimers will not separate again in the gaseous phase. The task of the matrix substance is therefore to keep the analyte molecules in a finely distributed form on the sample support plate (21); to absorb laser light from the pulse of laser light (25), and thereby desorb the sample material in such a way that the analyte molecules are mostly undamaged and individually transferred, either ionized or neutral, to the gaseous state; and