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# Measurement of inflammation in man and animals by radiometry

A. J. COLLINS AND E. F. J. RING*

*Pharmacology Group, School of Pharmacy, University of Bath*

## Summary

1. A radiometer is described, which is sensitive to infrared radiation in the range 0–25 $\mu$m, and which, after calibration with a black body standard can be used as a non-contact, fast reading thermometer.

2. An example of acute joint inflammation in a patient with rheumatoid arthritis is described. The temperatures over the joint measured by radiometry, followed inflammatory changes in the joint effusion.

3. Using rats, the method of measuring inflammation by radiometry was compared with measurements of increase in joint size. Changes measured by radiometry preceded changes shown by increase in joint size.

4. The radiometer method was able to demonstrate the effect of an anti-inflammatory drug, given orally, against carrageenin inflammation.

5. The procedure was found to be an accurate means of measuring inflammation and the anti-inflammatory effects of drugs. It was faster and less tedious than the other methods for the quantitative measurement of inflammation in man and animals.

## Introduction

The clinical assessment of inflammation and the effects of anti-inflammatory therapy in man is difficult to quantitate, and the results often cannot be directly compared with findings from animal experiments. Of the physical signs of inflammation apparent in patients with rheumatoid arthritis, and other forms of inflammatory joint disease, Collins & Cosh (1970) have measured the temperature of the skin overlaying the affected joint by radiometry, and compared the results with some biochemical features of the joint effusion. Using more complex thermographic equipment it is possible to obtain an infrared picture of an inflamed area displayed in shades of black and white on an oscillograph tube (Ring & Collins, 1970). Both methods are possible because of the raised skin temperature over an affected inflamed part, and because skin acts as an almost perfect black body, radiating infrared energy in the range 2–20 $\mu$m (Hardy, 1934). This report attempts to show how the simpler method of radiometry can, by measuring the temperature of the skin over an inflamed area, follow both the physical and biochemical changes of an inflammatory response in man, and that the same method is suitable for measuring inflammation, and the effect of anti-inflammatory agents, in animals.

* *Clinical Research Department, Royal National Hospital for Rheumatic Diseases, Bath*

## Methods

### *Infrared radiometer*

The radiometer used in this work has been described in part by Ring & Cosh (1968). The instrument is manufactured by Heimann GMBH, Wiesbaden, G.F.R. (U.K. Agents, Guest International, Brigstock Road, Thornton Heath, Surrey). It comprises an infrared detector head which is easily movable, and temperature read-out through a meter or plotter package. Radiation in the range 0–25 $\mu$m, which encompasses the known infrared emissivity of skin, is detected by the stabilized bolometer. The scale was calibrated to read temperatures in the range 11–41 $\pm$ 0·2° C, and 26–41 $\pm$ 0·1° C. By plotting the scale calibration against a standard black body radiator at various temperatures, a satisfactory straight line relationship was obtained (Fig. 1). The instrument could therefore be used as a direct reading non-contact thermometer. The response time of the machine was less than 3 s per reading, the lag time being entirely due to the degree of damping used on the meter. The machine is robust, and for all practical purposes was found to be drift free over several months.

### *Area and temperature read, with distance from detector*

The angle of incoming radiation to the infrared detector is 6°. At 10 cm from the subject the detector reads the temperature of a circle 1 cm in diameter. This distance/temperature relationship remained constant, with a small fall off in the temperature reading when the distance of the detector from the source is greater than 20 cm (Fig. 2).

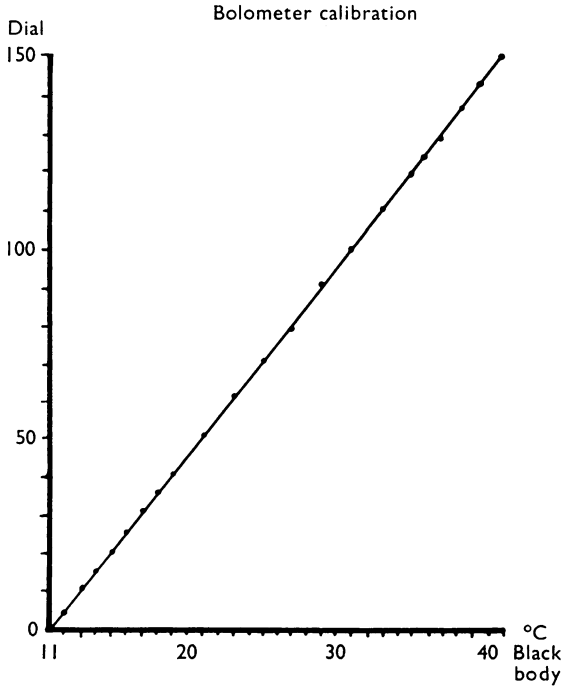


FIG. 1. Curve showing the relationship between a standard black body radiator and bolometer dial readings.

*Temperature readings from human joints*

The human knee joint is a convenient site for the study of the inflammatory response in man. The easy access it affords to the intra-articular space allows easy withdrawal of the synovial effusion produced during acute episodes of inflammation associated with rheumatoid and other arthritides. The patella forms a convenient reference point from which to take serial temperature readings, providing the readings are taken with the leg in full extension. For these reasons, we have mainly studied inflammation of the knee.

Subjects for aspiration of the joint and assessment of the inflammation by radiometry were placed in a temperature controlled room, maintained at 20° C. The clothing covering the joint was removed and the area to be measured was allowed to come to equilibrium with the room temperature for 20 minutes. Temperature readings were taken at 10 cm above a point marked at the centre of the patella. The joint was then aspirated to dryness, and the volume of the synovial fluid noted. A sample of the synovial fluid was centrifuged at 20,000 *g* for 30 min at 4° C to precipitate cellular matter and debris. The cell-free fluid was then stored at −20° C until required. A total white cell count was made on a sample of the uncentrifuged fluid.

Injection of intra-articular anti-inflammatory compounds if they were required, followed aspiration of the joint fluid.

*Temperature readings from inflamed rat joints*

Male Wistar rats (Fisons Pharmaceuticals) of body weight 200–250 g were used. Inflammation was produced either by injection of 0·1 ml of Freund's adjuvant, or by injection of 0·1 ml of a 1·0% solution of carageenin into the left hind foot pad. Batches of five animals were kept in cages with a minimum of sawdust.

The temperature of the foot pad was measured by presentation of the foot to the fixed radiometer detector head (Fig. 3). On contact with the detector there was a

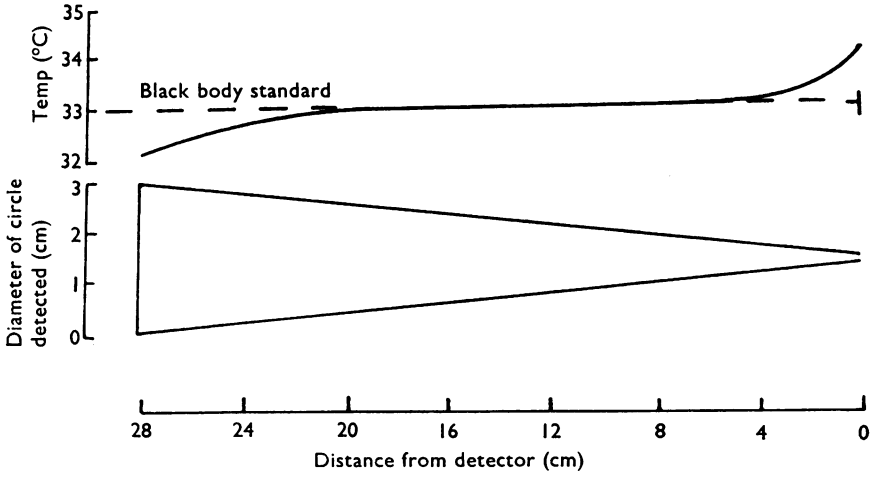


FIG. 2. The upper curve shows the distance/temperature relationship of the bolometer with a heated black body standard, maintained at 33° C. In the lower illustration the diameter of the area detected by the bolometer (represented by the vertical distance between the two lines) is plotted against distance from the detector.

tendency for the rats to grasp the radiometer head with the foot, thus making the contact quite firm. Although slightly higher readings were obtained by this method, this error was constant for all the animals used. The anti-inflammatory compound, Azapropazone (5-(dimethylamino)-9-methyl-2-propyl-1H-pyrazolo (1,2,4) benzetriazine-1, 3(2H)dione dihydrate) (A. H. Robins Co. Ltd.), was given to a test group of rats orally, 30 min before injection of carageenin, and the temperature of the injected foot measured up to 72 h after injection.

The protein content of the aspirated fluids was measured by the method of Lowry, Rosebrough, Farr & Randall (1951).

The circumference of the ankle joint was measured with a modified ' Geigy ' finger measuring loop.

**Results**

As it was not possible to demonstrate dose related drug effects on human subjects, changes produced by single injections only are reported. The results shown in Fig. 4 were obtained from a woman of 69 years with a 20 year history of rheumatoid arthritis, who developed acute inflammation and an effusion in the right knee. The joint temperature was recorded and the joint aspirated. On the days shown (Fig. 4) two intra-articular injections of hydrocortisone, each of 50 mg were given on days 8 and 13, following temperature recording and aspiration.

The observed fall in temperature 24 h after the second intra-articular injection of 50 mg hydrocortisone was effectively demonstrated by radiometry. The anti-

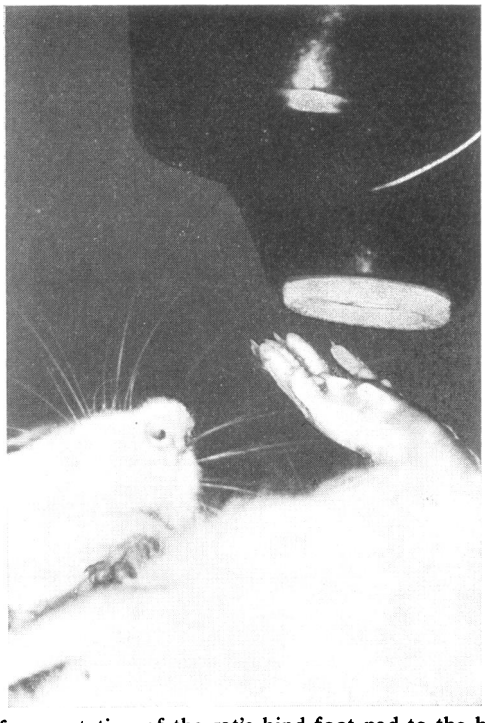

FIG. 3. The method of presentation of the rat's hind foot pad to the bolometer detector head.

inflammatory effect of the hydrocortisone was such that the volume of the joint effusion withdrawn fell to less than 0·25 ml, making determination of protein and the cell content of the fluid impractical. The reappearance of a joint effusion, with a sharp increase in joint temperature, was detected on day 23. The joint temperature and effusion volume were comparable with the results from day 13. No cells were found in the fluid and the protein concentration was still below the highest recorded level.

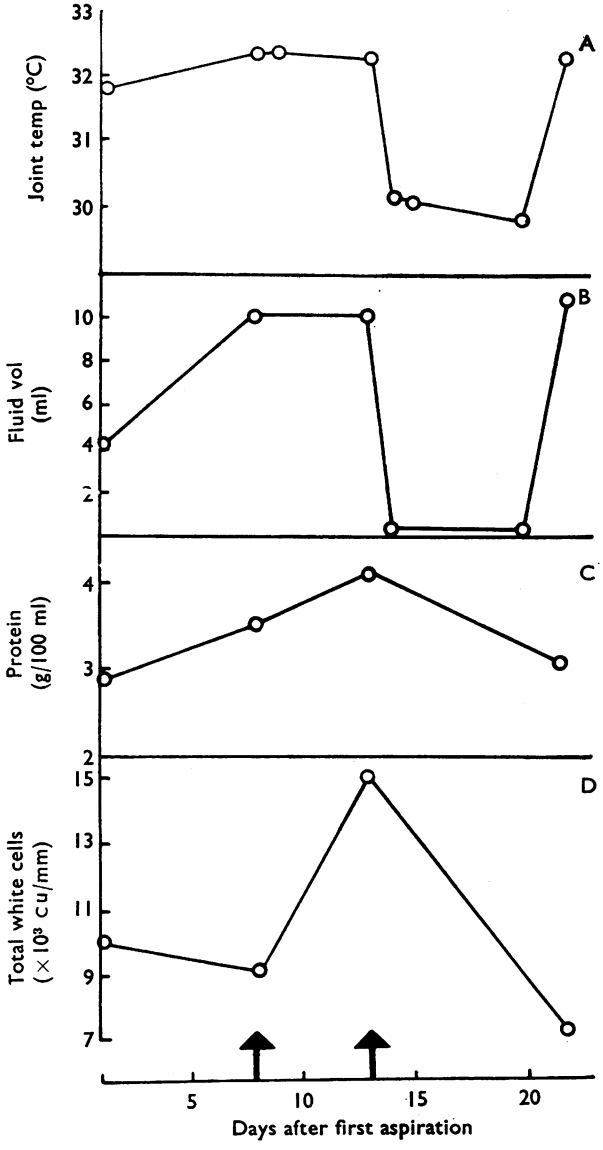


FIG. 4. Comparison of joint temperature with changes in the synovial effusion of a knee joint during the acute inflammation of rheumatoid arthritis. A. Joint temperature, taken by radiometry, from a point at the centre of the patella. B. Volume of the aspirated joint effusion. C. Protein content of the synovial fluid, after centrifugation to remove cellular material. D. Synovial fluid white cell count. Two injections of hydrocortisone (50 mg in 1 ml) were given, intra-articularly on the days marked by arrows.

*Comparison of joint size and of foot temperature measurements by radiometry, in rats injected with Freund's adjuvant*

Measurements of temperature and ankle size of both hind feet in a group of thirty rats were made before injection of Freund's adjuvant into the left hind foot pad, and subsequently for 20 days. The expected inflammation appeared in the uninjected left foot after 10 days and was recorded by both methods of measurement (Fig. 5). There was a good correlation between the two methods of measuring the same inflammation, except that temperature increases tended to precede changes shown by increase in size of the joint. A rapid increase in temperature due to the initial injection into the left foot pad was accompanied by a transient increase in temperature of the uninjected foot and was probably due to a systemic temperature increase due to the injection of a foreign protein. After 20 days neither the injected nor the contralateral foot had reached the initial temperature caused by adjuvant

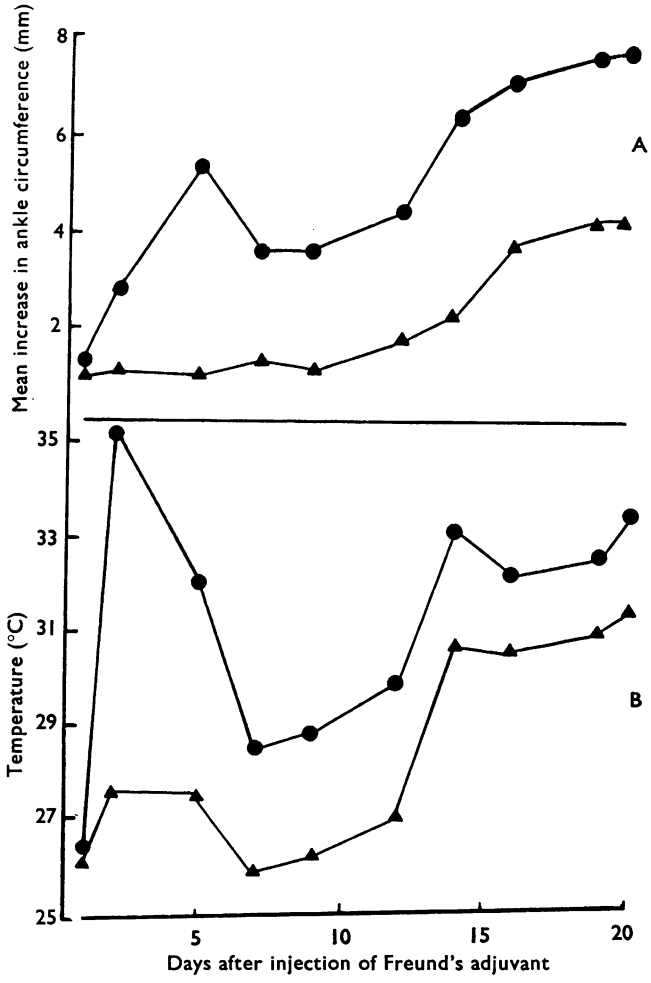


FIG. 5. Measurement of an immune inflammation in a group of thirty male Wistar rats, by: (A) measurement of ankle circumference of both hind feet and (B) radiometry of both hind foot pads. (●——●), Left hind foot, (▲——▲), right hind foot. Inflammation was produced by injection of 0·1 ml of Freund's adjuvant into the left hind foot pad immediately after the recording of the size of both hind ankles and the temperatures of both hind foot pads.

injection, while the ankle circumference measurements had surpassed the initial rise. After the first rise in temperature due to injection, the temperature of the hind feet showed an almost identical pattern of temperature increase.

*Measurement of an anti-inflammatory effect by radiometry*

The temperature of the left foot pad of forty male Wistar rats was recorded immediately before the animals were split into four groups of ten. Three groups of ten rats were given Azapropazone, 50, 100 and 200 mg/kg, in 1·0 ml of 0·1 N NaOH, orally. A control group of animals received 1·0 ml of 0·1 N NaOH by the same route. After 30 min all animals were injected into the left hind foot pad with 0·1 ml of 1% carageenin in sterile saline. The temperatures of both hind foot pads were taken at the time intervals shown in Fig. 6.

The inflammation caused by injection of carageenin in the control group was clearly shown, and the dose related anti-inflammatory action of Azapropazone was demonstrated.

## Discussion

Joint inflammation in man and animals is marked by several parameters such as pain swelling, immobility, and a rise in temperature of the affected part. Since human skin, irrespective of its pigmentation, is an almost perfect radiator of infrared radiation, there is a direct relationship between the temperature and emissivity of this organ. The one example we have quoted of human joint inflammation is typical

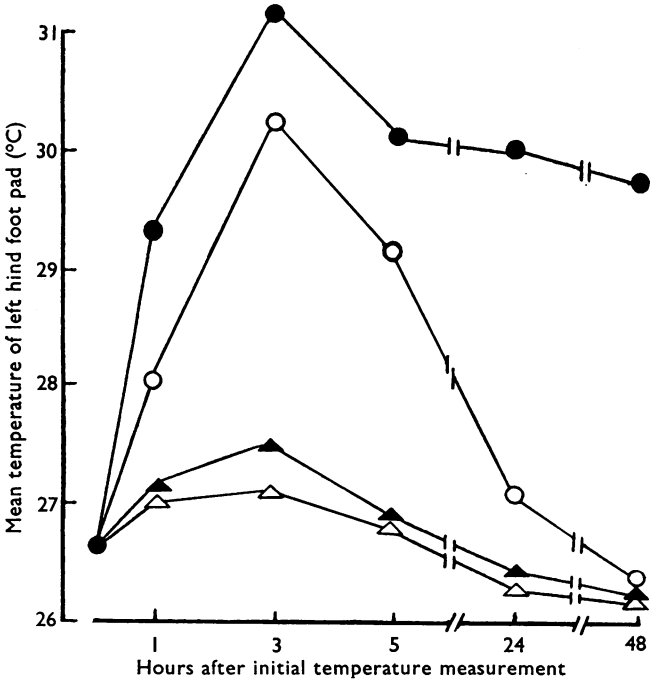


FIG. 6. Anti-inflammatory effect of Azapropazone, given orally against an injection of 0·1 ml of a 1% solution of carageenin into the left hind foot pad of male Wistar rats. (●——●), Control, (○——○), Azapropazone, 50 mg/kg, (▲——▲), Azapropazone, 100 mg/kg, (△——△), Azapropazone, 200 mg/kg. The initial temperature was recorded immediately before the animals were given Azapropazone and 30 min before the injection of carageenin.

of the relationship we have found between joint temperature, measured by radiometry or thermography, and the physical and biochemical inflammatory changes in joint effusion. It was of interest that the first injection appeared only to halt the increase of temperature and the synovial fluid volume, while the protein and cell levels of the fluid increased. A fall in joint temperature and fluid volume was not recorded until after the second injection, which was then accompanied by a drop in protein, and cell levels in the effusion fluid.

It is of particular importance that the pharmacology of anti-inflammatory compounds found in animals should be comparable with results obtained in man. The method of measuring joint temperature by radiometry provides a relatively simple, quantitative method of achieving this end. In contrast, the contact thermistor is less reliable for measuring surface temperature over inflamed tissue, because of its slow response time. The probe has to be secured to the skin, causing probable errors in the recording of temperature.

We have used the infrared technique in assessing inflammation in human joints as it has proved a convenient and accurate method. The technique also has several advantages in the quantitative measurement of inflammation in animals. For routine measurement of inflammation the procedure is faster than methods of measuring joint volume or circumference. Where large numbers of animals are concerned the time saving is appreciable. As a non-contact method, requiring only exposure of the inflamed area to the detector, the procedure does not require the careful alignment of the animal to the measuring instrument. The simplicity of the operation allows the method to give reliable results in the hands of an inexperienced operator. The increase in temperature due to the inflammatory process can in part be attributed to metabolic change with the local increase in blood flow being the major contributing factor. This technique may therefore be applied not only to the screening of anti-inflammatory compounds, but in other cases where measurements of local blood flow are required.

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## REFERENCES

COLLINS, A. J. & COSH, J. A. (1970). Temperature and biochemical studies of joint inflammation. *Ann. rheum. Dis.*, **29**, 386–392.

HARDY, J. D. (1934). The radiation of heat from the human body. *J. clin. Invest.*, **13**, 615–620.

LOWRY, O. H., ROSEBROUGH, N. J., FARR, A. L. & RANDALL, R. J. (1951). Protein measurement with folin phenol reagent. *J. biol. Chem.*, **193**, 265.

RING, E. F. J. & COLLINS, A. J. (1970). Quantitative thermography. *Rheum. Phys. Med.*, **x**, 337–341.

RING, E. F. J. & COSH, J. A. (1968). Skin temperature measurement by radiometry. *Br. med. J.*, **4**, 448.

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